Supplementary Online Content

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This supplementary material has been provided by the authors to give readers additional information about their work.

eTable 1. Baseline Characteristics for Patients Not Included in the Final Analysis Plan

		Doxycycline (N = 4)	Placebo (N = 3)
Demographics			
Age (years)	Mean (std dev)	72.8 (11.4)	79.3 (1.5)
		n (%)	n (%)
Gender	Female	1 (25%)	1 (33%)
	Male	3 (75%)	2 (67%)
Health Status			
Smoking Status	Current Smoker	2 (50%)	1 (33%)
	Former Smoker	2 (50%)	1 (33%)
	Never Smoked	0 (0%)	1 (33%)
Coronary Artery Disease		1 (25%)	1 (33%)
Congestive Heart Failure		0 (0%)	0 (0%)
Atrial Fibrillation		0 (0%)	1 (33%)
Stroke		0 (0%)	0 (0%)
Cancer		1 (25%)	1 (33%)
Chronic Obstructive Pulmonary Disease		1 (25%)	0 (0%)
Diabetes Mellitus		1 (25%)	0 (0%)
Hypercholesteremia		1 (25%)	3 (100%)
Any family members know to have been diagnosed with abdominal aortic aneurysm		1 (25%)	0 (0%)
Medication			
Beta Blocker		1 (25%)	3 (100%)
ACE Inhibitor		3 (75%)	2 (67%)
Angiotensin Receptor Blocker		1 (25%)	0 (0%)
Calcium Channel Blocker		1 (25%)	0 (0%)
Diuretics		0 (0%)	1 (33%)
Use of Any anti-hypertensive		4 (100%)	3 (100%)
Daily Aspirin		2 (50%)	3 (100%)
Other antiplatelet		0 (0%)	1 (33%)
Aspirin or other antiplatelet		2 (50%)	3 (100%)
Statin		0 (0%)	3 (100%)
Aneurysm Maximal Transverse Diameter			
Overall	N	4	3
	Mean (std dev)	4.0 (0.4)	4.2 (0.6)
	Median	4.1	3.9
	25th, 75th %-ile	3.7, 4.3	3.7, 4.9
Men	N	3	2
	Mean (std dev)	4.2 (0.2)	4.3 (0.8)
	Median	4.3	4.3
	25th, 75th %-ile	3.9, 4.4	3.7, 4.9
Women	N	1	1
	Mean (std dev)	3.5 ( )	3.9 ( )
	Median	3.5	3.9
	25th, 75th %-ile	3.5, 3.5	3.9, 3.9

eTable 1. Baseline Characteristics for Patients Not Included in the Final Analysis Plan		Doxycycline (N = 133)	Placebo (N = 128)
Aneurysm Volume			
Overall	N	4	3
	Mean (std dev)	81.7 (13.1)	81.5 (23.0)
	Median	84.5	74.9
	25th, 75th %-ile	73.5, 90.0	62.5, 107.1
Men	N	3	2
	Mean (std dev)	87.8 (5.9)	84.8 (31.5)
	Median	85.4	84.8
	25th, 75th %-ile	83.5, 94.5	62.5, 107.1
Women	N	1	1
	Mean (std dev)	63.4 ( )	74.9 ( )
	Median	63.4	74.9
	25th, 75th %-ile	63.4, 63.4	74.9, 74.9

eAppendix 1. Sex by Treatment Interaction

In N-TA³CT we stratified randomization by sex, and the Statistical Analysis Plan (SAP) specifies that sex by treatment interaction will be assessed in the primary outcome analysis. We committed to removing the interaction term from the primary, full analysis data set multiple imputation regression model if interaction is not statistically significant (i.e., $p \leq .05$) and to evaluate treatment results separately for men and women if the interaction were significant.

The multiple imputation model of normal scores corresponding to two-year change in abdominal aortic aneurysm maximal transverse diameter (MTD) (accounting for deaths and repairs as well) resulted in a positive beta coefficient for the treatment by sex interaction, indicating a trend toward a less beneficial/more detrimental effect of doxycycline on normal scores in women than in men that was not significant according to our *a priori* criteria; $p=.056$.

To further investigate, we estimated treatment effects separately in men and women. In the small group of women the coefficient of treatment effect was positive, indicating a trend toward a detrimental effect of doxycycline ($p=.07$). In men the coefficient was slightly negative but close to zero, suggesting no doxycycline effect ($t=-0.25$, $p=.80$). There is no convincing evidence of an effect of doxycycline on maximal transverse diameter growth in men or women and insufficient evidence of a difference between men and women to conclude that the analysis should be conducted separately for men and women.

We performed corresponding analyses of the actual MTD measurements in the per protocol population to assess whether this evaluation was sensitive to imputation, application of rank/normal scores and lack of adherence in the full data set analysis. The estimate of sex by treatment interaction beta coefficient was 0.089 cm with a standard error of 0.146 (95% Confidence Interval -.199 to 0.377), $t=0.61$ ($p=.54$). There is insufficient evidence of a difference between men and women to challenge the interpretation of the primary, full analysis data set multiple imputation regression model.

We concluded that in the primary analysis results for men and women should be analyzed together instead of separately.

eTable 2. MMP-9 and CRP Levels

			Doxycycline	Placebo	Total
Full Analysis Population					
MMP-9 Levels (ng/ml)	Baseline	N	128	124	252
		Mean (std dev)	54.6 (53.2)	55.0 (51.1)	54.8 (52.1)
		Median (25th, 75th %-ile)	37.2 (27.3, 55.5)	38.4 (27.0, 62.7)	38.0 (27.3, 57.0)
		10th, 90th %-ile	20.7, 107.8	22.1, 107.0	21.8, 107.0
		Min, Max	13.5, 322.8	9.5, 350.1	9.5, 350.1
	6 Month	N	108	111	219
		Mean (std dev)	60.9 (85.3)	61.9 (61.8)	61.4 (74.2)
		Median (25th, 75th %-ile)	39.2 (24.9, 62.3)	37.9 (25.8, 74.5)	38.2 (25.5, 64.4)
		10th, 90th %-ile	18.5, 110.5	21.1, 113.9	20.2, 113.9
		Min, Max	12.4, 727.4	13.1, 378.8	12.4, 727.4
	1 Year	N	111	100	211
		Mean (std dev)	63.0 (90.1)	61.0 (91.5)	62.1 (90.5)
		Median (25th, 75th %-ile)	31.3 (23.1, 59.4)	31.9 (26.0, 50.5)	31.7 (23.9, 55.7)
		10th, 90th %-ile	17.5, 145.1	20.4, 116.2	18.4, 136.4
		Min, Max	3.7, 630.6	10.7, 691.5	3.7, 691.5
	1 Year, 6 month	N	88	89	177
		Mean (std dev)	48.8 (43.9)	68.3 (86.7)	58.6 (69.3)
		Median (25th, 75th %-ile)	34.4 (22.9, 53.3)	36.1 (25.8, 68.6)	35.2 (24.2, 60.5)
		10th, 90th %-ile	19.0, 106.6	21.6, 146.2	20.3, 119.8
		Min, Max	11.5, 239.6	8.0, 450.6	8.0, 450.6
	2 Year	N	86	83	169
		Mean (std dev)	53.8 (54.8)	52.8 (41.9)	53.3 (48.7)
		Median (25th, 75th %-ile)	34.0 (27.6, 53.2)	37.0 (23.8, 61.5)	35.0 (26.2, 57.1)
		10th, 90th %-ile	21.9, 113.5	19.9, 108.6	20.3, 111.4
		Min, Max	14.1, 358.2	7.9, 215.1	7.9, 358.2
	2 Year, 6 month	N	7	13	20
		Mean (std dev	31.3 (5.0)	31.7 (13.2)	31.5 (10.9)
		Median (25th, 75th %-ile	30.6 (26.8, 33.7)	31.7 (21.8, 38.3)	31.1 (24.8, 38.1)
		10th, 90th %-ile	26.3, 40.6	16.4, 48.4	16.7, 46.3
		Min, Max	26.3, 40.6	13.9, 56.1	13.9, 56.1
CRP (mg/l)	Baseline	N	128	124	252
		Mean (std dev)	5.4 (13.6)	3.6 (4.9)	4.5 (10.3)
		Median (25th, 75th %-ile)	2.5 (1.3, 5.2)	2.2 (1.2, 4.2)	2.4 (1.2, 4.6)
		10th, 90th %-ile	0.7, 11.8	0.6, 8.3	0.7, 9.5
		Min, Max	0.3, 145.6	0.3, 43.1	0.3, 145.6
	6 Month	N	108	111	219
		Mean (std dev)	5.4 (12.7)	4.5 (10.7)	4.9 (11.7)
		Median (25th, 75th %-ile)	1.8 (1.1, 3.8)	2.3 (1.2, 4.8)	2.1 (1.1, 4.6)
		10th, 90th %-ile	0.6, 8.6	0.8, 7.4	0.7, 7.8
		Min, Max	0.3, 79.0	0.3, 110.5	0.3, 110.5
	1 Year	N	111	100	211
		Mean (std dev)	3.5 (4.8)	4.1 (6.9)	3.8 (5.9)
		Median (25th, 75th %-ile)	2.0 (1.0, 3.8)	2.4 (1.2, 4.6)	2.2 (1.1, 4.3)
		10th, 90th %-ile	0.6, 7.7	0.6, 8.3	0.6, 8.1
		Min, Max	0.3, 31.8	0.4, 63.2	0.3, 63.2
CRP (mg/l)	1 Year, 6 month	N	88	89	177
		Mean (std dev)	3.5 (6.5)	5.7 (15.1)	4.6 (11.6)
		Median (25th, 75th %-ile)	1.7 (0.8, 3.3)	2.7 (1.3, 4.6)	1.9 (1.0, 3.8)
		10th, 90th %-ile	0.5, 6.4	0.9, 7.3	0.6, 6.6
		Min, Max	0.1, 42.5	0.1, 124.0	0.1, 124.0

			Doxycycline	Placebo	Total
	2 Year	N	86	83	169
		Mean (std dev)	5.9 (15.6)	4.0 (5.3)	5.0 (11.7)
		Median (25th, 75th %-ile)	1.6 (1.0, 4.3)	2.2 (1.0, 4.3)	1.9 (1.0, 4.3)
		10th, 90th %-ile	0.5, 7.9	0.7, 10.5	0.5, 10.5
		Min, Max	0.1, 120.7	0.2, 26.4	0.1, 120.7
	2 Year, 6 month	N	13	22	35
		Mean (std dev)	2.2 (1.4)	5.3 (7.2)	4.1 (5.9)
		Median (25th, 75th %-ile	1.8 (0.9, 3.4)	2.6 (1.2, 6.8)	2.3 (1.2, 5.0)
		10th, 90th %-ile	0.5, 3.5	0.8, 8.6	0.8, 8.0
		Min, Max	0.4, 5.0	0.5, 33.5	0.4, 33.5

Per Protocol Population			Doxycycline	Placebo	Total
MMP-9 Levels (ng/ml)	Baseline	N	67	72	139
		Mean (std dev)	54.1 (51.4)	54.2 (52.5)	54.2 (51.8)
		Median (25th, 75th %-ile)	35.8 (28.1, 55.2)	38.9 (27.0, 62.7)	37.6 (27.4, 57.2)
		10th, 90th %-ile	22.6, 111.1	21.1, 104.8	21.8, 107.0
		Min, Max	15.3, 305.4	9.5, 350.1	9.5, 350.1
	6 Month	N	66	71	137
		Mean (std dev)	65.4 (103.3)	67.5 (70.2)	66.5 (87.4)
		Median (25th, 75th %-ile)	38.1 (25.1, 61.9)	37.9 (28.0, 86.3)	37.9 (26.3, 65.0)
		10th, 90th %-ile	20.2, 110.5	20.9, 152.7	20.7, 113.9
		Min, Max	12.4, 727.4	13.1, 378.8	12.4, 727.4
	1 Year	N	66	68	134
		Mean (std dev)	51.8 (65.6)	65.5 (106.0)	58.8 (88.4)
		Median (25th, 75th %-ile)	30.3 (23.1, 45.5)	32.1 (25.4, 48.4)	31.4 (23.7, 47.6)
		10th, 90th %-ile	17.4, 125.5	18.9, 143.2	18.4, 125.5
		Min, Max	3.7, 478.5	10.7, 691.5	3.7, 691.5
	1 Year, 6 month	N	58	65	123
		Mean (std dev)	50.7 (50.5)	73.5 (97.4)	62.7 (79.4)
		Median (25th, 75th %-ile)	31.1 (22.2, 51.1)	35.2 (25.8, 68.6)	34.0 (23.8, 60.5)
		10th, 90th %-ile	17.3, 119.8	21.6, 184.1	20.3, 142.6
		Min, Max	11.5, 239.6	8.0, 450.6	8.0, 450.6
	2 Year	N	58	56	114
		Mean (std dev)	57.5 (59.0)	50.4 (41.4)	54.0 (51.0)
		Median (25th, 75th %-ile)	34.5 (29.2, 58.0)	36.1 (25.4, 57.7)	35.0 (26.8, 58.0)
		10th, 90th %-ile	21.9, 126.0	19.5, 106.6	21.2, 113.5
		Min, Max	14.1, 358.2	7.9, 215.1	7.9, 358.2
	2 Year, 6 month	N	6	9	15
		Mean (std dev)	31.8 (5.3)	28.0 (10.2)	29.5 (8.6)
		Median (25th, 75th %-ile)	31.7 (26.8, 33.7)	26.8 (21.8, 36.0)	30.6 (23.4, 36.0)
		10th, 90th %-ile	26.3, 40.6	13.9, 44.3	16.4, 40.6
		Min, Max	26.3, 40.6	13.9, 44.3	13.9, 44.3
CRP (mg/l)	Baseline	N	67	72	139
		Mean (std dev)	5.7 (17.7)	3.4 (3.1)	4.5 (12.5)
		Median (25th, 75th %-ile)	2.3 (1.1, 5.5)	2.4 (1.2, 4.1)	2.3 (1.2, 4.7)
		10th, 90th %-ile	0.5, 10.6	0.9, 7.2	0.7, 8.4
		Min, Max	0.3, 145.6	0.3, 14.8	0.3, 145.6
CRP (mg/l)	6 Month	N	66	71	137
		Mean (std dev)	3.2 (8.0)	3.8 (3.4)	3.5 (6.1)
		Median (25th, 75th %-ile)	1.6 (0.9, 3.1)	2.7 (1.3, 5.4)	2.1 (1.1, 4.3)
		10th, 90th %-ile	0.5, 5.5	0.7, 7.8	0.6, 7.1
		Min, Max	0.3, 65.5	0.4, 16.2	0.3, 65.5
	1 Year	N	66	68	134
		Mean (std dev)	2.9 (4.3)	4.4 (7.7)	3.7 (6.3)
		Median (25th, 75th %-ile)	1.7 (0.8, 3.3)	2.7 (1.3, 5.3)	2.2 (1.1, 4.0)
		10th, 90th %-ile	0.5, 5.1	0.7, 8.1	0.6, 6.6
		Min, Max	0.3, 30.7	0.4, 63.2	0.3, 63.2
	1 Year, 6 month	N	58	65	123
		Mean (std dev)	3.1 (5.6)	6.6 (17.4)	4.9 (13.3)
		Median (25th, 75th %-ile)	1.2 (0.7, 3.1)	2.6 (1.3, 4.2)	1.9 (1.0, 3.9)
		10th, 90th %-ile	0.4, 5.7	0.9, 6.7	0.6, 6.4
		Min, Max	0.2, 35.9	0.1, 124.0	0.1, 124.0

			Doxycycline	**Placebo**	**Total**
	2 Year	N	58	56	114
		Mean (std dev)	3.2 (6.5)	4.6 (5.5)	3.9 (6.1)
		Median (25th, 75th %-ile)	1.5 (0.7, 3.4)	2.6 (1.1, 5.2)	2.0 (1.0, 4.0)
		10th, 90th %-ile	0.5, 6.1	0.8, 11.9	0.5, 10.4
		Min, Max	0.1, 47.0	0.2, 26.4	0.1, 47.0
	2 Year 6 month	N	10	16	26
		Mean (std dev)	1.6 (1.1)	6.3 (8.2)	4.5 (6.8)
		Median (25th, 75th %-ile	1.5 (0.8, 2.1)	3.6 (1.5, 8.0)	2.0 (1.2, 5.7)
		10th, 90th %-ile	0.5, 3.5	0.8, 14.2	0.5, 8.6
		Min, Max	0.4, 3.5	0.5, 33.5	0.4, 33.5

eTable 3. Frequency of Expected Adverse Events

Symptom	Expected Adverse Events		Difference	(95% CI)	p-value
	Doxycycline N=129	Placebo N=125			
Any symptom reported	121 (93.8%)	111 (88.8%)	5.0%	(-1.9 – 11.9 %)	.16
Frequent joint pain	84 (65.1%)	79 (63.2%)	1.9%	(-9.9 – 13.7 %)	.75
Frequent gastric or intestinal upset	58 (45.0%)	56 (44.8%)	0.2%	(-12.1 – 12.4 %)	.98
Frequent bleeding or bruising	47 (36.4%)	54 (43.2%)	-6.8%	(-18.8 – 5.3 %)	.27
Frequent spells of dizziness	34 (26.4%)	37 (29.6%)	-3.2%	(-14.3 – 7.8 %)	.56
Skin rash or hives	41 (31.8%)	29 (23.2%)	8.6%	(-2.3 – 19.5 %)	.13
Visual disturbance	26 (20.2%)	43 (34.4%)	-14.2%	(-25.1 – -3.4 %)	.01
Moderate to severe sunburn	38 (29.5%)	14 (11.2%)	18.3%	(8.6 – 28.8 %)	< .001
Frequent headaches	27 (20.9%)	20 (16.0%)	4.9%	(-4.6 – 14.5 %)	.31
Tooth discoloration	12 (9.30%)	18 (14.4%)	-5.1%	(-13.0 – 3.8 %)	.21
Fever	11 (8.50%)	18 (14.4%)	-5.9%	(-13.7 – 1.9 %)	.14
Other symptom requiring study drug dose adjustment or discontinuation	27 (20.9%)	27 (21.6%)	-0.7%	(-10.7 – 9.4 %)	.90

eTable 4. Doxycycline Levels

Measurement	Visit		Full Analysis Population	Per-Protocol Population
Doxycycline (ng/mL)	6 Month	N	102	66
		Mean (std dev)	3099.8 (2195.0)	3681.2 (2138.4)
		Median (25th, 75th %-ile)	2795.0 (1189.7, 4759.6)	3523.7 (2193.4, 4982.1)
		10th, 90th %-ile	415.5, 6077.5	797.7, 6854.0
		Min, Max	0.0, 9362.4	0.0, 9362.4
	1 Year	N	79	52
		Mean (std dev)	3165.0 (2355.4)	3729.0 (2285.2)
		Median (25th, 75th %-ile)	2998.0 (1164.2, 4742.3)	3740.0 (1707.1, 5135.8)
		10th, 90th %-ile	0.0, 6226.8	1099.7, 6531.9
		Min, Max	0.0, 9689.4	0.0, 9689.4
	1 Year, 6 month	N	48	34
		Mean (std dev)	2844.8 (2412.9)	3172.8 (2513.0)
		Median (25th, 75th %-ile)	1972.2 (665.8, 5050.3)	2410.9 (988.1, 5497.5)
		10th, 90th %-ile	0.0, 6196.7	493.0, 6345.5
		Min, Max	0.0, 8554.0	0.0, 8554.0
	2 Year	N	36	25
		Mean (std dev)	2390.0 (2517.4)	2750.7 (2539.8)
		Median (25th, 75th %-ile)	1881.9 (0.0, 4106.2)	2046.9 (370.3, 4626.5)
		10th, 90th %-ile	0.0, 6655.3	0.0, 6655.3
		Min, Max	0.0, 8858.2	0.0, 8858.2

Doxycycline assay was performed using an ultra-high performance liquid chromatography (UHPLC)-mass spectrometry (MS)/mass spectrometry (MS) method developed for this study because liquid chromatography methods used in animal studies returned false positive levels of doxycycline in specimens obtained at baseline.[31] Of 133 specimens from patients assigned to placebo, four (3%) were positive for doxycycline. These patients were not reported to be taking doxycycline and tracking of their study treatment kits found all to have been properly handled placebo treatments leaving errors in labeling or transcription in the clinical site or laboratory as a possibility.

Seelam RR, Terrin M, Hassan HE. Validated UHPLC-MS/MS method for quantification of doxycycline in abdominal aortic aneurysm patients. *Bioanalysis*. 2018;10(8):527-539.