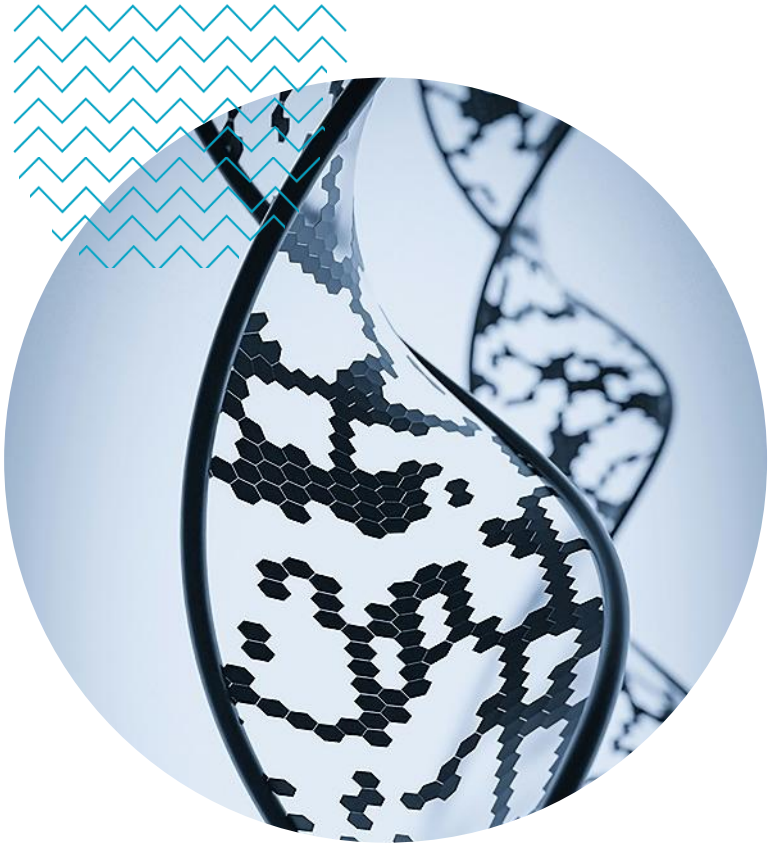
myNEO Therapeutics



Camyotopes

A novel class of tumor targets

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Camyotopes: a novel class of tumor targets

Introduction

Therapeutic cancer vaccines are increasingly gaining momentum, fueled by **promising results obtained with personalized vaccines**. Most notably, in an adjuvant setting, both Moderna and BioNTech have reported remarkable responses showing significant reduction in recurrence in high-risk melanoma and pancreatic cancer patients, respectively1,2. The positive data provides additional rationale for vaccinating cancer patients to elicit strong anti-tumor immune responses, and demonstrates that advanced data-driven technology platforms can be leveraged to discover the most appropriate vaccine targets.

Besides the computational obstacles associated with in-silico identification and selection of immunogenic personalized neoantigens, which many companies such as myNEO Therapeutics have tried to address, there are several other challenges that make personalized cancer vaccination difficult to incorporate into the Standard of Care for every cancer patient. These include logistics challenges, higher manufacturing costs, as well as a negative impact on the time from diagnosis to treatment of a patient.

Off-the-shelf vaccines provide an alternative strategy but have not yet been able to deliver clinical benefits on par with those obtained in a personalized setting. Off-the-shelf vaccine formulations target shared targets, i.e., tumor-associated (TAA) or shared tumor-specific (TSA) antigens. This type of vaccines is readily available (optimal diagnosis-to-treatment time) and facilitates prompt administration during the treatment window to avoid progression or interference from other treatments. Also, the cost of the therapy can be significantly lower since the product can be produced in large batches. Furthermore, patient selection (if even needed at all) can be performed in a fast and cost-efficient way by means of, for example, straightforward multiplex quantitative PCR (qPCR) analyses.

The lack of clinical benefit from off-the-shelf formulations can be attributed to several factors, the most important one being the scarcity of targets and thus a higher risk of immune escape. Furthermore, current shared vaccines struggle with limited coverage, and have therefore often shown ineffective for a large fraction of a given patient population. Indeed, although the well-known recurrent mutations have relatively high population frequencies – e.g., up to 50% for the BRAF V600E in certain sample cohorts – prevalence values are usually low (< 5%) for most recurrent mutation-derived neoantigens. This means that for most tumor types, the common off-the-shelf vaccines typically target one (highly) frequent neoantigen extended by a set of suboptimal low-prevalence neoantigens, achieving a maximum population coverage of typically 20-30%, mostly attributable to a single neoantigen for any given patient.

To overcome these shortcomings, novel antigen discovery avenues are being considered, such as the exploration of the dark genome. It is postulated and confirmed that tumors do not necessarily follow the borders of coding versus non-coding, dark genome that are typically observed in healthy cells.

Advanced AI analysis of the tumor epitope repertoire led to the discovery of tumor-specific long non-coding RNAs (camyoRNAs) containing one or more translatable small open reading frames (smORFs). Translation of these smORFs leads to camyopeptides which often contain epitopes (camyotopesTM) that are presented at the tumor cells' surface on MHC-I complexes and recognized by CD8+ T-cells (**Figure 1**). These camyotopes are of particular interest as immunotherapeutic targets because of their attractive characteristics: high tumor-specific abundance, first-in-class levels of coverage (sharedness) amongst cancer patients, favorable translation and presentation profiles, and high immunogenicity in comparative assays.

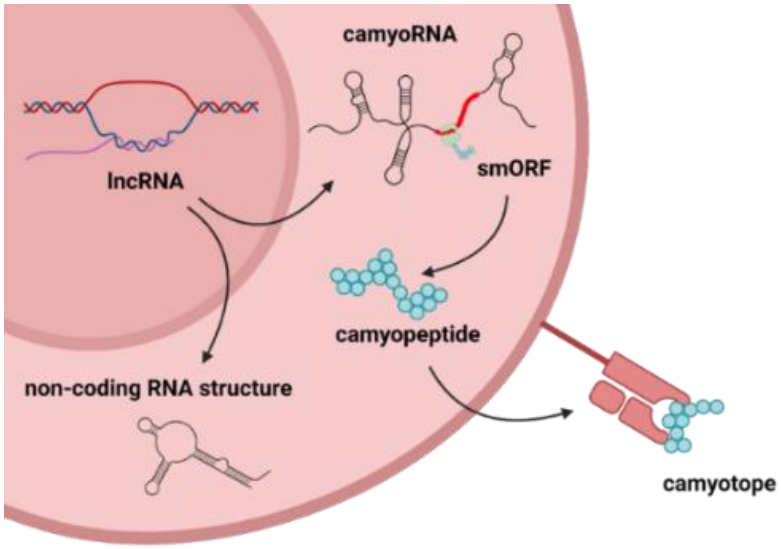

Figure 1. The biological process of camyotope presentation. A camyoRNA is a tumor-specific lncRNA comprising one or more translated smORFs that produce camyopeptides. These camyopeptides are processed through the proteasomal cleavage pathway into small epitopes (camyotopes) that are loaded and presented on MHC-I complexes on the cancer cell surface. These camyotope-MHC-I complexes can be recognized by cytotoxic CD8+ T-cells that induce a tumor-killing effect.

The potential of camyotopes

Camyotopes hold great potential over other shared antigen types such as shared SNVs (e.g., KRAS mutation) and canonical TAAs (e.g., MAGE-A3). Unlike shared SNVs, camyopeptides are not restricted to the occurrence of recurrent mutations within the cancer genome. Due to their tumor-specific expression profiles, any immunogenic camyopeptide derived from the translated smORFs can be targeted as a TAA in cancer therapies, thus significantly expanding the pool of targets. The tumor-specific nature of camyoRNAs also suggests their critical role in tumor initiation and progression, and combined with the multi-targeted immune responses that can be elicited against the large set of camyotopes present in tumor cells, this strongly reduces the chance of immune escape when targeting them as part of a vaccine.

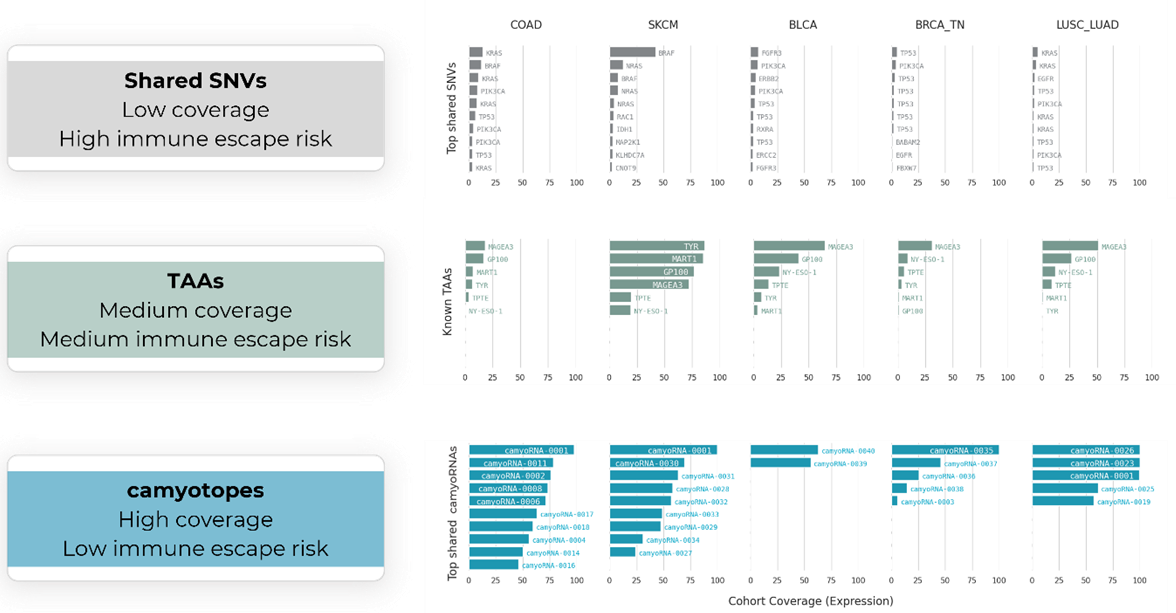

Figure 2. Per-epitope coverage of various off-the-shelf targets in different cancer populations. Based on mutation presence for shared SNVs, and on expression (>1TPM) for TAAs and camyotopes.

Furthermore, due to the abundance of camyotopes and their large coverage across patients as compared to shared SNVs and canonical TAAs, these drug products can improve the efficiency of off-the-shelf therapeutics by increasing population coverage and the number of targetable antigens per patient. Indeed, **Figure 2** indicates that across different cancer indications, a higher number of camyoRNAs is observed that is expressed across a wider portion of the cancer population: on average, camyoRNAs reach a coverage between 60% and 95%.

Additionally, camyoRNA expression is often limited to a specific tissue or tumor type, indicating camyoRNA-derived peptide targeting therapies could be associated with limited off-target effects. Extreme care must be taken when selecting suitable targets to avoid unwanted off-target effects, such as observed in past clinical trials using for example anti-*MAGE-A3* TCR gene therapies[3]. Fortunately, preliminary reports from ongoing clinical trials indicate that cancer vaccine therapies targeting for example *MAGE-A3*, *GP100*, *TYR,* and *NY-ESO-1* tend to induce more limited toxicity than treatments based on engineered cells[4-10]. As such, we expect that camyoRNAs that are sufficiently specific to the tumor and show expression profiles better than or similar to well-known protein-coding TAAs will likely have similar manageable toxicity profiles.

Besides tumor-specific expression, it was also observed that camyoRNA expression is stage-agnostic. Upon investigation of patient samples of different colorectal cancer stages, it was found that camyoRNA prevalence remained similar regardless of the stage, indicating that camyotope-based therapeutics can have relevance for both late- and early-stage patients.

Immunogenicity screening on healthy donor material of these targets revealed that a large fraction of camyotope pools exert multifunctional, highly immunogenic responses, both in high and low immune reactive donors, strongly outperforming common TAAs.

Conclusion

The magnitude of T-cell responses, the large pool of available targets positively impacting patient population size and significantly reducing immune-escape risk, as well as the limited predicted off-target toxicity, and simplified logistics showcase the unprecedented potential of the camyotopes versus other vaccine approaches (Table 1). Up to now, *in silico* and pre-clinical validation data has shown very promising results in support of further clinical efficacy studies.

Table **1** Comparison of various vaccine-based approaches.

	Patient population size	Immunogenicity	Immune-escape risk	Logistics	Safety
Prophylactic viral vaccine targets					
Viral antigens	++	+++	++	++	++
Therapeutic cancer vaccine targets					
Classical Tumor Assoc. Antigens	+- (30%)	-	-- 1 target /pt	++	+
Personalized mutational neoantigens	-- (1 patient)	+	+ 4-6 targets /pt	--	++
Camyotopes	++ (60-95%)	++	++ 6-8 eff targets /pt	++	+

It is noteworthy that off-the-shelf vaccine therapeutics targeting multiple shared antigens at once covering broad patient populations will very likely not be an effective strategy for every cancer indication and every patient cohort. In these situations, it is essential to explore other antigen targets to supplement the camyotopes (e.g., with targets originating from intron retention events, transposable elements, and alternative splicing events) thereby increasing the population coverage as well as lowering the risk of immune escape. Furthermore, off-the-shelf strategies can also be complemented with personalized, tailor-made vaccines in certain indications where shared antigen targets are too sparse.

To conclude, camyotopes have great potential as novel shared therapeutic targets and are an important addition to the arsenal of cancer treatments. They can be used as such or can be complemented with other shared and/or personalized antigen targets as well as can be combined with other immunotherapeutic agents such as checkpoint inhibitors to optimally prime and boost the immune system to attack the malignant cells.

References

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