Preclinical PET/CT imaging of colon-infiltrating CD4⁺ T cells with ^{89}Zr-df-IAB46 in a dextran sodium sulfate-induced mouse model of acute and chronic ulcerative colitis

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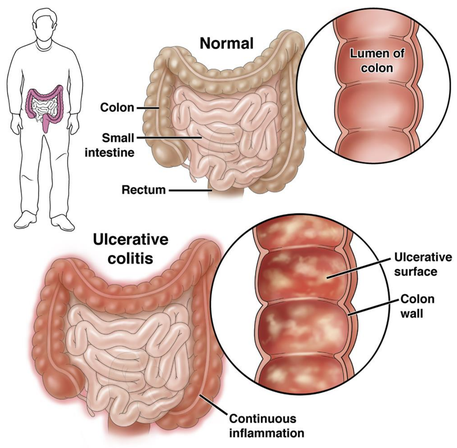
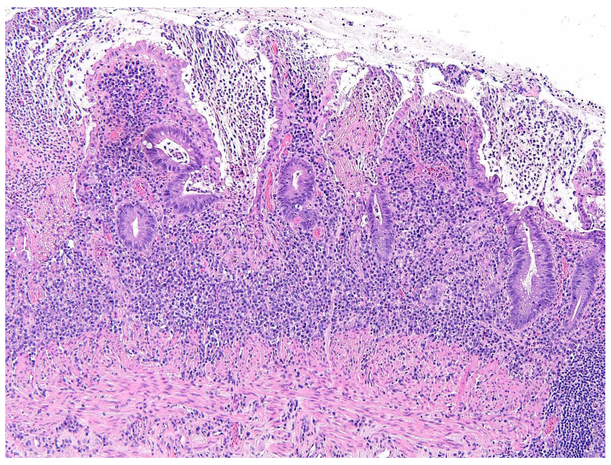
Background

Ulcerative colitis (UC) is a type of inflammatory bowel disease characterized by chronic inflammation of the large intestine, mainly mediated by CD4⁺ T lymphocytes (Geremia A. *et al.*, Autoimmun Rev. 2014;13:3–10). A probe for non-invasive PET imaging of CD4⁺ Th-cell dynamics could be used to identify the cell subsets responsible for colonic inflammation, as well as to assess response to therapy for clinical diagnostic and drug development applications. Here, the utility of anti-CD4 minibody (IAB46) as an immuno-PET probe to measure CD4⁺ T-cell levels *in vivo* was evaluated in a dextran sodium sulfate (DSS)-induced mouse model of UC.



(American Gastroenterological Association, 2022)

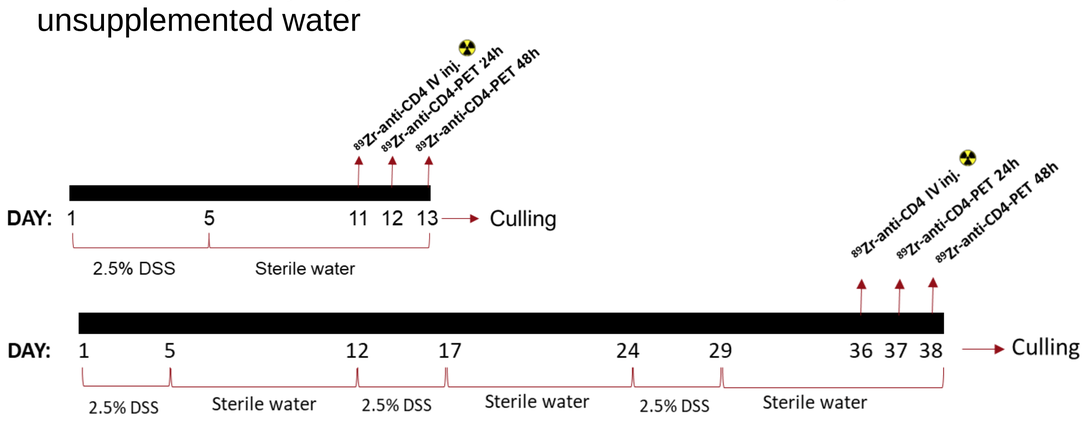


Severe active chronic colitis with multifocal colonic surface ulceration as shown by H&E staining (PathologyOutlines.com, Inc., 2023).

Methods

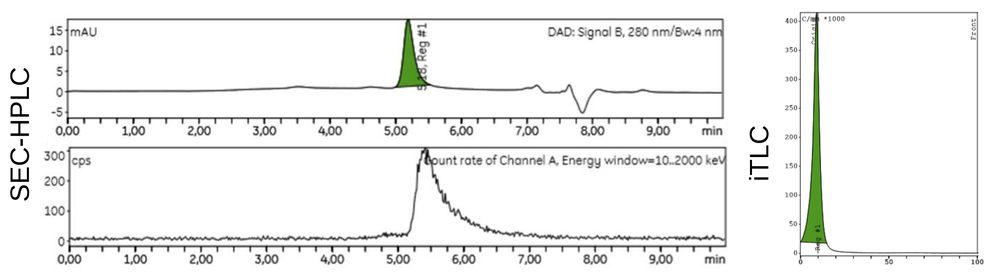
DSS-induced mouse model of UC

- C57BL/6J, F, 8-10 wo
- 1 or 3 x 5-day cycles of 2.5% DSS in drinking water + 7-day unsupplemented water



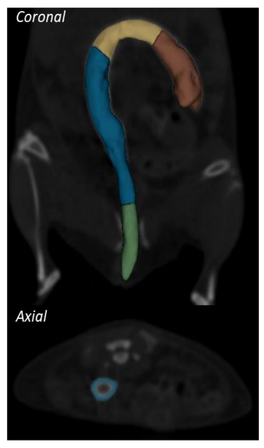
Radiosynthesis of ^{89}Zr-df-IAB46

- Anti-mouse CD4 minibody (ImaginAb, Inc.) conjugated to deferoxamine and radiolabeled with ^{89}Zr.
- RCP > 95%, SA 0.3 MBq/µg



In vivo PET/CT imaging

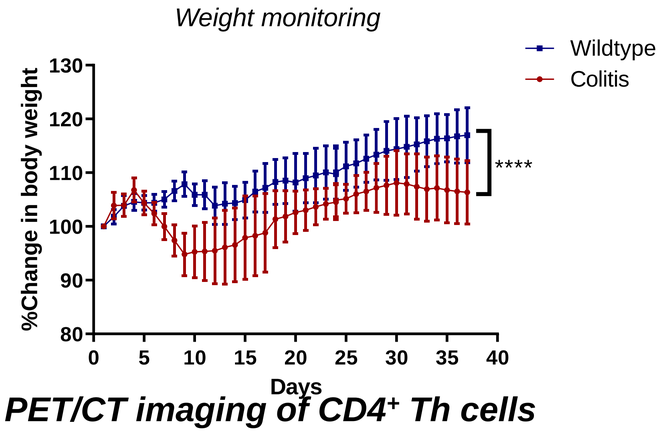
- 5 µg of ^{89}Zr-df-IAB46 (~1.5 MBq) injected intravenously to all mice.
- Negative controls: wildtype mice and colitic mice injected with 15-fold unlabeled df-IAB46 2 hours before tracer injection.
- Iohexol was injected intrarectally for contrast-enhanced CT imaging of the large intestine.
- PET images were quantified via VOI analysis using 3D fixed-volumes and semiautomatic iso-contouring methods.
- Excised colon tissue was examined by H&E and immunofluorescence staining.

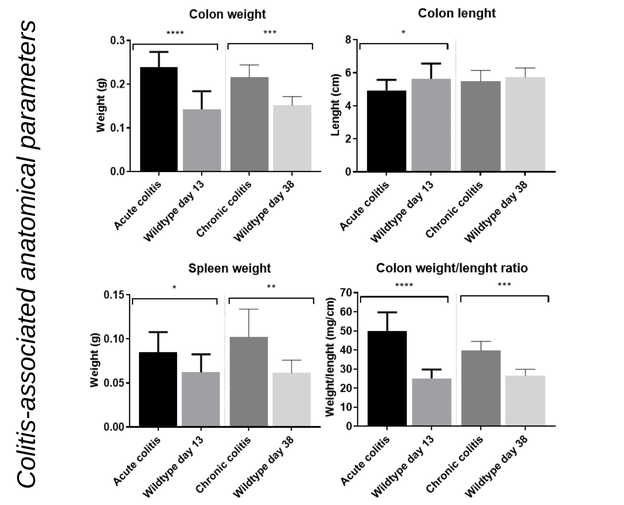


Colon segmentation on contrast-enhanced CT images

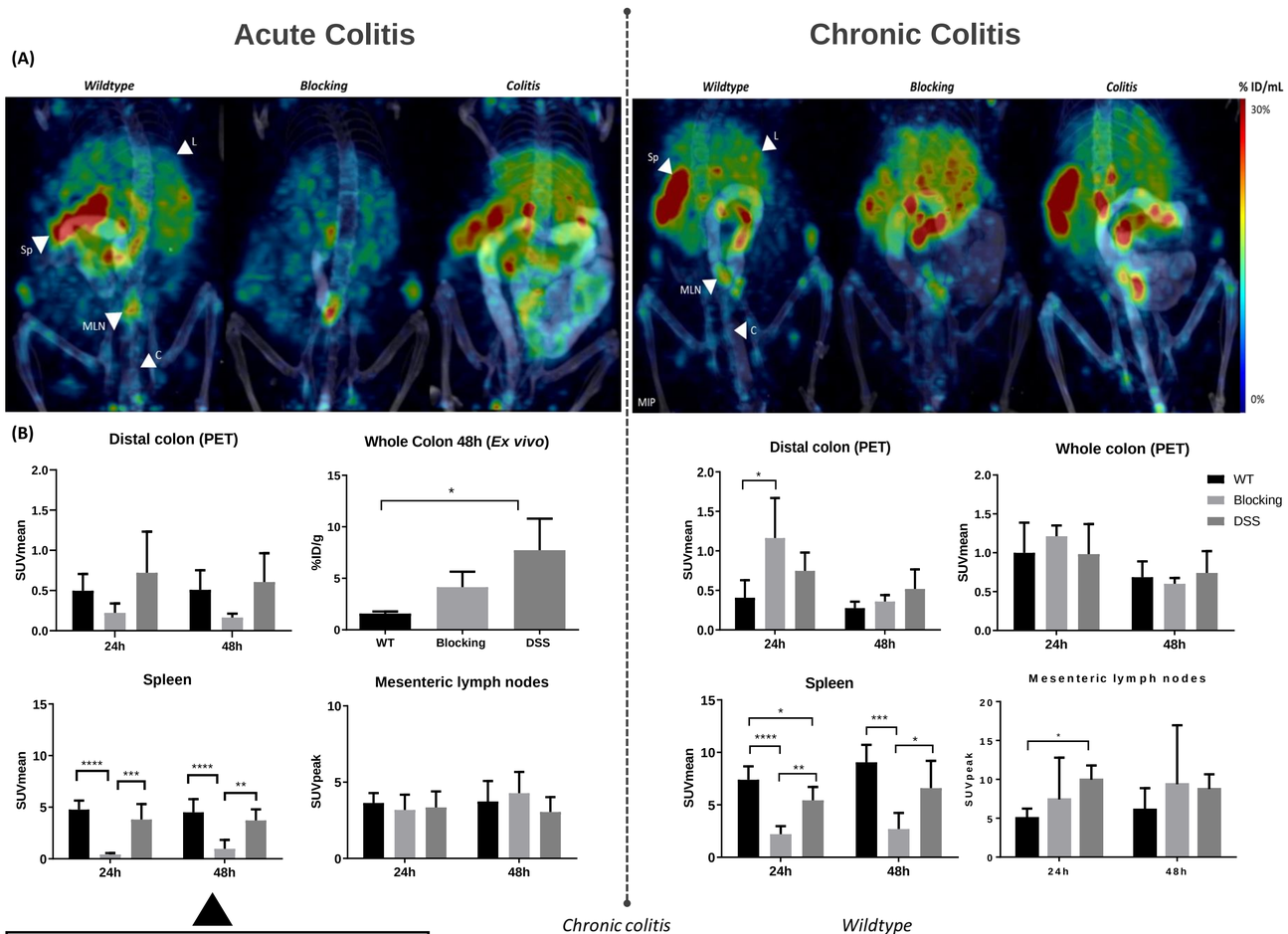
Results

DSS-induced colitis mouse model



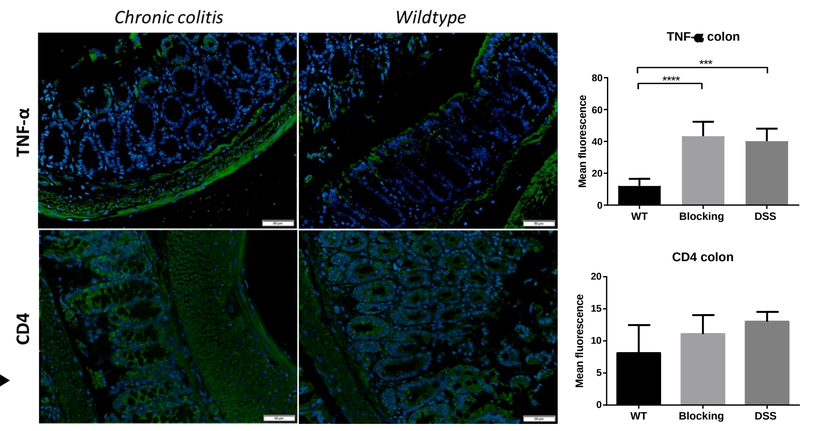


PET/CT imaging of CD4⁺ Th cells



(A) Coronal views of PET/CT MIPs corresponding to wildtype and colitic mice in the acute (*left*) and chronic phase (*right*) of the disease at 24h post-injection of ^{89}Zr-df-IAB46, showing accumulation in the spleen (Sp), mesenteric lymph nodes (MLN), liver (L) and colon (C). **(B)** PET image quantification.

Representative photomicrographs showing immunofluorescence staining of TNF-α (top) and CD4 (bottom) in colonic tissue from mice with chronic colitis and wildtype controls.



Conclusions

- ^{89}Zr-DFO-IAB46 accumulated mainly in secondary lymphoid organs and tissues, such as spleen and lymph nodes, and was primarily excreted into the bile.
- Splenic tracer uptake was found to be significantly higher in wildtype controls at 24h post-injection compared to mice with chronic colitis.
- Lymph node CD4 signal was significantly higher in DSS-treated mice at the chronic phase compared to wildtype controls but was not effectively blocked.
- PET signal was higher in the distal colon in both mice with acute and chronic colitis compared to wildtype littermates.

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