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ORIGINAL ARTICLE

[^{18}F]FDG uptake in axillary lymph nodes and deltoid muscle after COVID-19 mRNA vaccination: a cohort study to determine incidence and contributing factors using a multivariate analysis

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Abstract

Purpose Reactive FDG uptake in the axillary lymph nodes (ALN) and deltoid muscle (DM) after COVID-19 mRNA vaccination has been recognized, although the actual situation in the Japanese population remains unknown. To determine the incidence of reactive FDG uptake and its contributing factors, we retrospectively studied a cohort of subjects who were vaccinated at our hospital.

Methods Whole-body FDG-PET/CT examinations performed in 237 subjects out of 240 subjects with a definite history of COVID-19 vaccination (BNT162b2; BioNTech-Pfizer) were analyzed. Positivity and SUVmax of FDG uptake in the ALN and DM ipsilateral to vaccination, various subject characteristics, and the grade of the pathological FDG-PET/CT findings were evaluated using a multivariate analysis.

Results FDG uptake in the ALN and DM ipsilateral to vaccination was seen in about 60% of the subjects even soon (0–4 days) after the first vaccination, with percentages reaching 87.5% and 75.0%, respectively, after the second vaccination. DM uptake had almost disappeared at around 2 weeks, while ALN uptake persisted for 3 weeks or longer. A multivariate analysis showed that a short duration since vaccination, a younger age, a female sex, and a low FDG-PET/CT grade (minimal pathological FDG uptake) contributed significantly to positive ALN uptake, while a short duration since vaccination and a female sex were the only significant contributors to positive DM uptake. This study is the first to identify factors contributing to positive FDG uptake in ALN and DM after COVID-19 vaccination.

Conclusion A high incidence of FDG uptake in ALN and DM was observed after vaccination. ALN uptake seemed to be associated with a younger age, a female sex, and minimal pathological FDG uptake. After vaccination, an acute inflammatory reaction in DM followed by immune reaction in ALN linked to humoral immunity may be speculated.

Keywords COVID-19 · mRNA vaccine · Axillary lymph node · Deltoid muscle · FDG · PET/CT

Abbreviations

FDG	2-Fluoro[^{18}F]-2-deoxy-D-glucose
PET/CT	Positron emission tomography combined with computed tomography
COVID-19	Coronavirus disease 2019
ALN	Axillary lymph node
DM	Deltoid muscle

Introduction

Nationwide, mass vaccination programs have been recognized as a key measure for controlling the COVID-19 pandemic. In Japan, the vaccination of healthcare workers using an mRNA vaccine (BNT162b2; BioNTech-Pfizer) started on February 17, 2021, and vaccination priority then spread to the elderly beginning in April and to the general population thereafter. Although the start of the vaccination program was relatively delayed, rapid catch-up is ongoing.

Naturally, we have faced to a lot of episodes and information not only of the adverse reactions but also of the influence on radiological imaging. Several case reports [1–7], radiology panel reports [8], suggestions [9], a few cohort studies [10–13], and a review [14] have already been

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reported on the influence of COVID-19 vaccination on radiological imaging, especially on FDG-PET/CT.

At our hospital, several recently vaccinated subjects with FDG uptake in a cluster of axillary/supraclavicular lymph nodes (ALN) and deltoid muscle (DM) have been encountered during routine FDG-PET/CT imaging. And, we have been impressed that available information has been still insufficient. Here, we analyzed a cohort of FDG-PET/CT subjects with a known vaccination history to determine the incidence, SUVmax, and subject characteristics contributing to reactive FDG uptake in ALN and DM after vaccination. The principle hypothesis of this study was that some subject factors may affect the incidence of vaccine-related events on FDG-PET/CT imaging, such as FDG uptake in the ALN and DM. This is a retrospective, cross-sectional single-center study.

Subjects and methods

Subjects

A total of 240 subjects with a definite history of COVID-19 vaccination who underwent a standard whole-body FDG-PET/CT examination between May 10, 2021, and July 21, 2021 were consecutively enrolled. Patients with various types of cancer or large vessel vasculitis (LVV) and healthy subjects undergoing a health check-up that included cancer screening using FDG-PET/CT were included. Subjects undergoing a brain FDG-PET/CT examination only or whose date of vaccination was uncertain were excluded from the analysis. One subject with left advanced breast cancer and ALN metastasis and one subject with lung cancer and left supraclavicular and axillary lymph-node metastasis who had been vaccinated in their left arms, ipsilateral to their disease, were excluded because of the inability to differentiate metastatic FDG uptake from reactive FDG uptake in the lymph nodes. Also, one subject underwent two FDG-PET/CT examinations during the study period; this subject's second examination was excluded. All other subjects underwent only one PET/CT examination during the study period. As a result, 237 FDG-PET/CT examinations were included in the analysis. These examinations were performed in 192 (81.0%) cancer patients, 43 (18.2%) healthy subjects, and 2 (0.9%) LVV patients.

FDG-PET/CT

All the subjects were interviewed before the FDG-PET/CT study and were asked about their vaccine history, surgical history, physical condition, claustrophobia, etc., as part of the regular pre-examination procedure. The FDG-PET/CT scans were performed according to the guidelines of the Japanese Society of Nuclear Medicine [15]. Briefly, after fasting for more than 5 h, a blood glucose measurement was performed. FDG-PET/CT imaging was then performed 90 min after the intravenous injection of 3.7 MBq/kg of FDG. A longer uptake time was used to avoid additional delayed scans and to remain within the upper limit of the guidelines. After a low-dose, non-contrast CT scan, PET data acquisition (2 min per bed position) was performed from the vertex to the upper thigh with the arms in a downward position. Noise filtering, decay correction, image reconstruction, and fusion image preparation were performed using the standard methods based on the manufacturer's recommendations. All the PET/CT machines used in this study (Discovery 600 Q-clear updated, 610, IQ5R, and IQ3R from GE Healthcare, Claritom uMi 780 from United Image Healthcare) were appropriately calibrated and well maintained according to the guidelines. All FDG-PET/CT images obtained for eligible subjects were retrospectively reviewed and evaluated in the present study.

Image interpretation

Using whole-body maximum intensity projection (MIP) images, the presence or absence of vaccine-associated FDG uptake in the axilla (including the supraclavicular and intrapectoral areas) and in the upper arm was determined by comparing each subject's left and right sides. A dot-like or clustered small nodular FDG uptake higher than the background activity in the axilla and localized FDG uptake in the proximal part of the DM higher than the background activity were considered suspicious FDG uptake. If suspicious FDG uptake was observed ipsilateral to the vaccination site on MIP images, the axial fusion images were evaluated and the presence of FDG uptake in the ALN or DM was confirmed to avoid the FDG uptake as a result of arthritis, muscle activity, or metastasis not related to vaccination. The SUVmax of vaccine-associated lesions was then measured. If no significant uptake was seen, the SUVmax was determined to be 0. The ALN was visually evaluated if the ALN was not enlarged beyond the normal range of that for reactive lymph nodes, but the sizes of individual nodes were not measured. Some representative ALN were smaller than 1 cm along the short axis. The shapes were round, oval, club, or string-like and exhibited a large variation, and having the normal hilum of the lymph node typically resembled a low-density wedge at the nodal periphery, but this was not easily seen on the low-dose CT images obtained using PET/CT.

We evaluated the extension of pathological FDG uptake considered to represent the spread of cancer, excluding FDG uptake in the ALN and DM that could be attributed to vaccination, using a grading system. The FDG-PET/CT findings were graded as follows: Grade 0, no pathological FDG uptake; Grade 1, early stage disease consisting of a small

and single lesion, such as a solitary FDG uptake nodule; Grade 2, moderately advanced disease, such as a primary lesion plus a few lymph-node metastases or equivalent status; and Grade 3, advanced disease, such as a large mass lesion and/or multiple metastatic lesions or equivalent status. This grading system was uniformly applied to all cancer patients at initial evaluation as well as re-staging, recurrence, and follow-up and to all non-cancer subjects based on the FDG-PET/CT findings. The aim of this grading system was to evaluate the health conditions of all the subjects using a single and simple method.

Subjects' information

The interval between vaccination and FDG-PET/CT examination (days), whether a first or second vaccination, vaccination in the left or right arm, fasting blood glucose level (FBG, mg/dL), body weight (BW, kg), body mass index (BMI), age, sex, and grade (0–3) of FDG-PET/CT findings were recorded for use in the analyses. After fixing the data, all the analyses were performed anonymously.

The study was performed in accordance with the clinical practice guidelines of the Helsinki Declaration. The study protocol was approved by the institutional review board (No. 513); because of the study's retrospective nature and the use of anonymized data, the need to obtain informed consent from each participant was waived.

Statistics

The Welch *t* test or Chi-square test was used for individual comparisons of each factor. For the multivariate analysis, a binominal logistic regression analysis was performed. A *p* value lower than 0.05 was considered statistically significant. BellCurve for Excel (Social Survey Research Information, Tokyo, Japan) was used for all the statistical analyses.

Results

Subject characteristics

Table 1 shows the characteristics of the study population. Overall, 37% of the subjects were women. The age distribution was skewed toward an older age because of vaccination priority given to elderly individuals over the age of 65 years. A few younger subjects including both medical workers and general population were also included. The distribution of injection sites was 11.8% (28/237) in the right arm and 88.2% (209/237) in the left arm. Thirty-five subjects had received a second vaccination, while 202 subjects had received a first vaccination, probably because the subject enrollment period coincided with an early phase of the local vaccination program.

Table 1 Characteristics of study population

Characteristics of study population ($n=237$)		
Sex	Female	88 (37.1%)
	Male	149 (62.9%)
Age (years)	Mean ± SD	72 ± 9.3
	Range	29—94
FBG (mg/dL)	Mean ± SD	106 ± 21
BW (kg)	Mean ± SD	58.1 ± 11.3
BMI	Mean ± SD	22.5 ± 3.4
Vaccination	Left arm	209 (88.2%)
	Right arm	28 (11.8%)
First vaccination (*n*)		202 (85.2%)
Days after vaccination	Mean ± SD	12.9 ± 8.7
Second vaccination (*n*)		35 (14.8%)
Days after vaccination	Mean ± SD	20.9 ± 19.7

FBG fasting blood glucose, *BW* body weight, *BMI* body mass index

Table 2 Subject category and grading of pathological FDG uptake lesion on FDG-PET/CT images

Subject category	Cancer patients	Health check-up*	LVV†	Total (%)
Grade 0	66	41	1	108 (45.6)
G 1	62	2**	0	64 (27.6)
G 2	26	0	1	27 (11.4)
G 3	38	0	0	38 (16.0)
Total (%)	192 (81.0)	43 (18.2)	2 (0.8)	237

See the descriptions of the grades in the Methods section. The grade will be simply described as FDG-PET/CT grade

†Large vessel vasculitis

*Our hospital provides several health check-up programs for cancer screening using FDG-PET/CT imaging, blood chemistry, endoscopic examination of stomach or colon, ECG, mammography, PAP tests, etc. A total of 43 subjects who underwent cancer screening using FDG-PET/CT imaging and had a history of COVID-19 vaccination were enrolled in this study

** As results of the cancer screening, two stage 1 lung cancers were discovered as solitary nodules with FDG uptake

Grading of FDG-PET/CT findings, subject category, and types of cancer

Table 2 shows the distributions of the grades of pathological FDG uptake lesion according to subject category (cancer patients, health check-up subjects, and LVV patients). In this study, we evaluated the subjects according to FDG-PET/CT grade, and not the subject category. The grade will be simply described as FDG-PET/CT grade.

Table 3 shows the types of cancer and the corresponding FDG-PET/CT grades. Thirty-eight types of cancer were included, with lung cancer comprising the largest group; the remaining subjects exhibited a variety of cancer types. Of note, the examinations were performed pre-therapy, post-therapy, or at the time of re-staging after chemotherapy or radiotherapy. Therefore, the TNM classification at the time of cancer diagnosis does not reflect the actual disease situation at the time of FDG-PET/CT imaging. Instead, a grading system for the FDG-PET/CT images was broadly applied to this heterogeneous population as well as non-oncological subjects.

Table 3 Types of cancer and corresponding FDG-PET/CT grades

Disease and number of patients	FDG-PET/CT Grade				Total
	0	1	2	3	
Malignant lymphoma	3		2	1	6
Multiple myeloma	1				1
Ocular sebaceous ca	1	1			2
Epipharyngeal ca	1				1
Mesopharyngeal ca	2				2
Hypopharyngeal ca	1	1			2
Maxillary sinus ca	1		1	1	3
Floor of the mouth ca	3				3
Tongue ca	5	3		1	9
Gingiva C	3	3		1	7
Submandibular ca	1		1		2
Parotid gland ca	1	1	1		3
Thyroid ca	1		1		2
Laryngeal ca	2	1			3
Lung ca	10	20	2	11	43
Thymic ca	1				1
Pulmonary artery sarcoma	1				1
Breast ca	2	3	1	3	9
Hepatocellular ca			1		1
Bile duct ca	1	4		2	7
Pancreas ca	2	3	2	2	9
Esophageal ca	3	5			8
Gastric ca	4	4	4	2	14
Gastric sarcoma	1				1
Cecal ca		2		1	3
Ascending and transverse colon ca		1	1	3	5
Sigmoid colon ca	2	1	1	1	5
Rectal ca	4	4	5	1	14
Renal ca				2	2
Ureter ca			1	1	2
Bladder ca				2	2
Prostate ca	6	3		1	10
Endometrial ca		1			1
Cervical ca				1	1
Ovarian ca	1				1
Soft tissue sarcoma	1	1	2		4
Malignant melanoma				1	1
Carcinoma unknown primary	1				1
Total	66	62	26	38	192

*ca. cancer

Positivity and SUVmax

The percentages of positive uptake in the ALN and DM on the FDG-PET/CT findings after vaccination were shown in Table 4. The respective percentages of positive uptake in the ALN and DM were 42.6% and 21.5% after either the first or second vaccination, 41.1% and 19.3% after the first vaccination, and 51.4% and 34.3% after the second vaccination; both of these percentages were higher after the second vaccination, compared with after the first vaccination, but only the difference in the DM was significant. The SUVmax of the positive ALN and DM after the second vaccination (4.79 ± 4.91 and 2.17 ± 1.02, respectively) were also higher than those after the first vaccination, but only the difference in ALN was significant, probably because of the small case number and the large variation that was seen in the second vaccination group.

The distributions of the SUVmax values in the ALN and DM according to the number of days since the first or second vaccination are shown in Fig. 1A–D. The graphs clearly showed that uptake in the ALN was higher and continued for longer after vaccination than that in the DM after both the first and second vaccinations. Both the ALN uptake and the DM uptake were higher after the second vaccination than after the first vaccination. A tendency toward a slightly higher ALN uptake in females can also be seen. Large individual variations are also noticeable. Please note that an SUVmax of 0 was regarded as no significant uptake, as described in the Methods.

The numbers and percentages of subjects with positive uptake according to the number of days after the first and second vaccinations are shown in Table 5. At 0–4 days after vaccination, the percentages of positive uptake in the ALN and DM were both over 60% after the first vaccination and as high as 87.5% and 75%, respectively, after the second vaccination; both percentages decreased during subsequent time periods. Uptake in the DM had almost disappeared at around 2 weeks after vaccination, while uptake in the ALN persisted for around 3 weeks or longer.

The correlation between FDG uptake in the ALN and DM is shown in Table 6. Positive uptake in the ALN was observed in 100 subjects, and 49% of these subjects also had positive uptake in the DM. Positive uptake in the DM was observed in 51 subjects, and 49 of these subjects also had positive uptake in the ALN. In 135 subjects, no uptake in either area was seen. After the exclusion of the double-negative subjects, the SUVmax in the ALN was

Table 4 FDG uptake and SUVmax in the axillary lymph nodes (ALN) and deltoid muscle (DM) on FDG-PET/CT findings obtained after vaccination

	All vaccinations		First vaccination		Second vaccination	
	ALN	DM	ALN	DM	ALN	DM
Positive	101	51	83	39	18	12
Negative	136	186	119	163	17	23
Total	237	237	202	202	35	35
Percentage of subjects with positive uptake	42.6%	21.5%	41.1%	19.3%	51.4%*	34.3%**
SUVmax of positive foci (mean &SD)			2.88 ± 1.51	1.76 ± 0.58	4.79 ± 4.91†	2.17 ± 1.02‡
Range of SUVmax (Max–Min)			(7.9–1.1)	(3.2–1.0)	(21.5–1.4)	(4.9–1.2)

Compared with first vaccination

*$p = 0.25$

**$p = 0.046$

†$p = 0.004$

‡$p = 0.094$

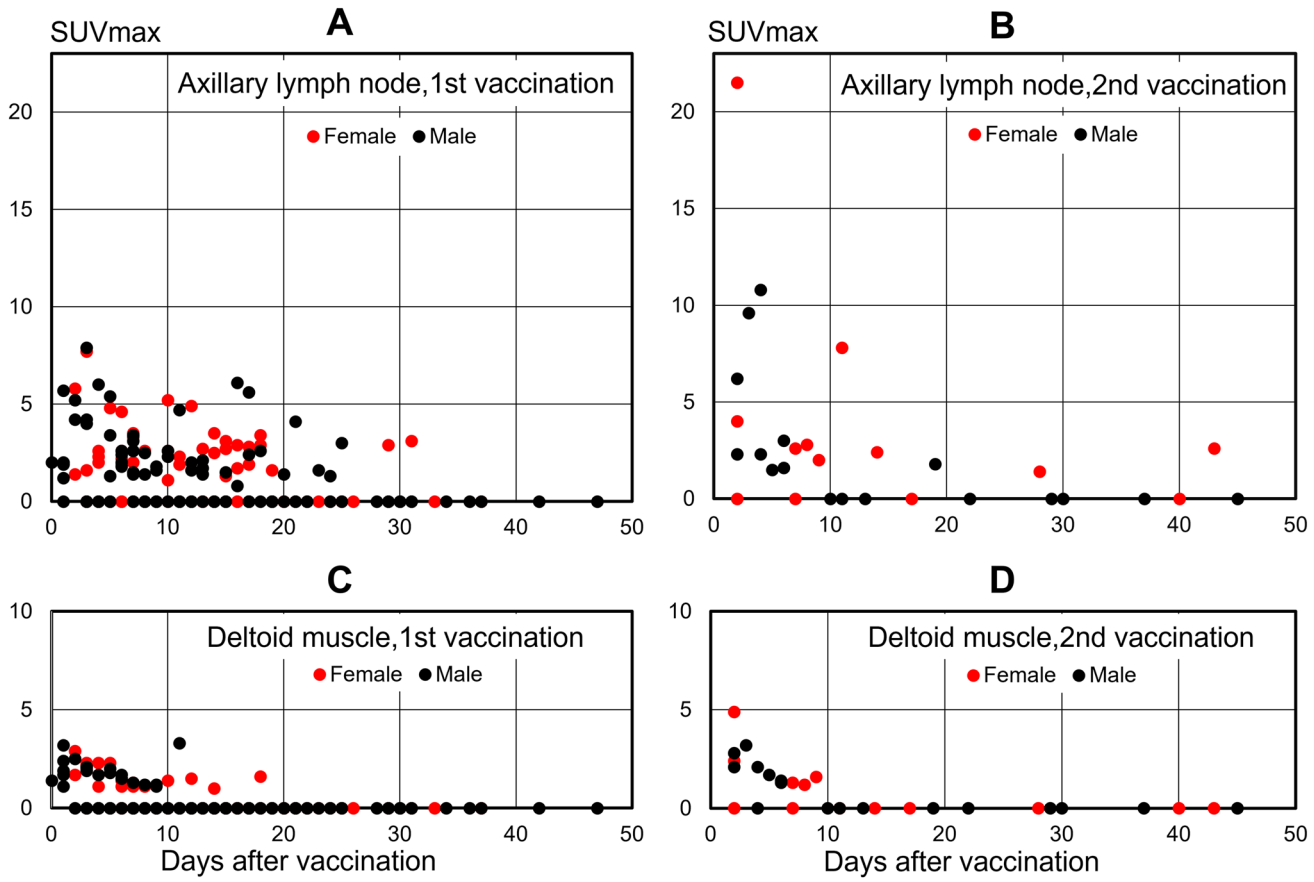

Fig. 1 SUVmax in axillary lymph nodes and deltoid muscle on FDG-PET/CT images obtained after vaccination. **A** SUVmax of axillary lymph nodes after first vaccination. **B** SUVmax of axillary lymph nodes after second vaccination. **C** SUVmax of deltoid muscle after first vaccination. **D** SUVmax of deltoid muscle after second vaccination. Please note that an SUVmax of 0 was regarded as no significant uptake, as described in the Methods

Table 5 Positive uptake rate and days after first and second vaccinations

Days after first vaccination	0–4	5–9	10–14	15–19	20–24	25–29	30–
Total subjects	31	49	47	36	17	9	13
Positive ALN	19	25	17	15	4	2	1
Positive DM	19	15	4	1	0	0	0
% positive ALN	61.3	51.0	36.2	41.7	23.5	22.2	7.7
% positive DM	61.3	30.6	8.5	2.8	0.0	0.0	0.0
Days after second vaccination	0–4	5–9	10–14	15–19	20–24	25–29	30–
Total subjects	8	7	5	2	1	2	10
Positive ALN	7	6	2	1	0	1	1
Positive DM	6	6	0	0	0	0	0
% positive ALN	87.5	85.7	40.0	50.0	0.0	50.0	10.0
% positive DM	75.0	85.7	0.0	0.0	0.0	0.0	0.0

ALN axillary lymph nodes, *DM* deltoid muscle

Table 6 Correlation between FDG uptake in axillary lymph nodes (ALN) and deltoid muscle (DM)**

	Positive ALN	Negative ALN	Total
Positive DM	49	2*	51
Negative DM	51	135	186
Total	100	137	237

*A 71-year-old male with hypopharyngeal cancer and FDG grade 1 at 1 day after his 1st vaccination and a 75-year-old female with lung cancer and FDG grade 1 at 3 days after her 1st vaccination were both DM positive and ALN negative. A specific cause for these finding could not be found in either case

**See the Results for the correlation between the SUVmax in the ALN and the DM

significantly and linearly correlated with that in the DM ($ALN = 1.069DM + 2.177$, $n = 102$, $r = 0.435$, $p < 0.001$).

Figure 2 shows typical FDG-PET/CT images of positive reactive FDG uptake in the ALN and DM after vaccination.

Analysis of contributing factors

A further analysis of possible contributing factors was performed among subjects who had received their first vaccination. Subject characteristics individually compared according to positive or negative FDG uptake in the ALN are shown in Table 7. The sex distribution, subject age, days after vaccination, and distribution of FDG-PET/CT grade differed significantly between the negative and positive ALN uptake groups. No significant differences in the site of injection (left or right), FBG, BW, or BMI were seen between the two groups.

The contributions of each factor to positive ALN uptake were analyzed using a multivariate analysis. Table 8 shows the results of a binominal logistic regression analysis for positive ALN. Significant contributing factors were a short duration since vaccination, a younger age, a female sex, and a low FDG-PET/CT grade. Please note that for the logistic regression analysis, the FDG-PET/CT grade was re-scored as follows: 0 for grades 0 and 1, and 1 for grades 2 and 3. In other words, a grade of 0 or 1 was a significant contributor to positive ALN uptake.

Table 9 shows factors contributing to positive DM uptake. Significant factors were a short duration since vaccination and a female sex. Age did not reach a significant level, and the FDG-PET/CT grade did not seem to be correlated with positive DM uptake.

An additional multivariate analysis examining contributing factors among subjects who had received either a first or a second vaccination was also performed. Significant contributing factors to positive ALN uptake were a second vaccination status, a short duration since vaccination, a younger age, and a low FDG-PET/CT grade. A female sex was not a significant contributor in this analysis. For positive DM uptake, a short duration since vaccination, a younger age, and a female sex were significant contributors. Because of the similarity of the results to those of the analysis among subjects receiving a first vaccination only, the data are not shown. The present study focused on subjects who had received a first vaccination, as this group comprised 85% of all the subjects, and limited data were available for subjects who had received a second vaccination. The subsequent discussion, therefore, will be based on the results of our analysis of subjects who had received a first vaccination.

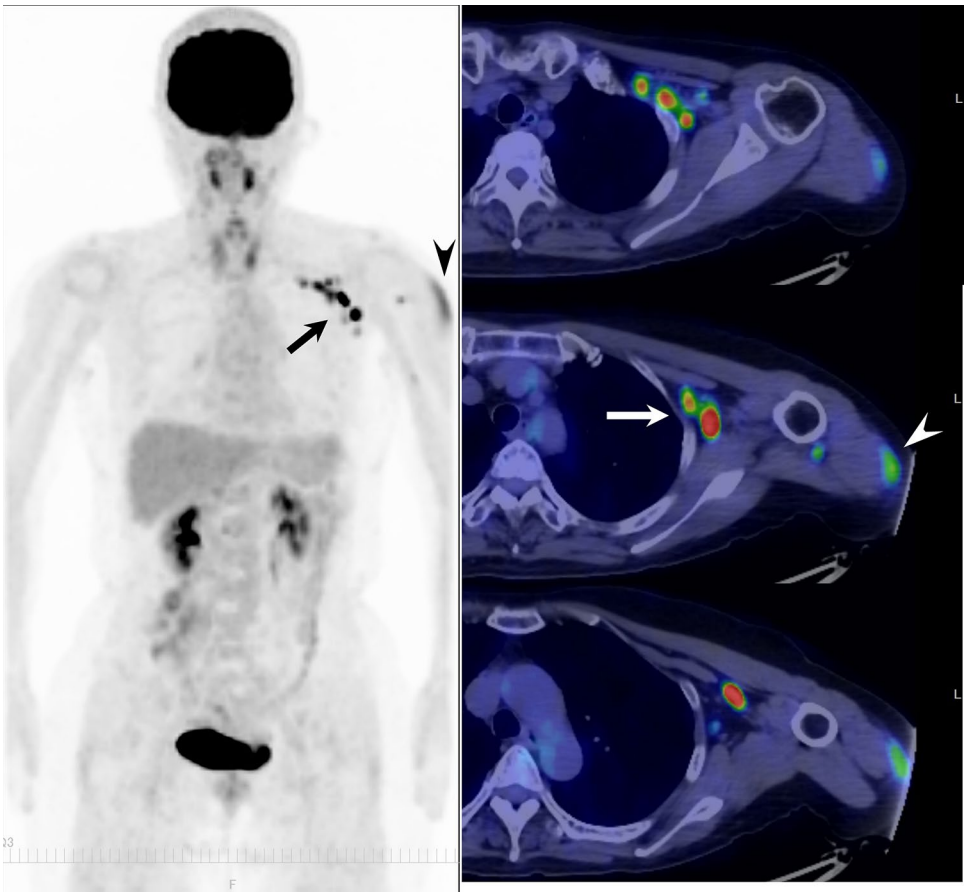

Fig. 2 Typical case of reactive FDG uptake after mRNA COVID-19 vaccination. A 64-year-old female underwent a follow-up FDG-PET/CT examination after receiving surgery for appendiceal cancer. She had received a second vaccination in her left arm 2 days before the FDG-PET/CT examination, and had complained of pain in her left shoulder and arm. The images showed a very high FDG uptake in a cluster of small left axillary lymph nodes, with normal structures, and the SUVmax was as high as 21.5 (arrow). The left deltoid muscle also showed FDG uptake with an SUVmax of 4.9 (arrowhead). In her right lower abdomen, a peritoneal nodule with FDG uptake of 3.9 was found and was suspected to be a peritoneal metastasis. The disease was determined to be Grade 1. An MIP image is shown on the left, and fusion axial images focusing on left axilla are shown on the right

Discussion

We have found that mRNA COVID-19 vaccination induced FDG uptake in the ALN and DM in about 60% of subjects even soon (0–4 days) after the first vaccination, with percentages reaching 87.5% and 75.0%, respectively, after the second vaccination. DM uptake had almost disappeared at around 2 weeks, while ALN uptake persisted for 3 weeks or longer. Large individual variations in reactive FDG uptake were observed, suggesting the contributions of various individual factors. Accordingly, various subject characteristics were analyzed, and a multivariate analysis showed that a short duration since vaccination, a younger age, a female sex, and a low FDG-PET/CT grade were significant contributors to positive ALN uptake, while a short duration since vaccination and a female sex were the only contributors to positive DM uptake. This is the first study to identify the contributions of these factors to FDG uptake in the ALN and DM after COVID-19 vaccination.

After an initial case report by Eifer and Eshet [1], several cases with FDG avid lymph adenopathy have been reported, some of which were biopsy-proven because of suspected lymph-node metastasis [2–7]. This situation has been widely recognized as a new diagnostic challenge in the era of mass vaccination with the new mRNA COVID-19 vaccines. A radiology panel report and others [8, 9] have stated that adenopathy is a frequent finding after COVID-19 vaccination, that vaccination should not be delayed because of upcoming imaging, and that imaging should be scheduled before the first or at least 6 weeks after the final vaccination, whenever possible. Meanwhile, Cohen et al. [10] reported a large cohort study from Israel in which hypermetabolic lymph nodes were observed in 45.6% of all the vaccinations group, 36.4% of the first vaccination group, and 53.9% of the second vaccination group; furthermore, a lower incidence was observed during the first 5 days or in the 3rd week after the first vaccination and beyond 20 days after the second vaccination. Therefore, they recommended these periods as the time window for imaging studies.

In our Japanese cohort, the incidence of hypermetabolic lymph nodes was higher than that in the above-mentioned report [10] after both the first vaccination and the second vaccination. Furthermore, we did not observe a lower incidence during the first 5 days; rather, the highest incidence was observed during the first 5 days. One of the differences between these two cohorts may be the vaccine dose per kg of body weight. The standard dose for the BNT162b2 vaccine from BioNTech-Pfizer is 30 µg/0.3 mL (100 µg/mL)

Table7 Subject characteristics compared individually according to negative and positive axillary lymph-node status after first vaccination

Item (mean ± SD)	Axillary lymph node		*p*
	Negative	Positive	
Total	119	83	
Female	35	38	0.017*
Male	84	45	
Age (years)	74.1 ± 8.1	69.3 ± 9.6	0.001**
Left arm	103	78	
Right arm	16	5	0.09
Days after injection	15.4 ± 9.2	10.0 ± 6.8	< 0.001**
FBG (mg/dL)	106 ± 22	105 ± 16	0.71
BW (kg)	58.0 ± 11.4	58.6 ± 11.6	0.77
BMI	22.4 ± 3.6	22.6 ± 3.5	0.71
FDG-PET/CT grade mean ± SD	1.14 ± 1.11	0.75 ± 0.99	
Grade 0	46	45	
G 1	31	23	
G 2	21	6	
G 3	21	9	0.043†

*Statistically significant, Chi-squared test

**Statistically significant, Welch *t* test

†The distributions of each grade according to negative and positive status were compared using the Chi-squared test, and a statistically significant difference was seen between these two groups

per person. Therefore, the dose is not calculated per kg of body weight. The median body weight of our cohort was 57.5 kg, and the BMI was 22.3, while we do not have data for the above-mentioned cohort, and the vaccine dose per kg may be larger in our cohort. This might be one of the reasons explaining the higher incidence of reactive lymphadenopathy in our cohort. Another possible reason may be the grade of pathological FDG uptake (FDG-PET/CT grade). In our cohort, 45.6% of the subjects had grade 0 findings (no FDG-positive lesion), and 27.6% had grade 1 findings. Thus, our cohort appeared to have relatively mild disease, compared with other cohort studies. As we showed in a multivariate analysis, a low FDG-PET/CT grade was a significant contributor to positive ALN uptake. Recently, Eshet et al. [16] reported relatively late response data in which 29% of subjects had positive ALN uptake at 7–10 weeks after a second vaccination. These findings suggest that a time window of more than 6 weeks after a second vaccination might not be appropriate for imaging studies.

We agree with the report by Shah et al. [4] that information regarding vaccination history, site of injection, and the nature of vaccine-associated lymph adenopathy is important; furthermore, clusters of small lymph nodes with a normal structure (preserved fat in the hilum of the lymph node) may help prevent misinterpretations of reactive lymphadenopathy. However, Cohen et al. [10] reported that they could not differentiate vaccine-associated lymphadenopathy from metastatic lymphadenopathy in 14.8% of vaccinated patients. Breast cancer and lymphoma were the leading diseases in this category, but no significant differences among various types of diseases were noted in their cohort. Thus, the differentiation of reactive lymph nodes may vary depending on the patient's disease status.

The distribution of injection sites in our study was 11.8% in the right arm and 88.2% in the left arm, since most subjects wished to be vaccinated on their non-dominant side. The percentage of left-dominant subjects is reportedly about 10% of the general population [17]. We had one case with breast cancer and one case with lung cancer and ALN metastasis who were vaccinated ipsilaterally to their diseases. The FDG-PET/CT images showed a mixture of vaccine-associated and metastatic lymphadenopathy, and these two cases were excluded from the analysis. Macintosh et al. [9] recommended that vaccine administration should be performed contralateral to a unilateral cancer to avoid confounding FDG uptake on the side with cancer. In our study,

Table 8 Results of multivariate analysis (binominal logistic regression analysis): factors contributing to a positive axillary lymph-node status after first vaccination

Variables	Odds ratio	95% confidence interval	*p* value
Left (0) or right (1) arm	0.381	0.111–1.313	0.126
Days after first vaccination	0.892	0.848–0.937	< 0.001*
Age (years)	0.913	0.869–0.960	< 0.001*
Female (0) or male (1)	0.357	0.129–0.986	0.047*
FBG (mg/dL)	1.007	0.991–1.024	0.401
BW (kg)	1.012	0.938–1.092	0.761
BMI	0.966	0.785–1.189	0.746
FDG-PET/CT grade**	0.413	0.190–0.899	0.026*

Considering the number of data points, an analysis of the first vaccination group only is shown. See the text for the analysis of all the subjects

*Statistically significant

**Re-scored as Score 0: grades 0 and 1, and Score 1: grades 2 and 3

Table 9 Factors contributing to positive deltoid muscle status after first vaccination

Variables	Odds ratio	95% confidence interval	*p* value
Left (0) or right (1) arm	0.366	0.036–3.707	0.395
Days after first vaccination	0.665	0.577–0.767	< 0.001*
Age (years)	0.945	0.891–1.002	0.057
Female (0) or male (1)	0.164	0.035–0.765	0.021*
FDG-PET/CT grade **	0.932	0.307–2.834	0.902

*Statistically significant

**Re-scored as Score 0: grades 0 and 1, and Score 1: grades 2 and 3

Data for FBS, BW, and BMI were omitted because of the absence of a significant contribution, similar to Table 6

9 patients with breast cancer, a fibrosarcoma, or a melanoma on the arm were vaccinated contralateral to their disease, and vaccine-associated ALN was successfully evaluated in these patients.

The mechanism of FDG uptake by the ALN has not been clarified. Eifer et al. and Cohen et al. both showed that a low FDG uptake by the lymph nodes was associated with an older age, while Eifer et al. reported an association with immunosuppressive treatment and hematologic diseases [12, 18]. In our study, the correlation of a younger age and a low FDG-PET/CT grade to positive ALN was consistent with the findings of these previous reports. We also showed, for the first time, that a female sex was a significant contributor to positive ALN uptake.

FDG uptake in DM was strongly correlated with the number of days since vaccination and with a female sex, but it was not related to the FDG-PET/CT grade. Eifer et al. [12] reported that age nor immunosuppressive treatment, etc. was not correlated to DM uptake. These findings were consistent with those of our study, but our study is the first to report an association between FDG uptake in the DM and a female sex.

Maeda et al. [19] reported a higher incidence of adverse reactions, including muscle pain, in females than in males, and this outcome may be consistent with our results. FDG uptake in the DM might reflect an inflammatory reaction that precedes lymph-node activation for antibody production. A recent study examining Japanese healthcare workers showed that the BNT162b2-elicited SARS-Cov-2-neutralizing antibody activities and IgG levels on day 28 after the first vaccination and the pain scores following the second vaccination were greater in woman than in men [19]. Seban et al. [20] considered vaccine-induced FDG uptake to be a marker of immune reaction. Turner et al. [21] reported that after vaccination, a persistent germinal center B-cell response was induced in the ALN, enabling the generation of humoral immunity. An elevation in glucose metabolism in the lymph nodes may reflect the immune reaction evoked by the mRNA vaccine, which probably involves antibody production. This hypothesis may explain the higher incidence of positive ALN uptake in females, younger subjects, and subjects with low FDG-PET/CT grades. However, a direct correlation between antibody production and vaccine-associated FDG uptake by lymph nodes has not yet been demonstrated.

Skawran et al. [13] reported that vaccination-induced FDG uptake in the ALN was more frequent for the mRNA-1273 vaccine (Moderna) than for the BNT162b2 vaccine (Pfizer). Puranik et al. [22] compared the differences of these two full-length Spike protein-encoding mRNA vaccines, which are both highly effective against COVID-19 infection and preventing hospitalization, with a slightly higher effectiveness observed for the Moderna vaccine. The Pfizer vaccine is administered as two 30 µg/0.3 mL (100 µg/mL) doses administered 21 days apart, and the Moderna vaccine is administered as two 100 µg/0.5 mL (200 µg/mL) doses administered 28 days apart. This means that the Moderna vaccine provides three times more mRNA copies of the Spike protein than the Pfizer vaccine, which could result in a more effective priming of the immune response. Also, adverse effects, such as myalgia and arthralgia, were observed more frequently after vaccination with the Moderna vaccine than after the Pfizer vaccine, and it can be speculated that this increased reactogenicity is paralleled by an increased immunogenicity. There are differences in the lipid composition of the nanoparticles used for packaging the mRNA content of the Moderna and Pfizer vaccines, but the influence of these differences seems to be small. The different doses of mRNA may explain the differences in the frequency of FDG uptake in the ALN after vaccination with the Pfizer and Moderna vaccines. These findings may support our assumption that FDG uptake in the ALN after vaccination reflects the immune response to mRNA vaccination, rather than being a non-specific inflammatory reaction.

This study did not intend to provide new diagnostic criteria for the differentiation of vaccine-associated FDG uptake in the ALN from FDG uptake in metastatic lymph nodes. If readers combine the known characteristics of vaccine-associated FDG uptake in ALN and our results, their understanding of vaccine-associated events will be deepened, possibly aiding diagnostic considerations. FDG-PET/CT is not just

an image, but a mirror of physiology, and understanding this nature is essential for improving diagnosis. The COVID-19 mRNA vaccine is the first mRNA medicine to be applied clinically, and many new vaccines and therapeutic molecules using mRNA techniques are under investigation with the aim of future applications [23]. Future response evaluations using FDG-PET/CT may refer to the mRNA vaccine-associated events described in this study.

Limitations

Our analysis focused on subjects who had received their first vaccination. Because the subject enrollment was performed during the early phase of a local vaccination program, the number of subjects who had received a second vaccination was insufficient. The incidence of FDG uptake in the ALN after vaccination is reportedly higher after the second vaccination than after the first vaccination, and our limited data confirmed this finding. Further data and analysis including sufficient numbers of subjects who have received a first vaccination as well as those who have received a second vaccination might yield additional knowledge.

Our cohort included mostly elderly subjects (median age, 72 years; range, 29–94 years) because of the prioritization of vaccination for the elderly and for medical worker. The inclusion of young subjects in future studies might yield new information.

As described in the Subjects section, two patients with ALN metastasis and FDG uptake were excluded from the study population. All the ALN findings were regarded as vaccine-associated FDG uptake, and nodular uptake or a cluster of small hot spots was not regarded as indicating a malignancy. No biopsies were performed, because both the radiologist and the physician in charge did not consider a biopsy to be necessary. Actually, many of the cancer patients were followed in our hospital after the study period, and no cases with ALN metastasis were observed. However, the lack of follow-up in some of the subjects is a study limitation.

This study was a retrospective analysis, which is a limitation of this study. Future prospective studies including an analysis of the production of neutralizing antibodies to the COVID-19 virus would be useful for elucidating the in vivo vaccine-reaction mechanism.

Conclusion

We observed a high incidence of positive FDG uptake in the ALN and DM during FDG-PET/CT examinations performed after mRNA COVID-19 vaccination. A short duration since vaccination, a younger age, a female sex, and a low FDG-PET/CT grade contributed significantly to positive ALN uptake, while a short duration since vaccination and a female sex contributed to positive DM uptake. After vaccination, an acute inflammatory reaction in the DM followed by an immune reaction in the ALN linked to humoral immunity may be speculated.

Appendix

Spleen uptake

Elevated FDG uptake in the spleen associated with COVID-19 vaccination has been reported by Steinberg [2] and Seban [20]. However, its incidence has never been reported. We additionally analyzed the incidence of FDG uptake in the spleen in our cohort. We defined an elevated splenic FDG uptake as a mean SUV in the spleen that was higher than the mean SUV of the liver in a representative axial slice of an FDG-PET/CT image. We observed elevated splenic FDG uptake in 6 out of 237 subjects (2.5%). All 6 had positive ALN; splenic uptake was observed in 6% of all the subjects with positive ALN. The mean S/L ratio was 1.10; there were 5 females and 1 male, and the mean age was 69 years. Two subjects had received their first vaccination, and 4 had received their second. Four subjects had FDG-PET grade 0, one had grade 1, and one had grade 3. The subject characteristics were similar to those observed for subjects with positive ALN. Elevated FDG uptake in the spleen after vaccination seems to be uncommon, and its contributing factors remain to be clarified.

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Declarations

Conflict of interest Kiichi Ishiwata (Fukushima Medical University) has received an endowment from the Southern TOHOKU Research Institute for Neuroscience. The other authors have no conflicts of interest related to this article.

References

1. Eifer M, Eshet Y. Imaging of COVID-19 vaccination at FDG PET/CT. Radiology. 2021;299(2):E248. https://doi.org/10.1148/radiol.2020210030.
2. Steinberg J, Thomas A, Iravani A. F-fluorodeoxyglucose PET/CT findings in a systemic inflammatory response syndrome after

COVID-19 vaccine. Lancet. 2021;397(10279):e9. https://doi.org/10.1016/S0140-6736(21)00464-5.
3. Nawwar AA, Searle J, Hagan I, Lyburn ID. COVID-19 vaccination induced axillary nodal uptake on [18F]FDG PET/CT. Eur J Nucl Med Mol Imaging. 2021;48(8):2655–6. https://doi.org/10.1007/s00259-021-05274-7.
4. Shah S, Wagner T, Nathan M, Szyszko T. COVID-19 vaccine-related lymph node activation - patterns of uptake on PET-CT. BJR Case Rep. 2021;7(3):20210040. https://doi.org/10.1259/bjrcr.20210040.
5. Brown AH, Shah S, Groves AM, Wan S, Malhotra A. The challenge of staging breast cancer with PET/CT in the era of COVID vaccination. Clin Nucl Med. 2021. https://doi.org/10.1097/RLU.0000000000003683.
6. Xu G, Lu Y. COVID-19 mRNA vaccination-induced lymphadenopathy mimics lymphoma progression. Clin Nucl Med. 2021;46(4):353–4.
7. Ahmed N, Muzaffar S, Binns C, Llyas MW, Usmani S. COVID-19 vaccination manifesting as incidental lymph nodal upatake on 18F-FDG PET/CT. Clin Nucl Med. 2021;46(5):435–6.
8. Becker AS, Perez-Johnston R, Chikarmane SA, Chen MM, Homsi ME, Feigin KN, et al. Multidisciplinary recommendations regarding post-vaccine adenopathy and radiologic imaging: radiology scientific expert panel. Radiology. 2021;300(2):E323–7. https://doi.org/10.1148/radiol.2021210436.
9. McIntosh LJ, Bankier AA, Vijayaraghavan GR, Licho R, Rosen MP. COVID-19 vaccination-related uptake on FDG PET/CT: an emerging dilemma and suggestions for management. AJR Am J Roentgenol. 2021;4:1–9. https://doi.org/10.2214/AJR.21.25728.
10. Cohen D, Krauthammer SH, Wolf I, Even-Sapir E. Hypermetabolic lymphadenopathy following administration of BNT162b2 mRNA Covid-19 vaccine: incidence assessed by [^{18} F]FDG PET-CT and relevance to study interpretation. Eur J Nucl Med Mol Imaging. 2021;48(6):1854–63. https://doi.org/10.1007/s00259-021-05314-2.
11. Bernstine H, Priss M, Anati T, Turko O, Gorenberg M, Steinmetz AP, Groshar D. Axillary lymph nodes hypermetabolism after BNT162b2 mRNA COVID-19 vaccination in cancer patients undergoing 18F-FDG PET/CT: a cohort study. Clin Nucl Med. 2021;46(5):396–401. https://doi.org/10.1097/RLU.0000000000003648.
12. Eifer M, Tau N, Alhoubani Y, Kanana N, Domachevsky L, Shams J, et al. COVID-19 mRNA vaccination: age and immune status and its association with axillary lymph node PET/CT uptake. J Nucl Med. 2021. https://doi.org/10.2967/jnumed.121.262194.
13. Skawran S, Gennari AG, Dittli M, Treyer V, Muehlematter UJ, Maurer A, et al. [^{18} F]FDG uptake of axillary lymph nodes after COVID-19 vaccination in oncological PET/CT: frequency, intensity, and potential clinical impact. Eur Radiol. 2021;22:1–9. https://doi.org/10.1007/s00330-021-08122-2.
14. Minamimoto R, Kiyomatsu Y. Effects of COVID-19 vaccination on FDG-PET/CT imaging: a literature review. Glob Health Med. 2021;3(3):129–33.
15. Japanese Society of Nuclear Medicine (2020) Clinical guideline for FDG-PET, PET-CT 2020. (in Japanese)
16. Eshet Y, Tau N, Alhoubani Y, Kanana N, Domachevsky L, Eifer M. Prevalence of increased FDG PET/CT axillary lymph node uptake beyond 6 weeks after mRNA COVID-19 vaccination. Radiology. 2021;27: 210886. https://doi.org/10.1148/radiol.2021210886.
17. Hardyck C, Petrinovich LF. Left-handedness. Psychol Bull. 1977;84:385–404.
18. Cohen D, Hazut Krauthammer S, Cohen YC, Perry C, Avivi I, Herishanu Y, Even-Sapir E. Correlation between BNT162b2 mRNA Covid-19 vaccine-associated hypermetabolic lymphadenopathy and humoral immunity in patients with hematologic malignancy. Eur J Nucl Med Mol Imaging. 2021;8:1–10. https://doi.org/10.1007/s00259-021-05389-x.
19. Maeda K, Amano M, Uemura Y, Tsuchiya K, Matsushima T, Noda K, et al. Correlates of neutralizing/SARS-CoV-2-S1-binding antibody response with adverse effects and immune kinetics in BNT162b2-vaccinated individuals. medRxiv. 2021. https://doi.org/10.1101/2021.07.27.21261237 (**Preprint**).
20. Seban RD, Champion L, Yeh R, Schwartz LH, Dercle L. Assessing immune response upon systemic RNA vaccination on [18F]-FDG PET/CT for COVID-19 vaccine and then for immuno-oncology? Eur J Nucl Med Mol Imaging. 2021;23:1–2. https://doi.org/10.1007/s00259-021-05468-z.
21. Turner JS, O'Halloran JA, Kalaidina E, Kim W, Schmitz AJ, Zhou JQ, et al. SARS-CoV-2 mRNA vaccines induce persistent human germinal centre responses. Nature. 2021;596(7870):109–13. https://doi.org/10.1038/s41586-021-03738-2.
22. Puranik A, Lenehan P, Silvert E, Niesen M, Corchado-Garcia J, O'Horo J, et al. Comparison of two highly-effective mRNA vaccines for COVID-19 during periods of alpha and delta variant prevalence. medRxiv. 2021. https://doi.org/10.1101/2021.08.06.21261707 (**preprint**).
23. Weng Y, Li C, Yang T, Hu B, Zhang M, Guo S, et al. The challenge and prospect of mRNA therapeutics landscape. Biotechnol Adv. 2020;40: 107534. https://doi.org/10.1016/j.biotechadv.2020.107534.