In Vivo Visualization of Electro-assisted Delivery of Nanoparcticles Using Optical Imaging

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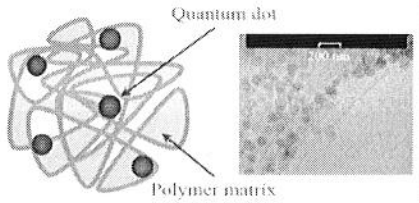
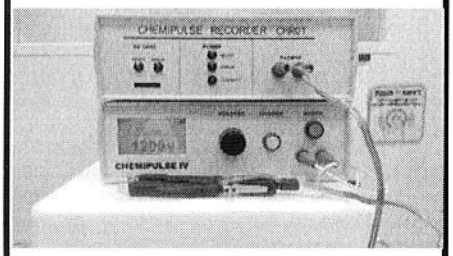


Scheme of polymersomes embedding quantum dots. TEM image shows the size of nanoparticles.

Abstract

The present study was designed to investigate whether electroporation can facilitate the delivery of drugs inside tumors, using quantum dot (QD)-loaded polymersomes as a model. The main goal was to increase the local concentration of anticancer drugs avoiding side-effects. The experiments were performed on colon-cancer grafted mice (*Balb/c*) using *Maestro EX Imaging System*. Electroporation facilitated the delivery of nanoparticles inside the tumor. A significant difference in the fluorescence intensity between electroporated and non-electroporated mice was observed in cancer area even 24 hours after treatment with nanoparticles. The data suggest that electro-assisted delivery of size-controlled long-circulating polymersomes in cancer is a promising therapeutic strategy, especially for treatment of solid tumors.

Methods



Chemipulse IV

- bipolar pulses
- large voltage control in the limits of 100-2200 V
- protection against electrical hazards
- electrotreatment with16 biphasic pulses, 50+50 µs duration, 20 µs pause between both phases
- parallel stainless steel electrodes were used
- electric pulse with intensity of 1200 V/cm was applied.

Water-soluble polymersomes were prepared from chemically modified chitosan and labeled with QD^{705} via carbodiimide chemistry. The nanoparticles were characterized by transmission electron microscopy, dynamic light scattering and fluorescent spectroscopy.
All experiments on animals were conducted in accordance with the guidelines of the Physiological Society of Japan and were approved by the Animal Care and Use Committee of the National Institute of Radiological Sciences, Chiba, Japan.
Balb6 nude mice (21 ± 2 g) were used. *Conol26* cells [$1x10^5$ in 10 mL phosphate-buffered saline (PBS), pH 7.4] were inoculated subdermally in the left or right hindpaw. All measurements were performed ~9-10 days after inoculation, when the tumor size was ~100 mm^3. The mouse was anesthetized with 1.5% isoflurane. The tail veil was catheterized for administration of nanoparticles and the mouse was fixed in the camera of the *Maestro EX Imaging System*. Nanoparticles were injected intravenously (i.v.) via the tail vain (single dose – 80 nmol; 100 mL volume).
The body autofluorescence and QD fluorescence was registered at excitation filter 435-480 nm and emission filter 700 nm longpass.
The data were analyzed by *Living Image In Vivo Imaging* software (*Maestro* version 2.10.0).

Experimental results

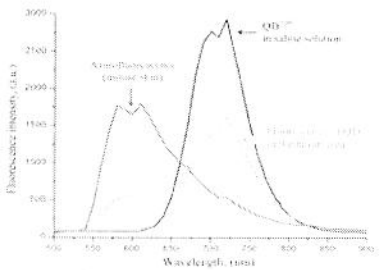


Figure 1. Fluorescence spectra of QD^{705} in saline solution (on phantom), fluorescence spectra (autofluorescence) of mouse body detected before injection of QD^{705}, and fluorescence spectra of mouse body detected after i.v. injection of QD^{705} in mouse.

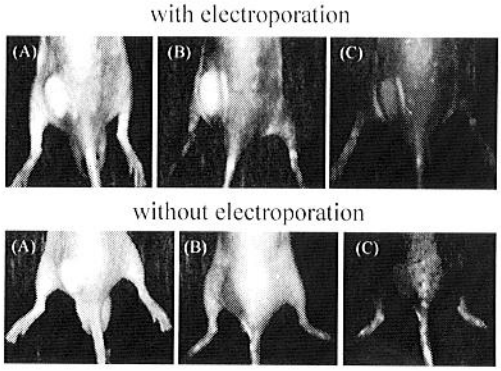


Figure 2. Images of colon cancer-grafted mice obtained: 2 min (A), 3 hours (B) and 24 hours (C) after i.v. injection of QD^{705}-labelled polymersomes with electroporation (upper panel) and without electroporation (lower panel).

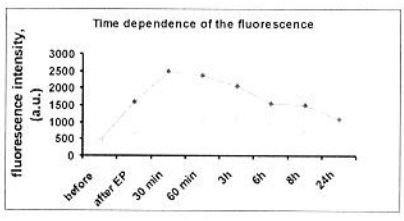


Figure 3. Time dependence of the fluorescence with electroporation (red line) and without electroporation (green line).

Acknowledgements

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