Tumor Mutational Burden (TMB) and Neoantigen Load (NAL) estimation model using targeted Next Generation Sequencing (NGS) gene panels

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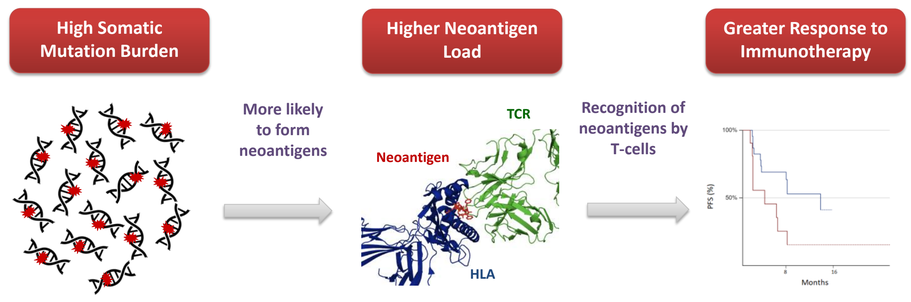
Introduction

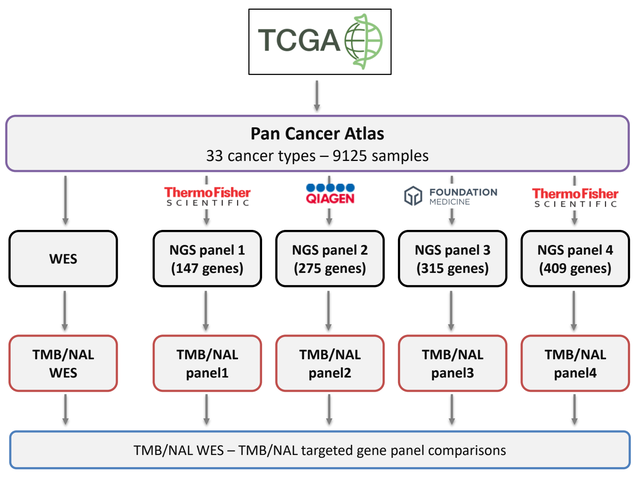
Tumor Mutational Burden (TMB) or Load (TML) is an emerging, independent biomarker [11] of outcomes with immunotherapy in multiple tumor types [1-6,10]. It is measured as the total number of somatic mutations that exist within a tumor's genome as usually determined by Whole Exome Sequencing (WES). A subset of these mutations may result in an expressed protein, termed neoantigen, that is not recognized by the host's immune system as self, and therefore has the potential to be immunogenic, leading to an anti-tumor immune-mediated response. Measurements of TMB (Mutations per megabase (Muts/MB)) from comprehensive gene panels [7,9] are strongly reflective of measurements from WES and provide a feasible, cost- and time- effective approach in clinical practice.

The aim of this study was the construction of a mutational burden and Neoantigen load (NAL) estimation model that can be used for the prediction of immunotherapy treatment response.

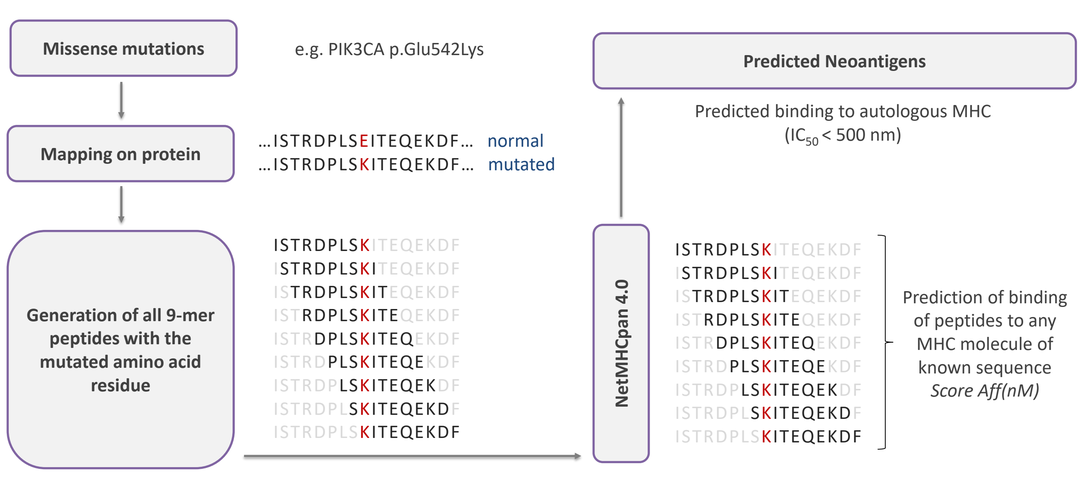


Methods

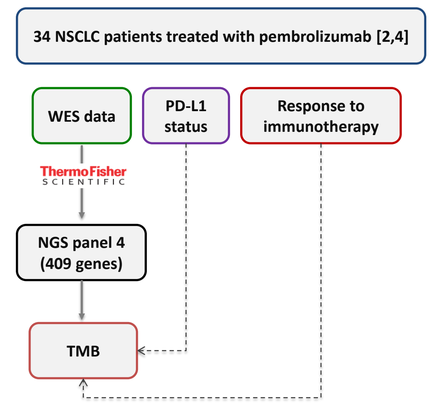
The methodology used for the selection of the optimal size the multi-gene panel approach:



The methodology used for Neoantigen prediction from SNVs:

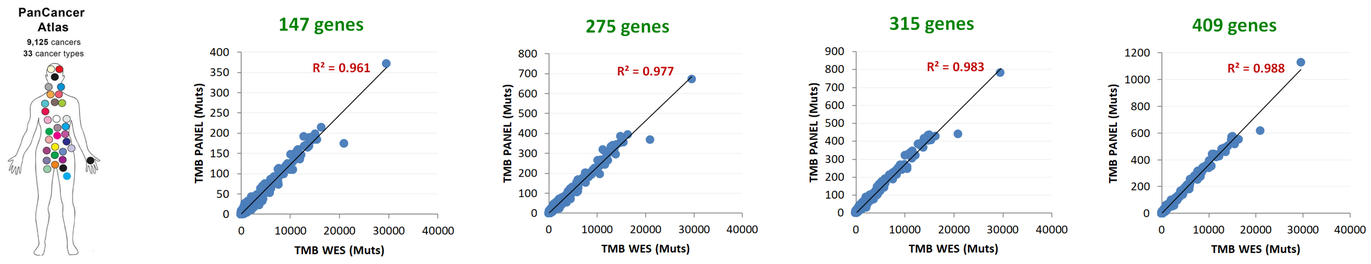


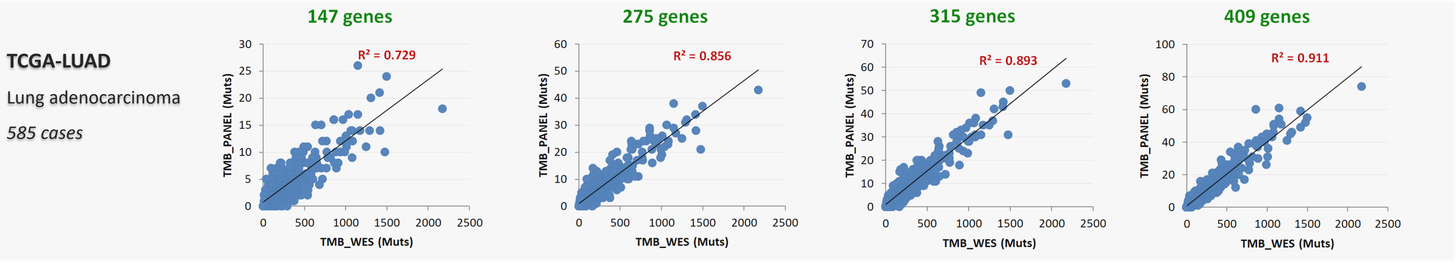
Data of response to immunotherapy for lung cancer were used to assess the predictive value of the approach on real treatment data:

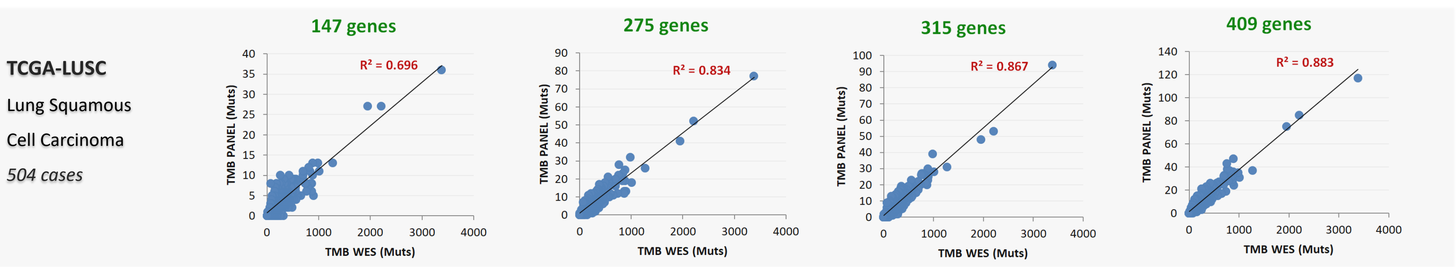


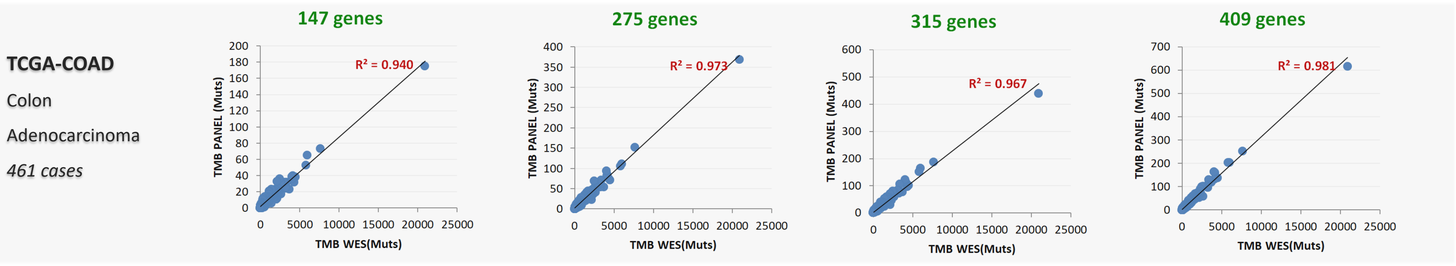
Results

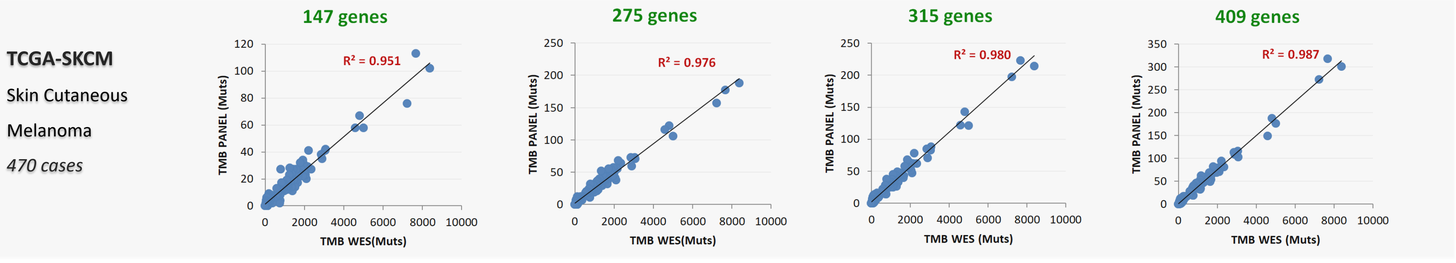
The results from the simulated application of different approaches on TCGA data. In each case TMB values were computed using only the fraction of mutations detectable by each multi-gene panel approach and were compared to the actual TMB value obtained from the WES data. Similar comparisons for NAL show that the higher number of genes used in the approach results to more correlated NAL values compared to WES data.

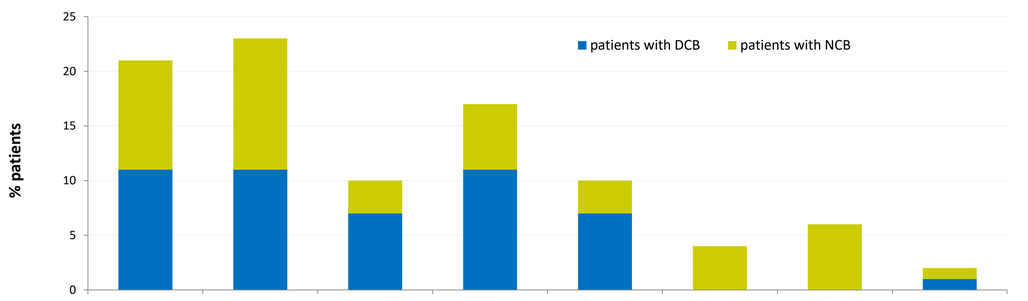






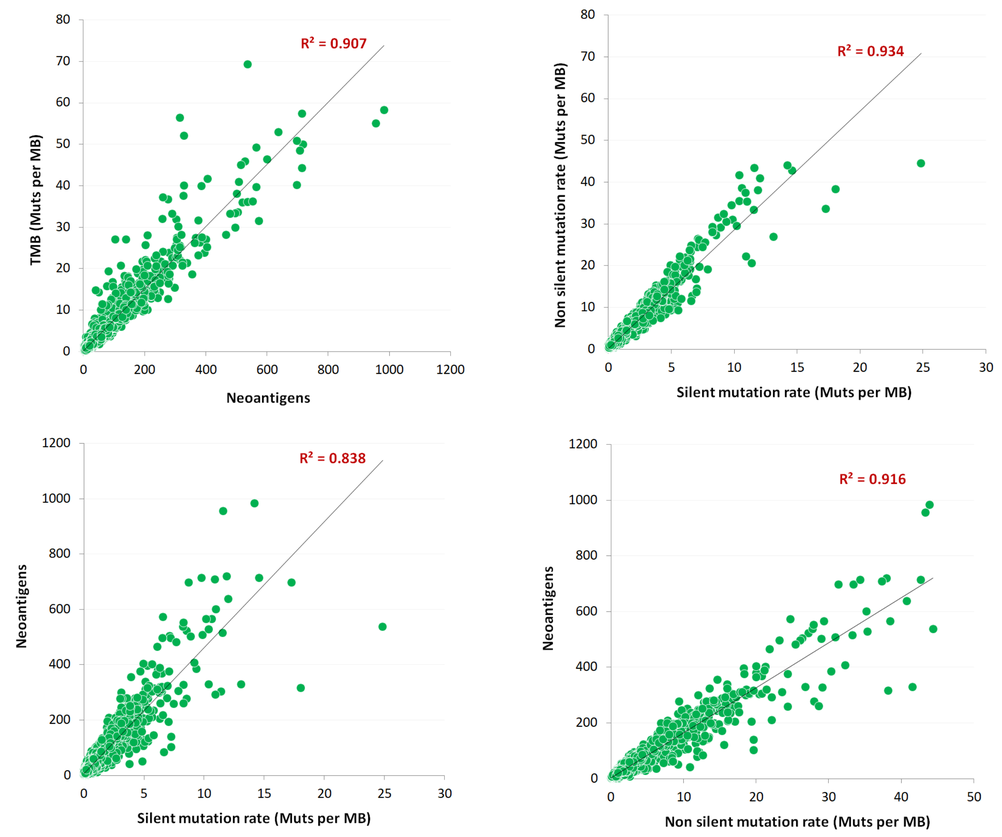






%DCB	52%	48%	70%	65%	70%	NA	NA	50%

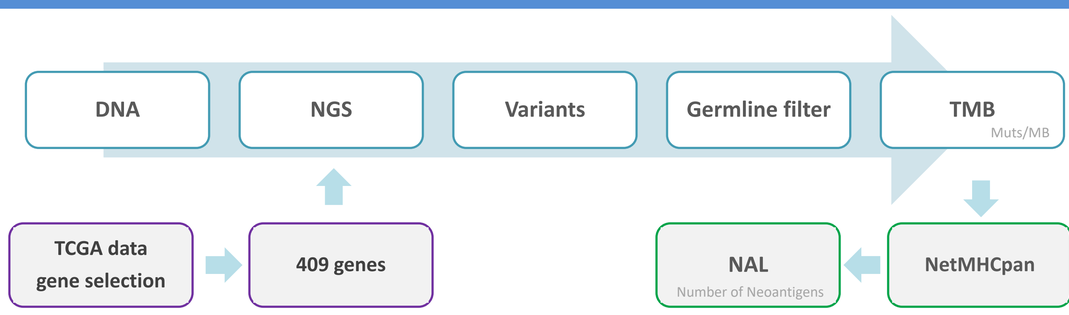
Results of the clinical utility of the TMB value obtained from the multi-gene panel approach of 409 genes for the 34 NSCLC patients with data about their response to immunotherapy (pembrolizumab). A composite of TMB and PD-L1 expression values may be most helpful in identifying with precision patients most likely to benefit. TMB high is defined as >=17 Muts/MB.



The Neoantigen load (NAL) of the PanCancer Atlas samples compared to the their total mutational rate, as well as the silent and non silent mutation rates. A higher number of mutations results to a higher number of neoantigens.

Discussion

Somatic mutation data from TCGA's Pan-Cancer Atlas were analyzed for the development of a computational framework that accurately assesses TMB and NAL from a gene panel with NGS. Comparisons of TMB with the predicted number of neoantigens (NAL) shows that tumors with a high mutation burden may have a higher rate of neo-antigens which, in principle, would be expected to be more immunogenic than tumors with comparatively low mutation burden. The silent mutation rate also correlates with the predicted number of neoantigens, supporting the inclusion of synonymous mutations in the TMB calculation approach. As noted before [9], while synonymous mutations are not likely to be directly involved in creating immunogenicity, their presence is a signal of mutational processes that will also have resulted in nonsynonymous mutations and neoantigens elsewhere in the genome. The computational pipeline described is used to tailor a designed targeted NGS cancer panel for estimation of TMB and NAL or can be adopted by custom NGS gene panels to guide the employment of targeted therapies towards a personalized use of immunotherapy in cancer.



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