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SEÇÃO: ARTIGOS

## The effect of the “K” technique on levels of anxiety and salivary cortisol in adolescents

*O efeito da técnica “K” nos níveis de ansiedade e cortisol salivar em adolescentes*  
*El efecto de la técnica “K” sobre la ansiedad y los niveles de cortisol salivales en adolescentes*

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**Abstract:** This research presents theoretical arguments of a new approach called Technique “K,” which aims to reduce student’s anxiety before a cognitive challenge. Through a randomized, controlled clinical trial, we evaluate the Beck Anxiety Inventory and salivary cortisol. We compared the results in the last year of high school, who were facing school tests, before and after the application of the “K” technique (intervention group) and placebo (control group). Statistical analyses consisted of using the Chi-square, Spearman and the Wilcoxon test. The principal component analysis observed in the case group, - 10.50 (-18.25; -4.75), was statistically higher than the control group, -6.00 (-8.75; -1.25). It is not possible to identify statistical significance ( $p = 0.462$ ) in the variation of salivary cortisol concentration. The results suggest the effectiveness of the “K” technique in managing anxiety in high school students. Although, future research is needed to extend the preliminary data obtained in this study.

**Keywords:** anxiety; learning; cognition.

**Resumo:** Esta pesquisa apresenta argumentos teóricos de uma nova abordagem denominada Técnica “K”, visando reduzir a ansiedade do aluno diante de um desafio cognitivo. Por meio de um ensaio clínico randomizado e controlado, avaliamos o Inventário de Ansiedade de Beck e o cortisol salivar. Comparamos os resultados dos alunos, que estavam enfrentando provas escolares, antes e após a aplicação da técnica “K” (grupo intervenção) e placebo (grupo controle). As análises estatísticas consistiram na utilização do Qui-quadrado, Spearman e teste de Wilcoxon. A análise de componentes principais observada no grupo caso, -10,50 (-18,25; -4,75), foi estatisticamente superior ao grupo controle, -6,00 (-8,75; -1,25). Não é possível identificar significância estatística ( $p = 0,462$ ) na variação da concentração de cortisol salivar. Os resultados sugerem a eficácia da técnica “K” no manejo da ansiedade em estudantes do ensino médio. No entanto, pesquisas futuras são necessárias para estender os dados preliminares obtidos neste estudo.

**Palavras chave:** ansiedade; aprendizado; cognição.

**Resumen:** Esta investigación presenta argumentos teóricos para un nuevo enfoque denominado Técnica “K”, con el objetivo de reducir la ansiedad de los estudiantes frente a un desafío cognitivo. A través de un ensayo clínico aleatorizado y controlado, evaluamos el Inventario de Ansiedad de Beck y el cortisol salival. Comparamos los resultados de los estudiantes, que se enfrentaban a pruebas escolares, antes y después de la aplicación de la técnica “K” (grupo de intervención) y placebo (grupo control). Los análisis estadísticos consistieron en utilizar la prueba de Chi-cuadrado, Spearman y Wilcoxon. El análisis de componentes principales observado en el grupo de casos, -10,50 (-18,25; -4,75), fue estadísticamente superior al grupo control, -6,00 (-8,75; -1,25). No es posible identificar significación estadística ( $p = 0,462$ ) en la variación de la concentración de cortisol salival. Los resultados sugieren la efectividad de la técnica “K” en el manejo de la ansiedad en estudiantes de secundaria. Sin embargo, se necesitan



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futuras investigaciones para ampliar los datos preliminares obtenidos en este estudio.

**Palabras clave:** ansiedad; aprendizaje; cognición.

## Introduction

Anxiety is a complex response in which individuals try to anticipate events due to their potential risk (American Psychiatric Association, 2014; Barker et al., 2018). It is a result of a delicate combination of emotional, physiological, cognitive, and behavioral aspects. In an evolutive perspective, the attentional focus is maintained in dangerous future events and prepares the individual to fight/flight responses (Barker et al., 2018; Koutsimani, Montgomery, & Georganta, 2019).

The biological aspects of stress and anxiety processes involve the sympathetic nervous system and the Hypothalamic, Hypophysis, Adrenal (HHA) axis. HHA is also responsible for recovering homeostasis (Kauer-Sant'Anna, Brietzke, & Quevedo, 2011; Rotta, Ohlweiler, & Riego, 2016). The production of Corticotrophin-releasing hormone (CRH) and antidiuretic hormone or vasopressin (ADH) increases the secretion of ACTH (adrenocorticotrophic hormone), cortisol (glucocorticoid synthesized from cholesterol and secreted by the adrenal cortex), adrenaline, vasopressin, so-called "stress hormones" (Kauer-Sant'Anna et al., 2011; Rotta et al., 2016). These hormones, in high levels, have a direct effect on attention bias and memory and may lead to memory deficits and reduced neuroplasticity (Izquierdo, 2011).

Attentional Control Theory (ACT) (Eysenck & Derakshan, 2011; Eysenck, Derakshan, Santos, & Calvo, 2007) is a broad model about the influences of anxiety in cognitive functioning. According to ACT, dangerous stimuli may influence attention control systems, leading to deficits in executive function and, by so, impairment in cognitive tasks. According to this, the focus shifted to aversive stimuli rather than controlling attention to other stimuli, influencing performance and cognition (Shi, Sharpe, & Abbott, 2019). Anxiety leads to physical symptoms and increased focus shifting, affecting learning in college students (Shin & Liberson, 2010; DeSousa, Moreno, Gauer, Manfro,

& Koller, 2013; Syokwaa, Aloka, & Ndunge, 2014). An example of the relationship between anxiety and cognitive performance was reported by Hood, Pulvers, Spady, Kliebenstein and Bachand (2015), suggesting deficits in working memory as a function of anxiety levels, mediated by cortisol levels.

Several techniques are effective in treating anxiety disorders. Some of these are: relaxation technique (Wernder-Seidler, Perry, Yael, Newby, & Christensen, 2017; Putwain & von der Embse, 2021), cognitive restructuring (Garnefski & Kraaij, 2018; Grist, Croker, Denne, & Stallard, 2019; de Hullu, Sportel, Nauta, & de Jong, 2017; Putwain & von der Embse, 2021; Yusufov, Nicoloso-SantaBarbara, Grey, Moyer, & Lobel, 2019), exposure techniques/systematic desensitization (Putwain & von der Embse, 2021; Wernder-Seidler et al., 2017) and body relaxation/meditation (Putwain & von der Embse, 2021; Wernder-Seidler et al., 2017; Yusufov et al., 2019). However, there is still an essential lack of understanding in how anxiety techniques should be combined during their use.

The techniques help people deal with symptoms even though they fulfill the criteria for anxiety disorder. Other studies suggest techniques that help with cognitive challenges and anxiety symptoms. Use of imagery techniques on health anxiety and biological changes (Tolgou et al., 2018), mindfulness and the impact on cortisol levels (Manigault, Woody, Zoccola, & Dickerson, 2018), and cortisol as a marker for improvement in mindfulness-based stress reduction (Matousek, Dobkin, & Pruessner, 2010), are examples of these. Despite the promising results of these techniques, some limitations are considered regarding the use of cortisol as a biomarker (Manigault et al., 2018; Matousek et al., 2010; Tolgou et al., 2018), considering that many variables (such as cortisol sensitivity during measuring, time of collection, the amount collected, other biomarkers that influences cortisol) may contribute to biased results.

This study aimed to evaluate the impact of a technique to decrease anxiety in students facing a cognitive challenge. We expect to observe lower levels of anxiety in the intervention group compared to the control one. We also expect

lower cortisol levels in the case group compared to control, following the hypothesis that there can be an interaction between techniques that help during cognitive challenges and biological changes.

## Methods

### Participants

The sample consisted of 30 students, 17 girls and 13 boys (56.7% female) from a senior high school. Students who consumed any drink or food within thirty minutes before cortisol collection had any oral lesion / dental treatment or brushed their teeth within the last three hours were considered exclusion criteria (Jones, Watkins, Hand, Warren, & Cowen, 2000). The control group (group 1) consisted of 16 students (seven girls and nine boys). The case group (group 2) was composed of 14 students (10 girls and four boys).

### Measures

#### *Beck Anxiety Inventory (BAI)*

Each participant had their anxiety levels analyzed through BAI. The scale has 21 items that can verify the affective and cognitive aspects of anxiety somatically. Each item has four possible answers to the questions about anxiety, being 0 (absolutely not), 1 (slightly: did not bother me much), 2 (moderately: it was very unpleasant, but I could bear it), and 3 (severely: I could hardly bear it). In the end, the combined alternatives can range from 0 to 63, with 0 to 10 being the minimum level of anxiety; 11 to 19, light level; 20 to 30, moderate level and 31 to 63, severe/high level. The copyrights of the questionnaire are Beck, Epstein, Brown and Steer (1988). This questionnaire was translated and validated for use in Brazil (Cunha, 2001).

#### *Salivary Sample Collection for the quantification of cortisol*

The salivary sample was collected through

the spit in an Eppendorf, according to the expectoration technique. After drying the mouth, the individual should remain for three minutes without swallowing and, in the end, spits all saliva stored in the mouth in the collection tube. The samples were collected after the students' lunch break, respecting the indications of the literature (Jones et al., 2000).

Cortisol analysis was performed using the Elecsys Cortisol II kit from Roche Diagnostics GmbH, with the technique Electrochemiluminescence by competition. Its reference values are: morning (6 to 10 am) <0.783 ug / dL, late (16 to 20pm) <0.243 ug / dL, midnight <0.208 ug / dL (Roche Diagnóstica Brasil Ltda, 2015).

We performed the analysis following the kit manufacturer's guidelines, with a total duration of 18 minutes per test. 1st incubation: 10 µl of the sample incubated with a specific anti-cortisol biotinylated antibody and a ruthenium complex-labeled cortisol derivative. Depending on the concentration of the analyte in the sample and the formation of the respective immune complex, the binding site of the labeled antibody is occupied in part with the sample analyte and in part with the ruthenium-labeled hapten. 2nd incubation: after adding the streptavidin-coated microparticles, the complex that was formed binds to the solid phase through the interaction of biotin and streptavidin.

Reaction mixture aspirated into the reading cell, where the microparticles are magnetically attached to the surface of the electrode. We removed the unbound elements with ProCell / ProCell M. Applying an electric current to the electrode induces a chemiluminescent emission measured by a photomultiplier.

The results are determined based on a calibration curve explicitly generated by the analyzer through a 2-point calibration and on the main curve in the reagent bar code (Roche Diagnóstica Brasil Ltda, 2015). The technique was performed by the Hospital de Clínicas de Porto Alegre.

## Procedures

### *"K" Technique*

Even though anyone can conduct the technique, a trained psychologist managed the technique in this study. The "K" technique was an intervention designed specifically for students during school tests. "K" Technique is a result of mixed techniques that are generally in use to face anxiety in "cognitive challenge", such as breathing and relaxation techniques (Putwain & von der Embse 2021; Wernder-Seidler et al., 2017; Yusuf et al., 2019), cognitive restructuring (de Hullu et al., 2017; Garnefski & Kraaij, 2018; Grist et al., 2019; Putwain, & von der Embse, 2021; Yusuf et al., 2019).

In a classroom with open space, the participants were arranged in lines to avoid colliding. The application of all steps took around 20 minutes, guided by the psychologist. The explanation that the technique had nine stages (induction; perceptual position; search for the situation; where else the situation happens; internal and external tools; coping; new solution; a bridge to the future; return) and could help them to deal with the cognitive challenge that they would face was an important part. All the steps follow a logical sequence. First of all, helping the participants relax and create a safe place is essential to diminish the tension and offer comfort (Blackwell, 2021; Schwarz et al., 2020). In second, using several sensory parts of the body increases emotional learning and cognitive changes (Hoffmann, Brackett, Bailey, & Willner, 2020; Jones, McGarrah, & Kahn, 2019; Likhnik & Johansen, 2019; Smith, Killgore, Alkozei, & Lane, 2018). Third, memories of past experiences impact cognitive difficulties and their emotional reaction (Barch, Harms, Tillman, Hawkey, & Luby, 2019; Engen & Anderson, 2018; Tyng, Amin, Saad, & Malik, 2017). Knowing that searching for these memories is also an important step (Daros et al., 2021; Sloan et al., 2017). Fourth, it is essential to assist in searching and constructing strategies to deal with these challenges in the present and future (Daros et al., 2021; Sloan et al., 2017). Fifth, to consolidate the entire experience, visualize

a future moment where the students use these learnings is also fundamental (Saulsman, Ji, & McEvoy, 2019). Coming back again to a safe place is again recomforting and creates a state of less tension (Holmes, Arntz, & Smucker, 2007; Saulsman et al., 2019). Participants maintain their eyes closed during the intervention. All the "K" Technique steps are in Figure 1.

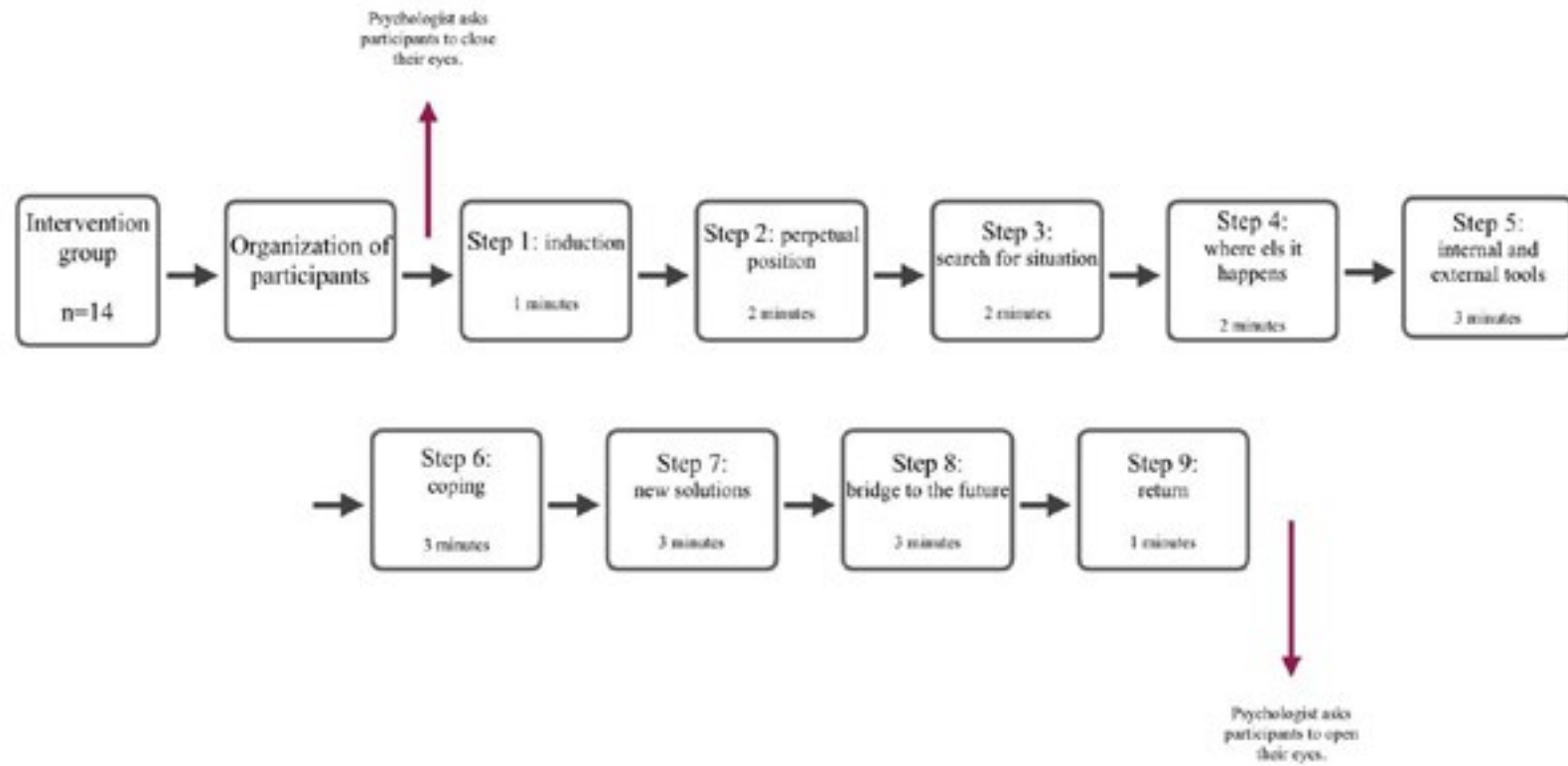


Figure 1 - Steps of the "K" Technique

## Experimental Design

This study design was a randomized, blind, controlled clinical trial. The randomization occurred in blocks. The subjects were listed in alphabetical order and, later, numbered. After the numbering of each participant, 15 participants for the control group and 15 for the case group were randomly assigned (Hulley, Cummings, Browner, Grady, & Newman, 2015).

Groups were separated, and no one knew about the technique or what it was proposing. They were guided by a qualified researcher on the filling out of the BAI to clear their doubts. The students collectively filled out the BAI, and directly after that, the first saliva sample took place for future salivary cortisol analysis. After the completion of both collections, we applied the "K" Technique. The students were standing with enough space to take a few steps forward. All activity took place with eyes closed for the best concentration with an average duration of 20 minutes. Students answered the questionnaire again, and the second sample of saliva was collected. Only two participants did not complete the cortisol sample exam.

The control group participants, separated from the case group so as not to have contact with the technique, were guided by a second trained researcher on the filling out of the BAI so that they could clear their doubts. The application of the BAI took place collectively, and directly after that, the first sample of saliva collection occurs. During the 20 minutes, they talked about future and professional choices, called placebo intervention. After the stipulated time, they filled out the questionnaire again, and the second saliva sample was collected.

All participants whose parents/guardians agreed and signed the Informed Consent Form. The local Ethics Committee approved the study (protocol number 16-0494).

## Data analysis

Parametric variables are presented as means  $\pm$  standard deviation, and non-parametric variables were shown in median and interquartile intervals

25 and 75%. For association tests, we used the Chi-square test and correlations using Spearman. For the comparisons between the before and after test scores, in other words, paired dependent results, we used the paired Wilcoxon test. The p values considered significant were less than 5%. To estimate the effect of the size, we computed the Hedge's g measure for the mean BAI scores of the two groups. The p values considered significant were less than 5%.

## Results

### BAI score in case and control group

Variation in BAI scores from before and after intervention in the case group median (interquartile range) (-10.50 (-18.25; -4.75)) was statistically higher than the values obtained in the control group (-6.00 (-8.75; -1.25)) (Figure 1). BAI scores in case group and control group presented statistically significant difference when pre intervention was compared to post intervention.

The Hedges g effect size computed for the differences between the case and control group's BAI scores was 0.5, which indicates a medium effect size according to the statistical literature (Lakens, 2014).

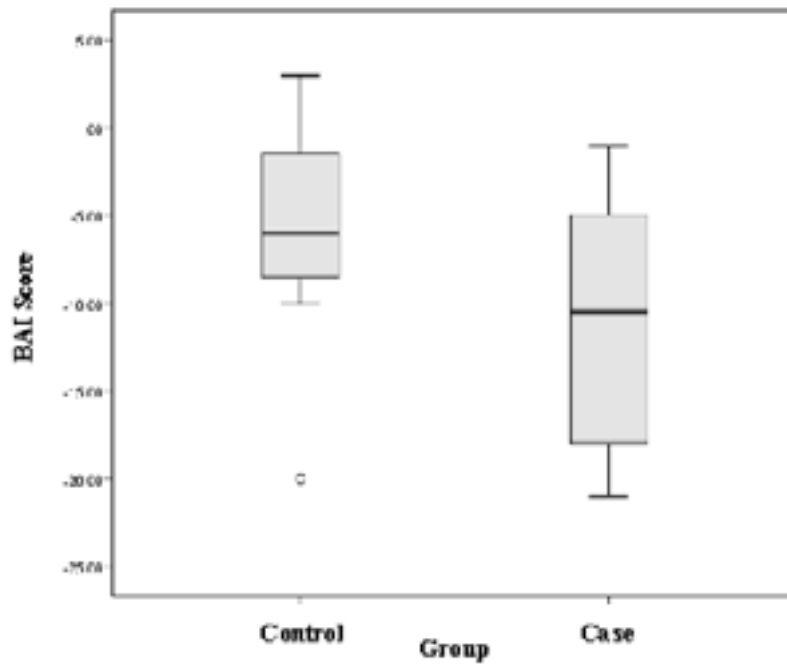


Figure 2 - The BAI (Beck Anxiety Inventory) scores before and after for the Case and Control Group,  $p = 0.025$

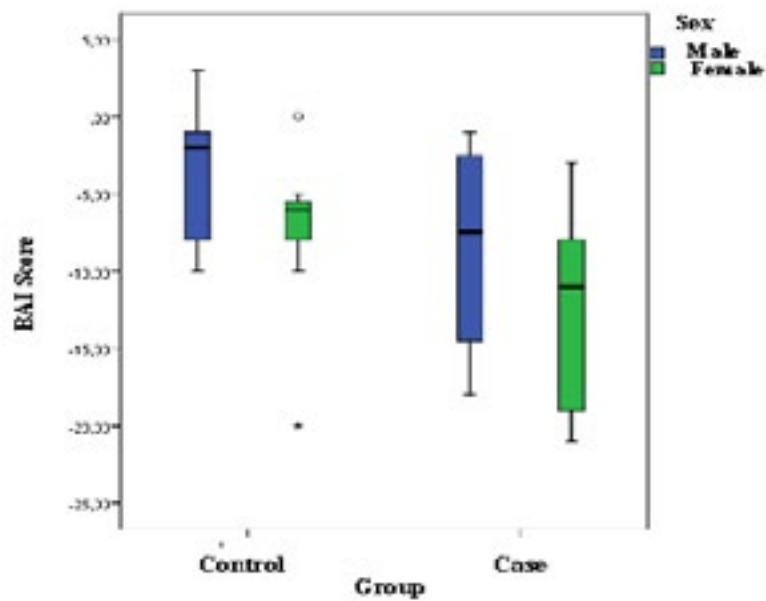


Figure 3 - The median variation of the BAI (Beck Anxiety Inventory) scores before and after for the Case and Control Group stratified by sex

### Salivary cortisol concentration in case and control group

Salivary cortisol levels in the Case group, comparing before, median (interquartile range), 0.28 (0.19-0.35), and after 0.18 (0.13-0.20), the application of the "K" technique, presented a statistically significant difference ( $p = 0.03$ ).

The levels of salivary cortisol in the control group, comparing previously, 0.29 (0.16-0.47), and after, 0.19 (0.15-0.24), the Placebo Intervention presented a statistically significant difference ( $p = 0.02$ ).

Comparing the variation of salivary cortisol levels in the case group, -0.1050 (-0.2075; -0.0400), with that of the control group, -0.0800 (-0.1800; -0.0250), it is not possible to identify statistical significance ( $p = 0.462$ ).

Variation in salivary cortisol levels was not significant when stratified by sex or grades.

### Discussion

The present study aimed to verify the effects of the "K" technique, based in breathing, relaxation techniques, psychoeducation and cognitive restructuring, in anxiety-related symptoms and cortisol levels on students facing school exams. We expected to observe lower levels of anxiety in the case group compared to the control one. We also expected lower cortisol levels in the case group compared to the control group. Considering the results, only the first hypothesis was corroborated through data.

In attempting to anticipate events, anxiety affects emotional, biological, and cognitive aspects that influence learning activities (Barker et al., 2018). One of the essential points in anxiety is the self-perception about symptoms and intensity (Hood et al., 2015; Moreno, Ávila-Souza, Gomes, & Gauer, 2015; Shi et al., 2019). As explained through Attentional Control Theory, dangerous stimuli influence attention control systems, leading to deficits in executive function and, by so, impairment in cognitive tasks (Silva, 2018; Shi et al., 2019). One of the measures chosen in the study (scores in Beck Anxiety Inventory) considers the affective, cognitive, and bodily self-perception of

anxiety (Cunha, 2001). The students participating in this study had, initially, a powerful perception of their anxiety. Insofar as the "K" technique was applied, it is possible that their perception of its symptoms reduced consistently. Such events suggest that the technique can be an useful tool to help participants manage anxiety and their involvement with emotions, cognitions, and physical sensations.

On the other hand, Salivary cortisol, which does not consider aspects of self-perception, is a well-known biological marker in studies on anxiety (Chojnowska, Ptaszyńska-Sarosiek, Kępka, Knaś, & Waszkiewicz, 2021; Joseph, Jiang, & Zilioli, 2021; Dhama et al., 2019; Roos et al., 2021). Still, it has several limitations regarding cortisol sensitivity during measuring, time of collection, the amount collected, other biomarkers that influence cortisol (Chojnowska et al., 2021; Dhama et al., 2019; Joseph et al., 2021; Roos et al., 2021). Anxiety-induced biological changes are known and studied (Barker et al., 2018). Although studies have shown a possible reduction in cortisol through techniques (Haugland et al., 2020; Manigault et al., 2018; Matousek et al., 2010), our results point to a possible limitation regarding this connection. In our study, we found a decrease in cortisol levels in the case and control groups. Because both variations are similar, their comparison was not statistically significant. Therefore, the "K" technique does not seem to specifically impact the reduction of salivary cortisol in students in our study. Another hypothesis is that the simple idea of doing something, the called placebo effect, may also have reduced cortisol in control.

The "K" technique seems to have a more impactful effect on symptoms, emotions, and cognitions than biological changes. Such results are similar to those found in studies on techniques that consider aspects of self-perception of anxiety, coping skills, motivation, and personality traits that demonstrate the importance of students managing their anxiety before exams and cognitive challenges at school and universities (de Hullu et al., 2017; Haugland et al., 2020; Putwain & von der Embse, 2021; Yusuf et al., 2019).



Thus, young people and adults can build more robust alternatives for cognitive, emotional, and behavioral coping with anxiety.

Despite similarities with the studies mentioned above, our research presents some points of difference that we consider essential. The studies mentioned present techniques such as breathing and relaxation techniques (Putwain & von der Embse, 2021; Werner-Seidler et al., 2017; Yusuf et al., 2019), cognitive restructuring (Garnefski & Kraaij, 2018; Grist et al., 2019; de Hullu et al., 2017; Putwain & von der Embse, 2021; Yusuf et al., 2019) used individually and not combined. Learning and changing psychological processes benefit from using of techniques that are more comprehensive about the individual's functioning (David, Cristea, & Hofmann, 2018; Feder, Fred-Torres, Southwick, & Charney, 2019; Phelps & Hofmann, 2019). Therefore, it is essential to combine techniques that encompass cognitions, emotions, and physical sensations in our case. With this in mind, we combined breathing/relaxation, cognitive restructuring techniques and added the experiential/experiential aspect (through changes in position and by keeping the eyes closed) to maximize gains. Experiencing/experiencing is part of the items listed as fundamental in the "3rd wave" of cognitive-behavioral therapy and has been widely studied, aiming at even more effective therapeutic gains (Garay, Korman, Keegan, 2015; Hayes & Hofmann, 2021).

Typically, in previous studies, protocols are based in five-weeks interventions (Burckhardt, Manicavasagar, Batterham, & Hadzi-Pavlovic, 2016; Caldwell et al., 2019, 2021; Feiss et al., 2019; Gee et al., 2020; Haugland et al., 2020; Johnstone et al., 2016; Werner-Seidler, Perry, Calear, Newby, & Christensen, 2017). Such duration can cause conflict with the school calendar and possible unforeseen circumstances in the study environment (Burckhardt et al., 2016; Caldwell et al., 2019, 2021; Feiss et al., 2019; Gee et al., 2020; Haugland et al., 2020; Johnstone et al., 2016; Werner-Seidler et al., 2017) and impair the learning and continuity of skills development through the protocols (Burckhardt et al., 2016; Caldwell et al., 2019, 2021;

Feiss et al., 2019; Gee et al., 2020; Haugland et al., 2020; Johnstone et al., 2016; Werner-Seidler et al., 2017). Furthermore, long protocols, such as the 5-week protocol, can result in fear of participation, avoidance of the study, and lack of commitment (Burckhardt et al., 2016; Caldwell et al., 2019, 2021; Feiss et al., 2019; Gee et al., 2020; Haugland et al., 2020; Johnstone et al., 2016; Werner-Seidler et al., 2017). Taking this into account, we opted for a single intervention, another difference in our study. Through a briefer intervention, it is possible to adapt to different school scenarios and favor student participation and, consequently, their gains from the intervention.

Another point that we believe to be crucial is the self-application of the learned technique to reinforce and improve gains. Although a long protocol presents significant results on management involving anxiety (Caldwell et al., 2021; Gee et al., 2020; Haugland et al., 2020), studies indicate that its self-application still becomes dependent on an instructor (Caldwell et al., 2021; Gee et al., 2020; Haugland et al., 2020). In our case, one of the objectives was to encourage students to apply the technique after their experience continuously by themselves, by reading the material written in the technique and mentally using the imaginary components to recall the experiences or using audio recorded with the technique.

## Conclusion

Despite potential results, our study has some important limitations. Although we have an encouraging result, this is only a preliminary study and deserves many adjustments until we can affirm the effectiveness of the "K" Technique effect. Some. For example, in the control group, the final anxiety scores measured through the BAI, comparing before and after the activity, were also considered statistically significant, suggesting that placebo intervention may have influenced the students by reducing the scores. Also, the procedure to collect cortisol material, spit in an Eppendorf, might be a stressor and affect salivary cortisol results so that future studies might choose another collection form. Due to the quick

variation of cortisol, it might be interesting for future surveys to choose the heart rate or breathing measurements for comparison. Another limitation of our study was not to consider academic results, social/demographic, and personality aspects. These are also essential aspects of understanding the impacts of these variables on anxiety and performance.

Nonetheless, these results point to a possible efficacy of the technique developed, which we call the "K" technique, in reducing anxiety symptoms when compared to control group. It is a brief technique, with potential to be spread in schools considering time of application and possibility of self-application when tutored. Future research that replicates these findings, expand them to other populations and that control other variables in regard to anxiety management in context of cognitive challenge in adolescence are desired and opportune.

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