

## Tumor-infiltrating lymphocytes for melanoma immunotherapy

Oncological routine has incorporated the regular use of monoclonal antibodies targeting checkpoints of T cell function such as ipilimumab, which blocks cytotoxic T-lymphocyte-associated protein 4 (CTLA-4), or pembrolizumab and nivolumab, which both inhibit programmed cell death 1 (PD-1). Such immune checkpoint inhibitors have substantially improved recurrence-free survival (RFS) in patients with advanced stage melanoma.<sup>1</sup> Moreover, the development of molecularly targeted therapies such as the BRAF inhibitor dabrafenib and the mitogen-activated protein inhibitor trametinib has extended treatment options for melanoma patients with advanced metastatic disease.<sup>2</sup>

Based on its survival benefits, ipilimumab was the first immune checkpoint blocker approved for the treatment of advanced melanoma.<sup>3</sup> However, high-grade immune-related adverse effects are observed in 23% of patients.<sup>4</sup> Moreover, in a large clinical trial enrolling 834 advanced melanoma patients, pembrolizumab was shown to cause prolonged progression-free and overall survival with less high-grade toxicity than ipilimumab.<sup>4</sup> For this reason, currently, ipilimumab is used as second-line treatment in patients with metastatic melanoma, whereas PD-1 inhibitors alone or in combination with ipilimumab have been moved to first-line, inducing responses in 45% or 58% of patients, respectively. Nonetheless, double immune checkpoint blockade targeting PD-1 plus CTLA-4 is associated with a high incidence of severe adverse effects and is currently recommended primarily for patients with poor prognostic factors.

An activating mutation in the B-Raf proto-oncogene serine/threonine kinase (BRAF) gene (*BRAF*<sup>V600E</sup>) is present in more than 50% of melanoma patients. Thus, combination of dabrafenib plus trametinib is yet another treatment option for melanoma harboring such mutation. Although this therapy is associated with a high initial response rate, most patients develop resistance over time.<sup>2</sup> Further combination approaches involving BRAF inhibition plus immune checkpoint blockade as well as the use of novel immune checkpoint blocking antibodies targeting lymphocyte-activation gene 3 (*LAG-3*) LAG3 have shown promising response rates. Thus, combination of anti-PD-1 and anti-LAG3 monoclonal antibodies has been associated with objective responses in 16% of patients with refractory disease but follow-up data are still missing.<sup>5</sup> Nevertheless, as it stands the efficacy of both immune checkpoint inhibition or targeted approaches for patients with advanced stage melanoma remains limited and despite optimal treatment about half of the patients will eventually die from the disease.

Pioneered by Steven Rosenberg and colleagues, adoptive cell therapy with tumor-infiltrating lymphocytes (TILs) has been

developed. This approach necessitates the *ex vivo* outgrowth and expansion of TILs, followed by their intravenous reinfusion into patients that have undergone preparative lymphodepletion by chemotherapy. The administration of recombinant human interleukin-2 (rhIL2) can be used to enhance the *in vivo* expansion of TILs. TIL therapy proofed safety and evidenced efficacy in phase 1–2 trials with 20% durable complete remission in patients with advanced stage melanoma and achieved objective responses in 36% of patients with advanced melanoma, including in anti-PD-1 refractory disease.<sup>6–9</sup>

Thus far, a direct comparison of TILs with standard immunotherapy has been missing. In a recent study published in the *The new England Journal of Medicine*, Rohaan et al. presented the results of a multicenter, open-label, randomized phase 3 clinical trial comparing ipilimumab with TIL therapy as first- or second-line treatment in patients with advanced unresectable stage IIIC or IV melanoma. A total of 168 patients, among whom 86% presented with disease refractory to anti-PD-1, were randomly assigned to TIL therapy or ipilimumab treatment.<sup>5</sup>

The median progression-free survival was 7.2 months (95% confidence interval [CI], 4.2–13.1) for patients receiving TIL therapy and 3.1 months (95% CI, 3.0–4.3) for those treated with ipilimumab. Objective response rate was 49% (95% CI, 38–60) and 21% (95% CI, 13–32) and median overall survival 25.8 months (95% CI, 18.2 to not reached) and 18.9 months (95% CI, 13.8–32.6) in TIL *versus* ipilimumab therapy, respectively. Of note, all patients who received TIL therapy experienced mainly chemotherapy-related high-grade adverse effects such as myelosuppression.<sup>5</sup> Altogether, the trial led to the conclusions that non-cryopreserved, autologous TILs prepared from resected melanoma metastases by standard rapid expansion protocol are more efficient than ipilimumab in extending the progression-free survival of advanced melanoma patients.<sup>5,10</sup>

As a caveat, the complex logistics of cell therapies, as well as the limited availability of bioactive TILs, both may limit the general applicability of TIL therapy. Moreover, treatment-related adverse effects are frequent, requiring careful monitoring and management by the treating oncologist. Irrespective of these disadvantages, the results by Rohaan *et al.*<sup>5</sup> underline the feasibility and utility of personalized autologous TIL therapy for advanced melanoma patients in specialized centers. Future studies must evaluate the optimization of TIL preparation as well as the combination of TIL therapy with targeted treatments or immunotherapies, including checkpoint inhibitors targeting CTLA-4 and the PD-1/PD-L1 interaction. Moreover, it will be important to further develop TIL-based therapies to other, more frequent malignancies than melanoma.<sup>11–13</sup>

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## Disclosure statement

OK is a cofounder of Samsara Therapeutics. GK has been holding research contracts with Daiichi Sankyo, Eleor, Kaleido, Lytix Pharma, PharmaMar, Osasuna Therapeutics, Samsara Therapeutics, Sanofi, Tollys, and Vascage. GK has been consulting for Reithera. GK is on the Board of Directors of the Bristol Myers Squibb Foundation France. GK is a scientific co-founder of everImmune, Osasuna Therapeutics, Samsara Therapeutics and Therafast Bio. GK is in the scientific advisory boards of Hevolution, Institut Servier and Longevity Vision Funds. GK is the inventor of patents covering therapeutic targeting of aging, cancer, cystic fibrosis and metabolic disorders. GK's brother, Romano Kroemer, was an employee of Sanofi and now consults for Boehringer-Ingelheim. LZ has held research contracts with Glaxo Smyth Kline, Incyte, Lytix, Kaleido, Innovate Pharma, Daiichi Sankyo, Pilege, Merus, Transgene, 9 m, Tusk and Roche, was on the on the Board of Directors of Transgene, is a cofounder of everImmune, and holds patents covering the treatment of cancer and the therapeutic manipulation of the microbiota. The funders had no role in the design of the study; in the writing of the manuscript, or in the decision to publish the results.

## Data availability statement

All data leading to the conclusion presented in this paper are annexed.

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