New formulation of EPR oxygen sensors dispersed in thermo-reversible hydrogels

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Introduction:

In vivo EPR oximetry is a powerful method to measure oxygenation of tissues. The use of paramagnetic oxygen-sensing probes such as charcoals and carbon black in aqueous suspension has been described extensively (1,2,3). In order to minimize diffusion and migration of paramagnetic particles *in vivo* (4), we developed a new type of thermo-reversible hydrogel (poloxamer) containing charcoal particles. This system has an interesting property: it is fluid at 5°C but becomes a gel at 37°C (*in vivo*). The purpose of this study was to examine the performance of this system in muscle and tumors.

Materials and Methods:

Charcoal suspension (100mg/ml) was prepared in 25% (w/v) polymer (poloxamer F-127) solution at 5°C. The properties of this oxygen sensor were characterized in vivo after injection in the gastrocnemius muscle of NMRI mice with 50 µl of char suspension at 5°C. Muscle pO2 was measured by EPR oximetry with a 1.2 GHz spectrometer after anesthesia of mice using isoflurane (1.8% v/v). The response of this oxygen sensor to changes in pO2 was evaluated by transiently interrupting the blood flow to the leg (ligation). Histological studies were performed using CD11B labeling to assess the presence of inflammatory reaction. The ability of the oxygen sensor to report on changes in pO2 was also verified in TLT tumors implanted in NMRI mice before and after breathing carbogen (95% O_2 / 5% CO_2).

Results:

The high viscosity of this suspension, especially at 37°C, ensures localization without diffusion or migration of particles. *In vivo* measurements indicate that the pO_2 values recorded in the muscle were stable for at least one month (fig 1). In tumors, the study indicated that the oxygen sensor was able to detect a large (typically 500%) increase in pO_2 after carbogen breathing (baseline of 10.5 mm Hg).
Histological studies indicate that the inflammatory reaction was minimal and strictly localized near the particles.

Discussion

We developed a thermo-reversible hydrogel that decreases drastically the diffusion of particles. Localized injections are easy to perform and the gellification occurs immediately after the injection. Using this system we controlled the injection location very precisely in target tissues. Since this poloxamer is already used in drug delivery systems in humans, the present system could be used in clinical EPR oximetry studies.

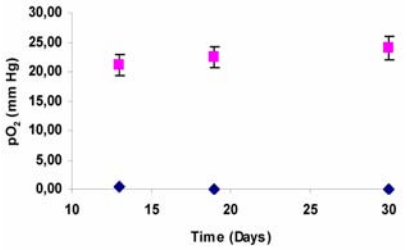

Fig.1: In vivo stability of the response of the oxygen sensor to changes in pO2. Measurements were recorded in muscle before (pink) and after (blue) interruption of the blood flow (ligation of the leg). Note the stability of response for at least 1 month.

Fig.2: CD11B-labeled histological section, one week after injection of the charcoal in F127 poloxamer suspension. Note the very weak inflammatory reaction surrounding the charcoal used as oxygen sensors.

References:

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