ORIGINAL RESEARCH

Humoral and cellular responses to mRNA-based COVID-19 booster vaccinations in patients with solid neoplasms under active treatment

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Background: Patients with cancer are at high risk for severe coronavirus disease 2019 (COVID-19) infection. Knowledge regarding the efficacy of the messenger RNA (mRNA) vaccines in actively treated cancer patients is limited as they had been excluded from the pivotal studies of these vaccines. We evaluated humoral and cellular immune responses in cancer patients after double vaccination and a booster dose and identified disease- and treatment-related factors associated with a reduced immune response. We also documented the number and outcome of breakthrough infections.

Patients and methods: Patients with metastatic solid malignancies undergoing active treatment were included if they had received two doses of the severe acute respiratory syndrome coronavirus 2 mRNA vaccines BNT162b2 or mRNA-1273 and a booster dose. Other causes of immunosuppression and previous COVID-19 infections (positive anti-nucleocapsid titers) were exclusion criteria. Anti-spike antibodies, neutralizing antibodies (nAbs) and T-cell responses were assessed about 6 months after the two-dose vaccination and 4 weeks after the booster.

Results: Fifty-one patients had pre-booster and 46 post-booster measurements. Anti-spike titers after two vaccine doses were highly variable and significantly lower in older patients, during treatment with chemotherapy compared to targeted and endocrine treatments and in patients with low CD4+ or CD19+ cell counts. The booster dose led to a significant increase in anti-spike antibodies and nAbs, achieving almost uniformly high titers, irrespective of baseline and treatment factors. The cellular immune response was also significantly increased by the booster, however generally more stable and not influenced by baseline factors and treatment type. Seventeen patients (33%) experienced breakthrough infections, but none required hospital care or died from COVID-19.

Conclusions: An mRNA vaccine booster dose is able to increase humoral and cellular immune responses and to overcome the immunosuppressive influence of baseline and treatment factors in cancer patients. Breakthrough infections were uniformly mild in this vaccinated high-risk population.

Key words: COVID-19, mRNA vaccine, solid tumors, humoral immunity, cellular immunity

INTRODUCTION

Vaccinations are an essential part of supportive therapy for cancer patients and recommended as part of routine care.[1] This also applies to the recently available messenger RNA (mRNA)-based vaccines against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), especially because

patients with neoplastic diseases represent a particularly vulnerable patient group. They bear a significantly higher risk for both acquiring SARS-CoV-2 infections and suffering a more severe course of coronavirus disease 2019 (COVID-19) compared to individuals without cancer.[2,3]

Knowledge on the efficacy of the mRNA-based SARS-CoV-2 vaccines in patients with actively treated solid malignant diseases, however, is still emerging as these patient groups had been excluded from the pivotal studies leading to approval of these vaccines.[4,5]

Initial reports indicate that the humoral response after two mRNA vaccine doses (i.e. antibody titers against the spike protein) is impaired in cancer patients compared to healthy age-matched controls.[6-8] Older age, male sex, type of mRNA vaccine and ongoing use of steroids[9,10] have so far

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been discussed as factors associated with a reduced humoral response in solid cancer patients. Patients undergoing chemo- or immunotherapy have also been reported to display lower levels of antibodies in comparison to patients who received an endocrine therapy or no therapy.[7,11,12]

Apart from antibodies, SARS-CoV-2-specific T-cells are also an integral part of the immune response.[13,14] The cellular immune response after two vaccine doses[15,16] or an active COVID-19 infection[17,18] has also been reported to be reduced in cancer patients, though less than the humoral response. This is of great importance as T-cell-mediated immunity is generally assumed to be a more robust correlate for protection, including against severe COVID-19 and variants of SARS-CoV-2.[9,19] So far, risk factors for lower cellular responses in patients with solid tumors are not well defined. The type of anti-neoplastic treatment and the use of steroids within 15 days of vaccination have been postulated as influencing factors.[9,20]

After only a few months, antibody levels against SARS-CoV-2 decrease substantially in cancer patients, as well as in individuals without cancer even after double vaccination.[21,22] Consequently, booster campaigns have been launched in many countries. According to previous studies, booster vaccinations can restore the diminishing humoral responses in solid tumor patients[23-26]; however absolute antibody titers were still lower than in healthy controls.[16,27] The level of neutralizing antibodies (nAbs), too, can be increased with the booster vaccination,[28-30] even in patients without detectable nAbs after two doses.[31] So far, only limited data are available on T-cell responses after the third mRNA-based vaccine.[16,23,31]

In our study, we focused on a population of patients with metastatic solid malignancies all undergoing active cancer treatment at the time of vaccination. We investigated the humoral and in addition cellular immune response 6 months after double vaccination with the BNT162b2 or mRNA-1273 SARS-CoV-2 vaccines and the effect of a third booster dose. Additionally, we aimed to identify disease- and treatment-related factors within this population associated with a reduced humoral and cellular immune response and to document the number and outcome of breakthrough infections.

PATIENTS AND METHODS

Setting/patients

For this prospective cohort study, we recruited patients with a documented metastatic solid malignancy at the Department of Medical Oncology and Haematology of the Cantonal Hospital of St. Gallen, Switzerland. For enrollment, patients had to have received two doses of the approved SARS-CoV-2 mRNA vaccines BNT162b2 or mRNA-1273 and be under active systemic treatment at the time of the initial two-dose vaccination. Patients were excluded if they had any other comorbidity known to be associated with an immunosuppression (e.g. human immunodeficiency virus

infection) or if they received immunosuppressive treatment for a reason other than neoplastic disease. Known previous COVID-19 (either symptomatic or asymptomatic and documented by positive anti-nucleocapsid titers) excluded patients from participation. Information on patient demographics, type of solid neoplasm, anti-neoplastic treatment, steroid co-medication, vaccines given and potential SARS-CoV-2 breakthrough infections was obtained from clinical records and by actively interviewing patients. The mean follow-up between booster vaccination and last contact was 170 days.

Blood samples were collected ∼6 months [median time 211 days, interquartile range (IQR) = 189.0-240.5 days] after the second (baseline, pre-booster) and ∼4 weeks (median time 34.5 days, IQR = 28.0-41.0 days) (post-booster) after the third vaccination to determine humoral and cellular immune responses as well as lymphocyte differentiation and levels of immunoglobulins. For comparison and validity of the pre-booster results, a cohort of 22 healthy age-matched volunteers was recruited.

This research project was conducted in accordance with the Declaration of Helsinki and the principles of Good Clinical Practice. Ethical approval had been granted for the project and use of clinical patient data (BASEC 2021-01062) before data collection and analysis, and patients gave written informed consent for participation. The study protocol is provided in the Supplementary Material, available at https://doi.org/10.1016/j.esmoop.2022.100587.

The literature search was carried out repeatedly on PubMed between February and June 2022 using the following keywords: solid tumor, COVID-19, SARS-CoV-2, vaccine, neutralizing antibodies, T-cells, BNT162b2 and mRNA-1273.

Measurement of humoral and cellular immune response

Antibodies against SARS-CoV-2 viral proteins nucleocapsid (anti-N) and spike (anti-S) were measured in plasma by electrochemiluminescence immunoassay (Roche Elecsys®, Rotkreuz, Switzerland[32]). Positivity was defined as either a cut-off index (COI) of 1 COI/ml and above for anti-N and/or a binding antibody unit (BAU) of 0.8 BAU/ml and above for anti-S. To rate the humoral response following vaccination, the following classification has been suggested for cancer patients by Barrière et al.[33] Responders: anti-S >260 BAU/ml, low responders: anti-S 40-260 BAU/ml and non-responders: anti-S <40 BAU/ml. It is applied in this study.

The presence of SARS-CoV-2-specific T-cells was detected using the enzyme-linked immunospot assay on cryopreserved peripheral blood mononuclear cells (PBMCs). PBMCs were rapidly thawed, incubated overnight and stimulated for 19 h with 15 mers of overlapping peptides (Peptide Solutions, JPT, Berlin, Germany) of SARS-CoV-2 spike protein. The number of interferon-γ-producing cells was quantified as spot-forming cells (SFC) per 10^6 PBMCs.

A surrogate virus neutralization test (Genscript, Piscataway, Piscataway, NJ) with enzyme-linked immunosorbent

assay was used to detect nAbs blocking the interaction between the viral receptor-binding domain of the S glycoprotein (wild type and variants) and the human cell surface receptor angiotensin-converting enzyme-2. A cut-off value of $\geq$30% recommended by the manufacturer indicates the presence of SARS-CoV-2 nAbs.

Statistical analysis

Anti-S levels were plotted and evaluated on a logarithmic scale because of the skewed distribution. Concentrations >5000 BAU/ml were set to 5000 BAU/ml, and those <0.4 BAU/ml were set to 0.4 BAU/ml. Associations of pre-booster anti-S titers with numeric variables were tested using Spearman rank correlation. Differences between two groups were tested using the Mann—Whitney U test (Wilcoxon rank sum test), and differences between more than two groups were tested using the Kruskal—Wallis rank sum test. Differences between two time points were tested using the Wilcoxon paired-samples test. The combined effect of variables on log anti-S concentrations was tested with multiple regression. Due to the small number of patients and variety of solid tumors, analysis was conducted based on the mode of treatment rather than the type of solid malignancy.

RESULTS

Patient and disease characteristics

A total of 56 patients were initially recruited for this study, 5 were excluded before the pre-booster analysis due to withdrawal of consent or because of a previous asymptomatic COVID-19 infection documented by positive anti-N (Supplementary Figure S1, available at https://doi.org/10.1016/j.esmoop.2022.100587). The remaining 51 patients formed the baseline study group and were included for pre-booster analysis. Post-booster analysis was carried out in 46 patients as 2 of the 51 patients did not want to undergo a booster vaccination and 3 others had symptomatic or asymptomatic COVID-19 infections between baseline and follow-up blood collection.

Baseline patient and disease characteristics are shown in Table 1. The median age at first vaccination was 69 (range 32-86) years, and 73% of patients ($n = 37$) were male. Most patients had prostate cancer (43%, $n = 22$), non-small-cell lung cancer (22%, $n = 11$) or colorectal cancer (18%, $n = 9$). Other solid tumors included urothelial carcinoma (6%, $n = 3$), mesothelioma (4%, $n = 2$), renal cell carcinoma (4%, $n = 2$), sarcoma (2%, $n = 1$) and small-cell lung cancer (2%, $n = 1$). All patients had metastatic disease undergoing active treatment as per protocol inclusion criteria. The most common mode of anti-neoplastic treatment was anti-hormonal therapy (41%, $n = 21$), followed by chemotherapy (26%, $n = 13$) and combined chemo-immunotherapy (16%, $n = 8$). The other patients received either targeted therapy with tyrosine kinase inhibitors (TKIs) (10%, $n = 5$) or immunotherapy alone (8%, $n = 4$).

Table 1. Baseline patient and disease characteristics ($n = 51$)

Characteristics	Value
Median age (range), years	69 (32-86)
Males, % (n)	73 (37)
Females, % (n)	27 (14)
Solid malignancy, % (n)	
Prostate carcinoma	43 (22)
NSCLC	22 (11)
Colorectal carcinoma	18 (9)
Urothelial carcinoma	6 (3)
Mesothelioma	4 (2)
Renal cell carcinoma	4 (2)
Sarcoma	2 (1)
SCLC	2 (1)
Treatment, % (n)	
Chemotherapy	26 (13)
Immunotherapy (mono)[a]	8 (4)
Chemo-immunotherapy[b]	18 (8)
Endocrine therapy	41 (21)
Targeted therapy with tyrosine kinase inhibitors	10 (5)
Co-medication, % (n)	
Dexamethasone	37 (19)
Prednisone	12 (6)
Antibiotics[c]	10 (5)
Influenza vaccination in current season, % (n)	67 (34)
SARS-CoV-2 vaccination, % (n)	
BNT162b2 (Pfizer/BioNTech)	80 (41)
mRNA-1273 (Moderna)	20 (10)

NSCLC, non-small-cell lung cancer; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; SCLC, small-cell lung cancer.
[a]3× Pembrolizumab, 1× durvalumab.
[b]4× Pembrolizumab, 1× durvalumab, 2× atezolizumab, 1× avelumab.
[c]During 3 months before vaccination.

Additionally, 37% of patients ($n = 19$) took dexamethasone as co-medication with their treatment (mean monthly dose of 14 mg), 12% ($n = 6$) took prednisone (mean daily dose of 15 mg) and 10% ($n = 5$) required antibiotics during the 3 months before vaccination. Most patients were also vaccinated against influenza during the last season (67%, $n = 34$). Eighty percent ($n = 41$) of patients received the BNT162b2 vaccine, while the other 20% ($n = 10$) were vaccinated with mRNA-1273.

Humoral response and cellular response after two vaccine doses

The median pre-booster anti-S concentration among the baseline study cohort was 295 BAU/ml. For internal control, the pre-booster anti-S titers of the study cohort were compared with those of a healthy, age-matched cohort (median age 67.5 years, $n = 22$). Anti-S titers of the tumor patients were significantly lower than those of the healthy controls (295 BAU/ml versus 912 BAU/ml, $P = 0.0001$) (Supplementary Figure S2, available at https://doi.org/10.1016/j.esmoop.2022.100587). According to the classification by Barrière et al.,[33] 52.9% ($n = 27$) of the study cohort were grouped as responders, 29.4% ($n = 15$) as low responders and 17.6% ($n = 9$) as non-responders after two vaccine doses. This contrasts with the control cohort in which 86.4% ($n = 19$) were classified as responders, 13.6% ($n = 3$) as low responders and none as non-responder.
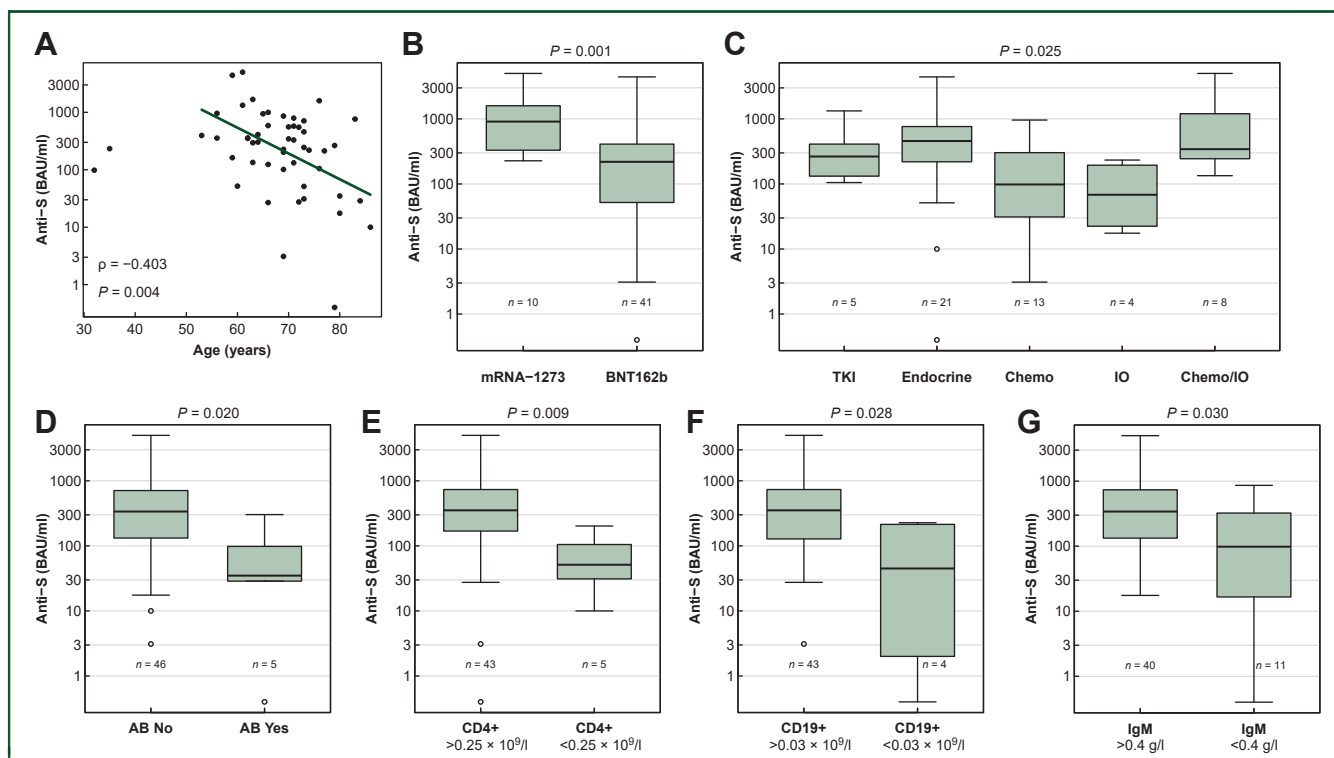
Figure 1. Humoral response after double vaccination in relation to baseline and disease characteristics. Anti-S titer (log scale) after double vaccination in solid tumor patients as a function of (A) age (linear regression line for illustration, only for patients aged >50 years), (B) type of vaccine, (C) systematic therapy, (D) antibiotics intake during the 3 months before vaccination, (E) CD4+ T-cell count, (F) CD19+ B-cell count and (G) IgM levels. P value indicates the significance of the Spearman rank correlation (ρ) for A or the significance of difference in Wilcoxon rank sum test for B-G.
AB, antibiotics; BAU, binding antibody unit; IgM, immunoglobulin M; IO, treatment with immune checkpoint inhibitors; TKI, targeted treatment with tyrosine kinase inhibitors.

We further examined predictors of lower antibody response in the study participants. Anti-S levels of cancer patients were negatively correlated with age ($P = 0.004$) (Figure 1A). Patients vaccinated with mRNA-1273 displayed significantly higher antibody levels compared to those vaccinated with BNT162b2 (910.5 BAU/ml versus 219.0 BAU/ml, $P = 0.001$) (Figure 1B). We also found differences in humoral immune response depending on the type of anti-neoplastic treatment ($P = 0.025$) (Figure 1C) with patients undergoing endocrine or targeted therapy showing the highest anti-S titers with 457 BAU/ml and 264 BAU/ml, respectively. Although these two treatment modalities are not considered as immunosuppressive as chemotherapy, patients who were treated with an endocrine or TKI therapy still displayed lower humoral immune responses compared to healthy controls (402.5 BAU/ml versus 912.0 BAU/ml, $P = 0.012$) (Supplementary Figure S3, available at https://doi.org/10.1016/j.esmoop.2022.100587).

Patients undergoing chemotherapy or immunotherapy generally showed lower anti-S titers with 98.2 BAU/ml and 95.4 BAU/ml only, respectively. Interestingly, patients treated with a combination of chemo- and immunotherapy again showed higher anti-S titers (343 BAU/ml). The use of antibiotics during the 3 months before vaccination ($n = 5$) was also correlated with lower anti-S (median 34.9 BAU/ml) compared to 46 participants without antibiotics (337.5 BAU/ml, $P = 0.020$) (Figure 1D). We found no significant association with sex, co-treatment with dexamethasone or

prednisone and concurrent influenza vaccination (Supplementary Figure S4A-D, available at https://doi.org/10.1016/j.esmoop.2022.100587).

As many patients with metastatic malignancies show underlying lymphopenia, we assessed the correlation of total lymphocyte, CD4+ T- and CD19+ B-cell counts and immunoglobulin levels with anti-S titers. We found significantly lower anti-S titers in patients with CD4+ T-cell levels $<0.25 \times 10^9$/l (352 BAU/ml versus 51.3 BAU/ml, $P = 0.009$) (Figure 1E) and CD19+ B-cell levels $<0.03 \times 10^9$/l[34] (352 BAU/ml versus 106.0 BAU/ml, $P = 0.028$) (Figure 1F). However, we observed no significant correlation with a total lymphocyte count $<1.0 \times 10^9$/l (303 BAU/ml versus 202.0 BAU/ml, $P = 0.371$) (Supplementary Figure S4E, available at https://doi.org/10.1016/j.esmoop.2022.100587). While immunoglobulin G (IgG) levels <7.0 g/l had no significant effect on anti-S titers (Supplementary Figure S4F, available at https://doi.org/10.1016/j.esmoop.2022.100587), low IgM titers <0.4 g/l were associated with a worse humoral response (341 BAU/ml versus 98.2 BAU/ml, $P = 0.030$) (Figure 1G).

Most of the variables with significant influence in the individual tests also had a significant effect in the multiple regression analysis (except from use of antibiotics and CD4+ T-cell count). The most important factors remained age ($P = 0.0013$), type of vaccine ($P = 0.0006$) and type of anti-neoplastic treatment ($P = 0.0043$).

In contrast to the humoral response, we found no significant difference in cellular immunity between cancer
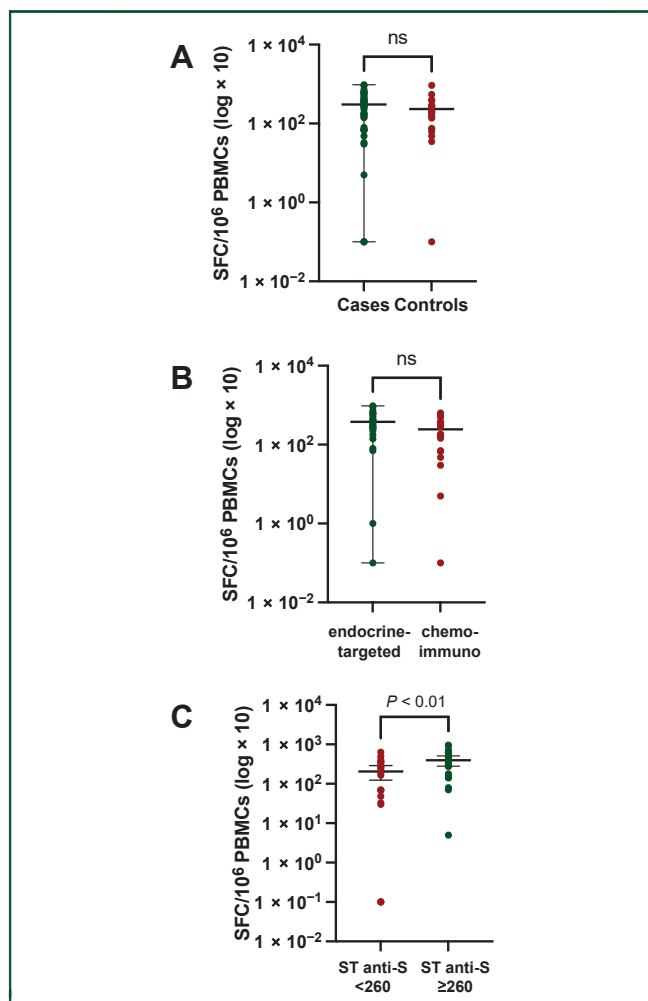
Figure 2. T-cell response after double vaccination. Log scale of SARS-CoV-2-reactive T-cells (spot-forming cells = SFC/10[6] PBMCs) after double vaccination in (A) solid tumor patients compared to healthy controls, (B) in relation to the type of treatment [endocrine or targeted (TKI) versus chemo- and/or immunotherapy] and (C) according to humoral response; *P* value indicates the significance of the Mann–Whitney test.

BAU, binding antibody unit; IO, treatment with immune checkpoint inhibitors; NS, non-significant; PBMCs, peripheral blood mononuclear cells; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; TKI, targeted treatment with tyrosine kinase inhibitors.

patients and healthy controls (279 SFC/10[6] versus 193 SFC/10[6] PBMCs, *P* = 0.4) (Figure 2A) after two vaccinations. Factors identified as having a significant impact on the humoral response such as age, vaccine type, use of antibiotics, lymphocyte counts and immunoglobulins all had no effect on the cellular response (Supplementary Figure S5, available at https://doi.org/10.1016/j.esmoop.2022.100587). Cellular immunity was also not influenced by treatment modality (Figure 2B, Supplementary Figure S6, available at https://doi.org/10.1016/j.esmoop.2022.100587). However, patients displaying a strong humoral response (responders with anti-S ≥260 BAU/ml) also had significantly higher SARS-CoV-2-specific T-cells compared to low responders (330 SFC/10[6] versus 185 SFC/10[6] PBMCs, *P* = 0.01) (Figure 2C).

Effect of booster dose

After the booster vaccination, we observed a significant increase in the anti-S antibody levels, with a median increase from 279.5 BAU/ml (pre-booster, T1) to >5000 BAU/ml (post-booster, T2) (*P* < 0.0001, Figure 3A). According to the classification suggested by Barrière et al.,[33] 95.7% (*n* = 44) of the study population were now responders compared to only 4.3% (*n* = 2) of non-responders. Both non-responders showed some of the predictors which we had identified as being associated with an impaired response after two doses of the vaccine. They were older than the median age (79 and 73 years). One received an endocrine therapy and as co-treatment both prednisone and antibiotics. This patient also displayed low CD19+ B-cells, IgG and IgM levels. The other patient received chemotherapy and displayed low total lymphocyte, CD4+ T-cell and IgG counts. Both patients had very low or no humoral response after the first two vaccinations.

We also observed a significant increase in SFC/10[6] PBMCs after the booster dose (median increase from 279 SFC/10[6] PBMCs to 310 SFC/10[6] PBMCs, *P* = 0.002) (Figure 3B). Only one patient showed no increase in T-cells after the booster. This patient (who received an endocrine therapy, prednisone and antibiotics, and had low levels of CD19+ B-cells, IgG and IgM) also did not show any humoral response after the second or third vaccination. None of the other baseline factors had significant impact on post-booster SARS-CoV-2-specific T-cell levels (Supplementary Figure S5, available at https://doi.org/10.1016/j.esmoop.2022.100587).

nAbs also showed a significant median rise from 59% to 97% (*P* < 0.0001) after the booster (Figure 3C). Only four patients showed low levels of nAbs after the booster dose. Interestingly, two of these had very high post-booster anti-S levels of >5000 BAU/ml.

Breakthrough infections

During this study, 13 patients (25%) suffered from symptomatic infections with SARS-CoV-2. Two patients experienced a breakthrough infection after the second vaccination (mean time between second vaccination and infection: 261 days). These two patients were excluded from the post-booster analysis. Eleven patients got infected after the third vaccination (mean time between booster vaccination and infection: 100 days).

Based on sequencing data from the region of our department (Eastern Switzerland), the period before 27 December 2021 was defined as Delta-dominant, and the period after this date as dominant for the Omicron variant.[35] Two patients (one before and one after the booster) got infected during the Delta-dominant period, and the remaining 11 during the Omicron-dominant period.

All 13 patients experienced varying degrees of influenza-like symptoms such as fever, headaches, fatigue, sore throat, cough and muscle aches. Two patients also reported difficulty in swallowing. The infected patients recovered within 2-10 days except for one patient who suffered from a dry cough for a few weeks and one patient who was hoarse for roughly 6 weeks. Another patient who experienced fatigue for about a month (infected before booster) later had a second SARS-CoV-2 infection, from which he recovered
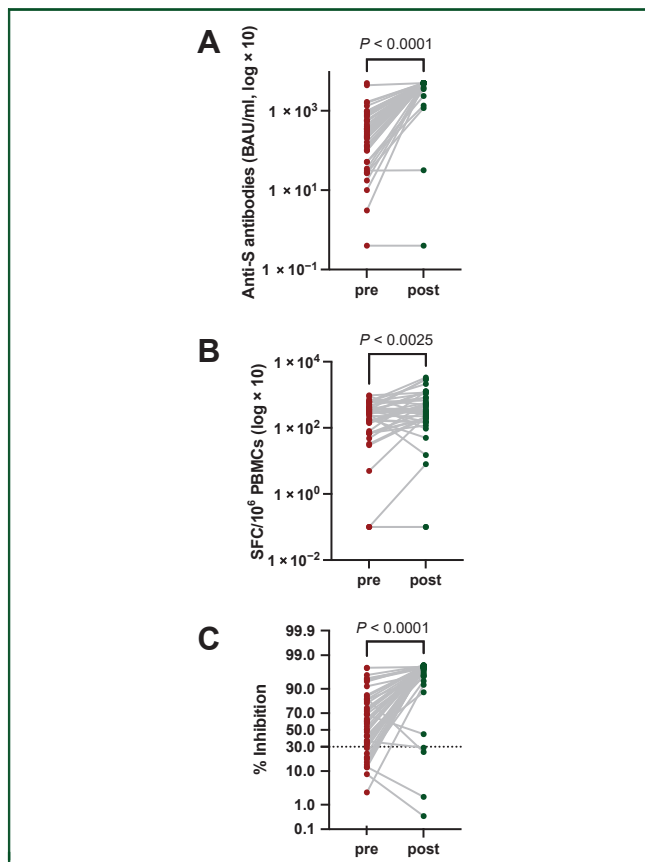
Figure 3. Effects of booster vaccination. On (A) the humoral, (B) the cellular and (C) the neutralizing antibody response against wild type and variants of the virus. *P* value indicates the significance of the Wilcoxon matched-pairs signed rank test.
BAU, binding antibody unit; PBMCs, peripheral blood mononuclear cells; SFC, spot-forming cells; T1, pre-booster; T2, post-booster.

within 10 days. Only one patient partially lost the sense of taste and smell for roughly 10 days.

Another four patients (8%) had asymptomatic, antibody-confirmed, COVID-19 infections, which were detected only by anti-N testing, either after the second ($n = 3$) or the third vaccination ($n = 1$). None of the patients recalled having experienced any symptoms which might have indicated an infection with SARS-CoV-2.

None of the patients in our cohort required hospital care or died from COVID-19.

DISCUSSION

In this study, we assessed the humoral and cellular immune response after two and three doses of the common mRNA vaccines against SARS-CoV-2 in a cohort of patients with metastatic solid malignancies all undergoing active systemic treatment at the time of vaccination. In addition, we carried out an extensive review of the existing literature and provide a summary in Table 2.

We found that after two doses of the vaccines, humoral responses were highly variable and significantly influenced by baseline and disease factors such as age, SARS-CoV-2 vaccine, treatment modality, number of CD4+ and CD19+ lymphocytes and IgM titers. This effect diminished after the

booster, achieving almost uniformly high anti-S titers irrespective of baseline characteristics. Our data therefore corroborate the high immunogenicity of a third (booster) dose on the serological level even in patients who failed a standard after the prior two-dose vaccination scheme. This observation is in line with several previous reports (Table 2) all describing high rates of seropositivity and a significant increase of antibodies after the booster.

SARS-CoV-2-specific T-cells in our cohort also showed a significant increase after the booster dose. However, cellular immunity was largely preserved irrespective of baseline factors and treatment modality. In contrast to the humoral response, we also could not observe a difference between cancer patients and healthy controls and different treatment modalities. Published data on T-cell response have so far been scarce and partly contradictory. While Lasagna and colleagues[36] reported higher T-cell responses in patients treated with immunotherapy, a recent study by Corradini et al.[37] documented comparable T-cell responses across different subgroups of fragile and immune-compromised patients.

However, findings of our study are reassuring. Having found a significant increase of anti-S antibodies, nAbs and SARS-CoV-2-specific T-cells after the booster, we can also provide the clinical follow-up of these patients. We documented a substantial number of symptomatic and asymptomatic breakthrough infections (in total 33% of the cohort) even after three doses of the vaccine. Although breakthrough infections occurred mainly during the Omicron-dominant period, disease courses were uniformly mild with only influenza-like symptoms. None of the patients in our cohort required hospital admission or died from COVID-19, which underlines the efficacy of the vaccine in a patient population at high risk of severe infection and death from COVID-19 when unvaccinated.

With high variability of humoral immune response and reported waning over time,[24,25] our clinical data also support the assumption that T-cell-mediated immunity might be a more robust correlate for vaccine protection against severe COVID-19. However, more data are needed to understand how T-cell responses change over time in the months after the booster dose.

Strengths and limitations

Limitations of our study are the small number of patients and that patients in our cohort were vaccinated with BNT162b2 or mRNA-1273. However, both are mRNA vaccines and it reflects a real-world clinical situation. Another limitation is that about half of the patients in our cohort received endocrine therapy or treatment with a TKI which is generally not considered to cause the same degree of immunosuppression as chemotherapy. Therefore, our results might be less generalizable to a broader population of solid tumor patients receiving chemotherapy only. However, while we observed differences in the humoral response, cellular immunity did not differ depending on treatment modality.

Table 2. Summary of existing data concerning analyses after SARS-CoV-2 booster vaccinations in patients with solid cancers

Reference	Vaccine type	Timepoint of analysis after third dose	Humoral response	Neutralizing antibodies	T-cell response	No. of solid cancer patients (and treatment information)	No. of hematological patients	No. of healthy controls	Main results
Debie et al. *Eur J Cancer.*[24]	BNT162b2	4 weeks	Yes	No	No	Total 115; 96 (83%) on active anticancer treatment	14 under rituximab 10 after HSCT	0	Significantly higher humoral response after booster for all cohorts except rituximab treated
Corradini et al. *Clin Infect Dis.*[37]	BNT162b2, mRNA1273	3 and 4 weeks	Yes	Yes	Yes	37; No/low/medium intensity treatment categories	19	67	Significantly higher humoral and cellular response and neutralizing antibody levels after booster; humoral response and neutralizing titers affected by therapy intensity (targeted versus chemo), cellular response not affected (but lower than in healthy controls)
Di Noia et al., *Ann Oncol.*[38]	BNT162b2	4 weeks	Yes	No	No	407; 366 (90%) on active anticancer treatment (therapy type, antibodies, chemo/antibody, IO, targeted, hormonal)	0	0	98.8% seropositive after booster, significant increase of humoral response after booster; only use of steroids significantly associated with lower antibody levels, therapy type without difference
Fendler et al. *Cancer Cell.*[31]	BNT162b2, ChAdOx1	23 days	No	Yes	Yes	115; 61% on active anticancer treatment	84	0	Significant increase of neutralizing antibodies and cellular response after booster, also for patients with undetectable levels after 2 doses; worse response for hematological patients.
Fendler et al. *Lancet.*[28]	BNT162b2, ChAdOx1	23 days	No	Yes	No				Significant increase in levels of neutralizing antibodies after booster; Omicron (90% seropositive), Delta (97%) and WT (99%); risk factors for worse response only hematological malignancy (no influence of age, sex, vaccine type)
Fenioux et al. *JAMA Oncol.*[39]	BNT162b2	4 weeks	Yes	No	No	36; 30 with chemotherapy, 6 with targeted therapy	0	0	Significantly higher humoral response after booster
Gounant et al. *J Thorac Oncol.*[26]	BNT162b2	21 days	Yes	No	No	26; only thoracic cancer (mixed population with localized and metastatic disease, under active treatment/no treatment)	0	0	Significantly higher humoral response after booster, 88.5% with seroconversion
Lasagna et al. *EMSO Open.*[36]	BNT162b2	21 days	Yes	Yes	Yes	142 (humoral analysis), 77 (cellular analysis), 10 (neutral Ab analysis)	0	0	Significantly higher humoral response after booster, no effect of age or treatment type

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Table 2. Continued

Reference	Vaccine type	Timepoint of analysis after third dose	Humoral response	Neutralizing antibodies	T-cell response	No. of solid cancer patients (and treatment information)	No. of hematological patients	No. of healthy controls	Main results
						all receiving chemo and/or IO			on humoral response; significant increase of cellular response after booster, T-cell non-responders significantly older, higher T-cell response among IO-only treated patients; neutralization of Omicron variant significantly lower
Ligumsky et al. *Lancet Oncol.*[27]	BNT162b2	4 weeks	Yes	No	No	72; All on active anticancer treatment	0	144	Significantly higher humoral response after booster, but lower than in healthy controls
Mair et al. *Eur J Cancer.*[40]	BNT162b2, mRNA1273	15-18 days	Yes	No	No	266; Majority on active anticancer treatment	173	41	Significantly higher humoral response after booster; significantly worse response in hematological patients after B-cell-targeted agent; patients undergoing chemo with significantly lower antibody levels compared to patients without active treatment; antibody levels correlated with CD19 and CD56 counts
Naranbhai et al. *Cancer Cell.*[29]	BNT162b2, mRNA1273, Ad26.COV2.S	14 days or more	No	Yes	No	13	0	0	Booster enhances neutralization of Alpha, Beta, Gamma and Delta variants
Oosting et al. *Lancet Oncol.*[41]	mRNA1273	28 days	Yes	Yes	Yes	47; 8 on IO, 30 with chemotherapy, 9 combined chemo/IO treatment	0	1	Higher humoral and cellular response after booster, lower neutralizing antibodies against Omicron compared to WT
Peeters et al. *ESMO Open.*[42]	BNT162b2	4 weeks	Yes	Yes	No	147; all on active anticancer treatment	41	40	Significantly higher humoral response after booster; solid cancer patients under endocrine/targeted therapy with similar response as healthy controls; fewer high responders under chemo; hematological patients with significantly lower antibody levels than solid cancer patients; neutralizing antibodies against WT variant significantly lower in patients under chemo, IO and hematological patients
Rottenberg et al. *JAMA Oncol.*[25]	BNT162b2	Median of 13 days	Yes	No	No	37; all on active anticancer treatment, 11 (30%) non-metastatic, 19 (51%) under chemo	0	0	Significantly higher humoral response after booster; anti-S antibody levels after second dose and older age correlated with higher antibody levels after booster

Continued

Table 2. Continued									
Reference	Vaccine type	Timepoint of analysis after third dose	Humoral response	Neutralizing antibodies	T-cell response	No. of solid cancer patients (and treatment information)	No. of hematological patients	No. of healthy controls	Main results
Shapiro et al. *Cancer Cell.*[23]	BNT162b2, mRNA1273	4 weeks	Yes	No	Yes	31; some on active cancer treatment	57	0	Solid cancer patients with better post-booster antibody levels than hematological patients; 50% seroconversion and detectable T-cell response in patients without immunity after two doses
Shroff et al. *Nat Med.*[16]	BNT162b2	7 days	Yes	Yes	Yes	20	0	50	Significantly higher humoral response after booster and higher neutralizing antibody levels, no increase in cellular response
Valanparambil et al. *J Clin Oncol.*[43]	BNT162b2, mRNA1273	5-60 and 60-110 days	Yes	Yes	No	14 patients 5-60 days post-booster, 10 patients 60-110 days post-booster; all NSCLC, most on active therapy	0	53	Significantly higher binding and neutralizing antibody titers to the WT and Omicron variant after booster, however 5-7× decrease in both after 2-4 months post-booster
Wagner et al. *Front Immunol.*[44]	BNT162b2, mRNA1273	4 weeks	Yes	Yes	No	63 (breast or lung cancer) 49 (78%) under active treatment	0	66	Significantly higher antibody levels and neutralizing capacity after booster
Zeng et al. *Cancer Cell.*[30]	BNT162b2, mRNA1273	2-112 days (median 47 days)	No	Yes	No	27	0	0	Significantly higher neutralizing response against Omicron after booster, independent from treatment type

Ab, antibody; HSCT, hematopoietic stem cell transplantation; IO, treatment with immune checkpoint inhibitors; NSCLC, non-small-cell lung cancer; WT, wild type.

Finally, we were—based on the assay used—not able to differentiate nAbs against different viral variants (e.g. Omicron). Nevertheless, we can demonstrate that the vaccination induces nAbs in our study population.

The strength of our study is the strict selection of metastatic solid cancer patients all undergoing active systemic treatment at the time of vaccination and having had no prior COVID-19 infection. Most previous studies examined mixed populations of metastatic and non-metastatic patients not all undergoing active treatment. This may result in substantial differences within the study population in terms of risk of severe infection as well as baseline immunological status. Another strength of our study is the completeness of clinical data, allowing us to test for and identify risk factors associated with a reduced response to SARS-CoV-2 vaccines and to report the clinical outcome of breakthrough infections. Finally, we were able to characterize not only humoral immunity (including nAbs) but also SARS-CoV-2-specific cellular immunity, which is less accessible in routine practice because it is more laborious.

Conclusion

Our data underline the efficacy of the booster vaccination against SARS-CoV-2 in actively treated patients with metastatic solid malignancies. Humoral and cellular immune responses were enhanced by the booster dose. Even though there was a substantial number of breakthrough infections at the time of the Omicron wave, these were uniformly mild with no hospital admission or fatalities related to COVID-19. We identified certain baseline and treatment factors associated with a reduced humoral immune response after two doses of the vaccines. These can largely be overcome by the booster dose and are not affecting T-cell immunity. More data are needed to understand how humoral and cellular immune responses persist in the months after the booster dose.

DISCLOSURE

The authors have declared no conflicts of interest.

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