Synthesis, Radiolabelling and Biological evaluation of New cyclo-peptide as Imaging Agent of $CXCR_4$ Receptor

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Introduction

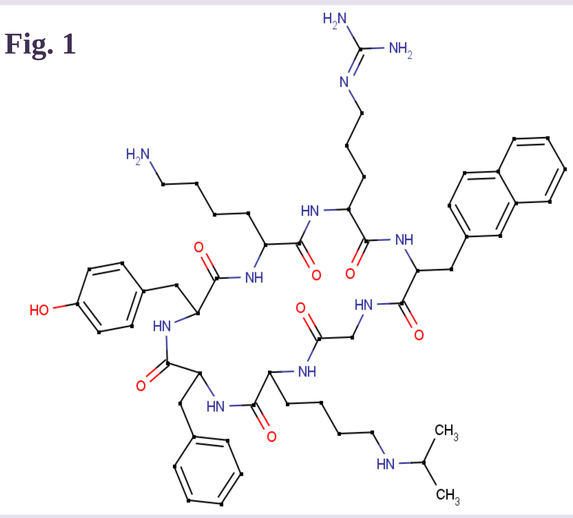
$CXCR_4$ (chemokine receptor 4) is overexpressed in many types of tumor and promotes cancer metastasis[1,2]. Expression level of $CXCR_4$ is predictive for the metastatic potential of a given tumor type and arbitrate organ-specific metastasis[3]. Numerous $CXCR_4$-targeted radiolabelled antagonists have been in vivo evaluated as $CXCR_4$ imaging agents by PET and SPECT imaging techniques [4-6]. In this study, a new analogue of LY251029 containing a HYNIC-coupled hexapeptide has been developed and radiolabeled by $[^{99m}Tc]TcO_4^-$.

Result and Discussion

Solid Phase Peptide Synthesis:

The peptide (Fig. 1) successfully was Synthesized, purified and Characterized using RP-HPLC and LC-MS spectrums.
The m/z peaks were appeared at (m/z: 1154.60, 578.10, 385.80).

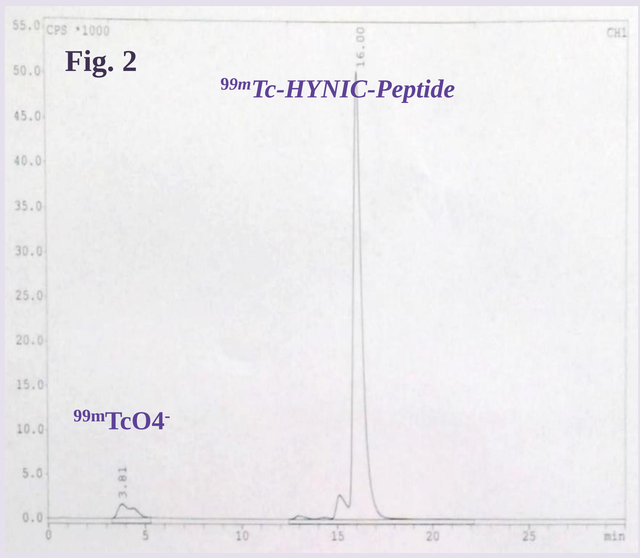
Fig. 1



Radio-labelling & Partition Coefficient:

The analytical HPLC with gamma detector and output chromatogram showed main radiolabeled peak at 16:00 min along with a small peak at 3.81 min (Fig 2).
log P = -2.04 ± 0.09
Specific activity
= 0.256 ± 0.05 MBq/μg

Fig. 2

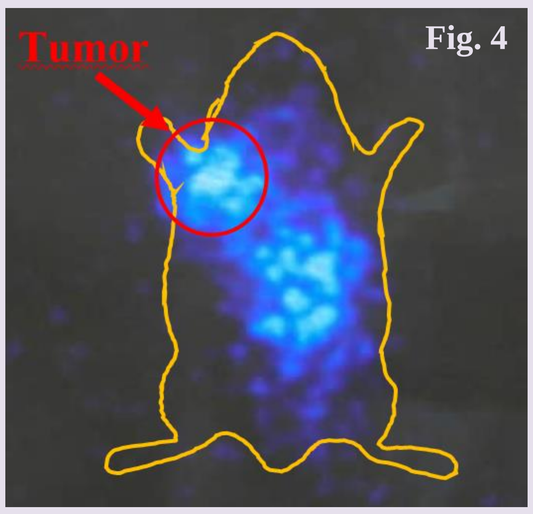


Result and Discussion

Imaging

Planar whole-body image of tumoral mouse at 30 min post injection of $[^{99m}Tc]$HYNIC-Peptide is displayed in ***Fig.4.***
Scintigraphy studies demonstrated accumulation of radioligand mainly in tumor tissue.
Whole body posterior planar imaging of B16-F10 tumoral mouse at 30 min post injection of $[^{99m}Tc]$HYNIC-peptide using gamma camera.

Fig. 4



Result and Discussion

In vitro and in vivo stability studies:

The $[^{99m}Tc]$-HYNIC-peptide showed appropriate *in-vitro* and *in-vivo* serum stability and low protein binding which is required to increase the ligand availability to the target tissue. It indicated more than 95% stability for 24 hr and 4 hr incubation at room temperature and 37 °C, respectively.

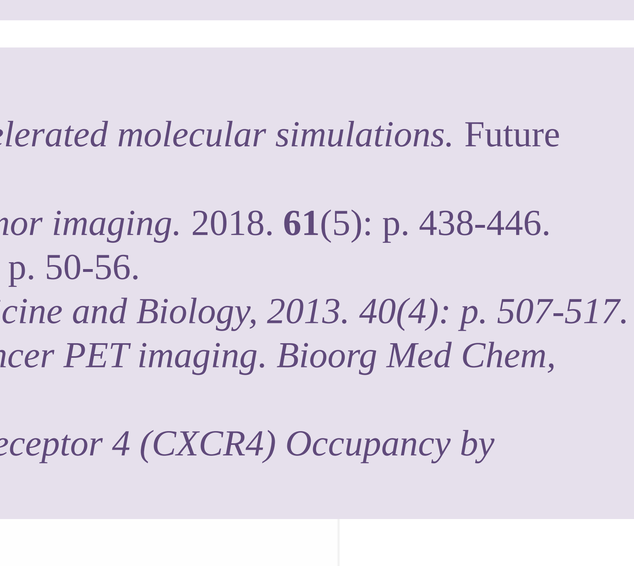
Biodistribution Studies:

The Biodistribution evaluations pointed to rapid clearance in blood from 4.87 ± 0.91 %ID/g at 15 min to 0.82 ± 0.08 at 60 min post injection that is in accordance with low protein binding of designed peptide. The whole-body clearance proceeded via urinary system which was predictable because of higher hydrophilicity of radioligand (Log p = -2.04 ± 0.09).

The tumors tissue uptake in presence of Plerixafor as $CXCR_4$ receptors antagonist, was diminished from 3.49 ± 0.27 to 1.84 ± 0.13 %ID/g which confirmed the specific binding of radioligand to the receptor.

Figure 3. Shows the best time to image is the highest uptake of radioligand in target tissue which is 30 min post injection.

Fig. 3



Conclusion:

In this study the synthesized and radio-labelled cyclo-peptide pointed to great radiochemical purity and even up to 24 hrs stability.
High accumulation of radioligand in tumor tissue and excretion via kidney were observed. Moderate affinity to tumor cells and higher uptake at 4 hours post injection should be optimized by next studies and potential candidate.

References:

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Acknowledgment: