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Development of FAPI-CAI heterodimeric theranostic agents for tumor microenvironment targeting: preliminary radiolabeling studies with [^{18}F]AlF

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Aim/Introduction: Theranostics employ structurally identical radiotracers for PET imaging and targeted radiotherapy[1]. Monospecific tracers often underperform due to tumor heterogeneity and microenvironment complexity, whereas heterodimeric agents targeting multiple biomarkers improve specificity, uptake, and retention[2,3]. This study aims to develop FAPI-CAI heterodimer targeting fibroblast activation protein and carbonic anhydrases IX/XII and to optimize their radioactive labelling with [^{18}F]AlF together with the corresponding FAPI and CAI monomers.

Materials and Methods: The FAPI-CAI dimer and its monomeric counterparts (FAPI-04 and CAI) were radiolabeled with ^{18}F using the aluminum fluoride ([^{18}F]AlF) approach. Radiolabeling conditions were systematically optimized by varying key parameters, including precursor amount and cosolvent composition, to maximize radiochemical conversion, followed by improvement of the purification protocol for the final radiotracers. Quality control was conducted using radio-TLC and radio-HPLC. Radiochemical conversion, yield, and purity, along with key physicochemical and biological properties, were assessed.

Results: Optimization of the [^{18}F]AlF labeling protocols yielded radiochemical conversion (RCC) of 40.86 ± 2.09 for [^{18}F]AlF-FAPI-04, with an RCY of 28.64 ± 2.22 and radiochemical purity ≥97%. Radiolabeling conditions for [^{18}F]AlF-CAI were also optimized, and it was observed that following preparative HPLC purification of the NOTA-CAI precursor, in addition to the removal of radiochemical impurities, an increase in RCC occurred, from 35% using 160 nmol precursor to 55.74% using 40 nmol precursor after the additional purification step. [^{18}F]AlF-FAPI-CAI showed greater challenges, with an RCC of 18% and radiolabeled impurities. In parallel, an optimized solid phase extraction protocol was developed, enabling recovery of 75 ± 5% of the tracer in only 600 µL.

Conclusions: This study establishes an optimized [^{18}F]AlF radiotracer framework, highlighting precursor purity as key for labeling efficiency and radiotracer quality, while structural constraints in the FAPI-CAI dimer may limit both radiochemistry and target binding, supporting future linker optimization.

[1] H. H. Tran, A. Yamaguchi, and H. C. Manning, Eur. J. Nucl. Med. Mol. Imaging 2025, 52, 2685–2709.
[2] B. Judmann, D. Braun, B. Wängler, R. Schirrmacher, G. Fricker, and C. Wängler, Pharmaceuticals 2020, 13, 173.
[3] C. Chambers, B. Chitwood, C. J. Smith, and Y. Miao, iRADIOLOGY 2024, 2, 128–155.