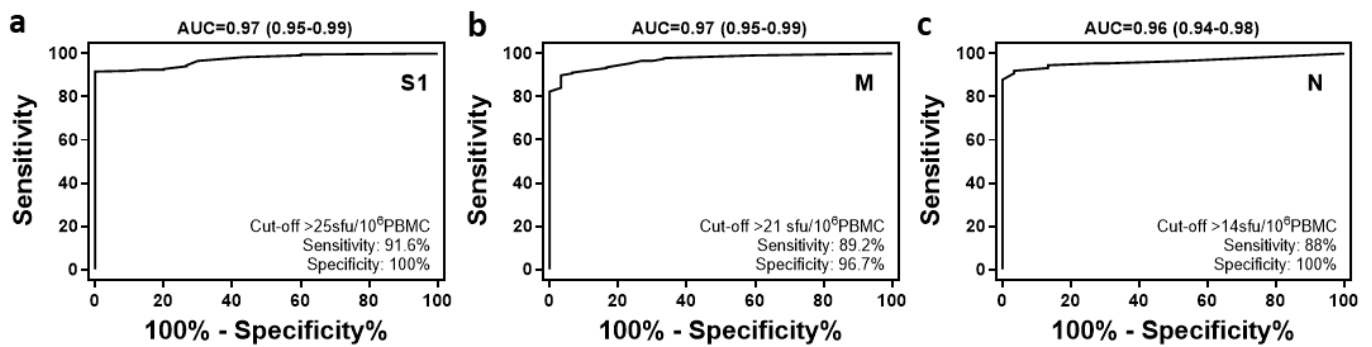


Supplemental Digital Content

SDC Methods

Diagnostic accuracy in terms of sensitivity and specificity of the cut-off values for IFN- γ -producing SFUs/ 10^6 PBMCs applied to define positive responses to different viral antigens in the IFN- γ FluoroSpot assay: **(a)** S1 domain of the spike (S) glycoprotein, **(b)** membrane (M) protein, and **(c)** nucleoprotein (N). AUC: area under the receiver operating characteristic curve; IFN- γ : interferon- γ ; PBMC: peripheral blood mononuclear cell; SFU: spot forming unit.



SDC Results

Table S1. Correlation between laboratory values at the baseline (pre-vaccination) assessment, and the number of S protein-specific IFN- γ -producing SFUs per 10^6 PBMCs by the IFN- γ FluoroSpot assay and the serum neutralizing titers against the S protein by the hACE-2/spike antibody inhibition method, both measured at two weeks after the completion of the full vaccine series.

	N	S protein-specific IFN- γ -producing SFUs/ 10^6 PBMCs after vaccination		Neutralizing titers against SARS-CoV-2 S protein after vaccination	
		Spearman's rho correlation coefficient	<i>P</i> -value ^a	Spearman's rho correlation coefficient	<i>P</i> -value ^a
Total CD3 ⁺ T-cell count	38	0.399	0.013	0.229	0.167
Total CD4 ⁺ T-cell count	38	0.369	0.023	0.063	0.707
Total CD8 ⁺ T-cell count	38	0.275	0.095	0.358	0.027
B-cell count	38	0.143	0.391	0.099	0.555
NK cell count	38	-0.156	0.349	0.135	0.419
IgG level	40	-0.037	0.819	0.014	0.931
IgA level	40	-0.135	0.406	-0.093	0.567
IgM level	40	-0.288	0.071	-0.178	0.272
eGFR	42	0.156	0.324	0.375	0.014
Tacrolimus trough level	35	-0.089	0.612	-0.058	0.739
MPA trough level	10	-0.018	0.960	-0.365	0.299
mTOR inhibitor trough level	8	-0.263	0.528	-0.342	0.406

eGFR: estimated glomerular filtration rate (MDRD-4 equation); IFN- γ : interferon- γ ; MPA: mycophenolic acid; mTOR: mammalian target of rapamycin; PMBC: peripheral blood mononuclear cell; S: SARS-CoV-2 spike glycoprotein; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; SFU: spot forming unit.

^a Bold characters denote significant *P*-values.

Table S2. *Sensitivity analysis excluding SOT recipients with natural immunity before vaccination (n = 3):* Comparison of clinical characteristics between recipients that developed or not SARS-CoV-2-specific cell-mediated immunity (>25 S protein-specific IFN- γ -producing SFUs/10⁶ PBMCs) or detectable serum neutralizing activity against the S protein (hACE-2/spike antibody inhibition ELISA-based method) at two weeks after the completion of the full mRNA-1273 vaccine series.

Variable	SARS-CoV-2-specific cell-mediated immunity			Serum neutralizing activity against SARS-CoV-2 S protein		
	Vaccine response (n = 22)	No vaccine response (n = 17)	P-value	Vaccine response (n = 11)	No vaccine response (n = 28)	P-value
Age of recipient, years [mean \pm SD]	51.8 \pm 12.5	52.0 \pm 10.6	0.960	50.5 \pm 12.3	52.5 \pm 11.5	0.637
Gender of recipient (male) [n (%)]	14 (63.6)	10 (58.8)	0.759	6 (54.5)	18 (64.3)	0.574
Smoking habit [n (%)]	5 (22.7)	2 (11.8)	0.438	2 (18.2)	5 (17.9)	1.000
Comorbidities [n (%)]						
Hypertension	15 (68.2)	14 (82.4)	0.315	7 (63.6)	22 (78.6)	0.424
Dyslipidemia	5 (22.7)	5 (29.4)	0.635	3 (27.3)	7 (25.0)	1.000
Diabetes mellitus	3 (13.6)	6 (35.3)	0.111	3 (27.3)	6 (21.4)	0.693
Cardiovascular disease	9 (40.9)	4 (23.5)	0.254	5 (45.5)	8 (28.6)	0.453
Obesity	3 (13.6)	3 (17.6)	1.000	2 (18.2)	4 (14.3)	1.000
Chronic pulmonary disease	2 (9.1)	0 (0.0)	0.495	1 (9.1)	1 (3.6)	0.490
Venous thromboembolic disease	4 (18.2)	3 (17.6)	1.000	1 (9.1)	6 (21.4)	0.649
Ethnicity other than Caucasian [n (%)]	3 (13.6)	3 (17.6)	1.000	11 (100.0)	22 (78.6)	0.158
Type of transplantation [n (%)]			0.198			0.022
Kidney (including double organ)	14 (63.6)	14 (82.4)		5 (45.5)	23 (82.1)	
Liver	8 (36.4)	3 (17.6)		6 (54.5)	5 (17.9)	

Previous solid organ transplantation [n (%)]	2 (9.1)	3 (17.6)	0.636	3 (27.3)	2 (7.1)	0.125
Time interval since transplantation, years [median (IQR)]	2.6 (1.3 – 5.6)	1.9 (0.8 – 3.2)	0.092	2.9 (1.9 – 3.4)	1.9 (0.8 – 5.2)	0.346
Immunosuppression regimen containing [n (%)] ^a						
Tacrolimus	18 (81.8)	16 (94.1)	0.363	9 (81.8)	25 (89.3)	0.609
Trough serum levels, ng/mL [mean ± SD]	8.2 ± 3.7	9.1 ± 2.7	0.430	7.8 ± 4.6	8.9 ± 2.6	0.384
MMF/MPS	17 (77.3)	11 (64.7)	0.387	6 (54.5)	22 (78.6)	0.234
Trough serum levels, ng/mL [mean ± SD]	4.2 ± 2.4	3.9 ± 1.9	0.899	3.6 ± 1.9	4.3 ± 2.3	0.714
mTOR inhibitor	4 (18.2)	3 (17.6)	1.000	3 (27.3)	4 (14.3)	0.379
Trough serum levels, ng/mL [mean ± SD]	6.4 ± 1.9	6.6 ± 4.4	0.940	6.7 ± 2.0	6.1 ± 3.7	0.779
Prednisone	15 (68.2)	15 (88.2)	0.251	6 (54.5)	24 (85.7)	0.038
Laboratory and immunological parameters [mean ± SD] ^a						
eGFR, mL/min/1.73 m ²	58.7 ± 20.6	54.6 ± 20.7	0.539	67.7 ± 22.9	52.7 ± 18.1	0.037
CD3 ⁺ T-cell count, cells/μL	1049 ± 488	738 ± 382	0.039	1112 ± 492	846 ± 446	0.129
CD4 ⁺ T-cell count, cells/μL	542 ± 302	334 ± 208	0.032	478 ± 292	447 ± 285	0.777
CD8 ⁺ T-cell count, cells/μL	470 ± 291	368 ± 233	0.269	595 ± 314	364 ± 225	0.019
B-cell count, cells/μL	123 ± 88	108 ± 90	0.616	142 ± 116	107 ± 74	0.301
NK cell count, cells/μL	172 ± 136	224 ± 187	0.341	193 ± 114	194 ± 175	0.983
Serum IgG levels, mg/dL	1043 ± 268	1109 ± 303	0.496	1063 ± 237	1075 ± 300	0.912
Serum IgA levels, mg/dL	233 ± 110	302 ± 140	0.100	216 ± 106	280 ± 131	0.176
Serum IgM levels, mg/dL	94 ± 72	146 ± 87	0.056	91 ± 43	126 ± 92	0.257

eGFR: estimated glomerular filtration rate (MDRD-4 equation); IQR: interquartile range; MMF/MPS: mycophenolate mofetil or mycophenolate sodium; mTOR: mammalian target of rapamycin; NK: natural killer; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2; SD: standard deviation.

^a At the time of the administration of the first dose of the mRNA-1273 vaccine.

Table S3. Sensitivity analysis excluding SOT recipients with natural immunity before vaccination

(n = 3): Comparison of the number of S protein-specific IFN- γ -producing SFUs per 10⁶ PBMCs assessed by the IFN- γ FluoroSpot assay and serum neutralizing titers against the S protein determined with a hACE-2/spike antibody inhibition ELISA-based method at two weeks after the completion of vaccination by type of transplantation and immunosuppression regimen.

Variable [n]	S protein-specific IFN- γ -producing SFUs per 10 ⁶ PBMCs [median (IQR)]	P-value	Serum neutralizing titers against the S protein (1/dilution) [median (IQR)]	P-value
Type of transplantation		0.078		0.046
Kidney (n = 28)	25.8 (8.3 – 94.2)		0.0 (0.0 – 0.0)	
Liver (n = 11)	175.0 (15.0 – 1116.7)		32.0 (0.0 – 90.0)	
Immunosuppression regimen containing tacrolimus		0.054		0.887
Yes (n = 34)	29.2 (7.9 – 120.0)		0.0 (0.0 – 32.3)	
No (n = 5)	190.0 (94.2 – 691.7)		0.0 (0.0 – 22.0)	
Immunosuppression regimen containing prednisone		0.054		0.100
Yes (n = 30)	25.8 (7.9 – 102.1)		0.0 (0.0 – 0.0)	
No (n = 9)	175.0 (23.3 – 1151.7)		32.0 (0.0 – 71.0)	
Immunosuppression regimen containing an antimetabolite		0.145		0.185
Yes (n = 29)	40.0 (10.8 – 186.7)		0.0 (0.0 – 0.0)	
No (n = 10)	21.7 (1.7 – 90.0)		15.0 (0.0 – 57.3)	
Immunosuppression regimen containing mTOR inhibitor		0.507		0.629
Yes (n = 7)	28.3 (1.7 – 180.0)		0.0 (0.0 – 32.0)	
No (n = 32)	35.8 (9.6 – 172.1)		0.0 (0.0 – 22.5)	
No. of immunosuppressive agents		0.124		0.346
One- or two-drug regimen (n = 11)	155.0 (15.0 – 483.3)		0.0 (0.0 – 39.0)	
Triple regimen (n = 28)	25.8 (7.1 – 106.3)		0.0 (0.0 – 0.0)	

IFN- γ : interferon- γ ; mTOR: mammalian target of rapamycin; PMBC: peripheral blood mononuclear cell; S: SARS-CoV-2 spike glycoprotein; SFU: spot forming unit.

Figure S1. Polyfunctional SARS-CoV-2-specific cell-mediated responses expressed as S protein-specific IL-2/IFN- γ -producing SFUs at baseline (pre-vaccination), after the first dose of the mRNA-1273 vaccine (4 weeks apart), and at two weeks after the completion of the full vaccine series in SOT recipients. No cut-off value for assay positivity was established. Data were \log_{10} transformed for graphical representation. Red bars and whiskers represent median values and interquartile ranges, respectively. Comparisons between repeated measures were performed with the Wilcoxon signed-rank test. IFN- γ : interferon- γ ; IL-2: interleukin-2; PMBC: peripheral blood mononuclear cell; S: SARS-CoV-2 spike glycoprotein; SFU: spot forming unit.

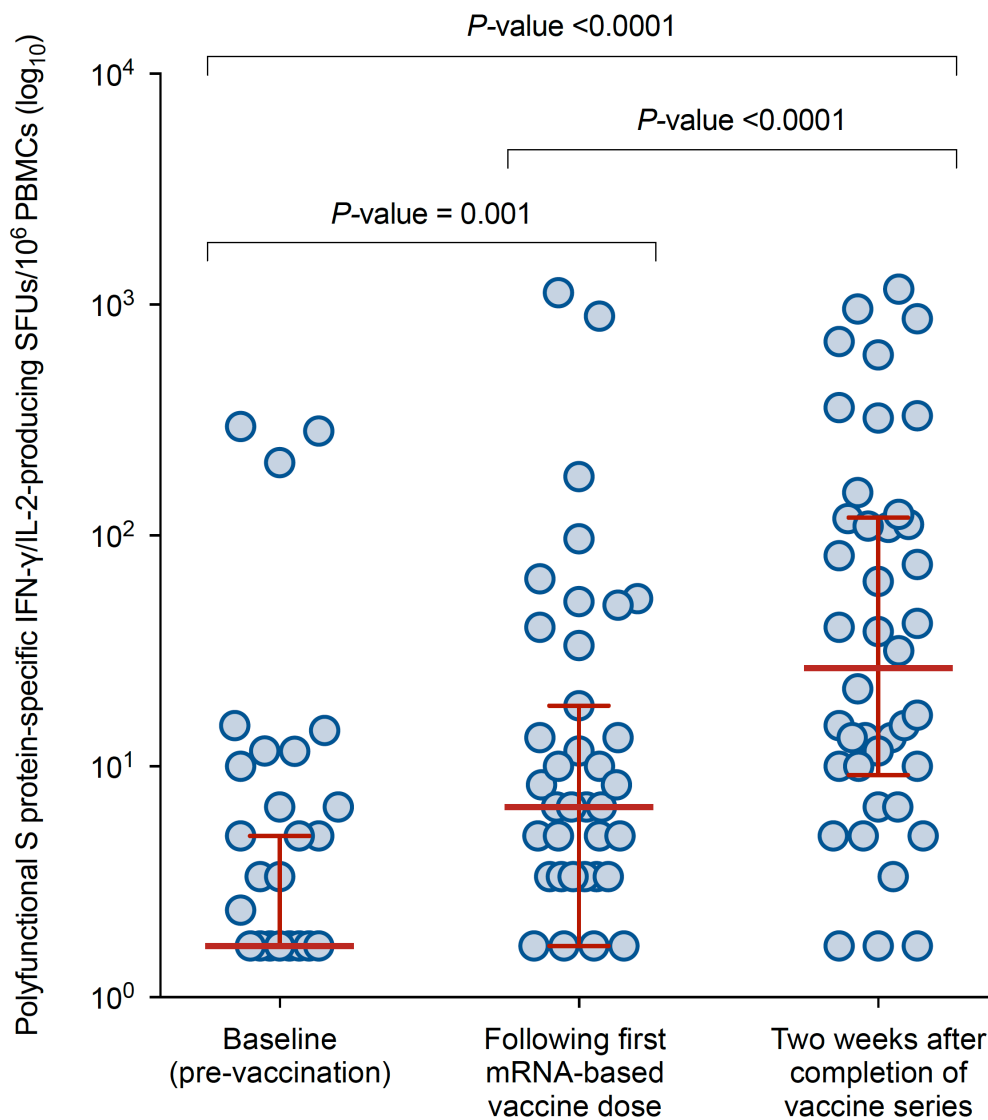
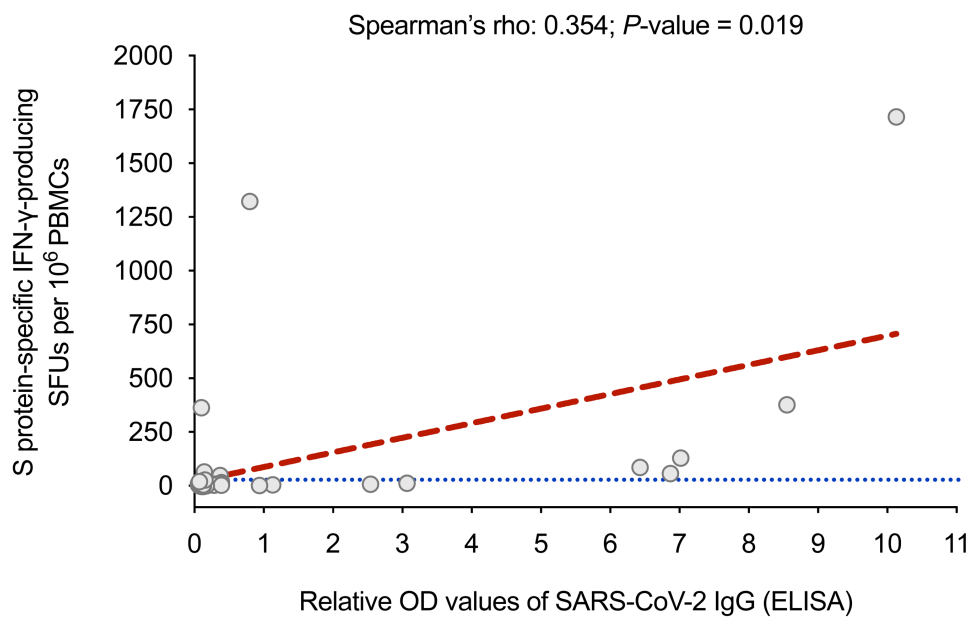


Figure S2. Correlation between the number of S protein-specific IFN- γ -producing SFUs per 10^6 PBMCs assessed by the IFN- γ FluoroSpot assay and semiquantitative results of SARS-CoV-2 IgG ELISA expressed as the relative OD (i.e. ratio of the OD value of the sample over the OD value of the calibrator) in SOT recipients: **(a)** after the first dose of the mRNA-1273 vaccine; **(b)** at two weeks after the completion of the full vaccine series. The cut-off value for positivity in the IFN- γ FluoroSpot assay (>25 SFUs/ 10^6 PBMCs) is denoted by the blue dotted line. IFN- γ : interferon- γ ; OD: optical density; PMBC: peripheral blood mononuclear cell; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2.

a)



b)

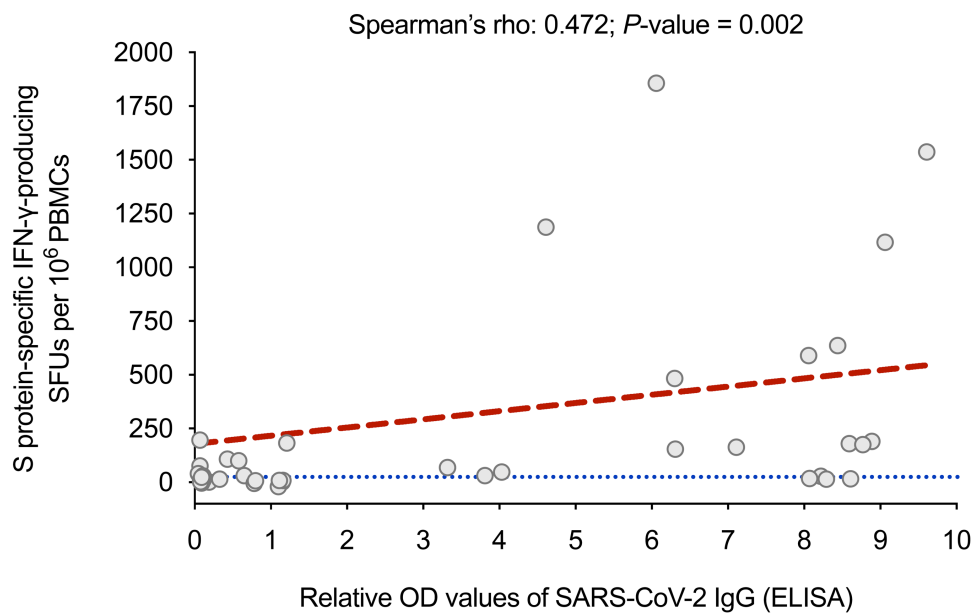


Figure S3 [next page]. Comparison of the number of S protein-specific IFN- γ -producing SFUs per 10^6 PBMCs assessed by the IFN- γ FluoroSpot assay at two weeks after the completion of full vaccine series according to: **(a)** transplant type; **(b)** use of tacrolimus; **(c)** use of prednisone; **(d)** use of antimetabolite; **(e)** use of mTOR inhibitor; and **(f)** number of immunosuppressive agents. The cut-off value for assay positivity (>25 SFUs/ 10^6 PBMCs) is denoted by the blue dotted line. Data were \log_{10} transformed for graphical representation. Red bars and whiskers represent median values and interquartile ranges, respectively. IFN- γ : interferon- γ ; mTOR: mammalian target of rapamycin; PMBC: peripheral blood mononuclear cell; S: SARS-CoV-2 spike glycoprotein; SFU: spot forming unit.

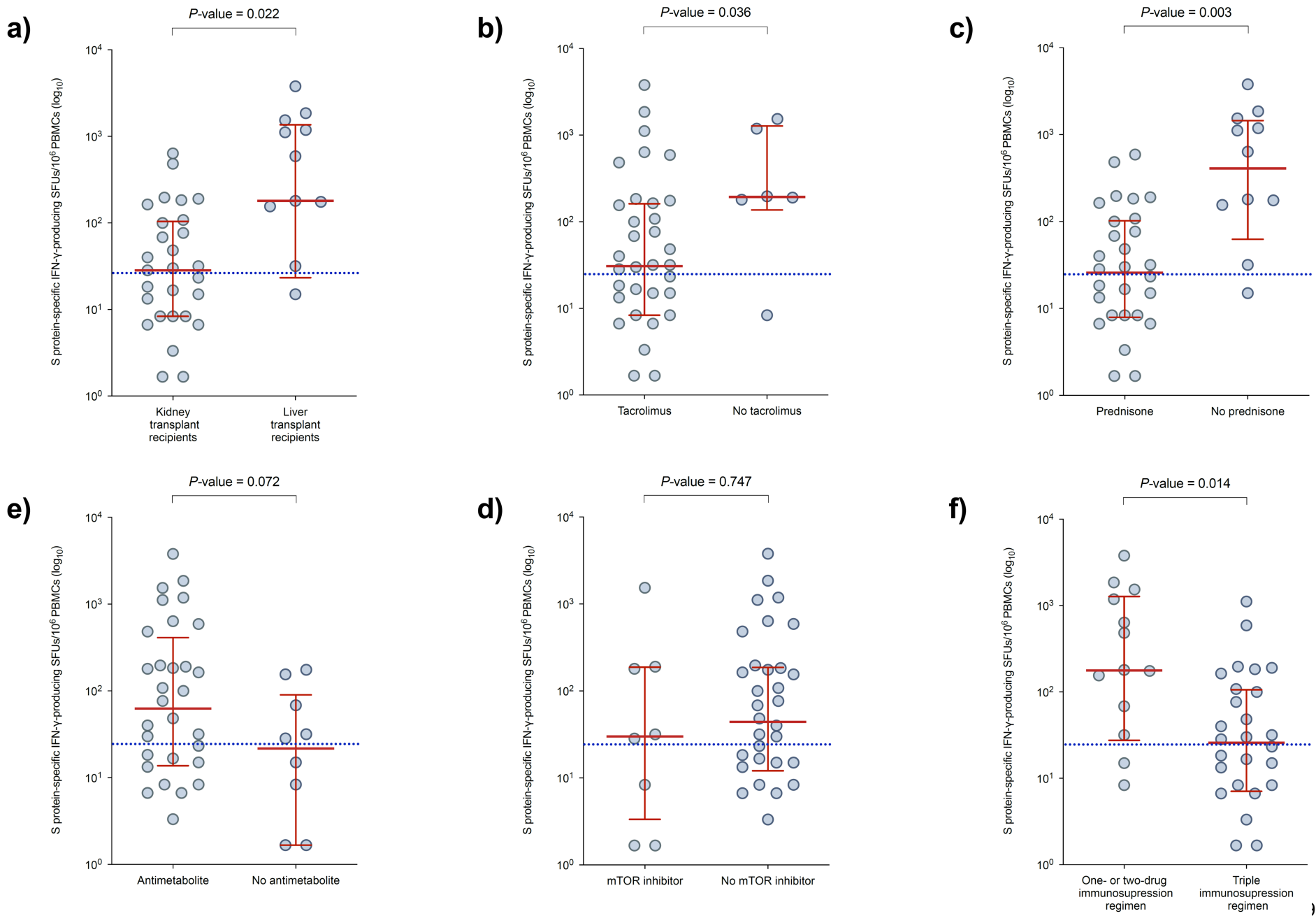


Figure S4 [next page]. Comparison of serum neutralizing titers against the SARS-CoV-2 S protein determined with a hACE-2/spike antibody inhibition ELISA-based method at two weeks after the completion of vaccination according to: **(a)** transplant type; **(b)** use of tacrolimus; **(c)** use of prednisone; **(d)** use of antimetabolite; **(e)** use of mTOR inhibitor; and **(f)** number of immunosuppressive agents. The cut-off value for assay positivity ($>1/10$) is denoted by the blue dotted line. Red bars and whiskers represent median values and interquartile ranges, respectively. mTOR: mammalian target of rapamycin; S: SARS-CoV-2 spike glycoprotein.

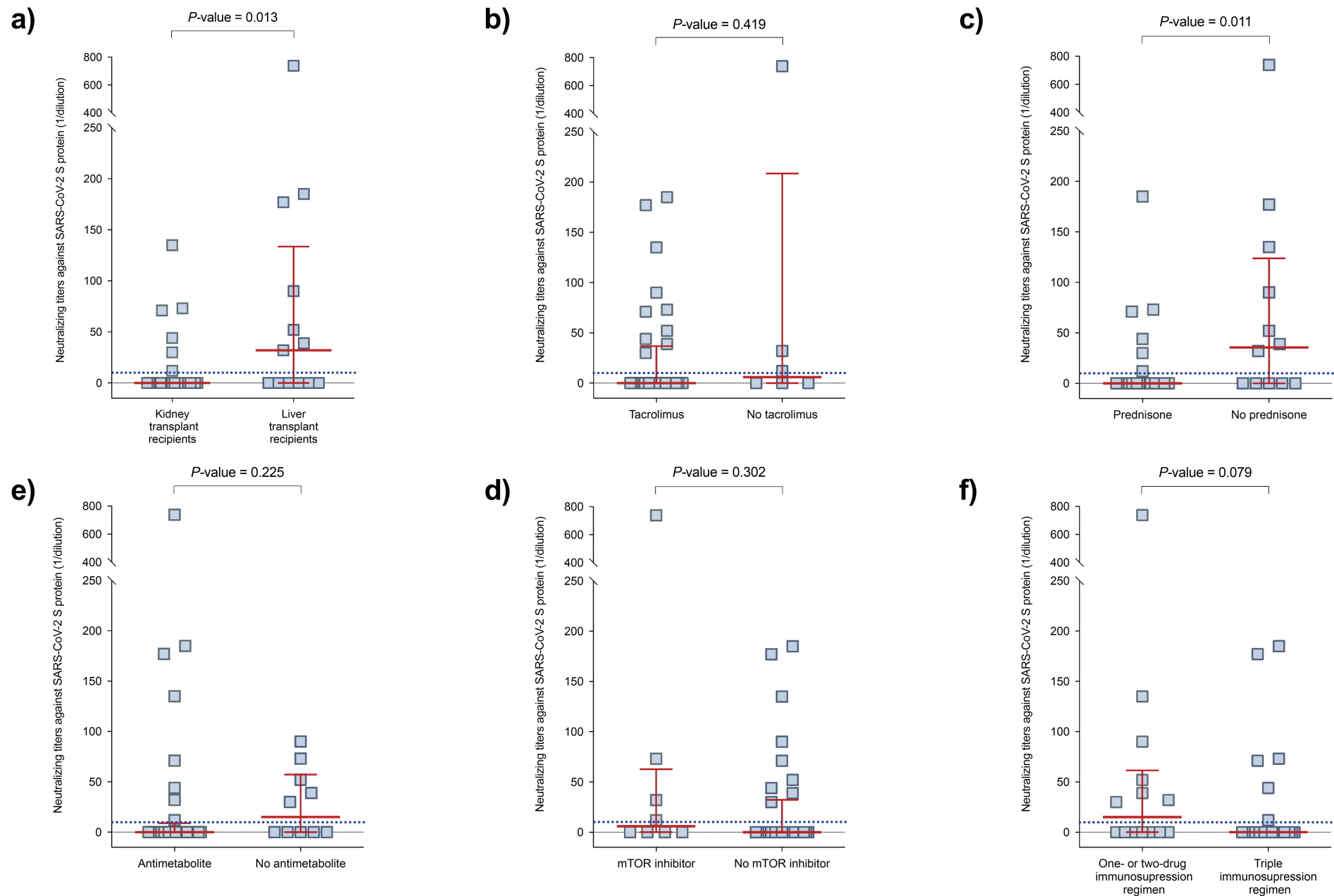
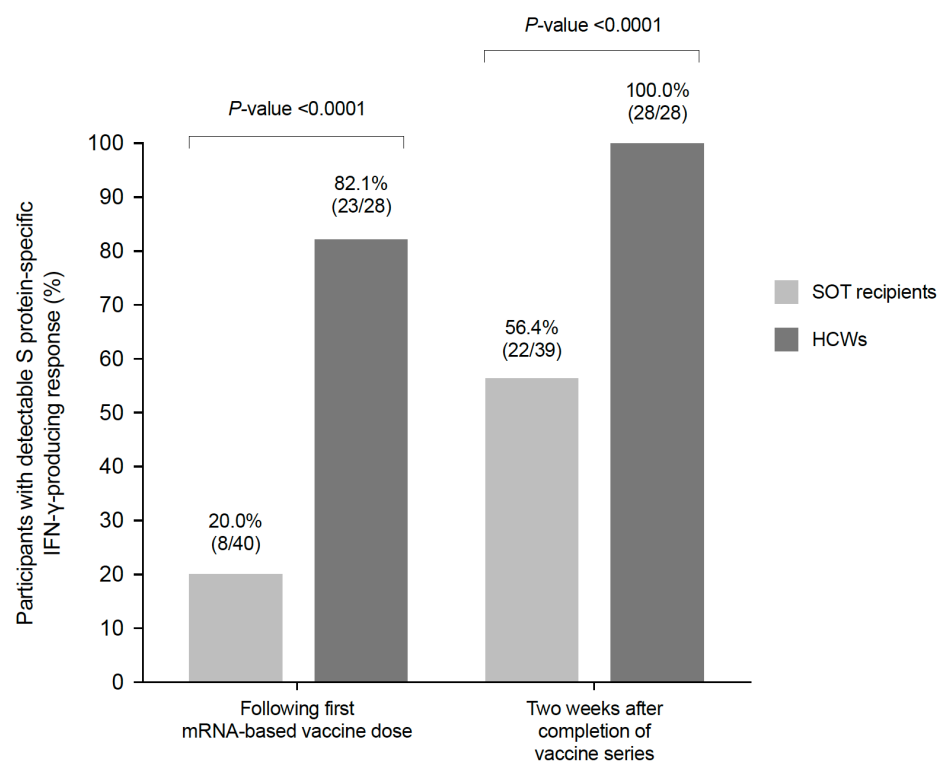


Figure S5 [next page]. *Sensitivity analysis excluding SOT recipients with natural immunity before vaccination (n = 3): SARS-CoV-2-specific cell-mediated immunity in SOT recipients and HCWs that received the mRNA-1273 and BNT162bs vaccines, respectively: (a) proportion with detectable S protein-specific IFN- γ -producing response after the first vaccine dose vaccine (i.e. four and three weeks apart for mRNA-1273 and BNT162b2) and at two weeks after the completion of vaccination; (b) kinetics of the number of S protein-specific IFN- γ -producing SFUs at baseline (pre-vaccination) and at the same time points. Red bars and whiskers represent median values and interquartile ranges, respectively. The cut-off value for positivity in the IFN- γ FluoroSpot assay (>25 SFUs/ 10^6 PBMCs) is denoted by the blue dotted line. Comparisons between repeated measures were performed with the McNemar test or the Wilcoxon signed-rank test, as appropriate. HCW: healthcare worker; IFN- γ : interferon- γ ; PMBC: peripheral blood mononuclear cell; S: SARS-CoV-2 spike glycoprotein; SFU: spot forming unit.*

a)



b)

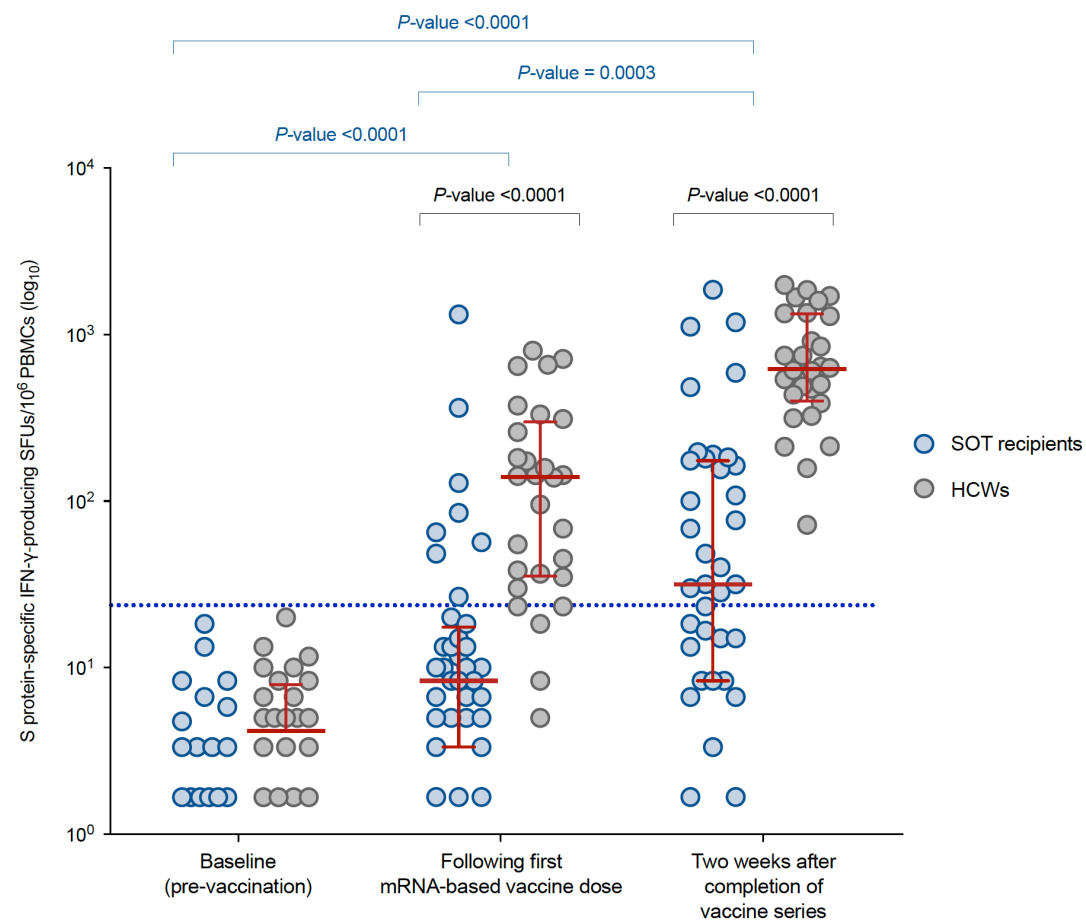


Figure S6. Sensitivity analysis excluding SOT recipients with natural immunity before vaccination ($n = 3$): SARS-CoV-2-specific humoral immunity elicited by the mRNA-1273 vaccine in SOT recipients: **(a)** proportion of patients with IgG antibodies targeting the SARS-CoV-2 S protein assessed by commercial ELISA after the first vaccine dose vaccine and at two weeks after the completion of vaccination; **(b)** proportion with serum neutralizing activity against the S protein analyzed with an in-house hACE-2/spike antibody inhibition ELISA-based method at both time points. Comparisons between repeated measures were performed with the Wilcoxon signed-rank test. S: SARS-CoV-2 spike glycoprotein; SARS-CoV-2: severe acute respiratory syndrome coronavirus 2.

