

RADIOLABELLING AND BIODISTRIBUTION STUDIES IN AN OPEN WOUND ANIMAL MODEL OF NEGATIVELY CHARGED POLYESTIRENE MICROSPHERES

Álvaro Erhard, María Collantes, Garazi Gainza, Luis Correa, Eloísa Mediavilla, Johana Rodriguez, Jose Javier Aguirre, Margarita Ecay, Iván Peñuelas.

PolyHeal® Micro, composed by Negatively Charged Microspheres (NCMs), is a device that promotes healing. It is indicated for the treatment of chronic and hard-to-heal wounds^{[1][2]}. The aim of this work was to radiolabel NCMs with ^{99m}Tc for biodistribution studies in an open wound animal model to confirm their lack of dermal absorption and demonstrate their safety.

Materials and Methods

NCMs were radiolabeled with SnCl₂-reduced ^{99m}Tc-pertechnetate in an oxygen-free atmosphere. Colloids and radiolabeling yield were measured by radiochromatography (ITLC-SG, mobile phase: acetone) after centrifugation. Stability experiments were carried out in NaCl 0.9% and human plasma solutions for up to 24 h; 0.1 mM DTPA was used for competition studies to define the specificity of the radiolabeling.

An interscapular surgical wound was made in female Wistar rats (n=22) under anesthesia. ^{99m}Tc-NCMs (0.0175 mg/animal; 6.5±0.5 MBq) or free ^{99m}Tc-pertechnetate (control, 6.1±0.75 MBq) were placed in the wound and covered with a dressing. We analyzed a single dose model treated once (G1) to study dermal absorption; and a multiple dose model, treated daily during 11 days (G2) to analyze the activity of phagocytic system activity on NCMs. SPECT/CT imaging was carried out at 1, 2, 12 and 24 hours immediately after administration (G1) or starting on day 10 (G2). Quantification was carried out by drawing VOIs over CT images transferred to SPECT. At the end point animals were euthanized, organs/tissues extracted and radioactivity measured in a gamma counter, calculating the percentage of injected dose (%ID/organ).

Results

NCMs showed a mean size of 5.18 µm and were negatively charged (-93 mV). Radiolabelling yield was >99% after optimization (10' reaction, 0.5 mg/ml SnCl₂), with no colloids (0.6±0.4), and was stable until 24 h in NaCl 0.9% (92.2±3.6) and plasma (97.3±2.5). 88.9±1.6 yield was obtained after DTPA challenge.

SPECT/CT studies showed that more than 99% of ^{99m}Tc-NCMs remained in the wound area for at least 24h (G1 & G2) exerting a local effect. Most of the NCM, were retired with the dressing removal, (82.1±6.2%), and some NCM remained in the skin surrounding the wound (2.9±2.4%) on group 2, No activity in lymph nodes was found by *ex vivo* studies suggesting absence of phagocytosis. ^{99m}Tc-pertechnetate was absorbed as expected and mainly in stomach and intestines.

Conclusions

NCMs radiolabeling was found to be simple and stable. No systemic absorption was found on the wound as seen in SPECT/CT imaging either after single or multiple administrations of NCMs. There was no evidence of MCN accumulation any organ which could be associated with a MCN phagocytosis exerted by macrophages

¹ Polyheal Micro instructions

²Shoham Y. Et al.J WoundCare. 2013;22(3):144-55