Clinical Cancer Research

Anti-CD137 and PD-1/PD-L1 Antibodies En Route toward Clinical Synergy

Elisabeth Pérez-Ruiz[1,2,3], Iñaki Etxeberria[1,2], Maria E. Rodriguez-Ruiz[1,2,4,5], and Ignacio Melero[1,2,4,5]

T-cell costimulation and coinhibition can be respectively exploited by blocking and agonist mAbs. Both strategies can be synergistically combined in mouse models. Early clinical results from combinations of anti–PD-1 mAbs in conjunction with agonist anti-CD137 (4-1BB) mAbs show excellent safety and promising efficacy. *Clin Cancer Res; 23(18); 5326–8. ©2017 AACR.*

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In this issue of *Clinical Cancer Research (CCR)*, Tolcher and colleagues report on the dose-escalation combination of the anti-CD137 agonist mAb utomilumab with standard 2 mg/kg every-3-week doses of the anti–PD-1–blocking mAb pembrolizumab, as tested in a phase Ib study for patients with a variety of solid tumors (1).

CD137 (4-1BB)-based cancer immunotherapy started with experiments in transplanted mouse tumors that showed CD8 T-cell mediated complete rejections of established tumors upon administration of agonist anti-CD137 mAbs, irrespective of the ability of such antibodies to inhibit binding of CD137 to its natural ligand (CD137L or 4-1BBL; ref. 2). CD137 (4-1BB, TNFRSF9) is a surface glycoprotein that belongs to the TNFR family and was discovered by Kwon and colleagues (3) as a T-cell costimulatory molecule that, importantly, was expressed on antigen-primed T lymphocytes, but not on their resting counterparts. It is also functionally expressed on activated natural killer (NK) cells, where its stimulation enhances antibody-dependent cellular cytotoxicity, and on dendritic cells and other leucocytes, where its function is less well studied.

Costimulation via CD137 upon binding to agonist mAb protects T lymphocytes from cell death, promotes memory differentiation, enhances effector functions, and favors clonal expansion (4). Clinical exploitation of such functions was started by scientists at Bristol-Myers Squibb with the development of urelumab, a non–ligand-blocking, antihuman CD137 mAb of the IgG4 subclass (5). Consistently with an excellent preclinical safety record, dose-escalation clinical trials revealed no unacceptable toxicity of repeated doses of up to 5 mg/kg, and some clinical objective responses both in melanoma and non-melanoma patients were observed (5). However, in the following phase Ib/II trials, serious liver inflammation was found in an important fraction of treated patients, thus precluding further clinical development at such dose levels (5).

Liver inflammation with potent CD137 agonist mAbs can be modeled in mice showing polyclonal periportal infiltration of T cells dominated by a $CD8^+$ component, even if such side effects are mild in mice (6). Failure of cynomolgus macaques experiments to preclinically identify such a problem is probably related to the lower affinity of urelumab for the nonhuman primate target molecule. As a result of these safety problems, urelumab was dose reescalated to be declared safe at a much lower dose level, comprising a flat dose regimen of 8 mg/kg given with 3-week intervals that questionably would provide enough receptor triggering (7). In a parallel effort, Pfizer developed utomilumab, an anti-CD137 mAb of the IgG2 class, which prevents CD137–CD137L binding and reportedly acts as a CD137 agonist in several functional *in vitro* and *in vivo* assays. It also shows an excellent preclinical safety profile based on experiments including cynomolgus macaques (8).

In contrast to urelumab, utomilumab showed no dose-limiting toxicities in regimens with doses up to 10 mg/kg and showed early evidence of clinical activity in dose-escalation studies, including patients with refractory non-Hodgkin lymphoma who were cotreated with rituximab (9, 10). In our side-by-side comparison experiments, both utomilumab and urelumab exert agonist activity in terms of CD137 toward NF-κB activation signaling and ensuing T-cell costimulation, even though the agonist activity of urelumab on the receptor is much more marked.

Cancer immunotherapy is at the crest of a revolution spawned by the efficacy of results from PD-1 and PD-L1 blockade across a broad spectrum of malignant diseases when given as a single-agent therapy or in combination. In mouse models that resist both treatment with anti–PD-1–blocking agents and CD137 agonists, the combination of these two immunotherapy agents exerts powerful synergistic effects (7, 11, 12) that correlate with stronger T-cell responses against tumor antigens. In part, the synergy comes from the fact that a fraction of tumor-infiltrating T lymphocytes coexpress on their surface both PD-1 and CD137, giving rise to a combined effect on individual T cells, which is very prominent even for very poorly immunogenic mouse cancer models (13). Interestingly, CD137 and PD-1 seem to be expressed mainly by CD8 T lymphocytes recognizing tumor neoantigens (14).

[1]Program of Immunology and Immunotherapy, Center for Applied Medical Research (CIMA), Pamplona, Spain. [2]Navarra Institute for Health Research (IDISNA), Pamplona, Spain. [3]Servicio de Oncología Médica, Hospital Costa del Sol, Marbella, Spain. [4]Servicio de Inmunología e Inmunoterapia, Clínica Universidad de Navarra, Pamplona, Spain. [5]CIBERONC (Centro de Investigación Biomédica en red de Oncología), Madrid, Spain.

Corresponding Author: Ignacio Melero, University of Navarra and Instituto de Investigacion Sanitaria de Navarra (IDISNA), Avenida Pio XII, 55, Pamplona, Navarra 31008, Spain. E-mail: imelero@unav.es

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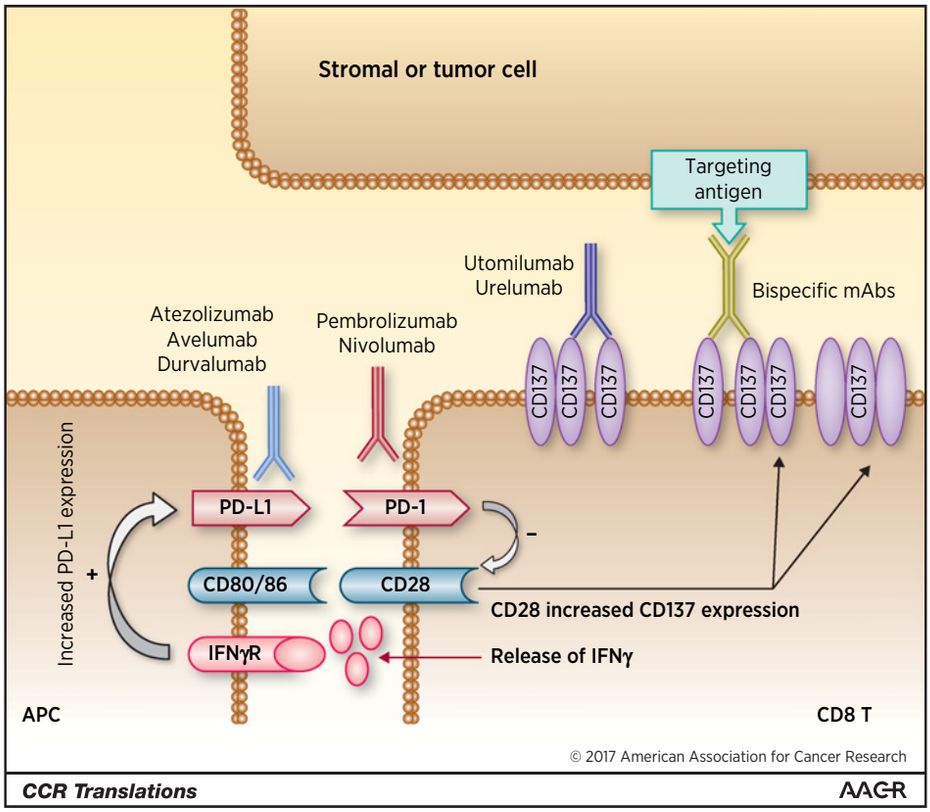

Figure 1.
Schematic representation of mechanisms leading to the synergy of anti-CD137 and anti–PD-1 immunostimulatory mAbs. Conceivably, the same tumor-reactive T cells are released from the inhibitory influence of PD-1 on CD28 and T-cell receptor (TCR) signaling while receiving active costimulation via CD137 to keep them alive and fitter to perform their antitumor function. Pharmacodynamic interactions occur between both mAb-targeted molecules as for example: (i) PD-1 blockade and CD28 signaling lead to increased expression of CD137 and (ii) CD137-enhanced INFγ production would lead to more intense PD-L1 expression in the tumor microenvironment. As depicted, targeted forms of CD137 agonists to the tumor microenvironment have been designed, including bispecific antibodies and other biomolecule formats. These smarter, local CD137 agonists will be safer and more potent. Such agents are also to be combined with PD-1 blockade. APC, antigen-presenting cell.

Prompted by their preclinical efficacy, both urelumab and utomilumab are currently being tested for safety and efficacy in combination with the anti–PD-1 mAb nivolumab or utomilumab. To date, no publications reporting results from clinical trials combining urelumab and nivolumab are available. At the 2016 SICT meeting (15), information was presented indicating the excellent tolerability of urelumab given at an 8-mg flat dose every 3 weeks, along with the standard doses of nivolumab. The most striking efficacy result was a high overall response rate of up to 47% in metastatic melanoma cases, with less than 1% of tumor cells expressing PD-L1. Even if some of these early results could be explained by nivolumab efficacy alone, the results in cases without PD-L1 expression warrant further development of the combination. However, it must be kept in mind that the doses of urelumab in the trials are extremely low. Thus, much of the efficacy in this combination scheme might be jeopardized by low dosing of the CD137 agonist component.

In this issue of *CCR*, the first phase I results of the utomilumab plus pembrolizumab combination are released. Again, safety results upon utomilumab dose escalation in combination with pembrolizumab are excellent and consistent with the side effects expected for pembrolizumab alone. It is too early to say whether additional clinical benefit exists, but complete responses in a case of a small-cell lung cancer and in a patient with renal cell carcinoma hold much hope. More intriguingly, a patient with anaplastic thyroid cancer experienced a durable, deep partial response. Unfortunately, insufficient immunobiology correlative studies were performed in connection with the trial, and it is not possible to conclude whether utomilumab plus pembrolizumab is modifying cancer immunity differently than pembrolizumab as a single agent.

Pharmaceutical and biotechnology industries are very actively pursuing ever smarter strategies to confine CD137 costimulation to the tumor microenvironment and draining lymph nodes. These approaches should bypass liver toxicity. Strategies to exploit this costimulatory pathway are to be deployed together with the PD-1 blockade backbone given the clinical efficacy of PD-1 monotherapy and preclinical evidence for therapeutic synergy in the anti-PD-1 plus anti-CD137 mAb combinations. Patients predicted to benefit from this combined approach are (i) those suffering from a disease not amenable to PD-1/PD-L1 blockade, (ii) those with tumors failing to show PD-L1 staining in the biopsy, and (iii) those who have already progressed to PD-1/PD-L1–blocking agents. Such cases in our opinion constitute the precise target population to demonstrate efficacy of the combination.

Biomarker discovery must progress in parallel. To begin, coexpression of CD137 and PD-1 in the pretreatment biopsy needs to be explored. Other parameters may narrow down who will ultimately benefit the most from this treatment. Of note, in patients treated with the combination, pharmacodynamic interactions are postulated to take place, including increases in CD137 expression upon PD-1 blockade (13) and increases in PD-L1 expression due to CD137-costimulated INFγ production (Fig. 1).

Disclosure of Potential Conflicts of Interest

I. Melero is a consultant/advisory board member for Alligator, AstraZeneca, Bayer, Bristol-Myers Squibb, Incyte, Lilly, and Roche and reports receiving commercial research grants from Bioncotech, Bristol-Myers Squibb, and Roche. No potential conflicts of interest were disclosed by the other authors.

Authors' Contributions

Conception and design: E. Pérez-Ruiz, M.E. Rodriguez-Ruiz, I. Melero
Development of methodology: E. Pérez-Ruiz
Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): E. Pérez-Ruiz
Writing, review, and/or revision of the manuscript: E. Pérez-Ruiz, I. Etxeberria, M.E. Rodriguez-Ruiz, I. Melero
Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): E. Pérez-Ruiz, I. Melero

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References

1. Tolcher AW, Sznol M, Hu-Lieskovan S, Papadopoulos KP, Patnaik A, Rasco DW, et al. Phase Ib study of utomilumab (PF-05082566), a 4-1BB/CD137 agonist, in combination with pembrolizumab (MK-3475) in patients with advanced solid tumors. Clin Cancer Res 2017;23:5349–57.
2. Melero I, Shuford WW, Newby SA, Aruffo A, Ledbetter JA, Hellström KE, et al. Monoclonal antibodies against the 4-1BB T-cell activation molecule eradicate established tumors. Nat Med 1997;3:682–5.
3. Vinay DS, Kwon BS. 4-1BB signaling beyond T cells. Cell Mol Immunol 2011;8:281–4.
4. Melero I, Hirschhorn-Cymerman D, Morales-Kastresana A, Sanmamed MF, Wolchok JD. Agonist antibodies to TNFR molecules that costimulate T and NK cells. Clin Cancer Res 2013;19:1044–53.
5. Segal NH, Logan RF, Hodi FS, McDermott D, Melero I, Hamid O, et al. Results from an integrated safety analysis of urelumab, an agonist anti-CD137 monoclonal antibody. Clin Cancer Res 2017;23:1929–36.
6. Dubrot J, Milheiro F, Alfaro C, Palazón A, Martinez-Forero I, Perez-Gracia JL, et al. Treatment with anti-CD137 mAbs causes intense accumulations of liver T cells without selective antitumor immunotherapeutic effects in this organ. Cancer Immunol Immunother 2010;59:1223–33.
7. Ascierto PA, Simeone E, Sznol M, Fu Y-X, Melero I. Clinical experiences with anti-CD137 and anti-PD1 therapeutic antibodies. Semin Oncol 2010;37:508–16.
8. Fisher TS, Kamperschroer C, Oliphant T, Love VA, Lira PD, Doyonnas R, et al. Targeting of 4-1BB by monoclonal antibody PF-05082566 enhances T-cell function and promotes anti-tumor activity. Cancer Immunol Immunother 2012;61:1721–33.
9. Segal NH, Gopal AK, Bhatia S, Kohrt HE, Levy R, Pishvaian MJ, et al. A phase 1 study of PF-05082566 (anti-4-1BB) in patients with advanced cancer. J Clin Oncol 32, 2014 (suppl; abstr 3007).
10. Gopal AK, Bartlett NL, Levy R, Houot R, Smith SD, Segal NH, et al. A phase I study of PF-05082566 (anti-4-1BB) + rituximab in patients with CD20 + NHL. J Clin Oncol 33, 2015 (suppl; abstr 3004).
11. Wei H, Zhao L, Hellstrom I, Hellstrom KE, Guo Y. Dual targeting of CD137 co-stimulatoy and PD-1 co-inhibitory molecules for ovarian cancer immunotherapy. Oncoimmunology 2014;3:e28248.
12. Palazon A, Martínez-Forero I, Teijeira A, Morales-Kastresana A, Alfaro C, Sanmamed MF, et al. The HIF-1α hypoxia response in tumor-infiltrating T lymphocytes induces functional CD137 (4-1BB) for immunotherapy. Cancer Discov 2012;2:608–23.
13. Morales-Kastresana A, Sanmamed MF, Rodriguez I, Palazon A, Martinez-Forero I, Labiano S, et al. Combined immunostimulatory monoclonal antibodies extend survival in an aggressive transgenic hepatocellular carcinoma mouse model. Clin Cancer Res 2013;19:6151–62.
14. Parkhurst M, Gros A, Pasetto A, Prickett T, Crystal JS, Robbins P, et al. Isolation of T-cell receptors specifically reactive with mutated tumor-associated antigens from tumor-infiltrating lymphocytes based on CD137 expression. Clin Cancer Res 2017;23:2491–505.
15. Masserelli E, Segal NH, Ribrag V, Melero I, Gangadhar TC, Urba W, et al. Clinical safety and efficacy assessment of the CD137 agonist urelumab alone and in combination with nivolumab in patients with hematologic and solid tumor malignancies. J Immunother Cancer 2016;4(Suppl 1):O7.