Characterising 111In-anti-γH2AX-TAT in Targeting the DNA Damage Signal Associated with Wnt Activated Colorectal Cancer

M. Konstantinou1 , J. Knight2 , T. Hay1 , P. Shaw1 , M. Smalley1 , B. Cornelissen2 , A. R. Clarke1 ;

1 Cardiff University, Cardiff, UNITED KINGDOM, 2 University of Oxford, Oxford, UNITED KINGDOM.

Objectives: Colorectal cancer (CRC) is the second most commonly diagnosed cancer has a poor 60% 5-year survival rate. The Wnt signalling pathway is fundamental for homeostasis of the intestinal epithelium. Its deregulation drives development of CRC and induces DNA damage. Histone-2AX (H2AX) is a component of the nucleosome whose phosphorylated form, γH2AX, is a marker of DNA damage. The VillinCreERApcfl/ fl mouse model is an inducible genetically engineered mouse model of CRC, where tamoxifen-induced deletion of the APC gene induces Wnt signalling and precipitates a crypt-progenitor-like phenotype. Here, we assess whether the DNA damage generated in this CRC mouse model with Wnt signalling deregulation can be targeted using 111In-anti-γH2AX-TAT, a γH2AX-imaging agent we developed previously.

Methods: 111Inanti-γH2AX-TAT, based on anti-γH2AX antibodies conjugated to the cell-penetrating peptide TAT to allow cellular internalisation and nuclear localisation, was produced as previously described. The VillinCreERApcfl/ fl model was induced by tamoxifen three times in a day by either intraperitoneal (IP) injection (80 mg/kg) or gavage (60 mg/kg). γH2AX immunohistochemical analysis was performed three days post induction to assess γH2AX levels. Three days post induction, another group of mice was injected intravenously with 111In-anti-γH2AX-TAT or isotype control RIC (5 μg/mouse; 1 MBq/μg). At 24h post-injection SPECT/CT imaging and biodistribution studies were performed.

Results: Immunohistochemical analysis showed that on day three post-induction using either IP injection or gavage, overall intestinal γH2AX levels were significantly elevated compared to controls. However, the intensity of γH2AX staining was higher by IP injection compared to gavage. In vivo experiments highlighted that 111In-anti-γH2AX-TAT uptake is more prominent in the induced intestine (2.46±0.57% ID/g) compared to controls (1.41±0.29% ID/g, p