

Botulinum Toxin potentiates cancer radiotherapy and chemotherapy

R. Ansiaux¹, C. Baudelet¹, G. O.Cron¹, J. Segers¹, C. Dessy², P. Martinive², J. De Wever², J. Verrax³, V. Wauthier³, N. Beghein¹, V. Grégoire⁴, P. Buc Calderon³, O. Feron², B. Gallez¹

¹Biomedical Magnetic Resonance Unit, Catholic University of Louvain, Woluwe, Brussels, Belgium, ²Laboratory of Pharmacology and Therapeutics, Catholic University of Louvain, Woluwe, Brussels, Belgium, ³Pharmacokinetic, Metabolism, Nutrition and Toxicology Unit, Catholic University of Louvain, Woluwe, Brussels, Belgium, ⁴Laboratory of Molecular Imaging and Experimental Radiotherapy, Catholic University of Louvain, Woluwe, Brussels, Belgium

Introduction

Structural and functional abnormalities in the tumor vascular network are considered factors of resistance of solid tumors to cytotoxic treatments. To increase the efficacy of anti-cancer treatments, efforts must be made to find new strategies for transiently opening the tumor vascular bed in order to alleviate tumor hypoxia (source of resistance to radiotherapy) and improve the delivery of chemotherapeutic agents. We hypothesized that *Botulinum Neurotoxin* type A (BoNT-A) could interfere with neurotransmitter release at the perivascular sympathetic varicosities, leading to inhibition of the neurogenic contractions of tumor vessels and therefore improving tumor perfusion and oxygenation.

Materials and Methods

To test this hypothesis, BoNT-A was injected locally into mouse tumors (fibrosarcoma FSaII, hepatocarcinoma TLT) and electron paramagnetic resonance (EPR) oximetry was used to monitor pO₂ in vivo repeatedly for four days. Additionally, contrast-enhanced magnetic resonance imaging (MRI) was used to measure tumor perfusion in vivo. Finally, isolated arteries were mounted in wire-myograph to monitor specifically the neurogenic tone developed by arterioles that were co-opted by the surrounding growing tumor cells.

Results

Using these tumor models, we demonstrated that local administration of BoNT-A (2 sites, dose 29 U.kg⁻¹) substantially increases tumor oxygenation (Fig.1) and perfusion (Fig.2), leading to a substantial improvement in the tumor response to radiotherapy (20 Gy of 250 kV RX; regrowth delay for doubling the tumor size was increased of 4.7±1 days for pre-treated group) and chemotherapy (cyclophosphamide 50mg/kg; regrowth delay for doubling the tumor size was increased of 3.9±0.6 days for pre-treated group).

Discussion

This observed therapeutic gain results from an opening of the tumor vascular bed by BoNT-A, since we demonstrated that BoNT-A could inhibit neurogenic tone in the tumor vasculature.

In conclusion, the opening of the vascular bed induced by BoNT-A offers a way to significantly increase the response of tumors to radiotherapy and chemotherapy. In vivo EPR and DCE-MRI are powerful tools to identify new therapeutic ways that can sensitize tumors to cytotoxic treatments.

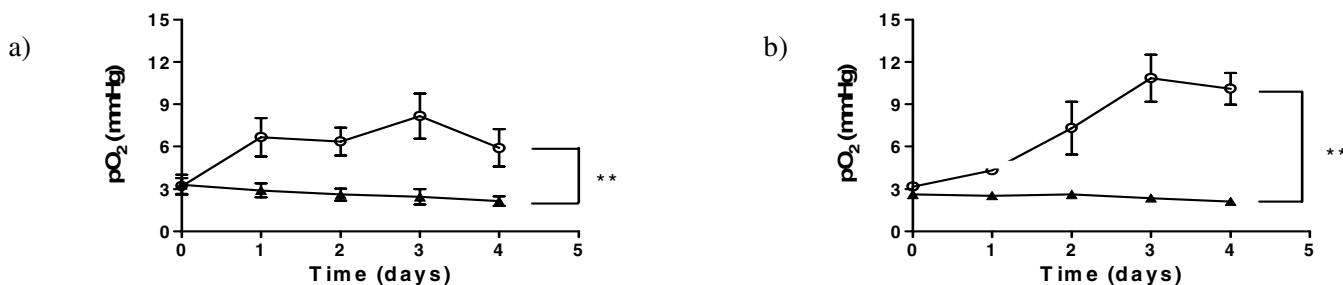


Fig.1 Effect of a single intratumoral injection of BoNT-A on FSaII tumor (a) and TLT tumor (b) oxygenation monitored by EPR Oximetry. $\blacktriangle$ control group (n=8 and 4 for FSaII and TLT respectively). $\circ$, treated group (n=10 and 4 for FSaII and TLT respectively). Note the significant increase in pO₂ 24 hours after the injection with a maximum on day 3. Points, mean; bars, SE; **, P<0.01.



Fig.2 a) Typical MRI images of FSaII tumors showing the perfused pixels 3 days after treatment or control. b) Mean percentage of perfused pixels for treated (n=3) and control group (n=4). Columns, mean; bars, SE. **, P<0.01; ns, not significant.