Overcoming the blood-brain-barrier by a linear 7-mer peptide, IF7, with binding specificity to Annexin A1 in brain tumors

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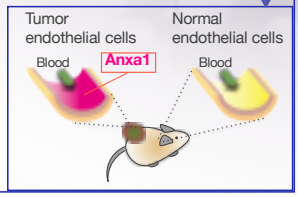
Summary

Brain malignancies are difficult to eradicate, as chemotherapeutics injected intravenously cannot reach cancer cells in stroma due to the blood-brain barrier (BBB). Previously we identified a linear 7-mer peptide that we designate IF7 binds to the N-terminal domain of annexin A1 (Anxa1) (1). Although Anxa1 is normally expressed intracellularly in numerous cell types, Anxa1 is found on the endothelial cell surface in malignant tumors (2). When fluorescently labeled IF7 was injected intravenously into brain tumor model mice, IF7 reached tumor vasculature and targeted tumor cells in stroma, overcoming the BBB (3). In a dual tumor mouse model harboring subcutaneous and brain tumors, IF7-conjugated to the anti-cancer drug SN-38 suppressed growth of both tumors. In a brain metastatic model of syngeneic melanoma, tumors continued shrinking after IF7-SN38 administration. When melanoma cells were injected subcutaneously into recovered mice, CD8+ cytotoxic T cells infiltrated the injection site, suggesting a heightened immune response against tumor cells (3). These results suggest that IF7-SN38 can overcome BBB and efficiently suppress growth of malignant brain tumors and that high efficacy of IF7-SN38 therapy may lead an immunotherapeutic response by the host. IF7Cure Inc. is preparing for the first-in-human clinical trial of IF7-SN38 on glioblastoma patients.

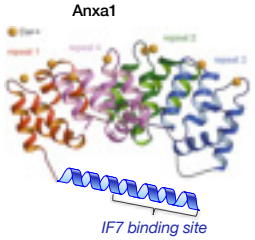
Introduction

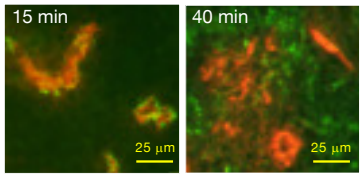
- Annexin A1 (Anxa1) is the most specific tumor vascular surface marker (2)
- Previously we reported that carbohydrate-mimetic IF7 peptide targets to tumor vasculature through Anxa1 and delivers anti-cancer drug to tumor stroma (1).
- Anxa1 transports its ligand, IF7, by transcytosis through the endothelial cell (1).

Hypothesis:
Intravenouly injected IF7-conjugated anti-cancer targets the brain tumor vasculature and crosses the blood-brain-barrier. IF7-conjugated anti-cancer drug eradicates the brain tumors in the mouse model (3).

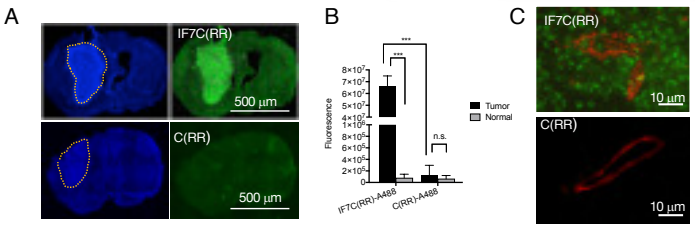


IF7 crosses tumor endothelial cells via transcytosis



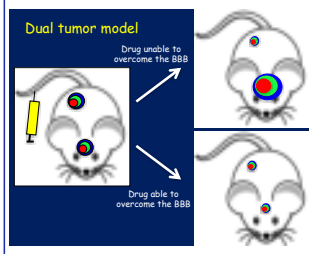


Alexa488 (green fluorescence)-labeled IF7 injected intravenously to colon tumor model mouse. Tumor section of 15 min after injection shows IF7 at the abdominal side of vasculature and that of 40 min shows IF7 in the stroma. Vasculatures were immunostained for endothelial cell marker CD31 (red). (1).



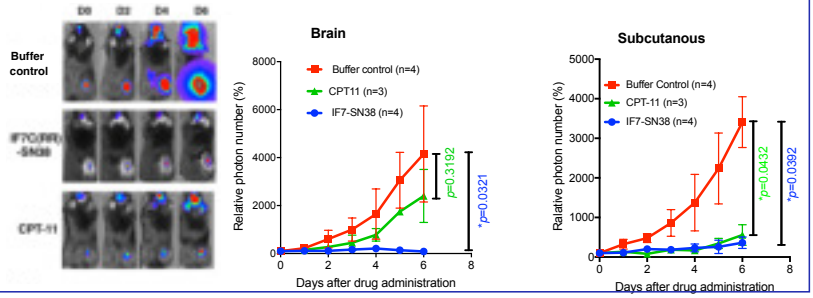
A and **B**: Brain tumor targeting of IF7 peptide (green) injected intravenouly to brain tumor model mice. **C**: Brain tumor tissue sections indicating IF7 penetration to tumor stroma and cancer cells. (3)

Comparable therapeutic efficacy of IF7-SN38 on brain and subcutaneous tumors

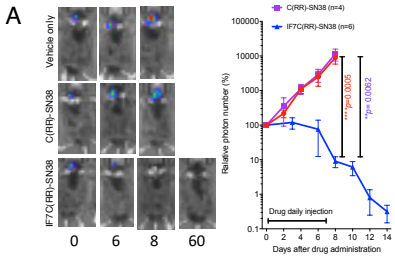


Left: Dual tumor mouse model to test location-dependent therapeutic efficacy of intravenously-injected IF7-SN38.

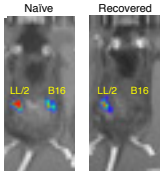
Right: B16-Luc melanoma cells were injected into the brain and under the skin of a single syngeneic C57BL/6 mouse. Once tumors had grown to photon number 1e+5, either IF7-SN38 (7.0 μmoles/kg, blue lines in graphs), irinotecan (CPT-11) (50 μmoles/kg, green lines in graphs) or PBS (Buffer control, red lines in graphs) was injected intravenously daily for 6 days. (3)

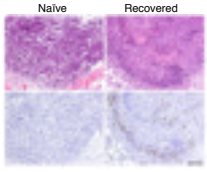


Immune response against tumor cells after eradication of brain tumor by IF7-SN38



B16-Luc tumors in C57BL/6 mice





Subcutanous B16-Luc cell injection site immunostained for CD8 (cytotoxic T cell marker).

A: IF7-SN38 2.5 μmoles/kg formulated in 10% Solutol HS15 in water was injected intravenously daily for 7 days. Note that brain tumor keep shrinking after ceasing the drug injection

B: Growth of syngeneic LL/2-Luc and B16-Luc cells in naïve mice and in mice whose tumors had been eradicated by IF7C(RR)-SN38 treatment, 4 days after subcutaneous injection.

D: Immunohistochemistry with anti-CD8 antibody of B16-Luc cells at subcutaneous injection sites, 20 hours after B16-Luc cell injection. Scale bars: 100 μm. (3)

Conflict of interest

The author Michiko N. Fukuda is the founder of IF7Cure Inc., of which the mission is to initiate clinical trial of IF7-SN38 for commercialization of IF7-SN38. She owns stock of the company.

References

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