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Inhaled dry powder cisplatin increases antitumour response to anti-PD1 in a murine lung cancer model

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Introduction

Despite progress in targeted therapy and immunotherapy, lung cancer remains the leading cause of cancer-related deaths worldwide. The combination of chemotherapy and immunotherapy, is now considered a standard treatment option for most lung cancer patients not eligible for targeted therapy.

Cisplatin is one of the most successful chemotherapeutic agents in the world and is used in the treatment of lung cancer. Inhaled chemotherapy offers a solution to the problem of systemic toxicity observed with intravenous administration. Cisplatin dry powder for inhalation (CIS-DPI) was developed to address limitations of nebulized cisplatin and intravenous chemotherapy [1]. This study aims to test the efficacy of CIS-DPI with the immune checkpoint inhibitor anti-PD1 in a murine lung cancer model.

Results & discussion

- CIS-DPI demonstrated a dose-dependent antiproliferative activity *in vivo*. Immunohistochemistry analysis revealed upregulation of PD-L1 following CIS-DPI treatment, which is in line with results observed in lung cancer patients following intravenous cisplatin injection [2].
- The combination of CIS-DPI and anti-PD1 significantly reduced tumour growth compared with anti-PD1 monotherapy (Figure 1A-B).
- The combination group significantly prolonged median survival by 57% (from 24 to 33 days).

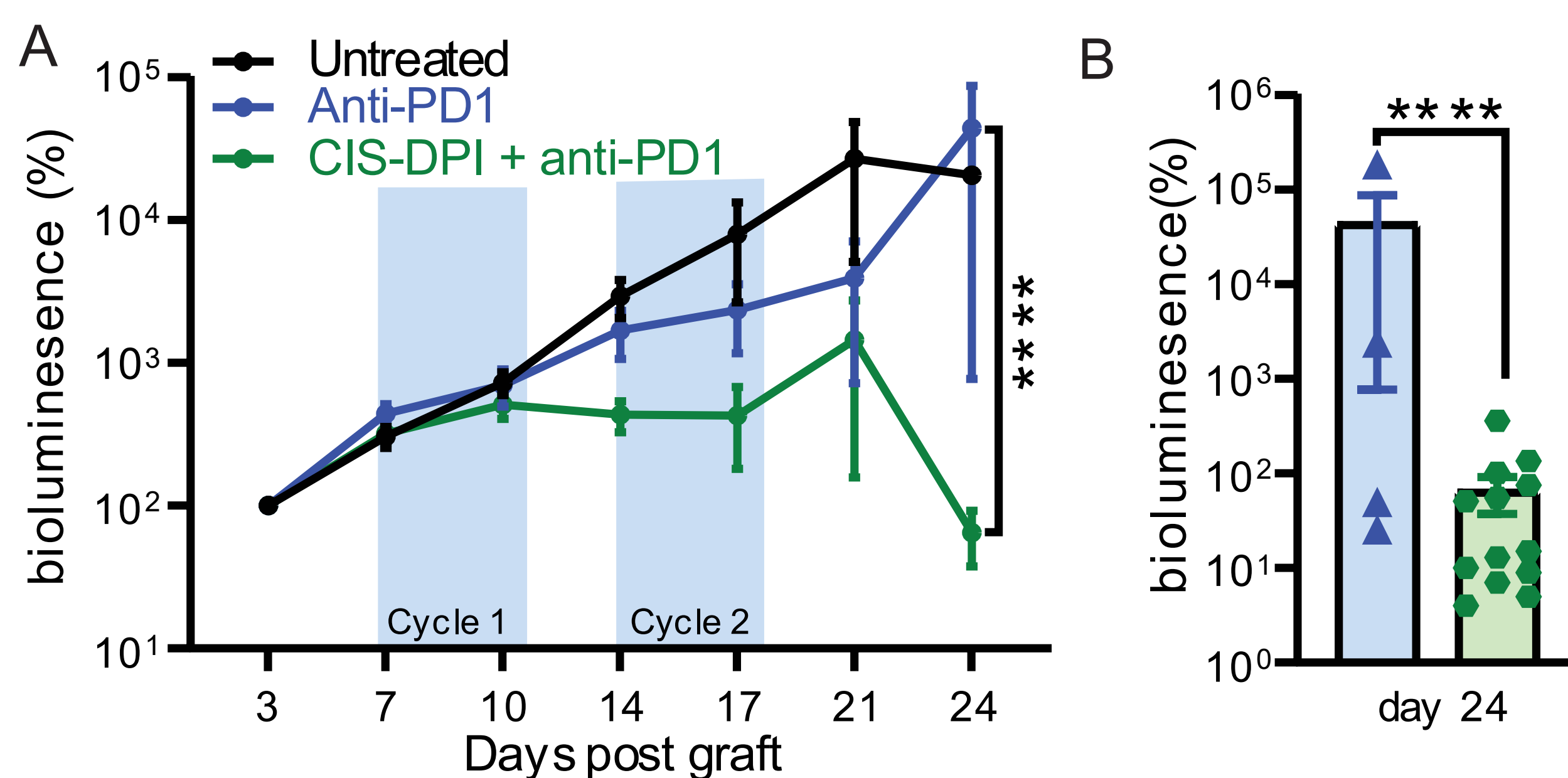


Figure 1. Tumour growth measured by bioluminescence was expressed as percentage (%) of signal measured on day 3. Mice were left untreated (n=7) or received the anti-PD1 (n=11) or the combination treatment CIS-DPI 1 mg/kg + anti-PD1 (n=16). Lines represent mean ± SEM. Statistics are two-way ANOVA multiple comparisons on day 24 (A-B). (B) Individual tumour growth values (% day 3) ± SD of mice in anti-PD1 and combination groups at day 24.

- The analysis of the tumour immune infiltrate by flow cytometry revealed significantly more dendritic cells from subset 1 (DC1), a subset specialised in presenting tumour antigens to T-cells in the combination group compared with the untreated (Figure 2A).
- There were significantly more CD8 T lymphocytes, a T cell subset involved in killing cancer cells, in the combination group compared with the untreated (Figure 2B).
- Increase in DC1 and CD8 T cells reflects the initiation of an anti-tumour immune response.

Material & methods

- The M109-luc2 orthotopic lung carcinoma model was implanted in BALBc mice.
- Bioluminescence imaging (BLI) was used to track tumour growth over time.
- CIS-DPI was delivered by endotracheal administration at the maximum tolerated dose (MTD) of 1 mg/kg cisplatin.
- Analysis of tumour immune infiltrate was performed by flow cytometry and immunohistochemistry (IHC) after 1 treatment cycle.

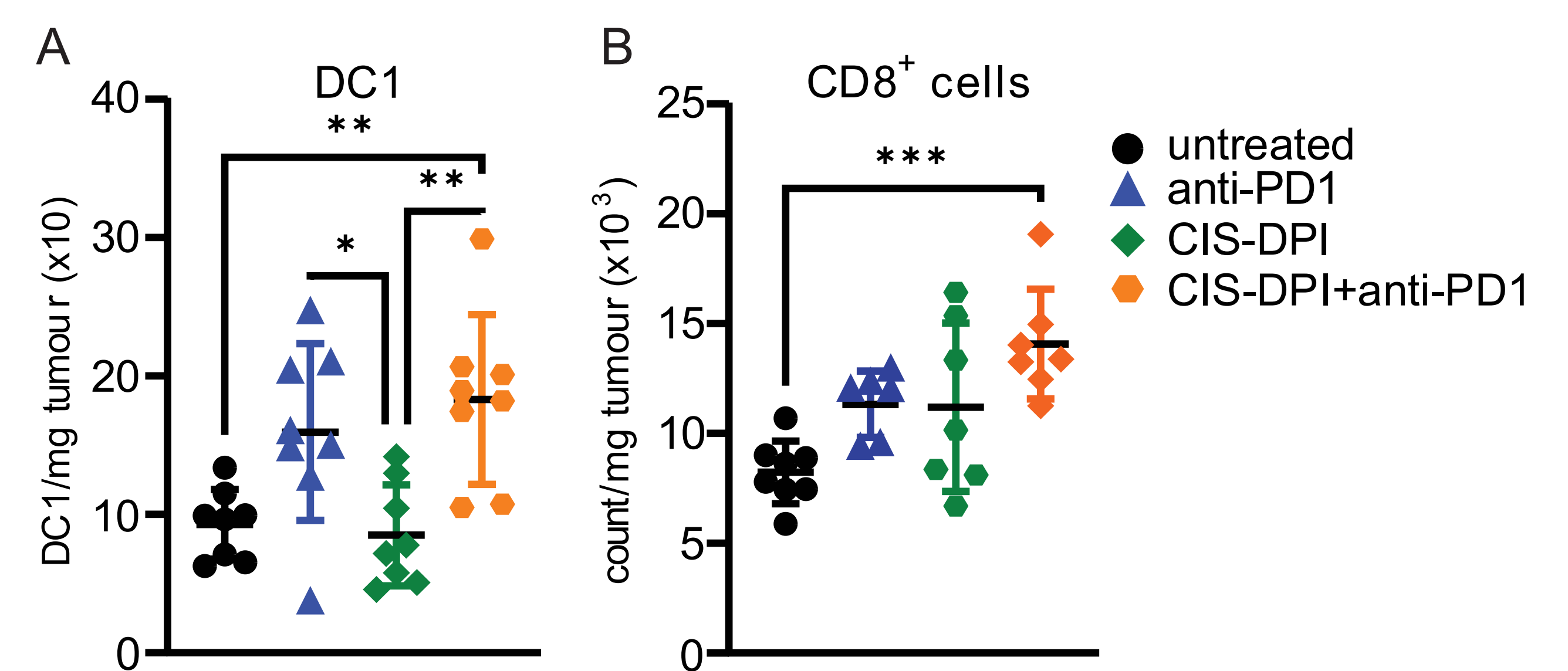
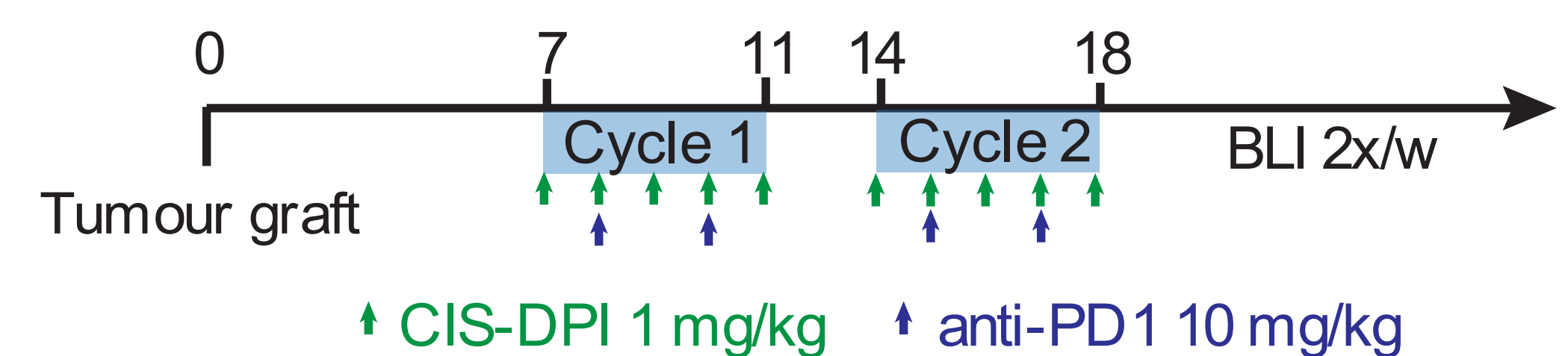


Figure 2. Analysis of tumour infiltrated immune cells. (A) Number of DC1 (CD103⁺) among CD11c⁺MHCII^{high} DCs on day 13, and of CD8⁺ T cells (B) on day 15 (2 and 4 days after the last dose of treatment respectively).

- IHC analysis on tumours confirmed that CIS-DPI monotherapy induced CD8 T cell recruitment after 1 treatment cycle. In addition, the combination induced the recruitment of significantly more CD8 T cells compared with anti-PD1 monotherapy (Figure 3A-B).
- The combination of CIS-DPI and anti-PD1 induced a significant upregulation of PD-L1 in the tumour (Figure 3C). This observation correlates with the PD-L1 upregulation following neoadjuvant cisplatin chemotherapy observed in NSCLC patients and is a predictive biomarker of response to anti-PD1 in patients [2-3].

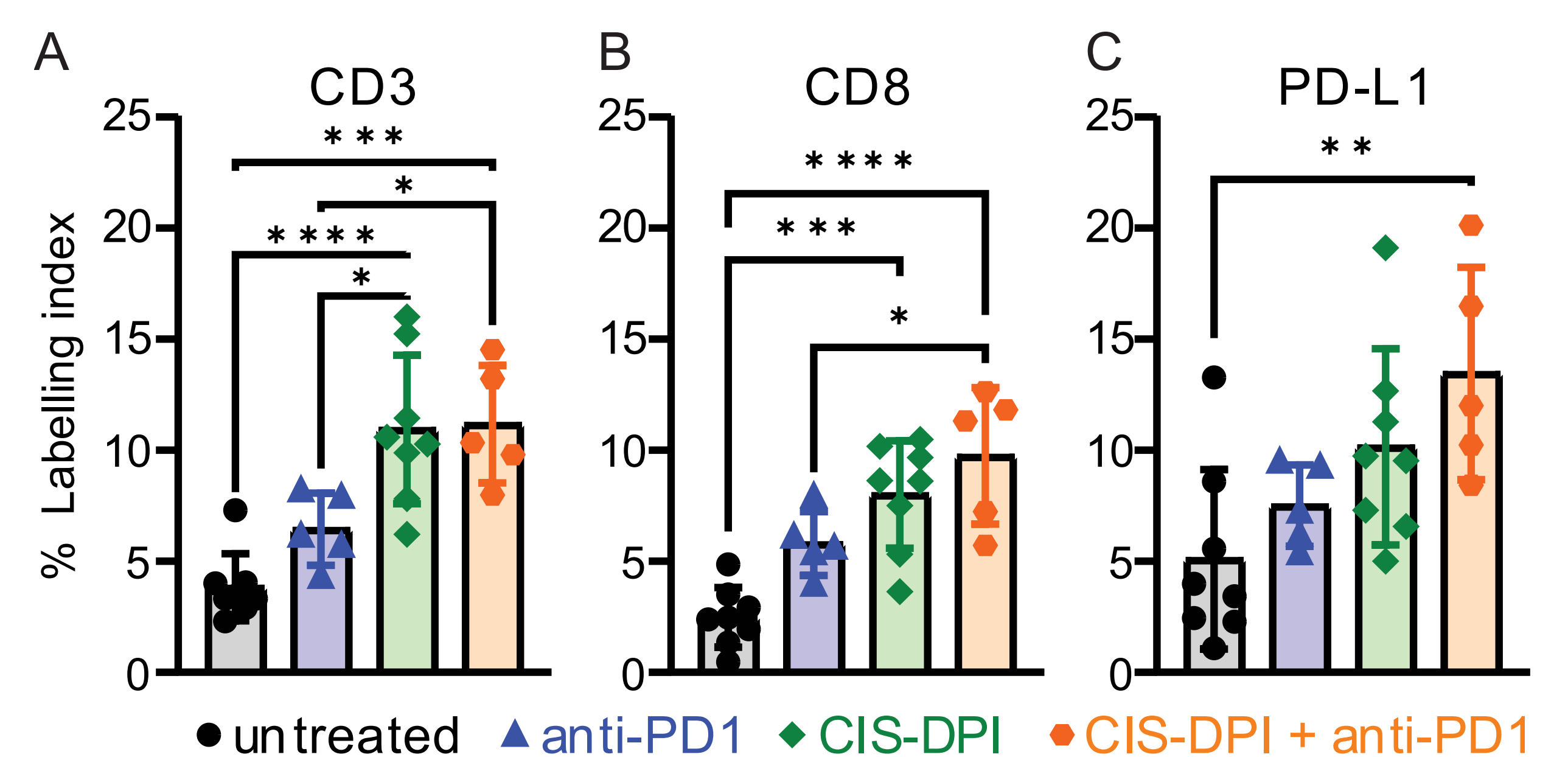


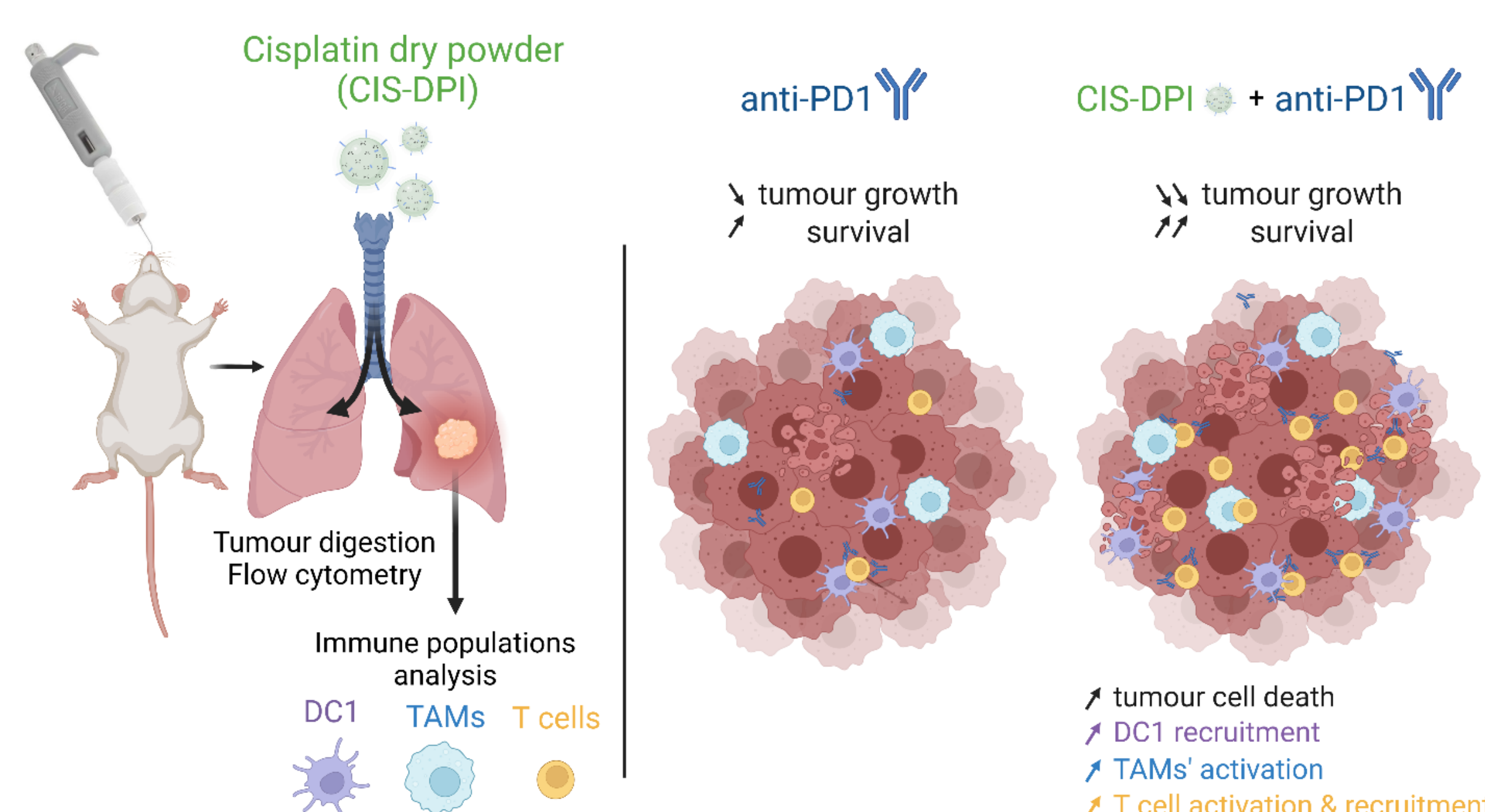
Figure 3. Immunohistochemistry of M109 tumours treated as indicated at day 15. The Percentage of positive (A) CD3, (B) CD8 and (C) PD-L1 is represented as labelling index. Statistics are two-way anova multiple comparisons between groups. P*[<]0,05, **p[<]0,01, ***p[<]0,001, ****p[<]0,0001

Conclusions & perspectives

CIS-DPI demonstrated a rapid cytotoxic and immunogenic effect and enhanced the anti-tumour immune response when combined with anti-PD1.

Combining CIS-DPI with immune checkpoint inhibitors (ICIs) promises higher efficacy, with a better safety profile than cisplatin iv [1], and a better quality of life for patients who could take their treatment at home with this novel treatment modality.

A phase I/IIa clinical study is due to start in 2023 to investigate the safety of CIS-DPI in NSCLC patients. It remains to be determined which dose levels and regimens would be optimal to induce the strongest cytotoxic and immunogenic effect: when combined with ICI, such as anti-PD1.



References

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