MONOCLONAL ANTIBODIES IN IMMUNODIAGNOSIS AND IMMUNOTHERAPY
Volume 41, Number 1, 2022

DOI: 10.1089/mab.2021.0056

TrMab-6 Exerts Antitumor Activity in Mouse Xenograft Models of Breast Cancers

Tomohiro Tanaka,[1] Tomokazu Ohishi,[2] Masaki Saito,[3] Manabu Kawada,[2] Mika K. Kaneko,[1] and Yukinari Kato[1,3]

Trophoblast cell surface antigen 2 (TROP2) has been reported to be overexpressed in many cancers, and is involved in cancer cell proliferation, invasion, and metastasis. We previously developed a highly sensitive anti-TROP2 monoclonal antibody (mAb) (clone TrMab-6; mouse IgG_{2b}, kappa) using a Cell-Based Immunization and Screening method. TrMab-6 is useful for investigations using flow cytometry, Western blotting, and immunohistochemistry and possesses antibody-dependent cellular cytotoxicity (ADCC) and complement-dependent cytotoxicity (CDC) against TROP2-expressing triple-negative breast cancer (TNBC) cell lines, such as MDA-MB-231 and MDA-MB-468. This study investigated whether TrMab-6 possesses *in vivo* antitumor activities via ADCC/CDC activities using mouse xenograft models of TNBC cell lines. *In vivo* experiments on MDA-MB-231 and MDA-MB-468 xenografts revealed that TrMab-6 significantly reduced tumor growth compared with normal mouse IgG treatment. The findings of this study suggest that TrMab-6 is a promising treatment option for TROP2-expressing TNBC.

Keywords: TROP2, monoclonal antibody, antitumor activity, triple-negative breast cancer

Introduction

Triple-negative breast cancer (TNBC) is an aggressive subtype of breast cancer. It is defined by the absence of estrogen receptor, progesterone receptor, and human epidermal growth factor receptor 2 (HER2) expressions.[1,2] TNBC formation is found in 10%–20% of breast cancers and frequently occurs in young and African American women.[3–5] Patients with TNBC have a worse prognosis than those with other breast cancers, with more than 30% developing metastatic diseases or recurring cancer.[6,7] Younger patients with TNBC have short disease-free survival than older patients with TNBC.[3] Furthermore, ~30% of patients with metastatic TNBC have been recorded to have at least one tumor metastasis to the brain.[8]

Surgery, radiotherapy, and platinum-based chemotherapy are primarily selected as the standard treatment for TNBC. Molecular targeted drugs, such as poly ADP-ribose polymerase inhibitors, phosphoinositide 3-kinase/Akt/mammalian target of rapamycin inhibitors, and cyclin-dependent kinase inhibitors, are also used as the basic treatment strategy for TNBC.[2,9,10] Immunotherapy is beginning to be introduced as a new treatment for breast cancers.[11,12]

The expression level of programmed cell death ligand 1 (PD-L1) is higher in TNBC than in other breast cancers.[13–15] PD-L1 expression is associated with malignancy and hormone receptor-negative breast cancer.[16] Programmed cell death 1/PD-L1-targeted antibodies, such as pembrolizumab and atezolizumab, have been actively tested on patients with breast cancer.[12,17,18]

Trophoblast cell surface antigen 2 (TROP2), a type I transmembrane glycoprotein, has been reported to be overexpressed in various solid tumors, including breast cancers. A higher expression of TROP2 is involved in tumor growth, invasion, and proliferation.[19–21] Particularly, TROP2 expression has been described to be upregulated in 80%–95% of

[1]Department of Antibody Drug Development, Tohoku University Graduate School of Medicine, Sendai, Japan.
[2]Microbial Chemistry Research Foundation, Institute of Microbial Chemistry (BIKAKEN), Numazu-shi, Japan.
[3]Department of Molecular Pharmacology, Tohoku University Graduate School of Medicine, Sendai, Japan.

TNBC.[22,23] TROP2 has been identified as one of the markers for epithelial cell adhesion molecule low/negative circulating tumor cells, which are derived from patients with metastatic TNBC.[24]

Sacituzumab govitecan (IMMU-132) is an antibody–drug conjugate (ADC) that combines an anti-TROP2 monoclonal antibody (mAb) with the irinotecan metabolite SN-38 as a payload.[25] The U.S. Food and Drug Administration (FDA) approved sacituzumab govitecan for treatment against metastatic TNBC and designated it a breakthrough therapy.[26,27] Another trial of this ADC was launched against some tumors, including hormone receptor-positive and HER2-negative breast cancers.[28]

We have previously developed an anti-TROP2 mAb (clone TrMab-6, mouse IgG_{2b}, kappa), which has been applied to various experiments, including flow cytometry, Western blotting, and immunohistochemistry.[29] TrMab-6 exerted *in vitro* antibody-dependent cellular cytotoxicity (ADCC) and complement-dependent cytotoxicity (CDC) activities or *in vivo* antitumor activities using mouse xenograft models of TROP2-overexpressed CHO-K1 (CHO/TROP2) or MCF7 breast cancer cells.[30] Furthermore, TrMab-6 exerted *in vitro* ADCC and CDC activities against TNBC cell lines, such as MDA-MB-231 and MDA-MB-468. However, whether TrMab-6 exerted *in vivo* antitumor activities, using mouse xenograft models of TNBC cell lines, has not been clarified.

In this study, we investigated whether TrMab-6 exerts *in vivo* antitumor activities using mouse xenograft models of TNBC cell lines, such as MDA-MB-231 and MDA-MB-468.

Materials and Methods

Cell lines

The breast cancer cell lines MDA-MB-231 and MDA-MB-468 were obtained from the American Type Culture Collection (Manassas, VA). MDA-MB-231 and MDA-MB-468 were cultured in Dulbecco's modified Eagle's medium (DMEM; Nacalai Tesque, Inc., Kyoto, Japan), supplemented with 10% heat-inactivated fetal bovine serum (Thermo Fisher Scientific, Inc., Waltham, MA), 100 U/mL of penicillin (Nacalai Tesque, Inc.), 100 μg/mL of streptomycin (Nacalai Tesque, Inc.), and 0.25 μg/mL of amphotericin B (Nacalai Tesque, Inc.), and incubated at 37°C in a humidified atmosphere containing 5% CO_2.

Primary antibodies

Purified mouse IgG (Cat. No. I8765) was purchased from Sigma-Aldrich (St. Louis, MO). TrMab-6 was purified using Protein G-Sepharose (GE Healthcare Biosciences, Pittsburgh, PA).

Antitumor activity of an anti-TROP2 mAb in a mouse xenograft model

A total of 32 five-week-old female BALB/c nude mice (mean weight, 15 ± 3 g) were purchased from Charles River Laboratories, Inc. (Kanagawa, Japan). The mice were divided into (i) MDA-MB-231-bearing mice and (ii) MDA-MB-468-bearing mice ($n = 16$ in each group). On day 7, each group was subdivided into two groups ($n = 8$ in each group) with equal mean tumor volume: a control mouse IgG-treated or TrMab-6-treated group. All animal experiments were performed following institutional guidelines and regulations to minimize animal suffering and distress in the laboratory. The Institutional Committee for experiments of the Institute of Microbial Chemistry (Permit No. 2021-015) approved the animal studies for antitumor activity.

The mice were maintained in a pathogen-free environment, on an 11-hour light/13-hour dark cycle at a temperature of $23°C \pm 2°C$ and humidity of $55\% \pm 5\%$, with food and water supplied *ad libitum* throughout the experiments. The mice were monitored for health and weight every 3 or 4 days. The experiments on mice were conducted after 4 weeks. Weight loss of >25% or tumor volume of >3000 mm^3 was identified as humane endpoints for euthanasia. At humane and experimental endpoints, the mice were euthanized by cervical dislocation, and death was verified by validating respiratory and cardiac arrest.

After an acclimation period of 1 week, these mice were used in experiments at 7 weeks of age (mean weight, 19 ± 2g). The cells (0.3 mL of 1.33×10^8 cells/mL in DMEM) were mixed with 0.5 mL of BD Matrigel Matrix Growth Factor Reduced (BD Biosciences, San Jose, CA). A total of 100 µL of this suspension (5×10^6 cells) was subcutaneously injected into the left flank of each animal. On day 7 postinoculation, 100 µg of TrMab-6 or control mouse IgG in 100 µL of phosphate-buffered saline (PBS) was intraperitoneally (i.p.) injected. Additional antibody inoculations were performed on days 14 and 21. All mice were euthanized by cervical dislocation 23 days after cell implantation, and tumor diameters and volumes were measured and recorded.

Statistical analysis

Data are expressed as mean ± standard error of the mean. Statistical analysis was conducted with ANOVA and Sidak's multiple comparison tests for tumor volume and mouse weight, and Welch's *t*-test for tumor weight. All calculations were performed using GraphPad Prism 7 (GraphPad Software, Inc.). $p < 0.05$ was considered statistically significant.

Results

Antitumor effect of TrMab-6 in mouse xenografts of a TROP2-expressing TNBC cell line (MDA-MB-231)

The tumor formation of 16 MDA-MB-231-bearing mice was observed on day 7 before the mice were divided into TrMab-6-treated and control groups. On days 7, 14, and 21, after MDA-MB-231 cell injections into the mice, either TrMab-6 (100 μg) or control mouse IgG (100 μg) was i.p. injected into the mice. Tumor volume was measured on days 7, 10, 14, 16, 21, and 23 after MDA-MB-231 cell injection. The TrMab-6-treated mice exhibited significantly less tumor growth on days 14 ($p < 0.01$), 16 ($p < 0.01$), 21 ($p < 0.01$), and 23 ($p < 0.01$) compared with the IgG-treated control mice (Fig. 1A).

On day 23, a 35% reduction in tumor volume was seen in the TrMab-6-treated mice (Fig. 1A). Tumors from the TrMab-6-treated mice weighed significantly less than tumors from the IgG-treated control mice on day 23 (50% reduction, $p < 0.01$; Fig. 1B). Figure 1C demonstrates each tumor. These results indicated that TrMab-6 reduced the growth of MDA-MB-231 xenografts, but did not contribute toward their total elimination. Total body weights did not significantly differ between the treatment and control groups (Fig. 1D). Figure 1E demonstrates the body appearance of the mice.

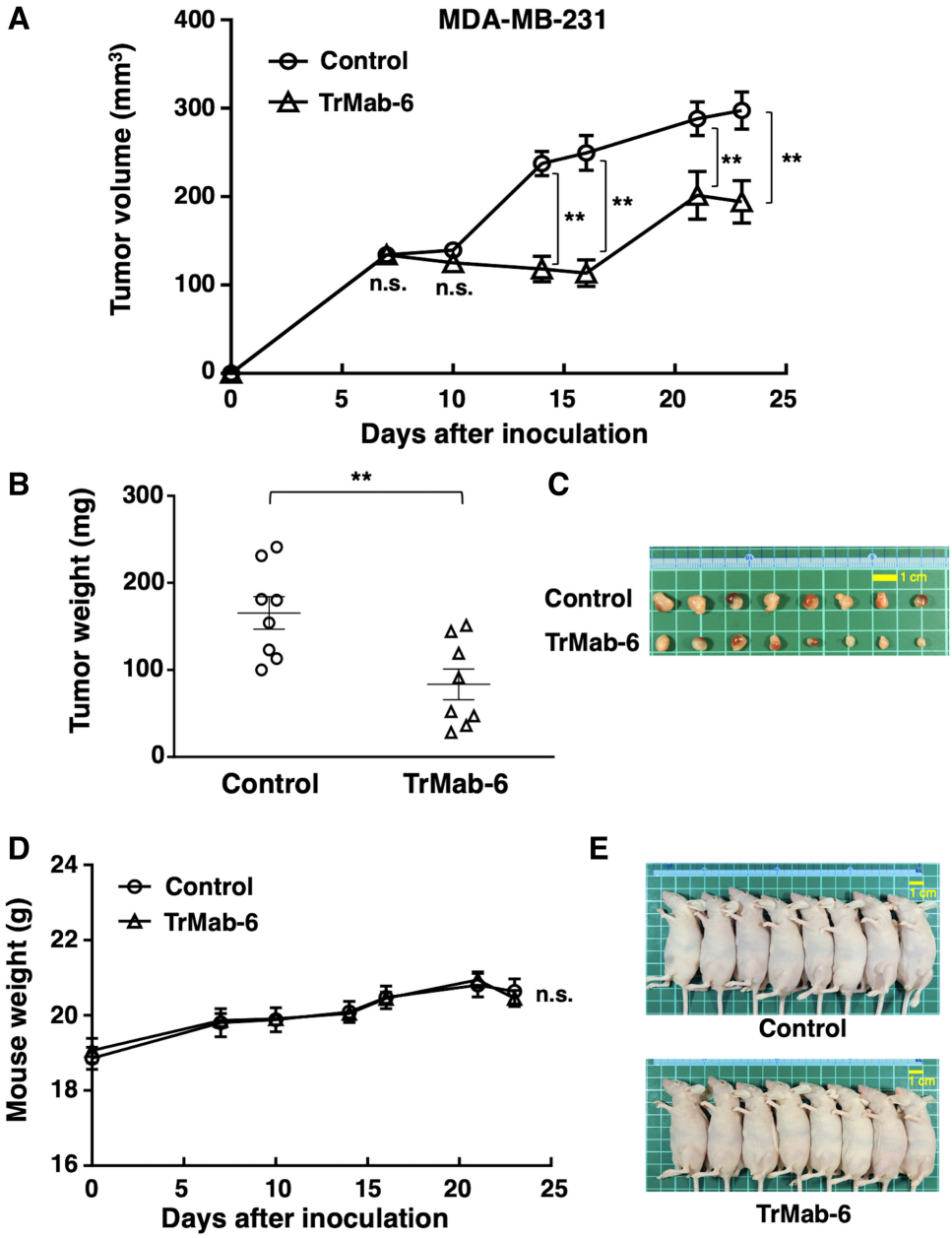

FIG. 1. Evaluation of the antitumor activity of TrMab-6 in MDA-MB-231 xenografts. **(A)** MDA-MB-231 cells (5×10^6 cells) were subcutaneously injected into the left flank. After day 7, 100 μg of TrMab-6 and control mouse IgG in 100 μL of PBS was i.p. injected into the treated and control mice, respectively. Additional antibodies were then injected on days 14 and 21. Tumor volume was measured on days 7, 10, 14, 16, 21, and 23. Values are mean ± SEM. Asterisk indicates statistical significance (**$p < 0.01$, ANOVA and Sidak's multiple comparisons test). **(B)** MDA-MB-231 xenograft tumors were resected from the TrMab-6 and control mouse IgG groups. Tumor weight on day 23 was measured from excised xenografts. Values are mean ± SEM. Asterisk indicates statistical significance (**$p < 0.01$, Welch's *t*-test). **(C)** Resected tumors of MDA-MB-231 xenografts from the control mouse IgG and TrMab-6 groups on day 23. Scale bar, 1 cm. **(D)** Body weights of the mice implanted with MDA-MB-231 xenografts were recorded on days 7, 10, 14, 16, 21, and 23. **(E)** Body appearance of mice on day 23. Scale bar, 1 cm. i.p., intraperitoneally; n.s., not significant; PBS, phosphate-buffered saline; SEM, standard error of the mean.

Antitumor effect of TrMab-6 in mouse xenografts of a TROP2-expressing TNBC cell line (MDA-MB-468)

The tumor formation of 16 MDA-MB-468-bearing mice was observed on day 7 before the mice were divided into TrMab-6-treated and control groups. On days 7, 14, and 21, after MDA-MB-468 cell injections into the mice, either TrMab-6 (100 μg) or control mouse IgG (100 μg) was i.p. injected into the mice. Tumor volume was measured on days 7, 10, 14, 16, 21, and 23 after MDA-MB-468 cell injection. The TrMab-6-treated mice exhibited significantly less tumor growth on days 10 ($p < 0.05$), 14 ($p < 0.01$), 16 ($p < 0.01$), 21 ($p < 0.01$), and 23 ($p < 0.01$) compared with the IgG-treated control mice (Fig. 2A).

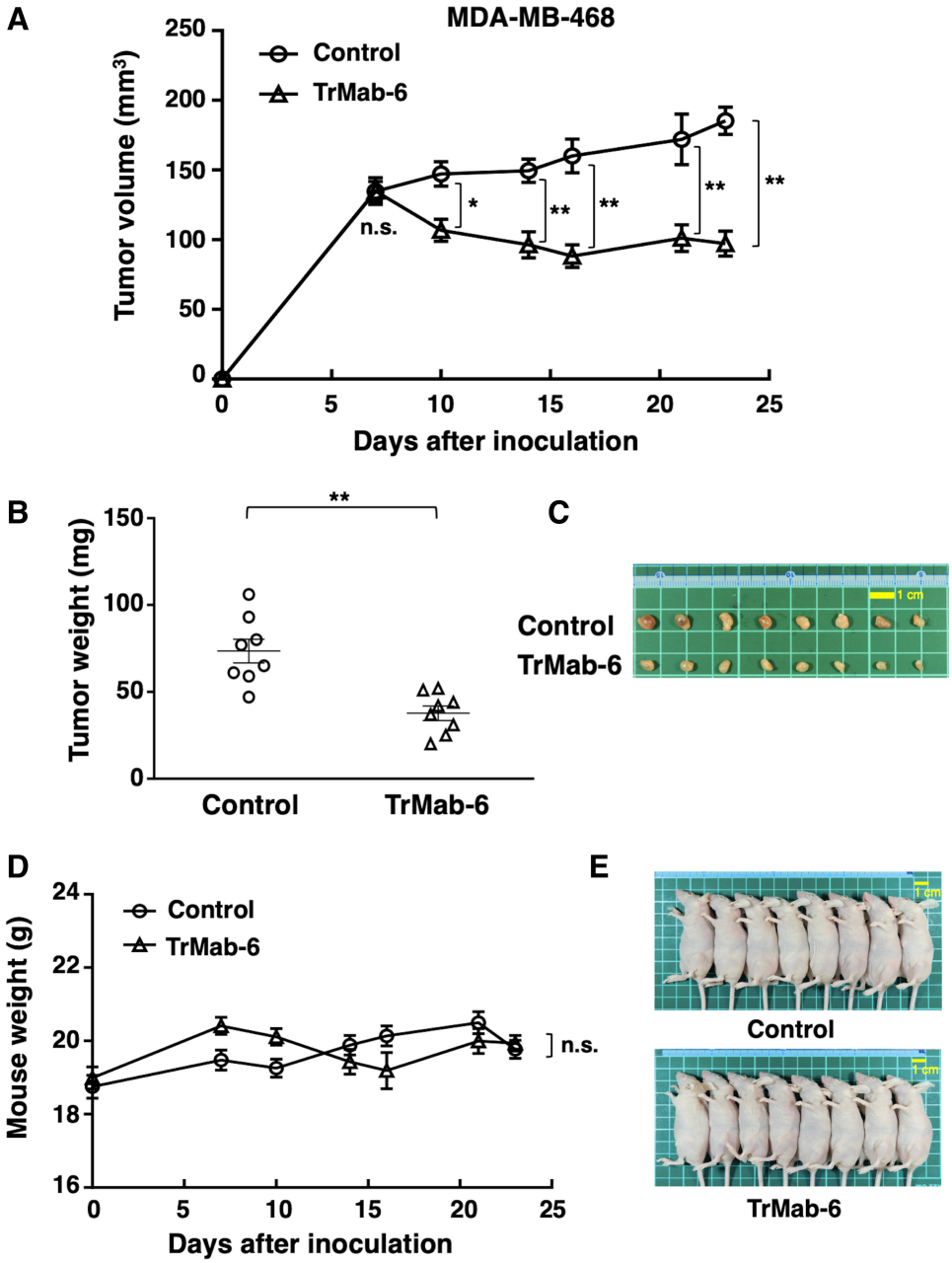

FIG. 2. Evaluation of the antitumor activity of TrMab-6 in MDA-MB-468 xenografts. **(A)** MDA-MB-468 cells (5×10^6 cells) were subcutaneously injected into the left flank. After day 7, 100 μg of the TrMab-6 and control mouse IgG in 100 μL of PBS was i.p. injected into the treated and control mice, respectively. Additional antibodies were then injected on days 14 and 21. Tumor volume was measured on days 7, 10, 14, 16, 21, and 23. Values are mean ± SEM. Asterisk indicates statistical significance (**$p < 0.01$, *$p < 0.05$, ANOVA and Sidak's multiple comparisons test). **(B)** MDA-MB-468 xenograft tumors were resected from the TrMab-6 and control mouse IgG groups. Tumor weight on day 23 was measured from excised xenografts. Values are mean ± SEM. Asterisk indicates statistical significance (**$p < 0.01$, Welch's *t*-test). **(C)** Resected tumors of MDA-MB-468 xenografts from control mouse IgG and TrMab-6 groups on day 23. Scale bar, 1 cm. **(D)** Body weights of the mice implanted with MDA-MB-468 xenografts were recorded on days 7, 10, 14, 16, 21, and 23. **(E)** Body appearance of mice on day 23. Scale bar, 1 cm. n.s., not significant.

On day 23, a 48% reduction in tumor volume was seen in the TrMab-6-treated mice (Fig. 2A). Tumors from the TrMab-6-treated mice weighed significantly less than those from the IgG-treated control mice on day 23 (49% reduction, $p < 0.01$; Fig. 2B). Figure 2C demonstrates each tumor. These results indicated that TrMab-6 reduced the growth of MDA-MB-468 xenografts, but did not contribute toward their total elimination. Total body weights did not significantly differ between the treatment and control groups (Fig. 2D). Figure 2E demonstrates the body appearance of the mice.

Discussion

Precision medicine has recently received considerable attention in personalized cancer treatment.[31,32] The concept of precision medicine targets proteins that drive carcinogenesis based on the abnormal molecular profiles of individual patients with cancer rather than the types of cancers, leading to the improved prognosis of patients.[33] Increasing evidence has indicated that TROP2 influences tumor development and progression. TROP2 overexpression enhances cancer cell proliferation and invasion, and predicts poor prognosis in solid tumors.[22,34,35] Alternatively, the inhibition of TROP2 expression impaired cell proliferation, migration, and invasion.[34,36] Therefore, TROP2 has been regarded as a potential target for cancer immunotherapy and precision medicine.

The therapeutic potential of mAbs and their derivatives has been well recognized for decades in treating various diseases, including cancer.[37] Ikeda et al. have established two recombinant fucosylated and defucosylated chimeric TROP2-targeting antibodies (cAR47A6.4.2 and cPr1E11, respectively) with a human IgG_1 constant region. Although both antibodies showed significant anticancer activities against human cancer cell lines *in vitro* and *in vivo*, only ADCC, but not CDC, was a substantive contributor to the efficacy of antibodies.[38]

As far as we know, TrMab-6 is the first anti-TROP2 mAb that shows anticancer activity *in vivo* (Figs. 1 and 2) and possesses both ADCC and CDC activities.[30] Stein et al. established RS7, a mouse IgG_1 mAb raised against human lung carcinoma, which could recognize human TROP2.[39–41] Since RS7 is rapidly internalized from the surface of cells after binding to TROP2, this antibody provided an opportunity to deliver cytotoxins into TROP2-expressing cells. The radiolabeled RS7 significantly showed antitumor activity *in vivo*,[42–45] and mutant ranpirnase, an amphibian RNase toxin-conjugated humanized RS7 (hRS7), inhibited cancer cell growth *in vivo*.[46,47] Sacituzumab govitecan is an FDA-approved first-in-class ADC consisting of hRS7, conjugated to a topoisomerase I inhibitor (SN-38) for treating advanced breast cancer.[48] These results indicate that antibody-based immunotherapy is a promising therapeutic strategy for TROP2-expressing cancers.

Intriguingly, TROP2 expression was detected in cancer cells and normal human tissues, including the prostate, cervix, lung, breast, uterus, kidney, skin, pancreas, liver, salivary grand, and esophagus.[49,50] These results suggest that TROP2 has important functions in normal tissues, thereby raising concerns that TROP2-targeting therapies may have unexpected effects on normal tissues. We recently developed a novel technology to produce a cancer-specific mAb (CasMab), which could react only to the antigen on cancer cells.[51] We have also successfully produced several CasMabs.[52–55] In the future, TROP2 CasMab production can be applicable as a basis for designing and optimizing potent immunotherapy modalities, such as ADCs and chimeric antigen receptor T cell therapy.

Authors' Contributions

T.T., T.O., M.S., and M.K. performed the experiments; M.K.K. designed the experiments; and T.T. and Y.K. wrote the article.

Author Disclosure Statement

No competing financial interests exist.

Funding Information

This research was supported, in part, by the Japan Agency for Medical Research and Development (AMED) under grant numbers JP21am0401013 (to Yukinari Kato) and JP21am0101078 (to Yukinari Kato).

References

1. Nielsen TO, Hsu FD, Jensen K, Cheang M, Karaca G, Hu Z, Hernandez-Boussard T, Livasy C, Cowan D, Dressler L, Akslen LA, Ragaz J, Gown AM, Gilks CB, van de Rijn M, and Perou CM: Immunohistochemical and clinical characterization of the basal-like subtype of invasive breast carcinoma. Clin Cancer Res 2004;10:5367–5374.
2. O'Reilly D, Sendi MA, and Kelly CM: Overview of recent advances in metastatic triple negative breast cancer. World J Clin Oncol 2021;12:164–182.
3. Liu Y, Xin T, Huang DY, Shen WX, Li L, Lv YJ, Jin YH, Song XW, Teng C, and Jiang QY: Prognosis in very young women with triple-negative breast cancer: Retrospective study of 216 cases. Med Oncol 2014;31:222.
4. Stead LA, Lash TL, Sobieraj JE, Chi DD, Westrup JL, Charlot M, Blanchard RA, Lee JC, King TC, and Rosenberg CL: Triple-negative breast cancers are increased in black women regardless of age or body mass index. Breast Cancer Res 2009;11:R18.
5. Prakash O, Hossain F, Danos D, Lassak A, Scribner R, and Miele L: Racial disparities in triple negative breast cancer: A review of the role of biologic and non-biologic factors. Front Public Health 2020;8:576964.
6. Dent R, Trudeau M, Pritchard KI, Hanna WM, Kahn HK, Sawka CA, Lickley LA, Rawlinson E, Sun P, and Narod SA: Triple-negative breast cancer: Clinical features and patterns of recurrence. Clin Cancer Res 2007;13:4429–4434.
7. Agostinetto E, Eiger D, Punie K, and de Azambuja E: Emerging therapeutics for patients with triple-negative breast cancer. Curr Oncol Rep 2021;23:57.
8. Jin J, Gao Y, Zhang J, Wang L, Wang B, Cao J, Shao Z, and Wang Z: Incidence, pattern and prognosis of brain metastases in patients with metastatic triple negative breast cancer. BMC Cancer 2018;18:446.
9. McCann KE, Hurvitz SA, and McAndrew N: Advances in targeted therapies for triple-negative breast cancer. Drugs 2019;79:1217–1230.
10. Liao M, Zhang J, Wang G, Wang L, Liu J, Ouyang L, and Liu B: Small-molecule drug discovery in triple negative breast cancer: Current situation and future directions. J Med Chem 2021;64:2382–2418.
11. Katz H, and Alsharedi M: Immunotherapy in triple-negative breast cancer. Med Oncol 2017;35:13.
12. Esteva FJ, Hubbard-Lucey VM, Tang J, and Pusztai L: Immunotherapy and targeted therapy combinations in metastatic breast cancer. Lancet Oncol 2019;20:e175–e186.
13. Soliman H, Khalil F, and Antonia S: PD-L1 expression is increased in a subset of basal type breast cancer cells. PLoS One 2014;9:e88557.
14. Mittendorf EA, Philips AV, Meric-Bernstam F, Qiao N, Wu Y, Harrington S, Su X, Wang Y, Gonzalez-Angulo AM, Akcakanat A, Chawla A, Curran M, Hwu P, Sharma P, Litton JK, Molldrem JJ, and Alatrash G: PD-L1 expression in triple-negative breast cancer. Cancer Immunol Res 2014;2:361–370.

15. Narang P, Chen M, Sharma AA, Anderson KS, and Wilson MA: The neoepitope landscape of breast cancer: Implications for immunotherapy. BMC Cancer 2019;19:200.
16. Ghebeh H, Mohammed S, Al-Omair A, Qattan A, Lehe C, Al-Qudaihi G, Elkum N, Alshabanah M, Bin Amer S, Tulbah A, Ajarim D, Al-Tweigeri T, and Dermime S: The B7-H1 (PD-L1) T lymphocyte-inhibitory molecule is expressed in breast cancer patients with infiltrating ductal carcinoma: Correlation with important high-risk prognostic factors. Neoplasia 2006;8:190–198.
17. Tolba MF, Elghazaly H, Bousoik E, Elmazar MMA, and Tolaney SM: Novel combinatorial strategies for boosting the efficacy of immune checkpoint inhibitors in advanced breast cancers. Clin Transl Oncol 2021;23:1979–1994.
18. Zhang J, Tian Q, Zhang M, Wang H, Wu L, and Yang J: Immune-related biomarkers in triple-negative breast cancer. Breast Cancer 2021;28:792–805.
19. McDougall AR, Tolcos M, Hooper SB, Cole TJ, and Wallace MJ: Trop2: From development to disease. Dev Dyn 2015;244:99–109.
20. Zaman S, Jadid H, Denson AC, and Gray JE: Targeting Trop-2 in solid tumors: Future prospects. Onco Targets Ther 2019;12:1781–1790.
21. Zeng P, Chen MB, Zhou LN, Tang M, Liu CY, and Lu PH: Impact of TROP2 expression on prognosis in solid tumors: A systematic review and meta-analysis. Sci Rep 2016;6: 33658.
22. Goldenberg DM, Stein R, and Sharkey RM: The emergence of trophoblast cell-surface antigen 2 (TROP-2) as a novel cancer target. Oncotarget 2018;9:28989–29006.
23. Son S, Shin S, Rao NV, Um W, Jeon J, Ko H, Deepagan VG, Kwon S, Lee JY, and Park JH: Anti-Trop2 antibody-conjugated bioreducible nanoparticles for targeted triple negative breast cancer therapy. Int J Biol Macromol 2018; 110:406–415.
24. Schneck H, Gierke B, Uppenkamp F, Behrens B, Niederacher D, Stoecklein NH, Templin MF, Pawlak M, Fehm T, and Neubauer H: EpCAM-independent enrichment of circulating tumor cells in metastatic breast cancer. PLoS One 2015;10:e0144535.
25. Seligson JM, Patron AM, Berger MJ, Harvey RD, and Seligson ND: Sacituzumab govitecan-hziy: An antibody-drug conjugate for the treatment of refractory, metastatic, triple-negative breast cancer. Ann Pharmacother 2021;55: 921–931.
26. Shaffer C: Trop2 deal heats up antibody-drug conjugate space in cancer. Nat Biotechnol 2021;39:128–130.
27. Bardia A, Hurvitz SA, Tolaney SM, Loirat D, Punie K, Oliveira M, Brufsky A, Sardesai SD, Kalinsky K, Zelnak AB, Weaver R, Traina T, Dalenc F, Aftimos P, Lynce F, Diab S, Cortés J, O'Shaughnessy J, Diéras V, Ferrario C, Schmid P, Carey LA, Gianni L, Piccart MJ, Loibl S, Goldenberg DM, Hong Q, Olivo MS, Itri LM, and Rugo HS: Sacituzumab govitecan in metastatic triple-negative breast cancer. N Engl J Med 2021;384:1529–1541.
28. Kalinsky K, Diamond JR, Vahdat LT, Tolaney SM, Juric D, O'Shaughnessy J, Moroose RL, Mayer IA, Abramson VG, Goldenberg DM, Sharkey RM, Maliakal P, Hong Q, Goswami T, Wegener WA, and Bardia A: Sacituzumab govitecan in previously treated hormone receptor-positive/HER2-negative metastatic breast cancer: Final results from a phase I/II, single-arm, basket trial. Ann Oncol 2020;31:1709–1718.
29. Sayama Y, Kaneko MK, and Kato Y: Development and characterization of TrMab-6, a novel anti-TROP2 monoclonal antibody for antigen detection in breast cancer. Mol Med Rep 2021;23:92.
30. Tanaka T, Ohishi T, Asano T, Takei J, Nanamiya R, Hosono H, Sano M, Harada H, Kawada M, Kaneko MK, and Kato Y: An anti-TROP2 monoclonal antibody TrMab-6 exerts antitumor activity in breast cancer mouse xenograft models. Oncol Rep 2021;46:132.
31. Tsimberidou AM, Fountzilas E, Nikanjam M, and Kurzrock R: Review of precision cancer medicine: Evolution of the treatment paradigm. Cancer Treat Rev 2020;86:102019.
32. Rodriguez H, Zenklusen JC, Staudt LM, Doroshow JH, and Lowy DR: The next horizon in precision oncology: Proteogenomics to inform cancer diagnosis and treatment. Cell 2021;184:1661–1670.
33. Jackson SE, and Chester JD: Personalised cancer medicine. Int J Cancer 2015;137:262–266.
34. Nieto MA, Huang RY, Jackson RA, and Thiery JP: EMT: 2016. Cell 2016;166:21–45.
35. Liu T, Liu Y, Bao X, Tian J, Liu Y, and Yang X: Overexpression of TROP2 predicts poor prognosis of patients with cervical cancer and promotes the proliferation and invasion of cervical cancer cells by regulating ERK signaling pathway. PLoS One 2013;8:e75864.
36. Zhao W, Jia L, Kuai X, Tang Q, Huang X, Yang T, Qiu Z, Zhu J, Huang J, Huang W, and Feng Z: The role and molecular mechanism of Trop2 induced epithelial-mesenchymal transition through mediated beta-catenin in gastric cancer. Cancer Med 2019;8:1135–1147.
37. Grilo AL, and Mantalaris A: The increasingly human and profitable monoclonal antibody market. Trends Biotechnol 2019;37:9–16.
38. Ikeda M, Kato K, Yamaguchi M, Hamada H, Nakamura K, and Sugimoto Y: Cell surface antibody retention influences in vivo antitumor activity mediated by antibody-dependent cellular cytotoxicity. Anticancer Res 2016;36:5937–5944.
39. Stein R, Basu A, Chen S, Shih LB, and Goldenberg DM: Specificity and properties of MAb RS7-3G11 and the antigen defined by this pancarcinoma monoclonal antibody. Int J Cancer 1993;55:938–946.
40. Stein R, Basu A, Goldenberg DM, Lloyd KO, and Mattes MJ: Characterization of cluster 13: The epithelial/carcinoma antigen recognized by MAb RS7. Int J Cancer Suppl 1994;8:98–102.
41. Shih LB, Xuan H, Aninipot R, Stein R, and Goldenberg DM: In vitro and in vivo reactivity of an internalizing antibody, RS7, with human breast cancer. Cancer Res 1995; 55:5857s–5863s.
42. Stein R, Goldenberg DM, Thorpe SR, and Mattes MJ: Advantage of a residualizing iodine radiolabel for radioimmunotherapy of xenografts of human non-small-cell carcinoma of the lung. J Nucl Med 1997;38:391–395.
43. Stein R, Govindan SV, Mattes MJ, Shih LB, Griffiths GL, Hansen HJ, and Goldenberg DM: Targeting human cancer xenografts with monoclonal antibodies labeled using radioiodinated, diethylenetriaminepentaacetic acid-appended peptides. Clin Cancer Res 1999;5:3079s–3087s.
44. Stein R, Govindan SV, Chen S, Reed L, Spiegelman H, Griffiths GL, Hansen HJ, and Goldenberg DM: Successful therapy of a human lung cancer xenograft using MAb RS7 labeled with residualizing radioiodine. Crit Rev Oncol Hematol 2001;39:173–180.

45. Stein R, Govindan SV, Mattes MJ, Chen S, Reed L, Newsome G, McBride BJ, Griffiths GL, Hansen HJ, and Goldenberg DM: Improved iodine radiolabels for monoclonal antibody therapy. Cancer Res 2003;63:111–118.
46. Chang CH, Gupta P, Michel R, Loo M, Wang Y, Cardillo TM, and Goldenberg DM: Ranpirnase (frog RNase) targeted with a humanized, internalizing, anti-Trop-2 antibody has potent cytotoxicity against diverse epithelial cancer cells. Mol Cancer Ther 2010;9:2276–2286.
47. Liu D, Cardillo TM, Wang Y, Rossi EA, Goldenberg DM, and Chang CH: Trop-2-targeting tetrakis-ranpirnase has potent antitumor activity against triple-negative breast cancer. Mol Cancer 2014;13:53.
48. Syed YY: Sacituzumab govitecan: First approval. Drugs 2020;80:1019–1025.
49. Stepan LP, Trueblood ES, Hale K, Babcook J, Borges L, and Sutherland CL: Expression of Trop2 cell surface glycoprotein in normal and tumor tissues: Potential implications as a cancer therapeutic target. J Histochem Cytochem 2011;59:701–710.
50. Trerotola M, Jernigan DL, Liu Q, Siddiqui J, Fatatis A, and Languino LR: Trop-2 promotes prostate cancer metastasis by modulating β(1) integrin functions. Cancer Res 2013;73:3155–3167.
51. Kato Y, and Kaneko MK: A cancer-specific monoclonal antibody recognizes the aberrantly glycosylated podoplanin. Sci Rep 2014;4:5924.
52. Yamada S, Kaneko MK, and Kato Y: LpMab-23: A cancer-specific monoclonal antibody against human podoplanin. Monoclon Antib Immunodiagn Immunother 2017;36:72–76.
53. Kaneko MK, Nakamura T, Kunita A, Fukayama M, Abe S, Nishioka Y, Yamada S, Yanaka M, Saidoh N, Yoshida K, Fujii Y, Ogasawara S, and Kato Y: ChLpMab-23: Cancer-specific human-mouse chimeric anti-podoplanin antibody exhibits antitumor activity via antibody-dependent cellular cytotoxicity. Monoclon Antib Immunodiagn Immunother 2017;36:104–112.
54. Miyazaki A, Nakai H, Sonoda T, Hirohashi Y, Kaneko MK, Kato Y, Sawa Y, and Hiratsuka H: LpMab-23-recognizing cancer-type podoplanin is a novel predictor for a poor prognosis of early stage tongue cancer. Oncotarget 2018;9:21156–21165.
55. Kaneko MK, Ohishi T, Kawada M, and Kato Y: A cancer-specific anti-podocalyxin monoclonal antibody (60-mG2a-f) exerts antitumor effects in mouse xenograft models of pancreatic carcinoma. Biochem Biophys Rep 2020;24:100826.

Address correspondence to:
Yukinari Kato
Department of Molecular Pharmacology
Tohoku University Graduate School of Medicine
2-1, Seiryo-machi, Aoba-ku
Sendai 980-8575
Japan

E-mail: yukinarikato@med.tohoku.ac.jp

Received: November 17, 2021
Accepted: January 11, 2022