Supplementary Online Content

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

eMethods 1. Biological characterization of the Adipose-Derived Stromal Vascular Fraction

Total viable nucleated cell recovery and cell viability were determined using the Nucleocounter NC100 instrument (ChemoMetec, Denmark). Cellular components within isolated Adipose-Derived Stromal Vascular Fraction (ADSVF) were identified by flow cytometry analysis (Beckman Navios instrument) according to the guidelines from the International Federation of Adipose Therapeutics and Science (IFATS) and International Society for Cellular Therapy (ISCT). A panel of makers including NucBlue as viability marker and fluorochrome-conjugated antibodies directed against cell surfaces antigens was used to characterize the different cell types present in the ADSVF (but without separating them before injection): CD14-FITC, CD90-FITC, CD146-PE, CD34-ECD, CD45-PC5. Corresponding isotype-matched control antibodies were used to determine the non-specific fluorescence. Debris, red blood cells and dead cells were excluded from the analysis. The frequency of Adipose-Derived Stem Cells (ADSC) was estimated using the Colony-Forming-Unit-Fibroblastic (CFU-F) clonogenic assay.

eMethods 2. Definition of vocal assessment's parameters

The Signal to noise ratio quantifies the aperiodic portion of the voice signal (the higher the signal the better).

The Vocal range is the difference between the maximum frequency and the minimum frequency.

The Maximum Phonation Time (MPT) and the Oral AirFlow (OAF) both quantify the glottal air leakage (if the leakage is important, MPT should be short and OAF high).

The Estimated subglottic pressure at the phonatory threshold is the minimum subglottic pressure needed to initiate and sustain vocal folds vibration.

The Voice Handicap Index (VHI) is composed of 3 parts (E = emotional subscale, F = functional subscale, P = physical subscale), with lower scores indicating a better voice perception. Patients were considered as responders if they achieved the Minimal Clinically Important Difference (MCID) defined by Jacobson as a shift in the total VHI score of at least 18[1].

SF36 is a generic quality-of-life scale consisting of 36 items describing eight dimensions (Physical Function, Social Function, Role limitations due to physical health problems, Role limitations due to emotional problems, Emotional Well-being, Vitality, Bodily Pain and General Health Perception[2]); and two summary measures: the Physical (PCS) and Mental (MCS) Component Summary scores[3]. Each dimension ranges from 0 to 100; the higher the score, the better the perceived state of health.

eTable 1. Characteristics of adipose tissue harvest and Adipose-Derived Stromal Vascular Fraction

Patient n°	Volume of adipose tissue harvested (ml)[a]	Volume of ADSVF injected in total (ml)	Number of ADSVF cells injected, after quality control (millions)	Viability (%)	*Leukocytes (%)*	*Macrophages (%)*	*Endothelial cells (%)*	*Pericytes (%)*	*Stromal cells (ADSC) (%)*	CFU-F (%)
1	135	1.00	13.60	82.5	15.7	3.8	5.2	32.2	43.1	2.8
2	340	0.90	12.16	86.0	28.5	8.2	2.7	8.3	52.3	3.4
3	100	1.25	2.63	85.8	34.5	12.8	5.6	9.5	37.7	5.3
4	190	1.00	7.20	85.5	19.3	13.0	5.1	37.1	25.5	1.3
5	100	1.00	10.30	93.4	42.8	10.1	2.2	6.0	38.8	8.0
6	190	1.00	6.48	80.6	30.0	8.7	6.8	21.8	32.7	3.9
7	120	1.00	2.23	82.1	25.1	6.8	14.1	26.5	27.7	2.2
8	190	1.00	7.80	80.0	30.0	6.6	8.8	17.6	37.0	2.7
Mean ± SD	171 ± 79	1.02 ± 0.10	7.8 ± 4.1	84.5 ± 4.3	*28.2 ± 8.5*	*8.7 ± 3.1*	*6.3 ± 3.8*	*19.9 ± 11.6*	*36.8 ± 8.6*	3.7 ± 2.1
Median	163	1.00	7.5	84.0	*29.3*	*8.5*	*5.4*	*19.7*	*37.4*	3.1
(min - max)	(100 - 340)	(0.90 - 1.25)	(2.2 - 13.6)	(80.0 - 93.4)	*(15.7 - 42.8)*	*(3.8 - 13.0)*	*(2.2 - 14.1)*	*(6.0 - 37.1)*	*(25.5 - 52.3)*	(1.3 - 8.0)

[a] net volume, after removal of infiltration liquid
ADSVF: Adipose-Derived Stromal Vascular Fraction
ADSC: Adipose-Derived Stromal / Stem cells

eTable 2. Changes in the self-evaluation, videolaryngostroboscopy, perceptive, acoustic and aerodynamic analyzes after injection of autologous ADSVF injection

		Baseline	1 month	Change (1 month-baseline)	6 months	Change (6 months-baseline)	12 months	Change (12 months-baseline)
Self Evaluation								
VHI (0-120)	Mean ± SD	79.5 ± 14.3	56.0 ± 26.0	-24.4 ± 20.5	48.1 ± 29.9	-31.4 ± 29.0	39.4 ± 28.2	-40.1 ± 21.5
	Median (range)	74.0 (61 ; 107)	64.0 (5 ; 84)	-22.0 (-56 ; -1)	45.5 (12 ; 92)	-44.5 (-62 ; 7)	37.0 (2 ; 80)	-40.5 (-66 ; -9)
VHI Functional (0-40)	Mean ± SD	22.3 ± 5.8	16.9 ± 8.8	-6.9 ± 7.1	12.4 ± 8.6	-9.9 ± 10.4	10.1 ± 9.2	-12.1 ± 7.8
	Median (range)	24.0 (12 ; 30)	21.0 (0 ; 26)	-5.0 (-16 ; 1)	12.5 (2 ; 25)	-11.0 (-22 ; 6)	6.5 (0 ; 26)	-11.0 (-22 ; -4)
VHI Physical (0-40)	Mean ± SD	30.3 ± 4.0	20.0 ± 7.1	-10.1 ± 8.7	20.1 ± 11.1	-10.1 ± 11.3	16.1 ± 9.2	-14.1 ± 8.3
	Median (range)	30.0 (27 ; 39)	21.0 (5 ; 27)	-9.0 (-25 ; 0)	19.0 (4 ; 33)	-14.5 (-26 ; 6)	16.0 (2 ; 28)	-13.0 (-28 ; -3)
VHI Emotional (0-40)	Mean ± SD	27.0 ± 10.0	19.1 ± 11.7	-7.4 ± 5.8	15.6 ± 12.6	-11.4 ± 10.1	13.1 ± 10.7	-13.9 ± 8.6
	Median (range)	27.0 (15 ; 38)	22.0 (0 ; 35)	-8.0 (-15 ; -1)	16.5 (1 ; 40)	-11.5 (-23 ; 3)	14.0 (0 ; 27)	-12.0 (-27 ; 1)
SF-36 - PCS (0-100)	Mean ± SD	51.6 ± 7.3	/	/	52.6 ± 2.7	0.9 ± 8.0	53.5 ± 4.2	1.1 ± 8.9
	Median (range)	52.4 (38.5 ; 60.8)			52.8 (48.8 ; 57.4)	-0.1 (-7.2 ; 14.3)	55.6 (46.7 ; 57.8)	1.8 (-10.6 ; 12.4)
SF-36 - MCS (0-100)	Mean ± SD	44.5 ± 13.4	/	/	40.0 ± 12.1	-3.7 ± 10.5	46.3 ± 11.3	0.5 ± 12.4
	Median (range)	50.4 (25.6 ; 59.5)			39.3 (26.4 ; 62.1)	-6.9 (-20.2 ; 10.6)	45.9 (35.0 ; 62.1)	6.5 (-24.5 ; 10.1)
Vidéolaryngostroboscopie [a]								
Glottal closure (0-3)	Mean ± SD	1.6 ± 0.9	/	/	/	/	1.0 ± 0.9	-0.6 ± 1.6
	Median (range)	2.0 (0.0 ; 3.0)					1.0 (0.0 ; 2.0)	-1 (-3.0 ; 2.0)
Regularity (0-3)	Mean ± SD	1.2 ± 0.7	/	/	/	/	0.9 ± 0.8	-0.3 ± 1.4
	Median (range)	1.3 (0.5 ; 2.0)					1.0 (0.0 ; 2.0)	0.0 (-2.0 ; 1.5)

Vibration (0-3)	Mean ± SD	1.3 ± 0.5	/	/	/	/	0.9 ± 0.8	-0.4 ± 1.2
	Median (range)	1.3 (0.5 ; 2.0)					0.8 (0.0 ; 2.0)	-0.3 (-2.0 ; 1.5)
Perceptive analysis								
Global grade (0-3)	Mean ± SD	2.1 ± 0.4	/	/	2.0 ± 0.5	-0.1 ± 0.4	1.8 ± 1	-0.4 ± 0.9
	Median (range)	2.0 (2.0 ; 3.0)			2.0 (1.0 ; 3.0)	0.0 (-1.0 ; 0.0)	2 (0.0 ; 3.0)	0.0 (-2.0 ; 1.0)
Roughness (0-3)	Mean ± SD	1.9 ± 0.6	/	/	1.5 ± 0.9	-0.4 ± 0.7	1.4 ± 1.1	-0.5 ± 0.8
	Median (range)	2.0 (1.0 ; 3.0)			1.5 (0.0 ; 3.0)	0.0 (-2.0 ; 0.0)	1.5 (0.0 ; 3.0)	0.0 (-2.0 ; 0.0)
Breathiness (0-3)	Mean ± SD	1.5 ± 1.1	/	/	1.6 ± 0.9	0.1 ± 0.4	1.5 ± 1.2	0.0 ± 0.5
	Median (range)	2.0 (0.0 ; 3.0)			2.0 (0.0 ; 3.0)	0 (0.0 ; 1.0)	1.5 (0.0 ; 3.0)	0.0 (-1.0 ; 1.0)
Acoustic analysis								
SNR (dB)	Mean ± SD	15.2 ± 4.1	9.8 ± 3.9	-0.7 ± 5.7	19.1 ± 2,6	3.9 ± 4.8	17.8 ± 1.5	2.6 ± 4.7
	Median (range)	15.9 (9.5 ; 21.3)	13.4 (9.8 ; 19.7)	-0.3 (-11.5 ; 8.4)	19.0 (15.8 ; 23.0)	3.3 (-2.3 ; 11.2)	17.9 (16.2 ; 21.1)	3.7 (-4.6 ; 8.2)
Vocal range (Hz)	Mean ± SD	380 ± 111	458 ± 101	109 ± 181	420 ± 91	99 ± 157	379 ± 84	-25 ± 158
	Median (range)	421 (209 ; 474)	456 (295 ; 593)	86 (-77 ; 384)	414 (316 ; 538)	107 (-61 ; 252)	375 (235 ; 518)	-17 (-239 ; 160)
Aerodynamic analysis								
MPT (s)	Mean ± SD	10.7 ± 6.2	10.3 ± 6.0	-0.4 ± 4.9	8.6 ± 5.5	-2.0 ± 3.6	10.6 ± 4.7	0.0 ± 4.4
	Median (range)	9.6 (3.1 ; 22.0)	10.4 (2.4 ; 21.0)	1.0 (-8.1 ; 5.0)	7.6 (3.6 ; 19.4)	-1.9 (-8.0 ; 3.4)	9.1 (3.8 ; 19.0)	1.7 (-8.4 ; 3.4)
OAF (cm^3/s)	Mean ± SD	302 ± 112	298 ± 113	-5 ± 153	344 ± 246	42 ± 169	282 ± 103	-20.6 ± 82.3
	Median (range)	296 (149 ; 519)	270 (160 ; 497)	25 (-334 ; 178)	275 (108 ; 919)	44 (-164 ; 400)	326 (138 ; 418)	-17 (-162 ; 89)
ESP (hPa)	Mean ± SD	8.0 ± 3.1	7.7 ± 2.2	-0.2 ± 2.0	8.1 ± 4.0	0.3 ± 4.3	9.6 ± 4.6	2.1 ± 3.3
	Median (range)	7.7 (5.0 ; 13.0)	7.2 (5.1 ; 11.5)	0.1 (-2.9 ; 2.7)	7.5 (3.5 ; 16.5)	0.7 (-6.6 ; 5.3)	8.3 (5.0 ; 17.0)	2.7 (-2.7 ; 5.8)

SD: Standard Deviation
PCS: Physical Component Summary score; MCS: Mental Component Summary score.
SNR: Signal-to-Noise Ratio; MPT: Maximum Phonation Time; OAF: Oral Air Flow; ESP: Estimated Subglottic Pressure at the phonatory threshold; dB: decibels; s: seconds; Hz: Hertz; hPa: hectopascals.

For videolaryngostroboscopy's parameters and GRB scale, 0 (minimum) means normal and 3 (maximum) severe disturbance.
Due to missing values: VHI at M1 (n=7), SF-36 at M6 and M12 (n=7), vocal range at baseline (n=6), M1 (n=7, n change=5), M6 (n=4, n change=3) and M12 (n change=6), and ESP at each time (n=7).
[a] Calculated with the mean of right and left vocal folds for each patient.

eTable 3. Individual patient data of vocal assessment and quality of life

Patient	Visit	Aerodynamic			Acoustic		GRB scale			Voice Handicap Index				SF-36									Videolaryngostroboscopy				
n°		OAF	ESP	MPT	VR	SNR	G	R	B	VHI	VHI_F	VHI_P	VHI_E	PF	SF	RP	RE	MH	VT	BP	GH	PCS	MCS	G	RR	RL	VR	VL
1	M0	319	13,0	3,1	460	21,3	2	1	2	85	21	27	37	85	38	100	67	24	45	74	77	61	26	2	2	1	2	1
1	M1	497	10,1	2,4	383	9,8				84	22	27	35															
1	M6	383	6,4	3,9	ND	19,0	2	1	2	92	19	33	40	90	63	100	67	20	30	41	52	54	28					
1	M12	157	15,7	3,8	329	16,7	2	0	2	66	15	24	27	80	88	100	100	24	20	42	67	50	36	2	2	1	3	1
2	M0	203	7,9	13,2	381	9,5	2	2	0	75	24	27	24	95	75	100	100	72	65	62	67	51	50	2	2	2	2	2
2	M1	243	5,6	6,9	467	17,9				68	21	25	22															
2	M6	244	4,7	8,0	ND	20,7	2	2	0	16	2	13	1	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND					
2	M12	198	7,3	7,8	460	17,7	1	1	0	9	2	7	0	100	100	100	100	92	85	100	97	58	58	0	0	0	0	0
3	M0	519	11,2	6,3	474	11,9	2	2	2	91	26	27	38	90	38	75	33	24	60	41	52	54	26	2	1	0	2	0
3	M1	185	11,5	3,7	ND	11,7				69	21	21	27															
3	M6	919	16,5	3,6	413	22,0	2	0	2	29	4	10	15	100	50	75	33	52	75	22	47	49	36					
3	M12	418	17,0	8,4	235	18,2	3	2	3	31	6	14	11	100	50	75	33	64	65	84	47	56	36	1	0	2	0	1
4	M0	272	5,0	10,9	469	17,7	2	2	2	73	12	31	30	100	100	100	100	92	95	100	92	57	59	2	2	1	2	1
4	M1	250	5,1	13,5	452	19,1				ND	ND	ND	ND															
4	M6	108	3,5	7,8	ND	23,0	2	2	2	72	18	33	21	100	50	75	100	56	25	41	82	51	39					

4	M12	344	10,8	14,3	356	16,7	2	2	2	43	7	18	18	65	50	75	67	32	45	22	82	47	35	1	0	0	1	0
5	M0	149	5,6	22,0	ND	18,2	2	2	1	71	25	30	16	85	75	0	100	68	55	62	62	39	52	1	1	0	2	0
5	M1	160	7,3	13,9	295	14,3				62	26	21	15															
5	M6	229	10,0	14,0	414	19,0	2	2	1	73	25	30	18	95	63	100	100	60	55	62	67	53	44					
5	M12	138	5,5	13,6	381	16,2	2	2	1	62	20	25	17	85	88	100	100	72	60	74	72	51	51	2	3	0	3	0
6	M0	259	7,7	5,4	286	17,0	2	1	2	61	16	30	15	100	100	100	100	84	80	100	87	57	56	3	2	2	2	2
6	M1	374	6,9	9,8	456	19,7				5	0	5	0															
6	M6	305	8,4	5,0	538	16,7	2	1	2	12	6	4	2	100	100	100	100	100	100	100	100	57	62					
6	M12	348	5,0	8,6	381	21,1	1	1	1	2	0	2	0	100	100	100	100	100	100	100	100	57	62	0	0	0	0	0
7	M0	344	5,3	8,3	209	11,1	3	3	3	107	30	39	38	61	38	100	100	16	70	41	31	46	35	0	0	1	0	1
7	M1	382	8,0	11,0	593	10,8				64	17	22	25															
7	M6	323	8,2	7,3	316	15,8	3	3	3	60	18	22	20	72	63	100	0	40	40	62	37	53	26					
7	M12	322	9,3	9,6	369	18,1	3	3	3	80	26	28	26	75	63	100	67	56	55	51	ND	ND	ND	2	2	2	2	2
8	M0	355	ND	16,0	ND	14,8	2	2	0	73	24	31	18	90	88	25	67	80	65	100	67	48	51	1	2	0	2	0
8	M1	289	7,0	21,0	561	12,4				40	11	19	10															
8	M6	242	6,7	19,4	ND	16,6	1	1	1	31	7	16	8	100	75	75	100	64	35	84	57	52	44					
8	M12	330	5,8	19,0	518	18,0	0	0	0	22	5	11	6	100	88	75	100	60	55	100	72	56	46	0	2	0	2	0

Aerodynamic analysis: OAF: oral airflow, ESP: estimated subglottic pressure, MPT: maximum phonation time; Acoustic analysis: VR: Vocal Range, SNR: signal-to-noise ratio; GRB scale: G: Global grade, R:

Roughness, B: Breathiness; VHI: Voice Handicap Index, VHI_F: functional subscale score, VHI_P: physical subscale score, VHI_E: emotional subscale score; SF-36: PF: Physical Function, SF: Social Function, RP: Role limitations due to physical health problems, RE: Role limitations due to emotional problems, MH: Emotional Well-being, VT: Vitality, BP: Bodily Pain, GH: General Health Perception, PCS: Physical Component Summary score, MCS: Mental Component Summary score; Videolaryngostroboscopy: G: Glottal closure, RR: Regularity of Right vocal fold, RL: Regularity of Left vocal fold, VR: Vibration of Right vocal fold, VL: Vibration of Left vocal fold

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