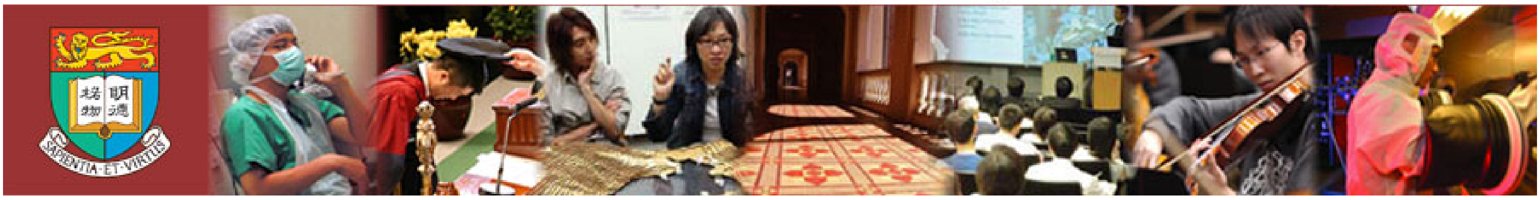

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Effect of a Chinese herbal tonic, 'watermelon frost', in preventing vocal fatigue

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ABSTRACT

This study aims at investigating the prophylactic use of 'watermelon frost' in preventing vocal fatigue, which is increasingly common among professional voice users such as teachers, singers and amateur karaoke singers. It is common to use the Chinese herbal tonic, 'watermelon frost' in treating acute pharyngitis and laryngitis in the Chinese community. Eighteen male and eighteen female participants (aged 17 - 24) were recruited and randomly allocated to either an experimental group which received 'watermelon frost', or to a placebo group which received 'wheat germ E powder' consecutively for seven days. All participants then took 120 minutes of continuous karaoke singing. The voice quality, as measured by phonetogram, fundamental frequency, jitter percentage, shimmer percentage, noise-to-harmonic ratio, subjective self-ratings of vocal qualities and functioning and perceptual evaluation did not show any significant changes after singing in both groups. The results do not provide evidence that 'watermelon frost' is effective in maintaining vocal qualities and functioning during the 120 minutes of singing.

INTRODUCTION

Vocal fatigue is commonly complained by individuals with voice disorders and those who need prolonged use of their voices (Kelchner, Lee & Stemple, 2003). Professional voice users such as singers, teachers and actors are particularly vulnerable to vocal fatigue (Welham & Maclagan, 2003). Karaoke-singing is a popular entertainment in Hong Kong among teenagers and adults. It requires extra vocal demands in pitch and phonation range control by these amateur singers, which are potentially damaging to vocal functions (Yiu & Chan, 2003). Vocal fatigue has been attributed to the excessive amount of voice use (Colton & Casper, 1996). Currently, there is not a clear definition of vocal fatigue in the literatures, although it is usually described symptomatically (Welham & Maclagan, 2003). Symptoms such as increased phonatory effort for continuing voice use, feeling of tiredness, harshness, soreness and tightness in the throat, talking in monopitch, deterioration if voice quality at the end of the day, and decreased flexibility in controlling pitch and loudness are reported (Colton & Casper, 1996; Gotaas & Starr, 1993; Kitch & Oates, 1994).

Auditory-perceptual findings of vocal fatigue

In addition to subjective descriptions, a number of studies have attempted to relate vocal fatigue with both auditory-perceptual and objective measurements. Induced vocal fatigue following prolonged vocal loading tasks was found to be associated with auditory-perceptual, acoustic, aerodynamic, larynscopic and video-stroboscpic changes. However, the findings are mostly inconclusive. Generally, an increased habitual fundamental frequency (F_0) was found from studies employing vocal loading tasks which lasted for 1 to 2 hours (Gelfer, Andrews &

Schmidt, 1991; Stemple, Stanley & Lee, 1995; Vilkman, Lauri, Alku, Sala & Sihvo, 1999). This is contrary to the findings of an early study conducted by Neils and Yairi (1987; cited in Welham et al., 2003) who found no significant change in habitual F_0 after 45 minutes of loud reading.

Acoustic findings related to vocal fatigue

The acoustic findings of vocal fatigue also vary among the literatures. Some studies (Burzynski & Titze, 1986; cited in Welham et al, 2003; Tirize & Scherer et al, 1991; cited in Welham et al, 2003) reported no significant changes in the perturbation measures: jitter, shimmer or harmonics-to-noise ratio (HNR). Verstraete, Forrez, Mertens and Debruyne (1993) also found no significant changes in jitter and shimmer in sustained vowel phonation at different fundamental frequencies after 25 minutes. On the contrary, Gelfer et al. (1991) found significant difference in jitter ratio in vowel prolongation in trained singers and lowered signal-to-noise ratio (SNR) in the untrained group after an hour-long reading task. Yiu and Chan (2003) also found a significant increase in jitter in their male participants and the highest fundamental frequency was significantly reduced in female participants of the non-hydration group after 86 minutes of karaoke-singing. The difference in findings may reveal that the amount of time and the intensity levels during vocal loading as important factors in measuring vocal fatigue. The participants in the study reported by Titze and his colleagues read aloud at their comfortable loudness. Also, participants in a study by Verstraete et al. (1993) phonated for a relatively short period of time (25 minutes) as compared with those in the studies of Gelfer et al. (60 minutes) and Yiu and Chan studies

(100 minutes). It is possible that the differences in these acoustic findings arose from the varied methodologies that were used to induce vocal fatigue. Therefore, the control of vocal intensity and vocal loading time is important in the investigation of vocal fatigue.

Aerodynamic findings related to vocal fatigue

The aerodynamic findings of vocal fatigue have also been investigated in a number of studies. Airflow rates and sub-glottal pressure have been employed to indicate vocal fatigue (e.g. Eustace, Stemple and Lee, 1996; Solomon & DiMattia, 2000; Solomon, Glaze, Arnold & van Mersbergen, 2003; and Chang & Karnell, 2004). Solomen et al. (2000, 2003) identified an increase in the phonation threshold pressure (PTP) which was defined as "the minmal lung pressure required to initiate and sustain vocal fold oscillation" with induced vocal fatigue. Chang and Karnell (2004) investigated the correlation between PTP and perceived phonatory effort (PPE). They identified a direct and moderately strong correlation between PTP and PPE in a 2-hour vocal fatiguing reading task, for the comfortable and low-pitch speaking condition (r=0.91) and the high-pitch condition (r=0.82).

Videostroboscopic findings related to vocal fatigue

Videostroboscopic findings reported in the literatures are contradictory regarding the relationship between vocal fatigue and the changes in the laryngeal appearance. Stemple et al (1995) identified anterior glottal chinks and incomplete glottal closure in their subjects after two hours of reading. A follow-up study conducted by Eustace et al. (1996) also found anterior, anterior and posterior and spindle-shaped glottal chinks. The study of Soloman and DiMatta (2000) also revealed a spindle-shaped vibratory closure pattern in three of four subjects in at least 1 second of the phonatory cycle.

The equivocal findings highlight the difficulties in quantifying vocal fatigue using objective measures alone. As suggested by some investigations, the use of both perceptual and objective measures and the evaluation of voice qualities in a multiparametric approach is advocated. (Ma & Yiu, in press; Yiu, Yuen, Whitehill and Winkworth, 2004).

Prevention of vocal fatigue

The prevention of vocal fatigue is important in high-risk groups mentioned earlier. Both vocal rest and hydration have shown to improve negative vocal changes associated with induced vocal fatigue. Verdolini, Sandage and Titze (1994) advocated the use of consecutive hydration treatment programme to improve voice and vocal fold condition in patients with nodules and polyps and hydration was found to reduce the phonatory effort. Yiu and Chan (2003) also found that hydration and vocal rest during karaoke-singing prolonged the singing time before vocal fatigue was reported.

Use of herbal medicine in managing dysphonia

In China, the use of over-the-counter Chinese herbal medicine and tonics are commonly employed by individuals with voice disorder. One of the medications frequently documented is ‘watermelon frost’. It is manufactured in different forms including insufflations, tablets and lozenges. The main ingredient in these manufactured products is watermelon frost, with other herbal ingredients including bulbus fritillariae cirrhosae, fructus momordicae, radix sophorae toninensis, rhizome belamcandae, radix scutellariae, herba menthae, indigo naturalis and borneolum syntheticum (see Appendix I for ingredient list).

Different forms of watermelon frost have been prescribed to treat the problems related to the throat. The therapeutic use of watermelon frost was first documented in the ancient medical literature, *Yang Yi Da Quan* (<<秠雍嶲斟>>), in Qing Dynasty (1644 A.D.). The effect of watermelon frost is believed to remove '*heat*', a kind of destructive energy disturbing the normal functioning of the five '*zangs*' (organs), the lung, spleen, kidney, liver and the heart. The five '*zangs*' are responsible for generating adequate '*qi*' which is essential for voice production. Watermelon frost is documented to help regain this '*qi*' after the normal functioning of the '*zangs*' are disturbed by '*heat*' (盲, 1998). It is wide-held belief that watermelon frost is good for the voice. Indeed, a number of reports have described the successful use of watermelon frost in treating throat problems including acute and chronic pharyngitis and laryngitis (咏, 2002; 噦, 徨 &縋, 1994; 咏, 櫨&咏, 1995). However, there is no study that investigated specifically the effectiveness of watermelon frost in preventing and treating voice disorders. The aim of this study was to investigate the prophylactic potential of watermelon frost for vocal fatigue. Watermelon frost was taken by participants prior to vocal fatiguing task - karaoke-singing. It was hypothesized that the use of watermelon frost helps to maintain or promote vocal functions. Therefore, the participants would be able to sing continuously without deterioration of vocal qualities and functioning, in terms of phonetogram, acoustic, perceptual and subjective self-rating measures.

METHOD

A treatment-placebo, double-blinded design was employed in the present study. One group received a seven-day-prophylactic treatment for vocal fatigue using watermelon frost.

The other group took a placebo-spray for the same period of time. The allocation of group was double-blind to the investigator and the participants in order to control the demand characteristics and experimenter effects. The placebo-spray was used to control the demand characteristics of the placebo group (Shaugnessy, Zechmeister & Zechmeister, 2003).

Materials

The watermelon frost employed in the present study was *Sanjin Watermelon Frost Insufflations* (倇霆躃磚鯧壼呫, see Appendix II). Wheat germ E powder (稟g 菰蛸 E 肧, see Appendix II) was used as placebo with the ingredients not expected to cause changes in vocal qualities. Both types of sprays were stored in original containers of watermelon frost to control for the appearance of the container and the amount of spray-intake between two groups.

Participants

A total of 36 participants, including 18 males and 18 females (mean = 21.4 years, standard deviation = 1.63 years, range =17-24 years) participated in the study. All participants were recruited from the investigator's social circle or from the Division of Speech and Hearing Sciences of the University of Hong Kong. Informed consents were collected from all participants (Appendix III). The following selection criteria were used:

1. Participants self-reported to have the experience of vocal fatigue after prolonged singing.
2. Participants had the experience of karaoke-singing, on average 1 to 2 times a month.
3. Participants were in healthy state and they did not have respiratory tract infection at the time of study. They were also not taking any medication at the time of study.
4. Participants self-reported and were perceptually judged to have normal voice quality by the investigator.

5. Participants had no history of using watermelon frost. Since the distinctive odor and taste of watermelon frost would not be demonstrated by the placebo used in this study, participants should not have used watermelon frost in the past to reduce their demand characteristics.
6. Participants were not professional voice users such as teachers, sales or singers.
7. Participants had not received formal training in singing and voice-use.
8. Participants were non-smokers and non-alcoholic drinkers.

Procedures

Participants were allocated to the experimental group or the placebo group. All of the participants completed three identical sets of voice assessments on two separate days scheduled one week apart. A singing task was carried out on the second assessment day. To control for the possible differences of voice quality within the day and the week, all participants were evaluated at the same time and same day of the week during the two visits. Participants of the experimental group received a tube of watermelon frost insufflations. They were instructed to apply it inside the oral cavity, as near to the throat (supra-laryngeal area) as possible, for three times a day and three sprayings each time. The dosage of intake was set according to the recommendation suggested by the pharmaceutical company and the reports of 咏(2002). The dosage was considered to be safe because no side-effect was cautioned by the manufacturer and reports. A recording sheet to record the total use of spray was given to each participant and they were required to complete the form before the second day of evaluation. Participants of the placebo group were given a tube of placebo-spray and were given the same instruction of use and recording from as those in the treatment group. All participants were cautioned of activities that might lead to vocal fatigue and changes in voice qualities (such as karaoke-singing, barbecuing, drinking of alcohol and intake of other medications) to minimize the effect of confounding factors. All participants reported to have completed the full use of spray during the week of spray-intake.

Measurement-points: Three measurement-points were collected. Baseline data (pre-application) was collected on the first day of assessment before the use of spray. Measurement 2 (pre-fatigue) was carried out a week after the use of the spray and before the singing task was carried out. Measurement 3 (post-fatigue) was carried out immediately after a continuous singing task lasting for 120 minutes. Each measurement consisted of data obtained form the followings:

Self-perceived voice subjective rating questionnaire: Six questions concerning the self-control of breathing, pitch, loudness and phonation were included (see Appendix VI). Participants were required to rate their voice qualities in these aspects using a five-point-rating scale (1 to 5) to collect subjective measurements of voice quality.

Voice recordings for acoustic analysis: Each participant was required to produce a prolongation of a vowel /a/ at comfortable pitch and loudness levels for five seconds in a soundproof room. The recordings were captured by Kay Elemetrics' Computerized Speech Lab (CSL, Kay Elemetrics, Lincoln Park, NJ) with a Shure SM 48 microphone. A mouth-to-microphone distance of 10 cm was kept among all participants. Three trials at each data-measurement point were recorded and then an averaged value was obtained.

*Voice recordings for perceptual analysis:*Each participant was required to read aloud a

Chinese sentence /pa pa ta k□ k□/ (father hits brother) at comfortable pitch and loudness levels for three times in a soundproof room. The sentence was chosen because all the words are made up of simple vowels and unaspirated vowels. Each recording was made three times. All the samples were analyzed perceptually in the way to be described below.

Voice recordings for phonetogram analysis: The voice range profiles (phonetograms) were recorded in a soundproof room. The Swell's real-time computerized phonetogram Phog 1.0 system from AB Nyvalla DSP and a microphone (AKG acoustics, C420) with a mouth-to-microphone distance of 5 cm were used to record the phonetograms. Each participant was required to sustain /a/ at musical tone C4 and produced the initial pitch at a comfortable loudness level, followed by reducing the loudness to the softest level. The pitch proceeded down the musical scale until the pitch at which the participant was unable to sustain /a/. Then the pitch level return to C4 and proceeded up the musical scale until the pitch at which the participant was unable to sustain /a/. The procedure was then repeated with /a/ sustained from comfortable loudness levels to the maximum loudness levels for each pitch level.

Vocal loading task - prolonged karaoke-singing: A karaoke singing task was administered to induce vocal fatigue in the participants. Each participant was informed of the list of songs in the singing task. The participants only sang songs which were appropriate for their gender and participants of the same gender sang the same list of songs.

Each participant was sitting in a quiet room with karaoke facility (Panasonic SV-VP35). Music videos with echo effects were played on a television and the participant was required to sing for 120 minutes continuously. Participants of each gender sang the same number of songs (40 songs) in the same predetermined sequence of songs. No drink or food was allowed until all measurements were completed.

Data analysis

Acoustic analysis

The analysis of vowel /a/ samples was performed by extracting the middle three-second portion. Jitter percentage, the shimmer percentage and noise-to-harmonic ratio were obtained using Kay Electmetrics' Computerized Speech Lab (CSL 4300B) and the Multidimensional Voice Program (MDVP 4305).

Phonetogram analysis

Seven parameters obtained for the voice range profiles were analyzed, mean fundamental range (Hz), maximum fundamental frequency (Hz), minimum fundamental frequency (Hz), maximum intensity (dB), minimum intensity (dB), mean loudness range(dB) and total area of voice range profile (dB semitone).

Perceptual analysis

Mean roughness and breathiness of each sample of the sentence /pa pa ta k□ k□/ were rated by an assistant professor and two postgraduate speech and hearing sciences students with at least a year of experience in conducting voice assessment. These two vocal qualities were analyzed because they are more reliable than other (Dejonckere, Obbens, deMoor & Wieneki, 1993) in the evaluation of dysphonia. They are also more reliable signs of vocal fatigue (Colton & Casper, 1996). The definitions of roughness and breathiness were explained to the judges before they rated the samples. In this study, roughness was defined as "irregular vocal quality, random fluctuations of glottal pulse and lack of clarity" while breathiness was defined as "audible sound of expiration, air escape and friction noise" (Chan & Yiu, 2002). Judges listened to the stimuli presented through a computerized program in a sound booth. A Creative Sound Blaster Extigy Signal Processing unit and a pair of headphones (Sennheiser, HD 25) were used to present the stimuli at a comfortable intensity level.

A paired comparison matching paradigm using synthesized Cantonese voice stimuli was administered to improve the reliability of perceptual voice rating in this study. This rating method was adopted because it was reported to increase the mean percentage of intra-rater agreement in perceptual voice rating (up to 0.94) (Yiu, Chan & Mok, submitted). An eight-point equal-appearing-interval (EAI) scale was used (see Appendix VI). The eight scale points (0-7) were used to represent normal to more dysphonic voice qualities of the voice anchors. The whole synthesized continua of roughness and breathiness were played twice before the rating task to familiarize the judges with the synthesized signals. Three practice trials of perceptual evaluation were given for the listener to familiarize with the procedures. The judges then select the synthesized anchor that best represented the level of breathiness and roughness of each of the samples. The judges were allowed to listen to the samples and anchors repetitively until they could give a rating. Fifteen percent of the stimuli were repeated once to determine the intra-rater reliability. The entire session lasted for one hour and 45 minutes and the ratings given by the judges were averaged to give a final rating for each sample.

RESULTS

Phonetogram analysis

Table 1 lists the mean results of the seven phonetogram measures for the experimental and placebo groups. Table 2 shows the results of factorial ANOVA of repeated measure covariates with the variable ‘sex’ was performed.

The mean maximum loudness of the experimental group was found to be significantly higher than the placebo group ($p<0.05$). The mean area of voice range profile was found to be significantly lowered after the singing task ($p<0.05$), a further F-test to obtain within-subjects contrasts revealed significantly reduced voice range profile areas between pre-fatigue and post-fatigue for both groups ($F=6.329$; $p<0.05$, see Figure 1 in Appendix VI for graphical presentation). In addition, interaction effect between time and gender was found ($p<0.05$), the reduction of area of voice range profile from pre-fatigue to post-fatigue period was significantly greater in male participants than female participants ($F=6.489$; $p<0.05$, see Figure 2).

Significant gender differences between male and female participants were found in maximum fundamental frequency, fundamental frequency range and maximum loudness ($p<0.05$). However, no significant difference and no interaction effect between time and groups were found between experimental and placebo groups ($p>0.05$; see Table 2).

Table 1. *Mean results of phonetogram measures*

	Experimental group			Placebo group		
	Mean			Mean		
	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)
Area of voice range profile (dB semitone)	1253.17 (178.52)	1298.61 (189.69)	1194.44 (202.80)	1162.89 (218.24)	1165.06 (228.89)	1078.67 (233.33)
Male	1313.00 (215.96)	1375.67 (190.66)	1224.67 (209.88)	1250.44 (192.66)	1215.22 (149.63)	1064.89 (220.41)
Female	1193.33 (114.12)	1221.56 (163.57)	1164.22 (203.21)	1075.37 (216.43)	1114.89 (288.58)	1092.44 (258.23)
Highest fundamental frequency (Hz)	972.62 (239.71)	998.87 (231.02)	990.34 (249.95)	953.31 (249.76)	905.95 (246.80)	946.54 (269.28)
Male	972.62 (239.71)	998.87 (231.02)	990.34 (249.95)	953.31 (249.76)	905.95 (246.80)	946.54 (269.28)
Female	1120.37 (252.65)	1167.92 (200.74)	1159.83 (238.28)	1136.16 (176.71)	1066.71 (219.69)	1159.83 (238.28)
Lowest fundamental frequency (Hz)	109.85 (29.63)	109.11 (30.68)	116.52 (33.35)	112.36 (36.93)	114.47 (36.73)	205.46 (361.80)
Male	82.64 (11.27)	80.40 (10.33)	85.88 (12.17)	78.94 (11.02)	81.93 (10.77)	91.72 (11.65)
Female	137.06 (8.59)	137.82 (6.24)	147.16 (10.13)	145.77 (16.27)	147.02 (19.20)	319.19 (498.92)
Fundamental frequency range (Hz)	862.77 (223.27)	889.761 (208.04)	873.83 (228.84)	840.96 (226.89)	791.47 (230.41)	741.09 (369.69)
Male	742.22 (100.66)	749.42 (93.92)	734.98 (105.95)	691.52 (158.81)	663.26 (146.86)	638.46 (141.20)
Female	983.31 (251.20)	1030.10 (197.07)	1012.68 (238.08)	990.39 (184.20)	919.67 (232.94)	843.72 (496.78)
Maximum loudness (dB)	110.44 (4.29)	112.22 (5.40)	112.61 (5.66)	108.28 (5.74)	109.11 (5.43)	109.50 (5.20)
Male	111.78 (3.96)	113.00 (4.61)	114.22 (4.29)	110.78 (5.47)	111.78 (4.02)	114.22 (4.29)
Female	109.11 (4.40)	111.44 (6.27)	111.00 (6.61)	105.78 (5.12)	106.44 (5.53)	111.00 (6.61)
Minimum loudness (dB)	59.67 (5.72)	59.83 (5.73)	61.22 (7.32)	60.00 (4.70)	62.13 (7.35)	61.56 (8.71)
Male	61.44 (7.28)	59.37 (7.00)	61.33 (9.26)	61.44 (5.46)	64.71 (9.43)	62.67 (12.28)
Female	57.89 (3.06)	60.33 (4.50)	61.11 (5.30)	58.56 (3.54)	59.56 (3.28)	60.44 (2.79)
Loudness range (dB)	50.78 (6.12)	52.39 (6.49)	51.39 (8.14)	48.28 (7.61)	46.98 (9.42)	47.94 (8.78)
Male	50.33 (7.83)	53.67 (8.08)	52.89 (10.80)	49.33 (9.68)	46.89 (7.24)	49.11 (11.69)
Female	51.22 (4.24)	51.11 (4.34)	49.89 (4.37)	47.22 (5.17)	7.07 (11.66)	46.78 (4.92)

Table 2. *Factorial ANOVA on phonetogram measures.*

	Main effect / interaction effects	F	df	p
Area of voice range profile (dB semitone)	Group	3.239	1	0.081
	Time	7.637	2	* 0.002
	Sex	2.379	1	0.132
	Time x group	0.543	2	0.586
	Time x sex	3.895	2	* 0.031
Highest fundamental frequency (Hz)	Group	0.900	1	0.350
	Time	1.510	2	0.236
	Sex	40.460	1	* 0.000
	Time x group	2.281	2	0.073
	Time x sex	2.851	2	0.119
Lowest fundamental frequency (Hz)	Group	1.338	1	0.256
	Time	0.160	2	0.853
	Sex	10.121	1	* 0.003
	Time x group	0.941	2	0.401
	Time x sex	0.493	2	0.615
Fundamental frequency range (Hz)	Group	2.070	1	0.160
	Time	0.015	2	0.985
	Sex	19.701	1	* 0.000
	Time x group	2.648	2	0.086
	Time x sex	0.049	2	0.952
Maximum loudness (dB)	Group	3.292	1	* 0.021
	Time	0.556	2	0.579
	Sex	5.833	1	* 0.021
	Time x group	0.095	2	0.909
	Time x sex	0.384	2	0.684
Minimum loudness (dB)	Group	0.282	1	0.599
	Time	0.100	2	0.905
	Sex	1.365	1	0.251
	Time x group	0.611	2	0.549
	Time x sex	0.555	2	0.579
Loudness range (dB)	Group	2.560	1	0.119
	Time	0.456	2	0.638
	Sex	0.428	1	0.517
	Time x group	1.231	2	0.305
	Time x sex	0.465	2	0.632

* significant at 0.05 level

Acoustic analysis

Table 3 lists the mean results of the four acoustic measures, fundamental frequency, jitter percentage, shimmer percentage and noise-to-harmonic ration for the experimental and placebo groups. Table 4 shows the results of factorial ANOVA of repeated measure covariates with the variable ‘sex’ was performed.

Significant gender difference was found between male and female participants ($p< 0.05$) and a significant interaction effect between time and gender was found for the jitter percentage ($p<0.05$). A further F-test performed identified a significantly higher jitter percentage for female participants at pre-application ($F=6.888$; $p<0.05$, see Figure 3 in Appendix VI for graphical presentation).

Table 3. *Mean results of acoustic measures*

	Experimental group			Placebo group		
	Mean			Mean		
	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)
Fundamental frequency (Hz)	200.03 (62.57)	211.57 (63.42)	218.55 (61.08)	209.52 (73.97)	214.40 (71.48)	218.00 (69.31)
Male	145.16 (27.50)	153.14 (24.06)	161.81 (22.60)	142.72 (34.95)	151.20 (39.82)	159.12 (40.30)
Female	254.91 (28.03)	269.99 (16.94)	275.29 (12.67)	276.32 (17.14)	277.60 (21.82)	276.87 (28.05)
Jitter percentage (%)	0.95 (0.46)	0.98 (0.48)	0.84 (0.50)	0.77 (0.54)	0.72 (0.45)	0.88 (0.80)
Male	0.84 (0.44)	1.16 (0.58)	0.87 (0.58)	0.42 (0.16)	0.46 (0.22)	0.85 (1.00)
Female	1.07 (0.47)	0.81 (0.28)	0.80 (0.44)	1.11 (0.57)	0.98 (0.48)	0.90 (0.63)
Shimmer percentage (%)	3.87 (1.64)	3.07 (0.85)	3.07 (0.94)	2.74 (1.08)	2.98 (1.04)	2.83 (1.15)
Male	3.40 (1.01)	3.32 (0.87)	2.87 (0.90)	2.36 (1.05)	2.68 (1.00)	2.67 (0.90)
Female	4.33 (2.05)	2.83 (0.81)	3.28 (1.00)	3.11 (1.03)	3.28 (1.04)	2.99 (1.34)
Noise-to-harmonic ratio	0.13 (0.02)	0.14 (0.05)	0.12 (0.01)	0.15 (0.11)	0.12 (0.02)	0.12 (0.01)
Male	0.14 (0.01)	0.13 (0.01)	0.12 (0.01)	0.17 (0.16)	0.13 (0.02)	0.13 (0.01)
Female	0.17 (0.16)	0.14 (0.07)	0.11 (0.01)	0.13 (0.01)	0.11 (0.02)	0.11 (0.01)

Table 4. *Factorial ANOVA on acoustic measures.*

	Main effect / interaction effects	F	df	p
Fundamental frequency (Hz)	Group	0.291	1	0.593
	Time	2.543	2	0.094
	Sex	180.762	1	* 0.000
	Time x group	1.613	2	0.215
	Time x sex	0.918	2	0.410
Jitter percentage (%)	Group	1.139	1	0.294
	Time	3.079	2	0.060
	Sex	0.272	1	0.177
	Time x group	0.846	2	0.438
	Time x sex	3.486	2	* 0.043
Shimmer percentage (%)	Group	2.927	1	0.096
	Time	0.947	2	0.399
	Sex	1.166	1	0.288
	Time x group	2.945	2	0.067
	Time x sex	1.924	2	0.162
Noise-to-harmonic ratio	Group	0.099	1	0.755
	Time	0.149	2	0.862
	Sex	0.302	1	0.587
	Time x group	0.615	2	0.547
	Time x sex	0.784	2	0.465

* significant at 0.05 level

Self-perceived voice subjective rating

Table 5 lists the mean results of the six measures of self-voice ratings, general voice use, breathing control, pitch control, ease of initiation of phonation and tiredness in the throat. Table 6 shows the results of factorial ANOVA of repeated measure covariates with the variable ‘sex’ was performed.

The experimental group was found to rate the general voice use significantly higher than the placebo group ($p<0.05$). The rating of general voice use of the two groups also changed significantly across time ($p<0.05$). An F-test to determine inter-subjects contrasts indicated significantly higher ratings of general voice use at post-fatigue period in the experimental group ($F=8.790$; $p< 0.05$; see Figure 4 in Appendix VI for graphical presentation). Moreover, females participants were found to give higher ratings for general voice use then male participants significantly at all time ($p<0.05$)

Table 5. *Mean results of self-perceived voice subjective rating*

	Experimental group			Placebo group		
	Mean			Mean		
	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)
General voice use	3.39 (0.70)	3.56 (0.71)	3.33 (0.77)	2.94 (0.73)	3.17 (0.62)	2.56 (0.78)
Male	3.22 (0.67)	3.67 (0.73)	2.89 (0.78)	2.67 (0.71)	2.89 (0.60)	2.22 (0.67)
Female	3.56 (0.73)	3.44 (0.73)	3.78 (0.44)	3.22 (0.67)	3.44 (0.53)	2.89 (0.78)
Breathing control	3.28 (0.90)	3.22 (0.94)	3.17 (0.71)	3.11 (0.76)	3.06 (0.73)	2.94 (0.73)
Male	3.11 (0.78)	3.56 (0.73)	3.00 (0.71)	3.22 (0.83)	3.22 (0.83)	3.00 (0.69)
Female	3.44 (1.01)	2.89 (1.05)	3.33 (0.71)	3.00 (0.71)	2.89 (0.60)	2.89 (0.73)
Loudness control	3.39 (1.04)	3.44 (0.88)	2.94 (0.73)	3.22 (0.81)	3.33 (0.67)	3.00 (0.69)
Male	3.17 (0.87)	3.22 (0.67)	2.89 (0.78)	3.22 (1.09)	3.44 (0.88)	3.11 (0.78)
Female	3.11 (1.17)	3.33 (0.77)	3.00 (0.71)	3.22 (0.44)	3.22 (0.44)	2.89 (0.60)
Pitch control	3.22 (0.81)	3.39 (0.85)	3.00 (0.84)	2.72 (1.12)	3.06 (0.94)	2.94 (0.64)
Male	3.00 (0.87)	3.44 (0.88)	2.56 (0.73)	2.56 (1.33)	3.00 (1.12)	2.78 (0.44)
Female	3.44 (0.73)	3.33 (0.87)	3.44 (0.73)	2.89 (0.93)	3.11 (0.78)	3.11 (0.78)
Ease of initiation of phonation	4.00 (0.84)	3.83 (0.86)	3.11 (1.08)	3.78 (1.01)	3.83 (0.86)	3.28 (1.18)
Male	3.67 (0.87)	3.44 (0.73)	2.67 (0.71)	3.56 (1.01)	3.78 (0.83)	2.78 (0.97)
Female	4.33 (0.71)	4.22 (0.83)	3.56 (1.23)	4.00 (1.00)	3.89 (0.93)	3.78 (1.20)
Tiredness in throat	3.61 (0.78)	3.50 (0.71)	3.11 (0.90)	3.67 (1.03)	3.61 (0.85)	2.78 (1.17)
Male	3.44 (0.88)	3.44 (0.73)	2.67 (0.87)	4.00 (0.87)	3.67 (1.00)	2.67 (1.12)
Female	3.78 (0.67)	3.56 (0.73)	3.56 (0.73)	3.33 (1.12)	3.56 (0.73)	2.89 (1.27)

Table 6. *Factorial ANOVA on self-perceived voice subjective rating*

	Main effect / interaction effects	F	df	p
General voice use	Group	9.852	1	* 0.004
	Time	4.292	2	* 0.022
	Sex	7.322	1	* 0.011
	Time x group	2.256	2	0.121
	Time x sex	1.191	2	0.317
Breathing control	Group	0.875	1	0.356
	Time	1.974	2	0.155
	Sex	0.315	1	0.578
	Time x group	0.020	2	0.980
	Time x sex	2.129	2	0.136
Loudness control	Group	0.120	1	0.310
	Time	1.117	2	0.340
	Sex	0.766	1	0.388
	Time x group	0.279	2	0.758
	Time x sex	0.243	2	0.786
Pitch control	Group	1.577	1	0.218
	Time	3.180	2	0.550
	Sex	2.465	1	0.126
	Time x group	1.029	2	0.369
	Time x sex	1.889	2	0.168
Ease of initiation of phonation	Group	0.006	1	0.937
	Time	2.634	2	0.087
	Sex	7.761	1	0.090
	Time x group	0.651	2	0.529
	Time x sex	0.789	2	0.463
Tiredness in throat	Group	0.064	1	0.802
	Time	3.599	2	0.039
	Sex	0.349	1	0.559
	Time x group	0.623	2	0.543
	Time x sex	1.463	2	0.247

* significant at 0.05 level

<u>Perceptual voice evaluation</u>

Reliability measures

Intraclass correlation coefficients for inter-rater reliability of breathiness were 0.87, 0.85 and 0.90 for pre-application, pre-fatigue and post-fatigue respectively ($p<0.05$). Intraclass correlation coefficients for inter-rater reliability of roughness were 0.66, 0.63 and 0.71 for pre-application, pre-fatigue and post-fatigue respectively ($p<0.05$). Mean intraclass correlation coefficients for intra-rater reliability among the three judges were 0.81 and 0.78 for breathiness and roughness respectively (p<0.05).

Mean breathiness and roughness ratings

Table 7 shows the mean results of perceptual analysis for the two groups, male and female participants respectively. Significant gender differences between male and female participants were identified by a Factorial ANOVA test of repeated measure covariates with the variable 'sex' (p<0.05; see Table 8).

Table 7. *Mean perceptual ratings of /pa pa ta k□ k□/*

	Experimental group			Placebo group		
	Mean			Mean		
	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)	Pre-application (Standard deviation)	Pre-fatigue (Standard deviation)	Post-fatigue (Standard deviation)
Breathiness (0-7 points)	0.51 (0.48)	0.61 (0.49)	0.65 (0.60)	0.50 (0.59)	0.42 (0.50)	0.34 (0.44)
Male	0.52 (0.53)	0.57 (0.59)	0.63 (0.66)	0.32 (0.52)	0.28 (0.45)	0.32 (0.48)
Female	0.51 (0.47)	0.65 (0.39)	0.68 (0.57)	0.68 (0.62)	0.56 (0.54)	0.50 (0.54)
Roughness (0-7 points)	0.74 (0.44)	0.60 (0.43)	0.68 (0.42)	0.96 (0.91)	0.67 (0.36)	0.65 (0.34)
Male	0.46 (0.23)	0.32 (0.29)	0.60 (0.36)	0.69 (0.39)	0.69 (0.35)	0.68 (0.38)
Female	1.22 (1.21)	0.89 (0.36)	0.75 (0.48)	1.22 (1.21)	0.65 (0.38)	0.63 (0.33)

Table 8. *Factorial ANOVA on the perceptual analysis of /pa pa ta k□ k□/*

	Main effect / interaction effects	F	df	p
Breathiness (0-7 points)	Group	1.280	2	0.266
	Time	0.381	1	0.686
	Sex	0.743	2	0.395
	Time x group	1.721	1	0.195
	Time x sex	0.502	2	0.610
Roughness (0-7 points)	Group	0.661	2	0.422
	Time	1.261	1	0.297
	Sex	7.342	2	* 0.011
	Time x group	0.448	1	0.643
	Time x sex	1.988	2	0.154

* significant at 0.05 level

DISCUSSION

The present study aimed to investigate the possible effects of watermelon frost on the vocal qualities and functioning in a group of male and female participants. The objective of the study was based on the traditional Chinese medicine hypothesis that watermelon frost would assist the generation of '*qi*' necessary for voice production in the five '*zangs*' (王, 1998). Vocal fatigue was therefore hypothesized to be reduced by the use of watermelon frost. Karaoke singing task was used to investigate whether vocal qualities and functioning were maintained or improved after using watermelon frost consecutively for a week. The results obtained from the data on acoustic measures, perceptual measures and subjective self-rating of vocal functions indicated no significance between experimental and placebo groups.

<u>Voice range profile</u>

There were also no significant interaction effects found between time and groups in all the seven phonetogram measures, therefore, no changes in the vocal qualities and functioning as reflected by both frequency and intensity measures were found between the experimental and placebo groups. Watermelon frost demonstrated no effect of maintaining vocal qualities as reflected in the voice range profiles.

The interaction effect between time and gender may only indicate that different levels of vocal fatigue were obtained from the two genders (see Figure 2 in Appendix VI), male participants were found to have greater reduction of area of voice range profile after singing for 120 minutes. It appeared that male participants were more vulnerable to vocal fatigue as revealed by this phonetogram measure.

The area voice range profile was significantly reduced for both groups after the singing task was conducted. Therefore, both groups exhibited vocal fatigue after the 120 minutes of singing. This finding is not consistent with the study by Yiu and Chan (2003) which also adopted a singing task to induce vocal fatigue. The control group in Yiu and Chan's study reported vocal fatigue was perceived after an average of 86 minutes of singing. As self-report measure was used to determine the time to fatigue the voice in this study, there was the risk of reporting vocal fatigue due to boredom in the task. A fixed vocal loading time is still preferred, because it is suggested that the risk of false-positive results can be avoided. However, as both groups in the present study exhibited vocal fatigue in 120 minutes and vocal fatigue can actually occur before the 120th minute. It is therefore suspected that the time for inducing vocal fatigue may be between 86 to 120 minutes. Further studies which aim at inducing vocal fatigue using singing tasks may need to consider the above suggested range of time for vocal fatigue to occur.

Although significant difference of change in the area of voice range profiles between groups was not obtained as no interaction effect between groups and time was obtained, there was an increase of 45 dB semitone (from 1253.17 to 1298.61 db semitone) in the area in the experimental group between pre-application and pre-fatigue data-points, when the placebo group did not change at all (from 1162.89 to 1165.06 dB semitone) during these period. It is probably that this phonetogram measure is more sensitive to reflect the effectiveness of watermelon frost on voice range profiles. The second possibility is that watermelon frost had improved the voice range profiles during the seven days of application before singing task was conducted, but only because the vocal loading time was set too long (120 minutes) as compared with other similar studies, for example, 86 minutes of singing in the study by Yiu & Chan, (2003), 1 hour of reading in study by Gelfer et. al.(1991), and 2 hours of reading in the study by Stemple at. al (1995). It may be possible that the effectiveness of watermelon frost was not able to extend in the long period of vocal loading and could not maintain good vocal functioning to prevent vocal fatigue, but it helped with improving vocal functioning at least after application. However, practice effect of using phonetogram repetitively would explain this possible increase again and therefore the interpretation of this increase in voice range profile areas should be careful.

A number of phonetogram measures revealed significant gender effects. However, these measures were gender-sensitive in their nature, for example, the highest and lowest fundamental frequencies and the frequency range per se. Therefore, the significant changes can be explained by the different gender nature.

<u>Acoustic measures:</u>

There was no significant changes in the four acoustic measures between experimental and placebo groups, probably indicating that the selected acoustic measures were not sensitive to measuring vocal fatigue or no significant vocal changes were obtained from the use of watermelon frost. The lack of significant differences may be attributed to individual differences in producing the vowel /a/ for acoustic analysis to be performed on. Although participants were told to phonate at their comfortable levels of pitch and loudness consistently for three trials at each measurement point, the three values obtained were not consistent with each other. It is therefore suggested that the reliability of acoustic measures was not high enough which caused the inconsistent values obtained. It is also doubted if the data obtained from acoustic measures reflected actual differences of vocal qualities in the two groups. As a result, data from acoustic measures should be interpreted with caution.

Subject self-rating of vocal qualities and functioning

There was no significant difference between the two groups in all the six parameters measuring the participant's subjecting ratings of their vocal qualities and functioning (see Table 6) and therefore, the subjective rating of voice in the two both did not differ. Watermelon frost was not thought to be effective in reducing vocal fatigue.

Perceptual analysis

Contrary to the hypothesis that changes in vocal qualities, breathiness and roughness, would be observed in the perceptual analysis of sentences produced at the three data-points, no significant difference was obtained between the two groups at all time (see Table 7). Although an 8 point-scale was adopted, all samples were rated under point 2 (see Table 6). It may be because the changes in roughness and breathiness were too small for the judges to detect and rate. The problem of undetectable tiny changes in roughness and breathiness may result in the lack of significant difference between groups. This problem is expected because normal participants without voice disorders were recruited in this study and thus undetectable changes in perceptual analysis were reasonable. Further studies investigating vocal fatigue may need to consider recruiting patients with voice disorders so that greater changes in voice qualities may be detected perceptually more easily. Moreover, the inter-rater reliabilities for rating roughness were rather low (0.66, 0.63 and 0.71 for the three data-points), therefore the averaged ratings for roughness among the three judges must be interpreted with caution, and it may only be fairly representative to the actual performance of the participants.

In summary, the present study failed to show that the herbal tonic ‘watermelon frost’ was able to maintain good vocal qualities and functioning during 120 minutes of karaoke-singing. Although it was hypothesized that watermelon frost was able to help generate the ‘qi’ which in turn assist voice production, result did not demonstrate that watermelon frost was useful to preserve nor improve vocal functioning in this study. The participants in the experimental group did not show to have less vocal fatigue than those in the placebo group. The vocal qualities and functions of the two groups as measured by both acoustic, perceptual measures and subjective self-ratings were similar, although a statistically insignificant increase in the area of voice range profile obtained after applying watermelon frost revealed its potential effectiveness in improving vocal functioning.

Limitation of the present study

Several methodological shortcomings in this study may affect the results obtained. The first limitation was the application method of watermelon frost. The participants were instructed to use the spray in the oro-pharyngeal area. It was possible that watermelon frost in the spray may not reach the larynx and the vocal folds for reaction to take place, and thus the effectiveness of watermelon frost was reduced and not reflected in this study.

The second limitation was the amount of application. In this study, every participant was given a tube of watermelon frost or placebo spray (3 grams). Although they were instructed to ensure enough amount of watermelon frost was pressed out from the tueb every time, it was difficult to measure the amount of watermelon frost applied. As a result, the varied amount of intake of watermelon frost may affect its effectiveness in different individuals.

The third limitation was the stimuli used in the singing task. Since both male and female participants were involved, songs sung by the two genders were selected as stimuli. However, it was difficult to ensure that songs for both gender required the same level of vocal performance, and therefore gender differences may not be well interpreted from the results of this study.

Recommendations for further researches

The use of a fixed amount of vocal loading time in order to induce vocal fatigue is still recommended as it is possible to prevent the problem of false-negative results raised by self-reporting measures adopted in previous studies. In addition, further studies might standardize the amount of watermelon frost used in each application. In this way, participants would be receiving the same amount of tonic before vocal loading task is conducted and its effectiveness can be ensured when standardized dosage is prescribed. Also, participants may be instructed to inhale watermelon frost from the oral cavity to the laryngeal area so as to allow watermelon frost to react on the laryngeal area more directly and easily. However, this should be dealt with care as the action of inhalation may cause choking and is therefore potentially dangerous for participants. Finally, further studies may also focus on the effectiveness of curative treatment by applying watermelon frost on dysphonic patients.

Conclusion

The present study attempted to investigate the effectiveness of 'watermelon frost' in the prevention of vocal fatigue. Results of this study showed no significant changes between the experimental group receiving 'watermelon frost' and the placebo group receiving 'wheat germ E powder'. 'Watermelon frost' was not demonstrated to have effectiveness in preventing vocal fatigue. Further researches recruiting dysphonic patients to study the curative effects of watermelon frost can be conducted.

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Appendix I: leaflet showing ingredients of *Sanjin Watermelon Frost Insufflation*

中國中藥保護品種　中國中藥名牌產品

三金西瓜霜 新一代 噴劑

（原名：復方西瓜霜）

【成　　份】西瓜霜、硼砂(煅)、黄芩、山豆根、冰片、青黛、甘草、黄 柏、黄連、浙貝母、射干、無患子果(炭)、薄荷腦、大黄。

【藥理作用】抗菌，抗病毒，消炎，解熱，消腫，止痛，止咳，祛痰，促進潰瘍愈合等。

【功　　能】清熱解毒，降火，清咽利喉，消腫止痛，止血。

【主　　治】咽喉、嗓音、口腔、上呼吸道等炎症疾患：咽喉腫痛，嗓音嘶啞，乳蛾喉痹，口舌生瘡或潰爛，牙齦腫痛，小兒鵝口瘡，口臭，急性咽喉炎，扁桃體炎，會咽炎，腮腺炎；口腔炎，口腔潰瘍，舌體炎，牙齦炎，牙周炎；鼻炎，支氣管炎等。兼治暗瘡、中耳炎、燙傷、燒傷、創傷或牙齦出血等。

【性　　狀】本品爲灰黄緑色粉末；氣香，味咸、甜、微苦而辛涼。

【用法用量】

1. 口腔、咽喉、鼻咽諸症：用本品直接噴于患處，一日 3 — 6 次，每次 2 — 4 噴。重症可兼服，每次 0.3 — 0.5 克，每日 1 — 2 次。
2. 牙痛：用本品噴（或用藥棉沾本品藥粉填塞）于齲齒孔中以及牙肉腫痛處，一日 3 — 6 次，每次 2 — 4 噴。重症可兼服，一次 0.3 — 0.5 克，每日 1 — 2 次。
3. 止血：以本品噴敷或將本品藥粉撒布于患處，用藥棉壓迫數分鐘或用紗布包扎，即可止血。
4. 燒傷燙傷：用適量食油調和本品藥粉，塗于患處，連用數次至痊愈爲止。
5. 暗瘡、蚊蟲叮咬、痱子、香港脚、中耳炎等：用本品藥粉塗擦或直接噴于患處，連用數次至痊愈爲止。

【規　　格】每瓶裝 3 克

【儲　　藏】密閉，防潮　　【有效期】三年

【地　　址】中國桂林市金星路 1 號　　【電　　話】86-773-5843251

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咽喉、嗓音、口腔疾患爲常見、多發性疾病，中醫學素有"咽喉口腔諸病皆源于火"之說。咽喉、嗓子、口腔、牙齦等主要病理變化——紅腫、疼痛、化膿等都歸爲火的表現。

三金西瓜霜噴劑，系中國有突出貢獻的中醫藥專家，中國藥典委員會委員鄒節明先生，集中國傳統古方、秘方、驗方之大成，經三十余年臨床用藥經驗，在本廠（原桂林中藥制藥廠）獨家生産的"桂峰"牌與"象鼻山"牌"復方西瓜霜"（海外銷售三十余年）的基礎上改進與創新的新一代産品，其功效更强，療效更顯著，服用更安全，經中國藥品行政管理部門批准，由高新技術企業——桂林三金藥業股份有限公司獨家生産。

本品以三金®西瓜霜爲主藥，配以浙貝母等道地名貴優質中藥材精制而成。本品主藥，即本廠獨家生産的中藥高科技産品三金西瓜霜，入心、胃、大腸經，具有清熱瀉火、消腫止痛等優良功效，尚含有人體所必需的鋅等數種微量元素和 18 種氨基酸，質量上乘，非同凡響；配以其他中藥的合理組方，科學精制，確保本品之功效奇特，療效突出，誠爲醫藥保健、居家旅行常備良藥。

特點

1. 功效奇特，作用顯著。本品抗菌譜廣，有優良的抗菌、抗病毒、抗炎消腫、止痛、止咳祛痰、促進潰瘍愈合等作用。
2. 療效高，應用範圍廣。對咽喉、嗓音、口腔、上呼吸道等多種炎症有奇效。
3. 見效快，降火、清熱、消炎止痛快速。一般噴藥后 5 — 10 分鐘即可止痛，噴藥數次症狀即可消除，患處周圍血象與體征即可恢復正常。
4. 對口腔潰瘍、口舌潰瘍、口腔炎、舌體炎、牙痛等有奇效。一般噴藥數次潰瘍即可愈合，用藥一次牙痛即可止痛或顯著改善。
5. 治防兼備，對咽喉、嗓音、口腔疾患，有治療與預防的雙重優良作用。對吸烟、飲酒、辛辣食物刺激、用嗓過度、疲勞等因素引起的咽干口燥、喉癢、干咳、聲音嘶啞、口臭等疾患均有優良的治療與預防作用。
6. 服用安全，老少皆宜。

香港總代理：
仁泰藥業有限公司

三金®
SANJIN
桂林三金藥業股份有限公司
中國・桂林

XG

Chinese version

China's Protected Traditional Chinese Medicine　Famous Traditional Chinese Medicine Brand Product

SANJIN WATERMELON FROST
NEW INSUFFLATIONS

[Ingredients] Mirabilitum Citrulli Praeparatum, Borcborax(calcined), Radix Scutellariae, Radix Sophorae Tonkinensis, Borneolum Syntheticum, Indigo Naturalis, Radix Glycyrrhizae, Cortex Phellodendri Rhizoma Coptidis, Bulbus Fritillariae Thunbergii, Rhizoma Belamcandae, Fructus Sapindi (charred), Mentholum, Radix et Rhizoma rhei.

[Pharmacological Action] Antibiosis, anti-virus, resolving heat, dispelling swelling, relieving pain, settling cough, dispelling phlegm, boosting ulcerative union, etc.

[Functions] Clearing heat and detoxicating, downbearing fire, clearing pharynx and disinhibiting larynx, diminishing inflammation, dispelling swelling and relieving pain, stanching bleeding.

[Main Indications] The inflammatory diseases of throat, throat voice, and upper respiratory tract: swelling and pain in the throat, hoarse voice, tonsillitis and pharyngitis, mouth and tongue sore or ulcerating sore, painful swollen gums, goose mouth sore in children, bad breath, acute ,chronic pharyngolaryngitis,amygdalitis,pharyngitis,parotitis,stomatitis,stomatocace,oral cavity inflammation, gingivitis, periodontitis, rhinitis, bronchitis etc . also may treat dark sore, Otitis Media, scald , burn, wound, or gingival bleeding, etc.

[Properties] This product is gray-yellow-green powder. Its smell is fragrant .Its taste is salty, sweet, light bitter and pungent-cool.

[Administration and Dosage]

1. For all diseases in oral cavity, throat, and rhinopharynx etc: spray this product on the affected part,3-6 times daily,2-4 spraying each time. For serious symptoms, the patient may take orally, 0.3-0.5g one time, 1-2 times daily.
2. Toothache: use this product to spray (or fill this product powder in by using cotton wool) on the dental caries and painful swollen gums,3-6 times daily, 2-4 spraying each time. For serious symptoms, the patient may take orally, 0.3-0.5g one time, 1-2 times daily.
3. Stanch bleeding: to spray or smear the product on the affected part, press on it for a few minutes by using cotton wool or bind it up with gauze. The bleeding may be stanched instantly.
4. Burn and scald: mix this product powder with some edible oil, smear it on the affected part, use it for numerous times in succession until it recovers.
5. Dark sore, bites of mosquito, prickly heat, Hong Kong foot, Otitis Media etc: smear the powder or spray on the affected part, use it for numerous times in succession until it recovers.

[Specification] 3g in a tube

[Storage] Sealed tightly, moisture-proof.　**[Validity]** 3 years.

[Add] 1 Gold Star Road, Guilin, China　**[Tel]** 86-773-5843251

[Fax] 86-773-5812416　**[Web Site]** http://www.sanjin.com.cn

The diseases in throat, throat voice, and oral cavity are commonly seen, frequently encountered inflammatory diseases. For traditional Chinese medical science, always there is such a saying "All diseases in throat and mouth result from fire (internal heat)".The main pathological changes of throat, voice, oral cavity and tooth: reddish swelling, pain and suppuration etc belong to the manifestations of "fire (internal heat)". Sanjin Watermelon Frost Insufflations is refined by Mr Zou Jie ming, a famous China Traditional Chinese Medicine Expert of Outstanding Contribution, a Member of China Pharmacopeia Committee, who gathered the good points of the ancient prescription, secret prescription, and proved effective prescription, integrated more than 30 years' clinical pharmaceutical experience, on the basis of "Compound Watermelon Frost" of "Kwei Feng Brand" and "Xiang Bi Shan Brand"(sold in overseas market for more than 30 years) solely produced by our factory (original Guilin Traditional Chinese Medicine Pharmaceutical Factory), improved and invented the new generation product .Its action is more strong ,curative effect is more obvious, more safe to take. It has been approved by Chinese drug administration departments .It is solely produced by the high-new technology enterprise------ Guilin Sanjin Pharmaceutical Co.,Ltd.

This product takes Sanjin® Watermelon Frost as main medicine, makes up with the genuine famous, rare quality traditional Chinese medicines of Momordica Grosvenori, Fritillariae Bulbus etc. The main medicine of the product is Sanjin Watermelon Frost, the high science and technology product of Traditional Chinese Medicine solely produced by the factory, has the good functions of clearing heat and draining fire, dispelling swelling and relieving pain. It contains numerous necessary micro elements such as zinc and 18 kinds of amino acids for human body. Its quality is high, different from ordinary products; The reasonable prescription is made up with other traditional Chinese medicines, is refined scientifically to assure its effect unique, and outstanding. It is the standing good medicine for medical healthcare, home and travel.

[Characteristics]

1. Unique effect, obvious action. This product is broad antibiotic, has good functions of antibiosis, anti-virus, diminishing inflammation and dispelling swelling, relieving pain, settling cough, dispelling phlegm, boosting ulcerative union etc.
2. High curative effect, broad application scope. It has specific effects to many kinds of the inflammatory diseases in throat, throat voice, and upper respiratory tract, etc.
3. Instant effect, downbearing fire, clearing heat, diminishing inflammation and relieving pain fast. Normally the pain may be relieved in 5-10 minutes after spraying the product. After spraying for numerous times its symptoms may be cleared, the signs of body and hemogram around the affected part may be recovered normally.
4. Has specific effects to stomatocace, mouth and tongue ulcer, stomatitis, tongue body inflammation, toothache etc. Generally after spraying numerous times, its ulcer may be healed instantly. For toothache, one time use may relieve pain or remit it obviously.
5. To treat and prevent at the same time, has double good actions of treatment and prevention for the inflammatory diseases in throat, throat voice, and oral cavity etc. For the symptoms: dry pharynx and mouth, itchy larynx, dry cough, hoarse voice, bad breath etc caused by smoking, drinking, the stimulation of pungent-hot foods, overworked throat, tiredness, etc. It has the good actions of treatment and prevention.
6. It's safe to take suitable for both old and young.

三金®
SANJIN
Guilin Sanjin Pharmaceutical Co.,Ltd.
Guilin • China

English version

Appendix II: ***Sanjin Watermelon Frost Insufflations*** **and** ***Wheat Germ E Powder (placebo)***

Name	Sanjin Watermelon Forst Insufflations	Wheat Germ E Powder
Appearance		
Ingredients	Mirabilitum Cirulli Praeparatum, Borcborax (calcined), Radix Scutellariae, Radix SoPhorae, Tonkinensis, Borneolum Syntheticum, Indigo Naturallis, Radix Glycyrrhizae, Cortex Phellodendri Rhizoma, Coptidis, Belbus Fritillariae Thunbergii, Rhizoma Belamcandae, Fructus Sapindi (Charred), Mentholum, Radix et Rhizoma rhei.	Wheat Germ
Manufacturer	Guilin Sanjin Pharmaceutical Co., Ltd.	Sweet Garden Biotechnology Food Co., Ltd.
Origin	China	Taiwan

Appendix III: Consent form

<u>參加研究者須知</u>

<u>INFORMATION SHEET FOR PARTICIPANTS</u>

西瓜霜預防聲線疲勞

Effects of watermelon frost on prevention of vocal fatigue

我們現正進行一項探討中藥西瓜霜對於預防聲線疲勞的研究，現誠意邀請閣下參與。
We wish to invite you to participate in a project which aims to investigate the preventive effects on vocal fatigue.

是項研究將需要閣下於喉部使用西瓜霜噴劑，為期一星期，目的是測試噴劑對人為引起的聲線疲勞所產生的預防作用。閣下將需要完成以下步驟:
The study will involve the use of watermelon frost spray in the oral-pharyngeal area for a period of one week. This is to determine whether watermelon frost is effective in preventing induced vocal fatigue. You will need to complete the following procedures:

1) 連續唱歌至少兩小時;
1) complete a two-hour-singing task;

2) 於喉部使用西瓜霜噴劑，每次劑量為三噴，每天三次，為期一星期;
2) use watermelon frost spray in the oral-pharyngeal area three times a day, applying three sprays each time, for a period of one week;

3) 完成聲線測試項目，包括:
4) complete a set of voice assessment procedures including:

i. 填寫一份自我聲線評估問卷;
i. complete self-perceived voice rating questionnaire;

ii. 發出一些單音及字句以用作錄音用途;
ii. Produce some syllables and read aloud some sentences for recording purposes;

iii. 發出不同的音量及音調，以紀錄聲量及音域變化;
iii. Produce voice at different pitches and loudness for documenting changes in your phonation and pitch range;

聲線測試將分三次進行，第一次測試將於閣下使用噴劑前一周完成，第二及第三次將於兩小時的歌唱測試之前和之後完成。三次的聲線測試項目均為相同。

These assessments will be carried out for three times, the first one will be carried out a week before the use of watermelon frost is started, the second and the third assessments will be completed prior to and after the singing task. All procedures will be the same for the three assessments.

所有提供的資料只供研究人員作為參考，而不會用作其他用途。參加者身分亦不會被公開。

All information provided by the participants will only be released to the investigators but the information will not be disclosed to any other people. The identity of the participants will not be revealed.

一切參與均屬自願性質，閣下亦可隨時提出終止。

Participation in this project is voluntary and the participant can withdraw from this project at any stage.

在此多謝閣下的參與。

We thank you for your interest and support.

閣下需填寫及簽署一份同意書。如有任何疑問，請聯絡以下人仕:

You will be asked to complete and sign the consent form. If you would like to ask further questions, please contact the investigator listed below:

姚文禮博士，香港大學 言語及聽覺科學學系 電話: (852) 28590583

Dr Edwin Yiu, Department of Speech and Hearing, The University of Hong Kong. Tel: (852.) 28590583

同意書
WRITTEN CONSENT FORM

本人(姓名)____

I (Name) ____

現同意參加「西瓜霜預防聲線疲勞」的研究。

I hereby consent to participate in the study entitled "Effects of watermelon frost on prevention of vocal fatigue".

本人明白此項研究的內容，其目的是測試中藥西瓜霜噴劑對預防人為的聲線疲勞的預防作用。 本人明白須於喉部使用噴劑，每天三次，每次三噴，為期一星期，完成一次歌唱測試及三次聲線測試。

I have read / understood the information about this study. I understand that the purpose of this study is to determine how effective watermelon frost spray in preventing vocal fatigue is. I understand that I have to apply three sprays in the oral-pharyngeal area for three times a day for a period one week. I also have to complete a singing task and three voice assessments.

本人已有足夠機會詢問清楚有關這項研究的內容。

I have been given the opportunity to ask questions about this study and they have been answered to my satisfaction.

本人現同意參加這項研究，而本人亦有權保留權利隨時終止參與此項研究。

I consent to participate in this study and understand that I have the right to withdraw at any time.

研究人員簽署 Investigator's signature	參加者簽署 Participant's signature
研究人員姓名 Investigator's name in block letter	參加者姓名 Participant's name in block letter
日期 Date	日期 Date

Appendix IV: Self-perceived voice subjective rating questionnaire

<u>自我評估用聲情況</u> **(I / II / III)**

參加者姓名**: _______________** 測試日期**:____/_____/____**

性別**: _____________**

年齡**: _____________**

請就閣下現時的用聲情況回答以下問題 (請於適用者打圈):

1. 整體上你對自己的聲線滿意嗎? (1 為最不滿意, 5 為最滿意)

   1-------------2-------------3-------------4-------------5

2. 你對自己發聲時的運氣情況滿意嗎? (1 為最不滿意, 5 為最滿意)

   1-------------2-------------3-------------4-------------5

3. 你對自己發聲時的聲量 (大小聲的運用) 滿意嗎? (1 為最不滿意, 5 為最滿意)

   1-------------2-------------3-------------4-------------5

4. 你對自己發聲時的音調 (高低音的運用) 滿意嗎? (1 為最不滿意, 5 為最滿意)

   1-------------2-------------3-------------4-------------5

5. 你發聲時感到困難嗎? (1 為非常困難, 5 為沒有困難)

   1-------------2-------------3-------------4-------------5

6. 你發聲時喉部感到疲勞嗎? (1 為非常疲勞, 5 為沒有疲勞)

   1-------------2-------------3-------------4-------------5

<u>Self – Perceived Subjective Voice Rating</u> (I / II / III)

Name of Participant: _______________ **Date of examination:____/_____/____**

Sex: _____________

Age: _____________

Please answer the following questions according to your voice use at the moment (please circle the most appropriate number):

1. Are you satisfied with your voice use in general? (1 as the least dissatisfied , 5 as the most satisfied)

   1-------------2-------------3-------------4-------------5

2. Are you satisfied with your breath control during phonation? (1 as the least dissatisfied , 5 as the most satisfied)

   1-------------2-------------3-------------4-------------5

3. Are you satisfied with your intensity control during phonation? (1 as the least dissatisfied , 5 as the most satisfied)

   1-------------2-------------3-------------4-------------5

4. Are you satisfied with your pitch control during phonation? (1 as the least dissatisfied , 5 as the most satisfied)

   1-------------2-------------3-------------4-------------5

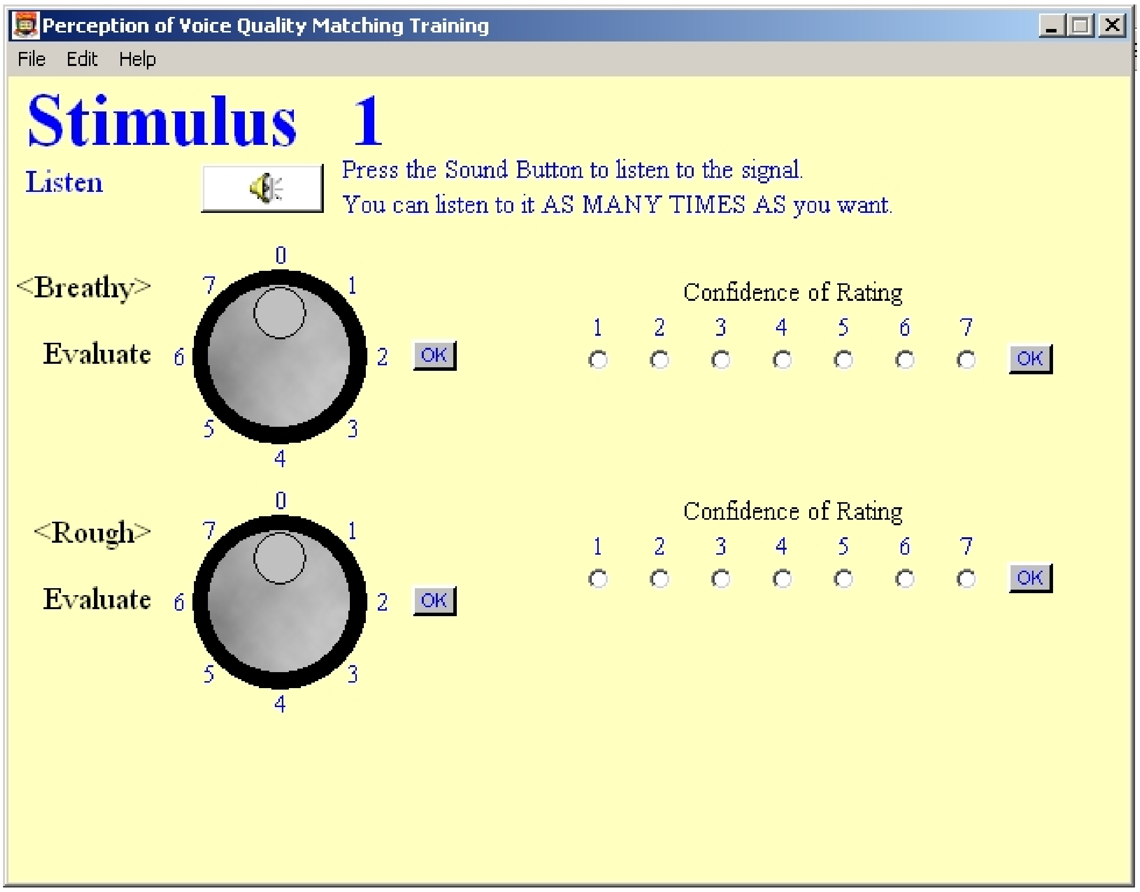
5. Do you find it difficult to initiate phonation? (1 as extremely difficult, 5 as no difficulty)

   1-------------2-------------3-------------4-------------5

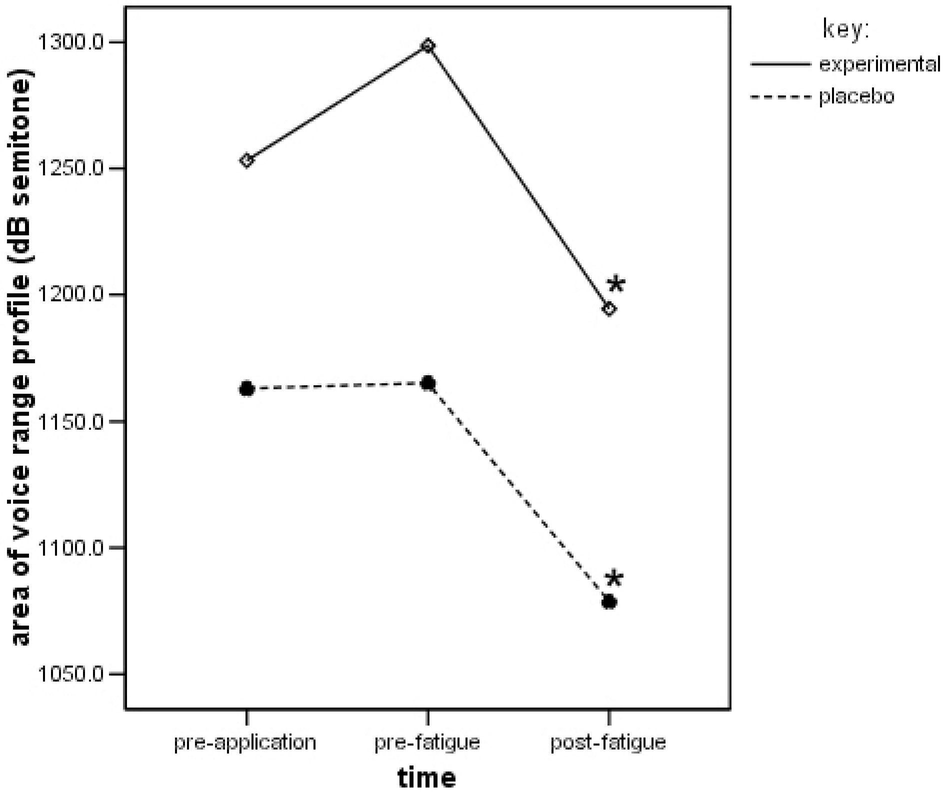
6. Are you feeling tiredness in your throat during phonation? (1 as extremely tired, 5 as no tiredness)

   1-------------2-------------3-------------4-------------5

Appendix V: Eight-point equal-appearing-interval (EAI) scale

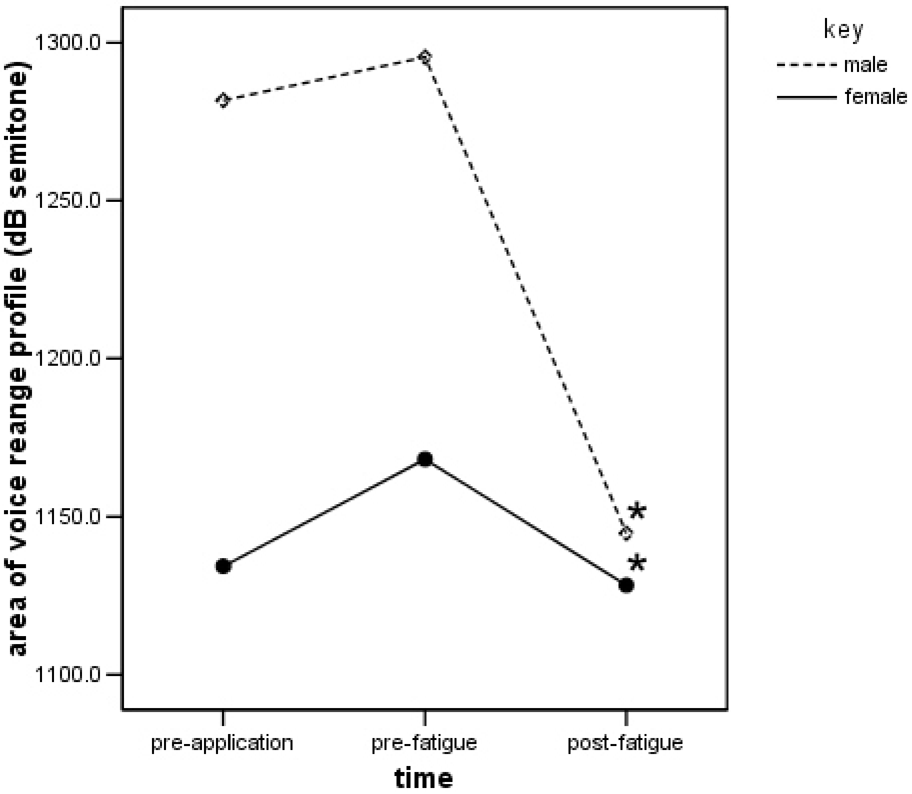


Appendix VI: Figures showing significant changes



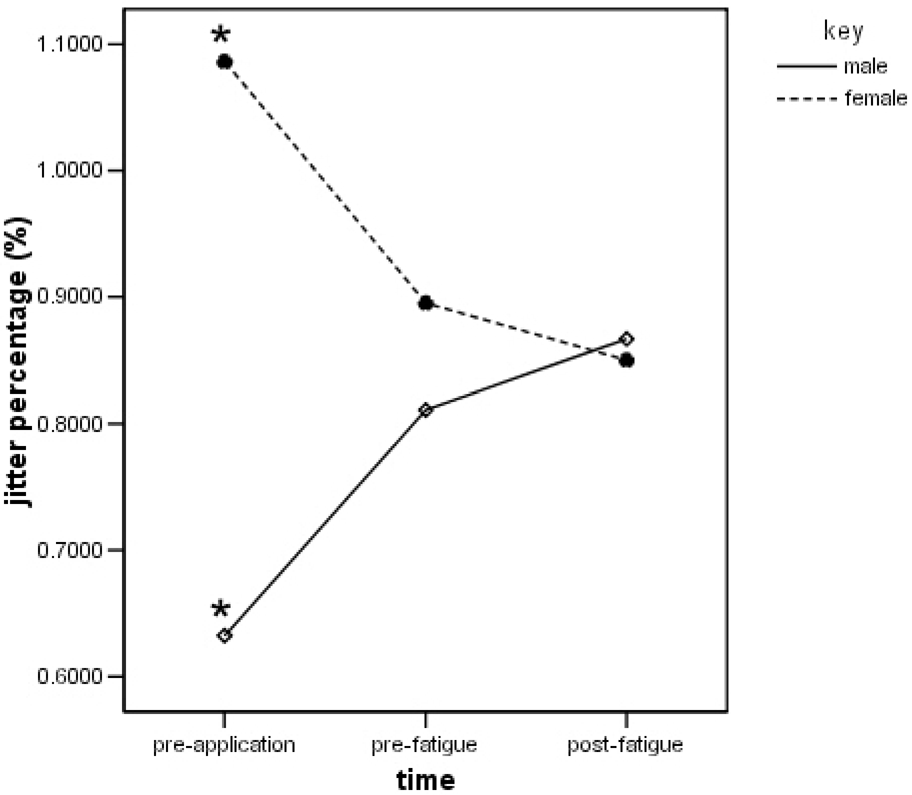
* significantly different at 0.05 level

Figure 1. *Changes in area of voice range profile (in dB semitone) between experimental and placebo groups*



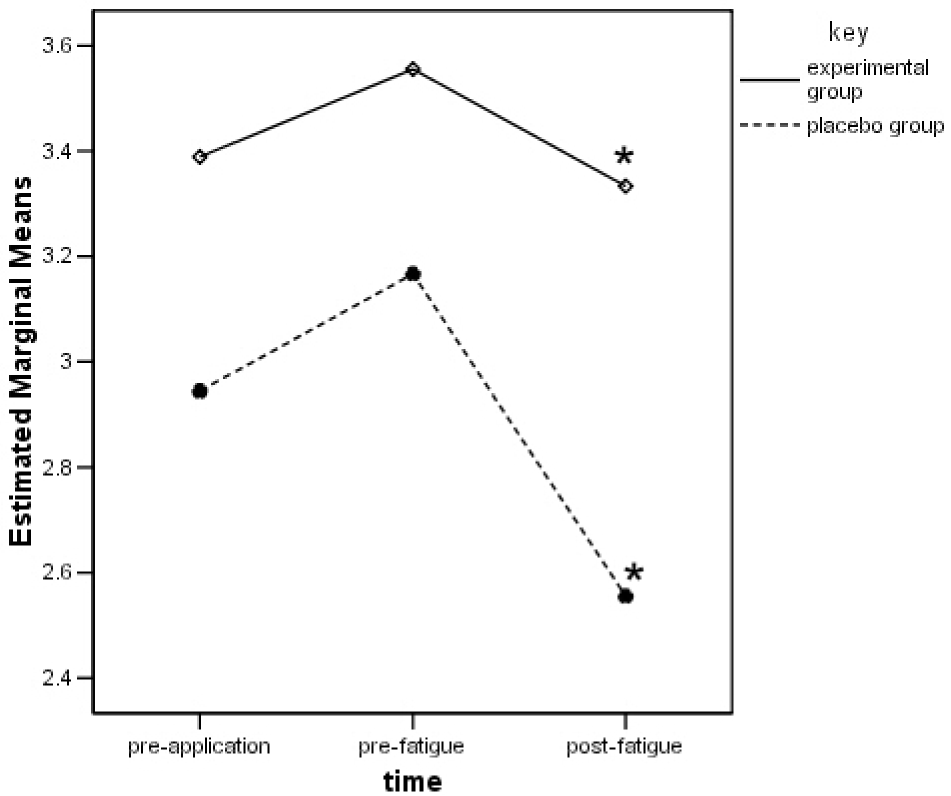
* significantly different at 0.05 level

Figure 2. *Changes in area of voice range profile (in dB semitone) between male and female participants.*



* significantly different at 0.05 level

Figure 3. *Changes in jitter percentage (%) between male and female participants*



* significantly different at 0.05 level

Figure 4. *Changes in ratings of general voice use between experimental and placebo groups*