

Investigation of the Effects of Relaxation and Respiratory Exercises on Anxiety in Thyroid Biopsy Patients

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Abstract

It is known that high stress levels increase the risk of complications in invasive procedures. The aim of this study is to investigate the effect of the relaxation and respiratory exercise on the anxiety state specific to the thyroid biopsy procedure in patients. A hundred randomly selected thyroid biopsy patients from 200 patients watched the Relaxation and Respiratory Exercise video (Group 1) and did the instructions, and the rest 100 patients were control group (group 2) who did not watch it. The State-Trait Anxiety Inventory state scale (STAI-I) and trait anxiety scale (STAI-II) were administered to all patients before the procedure. Group 1 watched the video just before the biopsy procedure. After the biopsy procedure, all the 200 patients rested for 15 minutes, and the STAI- I scale was filled again. When the STAI-II and pre-biopsy STAI-I questionnaire results of the patients were compared between two groups; study group (Group 1): Patients who were selected to watch the video, control group (Group 2): Patients who were not watching the video), no statistically significant difference was found ($p>0,005$). When the anxiety levels in the first STAI-I performed before the biopsy and the STAI-I tests performed after the procedure were compared, a statistically significant decrease was found in the anxiety levels of the SG ($p<0,001$). Anxiety levels of female patients were statistically significantly higher than male patients. Although thyroid biopsy is a minimally invasive procedure, it may cause anxiety in patients. Watching the video before the procedure resulted in a decrease in the anxiety levels of the patients.

Keywords: anxiety, biopsy, thyroid, relaxation, respiratory exercises

Öz

Tiroid Biyopsi Hastalarında Gevşeme ve Nefes Egzersizlerinin Anksiyete Üzerine Etkilerinin İncelenmesi

Yüksek stres düzeylerinin invaziv işlemlerde komplikasyon riskini artırdığı bilinmektedir. Bu çalışmanın amacı hastalarda gevşeme ve nefes egzersizlerinin tiroid biyopsisi işlemine özgü anksiyete durumuna etkisini araştırmaktır. İki yüz hastadan rastgele seçilen 100 tiroid biyopsisi hastası Gevşeme ve Nefes Egzersizi videosunu izleyip talimatları yaptı, geri kalan 100 hasta videoları izlemeyen kontrol grubuna alındı. Tüm hastalara işlem öncesi Durumluk-Sürekli Anksiyete Envanteri (DSAE) durumsal anksiyete ölçeği (DSAE-I) ve sürekli anksiyete ölçeği (DSAE-II) uygulandı. Grup 1 biyopsi işleminden hemen önce videoyu izledi. Biyopsi işleminden sonra 200 hastanın tamamı 15 dakika dinlendirildi ve DSAE-I skalası tekrar dolduruldu. Hastaların DSAE-II ve biyopsi öncesi DSAE-I anket sonuçları iki grup arasında karşılaştırıldığında istatistiksel olarak anlamlı bir fark bulunmadı ($p>0,005$). Biyopsi öncesi yapılan ilk DSAE-I ile işlem sonrası yapılan DSAE-I testlerindeki kaygı düzeyleri karşılaştırıldığında, video izletilen grubun kaygı düzeylerinde istatistiksel olarak anlamlı bir azalma bulundu ($p<0,001$). Kadın hastaların kaygı düzeyleri erkek hastalara göre istatistiksel olarak anlamlı derecede yüksekti. Tiroid biyopsisi minimal invaziv bir işlem olmasına rağmen hastalarda anksiyeteye neden olabilir. İşlem öncesi videonun izlenmesi hastaların anksiyete düzeylerinde azalma sağlayabilir.

Anahtar Kelimeler: anksiyete, biyopsi, tiroid, gevşeme, nefes egzersizleri

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INTRODUCTION

Fine-needle aspiration biopsy of the thyroid gland is safe, inexpensive, minimally invasive, and has high diagnostic value in nodular diseases of the thyroid (Gharib & Goellner, 1993; Papini et al., 2019). It has very few complications and is more diagnostic than any other method. Various studies have reported sensitivity ranging from 57% to 98%. The most important limitation is the non-diagnostic aspirates, which are encountered at a rate of 20% (Gharib, 1994; Mufti & Molah, 2012). In addition to the invasiveness of the procedure and the waiting time before the biopsy procedure, the meaning of the procedure for the patient, the lack of information about the procedure, the problems that may be experienced during and after the procedure, the pain during and after the procedure, or the interruption of daily life are among the causes of anxiety (Zisman, Leibovici, Kleinmann, Siegel & Lindner, 2001). Various methods are used to reduce the anxiety levels of patients. Informing the patient about all stages and listening to classical music before the biopsy are some of these methods (Deane and Gegner, 1998; Chang, Oh, Lee, Park, Seo, Jeong, & Kwon, 2015; Yeo, Cho, Oh, Park, & Park, 2013; Özdemir, Taşcı, Yıldızhan, Aslan, & Eser, 2019). Various studies have investigated the effect of breathing exercises on anxiety. It was determined that the patient's anxiety decreased significantly when relaxing breathing exercises were performed (Kim & Kim, 2005; Sellakumar, 2015; Bailey, 2010; Bastani, Hidarnia, Kazemnejad, Vafaei, & Kashanian, 2005).

One of the most frequently used tests in medicine for the measurement of anxiety is the State-Trait Anxiety Inventory (STAI) scale developed by Spielberger (2010). This test measures state and trait anxiety levels. While the state anxiety scale measures the anxiety level of the person at a certain moment and under certain conditions, the trait anxiety scale measures how the person generally feels regardless of the situation and conditions. In this study, we aimed to evaluate the effect of the "Muscle Relaxation and Respiratory Exercise" video published by the TPA on the anxiety level of patients who will undergo thyroid biopsy, using the STAI anxiety scale. Previous studies have investigated the effects of various factors such as classical music, aromatherapy or informative video on anxiety and pain levels in patients who will undergo diagnostic minimally invasive procedures such as breast biopsy, bone marrow biopsy, prostate biopsy or thyroid biopsy. (Abbaszadeh, Tabari, & Asadpour, 2020; Dell'Atti, 2021; Bennett, Swan, Gazelle, & Saksena, 2020; Ay, Ata, and

Oncu, 2022; Akin, 2021). From this point of view, we aimed to investigate the effect of relaxation and breathing exercise video on the level of anxiety in patients who were referred to our clinic and undergoing diagnostic thyroid biopsy. Our hypothesis is that the anxiety levels of the patients who are watched the video will be lower than the control group that is not watched.

MATERIALS AND METHODS

After obtaining the approval of the Ethics Committee of our hospital, euthyroid female and male patients aged 20–60, literate, without additional psychiatric disease, who had a thyroid biopsy appointment for thyroid nodule or mass etc. in the Interventional Radiology. Our study included 2 groups as control and study groups. The study group was designed as the group who watch the video before the biopsy procedure and the control group as the group that did not watch the video before the biopsy procedure.

To estimate the sample size, power analysis was performed considering studies with confidence level of 95% and test power of 95%, and the number of samples for each group was found to be 94, total 188. Considering some reduction due to various reasons and exclusion criteria, a total of 223 patients were planned to be included in the study.

After the ethical approval of the study, 223 patients who applied to our clinic for diagnostic thyroid biopsy were informed about the study. Twenty-three patients (10.3%) who did not want to participate in the study and did not meet the inclusion criteria were excluded. Consent was obtained from the patients who were informed about the study and agreed to participate. Before the biopsy procedure, computer-based randomization was used to divide the patients into two groups, and group 1 (n=100) as the study group and group 2 (n=100) as the control group were separated. Both groups were informed by one of the researchers. Both groups were filled with STAI-I. Afterwards, the patients in Group 1 were taken to a private room by the researcher to watch the video. Before the video, the patients were briefed about the video content in a non-medical language by the researcher. It was observed whether the patients did the breathing and relaxation exercises effectively.

A total of 200 patients (Table 1) were included in our study, and 100 of them watched the "Muscle Relaxation

and Respiratory Exercise” video on the website of the Turkish Psychiatric Association (TPA), based entirely on patient volunteerism and consent. In our study, we used the 23-minute muscle relaxation and breathing exercise video published by the TPA to deal with stress. The video consists of two parts. There are progressive muscle relaxation exercises in the first part of the video, and deep breathing exercises in the second part. Progressive muscle relaxation exercises reduce heart rate, respiratory rate, and blood pressure. Deep breathing exercises are one of the most frequently used methods to reduce anxiety. The video clearly explains how to do these exercises in the form of instructions.

Patients in the control group did not watch the video. The STAI anxiety scale was used to assess anxiety. State-Trait Anxiety Inventory scale is one of the widely used anxiety scales developed by Spielberg et al. in 1970 to measure state (STAI-I) and trait (STAI-II) anxiety (Spielberger, 2010). Adaptation of the scale to Turkish language, reliability and validity studies were done by Öner and Le Comte (Öner, and Le Compte, 1998; Büyükoztürk, 1997). In many studies, the STAI anxiety scale is accepted as the gold standard in pre-operative anxiety assessment (Tenenbaum, Furst, & Weingarten, 1985; Vadhanan, Tripaty, & Balakrishnan, 2017; Kindler, Harms, Amsler, Ihde-Scholl, & Scheidegger, 2000). While state anxiety defines the anxiety that a person feels because of the situation he/she is in, trait anxiety describes the anxiety that is not dependent on the situation and time in which the person is and is relatively continuous. There are a total of 20 questions in the state and trait scale with a scoring system between 1 and 4 in each. One point indicates that the situation stated in the question does not reflect themselves at all, while 4 points indicate that the stated situation completely reflects.

Before the biopsy procedure, all patients were given preliminary information about how the procedure would be performed and a personal information form was filled. Group 1 watched the video just before the biopsy procedure. Subsequently, after the video group patients watched the video, first the STAI state (STAI-I) scale and then the trait (STAI-II) anxiety scale were read to all patients by one-on-one interview method by the researcher. The places corresponding to the answers given by the patients were marked accordingly on the scale (Questionnaire 1). While the responses in the state anxiety scale are “never”, “sometimes”, “a lot”, “completely” according to the degree of

severity; the responses in the trait anxiety scale are “almost never”, “sometimes”, “often”, “almost always” according to the degree of frequency. All patients were taken to the biopsy area one by one and fine needle aspiration biopsy was performed on the thyroid under sterile conditions. After the patients rested for 15 minutes after the biopsy, the STAI-I scale was read once again to the patients by one-on-one interview method and the places on the scale corresponding to the answers given by the patient were marked (Questionnaire 2). At the end of the questionnaire, the STAI scores of the patients were calculated. There are direct and inverted phrases expressing positive and negative emotions in the scales. There are 10 inverted statements in the state anxiety scale and 7 in the trait anxiety scale. When scoring, the sum of the inverted expressions is subtracted from the sum of the direct expressions and a constant is added to this number. This is 50 for the steady state anxiety scale and 35 for the trait anxiety scale. The scores obtained from both scales are in the range of 20–80. A high score indicates a high level of anxiety, and a low score indicates a low level of anxiety.

Statistical Analysis

Statistical Package for Social Sciences (SPSS) program version 20 (IBM Inc., Chicago, IL) was used to analyze the data. Results were given as mean \pm standard error for continuous variables and as count and percentages (%) for categorical variables. Gender, age, educational status, family history of thyroid cancer and marital status were compared by chi-square test. We divided patients into two groups; Group 1: Patients who watched video, Group 2: Patients who did not watch video. The STAI-II questionnaire score of the groups were compared with independent sample t test. Anxiety reduction percentages were calculated. A p-value of less than 0.05 was considered statistically significant, and all analyses were two-tailed.

RESULTS

A total of 200 patients aged 20–60 years (mean 40.35 ± 12.18 years) were included in this study. Of the included patients, 60 were male and 140 were female. There was no significant difference between the patients in the video-watching ($n=100$) and non-watching ($n=100$) groups in terms of gender, age, educational status, and marital status. The STAI-II questionnaire score, which indicates the trait anxiety status of the patients participating in the study, was found to be in the range of 20–69

(mean 44.61 ± 9.27). The STAI-II questionnaire score of the first group who did not watch the video was in the range of 20–68 (mean 44.70 ± 9.23), while the score of the group who was watched video ranged 23–69 (mean 44.53 ± 9.36), and there was no statistically significant difference between the two groups ($p=0.89$) (see: Table 1).

The mean pre-biopsy anxiety score of the 200 patients included in our study was 47.19. The mean of the group that watched and did not watch the video was found to be 47.89 and 46.49, respectively.

When the results of the STAI-I questionnaire filled out before the biopsy procedure were compared, no statistically significant difference was found between the groups who watched and those who did not watch ($p>0.05$). When the results of the STAI-I questionnaire filled after the biopsy procedure were examined, it was shown that there was a decrease in the anxiety level in both groups compared to the pre-procedure, but the decrease was higher in the video-watched group, and this difference was statistically significant ($p<0.001$) (Table 2). Anxiety reduction percentages were found to be 14.5% in the group that did not watch, and 26.7% in the group that watched, and a statistically significant difference was found ($p<0.001$) (Table 3).

When the relationship between gender and STAI score was examined, it was seen that the results of the STAI-II questionnaire and the pre-procedure STAI-I questionnaire were higher in female patients than in male patients, and this difference between the two groups was statistically significant ($p<0.001$). When the relationship between gender and the results of the post-biopsy STAI-I questionnaire was examined within the video-watching and non-watching groups, the anxiety levels of female patients were higher than male patients in both groups; however, when the percentages of decrease in anxiety compared to the pre-biopsy level were compared, there was no significant difference between male and female patients ($p>0.05$).

When the relationship between age and anxiety levels was examined, it was seen that the STAI-II and pre-biopsy STAI-I scores were lower in patients under the age of 40 (47%) than in patients over the age of 40 (53%), and this difference was statistically significant ($p<0.05$). While the relationship between family history of thyroid Ca, smoking, coronary artery disease and post-procedure anxiety was not correlated; age, gender, educational status, job status, and marital status were significantly correlated with STAI-I ($p<0.05$).

Table 1: Patients' characteristics

	Group 1 (n=100)	Group 2 (n=100)
Ages (mean \pm SD)	40.2 \pm 12.51	41.3 \pm 11.25
Gender (female/male)	72/28	68/32
Education \leq highschool (%)	53	64
Marriage status (married/single) (%)	36/64	75/25
Working/employed (%)	52	45
Smokers (%)	14	25
Alcohol user (%)	3	5

Table 2: Preoperative and postoperative anxiety scores of the patients

	STAI-I (pre-biopsy)	STAI-I (post-biopsy)	P value
Group 1 (patients who watched video)	47.89 \pm 11.07	34.75 \pm 8.47	<0.001
Group 2 (patients who did not watch video)	46.49 \pm 11.19	39.55 \pm 10.14	<0.001

STAI: State-Trait Anxiety Inventory.
p<0.05 was considered as statistically significant.

Table 3: Comparison of some variables between groups

Parameters	Group 1 (patients who watched video)	Group 2 (patients who did not watch video)	P value
Number of patients	100	100	
Age (mean \pm SD)	40.2 \pm 12.51	41.3 \pm 11.25	0.618
Gender (female/male)	72/28	68/32	0.644
STAI-II	44.53 \pm 9.36	44.7 \pm 9.23	0.895
STAI-I (pre-biopsy)	47.89 \pm 11.07	46.49 \pm 11.19	0.375
STAI-I (post-biopsy)	34.75 \pm 8.47	39.55 \pm 10.14	<0.001
Percentage of STAI-I score reduction after biopsy	%26.73 \pm 9.55	%14.57 \pm 10.42	<0.001

STAI: State-Trait Anxiety Inventory; SD: Standart deviation.
p<0.05 was considered as statistically significant.

Of the patients included in the study, 117 (58.5%) were high school or below education level, and 83 (41.5%) were university graduates. When the relationship between education level and STAI score was examined, it was seen that STAI-II and pre-biopsy STAI-I scores were lower in university graduate patients than in patients with high school

or below education level, and this difference between the two groups was statistically significant ($p < 0.05$). When the relationship between the post-biopsy STAI-I scores and the education level was examined within the video-watching and non-watching groups; it was shown in both groups that the anxiety levels of the university graduate patients were lower than high school graduate or lower education level patients, and this was statistically significant. Furthermore, when the relationship between educational status and pre-to post-procedure anxiety reduction was examined within each video-watching and non-watching groups, no statistically significant difference was found ($p > 0.05$).

State-Trait Anxiety Inventory scores are generally classified as “no anxiety or low anxiety” (≤ 35), “moderate anxiety” (36–42) and “high anxiety” (≥ 42) (Marteau, and Bekker, 1992). Table 4 shows the number of patients in both groups with low, moderate, severe anxiety levels. In a study by Ay et al. it was shown that the decrease in anxiety scores in patients with high anxiety was higher than in patients with moderate and low anxiety (Ay et al., 2022). In our study, however, no significant difference was found in the decrease in anxiety scores between patients with high pre-biopsy anxiety score (STAI score ≥ 42) and patients with moderate or low anxiety score (STAI score < 42) ($p < 0.05$).

Table 4: State-Trait Anxiety Inventory scores of patients based on anxiety levels

STAI-I (pre-biopsy)	STAI-II (post-biopsy)			
	Group 1	Group 2	Group 1	Group 2
Low anxiety ≤ 35 (n)	15	19	53	37
Moderate anxiety 36–42 (n)	10	17	28	29
Severe anxiety ≥ 42 (n)	75	64	19	34

STAI: State-Trait Anxiety Inventory.

Thirty-five patients (17.5%) included in the study had a previous history of thyroid biopsy. When the STAI-II and pre-biopsy STAI-I results of patients with a previous biopsy history were compared with those of patients without a biopsy history, it was seen that these patients had higher anxiety levels, nonetheless the difference was not statistically significant. When the relationship of the previous biopsy history with the post-biopsy STAI-I scores or the percentage of anxiety score decrease compared to the pre-biopsy level was examined within video-watching or non-watching groups, there was no statistically significant difference ($p > 0.05$).

Nine patients included in our study had a family or personal history of thyroid cancer. When the relationship between anxiety levels and thyroid cancer history was examined, no statistically significant difference was found ($p > 0.05$).

Of the patients included in the study, 8 (4%) were alcohol users. When the relationship between alcohol and anxiety levels was examined, it was seen that the STAI-II and pre-biopsy STAI-I scores were higher in patients under the alcohol users than in non-alcoholic patients, and this difference was statistically significant ($p < 0.05$). When the relationship between the post-biopsy STAI-I scores and alcohol use was examined within the groups; no statistically significant difference was found ($p > 0.05$).

Of the patients included in the study, 39 (19.5%) were smokers. When the relationship between smoking and anxiety levels was examined, it was seen that the STAI-II and pre-biopsy STAI-I scores were higher in smoker patients than in non-smoker patients, and this difference was statistically significant ($p < 0.05$). When the relationship between the post-biopsy STAI-I scores and smoking was examined within the groups; no statistically significant difference was found ($p > 0.05$).

DISCUSSION

Since biopsy is an invasive procedure, it causes anxiety, and many studies have tried methods to reduce anxiety before invasive procedures (Abbaszadeh et al., 2020; Dell’Atti, 2021; Bennett et al., 2020; Ay et al., 2022; Akın, 2021). In their study, Michael Haun FNPC et al. evaluated the effect of listening to classical music for 20 minutes on anxiety, by measuring vital functions (blood pressure, heart rate, respiratory rate, etc.) and STAI state anxiety scale in 20 female patients waiting for breast biopsy. They reported anxiety level to be considerably lower in the group that listened to music (Haun, Mainous, & Looney, 2001; Cooke, Chaboyer, Schluter, & Hiratos, 2005; Jiménez-Jiménez, García-Escalona, Martín-López, De Vera-Vera, & De Haro, 2013). Similarly, in our study, it was observed that the anxiety levels of patients who underwent thyroid biopsy after watching the video were lower than those who did not.

In their study with 100 patients, Reza Shabanloei et al. evaluated the effect of music on post-procedural anxiety and pain in patients who will undergo bone marrow biopsy & aspiration and reported that both anxiety and

pain were lower in the group listening to music compared to the control group who did not listen to music (Shabanloei, Golchin, Esfahani, Dolatkah, & Rasouljan, 2010).

In our study, it was observed that the anxiety levels of the patients who watched a video of relaxing breathing exercises before the biopsy procedure were significantly lower than the group that did not watch.

Anxiety disorders are more common in women than in men (Pigott, 1999). However, only a small number of research have looked at whether women with anxiety disorders vary from men with the same disease in some ways. The reason for the increased vulnerability to anxiety for women remains largely unclear. According to recent findings, female reproductive hormones and associated cycles may be crucial (Pigott, 1999). Since neurotransmitter systems are major mediators of the anxiety response and estrogen and progesterone play crucial roles in their regulation, they have been linked to heightened susceptibility for anxiety disorders in women (Seeman, 1997; Shear, 1997; Reynolds, 2018). In our study, the anxiety level of women was found to be higher than that of men.

In our study, the anxiety level of the youth was lower than that of the elderly. In addition, the fact that young patients had more information about the procedure, complications of the procedure and biopsy results may have contributed to lower anxiety levels.

When the relationship between education level and anxiety level was examined in both groups who watched and did not watch the video, it was seen that the level of anxiety was lower in people with higher education levels. Today, with the developing technology and the increasing use of the internet, easy access to much information is provided (Wald, Dube, & Anthony, 2007). For this reason, it is estimated that people with a high level of education can have detailed information about the procedure and its results, and this makes the patients more comfortable.

Similarly, in the video-watching group, the anxiety level of people with higher education level was lower than those with low education level. The reason for this is estimated that the breathing exercises described in the video may have been performed more effectively by people with higher education levels. The level of anxiety was higher in patients who had a previous thyroid biopsy (although there was no statistically significant difference) than those

without a history of biopsy. When we designed our study, we expected a higher level of anxiety in patients who would undergo biopsy for the first time. However, in our study, the anxiety level was found to be higher in patients who had previously undergone thyroid biopsy. We would expect the anxiety level to be higher in patients with a family history or a personal history of thyroid cancer. However, in our study, no significant difference was found between the anxiety levels of those with a history of thyroid cancer and those without.

Epidemiological data indicate that smoking increases the risk of developing anxiety disorders (Zvolensky, Kotov, Antipova, & Schmidt, 2003). In the study of Morissette SB et al., in 527 patients, when smokers and non-smokers with anxiety disorders were compared, it was reported that smokers experienced more anxiety sensitivity, anxiety symptoms, depressed mood, negative affect and stress (Morissette, Brown, Kamholz, & Gulliver, 2006). However, when the literature is analyzed, it is seen that smoking is highly comorbid with anxiety disorders and clinical depression, and this relationship is affected by variables such as the age of the smoker and the level of nicotine addiction (Morrell & Cohen, 2006). In the study "Effect of music on level of anxiety in patients undergoing colonoscopy without sedation" by Ko et al., the relationships between blood pressure, BMI, history of hypertension, smoking, alcohol consumption and anxiety before colonoscopy examination were examined; however, no significant difference was found between any item and anxiety state (Ko et al., 2017).

Clinical reports show that people use alcohol as a coping strategy with social fears as well as with stress. A standard theory of why people drink—the tension relief hypothesis—implies that alcohol acts to reduce stress and anxiety. This can be seen as an incentive for people to drink alcohol (Book & Randall, 2002). When a person recovers from stress after consuming alcohol, they are likely to continue to use alcohol because of its stress-reducing properties. Whether alcohol actually reduces stress is debatable (Carrigan & Randall, 2003). In our study, the pre-procedural anxiety level of patients who used alcohol was found to be higher than those who did not use alcohol. However, this result may be misleading due to the small number of patients (4%) who used alcohol in the study.

There were some limitations in the study. The relaxing breathing exercise video we used in our study is very time consuming because it is 23 minutes. However, since

thyroid biopsy is a widely used method for diagnosis, the high number of patients causes very little time per patient. Although there were 200 patients in our study, there could have been more patients since it was a study based on a questionnaire. Since participation in the study and watching videos were voluntary, the video-watching group in our study population may have consisted of relatively more agreeable and positive people, which may have caused bias in our study. “Due to the low number of patients who used alcohol in the patients included in the study, the results of the relationship between alcohol use and anxiety levels may be insufficient. In addition, the amount of alcohol use (social drinker-dependent) may be effective. When the literature is examined, it is seen that the relationship between smoking and anxiety disorders is affected by variables such as the age of the smoker and the level of nicotine addiction.

CONCLUSIONS

In conclusion, in our study, it was determined that the patients experienced anxiety before and after the thyroid biopsy, and their anxiety levels were reduced by watching relaxation and respiratory exercise video. Controlled randomized studies with larger participants may provide valuable information.

Ethics Committee Approval: The study was approved by the Ethics Committee of Haydarpasa Numune Training and Research Hospital (date and number of approval: 2020 / KK/124).

Informed Consent: Informed consent was obtained from all individual participants included in the study.

Peer-review: Externally peer-reviewed.

Conflict of Interest: The authors declare no conflict of interest.

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