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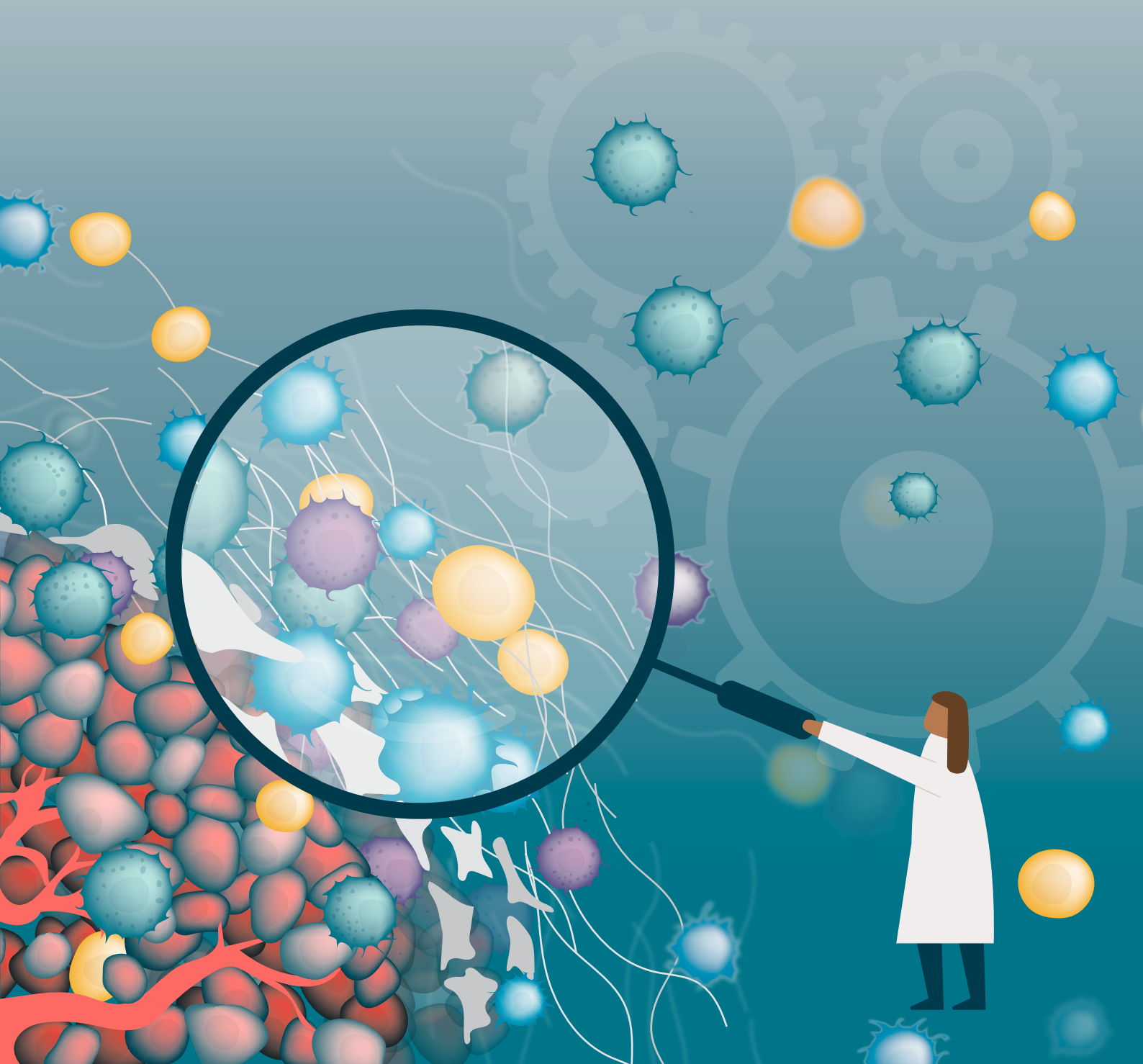
IMMUNO-ONCOLOGY INSIGHTS

SPOTLIGHT ON

Enhancing clinical development and patient stratification



CONTENTS

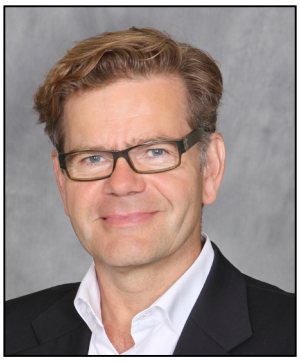
SPOTLIGHT Enhancing clinical development and patient stratification

ENHANCING CLINICAL DEVELOPMENT AND PATIENT STRATIFICATION

SPOTLIGHT

INTERVIEW

Challenges and innovations in patient selection, biomarkers, and personalized treatments within the I-O landscape



Lauren Coyle, Commissioning Editor of *Immuno-Oncology Insights*, speaks with **Oliver Rosen**, Chief Medical Officer of Akamis Bio, to discuss the impact of accurate patient selection on the effectiveness of I-O treatments and the limitations of current biomarkers in patient selection.

Immuno-Oncology Insights 2024; 5(3), 99–105

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Q Can you give us a brief overview of your background and what you are currently working on?

OR: During my academic career, I trained in malignant hematology and medical oncology at the Charité Hospital in Berlin, Germany. Currently, I serve as the Chief Medical Officer of Akamis Bio and have held leadership positions at several biotech companies over the past decade.

At Akamis Bio, we are developing a portfolio of therapeutics based on our Tumor-Specific Immuno Gene Therapeutics (T-SIGn) platform. These therapeutics are based on a replication-competent chimeric group B adenovirus, which enables intravenous (IV) delivery and selective replication in both primary and metastatic epithelial cancers. T-SIGn therapeutics are designed to home specifically to solid tumors following IV delivery and drive expression of therapeutic proteins to remodel the tumor microenvironment (TME) and trigger robust antitumor immune responses.

Our lead program features a CD40 agonist monoclonal antibody that can be safely administered, with its activity confined to the TME. Notably, in our first human study, we demonstrated the superiority of IV administration, as evidenced by enhanced virus persistence and systemic detection of transgene mRNA.

Q How does accurate patient selection impact the effectiveness of I-O treatments?

OR: The introduction of personalized medicine has revolutionized patient selection by focusing on genetic alterations, marking a significant departure from the traditional all-comer approach to chemotherapy. We anticipate that more precise patient selection for I-O treatments will enhance their real-world effectiveness. This approach also reduces unnecessary toxicity, enables smaller clinical trials, and ultimately accelerates patient access to new treatment options.

Currently, we utilize three types of biomarkers to guide treatment with PD-1 and PD-L1 inhibitors: PD-L1 expression, tumor mutation burden (TMB), and mismatch repair status. While the benefit of PD-L1-based patient selection varies across tumor types and treatment settings, it remains valuable in identifying patient populations most likely to benefit from these checkpoint inhibitors.

The use of PD-1 and PD-L1 inhibitors in patients with microsatellite instability or mismatch repair deficiency has been a significant breakthrough across multiple tumor types. Improved patient selection is expected to further enhance the effectiveness of existing I-O treatments, such as CTLA-4 and LAG-3 inhibitors, as well as emerging treatment options and combinations.

Q Further to that, what are the main challenges faced in the patient selection process?

OR: There are three primary challenges in the patient selection process. The first challenge is the limitations of our highly effective research model. While the field has developed sophisticated model systems that have advanced our understanding of cancer immunology, the insights gained from basic research do not always fully translate to human biology.

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"It takes considerable time to comprehend the biology of a potential biomarker in humans. Experimental medicines are often tested in heavily pretreated patients, where response rates to monotherapy are typically low."

One potential explanation is that the TME in preclinical studies, which develops for up to one month following tumor implantation, may not mirror the same immunological pathways and complexity as a TME in human malignancies, which develop over years.

The second challenge is our limited understanding of the biology of a potential biomarker. It takes considerable time to comprehend the biology of a potential biomarker in humans. Experimental medicines are often tested in heavily pretreated patients, where response rates to monotherapy are typically low. This low number of responders makes it difficult to distinguish between responders and non-responders, necessitating the gradual evolution of our understanding as more clinical trials are conducted. For example, PD-L1 expression testing was introduced early in clinical trials, with various thresholds for expression levels being assessed. The assessments included expression limited to tumor cells, known as the tumor proportion score, and expression on tumor-infiltrating cells, referred to as the combined positive score.

Given our understanding of ICIs, it is logical to consider TMB as a compelling biomarker for patient selection. However, there is still ongoing debate amongst experts regarding whether TMB is purely a predictive biomarker or also a prognostic one. Additionally, various cutoff levels have been proposed and tested. For example, the package insert of a PD-1 inhibitor indicates that adults and pediatric patients with unresectable or metastatic TMB-high cancers, who have progressed following prior treatments and have no satisfactory alternative options, are eligible for a PD-1 inhibitor if their TMB is equal to or greater than 10 mutations per megabase, as determined by an FDA-approved test.

A thorough understanding of the clinical value of PD-L1 expression and TMB is particularly important as we explore more I-O combination treatments. One example is the first prospective clinical validation of a TMB cutoff of 10 mutations per megabase or higher, used in non-small cell lung cancer for a combination treatment with a PD-1 and CTLA-4 inhibitor, compared to chemotherapy.

Finally, the third challenge is the lack of harmonization in biomarker testing. Access to PD-1 or PD-L1 inhibitors is often restricted by specific companion diagnostics, which are typically limited to certain agents. There is still significant variability between different assays in terms of both performance and cutoff points. Similarly, we need to improve the standardization of TMB assays, a challenge that has been widely recognized by the field. The Friends of Cancer Research assembled a TMB harmonization working group, which established a gold standard method for determining TMB and published recommendations for cutoff levels. While progress has been made in this area, it is now up to the broader community to implement these recommendations.

Q How do you balance the need for precision in patient selection with the practical limitations of current diagnostic tools?

OR: The community is well-aligned and ready to embrace creativity during this time of significant transition. While the example of combining PD-L1 expression and TMB is one approach to enhancing precision, incremental improvements in patient selection for I-O treatments will not be sufficient. Simply adding one biomarker after another is not a viable long-term strategy. Instead, we must explore new approaches to drug development.

One likely path forward involves parallel assessments that utilize current diagnostic tools alongside exploratory approaches capable of integrating AI-based high-throughput technologies. For example, this could include immunohistochemistry (IHC) assessments combined with multi-omics analyses, which could be either RNA or protein-based. However, such trials place a much higher burden on sample collection, which may not be appealing to every patient or feasible at every cancer center.

Patients who are actively involved in decision-making might find these trials particularly appealing. Additionally, the advanced state of analytical approaches available today, along with creative partnerships, gives hope that even smaller biotech companies can pursue these integrated approaches.

An exciting example of this is being pursued at Akamis Bio, where the team is working to expand existing diagnostic tools. The upcoming FORTRESS study in locally advanced rectal cancer includes circling tumor DNA (ctDNA) testing to determine whether adding the T-SIGn program to chemoradiotherapy provides clinical benefit. Longitudinal ctDNA data collected during the 12-week neoadjuvant treatment period will be made available to patients and their cross-functional treatment teams to help guide the best next step, whether that be consolidation chemotherapy, surgery, or a watch-and-wait approach.

There is growing evidence in colorectal cancer that ctDNA could play a crucial role in guiding decisions regarding the intensity and duration of treatment.

Q Can you discuss the limitations of current biomarkers in patient selection for I-O and how the discovery of new biomarkers could aid in treatment personalization?

OR: Unlike personalized treatment options, I-O presents a much higher level of complexity. Although targeted therapies can be highly effective, their benefits are often short-lived. In contrast, I believe that the complexity inherent in I-O holds the potential for greater rewards, particularly in the personalization of effective treatment selection. Reflecting on the development of targeted therapies, it is clear that predicting the next tumor mutation leading to secondary resistance is nearly impossible.

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"My prediction is that a combination of multiplexed IHC combined with RNA-based tests will be the way forward."

I am confident that we will soon be able to define distinct immunological subsets within TMEs. The initial classification into cold, excluded, and inflamed TMEs was a promising start, but it has not proven to be as actionable as anticipated. I do not believe that a single set of biomarkers will adequately address the current unmet medical need. While I strongly believe that tumor biopsies will be key to finding answers, I would be equally excited to discover a liquid biopsy solution for I-O.

My prediction is that a combination of multiplexed IHC combined with RNA-based tests will be the way forward. Additionally, AI-guided data integration from multiple slides of a tumor biopsy could help to address the well-documented issues of tumor heterogeneity. Although there have been some disappointments along this path, I believe it offers far more promise than the identification and validation of individual biomarkers.

Q How can different data streams be better harnessed and combined to monitor response and predict patient outcomes?

OR: Although IHC has long been the gold standard in diagnostics, I have encountered numerous cases of misdiagnosis. Pathologists increased the precision by adding new parameters and approaches. While AI may also make mistakes during its implementation, it is likely to lead to more accurate diagnoses overall. In I-O, patient selection will benefit from ongoing efforts to improve the assessment of clinical benefits, particularly through the use of new potential surrogate endpoints.

As previously mentioned, ctDNA is becoming an increasingly important tool for defining treatment duration, identifying early treatment failure, and recognizing patient subsets at risk for early relapse. This advancement will pave the way for new treatment settings, allowing for faster validation of innovative approaches or algorithms for patient selection.

Q Lastly, what are your goals and fears for the upcoming years?

OR: I am concerned that the fear of making mistakes may hinder us from implementing new approaches as swiftly as needed. My primary goal is to improve the lives of patients living with cancer. Therefore, at my company, our focus is on delivering proof of concept for the lead candidate of our T-SIGn platform through the planned FORTRESS study. With FORTRESS, we hope to identify new biomarkers in locally advanced rectal cancer

for future patient selection. As a community, I hope we will begin adopting new approaches and explore innovative partnerships in the pre-competitive space to enhance patient selection in the I-O space as soon as possible.

In addition to this, I acknowledge that combining different modalities is a significant challenge we currently face. On one hand, large biotech companies are often seen as having the tools and resources to conduct all necessary tests. On the other hand, small biotech companies are often the ones driving innovative programs. Additionally, technology companies also play a crucial role and must find ways to demonstrate their value. My hope is that we can find a way to bridge this gap by establishing innovative partnerships between small technology, diagnostics and biotech companies to address this unmet medical problem.

I remain optimistic that the increasing recognition of the need for biomarker harmonization, along with heightened scrutiny in drug development, will bring us closer together as a community. This will not only lead to incremental improvements but also to the development of entirely new solutions.

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IMMUNO-ONCOLOGY INSIGHTS

ENHANCING CLINICAL DEVELOPMENT AND PATIENT STRATIFICATION

SPOTLIGHT

INTERVIEW

Advancing cellular therapies: moving beyond the CAR-T landscape

Lauren Coyle, Commissioning Editor, *Immuno-Oncology Insights*, speaks with **Frank Borriello**, Founder and CEO, Alloplex Biotherapeutics, to discuss the challenges the I-O landscape faces in the cellular therapy landscape and the advantages that Alloplex's lead platform 'SUPLEXA cells' have in overcoming these challenges.

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Q Can you tell us about your background and what you are currently working on?

FB: I trained as an MD-PhD with a specialization in immunology at the Albert Einstein College of Medicine in New York. After that, I moved to Boston where I completed my residency as a clinical pathologist at the Brigham and Women's Hospital. It was then that I began to observe the significant vibrancy of the biotechnology industry and the emergence of I-O as a genuine therapeutic approach.

I, subsequently, transitioned into the industry sector, gaining experience with both large pharmaceutical companies and small biotech firms. My focus turned to external evaluation: a role that involves identifying and assessing promising assets, marshalling necessary resources, and ultimately supporting business development transactions. I served in this capacity at multiple companies, including Millennium, Takeda, Shire, and Baxalta which gave me a broad perspective into the factors that attract, enable, and drive transactions.

In 2015, Baxalta was acquired by Shire Pharmaceuticals while at the same time I was losing my brother to cancer. I saw an opportunity to refocus the remainder of my career in pursuit of an idea that had long intrigued me involving cellular vaccines to treat cancer. This led to the founding of Alloplex in 2016, and to the genesis of the concept that became 'SUPLEXA' our lead autologous, non-engineered cellular therapy for treating solid cancer tumors. In that time, we have achieved significant progress, transitioning from an initial concept to conducting a first-in-human clinical studies, with encouraging results now in hand. As we speak, we are approaching our pre-IND meeting with the US FDA and actively seeking investors to finance the next phase of SUPLEXA development.

Q What are the biggest challenges facing the I-O clinical landscape, and the cellular therapy landscape in particular?

FB: Despite experiencing a renaissance in scientific innovation with several groundbreaking therapies emerging, it is somewhat paradoxical that funding remains the rate-limiting step. As a result, not every promising idea can receive the financial support it deserves, necessitating careful decision-making by funding agencies, granting bodies, and investors.

The I-O field has come a long way from being a peripheral concept in cancer treatment to becoming central in contemporary medical practice. Chemotherapy, radiation, and surgery treatments remain key options for cancer patients; however, it is now widely accepted that the immune system plays a crucial role in maintaining the body free of infections and tumors and I-O has emerged as another key therapeutic pillar.

A critical issue in I-O arises from the reliance on using mouse models as a gating step to the human scenario and clinical trials. Many promising ideas are discarded prematurely as they do not perform well in murine models, despite their potential efficacy in the vastly different human clinical setting. The key purpose of murine models has traditionally been to mitigate toxicity risks and provide preliminary evidence to investors. However, this practice has—in my view—been misapplied with respect to cellular therapies because human cells can only display their true safety and efficacy within the context of a human body, where a myriad number of interactions can occur. To require the efficacy of human cells in murine models before progressing to human trials, as if they were just another pharmaceutical, is not supported by the known mechanisms of cellular action.

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"SUPLEXA cells have shown efficacy against solid tumors which represent about 90% of the overall oncology market."

Cellular therapy, leveraging the body's immune cells to perform their natural functions, emerges as a niche within a niche. Alloplex's approach involves activating the patient's immune cells *in vitro* to restore their full potential and then reintroducing them intravenously back to the patient.

The first positive clinical data from Alloplex has successfully demonstrated safety and efficacy of 'SUPLEXA therapeutic cells' not only in murine models but more dramatically in patients, therefore showing progress in navigating a key translation challenge.

Q What is the genesis of the SUPLEXA platform concept and how is it differentiated from other cell therapy approaches?

FB: SUPLEXA cells and CAR-T cells share little in common beyond the fact that both are cell therapies. The fundamental concepts underlying SUPLEXA cells originate from research conducted over decades, which demonstrated that activating even a single immune pathway has the potential to modulate the function of the patient's immune system. Building on this idea, we hypothesized that simultaneous activation of multiple immune pathways could progressively enhance the immune response even further. Our research has confirmed this hypothesis demonstrating that peripheral blood immune cells could be stimulated to proliferate robustly, produce a range of cytokine proteins capable of stimulating and recruiting other parts of the immune system, and most importantly, awaken their ability to specifically recognize and kill tumor cells.

Impressively, the dramatic immune activation required no genetic engineering, making manufacturing simpler, more robust, and reproducible compared to genetically engineered alternatives. In contrast, genetic engineering, while powerful and attractive, carries risks such as the potential for unintended genetic modifications that could lead to tumorigenesis.

While CAR-T cells have yielded significant developments in the I-O space, especially in treating liquid tumors, they have been less successful in solid tumors. The difference relates to the accessibility of tumor cells in the circulation compared to solid tumor cells shielded by a tumor microenvironment. In contrast, SUPLEXA cells have shown efficacy against solid tumors which represent about 90% of the overall oncology market.

One aspect that SUPLEXA cells and all current approved cell therapies for cancer have in common is that they are autologous. The alternative allogeneic approach has been pursued with enthusiasm for its perceived promise of commercial viability. However, this approach has

encountered numerous technical challenges and, despite significant investment, has yet to yield any approved product.

We believe the issues confronting allogeneic cell therapy are akin to those in the transplantation world. Transplants from identical siblings are known to pose no issues, but those from unrelated donors can lead to rejection and other complications. Similar principles apply to cellular therapy, underscoring the advantage of using autologous cells for the best therapeutic outcomes.

Q How have you gone about dissecting the mechanisms by which SUPLEXA cells work?

FB: SUPLEXA cells are natural cells that exist at a low level within the human body. We have discovered a method to produce these in large quantities while retaining their natural properties. They function based on immunologic principles that have been extensively studied for decades. Further, we have a comprehensive list of proteins to examine, each with well-documented functions elucidated by resolute scientists over the years.

When a protein is identified on the surface of our cells, its role can be easily inferred making it straightforward to understand a mechanism of action. For instance, there is a group of well-understood proteins expressed by activated immune cells that are responsible for tumor cell killing. SUPLEXA cells express unusually high levels of such proteins and as such are believed to be central to the mechanism through which SUPLEXA cells exert their confirmed cytotoxic effects against tumor cells.

Moreover, we have identified additional proteins that govern the migration of SUPLEXA cells within the body. These proteins are known as chemokine receptors. These receptors are well-documented, and the reagents for measuring their expression are readily available. Several chemokine receptors on the SUPLEXA cells have been identified that aid in guiding the cells to tumors, lymph nodes, and bone marrow—three critical immunological sites for effective tumor eradication.

It is important to emphasize that there is a wealth of knowledge to be gleaned from the proteins expressed on naturally occurring cells like SUPLEXA cells. We have employed advanced techniques to measure the expression of over 50,000 proteins in these cells. While it is not feasible to understand all of them immediately, we are systematically studying these proteins to gain a comprehensive understanding of the multiple mechanisms of action.

That said, it is not immediately necessary to fully understand all the cellular mechanisms before utilizing them in practical clinical applications. We have already observed clinical benefits from using SUPLEXA cells in a first-in-human clinical setting; remarkable as the Phase 1 study was only designed to verify safety and tolerability. Thus, while we continue to deepen our understanding of these cells, we can simultaneously advance our clinical work without interruption. We know enough about their functionality to proceed confidently with clinical applications, ensuring that we continue to make progress in treating patients.

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"Our laboratory results indicated that SUPLEXA cells killed all tumor types that they encountered."

Q Could you elaborate on the ongoing Phase 1 SUPLEXA clinical study at Alloplex and whether the pre-specified clinical endpoints have been met, specifically for safety and efficacy?

FB: When we initiated our Phase 1 trial, we faced a significant challenge—we did not know which type of cancer patients to enroll. Our laboratory results indicated that SUPLEXA cells killed all tumor types that they encountered. Consequently, we conducted a survey study, enrolling over 35 patients with 14 different tumor types. These patients were at the end-stage of their illness, having failed multiple prior therapies and exhausted all other options. They were offered SUPLEXA cells as a single-agent therapy. This distinction is crucial, as many other cellular therapies utilize chemotherapy in conjunction with or prior to administration, such as CAR-T therapy. In our trial, we used only SUPLEXA cells.

The results revealed that certain tumor types and specific patients responded well, while others did not. It is important to note that we did not expect to find a universal treatment for all cancers immediately. However, we observed significant benefits in patients with melanomas, colorectal, kidney, breast, and lung cancer, indicating a promising list of cancer types that show observable benefits to SUPLEXA cells.

The primary endpoint of the clinical trial was safety, assessing whether patients experienced any adverse side effects. From the results, patients did not experience any negative side effects. Many reported improvements in their quality of life, such as increased energy, the ability to return to work, spend time with their families, and engage in physical activities. These results are promising, even though the sample size was small.

We successfully met the primary safety endpoint and secondary efficacy endpoint, which was to observe any signs of therapeutic activity and improvements in patient well-being. We achieved these endpoints without any ambiguity. Additionally, we aimed to measure any changes in the blood composition of treated patients that would indicate an enhanced anti-tumor response. Within weeks of administering SUPLEXA cells, we observed and measured changes in the blood that suggested an improved immune response against tumors.

Our first-in-human Phase 1 clinical trial, conducted in Australia, has been successful, achieving all its prespecified endpoints. In addition, anecdotal reports of decreased fatigue and pain are very encouraging signs of patient quality of life benefits. We are now concluding the trial and will present a finalized report at an international conference later this year. The company is also preparing to commence a Phase 2 study next year.

Reflecting on the first day we administered the drug, it was a tense time due to the uncertainty of first-time administration, as any adverse reaction could occur unpredictably. However, we have now administered over 200 doses to individual patients without encountering

any concerning reactions. This consistency has reassured all stakeholders and now allows us to confidently inform future trial participants about the therapy's relative safety and efficacy profile.

Q Looking forward, what is the strategic path for Alloplex?

FB: Progress in this field requires active support and engagement. The challenge for Alloplex now extends beyond the science—it involves capturing the attention and imagination of motivated investors and collaborators.

Our foremost priorities are working closely with the FDA in the lead-up to the IND submission later in 2024, securing partners necessary for the manufacturing of the cells, and seeking investors who are aligned with our vision.

Cell therapy clinical trial develop relies heavily on the ability to make the product and select the correct patient population for testing. While more complex than manufacturing a pill, we have developed a robust and reproducible manufacturing process, grasping all its intricacies and nuances. The clinical strategy has also evolved so that we have now selected tumor types where we have observed single-agent activity in our first-in-human trial. Strategically, we have also decided that integrating SUPLEXA cells with existing beneficial treatments to enhance patient outcomes is the most logical path to drug approval as new therapies will not be deployed in isolation but in the context of existing treatment paradigms.

Conducting clinical trials requires substantial financial resources. We are currently engaged in discussion with potential partners and collaborators, hoping to form partnerships that will enable us to progress. In the challenging biotech investment environment, many promising companies may struggle to advance due to a lack of attention and capital, which are essential for taking the next logical step.

Q What are your main goals and fears for the upcoming years?

FB: We are living in both the best and worst of times. As an industry, we possess the capability to effectively treat certain cancers and the potential to address even more. Still, we lack the financial resources to implement these solutions as quickly as we would like.

Our main goal is to continue and expand upon our current efforts, with the confidence that comes from knowing the trajectory of the SUPLEXA cells program has been consistently positive for the past eight years. Our focus remains on developing a dataset that is both meaningful and enhances the company's value. This involves progressing to the next clinical trials as swiftly as possible—ideally conducting multiple trials if financially feasible—continuing to analyze SUPLEXA cells and demonstrating their comprehensive benefits. Everything these cells express are targets that researchers have long sought to incorporate in other cell therapies,

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and we achieve this naturally through our activation procedure. Securing the necessary funding is the only remaining barrier to realizing the potential of SUPLEXA cells as the science itself is currently locked down and derisked.

Our key task is to generate quality clinical data as required by regulatory authorities for approval, and we are preparing to discuss with the US FDA to do that.

BIOGRAPHY

FRANK BORRIELLO is the Chief Executive Officer of Alloplex Biotherapeutics Inc., Boston, MA, USA—a private company he founded based on an original concept. Dr Borriello obtained his MD and PhD degrees at the Albert Einstein College of Medicine, New York, NY, USA where he studied Class I MHC structure-function relationships. He continued training at the Brigham and Women's Hospital with a residency in clinical pathology and a laboratory research focus in cellular immunology and the B7-CD28 costimulatory pathway. He has an extensive background of over 20 years in the biotech/pharma industry spanning diverse roles such as clinical development (Wyeth), financial buy-side analyst (BB Biotech) and business development with Millennium, Takeda, Shire and finally at Baxalta as VP of Search and Evaluation until its acquisition in 2016. In 2016, he conceived of a novel differentiated, non-engineered approach to immuno-oncology (now called 'SUPLEXA' and protected by both issued patents and trade secrets) and established Alloplex Biotherapeutics to reduce the concept to practice. As Scientific Founder and CEO of Alloplex, Dr Borriello has led the organization through research, fundraising and business development, and into first-in-human clinical trial for the lead SUPLEXA cell therapy program which is currently underway in Australia. SUPLEXA is a potential first-in-class, individualized, pan cancer-cellular therapy comprised of highly activated cells with well-understood anti-tumor activity.

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AUTHORSHIP & CONFLICT OF INTEREST

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IMMUNO-ONCOLOGY INSIGHTS

CORRIGENDUM

Corrigendum to: Revolutionizing cancer treatment? A conversation on the potential of personalized cancer vaccines

Roy de Souza and David Hawke

In the version of this Interview initially published, Roy de Souza expressed his opinion on the challenges companies have faced in the development of personalized cancer vaccines (*Immuno-Oncology Insights* 2024; 5(1), 65–74; pp 68–69); however, this has since altered.

To reflect this change, we have amended the HTML and PDF versions of this article as of May 30, 2024 and which may be accessed here.

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