Objective: A reliable imaging modality to probe live bacterial infection is still an unmet clinical need as the current methods of detection rely on indirect measurements of infection like morphologic changes or host immune response [1]. We have demonstrated that radiolabeled D-amino acids (DAA) are specific markers for live bacterial infections *in vivo* [2]. Our continued pursuit of developing D-amino acids as PET imaging agents has led us to ^{11}C-D-alanine, which has demonstrated accumulation into examples of gram-positive and gram-negative bacteria in vivo in a murine myositis model. One of our current focuses is further evaluated ^{11}C-D-alanine in more clinically-relevant models. In this report, we describe the evaluation of ^{11}C-D-alanine in in vivo vertebral discitis osteomyelitis (VDO) model [3].

Methods: ^{11}C-D-alanine was synthesized in two steps using an adapted procedure used for L-alanine [4]. The radionuclide is installed using a catalytic asymmetric alkylation of a glycine Schiff base precursor and ^{11}C-methyl iodide. A VDO rat model was established by injection of a bioluminescent laboratory strain of S. aureus in between two vertebrae on the rat tails. The growth of the bacteria was monitored using optical imaging. On day 3 and day 10, rats were imaged using ^{11}C-D-alanine.

Results: 11C-D-alanine was synthesized in 39.2+/-3.3% radiochemical yield with a radiochemical purity of >95% and 94.6+/-1.2% stereochemical purity (n=20). Optical imaging validated growth of bacteria sequestered to the region of injection. A weak signal was observed *In vivo* PET imaging at day 3. However, the signal strength was significantly higher at day 10 post inoculation. Furthermore, clear erosion to the vertebrae is apparent in the CT at the site of inoculation.

Conclusions: ^{11}C-D-alanine has been successfully evaluated in a VDO rat model. The preliminary results suggest ^{11}C-D-alanine to a promising new tracer for direct detection of live bacterial infection in both myositis and VDO infections.

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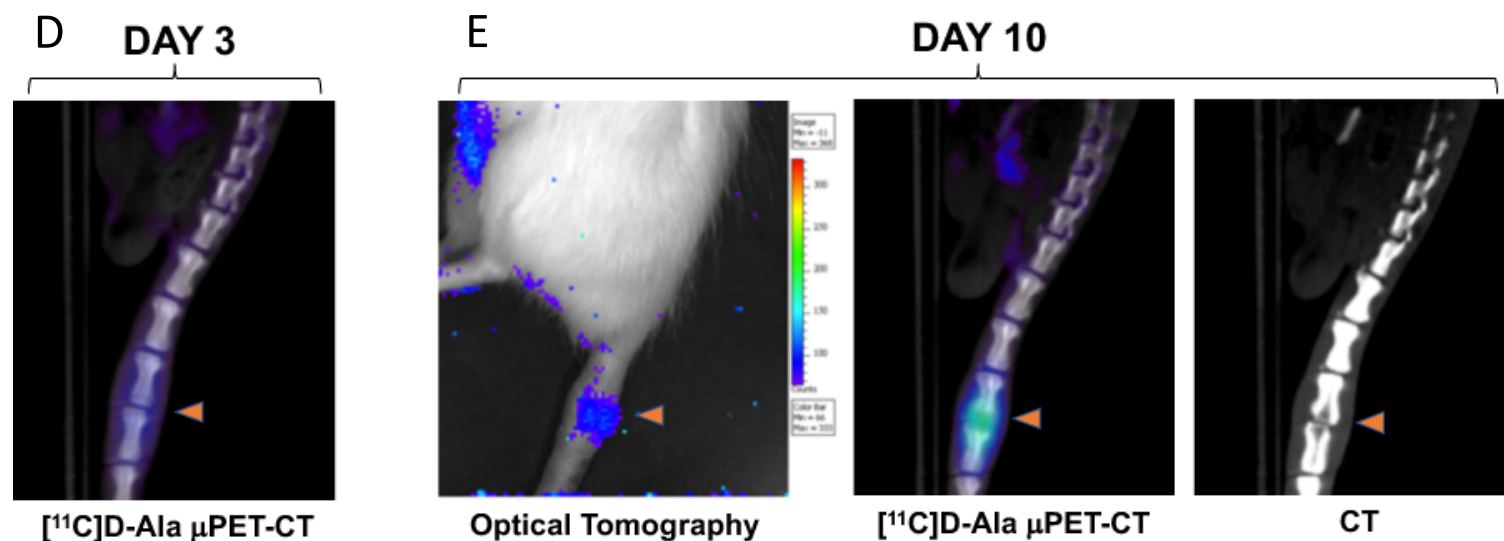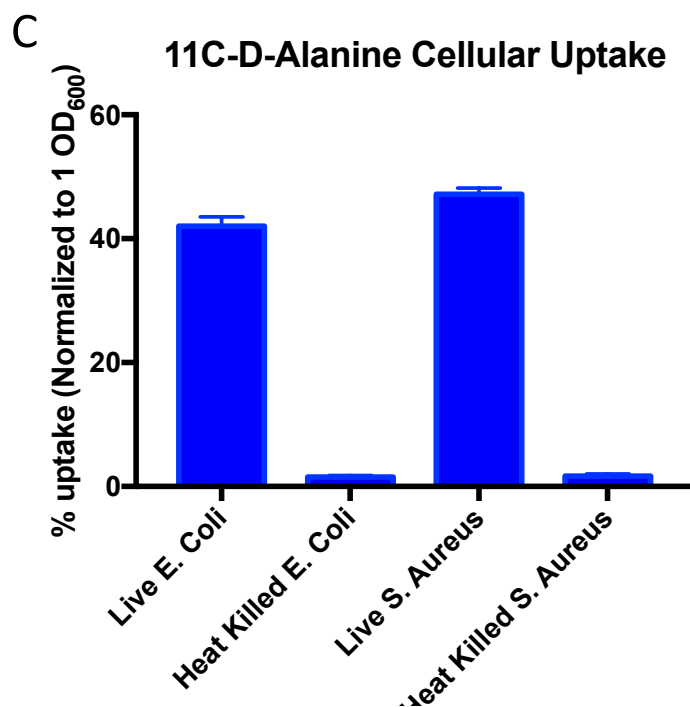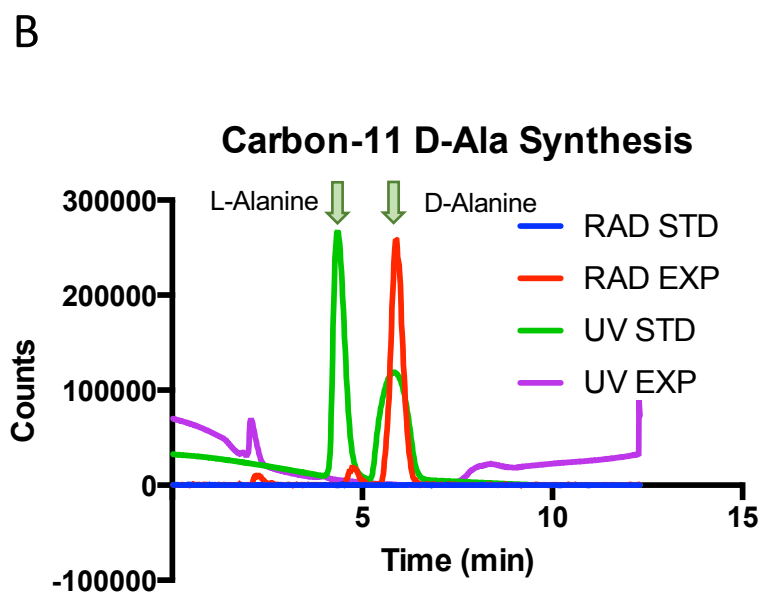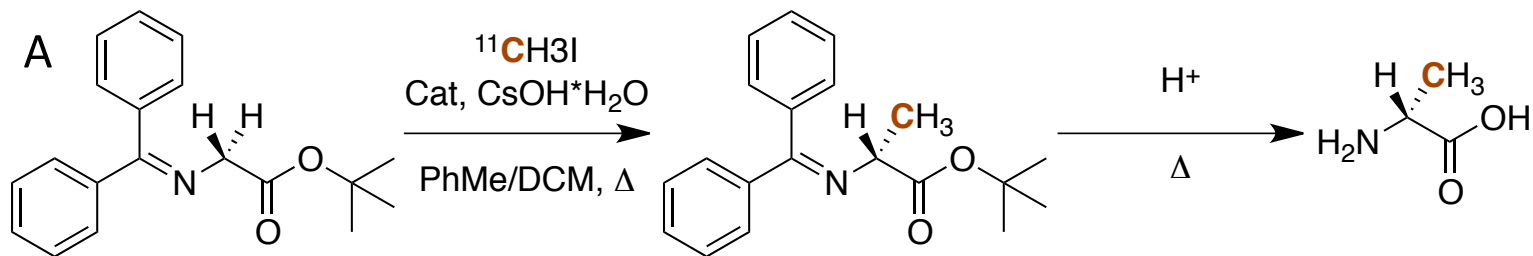
Figure. A) Two-step radio synthesis of 11C-D-alanine in 39.2+/-3.3% radiochemical yield with a radiochemical purity of >95% and 94.6+/-1.2% stereochemical purity (n=20). B) Chiral HPLC analysis of ^{11}C-D-alanine versus racemic standard. Green and blue traces are for the UV aned RAD detection of the racemic standards, and purple and red traces are for the UV and RAD detection of the ^{11}C-D-alanine, respectively. C) In vitro analysis of 11C-D-alanine with live and heat killed laboratory strains of *E. coli* and *S. aureus*. D) MicroPET/CT image of VDO rat model at day 3. Image were taken 90 minutes following a 2Ci tail vein injection of ^{11}C-D-alanine. E) Optical imaging and MicroPET/CT imaging of VDO rat model at day 10. Optical images were taken after PET/CT image. Micro PET/CT Images were taken 90 minutes following a 2Ci tail vein injection of ^{11}C-D-alanine.