

# CRT Licensing Opportunity



## $\alpha v\beta 6$ -binding Peptides for Tumour Targeting

- Novel and proprietary peptides with high affinity and selectivity for integrin  $\alpha v\beta 6$
- Lead peptide selectively targets  $\alpha v\beta 6$  tumours *in vivo* for imaging and therapy
- $\alpha v\beta 6$  is highly expressed in many tumours and clinically correlates with poor prognosis
- $\alpha v\beta 6$  identified as a key target in RAS-dependent tumours of high unmet medical need

BIOLOGICAL THERAPEUTICS/ DIAGNOSTICS | *In Vivo* Proof-of-Principle

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### Commercial Opportunity

The integrin  $\alpha v\beta 6$  is an exciting emerging target for both imaging and therapy across many common tumour types including pancreatic, breast, oesophagus, head and neck, skin, lung and ovarian. Each year an estimated 279,000  $\alpha v\beta 6$ -positive tumours are diagnosed in the US & UK alone. In addition,  $\alpha v\beta 6$  is recognised as an exciting target in fibrotic diseases.

Peptides with remarkable affinity and selectivity for  $\alpha v\beta 6$  have been identified and characterised (1). *In vitro* and *in vivo* data demonstrates the potential of these peptides as the basis for novel PET and SPECT imaging probes, tumour targeting agents and functional inhibitors of  $\alpha v\beta 6$ . The peptide inhibits  $\alpha v\beta 6$ -ligand binding with an IC<sub>50</sub> of 3 nM, an activity 1000-fold more selective for  $\alpha v\beta 6$  than for other RGD-directed integrins ( $\alpha v\beta 3$ ,  $\alpha v\beta 5$  and  $\alpha 5\beta 1$ ; Table 1).

Peptide	$\alpha v\beta 6$	$\alpha v\beta 3$	$\alpha v\beta 5$	$\alpha 5\beta 1$
A20FMDV2	3 ± 1 nM	>10 µM	>100 µM	>10 µM
[ <sup>19</sup> F]FBA-A20FMDV2	3 ± 1 nM	>10 µM	>10 µM	>10 µM

Table 1: IC<sub>50</sub> values comparing the ability of A20FMDV2 and [<sup>19</sup>F]FBA-A20FMDV2 to inhibit binding of biotinylated ligands to immobilized  $\alpha v\beta 6$ ,  $\alpha v\beta 3$ ,  $\alpha v\beta 5$ , and  $\alpha 5\beta 1$  integrins.

Patent family WO2007/039728 protecting the peptide consensus sequence is available for exclusive licensing or collaborative development.

### *In vivo* Imaging

The lead peptide ('A20FMDV2') has been validated *in vivo* for use in both PET and SPECT imaging with a variety of radiolabelling approaches. The labelled peptide shows rapid uptake and selective retention in xenograft tumours engineered to express  $\alpha v\beta 6$ , but not in their non- $\alpha v\beta 6$  expressing counterparts (2). Furthermore, A20FMDV2 has been used to successfully image both breast and pancreatic xenograft tumours endogenously expressing  $\alpha v\beta 6$  (3) and Figure 1). Based on these data, the peptide has potential for use as a novel imaging probe in these tumours and others where  $\alpha v\beta 6$  over-expression is observed. Cancer Research UK is currently funding a trial that will progress the peptides into Phase I for imaging solid tumours.

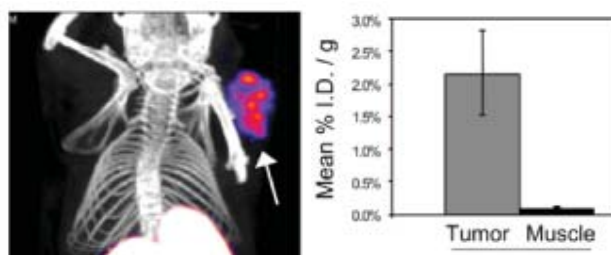


Figure 1. NanoSPECT/CT image of a breast cancer xenograft using <sup>111</sup>Indium-A20FMDV2 (left panel) and calculated % injected dose per gram of tissue for tumour versus muscle (right panel)

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## Therapeutic Tumour Targeting

Function-blocking abilities of the peptide have been demonstrated using  $\alpha v\beta 6$ -dependent tumour cell migration and invasion assays. In addition, A20FMDV2 mediates rapid tumour cell uptake, a key prerequisite for efficacious delivery of toxic payloads (Figure 2).

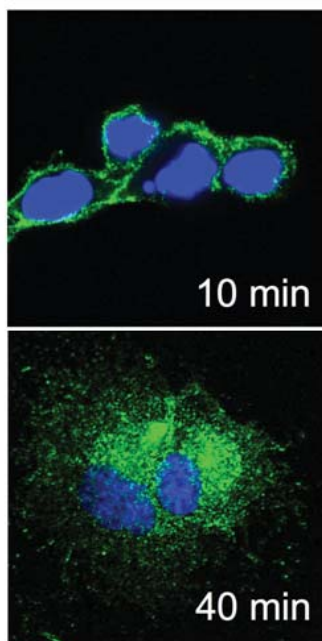


Figure 2. A20FMDV2 is rapidly internalised by  $\alpha v\beta 6$ -positive tumour cells

Recent studies have demonstrated that drug-conjugate versions of A20FMDV2 can selectively kill  $\alpha v\beta 6$  positive tumour cells *in vitro* (Table 2).

	$\beta 6$ +ve cell line	Matched $\beta 6$ -ve cell line	Ratio EC50 $\beta 6$ -ve/ +ve
EC50 (nM)	0.1132	24.73	218.5

Table 2. Toxin linked A20TJ peptide shows selective killing of cells expressing  $\alpha v\beta 6$ . Cell viability was measured by MTT assay after 72h of peptide-drug treatment

To demonstrate the potential of the peptides for therapeutic targeting of tumours *in vivo*, the A20FMDV2 sequence was incorporated into the HI loop of the adenovirus type 5 (Ad5) fiber knob. The resulting  $\alpha v\beta 6$ -retargeted virus exhibited up to 50-fold increases in CAR-independent transduction and up to 480-fold increased cytotoxicity *in vitro* (4). *In vivo*, the virus demonstrated enhanced tumour uptake and reduced liver sequestration. The peptides have also been out-licensed for the purpose of targeting drug-containing nanoparticles to tumours.

## Supporting Rationale

The expression of  $\alpha v\beta 6$  is restricted primarily to epithelial cells where it is expressed at low levels in healthy tissue and significantly up-regulated during wound healing, fibrosis and in tumorigenesis.  $\alpha v\beta 6$  has multiple regulatory functions in tumours including TGF- $\beta$  activation, cell proliferation, MMP production, cell invasion and survival. Antibody-mediated blockade of  $\alpha v\beta 6$  has been demonstrated to inhibit tumour growth *in vivo* (5), supporting the use of  $\alpha v\beta 6$ -targeted agents in cancer therapy. In cancer patients, elevated  $\alpha v\beta 6$  expression has been correlated with poor prognosis in tumours including colorectal, ovarian and lung. Numerous publications have identified  $\alpha v\beta 6$  as a key regulator of the epithelial to mesenchymal transition, and more recently  $\alpha v\beta 6$  has been linked with maintenance of a pluripotent cancer stem cell phenotype in oral cancer, and as a key member of a "K-Ras dependency signature" in lung and pancreatic tumours (6).

## References

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