

## **Supplementary Appendix**

### *Exclusion Criteria*

Exclusion criteria included (1) previous receipt of a *C difficile* vaccine or monoclonal antibody (mAb) therapy; (2) proven or suspected history of CDI; (3) unstable chronic medical condition(s); (4) serious chronic medical disorders; (5) any condition associated with prolonged bleeding, including receipt of antithrombotic, antiplatelet, or anticoagulants other than daily aspirin ( $\leq 325$  mg/d) within 30 days of enrollment; (6) any contraindication to vaccination; (7) immunodeficiency disorders or receipt of immunosuppressive drugs within 1 year of enrollment or treatment with systemic corticosteroids for  $\geq 7$  days within 90 days of enrollment through study conclusion; (8) blood or bone marrow disorder treated within the previous year or history of bone marrow transplant; (9) malignancy requiring treatment within the last 24 months; (10) receipt of oral or parenteral antibiotics within 1 month before enrollment; (11) receipt of blood products, including immunoglobulins, within 6 months before enrollment through study conclusion; (12) participation in other studies using investigative drugs within 30 days before enrollment throughout the study duration; (13) being directly involved in the study conduct or related to someone directly involved in the study conduct, or site staff members otherwise supervised by the study investigator; (14) residence in a nursing home, long-term-care facility, or requirement for semiskilled nursing care or assisted living; (15) infection with HIV, hepatitis B virus, or hepatitis C virus; or (16) had any other condition judged by the investigator to complicate study participation and/or results interpretation.

### *Reasons for Temporary Delay of Vaccination*

Vaccination could be temporarily delayed in the case of (1) acute illness (delay of 48 hours); (2) receipt of nonlive influenza vaccine (14 days) or any other vaccine (28 days); (3) receipt of systemic

antibiotics for an acute illness (72 hours); or (4) receipt of systemic corticosteroids for <7 days (1 month after cessation of treatment).

#### *Determination of the Neutralization Threshold*

Based on the phase 2 efficacy data from Merck/Medarex in which a combination of 2 mAbs against toxin A and toxin B was tested in patients who had experienced  $\geq 1$  episode of *C difficile*-associated diarrhea (CDAD) [34], it was inferred that toxin A-neutralizing and toxin B-neutralizing mAb levels above a threshold of 10  $\mu\text{g}/\text{mL}$  are associated with protection against CDAD. The “protective” 50% neutralization titer for each antibody was therefore estimated to be the antibody concentration at the protective threshold of 10  $\mu\text{g}/\text{mL}$  divided by the respective mAb half maximal inhibitory concentration (IC50; 50% neutralization titer = mAb concentration  $\div$  mAb IC50) given the similarity of the neutralization assays used in the field. Because Pfizer’s neutralization assay employs a reference standard, the 50% neutralization titers corresponding to the estimated thresholds were then experimentally bridged to Pfizer’s neutralization U/mL.

**Table S1. Percentages of subjects achieving prespecified levels of toxin A– and toxin B–specific neutralizing antibodies in the month and day regimens.**

	Toxin A–Specific Neutralizing Antibody			Toxin B–Specific Neutralizing Antibody		
	Placebo	100 µg <i>C diff</i>	200 µg <i>C diff</i>	Placebo	100 µg <i>C diff</i>	200 µg <i>C diff</i>
Sampling	n <sup>a</sup> /N <sup>b</sup> (%)	n <sup>a</sup> /N <sup>b</sup> (%)	n <sup>a</sup> /N <sup>b</sup> (%)	n <sup>a</sup> /N <sup>b</sup> (%)	n <sup>a</sup> /N <sup>b</sup> (%)	n <sup>a</sup> /N <sup>b</sup> (%)
Time Point	95% CI <sup>c</sup>	95% CI <sup>c</sup>	95% CI <sup>c</sup>	95% CI <sup>c</sup>	95% CI <sup>c</sup>	95% CI <sup>c</sup>
<b>Month Regimen</b>						
Day 1	1/53 (1.9) (0.0, 10.1)	1/163 (0.6) (0.0, 3.4)	2/158 (1.3) (0.2, 4.5)	2/53 (3.8) (0.5, 13.0)	4/163 (2.5) (0.7, 6.2)	7/158 (4.4) (1.8, 8.9)
Day 30	0/53 (0.0) (0.0, 6.7)	28/162 (17.3) (11.8, 24.0)	30/157 (19.1) (13.3, 26.1)	1/53 (1.9) (0.0, 10.1)	42/162 (25.9) (19.4, 33.4)	58/157 (36.9) (29.4, 45.0)
Day 37	1/53 (1.9) (0.0, 10.1)	71/163 (43.6) (35.8, 51.5)	89/157 (56.7) (48.6, 64.6)	2/53 (3.8) (0.5, 13.0)	47/163 (28.8) (22.0, 36.4)	60/157 (38.2) (30.6, 46.3)
Month 2	1/53 (1.9) (0.0, 10.1)	67/163 (41.1) (33.5, 49.1)	90/158 (57.0) (48.9, 64.8)	3/53 (5.7) (1.2, 15.7)	47/163 (28.8) (22.0, 36.4)	58/158 (36.7) (29.2, 44.7)
Month 6	1/53 (1.9) (0.0, 10.1)	25/163 (15.3) (10.2, 21.8)	32/158 (20.3) (14.3, 27.4)	0/53 (0.0) (0.0, 6.7)	32/163 (19.6) (13.8, 26.6)	44/158 (27.8) (21.0, 35.5)
Day 187	0/53 (0.0) (0.0, 6.7)	130/162 (80.2) (73.3, 86.1)	134/158 (84.8) (78.2, 90.0)	1/53 (1.9) (0.0, 10.1)	94/162 (58.0) (50.0, 65.7)	101/158 (63.9) (55.9, 71.4)
Month 7	1/53 (1.9) (0.0, 10.1)	160/163 (98.2) (94.7, 99.6)	151/158 (95.6) (91.1, 98.2)	4/53 (7.5) (2.1, 18.2)	122/163 (74.8) (67.5, 81.3)	138/158 (87.3) (81.1, 92.1)
Month 12	1/51 (2.0) (0.0, 10.4)	115/160 (71.9) (64.2, 78.7)	120/155 (77.4) (70.0, 83.7)	3/51 (5.9) (1.2, 16.2)	71/160 (44.4) (36.5, 52.4)	88/155 (56.8) (48.6, 64.7)
Month 18	2/51 (3.9)	75/154 (48.7)	81/152 (53.3)	4/51 (7.8)	53/154 (34.4)	72/152 (47.4)

	(0.5, 13.5)	(40.6, 56.9)	(45.0, 61.4)	(2.2, 18.9)	(27.0, 42.5)	(39.2, 55.6)
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Day Regimen						
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Day 1	0/56 (0.0)	2/171 (1.2)	1/164 (0.6)	3/56 (5.4)	8/171 (4.7)	6/164 (3.7)
	(0.0, 6.4)	(0.1, 4.2)	(0.0, 3.4)	(1.1, 14.9)	(2.0, 9.0)	(1.4, 7.8)
Day 8	0/56 (0.0)	8/171 (4.7)	11/165 (6.7)	2/56 (3.6)	17/171 (9.9)	19/165 (11.5)
	(0.0, 6.4)	(2.0, 9.0)	(3.4, 11.6)	(0.4, 12.3)	(5.9, 15.4)	(7.1, 17.4)
Day 15	0/56 (0.0)	29/171 (17.0)	41/164 (25.0)	2/56 (3.6)	52/171 (30.4)	61/164 (37.2)
	(0.0, 6.4)	(11.7, 23.4)	(18.6, 32.3)	(0.4, 12.3)	(23.6, 37.9)	(29.8, 45.1)
Day 30	0/56 (0.0)	45/171 (26.3)	60/164 (36.6)	2/56 (3.6)	51/171 (29.8)	61/164 (37.2)
	(0.0, 6.4)	(19.9, 33.6)	(29.2, 44.5)	(0.4, 12.3)	(23.1, 37.3)	(29.8, 45.1)
Day 37	7/56 (12.5)	117/171 (68.4)	141/165 (85.5)	1/56 (1.8)	51/171 (29.8)	64/165 (38.8)
	(5.2, 24.1)	(60.9, 75.3)	(79.1, 90.5)	(0.0, 9.6)	(23.1, 37.3)	(31.3, 46.7)
Month 2	2/56 (3.6)	105/171 (61.4)	141/165 (85.5)	1/56 (1.8)	50/171 (29.2)	61/165 (37.0)
	(0.4, 12.3)	(53.7, 68.7)	(79.1, 90.5)	(0.0, 9.6)	(22.5, 36.7)	(29.6, 44.8)
Month 4	1/55 (1.8)	50/170 (29.4)	86/164 (52.4)	0/55 (0.0)	45/170 (26.5)	51/164 (31.1)
	(0.0, 9.7)	(22.7, 36.9)	(44.5, 60.3)	(0.0, 6.5)	(20.0, 33.8)	(24.1, 38.8)
Month 7	0/54 (0.0)	25/166 (15.1)	50/159 (31.4)	0/54 (0.0)	40/166 (24.1)	42/159 (26.4)
	(0.0, 6.6)	(10.0, 21.4)	(24.3, 39.3)	(0.0, 6.6)	(17.8, 31.3)	(19.7, 34.0)
Month 13	1/54 (1.9)	16/158 (10.1)	40/155 (25.8)	1/54 (1.9)	31/158 (19.6)	48/155 (31.0)
	(0.0, 9.9)	(5.9, 15.9)	(19.1, 33.4)	(0.0, 9.9)	(13.7, 26.7)	(23.8, 38.9)

*C diff*=*Clostridium difficile* vaccine

<sup>a</sup>n=number of subjects achieving toxin A– or toxin B–specific neutralizing antibody levels  $\geq$  threshold.

<sup>b</sup>N=number of subjects with valid and determinate assay results for the specified neutralizing antibody.

<sup>c</sup>2-sided CIs are calculated based on the Clopper-Pearson method.