Multiple KRAS and changing mutations detected by cancer related KRAS DNA in patients with resected pancreas ductal adenocarcinoma during treatment with TG01/GM-CSF and gemcitabine (CT TG01-01)

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INTRODUCTION

TG01-01 was a Phase I/II open-label study to assess the safety, immune activation and clinical efficacy of TG01/GM-CSF vaccination in combination with adjuvant chemotherapy in patients (pts) (N=32) with resected pancreatic ductal adenocarcinoma (PDAC).

Pancreatic cancer is a major cause of cancer mortality globally, with 5-year survival rate less than 5%. Oncogenic mutations in KRAS, which drive cell growth and malignant transformation, occurs in more than 90% of pts with PDAC. The fact that KRAS mutations are expressed in high frequency in PDAC may be one of the reasons why chemotherapy and "targeted" drugs tested have failed to significantly increase survival.

TG01/GM-CSF is an injectable antigen-specific cancer immunotherapy targeted to treat pts with KRAS mutations. TG01 consists of a mixture of 7 synthetic peptides representing 7 of the most common codon 12 and 13 oncogenic mutations in KRAS associated with PDAC.

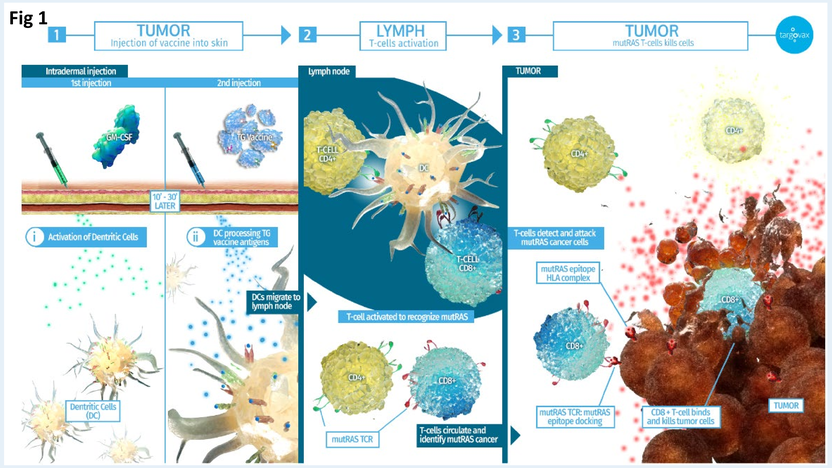
AIM

Pancreatic cancer is a heterogeneous and genetically unstable disease, meaning that more than one KRAS mutation may be present in pts[1-2]. Therefore, we have investigated if cancer related KRAS DNA from pts with resected PDAC had more than one KRAS mutation and if the mutation status changed during treatment with TG01/GM-CSF.

MECHANISM OF ACTION

TG01 induces KRAS mutant-specific T-cell responses, which are enhanced by co-administration of GM-CSF (Fig 1). The TG01 peptides activate both MHC class II restricted CD4+ helper T-cells and as MHC class I restricted CD8+ cytotoxic T-cells, which is necessary to sustain the CD8+ cytotoxic T-cell effect[3].

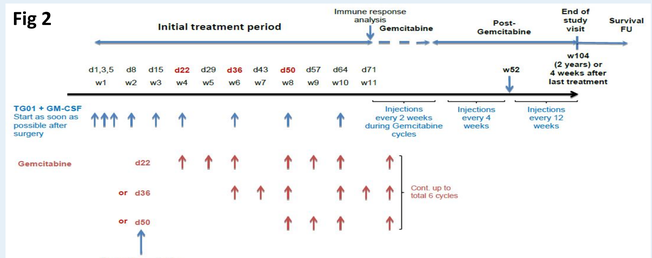
Fig 1



METHOD

Pts were eligible after a R0 or R1 PDAC resection. TG01 (0.7 mg intradermal injection) together with GM-CSF (0.03 mg) was initially given on day 1, 3, 5, 8, 15, 22 and 2-weekly until end of chemotherapy, 4-weekly up to 1 year and 12-weekly up to 2 yrs (n=19) (Fig 2). A modified treatment schedule (n=13) was introduced where no TG01/GM-CSF vaccinations were given during chemotherapy.

Fig 2



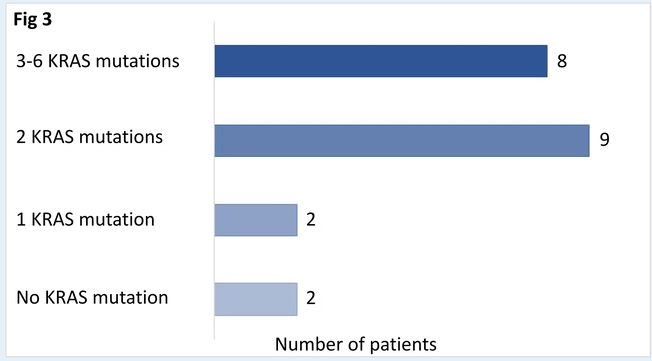
Plasma samples (up to 9) from 21 pts were collected. Cancer related KRAS DNA for 6 of the 7 mutations in the TG01 mixture were analyzed using ARMS: 12D, 12V, 12A, 12R, 12S and 12C.

The Amplification Refractory Mutation System (ARMS) is an allele-specific real time PCR method detecting any mutation involving single base changes or small deletions by comparing the apparent quantity of mutant sequence against the apparent quantity of all sequences[4]. ARMS is based on the use of sequence-specific PCR primers.

RESULTS

Initial results from tumor specimen showed that 16 out of 21 pts had a single KRAS mutation. When analyzing plasma using ARMS, 19 out of 21 pts had one or more KRAS mutations. 17 out of 21 pts (81%) had multiple mutations (up to 6 mutations) throughout the study (Fig 3). 12D and 12V mutations co-occurred in 17/21 (81%) of the pts. In one pt all 6 assessed KRAS mutations were detected during the course of the study.

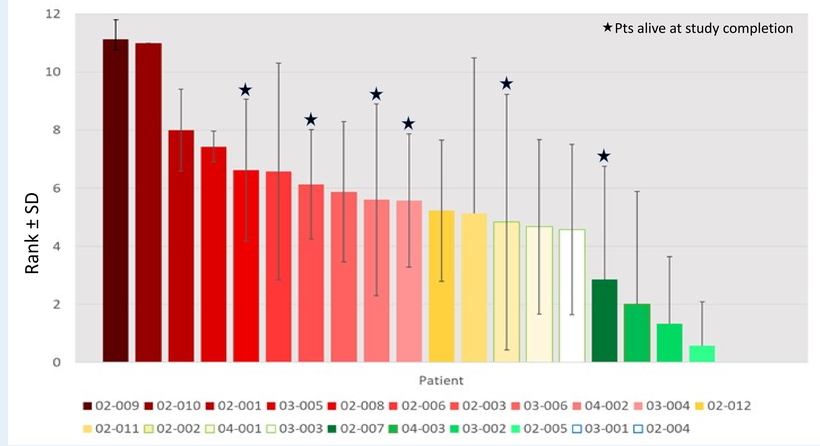
Fig 3



RESULTS

Pts were ranked according to mutation burden. The highest ranking is pts with highest mutation burden. The standard deviation was taken as a variability in mutation during treatment. A high mutation burden with low standard derivation could be taken as an indication of that the mutation burden did not change during treatment (Fig 4).

Fig 4



Changes in KRAS mutation status during treatment:

- In 5 pts all mutations were eliminated (up to 6 mutations) during treatment
- In 4 pts some but not all mutations were eliminated showing impact of treatment
- In 10 pts mutations were changed either by revealing new mutations or shift in mutations during treatment

- In 5/6 pts with KRAS mutation (n=21) still alive at study completion, the KRAS mutations were either eliminated or partly eliminated during treatment

CONCLUSION

We found that the great majority of pts with PDAC have multiple KRAS mutations and that some mutations change during the course of the study. Single mutation vaccines and small molecules targeting single mutations are therefore not likely to be effective while therapies targeting a mix of KRAS mutations such as TG01/GM-CSF should be more beneficial.

References:

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