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研究課題名（和文）The research of targeting tumor-associated macrophages, a new combination therapy strategy by macrophage polarization and immune checkpoint blockade

研究課題名（英文）The research of targeting tumor-associated macrophages, a new combination therapy strategy by macrophage polarization and immune checkpoint blockade

研究代表者
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研究者番号：４０８６８９６５

交付決定額（研究期間全体）：（直接経費）　　3,500,000円

研究成果の概要（和文）：βグルカンは、腫瘍に誘導内CD11b陽性細胞の抗腫瘍効果を増強させる。本研究では、PD-L1阻害抗体によるβグルカンの抗腫瘍効果の増強とその相乗効果メカニズムに注目した。マウスメラノーマモデルにおいて、PD-L1阻害抗体とβグルカンの併用は、βグルカン単独投与に比して腫瘍の縮小が確認された。βグルカンと抗PD-L1抗体投与は、CTLの増加および腫瘍浸潤白血球のT細胞機能増強のみならず、腫瘍浸潤CD11b陽性細胞活性の増強も見られた。メラノーマモデルにおいて、PD-L1阻害抗体は腫瘍に誘導されたCD11b陽性細胞の免疫チェックポイント機構を阻害することにより、βグルカンの抗腫瘍効果を高めた。

研究成果の学術的意義や社会的意義
Immunotherapy aimed at reversing the phenotype of tumor-recruited CD11b+ cells to promote the anti-tumor phenotype is a promising area of research, and further investigation is needed to establish its clinical value in cancer immunotherapy.

研究成果の概要（英文）：One of the major functions of tumor-recruited CD11b+ cells is suppressing the T-cell mediated anti-tumor immune response. β-glucan could enhance tumor-recruited CD11b+ cells anti-tumor effects. However, β-glucan could increase PD-1/PD-L1 expression on CD11b+ cells. These effects may be reversed by PD-1/PD-L1 block therapy. In the present study, we focused on the PD-1/PD-L1 blocked therapy enhanced the β-glucan antitumor effects, and their synergistic effects mechanism. In our mouse melanoma model, PD-L1 blocking antibody with β-glucan synergized tumor regression. After treatment with β-glucan and anti-PD-L1 antibody, tumor infiltrating leukocyte (TILs) were not only competent for the T cell function and CTL population but also showed enhanced tumor-recruited CD11b+ myeloid cells activity.PD-1/PD-L1 blocked therapy enhanced the β-glucan antitumor effects via blockade the tumor-recruited CD11b+ cells immune checkpoints.

研究分野：腫瘍診断および治療学関連

キーワード：PD-L1　Tumor microenvironment　CD11b+ cells　β-glucan

1. 研究開始当初の背景

In the tumor microenvironment (TME), one of the major functions of tumor-recruited CD11b+ cells are the suppression of the T-cell mediated anti-tumor immune response. β-glucan is a well-known immunostimulating agent with anti-tumor activities. β-glucan could convert the phenotype of tumor-recruited CD11b+ cells from the suppressive to the promotive, and enhanced their anti-tumor effects. β-glucan could convert the phenotype of tumor-recruited CD11b+ cells from the suppressive to the promotive, and enhanced their anti-tumor effects. However, β-glucan could enhance the PD-1/PD-L1 expression on CD11b+ cells, while PD-1/PD-L1 could inhibit CD11b+ cells phagocytosislead and lead to T cell exhaustion. These pro-tumor effects may be reversed by PD-1/PD-L1 block therapy. We think this is one of the important side effects of β-glucan. It is extremely important to reverse this side effect.

2. 研究の目的

In the present study, we focused on the efficacy of β-glucan anti-tumor therapy combined with anti-PD-L1 mAb treatment, and the mechanism of their synergistic effects could be fully verified.

3. 研究の方法

C57BL/6 mice were subcutaneously injected with MO4-Luc(B16-OVA) by applicants. After 8 days, Tumors bearing mice were intraperitoneally (i.p.; intraperitoneal injection.) treated with anti-murine anti-PD-L1mAb, β-glucan, or both agents. Tumor volumes were calculated using the formula for typical ellipsoid length × (width2) /2. We verify anti-PD-L1mAb enhances the antitumor effects of β-glucan in melanoma tumor model（Fig1）. We will analyze the mechanism and confirm this effect to another tumor model. For mechanism, we will confirm the effect of β-glucan and β-glucan + anti-PD-L1mAb on CD11b+ cells and TIL's T cells function. Applicant will isolate TILs, spleen cells and peritoneal exudate macrophages, analyze the effect of β- glucan on immune checkpoint PD-1/PD-L1. We isolate it from different group in tumor bearing mouse and analyze TAMs phenotype and functions such as cytokines (IFN-g, TNF-a, IL-12, IL-1β, IL-6. The TILs would isolate from different group. We will analyze different group TILs phenotype and functions such as cytokines (IFN-g, TNF-a, IL-2), cytolytic particles (Granzyme B, Perforin and CD107a) and T cell proliferation marker (Ki67).

4. 研究成果

In the present study, we focused on the efficacy of β-glucan anti-tumor therapy combined with anti-PD-L1 mAb treatment, and the mechanism of their synergistic effects could be fully verified（Fig1）. We verified the effect of β-glucan (i.e., inflammatory cytokine secretion of TNF-a, IL-12, IL-6, IL-1β and the expression of immune checkpoint PD-1/PD-L1) in naïve mouse peritoneal exudate $CD11b^+$ cells. In our mouse melanoma model, treatment with a PD-L1 blocking antibody with β-glucan synergized tumor regression. After treatment with β-glucan and anti-PD-L1 mAb antibody, tumor infiltrating leukocyte (TILs) not only showed a competent T cell function (CD107a, perforin, IL-2, IFN-γ and Ki67) (Fig2) and CTL population but also showed enhanced tumor-recruited $CD11b^+$ cell activity (IL-12, IL-6, IL-1β and PD-1). Therefore, immunotherapy aimed at reversing the phenotype of tumor-recruited CD11b+ cells to promote the anti-tumor phenotype is a promising area of research, and further investigation is needed to establish its clinical value in cancer immunotherapy.

Fig1

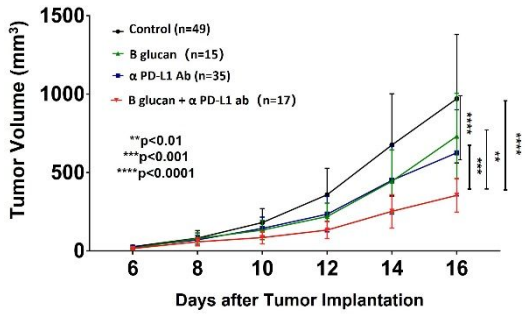
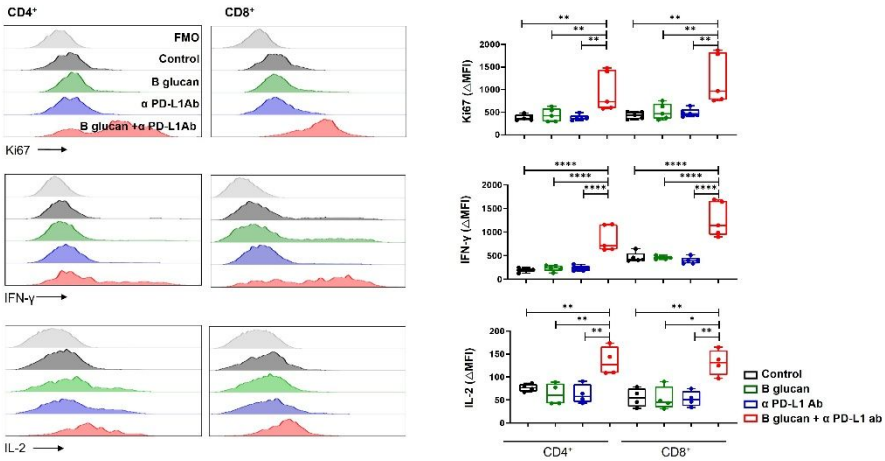


Fig2


CD4+
CD8+
FMO
Control
B glucan
α PD-L1Ab
B glucan +α PD-L1Ab
Ki67
IFN-γ
IL-2
Ki67 (△MFI)
IFN-γ (△MFI)
IL-2 (△MFI)
Control
B glucan
α PD-L1 Ab
B glucan + α PD-L1 ab
CD4+
CD8+

５．主な発表論文等

〔雑誌論文〕　計3件（うち査読付論文　3件／うち国際共著　3件／うちオープンアクセス　2件）

１．著者名 Hu Xin、Shui Yifang、Hirano Hiroshi、Kusano Kisato、Guo Wen-Zhi、Fujino Masayuki、Li Xiao-Kang	４．巻 72
２．論文標題 PD-L1 antibody enhanced β-glucan antitumor effects via blockade of the immune checkpoints in a melanoma model	５．発行年 2022年
３．雑誌名 Cancer Immunology, Immunotherapy	６．最初と最後の頁 719～731
掲載論文のDOI（デジタルオブジェクト識別子） 10.1007/s00262-022-03276-4	査読の有無 有
オープンアクセス オープンアクセスとしている（また、その予定である）	国際共著 該当する

１．著者名 Shui Yifang、Hu Xin、Hirano Hiroshi、Kusano Kisato、Tsukamoto Hirotake、Li Mengquan、Hasumi Kenichiro、Guo Wen-Zhi、Li Xiao-Kang	４．巻 101
２．論文標題 β-glucan from Aureobasidium pullulans augments the anti-tumor immune responses through activated tumor-associated dendritic cells	５．発行年 2021年
３．雑誌名 International Immunopharmacology	６．最初と最後の頁 108265～108265
掲載論文のDOI（デジタルオブジェクト識別子） 10.1016/j.intimp.2021.108265	査読の有無 有
オープンアクセス オープンアクセスではない、又はオープンアクセスが困難	国際共著 該当する

１．著者名 Shui Yifang、Hu Xin、Hirano Hiroshi、Tsukamoto Hirotake、Guo Wen-Zhi、Hasumi Kenichiro、Ijima Fumihiro、Fujino Masayuki、Li Xiao-Kang	４．巻 39
２．論文標題 Combined phospholipids adjuvant augments anti-tumor immune responses through activated tumor-associated dendritic cells	５．発行年 2023年
３．雑誌名 Neoplasia	６．最初と最後の頁 100893～100893
掲載論文のDOI（デジタルオブジェクト識別子） 10.1016/j.neo.2023.100893	査読の有無 有
オープンアクセス オープンアクセスとしている（また、その予定である）	国際共著 該当する

〔学会発表〕　計4件（うち招待講演　0件／うち国際学会　0件）

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２．発表標題 Enhancement of antitumor effects of PD-L1 blockade by 5-ALA/SFC via upregulation of exhausted T cells metabolism in mice
３．学会等名 第80回 日本癌学会学術総会
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Xin Hu, Weitao Que, Masayuki Fujino, Xiao-Kang Li

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Xin Hu, Masayuki Fujino, Xiao-Kang Li

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３．学会等名
第81回日本癌学会学術集会

４．発表年
2022年～2023年

〔図書〕　計0件

〔産業財産権〕

〔その他〕

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６．研究組織

	氏名 （ローマ字氏名） （研究者番号）	所属研究機関・部局・職 （機関番号）	備考

７．科研費を使用して開催した国際研究集会

〔国際研究集会〕　計0件

８．本研究に関連して実施した国際共同研究の実施状況

共同研究相手国	相手方研究機関