

Turn up the MIC(A/B): Amplifying stress-ligand recognition to target iPSC derived immune cell therapies against tumors and diseased cell states

John Goulding^{1*}, Alejandro J. Garcia¹, Martin P. Hosking¹, Peter Szabo¹, Bahram Valamehr^{1*}

¹Fate Therapeutics Inc., San Diego, CA, 92131, USA

*Author for correspondence: Email: bob.valamehr@fatetherapeutics.com; bobby.goulding@fatetherapeutics.com

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Commentary

Cancer is a multifaceted disease that is overwhelming in the breadth and scope of its genetic diversity, tissue pathology, and response to therapy [1]. In recent years, the development of novel chimeric antigen receptor (CAR) containing cell-based therapies has provided significant clinical benefit to patients with certain hematological malignancies [2]. The overall response rates (ORR) of Food and Drug Administration (FDA) approved autologous CD19 CAR T cell products for the treatment of refractory and relapsed B cell lymphomas have reached approximately 80% in defined patient populations, with durable remissions of greater than several years [3].

Regrettably, the deployment of these novel cell-based therapies against solid tumor indications has garnered much less enthusiasm due to disappointing clinical success [4,5]. The intrinsic adaptability of solid tumors makes them particularly elusive for effective targeted treatment, in part due to their extensive cellular heterogeneity and antigen diversity, their capacity to evade immune responses through antigen escape, and their ability to establish immune suppressive microenvironments [6]. Identifying and selecting appropriate tumor specific antigens that distinguish tumor cells from healthy tissue remains arguably one of the most essential requirements to develop efficacious solid tumor targeting cell-based therapies. A unique solution to this problem was recently described in our latest study, in which a novel acting CAR was developed to target a specific region of an inducible stress protein class, expressed on many cancers, that simultaneously prevents tumor immune evasion and provides specific tumor killing whilst leaving healthy tissue intact [7]. To develop a unique cell therapy product that overcomes the many challenges present in solid tumor indications, we combined the novel acting pan-tumor reactive CAR system with an 'off-the-shelf' allogenic cell therapy product, by generating multiplexed-engineered induced pluripotent stem cell-derived CAR natural killer (CAR-iNK) cells that contain synthetically enhanced functionality to promote anti-tumor potency, multiantigen capability and on-demand availability. This commentary aims to continue to explore the fundamental concepts of the original research article, while also discussing potential future applications for this innovative antigen targeting technology.

Stabilizing Stress Ligand Expression Presents a Unique Tumor Recognition Strategy

Existing cancer therapeutic modalities, such as chemotherapy, radiation, and monoclonal antibodies, are minimally tumor cell selective and detrimentally impact all exposed cells, healthy or diseased, which can result in significant 'off target' toxicities and collateral damage to healthy tissue. Despite providing improved precision capability, on-target off-tumor safety concerns have occurred in patients treated with autologous anti-CD19 and anti-B cell maturation antigen (BCMA) CAR T cells [8,9]. Their ability to provide deep depletion of both healthy and malignant B cells, and their capacity to react to low target antigen expression, has elicited prolonged immunocompromised states, cytokine

release syndrome (CRS) and accompanying immune effector cell-associated neurotoxicity syndrome (ICANS). In contrast to hematological malignancies, solid tumors typically arise from ecto- or endoderm derived cell lineages, meaning they share a myriad of self-antigens common to vital organs and connective tissue within the body. Early autologous CAR-T cell therapies targeting human epidermal growth factor receptor 2 (HER2), a mitogenic receptor tyrosine kinase highly expressed on the surface of many solid tumor indications, demonstrated significant reactivity to healthy tissues that also express HER2 on their surface. Low-level expression of HER2 on healthy lung epithelial cells triggered on-target off-tumor HER2 CAR T cell mediated CRS that resulted in a serious adverse drug event that culminated in a patient death [10]. Consequently, the need to effectively distinguish tumor cells from healthy tissue is critical for the development of safe and effective CAR targeted cell-based therapies against heterogeneous solid tumors. To achieve this, we leveraged elements of an evolutionary conserved innate immune surveillance axis that relies upon the natural killer group 2 member D (NKG2D) activating receptor's ability to recognize the stress ligand family members major histocompatibility complex class-I polypeptide-related sequence A (MICA) and B (MICB). MICA and MICB (MICA/B) belong to a polymorphic and polyallelic family of inducible stress ligands that are structurally similar and related to the human leukocyte antigen (HLA) A, B and C genes (human MHC I) [11]. Surface MICA/B expression is tightly controlled and augmented only in response to DNA damage or cellular stresses, which allows for context specific expression and immune cell recognition through detection via NKG2D [12-16].

Intriguingly, although MICA/B stress protein transcripts are detectable across many cancer indications, their surface expression is difficult to detect as they are often shed via a well-defined proteolytic process that takes place within the membrane-proximal alpha-3 domain in response to tumor cell proliferation and metastatic transition [17-19]. This cleavage event contributes to tumor immune evasion via MICA/B antigen loss, and the subversion of NKG2D-dependant effector function via binding of shed MICA/B to collectively facilitate tumor escape [20-22]. In contrast, MICA/B stress ligands are seldom detected on the surface of healthy cells, making them an attractive therapeutic CAR target. To this end, we hypothesized that restoring aspects of the dysfunctional tumor associated NKG2D:MICA/B immune surveillance axis would provide a novel therapeutic approach to synthetically deliver anti-tumor immunity that can effectively distinguish 'altered self' MICA/B expressing tumor cells from their surrounding 'normal self' healthy tissue. To achieve this, we developed a novel MICA/B specific CAR that targets the conserved membrane proximal alpha-3 domain of MICA/B, aptly referred to as 3MICA/B CAR, which can simultaneously prevent the shedding of surface MICA/B by sterically preventing the binding of the enzymes responsible for cleavage, thereby stabilizing surface MICA/B and circumventing cleavage-driven antigen escape [7,23,24]. As the initial proof of concept, the 3MICA/B CAR was expressed on the surface of primary T cells and an induced pluripotent stem cell (iPSC) derived NK cell (iNK) to establish that signaling via the 3MICA/B CAR in response to a range of surface MICA/B densities facilitates robust cytokine production and cytolytic killing across a broad collection of tumor types [7]. Critically, in contrast to natural NKG2D receptor or NKG2D CAR, this functionality occurred with high tumor-cell binding avidity and was maintained in the presence of natively shed MICA/B, as the

3MICA/B CAR is resistant to competitive binding inhibition from the cleaved membrane distal alpha 1-2 domain MICA/B subunits [7]. The 3MICA/B CAR tumor targeting approach is profoundly different to that of other CAR targeted cell therapies currently in pre-clinical and clinical development. Rather than defining and targeting a unique solid tumor associated antigen (TAA), a challenging task across the huge diversity and heterogenous nature of solid tumors, within and across tumor types, the 3MICA/B CAR is agnostic to tumor cell ontology and can detect an induced 'altered self' cell state associated with distress and 'disease'. The 3MICA/B CAR effectively acts as an upgraded synthetic immune surveillance system that is resistant to tumor antigen escape and immune evasion whilst simultaneously providing pan-allelic and pan-tumor recognition potential. A parallel approach using an alternative therapeutic modality that targets MICA/B stress ligands is currently being evaluated in the clinic (ClinicalTrials.gov Identifier: NCT05117476) and aims to provide additional safety data and validation of MICA/B as a promising clinical tumor target.

iPSC Derived Cell Based Therapies – The Future Starts Now

The dependency of autologous cell-based therapies on complex and costly manufacturing processes to generate individualized drug product, and the difficulties associated with incorporating multiple engineering steps, continues to drive rapid innovation and investigation into the potential efficacy and commercial application of allogeneic cell-based therapies. Despite initial setbacks, allogeneic based cell therapies continue to promise multi-editing potential, a more consistent product quality and potency profile, and a lower cost streamlined manufacturing timeline to allow for readily available 'off the shelf' on demand drug product [25]. Allogeneic NK cell adoptive transfer has demonstrated clinical benefit in patients with advanced cancers, however, there remains inherent limitations with respect to the absolute number of primary NK cells that can be isolated during apheresis, with marked inter-donor variability in quantity and quality [26,27]. To overcome these significant limitations, we have pioneered a platform for the large-scale manufacture of multi-edited iPSC derived effector immune cells. A crucial advantage of this approach is the ability to multi-plex the precise engineering of clonal master iPSC lines, thereby eliminating the stochastic editing variability associated with pool editing strategies, which provides a unique opportunity for genomic stability and integrated quality control, whilst simultaneously delivering a consistent, cost effective and scalable manufacturing process [28,29]. The capability to mitigate against unwanted genomic stability and integration events has recently been brought into focus following the concerns surrounding T cell malignancy risks attributed to existing autologous and allogeneic CAR T cell-therapy manufacturing and clinical application [30]. The ability to generate large quantities of highly efficacious multi-edited iPSC derived immune cells was highlighted in our study through the bi-allelic incorporation of the 3MICA/B CAR into a multi-edited iNK cell containing a high affinity non cleavable CD16 (hnCD16) receptor. Combining 3MICA/B CAR with synthetically enhanced antibody dependent cellular cytotoxicity (ADCC) augmented anti-tumor efficacy against tumors composed of cells expressing heterogenous tumor antigens. The benefits of multi-antigen targeting and multi anti-tumor attributes reflect the rapidly evolving landscape of cancer therapy, where the synergistic use of multiple anti-tumor therapeutic modalities in combination

can significantly improve outcomes. Collectively, combining an innovative iPSC based immune cell manufacturing platform with multiple novel anti-tumor modalities provides a glimpse of how the convergence of multiple pieces of synthetic and regenerative biotechnology is driving innovation within the cell-therapeutic arena.

Clinical Translation - From Oncology to Cellular Senescence Through Viral Infections

While the focus of our study was the application of the 3MICA/B CAR in the context of cancer immunotherapy, the principles and concepts highlighted in our research hold much broader clinical implications. The idea of restoring defective, or evaded immune surveillance, with a synthetically enhanced immune cell that can detect and eliminate stressed, diseased, or infected cells, in combination with therapeutic monoclonal antibodies to provide multi-antigen targeting, is not limited to the treatment of cancer. MICA/B expression has been implicated in a variety of acute and chronic viral infections, a consequence of convergent evolutionary selection between viral immune evasion strategies and host adaptation [31]. CD4 T cells upregulate surface NKG2D ligands following acute HIV infection, and elevated plasma levels of shed MICA/B is associated with dysfunctional NK cell responses often observed in treatment naïve chronically infected HIV patients [32,33]. Elevated plasma levels of MICA/B are also detected in patients with hepatitis B virus (HBV) and human cytomegalovirus infections (HCMV) [34,35]. Intriguingly, NK cell dysfunction and loss of NKG2D expression is observed in patients with severe coronavirus disease 2019 (COVID19) infections [36]. Although this study did not specifically measure soluble plasma MICA/B levels, a loss of NKG2D surface expression is a hallmark of soluble MICA/B:NKG2D receptor binding interaction. How precisely a 3MICA/B CAR containing cell-based immunotherapy might be utilized in an infectious disease setting remains without precedent, however the existence of significant unmet clinical need for patients with latent HIV infection and other incurable chronic viral infections is unquestionable. One approach we are actively pursuing is to leverage genotoxic chemotherapeutic agents known to provoke MICA/B expression, such as the HDAC inhibitor romidepsin in multiple myeloma, and mitoxantrone and epirubicin therapy in prostate and breast cancer patients respectively, to enhance the therapeutic window for 3MICA/B CAR therapy application in pre-clinical tumor and viral infection models [37,38]. While systemic chemotherapy has been demonstrated to increase MICA/B expression via genomic toxicity in both diseased and healthy tissues, the use of low dose radiation therapy to elicit a localized MICA/B tissue expression profile to minimize potential systematic effects on healthy tissues is also being investigated by ourselves and others [37]. Obviously, the use of non-cell targeting therapeutic modalities to induce surface MICA/B expression would elevate the risk of on-target off-tumor/diseased cell toxicities and will require careful patient and indication selection if utilized in combination of 3MICA/B CAR in a clinical setting.

Additional therapeutic opportunities to employ 3MICA/B CAR cell-based therapies exist in the context of age-related senescence, fibrosis, and prophylactic elimination of pre-cancerous cells.

Cellular senescence has been implicated in an increasing number of chronic diseases, such as obesity, cardiovascular disease, diabetes, tissue dysfunction, and age-related pathologies [39,40].

Senescent cells are characterized by permanent cell cycle arrest, metabolic dysfunction, and their ability to resist immune detection and elimination, however the precise mechanisms of why and how senescent cells accumulate during ageing and persist at sites of age-related pathologies remains unknown [41]. Age related immune dysfunction may contribute to incomplete elimination of senescent cells, and the secretion of pro-inflammatory mediators, as part of a senescence-associated secretory phenotype (SASP), is thought to be a key driver of fibrosis which can accelerate the spread of cellular senescence and establish immunosuppressive niches that initiate tumor genesis [39,42]. Despite the development of senolytics, drugs that 'selectively' remove senescent cells through inducing apoptosis, that have proven age-reversing properties and can eliminate senescent cells in small animal models, enduring safety concerns surrounding systemic 'off target' toxicities have promoted the interest in investigating the application of targeted cell-based therapeutic modalities in models of senescence. Predictably, identifying specific senescent cell associated antigens, that are not expressed on surrounding healthy tissues, presents similar challenges discussed above in the context of solid tumors. CAR T cells targeting the urokinase-type plasminogen activator receptor (uPAR), a GPI-anchored receptor implicated in degrading extracellular matrix through focusing urokinase (uPA) proteolytic activity, have demonstrated selective elimination of uPAR expressing tumor cells and alleviated liver fibrosis in pre-clinical animal models [43,44]. NKG2D ligands, including MICA/B have also been detected on the surface of senescent cells, however senescent cells, like tumor cells, can also evade NKG2D mediated immune surveillance [45]. Furthermore, NKG2D ligands are detected at higher levels in senescent skin fibroblasts of older patients relative to younger patients [46].

These observations were recently supported following the demonstration that NKG2D ligand expressing senescent cells in naturally ageing non-human primates can be selectively and safely eliminated by the adoptive transfer of NKG2D CAR T cells [45,47]. Intriguingly this study also demonstrated that a murine NKG2D CAR T cell reversed the occurrence of senescence-associated phenotypes in aged mice, a hallmark of many age-related senescence associated pathologies and a prerequisite for stopping progressive fibrotic pathologies. Collectively these studies suggest that senescent cells expressing stress ligand family members in aged humans could potentially be targeted using stress ligand specific CAR T cells to reverse disease phenotype. Ongoing clinical trials utilizing NKG2D-based CAR T cells for the treatment of cancer continue to show favorable safety and tolerability profiles in patients [48]. Taking into account our data highlighting the superior functionality of 3MICA/B CAR relative to NKG2D or NKG2D CAR recognition of the MICA/B stress ligand family, the idea of employing 3MICA/B CAR expressing iPSC derived T cells to eliminate pre-cancerous senescent cells, or slow/reverse progressive fibrotic age-related pathologies is a tantalizing thought.

In summary, targeting an inducible family of stress ligands in a manner that mitigates multiple mechanisms of tumor evasion can provide a novel approach to discriminate malignant cells from surrounding healthy tissue. The proof-of-concept data that we presented using 3MICA/B CAR represents a universally applicable cell therapy concept amenable to clinical application against an array of pathological indications. As we continue to develop and understand the 3MICA/B CAR concept, it is evident that this

pioneering technology, combined with iPSC-derived effector immune cells, has the potential to redefine cancer therapy and extend its reach to combat other diseases where immune recognition of stress ligands plays a pivotal role. It appears that 3MICA/B CAR has not only added to the ongoing battle against cancer but also opened a new chapter for the possibility of therapeutic and prophylactic synthetic cellular immune surveillance.

References

1. Hanahan D. Hallmarks of Cancer: New Dimensions. *Cancer Discov* 2022;12:31-46.
2. Cappell KM, Kochenderfer JN. Long-term outcomes following CAR T cell therapy: what we know so far. *Nat Rev Clin Oncol* 2023;1-13.
3. Schuster SJ, Bishop MR, Tam CS, Waller EK, Borchmann P, McGuirk JP, et al. Tisagenlecleucel in Adult Relapsed or Refractory Diffuse Large B-Cell Lymphoma. *New Engl J Med* 2019;380:45-56.
4. Rafiq S, Hackett CS, Brentjens RJ. Engineering strategies to overcome the current roadblocks in CART cell therapy. *Nat Rev Clin Oncol* 2020;17:147-67.
5. Marofi F, Motavalli R, Safonov VA, Thangavelu L, Yumashev AV, Alexander M, et al. CAR T cells in solid tumors: challenges and opportunities. *Stem Cell Res Ther* 2021;12:81.
6. D'Aloia MM, Zizzari IG, Sacchetti B, Pierelli L, Alimandi M. CAR-T cells: the long and winding road to solid tumors. *Cell Death Dis* 2018;9:282.
7. Goulding J, Yeh W-I, Hancock B, Blum R, Xu T, Yang B-H, et al. A chimeric antigen receptor uniquely recognizing MICA/B stress proteins provides an effective approach to target solid tumors. *Med* 2023;4:457-477.e8.
8. Oekelen OV, Aleman A, Upadhyaya B, Schnakenberg S, Madduri D, Gavane S, et al. Neurocognitive and hypokinetic movement disorder with features of parkinsonism after BCMA- targeting CAR-T cell therapy. *Nat Med* 2021;27:2099-103.
9. Hirayama AV, Turtle CJ. Toxicities of CD19 CAR-T cell immunotherapy. *Am J Hematol.* 2019;94:542-9.
10. Morgan RA, Yang JC, Kitano M, Dudley ME, Laurencot CM, Rosenberg SA. Case Report of a Serious Adverse Event Following the Administration of T Cells Transduced With a Chimeric Antigen Receptor Recognizing ERBB2. *Mol Ther* 2010;18:843-51.
11. Bahram S, Bresnahan M, Geraghty DE, Spies T. A second lineage of mammalian major histocompatibility complex class I genes. *Proc National Acad Sci* 1994;91:6259-63.
12. Raulet DH, Gasser S, Gowen BG, Deng W, Jung H. Regulation of Ligands for the NKG2D Activating Receptor. *Immunology* 2013;31:413-41.
13. Fuertes MB, Domaica CI, Zvirner NW. Leveraging NKG2D Ligands in Immuno- Oncology. *Front Immunol* 2021;12:713158.
14. Vantourout P, Willcox C, Turner A, Swanson CM, Haque Y, Sobolev O, et al. Immunological Visibility: Posttranscriptional Regulation of Human NKG2D Ligands by the EGF Receptor Pathway. *Sci Transl Med* 2014;6:231ra49.
15. Lanier LL. NKG2D Receptor and Its Ligands in Host Defense. *Cancer Immunol Res* 2015;3:575-82.
16. Groh V, Rhinehart R, Randolph-Habecker J, Topp MS, Riddell SR, Spies T. Costimulation of CD8 α T cells by NKG2D via engagement by MIC induced on virus-infected cells. *Nat Immunol* 2001;2:255-60.
17. Wang X, Lundgren AD, Singh P, Goodlett DR, Plymate SR, Wu JD. An six-amino acid motif in the alpha3 domain of MICA is the cancer therapeutic target to inhibit shedding. *Biochem Bioph Res Co* 2009;387:476-81.
18. Zarrabi K, Dufour A, Li J, Kescu C, Pulkoski-Gross A, Zhi J, et al. Inhibition of Matrix Metalloproteinase 14 (MMP-14)-mediated Cancer Cell Migration. *J Biol Chem* 2011;286:33167-77.
19. Gooz M. ADAM-17: the enzyme that does it all. *Crit Rev Biochem Mol* 2010;45:146-69.
20. Groh V, Wu J, Yee C, Spies T. Tumour-derived soluble MIC ligands impair expression of NKG2D and T-cell activation. *Nature* 2002;419:734-8.
21. Mincheva-Nilsson L, Baranov V. Cancer exosomes and NKG2D receptor-ligand interactions: Impairing NKG2D-mediated cytotoxicity and anti-tumour immune surveillance. *Semin Cancer Biol* 2014;28:24-30.
22. Chitadze G, Bhat J, Lettau M, Janssen O, Kabelitz D. Generation of Soluble NKG2D Ligands: Proteolytic Cleavage, Exosome Secretion and Functional Implications. *Scand J Immunol* 2013;78:120-9.
23. Andrade LF de, Tay RE, Pan D, Luoma AM, Ito Y, Badrinath S, et al. Antibody-mediated inhibition of MICA and MICB shedding promotes NK cell-driven tumor immunity. *Science* 2018;359:1537-42.
24. Lombana TN, Matsumoto ML, III JB, Berkley AM, Toy E, Cook R, et al. High-resolution glycosylation site-engineering method identifies MICA epitope critical for shedding inhibition activity of anti-MICA antibodies. *Mabs* 2018;11:75-93.
25. Depil S, Duchateau P, Grupp SA, Mufti G, Poirot L. 'Off-the-shelf' allogeneic CAR T cells: development and challenges. *Nat Rev Drug Discov* 2020;19:185-99.
26. Ciurea SO, Schafer JR, Bassett R, Denman CJ, Cao K, Willis D, et al. Phase 1 clinical trial using mBL21 ex vivo-expanded donor-derived NK cells after haploidentical transplantation. *Blood* 2017;130:1857-68.
27. Romee R, Rosario M, Berrien-Elliott MM, Wagner JA, Jewell BA, Schappe T, et al. Cytokine-induced memory-like natural killer cells exhibit enhanced responses against myeloid leukemia. *Sci Transl Med* 2016;8:357ra123.
28. Valamehr B, Robinson M, Abujarour R, Rezner B, Vranceanu F, Le T, et al. Platform for Induction and Maintenance of Transgene-free hiPSCs Resembling Ground State Pluripotent Stem Cells. *Stem Cell Rep* 2014;2:366-81.
29. Li Y, Hermanson DL, Moriarity BS, Kaufman DS. Human iPSC-Derived Natural Killer Cells Engineered with Chimeric Antigen Receptors Enhance Anti-tumor Activity. *Cell Stem Cell* 2018;23:181-192.e5.
30. FDA Investigating CAR-Related T-cell Malignancies. *Cancer Discov* 2023;OF1-OF1.
31. Baugh R, Khalique H, Seymour LW. Convergent Evolution by Cancer and Viruses in Evading the NKG2D Immune Response. *Cancers* 2020;12:3827.
32. Nolting A, Dugast A-S, Rihn S, Luteijn R, Carrington MF, Kane K, et al. MHC class I chain-related protein A shedding in chronic HIV-1 infection is associated with profound NK cell dysfunction. *Virology* 2010;406:12-20.
33. Matusali G, Tchidjou HK, Pontrelli G, Bernardi S, D'Ettorre G, Vullo V, et al. Soluble ligands for the NKG2D receptor are released during HIV-1 infection and impair NKG2D expression and cytotoxicity of NK cells. *Faseb J* 2013;27:2440-50.

34. Tong HV, Toan NL, Song LH, Bock C -T., Kreamsner PG, Velavan TP. Hepatitis B virus- induced hepatocellular carcinoma: functional roles of MICA variants. *J Viral Hepat* 2013;20:687-98.
35. Estes G, Luzón E, Sarmiento E, Gómez-Caro R, Steinle A, Murphy G, et al. Altered MicroRNA Expression after Infection with Human Cytomegalovirus Leads to TIMP3 Downregulation and Increased Shedding of Metalloprotease Substrates, Including MICA. *J Immunol* 2014;193:1344-52.
36. Varchetta S, Mele D, Oliviero B, Mantovani S, Ludovisi S, Cerino A, et al. Unique immunological profile in patients with COVID-19. *Cell Mol Immunol* 2021;18:604-12.
37. Tsai C-L, Yang P-S, Hsu F-M, Cheng A-L, Yu W-N, Cheng JC-H. Topoisomerase I Inhibition Radiosensitizing Hepatocellular Carcinoma by RNF144A-mediated DNA-PKcs Ubiquitination and Natural Killer Cell Cytotoxicity. *J Clin Transl Hepatol* 2023;11:614-25.
38. Fuertes MB, Domaica CI, Zvirner NW. Leveraging NKG2D Ligands in Immuno- Oncology. *Front Immunol* 2021;12:713158.
39. He S, Sharpless NE. Senescence in Health and Disease. *Cell* 2017;169:1000-11.
40. Riessland M. Cellular Senescence in Health, Disease and Aging: Blessing or Curse? *Life* 2021;11:541.
41. Sharpless NE, Sherr CJ. Forging a signature of in vivo senescence. *Nat Rev Cancer* 2015;15:397-408.
42. Chatterjee D, Chakrabarti O. Role of stress granules in modulating senescence and promoting cancer progression: Special emphasis on glioma. *Int J Cancer* 2022;150:551-61.
43. Amor C, Feucht J, Leibold J, Ho Y-J, Zhu C, Alonso-Curbelo D, et al. Senolytic CAR T cells reverse senescence-associated pathologies. *Nature* 2020;583:127-32.
44. Mauro CD, Pesapane A, Formisano L, Rosa R, D'Amato V, Ciciola P, et al. Urokinase-type plasminogen activator receptor (uPAR) expression enhances invasion and metastasis in RAS mutated tumors. *Sci Rep* 2017;7:9388.
45. Pereira BI, Devine OP, Vukmanovic-Stejic M, Chambers ES, Subramanian P, Patel N, et al. Senescent cells evade immune clearance via HLA-E-mediated NK and CD8+ T cell inhibition. *Nat Commun* 2019;10:2387.
46. Hasegawa T, Oka T, Son HG, Oliver-García VS, Azin M, Eisenhaure TM, et al. Cytotoxic CD4+ T cells eliminate senescent cells by targeting cytomegalovirus antigen. *Cell* 2023;186:1417-1431.e20.
47. Yang D, Sun B, Li S, Wei W, Liu X, Cui X, et al. NKG2D-CAR T cells eliminate senescent cells in aged mice and nonhuman primates. *Sci Transl Med* 2023;15:eadd1951.
48. Curio S, Jonsson G, Marinović S. A summary of current NKG2D-based CAR clinical trials. *Immunother Adv* 2021;1:ltab018.