Received:         2004.01.12           Accepted:         2004.07.05           Published:         2004.12.01	Early effects of smoking on the voice: A multidimensional study
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	Summary
Background:	The purpose of this study was to evaluate the effect of tobacco on the voice in a relatively early stage of the cigarette-smoking habit (<10 years).
Material/Methods:	A multi-parameter acoustic analysis tool, the Multi-Dimensional Voice Program <sup>™</sup> (MDVP), was used to obtain a set of 27 parameters from sustained vowel phonations of 134 non-dysphonic young adults (aged 20–29) of both genders, including smokers and non-smokers.
Results:	Some voice parameters were significantly altered in young smokers when compared with young non-smokers, probably as a consequence of histological changes caused by tobacco. Main differences were observed in Frequency Perturbation parameters (jitter, sPPQ) for both genders, in Fundamental Frequency parameters (Fo, Fhi, Flo) mainly in women, and in tremor parameters (ATRI, FTRI) in men. The number of cigarettes smoked per day was related to the Fundamental Frequency values in women and FTRI in men. A discriminant analysis correctly classified 70–75% of the subjects in each gender group as smokers and non-smokers.
Conclusions:	A combination of voice parameters seems to suggest a possible neurological effect of nicotine – or some other chemical component of tobacco – on the voice.
key words:	tobacco • smoking • voice • voice parameters • voice analysis • MDVP
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# BACKGROUND

Evidence that smoking tobacco is harmful for health has accumulated over the past several decades. Cigarette smoking has been positively associated with some 40 causes of morbidity and mortality in contemporary people [1–3]. The association between tobacco use and laryngeal pathology is beyond all doubt. For example, larvngeal carcinoma is a multifactorial disease, but smoking is the most important factor associated with this type of cancer [4-6]. Other less important pathologies are also related to tobacco [7]. In a recent study of risk factors in a group of patients affected by Reinke's edema [8], the authors found that both daily cigarette consumption and the duration of exposure to cigarette smoke were the main risk factors for the presence of edema and for its recurrence. A relationship between tobacco and laryngeal epithelial changes has also been proved [9].

Although the voice problems caused by pathologies typically associated with smoking are well known, research using objective parameters focused on the relation between tobacco and the voice is very scarce [10,11]. Recently, Damborenea et al. [12] carried out an observational study of acoustic voice parameters in a sample of non-dysphonic adult smokers and non-smokers. The results showed that some parameters differed between smokers and non-smokers. The fundamental frequency was lower in smokers, and average jitter and shimmer were higher in smokers.

Damborenea's study was based on a set of only five parameters (fundamental frequency, jitter, shimmer, harmonics-to-noise ratio, and normalized noise energy). The subjects were mature adult whose average age was 38 years, and ranged in age up to 69 years. In the present study, we put the following question: Does tobacco have any effect on voice at a relatively early stage of the smoking habit? To answer this question, a multidimensional analysis was performed on voice samples of young adults between 20 and 29 years of age, using a wide set of voice acoustic parameters obtained by a computer-based system. This system (MDVP<sup>TM</sup>) was chosen because recent literature has proved its robustness and reliability [13-14] and its value as a tool in both research and clinical applications [15-21]. Furthermore, an increased effort has been made over the last few years to obtain normative data for this analysis system [13,22-26].

## **MATERIAL AND METHODS**

The subjects were 134 young adult students of both sexes at University Jaume I (Spain), of whom 53 were men (aged 20–29 years, mean=22.0 years, s.d.=2.2 years) and 81 were women (aged 20–27 years, mean=21.6 years, s.d.=1.5 years). Of the males, 28 were non-smokers and 25 smokers (12 subjects smoked 10 or less cigarettes per day, and 13 subjects smoked more than 10 cigarettes per day). There were 40 female non-smokers and 41 smokers (23 subjects smoked 10 or less cigarettes per day, and 18 subjects smoked nore than 10 cigarettes per day. All these subjects participated in this study voluntarily.

Voice samples were recorded with a Shure SM58 microphone at a distance of about 15 cm from the mouth. The voice parameters were extracted with the Multi-Dimensional Voice Program<sup>™</sup> (MDVP) model 4305 from Kay Elemetrics Corporation [27], implemented in a Computerized Speech Lab (CSL model 4300B, Kay Elemetrics Corp, Pine Brook, New Jersey, USA).

In compliance with the MDVP operations manual, the speakers were asked to produce a sustained phonation of the vowel /a/ for 3 seconds at a comfortable pitch and loudness. The subjects were instructed to maintain as steady a phonation as possible. MDVP software can only work with two sample frequencies: 25 or 50 kHz. In this experiment, all voice samples were recorded at 50 kHz and directly stored in the host computer. The samples were recorded in a sound-proof room at the University Laboratories.

All voice samples were analyzed with the MDVP software and the following parameters were obtained:

- 1. Fundamental frequency parameters: Average Fundamental Frequency (Fo), i.e. the average value of all extracted period-to-period fundamental frequency values; Highest Fundamental Frequency (Fhi), i.e. the greatest of all extracted period-to-period fundamental frequency values; Lowest Fundamental Frequency (Flo), i.e. the lowest of all extracted values; Standard Deviation of Fo (STD), i.e. the SD of all extracted values; and Phonatory Fo-Range in semi-tones (PFR) for all extracted pitch periods. Voice break areas were excluded in identifying all frequency parameters.
- 2. Frequency perturbation parameters: Absolute Jitter (*Jita*)/µs/gives an evaluation in microseconds (µs) of the period-to-period variability of the pitch period within the analyzed voice sample. This measure is widely used in voice research [28] and is very sensitive to pitch variations occurring between consecutive pitch periods. Jitter Percent (Jitt) /%/: the relative period-to-period variability of the pitch period. Relative Average Perturbation (RAP) /%/: Introduced by Koike [29] this parameter gives the relative evaluation of the period-to-period variability of the pitch with a smoothing factor of 3 periods. Pitch Perturbation Quotient (PPQ) /%/: Introduced by Koike, Takahashi, and Calcatera [30], this gives the variability of the pitch period at a smoothing factor of 5 periods. Smoothed Pitch Perturbation Quotient (sPPQ) /%/: An evaluation of the long term variability of the pitch period within the analyzed voice sample, with a smoothing factor of 55 periods. RAP, PPQ and sPPQ have been extensively used in the last decade, since they are less sensitive to pitch extraction errors due to smoothing in their calculation. Fundamental Frequency Variation (vFo) /%/: The relative standard deviation of the fundamental frequency. It reflects the very long term variation of Fo within the analyzed voice sample. Any variations in the fundamental frequency are reflected in vFo, and this parameter increases regardless of the type of pitch variation, whether it be of the random or regular fluctuating type.
- **3. Amplitude perturbation parameters:** *Shimmer in dB* (*ShdB*) /dB/: Evaluation in dB of the period-to-period variability of the peak-to-peak amplitude within the analyzed voice sample. As in other parameters, voice break areas are excluded. As occurs with jitter, this parameter has been widely used in voice research. *Shimmer Percent*

(*Shim*) /%: Relative evaluation of the period-to-period variability of the peak-to-peak amplitude. *Amplitude Perturbation Quotient (APQ)* /%/: Introduced by Koike et al. [29], this gives a relative evaluation of the variability of the peak-to-peak amplitude at a smoothing of 11 periods. The smoothing reduces the sensitivity of APQ to pitch extraction errors. *Smoothed Amplitude Perturbation Quotient (sAPQ)* /%/: Evaluation of the long-term period-to-period variability of the peak-to-peak amplitude at a smoothing of 55 periods. *Peak-Amplitude Variation (vAm)* /%/: This gives the relative standard deviation of period-to-period calculated peak-to-peak amplitude, reflecting the very long term amplitude variations within the analyzed voice sample.

- 4. Noise parameters: Noise to Harmonic Ratio (NHR): A general evaluation of noise presence in the analyzed signal (such as amplitude and frequency variations, turbulence noise, subharmonic components or voice breaks). This is the ratio of non-harmonic energy in the range 1500-4500 Hz to the harmonic spectral energy in the range 70-4500 Hz. Voice Turbulence Index (VTI): Ratio of the non-harmonic energy in the range 2800-5800 Hz to the harmonic spectral energy in the range 70-4500 Hz. This parameter measures the relative energy level of high frequency noise, a new attempt to compute "breathiness" in the voice signal. Soft Phonation Index (SPI): Ratio of the harmonic energy in the range 70-1600 Hz to the harmonic energy in the range 1600-4500 Hz. This is very sensitive to the vowel formant structure. This parameter is not actually a measurement of noise, but its formula is similar to the above two parameters and is therefore, as in the MDVP manual, listed in the same category.
- 5. Tremor parameters: Fo-Tremor Frequency (Fftr) /Hz/: This shows the frequency of the most intensive low frequency Fo-modulating component in the tremor range. Amplitude Tremor Frequency (Fatr) /Hz/: This shows the frequency of the most intensive low frequency amplitude-modulating component in the tremor range. Frequency Tremor Intensity Index (FTRI) /%/: Ratio of the frequency magnitude of the most intensive low-frequency modulating component (Fo- tremor) to the total frequency magnitude of the analyzed signal. Amplitude Tremor Intensity Index (ATRI) /%/: Ratio of the amplitude of the most intensive low-frequency amplitude modulating component (amplitude tremor) to the total amplitude of the analyzed signal.
- 6. Parameters of Subharmonic components: Number of Subharmonic Segments (NSH): Number of subharmonic segments found during analysis. Degree of Subharmonics (DSH) /%/: Relative evaluation of subharmonic to Fo components in the analyzed sample.
- 7. Parameters of Voice irregularities: Number of Unvoiced Segments (NUV): Number of unvoiced segments detected during the analysis. Degree of Voiceless (DUV) /%/: Relative evaluation of non-harmonic areas in the voice sample.

## RESULTS

Table 1 shows the means and standard deviations of the voice parameters obtained with MDVP from smoking and non-smoking subjects, separated by gender. Comparisons

between smokers and non-smokers were made using the ttest (one-tailed), and the marginal 0.10 level was also examined. In general, the fundamental frequency parameters were clearly affected by smoking, especially in women. The mean fundamental frequency (Fo) of smokers was lower than in non-smokers. This difference reached a value of marginal significance of only p<0.10 in men (125.4 Hz for non-smokers vs. 119.4 Hz for smokers), but in women the difference (206.4 Hz for non-smokers vs. 192.4 Hz for smokers) was significant at p<0.01. The parameters Highest Fundamental Frequency (Fhi) and Lowest Fundamental Frequency (Flo) also showed lower values in smokers, and more so among women. Conversely, the variability of the fundamental frequency of voice - Standard Deviation of Fo (STD) and Phonatory Fo-Range in semi-tones (PFR) for all extracted pitch periods - seemed higher for smokers, but only approached significance at the level of 0.10 in men.

**Frequency perturbation parameters**, mainly *jitter* and *Smoothed Pitch Perturbation Quotient* (sPPQ), were affected in the voices of young smokers (Figure 1). Jitter was higher in smokers, especially in men (47.67 µs for non-smokers vs. 62.78 µs for smokers, p<0.05). The values of the sPPQ parameter were significantly higher in smoker subjects, both in men (0.61% in non-smokers vs. 0.74% in smokers, p<0.05) and in women (0.69% in non-smokers vs. 0.85% in smokers, p<0.05). The parameter *Fundamental Frequency Variation* (vFo), or the relative standard deviation of the fundamental frequency, was also higher in smokers vs. 1.25% in smokers, p<0.05).

The **Amplitude perturbation parameters**, however, do not seem to be affected by tobacco in an early stage of the smoking habit. Broadly speaking, the values were not significantly different for smokers and non-smokers, with the exception of the *Smoothed Amplitude Perturbation Quotient* (sAPQ), which reached a level of marginal significance of 0.10 in men. None of the **noise** parameters (NHR, VTI, SPI), **subharmonic components** (DSH, NSH) or **voice irregularities** (DUV, NUV) seemed to be affected by early action of tobacco consumption. Nevertheless, the tremor parameters FTRI (*Frequency Tremor Intensity Index*) and ATRI (*Amplitude Tremor Intensity Index*) seemed to be significantly modified by tobacco only in men (FTRI: 0.34% in non-smokers vs. 0.45% in smokers, p<0.05; ATRI: 3.15% in non-smokers vs. 4.54% in smokers, p<0.05).

In order to study the extent to which the voice parameters of young smokers differed from those of young non-smokers, a linear Discriminant Analysis was conducted across subjects within each gender; this analysis made use of voice parameters as discriminant variables. The greater the difference in the voice parameters between smokers and nonsmokers, the greater the efficiency of the parameters as prediction variables in the classification of each voice sample. The results of this analysis are shown in Table 2. It can be seen that the percentage of correct classification between smokers and non-smokers was 73.6% in men (Wilk's lambda 0.82, p<0.05), and 70.4% (Wilk's lambda 0.94, p<0.05) in women. When considering the variables that had a greater influence on the discriminant function, - i.e. the highest correlations, in absolute values, between MDVP parameters and discrimination function - we can see the emergence of the parameters concerned with the measurement of tremor

(a)	Non-S Total	mokers (n=68)	Smoke (n=	ers Total =66)	Non-Smoker Males (n=28)		Smoker Males (n=25)		Non-Smoker Females (n=40)		Smoker Females (n=41)			
				Fundam	ental Freq	luency para	ameters	:						
Fo (Hz)	173.0	(45.5)	164.7	(40.1)	125.4	(13.9)	119.4	(13.9)+	206.4	(25.5)	192.4	(20.7)**		
Fhi (Hz)	183.9	(51.7)	174.6	(44.6)	129.6	(14.8)	125.1	(16.7)	221.9	(28.9)	204.8	(24.7)**		
Flo (Hz)	162.6	(41.6)	153.2	(39.2)	120.7	(13.3)	113.4	(13.3)*	191.9	(26.5)	179.1	(26.7)*		
STD (Hz)	2.10	(1.19)	3.14	(8.21)	1.26	(0.30)	1.49	(0.76)+	2.69	(1.22)	4.14	(10.32)		
PFR (semit.)	3.04	(1.66)	3.18	(2.68)	2.32	(0.86)	2.76	(1.39)+	3.55	(1.89)	3.44	(3.22)		
Frequency Perturbation parameters:														
<b>Jita</b> (us)	46.45	(24.86)	58.02	(38.16)*	47.67	(18.39)	62.78	(41.22)*	45.60	(28.74)	55.11	(36.39)+		
Jitt (%)	0.78	(0.45)	0.93	(0.61)+	0.60	(0.24)	0.74	(0.50)+	0.91	(0.52)	1.04	(0.65)		
<b>RAP</b> (%)	0.47	(0.29)	0.55	(0.38)+	0.35	(0.15)	0.43	(0.31)	0.55	(0.33)	0.62	(0.40)		
<b>PPQ</b> (%)	0.46	(0.26)	0.54	(0.36)+	0.35	(0.14)	0.44	(0.29)+	0.54	(0.30)	0.60	(0.39)		
sPPQ (%)	0.66	(0.23)	0.81	(0.45)**	0.61	(0.12)	0.74	(0.29)*	0.69	(0.28)	0.85	(0.53)*		
<b>vFo</b> (%)	1.18	(0.51)	1.87	(4.71)	1.00	(0.19)	1.25	(0.57)*	1.31	(0.62)	2.25	(5.95)		
Amplitude Perturbation parameters:														
ShdB (dB)	0.34	(0.15)	0.34	(0.14)	0.33	(0.13)	0.31	(0.13)	0.35	(0.15)	0.36	(0.14)		
Shim (%)	3.88	(1.60)	3.91	(1.54)	3.83	(1.54)	3.61	(1.56)	3.92	(1.66)	4.05	(1.52)		
<b>APQ</b> (%)	2.93	(1.07)	3.00	(1.08)	2.98	(1.06)	3.01	(1.20)	2.89	(1.10)	3.00	(1.02)		
sAPQ (%)	4.93	(1.85)	5.30	(1.70)	4.62	(1.41)	5.22	(1.92)+	5.15	(2.10)	5.34	(1.57)		
<b>vAm</b> (%)	14.23	(7.13)	14.72	(6.76)	11.05	(5.08)	13.05	(6.63)	16.46	(7.56)	15.74	(6.72)		
					Noise pa	rameters:								
NHR	0.13	(0.02)	0.13	(0.02)	0.14	(0.02)	0.14	(0.01)	0.12	(0.02)	0.13	(0.03)		
VTI	0.05	(0.01)	0.05	(0.02)	0.05	(0.01)	0.06	(0.02)	0.05	(0.01)	0.05	(0.01)		
SPI	8.48	(4.78)	8.77	(4.42)	9.05	(3.33)	8.78	(3.47)	8.07	(5.58)	8.76	(4.95)		
					Tremor pa	arameters:								
Fftr (Hz)	2.88	(2.72)	2.99	(2.93)	2.72	(2.81)	2.86	(2.80)	2.99	(2.67)	3.07	(3.04)		
Fatr (Hz)	2.58	(1.43)	2.75	(1.64)	2.54	(1.82)	2.71	(1.79)	2.60	(1.11)	2.78	(1.55)		
<b>FTRI</b> (%)	0.38	(0.18)	0.58	(1.22)+	0.34	(0.15)	0.45	(0.22)*	0.40	(0.19)	0.65	(1.53)		
ATRI (%)	4.38	(2.58)	4.83	(2.59)	3.15	(1.17)	4.54	(3.08)*	5.21	(2.93)	5.00	(2.26)		
	Parameters of Subharmonic components:													
<b>DSH</b> (%)	0.64	(3.10)	0.71	(2.82)	0.11	(0.42)	0.04	(0.21)	1.02	(4.01)	1.12	(3.53)		
NSH	0.59	(2.97)	0.68	(2.72)	0.11	(0.42)	0.04	(0.20)	0.93	(3.85)	1.07	(3.40)		
	Parameters of Voice irregularities:													
<b>DUV</b> (%)	0.51	(3.01)	0.06	(0.30)	1.02	(4.62)	0.08	(0.29)	0.16	(0.69)	0.05	(0.32)		
NUV	0.50	(2.95)	0.06	(0.30)	1.00	(4.53)	0.08	(0.28)	0.15	(0.66)	0.05	(0.31)		

Table 1. Means and standard deviations (in parentheses) of the voice parameters obtained in MDVP from Smoker and Non-Smoker subjects.

(a) Parameter abbreviations are explained in the text;

+ mean difference between Smokers and Non-Smokers at a significance value p<0.10 (t-test, one tail);</li>
 \* mean difference between Smokers and Non-Smokers at a significance value p<0.05 (t-test, one tail);</li>
 \*\* mean difference between Smokers and Non-Smokers at a significance value p<0.01 (t-test, one tail);</li>





**Table 2.** Discriminant analysis within gender between voices<br/>of Smokers and Non-Smokers utilizing MDVP parameters<br/>as discriminating variables. The six highest correlations<br/>(in absolute values) between variables and discrimination<br/>function are presented. The last row shows the percentage<br/>of correct classification of voices.

(a)	Men	(n=53)	Women (n=81)			
Correlations variables vs discrimination function	ATRI sPPQ vFo Jita FIo FTRI	(0.430) (0.378) (0.357) (0.335) (0.331) (0.323)	Fhi         (-0.480)           Fo         (-0.427)           sPPQ         (0.336)           Flo         (-0.286)           PFR         (-0.257)           Jita         (0.241)			
Correct classification	7.	3.6%	70.4%			

<sup>(a)</sup> Parameter abbreviations are explained in the text

(only in men), **fundamental frequency** (mainly in women), and **frequency perturbation** (both genders).

To study the effect that the number of cigarettes smoked per day has on voice, smokers of each gender were separated into two groups: subjects that smoked 10 or less cigarettes per day, and subjects that smoked more than 10 cigarettes per day. Statistical tests revealed significant differences in fundamental frequency parameters for women smokers (Figure 2). Mean Fundamental Frequency (Fo) was 199.5 Hz for women who smoked 10 or less cigarettes per day ( $\leq 10W$ ), whereas it was 183.2 Hz for women who smoked more than 10 cigarettes per day (>10W), the difference being significant [t(39)=2.68; p<0.01]. A one-way between-subject analysis of variance (ANOVA), including the group of women non-smokers (0 cigarettes per day), revealed a main effect for the number of cigarettes smoked per day [*F*(2,78)=6.51, p<0.01]. Furthermore, Fhi and Flo parameters were significantly different for ≤10W and >10W



Figure 2. Mean values (Hz) of *Fhi* (Highest Fundamental Frequency), Fo (Average Fundamental Frequency), and *Flo* (Lowest Fundamental Frequency) as a function of the number of cigarettes smoked per day. Data separated by gender. (≤10): subjects who smoked 10 or less cigarettes per day. (>10): subjects who smoked more than 10 cigarettes per day.

[Fhi: 211.9 Hz vs. 195.9 Hz, t(39)=2.15, p<0.05; Flo: 188.6 Hz vs. 166.9 Hz, t(39)=2.79, p<0.01]. The ANOVA showed a main effect for the number of cigarettes smoked per day for Fhi [F(2,78)=6.10, p<0.01] and Flo [F(2,78)=6.12, p<0.01] parameters.

For men, fundamental frequency parameters were not affected by the number of cigarettes smoked per day. Fo, Fhi, and Flo values (Figure 2) were not significantly different for men who smoked 10 or less cigarettes per day ( $\leq 10M$ ) than for men who smoked more than 10 cigarettes per day (>10M). The only voice parameter that showed a (marginally) significant difference in men according to the number of cigarettes was the tremor parameter FTRI (Figure 3). FTRI was 0.36% for  $\leq 10M$  and 0.53% for >10M[t(22)=-1.85; p=0.07]; when the group of male non-smokers was included, an ANOVA showed a main effect for the number of cigarettes smoked per day [F(2,49)=4.75, p<0.05].

## DISCUSSION

The purpose of the present study was to determine whether tobacco consumption had any effect on voice at a relatively early stage of the smoking habit and, if so, to quantify this effect by means of objective measures provided by voice parameters. All the subjects studied were young adults of both sexes between 20 and 29 years of age (86% of the subjects were



Figure 3. Mean values of Frequency Tremor Intensity Index (*FTRI*) (%) for men as a function of the number of cigarettes smoked per day. Errors bars indicate ±1 standard error of the mean.

20-23 years old), and most of them had a smoking history shorter than one decade. At the same time, our interest was centered on using a very wide set of parameters classified in different voice dimensions, all of which had been extracted from a single vocalization. The advantage of a multiple parameter extraction is that different parameters are important for the analysis of different pathologies; if tobacco has produced some quantifiable early effect on the voice, the more comprehensive the parameter set used, the more likely it is to be detected. The multi-parameter system selected was the Multi-Dimesional Voice Program, or MDVP™, produced by Kay Elemetrics, because it is increasingly used in both research and clinical applications, and at the present moment it is a promising assessment tool. As stated by Kent et al. in a very recent study [19], "because MDVP holds the promise of standardized and rapid assessment of voice, it is of particular interest as a potential tool for the characterization of a voice disorder" (p.283).

Our results show that tobacco has an effect on some voice parameters in a relatively early stage of the smoking habit. It is clear that the **Fundamental Frequency Parameters** were affected by smoking, especially in women. The fundamental frequency (Fo) of voice from young women smokers was on average 14 Hz lower than in young women non-smokers – significant at the level of p<0.01. Moreover, the parameters *Highest Fundamental Frequency* (Fhi) and *Lowest Fundamental*  Frequency (Flo) showed significantly lower values in female smokers (on average 17 and 13 Hz, respectively). In addition, the number of cigarettes smoked per day had a clear effect on the fundamental frequency parameters of women's voices. Females who smoked more than 10 cigarettes per day showed a mean Fo 16 Hz lower than females who smoked 10 or less cigarettes per day. Also, Fhi and Flo parameters were significantly different between the two groups of women smokers. On the other hand, tobacco had a less clear effect on fundamental frequency parameters in young men. The Fo of voice from young male smokers was on average 6 Hz lower than in young male non-smokers, but this difference only reached marginal significance (p<0.10). The difference was clearer in the Lowest Fundamental Frequency (Flo) parameter, which dropped to 113.4 Hz for smokers, in contrast to 129.7 Hz for non-smokers - significant at the level of p<0.05. However, the number of cigarettes smoked daily by young men seems to have no effect on fundamental frequency parameters. Fo, Fhi, and Flo parameters were not significantly different for men who smoked more than 10 cigarettes per day versus men who smoked 10 or less cigarettes per day.

Previous literature has demonstrated the effects of long-term cigarette smoking on laryngeal health and the fundamental frequency values. Sorensen and Horii [10] found a significant difference between the Fo of male smokers (lower Fo) and male non-smokers in oral reading and spontaneous speech tasks. Differences between the Fo values of female smokers and non-smokers were not significant, but the same trend was evident. Results of the Fo analysis of sustained vowel phonations were not significant, but showed the same trend, that is, smokers had lower fundamental frequency values than non-smokers. Recently, Damborenea et al. [12] have found that Fo of the sustained vowel /a/ was lower for smokers than non-smokers (115 vs. 129 Hz for males, and 185 vs. 201 for females). Most of these subjects, who were mature adults with an average age of 38 years and ages that ranged up to 69 years old, had a long-term smoking habit.

It is believed that the reduction of the mean fundamental frequency of voice associated with smoking is a result of edema of the vocal folds caused by tobacco. In particular, Reinke's edema has been associated typically with smoking [8,31,32] and sometimes with vocal abuse and gastroesophageal reflux. Reinke's edema is an accumulation of fluid underneath the lining of the vocal fold - i.e. Reinke's space, or lamina propria - that generally causes extensive swelling and enlargement of the vocal folds. Individuals with Reinke's edema tend to have low-pitched voices (low Fo) and this is especially obvious in the female voice, which is typically higher in pitch. The low pitch results from the increased mass and size, which causes the vocal folds to vibrate at a lower frequency. Curiously, the pitch-lowering effect of cigarette smoking may be partly reversed after as few as 40 hours of smoking cessation, as Murphy and Doyle [11] demonstrated in a study with two subjects. These authors investigated Fo changes during smoking and no-smoking periods. Voice analyses were performed before, during, and after a 40-hour period of no-smoking, and the results showed a rise in Fo during the no-smoking period.

A long history of vocal-fold inflammation by tobacco and other factors is frequently made evident by a very lowpitched voice. Some female voices display such low pitches that women are driven to seek medical attention or surgical management more often than men [33]. According to our data, even a smoking habit of relatively short duration – less than a decade – seems to be enough to cause some degree of edema and, consequently, to reduce the vibration frequency of the vocal cords. This is particularly obvious in young women, in whom the reduction in hertz is similar to the reduction displayed by women with a longer history of smoking.

Scientific literature suggests a very clear relationship between changes of vocal cord epithelia and the quantity of tobacco consumed over a lifetime [34]. There is a significant difference in the thickness of the epithelium of the supraglottic region and vocal cords of smoking and drinking patients, when compared with the same tissues of nonsmokers and non-drinkers [9]. Furthermore, there is a significant difference in these tissues in heavy smokers, when compared with light smokers. The histological damage caused by tobacco may be acoustically evidenced by means of several voice parameters. The most obvious is the aforementioned lowering of the main fundamental frequency parameters, but the histological changes could alter other parameters. Zeitels et al. [33] found that patients with Reinke's edema (not necessarily caused by tobacco consumption) had lower Fo, and higher percentages of jitter and shimmer than normal subjects. Surgical reduction of the volume of the superficial lamina propia resulted in a significant elevation in Fo and improvement in perturbation measures. Damborenea et al. [12] studied five voice parameters in a sample of non-dysphonic mature adult subjects, including smokers and non-smokers. Acoustic analysis was performed with the Dr Speech Science.3.0 software on a sustained vowel /a/ recorded from each subject. The authors found that three parameters differed between smokers and non-smokers: the mean Fo was lower, and jitter (%) and shimmer (%) were higher in smokers than in non-smokers. The other two parameters, HNR (Harmonic-to-Noise Ratio), and Normalized Noise Energy (NNE) did not differ significantly. It is probable that the histological changes caused by tobacco smoke, particularly the more or less pronounced swelling and mass increase of the vocal folds caused by the edema, may have consequences on the regularity of vibration, thus increasing the Frequency-Perturbation and Amplitude-Perturbation measurements.

A relative short history of smoking - less than a decade - seems to be enough to significantly increase the periodto-period variability of the pitch period, but not the period-to-period variability of the peak-to-peak amplitude. Our data show that some Frequency Perturbation Parameters are affected in the voice of young smokers, i.e. jitter, vFo and, mainly, the Smoothed Pitch Perturbation Quotient (sPPQ). The sPPQ parameter gives an evaluation of the long-term variability of the pitch period, with a smoothing factor of 55 periods. A smoothing factor allows a parameter to be less sensitive to the short-term, usually random, variations occurring between consecutive pitch periods, and to be sensitive mostly to medium or long-term variations (depending on the factor). Our sPPQ values are significantly higher in smokers than in non-smokers (0.81% vs. 0.66%; p < 0.01), both in men (0.74% vs. 0.61%; p<0.05) and women (0.85% vs. 0.69%; p<0.05), although without exceeding the threshold of normality (1.02%, according to the MDVP Manual).

Relatively few studies have been conducted on the sPPQ parameter, but it seems that it is more sensitive than others to the effect of smoking on voice. The smoothing factor can be defined by the user (we used the default value, 55 periods). With a smoothing factor of 1 period, sPPQ is identical to Jitter Percent (Jitt). With a smoothing factor of 3, sPPQ is identical to the Relative Average Perturbation (RAP). With a smoothing factor of 5, sPPQ is identical to the Pitch Perturbation Quotient (PPQ). According to the information supplied in the MDVP Manual, at high smoothing factors (55 periods or more) sPPQ correlates with the intensity of the long-term pitch period variations. Studies of patients with Spasmodic Dysphonia [35] show that sPPQ with a smoothing factor set in the 45-65 period range has increased values when there are regular long-term pitch variations (frequency voice tremors). Finally, the parameter Fundamental Frequency Variation (vFo) is also higher in smoker subjects, but it only reaches significance for males.

The Amplitude perturbation parameters do not seem to be affected by tobacco, with the exception of the Smoothed Amplitude Perturbation Quotient (sAPQ, with a smoothing factor of 55 periods), which reaches a level of marginal significance for men. Neither does any parameter of noise (NHR, VTI, SPI), subharmonic components (DSH, NSH) or voice irregularities (DUV, NUV) seem to be affected by the early effect of tobacco consumption. However, the tremor parameters FTRI (Frequency Tremor Intensity Index) and ATRI (Amplitude Tremor Intensity Index) seem to be significantly increased by smoking in young men. In fact, ATRI and FTRI are included among the six variables with the highest correlations with the discriminant function between the voices of male smokers vs. male non-smokers. FTRI is the ratio of the frequency magnitude of the most intensive low-frequency modulating component (Fo tremor) to the total frequency magnitude of the analyzed signal. ATRI is the ratio of the amplitude of the most intensive low-frequency amplitude modulating component (amplitude tremor) to the total amplitude of the analyzed signal. The algorithm for tremor analysis determines the strongest periodic frequency and amplitude modulation of the voice. Tremor has both frequency and amplitude components; the relative intensity of both components is given by FTRI and ATRI, respectively. According to the MDVP Manual, the intensity and the regularity of the frequency tremors can be assessed by means of the tremor parameters and by using sPPQ (55 periods) in combination with vFo. If both sPPQ and vFo are high, there is a long-term periodic pitch variation, most likely a frequency tremor. Pathological values of these parameters are found in neurological voice disorders, such as Spasmodic Dysphonia [35]. Our young subjects have normal values in these parameters, but some differences emerge between smokers and non-smokers, especially in men. The fact that both tremor parameters (ATRI and FTRI) showed higher values for male smokers, along with the fact that the sPPQ, vFo and sAPQ are also higher (the latter only marginally) for smokers, suggests a possible neurological effect of nicotine or another chemical component of tobacco. Nevertheless, without concurrent physiological data, this hypothesis must be considered only tentative and these data should be interpreted with caution, since the tremor parameters have the lowest reliability of all MDVP parameters [13].

An additional question is why tremor parameters are significantly different only in men. Would a hypothetical neurological effect of nicotine – or other components of tobacco – be evident only in men? FTRI and ATRI have significantly higher values in male smokers compared with male nonsmokers (FTRI: 0.45 vs. 0.34%; ATRI: 4.54 vs. 3.15%), but in both cases they can be considered to be normal (for a summary of normative data see [19]). Female smokers do not have significantly higher FTRI/ATRI values than female non-smokers, but in both groups values are higher than in men (and more variable). Variability and a ceiling effect could be responsible for obscuring some differences in the tremor parameters within the female group.

A general picture is given by the results of the Discriminant Analysis performed within each gender group using the MDVP parameters as predictors. The percentage of correct classification between voices of smokers vs. non-smokers is in the 70–75% range for both genders. Considering the MDVP parameters that are better predictors of the discriminant function, we can observe that the **frequency perturbation parameters** are good predictors in both genders; **fundamental frequency parameters** are good predictors in young women, and **tremor parameters** are good predictors in young men.

#### **CONCLUSIONS**

In summary, our results show that a short duration of the smoking habit – less than a decade – has a clear effect on some voice parameters. Fundamental Frequency Parameters (Fo, Fhi, Flo) were lowered by smoking, mainly in women. The number of cigarettes smoked per day displayed a linear effect on these parameters in the female group. Frequency Perturbation Parameters (jitter, vFo, SPPQ) were significantly higher for the smoker participants. Specifically, sPPQ (smoothed Pitch Perturbation Quotient, with a smoothing factor of 55 periods) seems to be more sensitive than others to the effect of smoking on voice. Finally, vocal Tremor Parameters (FTRI, ATRI) seem to be significantly increased by smoking in young men.

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