A phase 1 mechanism of action study of intra-tumoural (IT) or intravenous (IV) administration of enadenotucirev, an oncolytic Ad11/Ad3 chimeric virus targeting colorectal cancer patients undergoing resection of primary tumour

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1. BACKGROUND
• To date, clinical success with oncolytic viruses tends to be associated with intra-tumoural (IT) administration and evidence for successful systemic delivery of viruses to tumour cells by intravenous (IV) infusion remains sparse. Pre-existing anti-viral antibodies have been implicated, but other forms of cellular and non-cellular mechanisms for viral inactivation have been demonstrated.

Enadenotucirev (EnAd or ColoAd1) is a tumour selective chimeric Ad11/Ad3 group B adenovirus that has demonstrated preclinical activity in a metastatic model of colorectal cancer (CRC) and in human tumour biopsies ex vivo. 2,3 Preclinical studies suggest that the prevalence of neutralising antibodies against group B adenoviruses is low, which may permit systemic delivery of viruses to tumour cells by intravenous (IV) infusion remains sparse. Pre-existing anti-viral antibodies have been implicated, but other forms of cellular and non-cellular mechanisms for viral inactivation have been demonstrated.

2. KEY OBJECTIVES
• Primary objective
  - Serological studies suggest that the prevalence of neutralising antibodies against group B adenoviruses is low, which may permit systemic delivery of viruses to tumour cells by intravenous (IV) infusion remains sparse. Pre-existing anti-viral antibodies have been implicated, but other forms of cellular and non-cellular mechanisms for viral inactivation have been demonstrated.

3. STUDY DESIGN
• Colon cancer patients scheduled for resection of primary tumour were randomized to either IT or IV administration of EnAd following intra-tumoural (IT) or intravenous (IV) delivery of therapeutic agents to tumour, lymph nodes and normal margins in resected tissues. The EnAd virus is transported back to the nucleus for final virus assembly where they tend to form crystal-like intra-nuclear inclusion bodies. Detection of virus protein inclusion bodies in the nucleus indicates the ability of virus to reach the tumour and to actively replicate within tumour cells – it is a marker of virus delivery and activity.

4. PRIMARY ENDPOINT - RATIONALE AND METHODS
Rationale for EnAd Hexon staining
- adenovirus structural proteins are expressed late during expression, only after replication has occurred.

IV cohort patients received 2.3 x 1012 vp x 3 doses
- Median age: 70.5 years (range 60 – 82)

All IV cohort patients received 1 x 1012vp x 3 doses
- Median time between 1st dose of EnAd and surgery: IV cohort: 16 days (range 8 – 51)

6. PATIENTS CHARACTERISTICS
• 10 patients treated: 5 IV and 5 IT
• 6 males / 4 females.
• Median age: 70.5 years (range 60 – 82)
• Median time between 1st dose of EnAd and surgery: IV cohort: 16 days (range 8 – 51)

5. KEY SELECTION CRITERIA
• Informed consent
• Age ≥ 18 years
• Histologically confirmed colorectal cancer
• Scheduled for resection of primary tumour and no previous history of metastatic disease
• Tumour size ≤ 3 cm in diameter
• Histologically confirmed colorectal cancer
• Surgery planned and feasible within 8 - 15 days of first EnAd administration (IT or IV)

7. RESULTS
Adenovirus hexon staining
- Clear evidence of viral activity (punctate brown staining of nuclear hexon protein) was found in tumour samples from 8 (80%) subjects following both IT and IV administration (Figure 4).

8. DISCUSSION
The aim of this phase 1 study was to examine the safety and preliminary efficacy of EnAd, an oncolytic virus, in patients scheduled for resection of primary colorectal cancer.

References

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Figure 7. Serum antibody titres over time in IT and IV cohorts

Figure 6. Post surgical tumour samples from two representative IV cohort patients.

Figure 5. Nuclear scoring (% hexon positive tumour cell nuclei) for multiple slices of post surgical tumour samples from two representative IT cohort patients.

Figure 4. Post surgical tumour samples IV cohort patient stained for hexon showing positive nuclear staining in tumour cells but negative staining in stromal and normal colon cells.

Figure 3. Electron micrograph of an A549 human lung cancer cell taken 72 hrs after infection with EnAd.