Supplementary methods and tables for

Dupilumab increases aspirin tolerance in NSAID-exacerbated respiratory disease (N-ERD)

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SUPPLEMENTARY METHODS

Additional study measures

Extent of nasal polyps was evaluated using a rigid 0° endoscope. After application of local anesthetic and decongestant, TPS was graded for each nasal cavity from 0 to 4 (0= no visible polyps, 1= small polyps in the middle meatus, 2= nasal polyps reaching below the lower border of the middle turbinate, 3= large polyps reaching the lower border of the inferior turbinate or polyps medial to the middle turbinate, 4= large polyps causing complete obstruction of the inferior nasal cavity) (1). Nasal secretions were collected at baseline and at week 24 and processed as previously described using nasosorptions (Nasosorption FX-I, Hunt Developments (UK) Limited, Midhurst, West Sussex, United Kingdom) (2). Urine was collected at baseline and week 24 after treatment. Blood eosinophil counts, serum total IgE levels and ECP were assessed at baseline and after 24 weeks and potential drug-related side effects were monitored at every visit. The study design is displayed in Fig. E1.

Sample size considerations:

The sample size of the study was calculated based on the half-width of the 95% confidence interval for the proportion of patients with an improvement in the primary endpoint between visit 1 (baseline) and visit 4 (24 weeks after treatment with dupilumab). With 30 patients and observed proportions of 50%, 70% or 80%, the respective half-width of the 95% confidence interval will be 18, 16, or 14 percentage points. Given the explorative nature of the study this is considered as a sufficient level of precision.

MSD Measurement

The following cytokines were assayed in nasal secretions using the electrochemiluminescence technology MSD multiplex U-Plex platform: Interleukin (IL)-1α, Il-1β, IL-1RA, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-12/IL-23p40, IL-12p70, IL-13, IL-15, IL-16, IL-17A, IL-17E/IL-25, IL-21, IL-22, IL-27, IL-33, Tumor Necrosis Factor-alpha (TNF-α), TNF-β, Interferon-gamma (IFN-γ), Thymic Stromal Lymphopoietin (TSLP), Eotaxin-1, Eotaxin-3, Thymus and Activation-Regulated Chemokine (TARC=CCL17), Vascular Endothelial Growth Factor-A (VEGF-A), Granulocyte Colony-Stimulating Factor (G-CSF) and Granulocyte-Macrophage

Colony-Stimulating Factor (GM-CSF). Nasal secretions of left and right nostrils were pooled prior to application to MSD plates. All measurements were performed according to the manufacturer´s instructions (https://www.mesoscale.com/en). Briefly, linkers were complexed with 200µL target antibody, diluted to 1x and plates were coated with 50µL of this solution on a shaker for one hour at room temperature, combinations of linkers with antibodies were noted. Plates were washed thrice with PBS 0.05% Tween 20 (PBS-T), dried and stored at 4°C for a maximum of seven days prior to use. Standards were freshly reconstituted and serially diluted in 4-fold steps in diluent 43 provided with the kit. 50µL sample or standard were incubated on the plate with agitation overnight at 4°C under a seal. Plates were washed thrice, detection antibodies were diluted with diluent 3 provided and 50µL of this mixture were applied to plates for one hour at room temperature with agitation under a seal. Plates were washed thrice with PBS-T and immediately measured in 150µL of MSD GOLD read buffer (provided) on a MESO SECTOR S 600 plate reader. DISCOVERY WORKBENCH software (MSD, Rockville, Maryland, USA) was inputted with batch-specific standard concentrations and sample analyte concentrations were extrapolated by the software.

Missing concentration values that resulted from readings below or above the detection limits were imputed using the mean of the estimated lower and higher detection limit values for the specific cytokine across all batch readings.

ELISA measurement of total IgE levels in nasal secretion

IgE levels within nasal secretions were measured by using the Human IgE ELISA Antibody Pair Kit (Stemcell Technologies, Vancouver, Canada) according to the manufacturer's instructions. Briefly, ELISA plates (i.e., 384 well plates, Nunc Maxisorp, Roskilde, Denmark) were coated with 1 µg/ml Human IgE Capture Antibody in PBS overnight at 4°C and blocked with PBS, 0.05% v/v Tween-20, 0.1% v/v BSA (incubation buffer). As a standard, purified Bet v 1 specific IgE (12.5 µg/ml) was used (3). Samples were diluted either 1:5 or 1:10 in incubation buffer. Samples and standards were added to the plate and then incubated overnight at 4°C. After washing, 0.3µg/ml Human IgE Biotinylated Detection Antibody was added and left for 1 hour at room temperature. Subsequently, the plates were washed and incubated with Streptavidin Alkaline Phosphatase for 1 hour at room temperature. After thorough washing, the 4-Nitrophenyl phosphate disodium salt hexahydrate (pNPP) substrate

solution (pNPP in 10% v/v Diethanolamine, 2% Sodium azide in H2O, pH 9.8) was added to the plate and plates were incubated for 1 hour at room temperature protected from light. OD levels were measure at 405 nm. All measurements were done in triplicates, background was subtracted.

ELISA measurement of 11β-Prostaglandin F2α and Leukotriene E4 levels in urine.

11β-Prostaglandin F2α and Leukotriene E4 levels in urine were measured using the Leukotriene E4 ELISA Kit (Cayman Chemicals, Michigan, USA) and 11β-Prostaglandin F2α (Cayman Chemicals, Michigan, USA) respectively, following the manufacturer's protocol similar to the ELISA described above. All measurements were done in triplicates, background was subtracted.

Statistics:

Data from clinical questionnaires were included in the analysis if they were greater than 80% complete and contained at least two patient visits. Missing values were imputed using the mean of the two closest visits (Number and percentage of imputed missing values: please refer to table E3). Mean and standard deviation as well as median and range were calculated for metric variables and absolute and relative frequencies were calculated for categorical variables. Tolerated aspirin dosages at baseline and after 24 weeks were summarized in a table.

McNemar's exact test was used for dichotomous data to assess significance between baseline and week 24 of matched samples. Patients with missing values in a particular outcome variable were excluded in the calculation of the respective summary statistics. Plots for secondary endpoints (TPS, TNSS, SNOT-22, UPSIT and ACT) are presented as mean difference from baseline (with 95% confidence intervals and *p* values). Statistical significance was assessed with non-parametric Wilcoxon signed-rank test apart from FEV1 and MEF50 where a two-sided t-test was used according to the assumption of normality. A significance level of 0.05 was applied.

Levels of cytokines and chemokines were not normally distributed as assessed by Shapiro-Wilks test. Statistical analysis on cytokines and chemokines was carried out for paired samples using the Wilcoxon signed-rank test and for non-paired samples (comparing aspirin-tolerant vs. aspirin-intolerant patients) using the Mann-Whitney *U* test.

Change in cytokine and chemokine levels from baseline to 24 weeks was presented as median percentage change from baseline with median absolute deviations. Differences in medians between aspirin-tolerant and aspirin-intolerant patients were assessed using the Mann-Whitney *U* test for independent samples.

Correlation plots of cytokines and chemokines with clinical parameters were drawn using percentage change from baseline and calculation of Spearman correlation coefficient.

Statistical analyses were conducted using Python 3.9.6 and R 4.1.0.

References:

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Table E1. Change from baseline to week 24 in clinical endpoints.

	Total				Aspirin-intolerant				Aspirin-tolerant			
Clinical Endpoint (range)	**Baseline, Mean (±SD[1]) Median (min-max)**	**Week 24, Mean (±SD) Median (min-max)**	**Mean difference from baseline Mean (±SD)**	***p* value**	**Baseline, Mean (±SD) Median (min-max)**	**Week 24, Mean (±SD) Median (min-max)**	**Mean difference from baseline Mean (±SD)**	***p* value**	**Baseline, Mean (±SD) Median (min-max)**	**Week 24, Mean (±SD) Median (min-max)**	**Mean difference from baseline Mean (±SD)**	***p* value**
TPS combined (0-8)	N=31 3.58 (2.42) 3 (0 – 8)	N=31 0.90 (1.45) 0 (0 – 4)	-2.68 (1.84)	p<0.001	N=13 4.46 (2.33) 5 (1 – 8)	N=13 1.62 (1.71) 1 (0 – 4)	-2.84 (1.61)	p<0.001	N=17 2.76 (2.28) 3 (0 – 7)	N=17 0.35 (1.00) 0 (0 – 4)	-2.41 (1.94)	0.001
UPSIT (0-40)	N=30 14.7 (8.68) 10.5 (6 – 37)	N=29 25.86 (8.07) 26 (10 – 40)	11.16 (9.54)	p<0.001	N=12 13.33 (8.62) 10 (6 – 37)	N=13 27.46 (9.04) 29 (13 – 40)	14.13 (9.19)	0.002	N=17 16.12 (8.85) 11 (8 – 33)	N=15 24 (7.09) 24 (10 – 35)	7.88 (8.17)	0.004
SNOT-22 (0-110)	N=30 41.43 (23.27) 40.25 (5 – 83)	N=30 14.7 (12.6) 12 (1– 66)	-26.73 (23.49)	p<0.001	N=13 46.85 (22.34) 42.5 (13 – 80)	N=13 14.35 (8.82) 13 (2 – 30)	-32.5 (19.76)	p<0.001	N=16 37.12 (25.54) 32 (5 – 83)	N=16 16.72 (16.01) 12 (1 – 68)	-20.4 (21.07)	0.001
TNSS (0-12)	N=30 5.27 (2.88) 5 (0 – 11)	N=31 1.81 (1.66) 2 (0 – 6)	-3.46 (3.39)	p<0.001	N=13 5.23 (3.06) 5 (1 – 11)	N=13 1.85 (1.68) 1 (0 – 6)	-3.38 (3.41)	0.008	N=16 5.12 (2.83) 5 (0 – 11)	N=17 1.65 (1.66) 2 (0 – 5)	-3.47 (3.47)	0.003
ACT (0-25)	N=29 21.07 (3.85) 22 (12 – 25)	N=27 23.56 (2.19) 25 (16 – 25)	2.49 (3.66)	p<0.001	N=13 20.23 (2.74) 21 (16 – 25)	N=10 22.90 (2.85) 24 (16 – 25)	2.67 (2.25)	0.004	N=15 21.53 (4.61) 23 (12 – 25)	N=16 23.88 (1.71) 25 (20 – 25)	2.35 (4.42)	0.03
FEV1[a] (%)	N=29 70.51 (8.87) 71.09 (51.13 – 86.58)	N=24 72.79 (8.19) 74.63 (51.99 - 84.88)	2.28 (5.44)	0.013	N=13 71.3 (8.58) 71.09 (51.13 – 82.71)	N=12 75.1 (6.42) 78.02 (64.78 – 84.59)	3.8 (5.24)	0.02	N=15 69.88 (9.64) 71.77 (53 – 86.58)	N=12 70.48 (9.34) 71.47 (51.99 – 84.88)	0.6 (5.32)	NS (0.3)
MEF50[b] (L/s)	N=29 2.40 (0.81) 2.29 (0.86 – 4.31)	N=24 2.66 (0.89) 2.50 (1.14 – 4.41)	0.26 (0.79)	0.016	N=13 2.29 (0.7) 2.29 (0.86 – 3.33)	N=12 2.88 (0.77) 2.64 (1.9 – 4.41)	0.59 (0.85)	0.03	N=15 2.46 (0.93) 2.23 (1.01 – 4.31)	N=12 2.43 (0.98) 2.25 (1.14 – 4.28)	-0.03 (0.66)	NS (0.29)
Mef75[c] (L/s)	N=23 4.69 (1.48)	N=18 4.89 (1.54)	0.2	p<0.001	N=9 4.6 (1.28)	N=9 5.27 (1.65)	0.67	0.006	N=13 4.68 (1.7)	N=9 4.5 (1.41)	-0.18	0.02

	4.85 (2.71 – 9.39)	4.52 (2.74 – 7.97)	(1.21)		4.75 (2.89 – 6.63)	5.4 (3.07 – 7.97)	(1.08)		4.85 (2.71 – 9.39)	4.12 (2.74 – 7.47)	(1.25)	
Eosinophils, absolute (cells/ µL)	N=28 0.42 (0.24) 0.4 (0.1 – 0.9)	N=28 0.63 (0.45) 0.55 (0 – 1.5)	0.21 (0.35)	0.003	N=11 0.51 (0.3) 0.5 (0.1 – 0.9)	N=11 0.61 (0.43) 0.5 (0 – 1.5)	0.1 (0.24)	0.28	N=16 0.37 (0.17) 0.35 (0.1 – 0.7)	N=16 0.69 (0.46) 0.65 (0.1 – 1.48)	0.32 (0.4)	0.021
Percentage of Eosinophils	N=28 5.67 (3.00) 4.6 (0.7 – 12)	N=28 7.38 (4.86) 7.6 (0.3 – 17)	1.71 (4.07)	0.01	N=11 6.3 (3.15) 6.7 (1.9 – 9.7)	N=11 7.74 (5.02) 7.0 (0.8 – 17)	1.44 (3.17)	0.07	N=16 5.27 (3.02) 4.3 (0.7 – 12)	N=16 7.57 (4.71) 8.55 (0.9 – 15)	2.3 (4.32)	0.039
Blood total IgE[d] (kU/L)	N=30 264.82 (264.71) 194.50 (26.9 – 1051)	N=31 99.69 (106.9) 64.2 (5.9 – 420)	-165.13 (184.06)	p<0.001	N=12 362.03 (324.97) 250.5 (62.3 – 1051)	N=13 116.46 (118.17) 87.5 (16.2 – 420)	245.57 (239.91)	0.02	N=17 206.79 (204.24) 146 (26.9 – 896)	N=17 88.96 (102.57) 40.1 (5.9 – 407)	117.83 (108.2)	p<0.001
Nasal secretion total IgE[d] (ng/µL)	N=31 1389.91 (1845.84) 528.87 (65.18 – 6813.47)	N=31 194.14 (358.54) 80.53 (53.61 – 1954.98)	-1195.77 (1777.02)	p<0.001	N=13 2275.51 (2343.61) 1652.23 (81.26 – 6813.47)	N=13 320.89 (533.59) 80.53 (55.25 – 1954.98)	-1954.62 (2256.51)	p<0.001	N=17 786.31 (1070.86) 293.6 (65.18 – 3728.98)	N=17 101.97 (70.75) 78.7 (53.61 – 336.66)	-684.34 (1040.22)	p<0.001
ECP[e] (µg/L)	N=23 57.23 (43.85) 39.9 (9.38 – 200)	N=22 106.25 (62.29) 109.5 (30.2 – 200)	49.02 (48.82)	p<0.001	N=10 54.24 (31.92) 47.7 (9.38 – 109)	N=10 114.24 (55.39) 127 (36.2 – 200)	60.0 (55.37)	p<0.001	N=12 61.36 (54.32) 38.5 (18.8 – 200)	N=12 99.59 (69.21) 72.55 (30.2 – 200)	38.23 (34.65)	p<0.001
Leukotriene E4	N=31 801.38 (673.48) 701.77 (10.64 – 2635.19)	N=31 506.19 (480.19) 319.87 (27.47 – 1024.72)	-295.19 (655.45)	0.02	N=13 825.37 (770.17) 792.48 (20.51 – 2635.19)	N=13 632.75 (577.27) 403.83 (27.47 – 2014.72)	-192.62 (485.35)	NS (0.31)	N=17 816.12 (620.59) 701.77 (10.64 – 2560.60	N=17 374.31 (358.69) 226.84 (67.38 – 1227.22)	-441.81 (703.33)	0.02
11β-Prostaglandin F2α	N=31 943.48 (864.94) 707.76 (29.93 – 3380.24)	N=31 454.92 (415.03) 249.32 (0.01 – 1363.05)	-488.56 (943.07)	0.03	N=13 853.87 (760.52) 674.56 (45.24 – 2500.24	N=13 515.68 (431.76) 449.03 (11.28 – 1363.05)	-338.19 (772.03)	NS (0.38)	N=17 1057.69 (954.38) 767.18 (29.93 – 3380.24)	N=17 376.07 (393.20) 167.40 (0.01 – 1276.7)	-681.62 (1003.91)	0.03

[1]SD: Standard deviation, [a]FEV1: Forced expiratory volume in one second, [b]MEF50: Maximal expiratory flow at 50%, [c]MEF75: Maximal expiratory flow at 75%, [d]Total IgE: total immunoglobulin E, [e]ECP: Eosinophil cationic protein, TPS: Total Polyp Score, UPSIT: University of Pennsylvania Smell Identification Test, SNOT: Sino-Nasal Outcome Test, TNSS: Total Nasal Symptom Score, ACT: Asthma Control Test

Table E2. Cytokine data, showing mean, standard deviation and *p* values for total patients and split into Aspirin-intolerant and Aspirin-tolerant groups at baseline and 24 weeks.

	Total (N=31)			**Aspirin-intolerant (N=13)**			**Aspirin-tolerant (N=17)**		
	Mean (±SD)			**Mean (±SD)**			**Mean (±SD)**		
	Baseline	**Week 24**	***p* value**	**Baseline**	**Week 24**	***p* value**	**Baseline**	**Week 24**	***p* value**
Eotaxin-1	604.65 (326.57)	339.29 (265.47)	p<0.001	646.03 (390.41)	366.37 (321.39)	NS (0.11)	591.55 (279.06)	332.1 (225.21)	0.001
Eotaxin-3	3161.94 (6944.9)	150.49 (107.42)	p<0.001	2528.66 (2405.85)	142.12 (87.55)	p<0.001	3680.26 (9244.51)	165.56 (119.28)	p<0.001
G-CSF	5083.04 (8733.07)	2615.67 (3521.18)	NS	6117.03 (11401.31)	2793.56 (4298.86)	NS	4568.97 (6555.58)	2618.82 (3001.96)	NS
GM-CSF	9.33 (7.63)	7.99 (4.48)	NS	11.52 (10.06)	8.06 (4.26)	NS	7.92 (5.08)	8.15 (4.8)	NS
IFN-γ	177.44 (671.33)	186.05 (540.82)	NS	66.29 (56.38)	48.27 (45.05)	NS	271.53 (906.49)	301.03 (718.16)	NS
IL-10	7.38 (7.72)	5.66 (5.86)	NS	10.2 (10.51)	4.68 (2.41)	NS	5.61 (4.05)	6.65 (7.55)	NS
IL-12p40	49.02 (47.34)	29.28 (17.47)	0.03	54.39 (48.26)	29.99 (19.55)	NS (0.13)	46.5 (48.79)	29.79 (16.27)	NS (0.21)
IL-12p70	5.01 (3.56)	5.06 (6.07)	NS	4.73 (3.34)	4.12 (4.68)	NS	5.48 (3.72)	5.9 (7.15)	NS
IL-13	43.53 (17.89)	33.8 (14.55)	0.03	41.69 (15.49)	31.9 (11.62)	NS (0.15)	45.71 (20.04)	36.12 (16.54)	NS (0.13)

IL-15	25.73 (17.26)	22.79 (13.95)	NS	26.54 (18.87)	21.86 (13.45)	NS	25.96 (16.68)	24.41 (14.52)	NS
IL-16	8274.43 (9458.6)	9236.96 (10314.08)	NS	10955.0 (12465.82)	8780.29 (8400.93)	NS	6590.28 (6319.19)	10050.08 (11906.81)	NS
IL-17A	65.33 (86.92)	33.17 (26.27)	0.04	100.13 (120.38)	35.07 (26.21)	NS (0.07)	41.85 (39.37)	33.12 (27.2)	NS (0.35)
IL-17E/IL-25	13.19 (6.56)	9.55 (7.37)	0.03	12.17 (7.2)	12.22 (6.51)	NS (1.0)	14.74 (5.26)	8.02 (7.55)	0.0044
IL-1RA	29066.16 (7360.98)	27722.82 (5734.71)	NS (0.06)	29964.26 (5844.54)	26854.85 (4456.54)	0.03	28531.05 (8637.19)	28436.05 (6750.6)	NS (0.61)
IL-1α	675.53 (1517.35)	560.84 (660.76)	NS	991.38 (2291.12)	644.0 (822.42)	NS	458.48 (487.91)	518.99 (543.44)	NS
IL-1β	2573.64 (7255.65)	4602.0 (20721.42)	NS	4892.74 (10873.38)	1057.76 (1100.06)	NS	947.77 (1569.66)	7575.68 (27987.85)	NS
IL-2	358.61 (339.38)	411.98 (375.15)	NS	447.83 (400.83)	461.77 (356.93)	NS	304.47 (286.87)	396.18 (396.34)	NS
IL-21	39.15 (36.89)	26.0 (14.69)	NS	53.03 (47.81)	25.68 (14.83)	NS	29.84 (23.61)	25.23 (15.16)	NS
IL-22	12.73 (55.95)	1.47 (0.81)	0.05	4.1 (4.71)	1.63 (1.08)	NS (0.14)	20.03 (75.68)	1.36 (0.56)	NS (0.06)
IL-27	33.46 (25.25)	26.3 (14.42)	NS	37.1 (23.62)	29.79 (14.46)	NS	31.93 (27.11)	24.16 (14.61)	NS
IL-3	117.39 (53.29)	119.38 (76.64)	NS (0.9)	97.96 (57.33)	99.59 (59.49)	NS (0.86)	128.8 (46.89)	131.19 (87.71)	NS (0.92)

IL-33	350.17 (728.43)	108.96 172.71	NS	625.84 (1046.7)	101.96 (111.13)	NS	110.6 (188.5)	102.3 (209.71)	NS
IL-4	9.35 (12.15)	11.9 (10.63)	NS	9.09 (8.82)	9.42 (6.62)	NS	10.02 (14.62)	14.43 (12.65)	NS
IL-5	736.3 (1667.52)	190.9 (210.62)	0.05	1437.03 (2437.47)	179.47 (136.9)	0.02	235.44 (264.76)	210.71 (257.67)	NS (0.85)
IL-6	499.03 (1130.93)	121.9 (195.71)	0.05	993.73 (1644.38)	157.71 (287.48)	0.04	146.96 (164.77)	100.56 (87.46)	NS (0.68)
IL-7	214.9 (136.12)	164.11 (109.8)	NS	211.96 (161.03)	164.54 (110.86)	NS	220.35 (122.75)	170.72 (111.69)	NS
IL-8	11159.01 (5785.64)	11610.46 (4198.59)	NS	9841.34 (4933.97)	11612.85 (4131.66)	NS	12291.37 (6438.25)	12024.78 (4115.77)	NS
IL-9	11.84 (12.94)	14.5 (16.44)	NS	14.82 (13.83)	16.06 (19.39)	NS	10.24 (12.34)	14.12 (14.53)	NS
CCL 17	205.06 (158.28)	98.6 (90.45)	0.001	235.67 (191.95)	118.2 (119.81)	NS (0.07)	185.21 (133.92)	87.76 (61.86)	0.01
TNF-α	20.05 (25.69)	12.65 (20.2)	0.02	26.0 (35.93)	9.43 (5.85)	NS (0.07)	16.5 (14.45)	15.47 (26.84)	NS (0.15)
TNF-β	3.26 (2.74)	2.5 (1.2)	NS	3.9 (3.62)	2.44 (0.94)	NS	2.86 (1.91)	2.64 (1.37)	NS
TSLP	42.89 (56.49)	19.93 (18.18)	0.03	55.47 (76.4)	23.66 (26.12)	NS (0.15)	31.71 (36.06)	17.81 (9.1)	NS (0.19)
VEGF	9943.77 (6885.19)	10994.2 (10197.55)	NS	9749.54 (6531.94)	11071.91 (6644.52)	NS	10114.36 (7537.63)	11416.28 (12549.39)	NS

Table E3. Missing values in clinical questionnaires.

Questionnaire (values assessed)	Missing values before imputation (n; %)	Imputed values (n; %)	Missing values after imputation (n; %)
ACT total (620)	61; 9.84%	2; 0.32%	59; 9.52%
V1 (155)	9; 5.81%	0; 0%	9; 5.81%
V2 (155)	21; 13.55%	1; 0.65%	20; 12.90%
V3 (155)	11; 7.10%	1; 0.65%	10; 6.45%
V4 (155)	20; 12.90%	0; 0%	20; 12.90%
SNOT total (2728)	106; 3.89%	32; 1.17%	74; 2.71%
V1 (682)	35; 5.13%	13; 1.91%	22; 3.23%
V2 (682)	37; 5.43%	7; 1.03%	30; 4.40%
V3 (682)	27; 3.96%	5; 0.73%	22; 3.23%
V4 (682)	7; 1.03%	7; 1.03%	0; 0%
TNSS total (496)	14; 2.82%	1; 0.2%	13; 2.62%
V1 (124)	4; 3.22%	0; 0%	4; 3.22%
V2 (124)	4; 3.22%	0; 0%	4; 3.22%
V3 (124)	5; 4.03%	0; 0%	5; 4.03%
V4 (124)	1; 0.81%	1; 0.81%	0; 0%

ACT: Excluded data from 12 visits in analysis, as referred to by the number of patients in each visit under the line plot (Figure 2E).
Values were imputed if questionnaire was >75% complete using a rationale and considering previous and following visits (last observation carried forward or mean of two visits).

SNOT: Excluded data from 4 visits in analysis, as referred to by the number of patients in each visit under the line plot (Figure 2C).
Values were imputed if questionnaire was >80% complete using a rationale and considering previous and following visits (last observation carried forward or mean of two visits).

TNSS: Excluded data from 3 visits. One value was imputed based on questionnaire >75% complete and using last observation carried forward.