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ORIGINAL RESEARCH

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Effects of the aroma of lemone verbena (*Aloysia citriodora* Paláu) essential oil on anxiety and the hemodynamic profile before cesarean section: A randomized clinical trial

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Abstract

Background: Aromatherapy is a noninvasive method used for alleviating anxiety. Lemon verbena (*Aloysia citriodora* Paláu, LV) has been frequently used in traditional medicine as an anxiolytic agent due to its pharmacological ingredients.

Objective: This randomized controlled trial aimed to assess the effects of inhaling essential oil of LV on the level of anxiety and subsequent hemodynamic changes before cesarean section.

Methods: The recent study was a randomized single-blind trial. Participants ($n = 84$) were randomly divided into two groups: LV essential oil (group A) and placebo (group B). The intervention group underwent aromatherapy using three drops of LV essential oil at a distance of 10 cm for 30 min. The placebo group received aromatherapy in a similar fashion. The State-Trait Anxiety Inventory of Spielberger questionnaire was administered before and 5 min after aroma inhalation. Vital signs were recorded before and after aromatherapy. Likewise, pain severity was assessed using the Numeric Rating Scale and vital signs were recorded. Data were analyzed using t-test, χ^2, and the Kolmogrov–Smirnov test through SPSS21 software.

Results: Anxiety level was significantly attenuated in group A after aromatherapy. Heart rate, respiratory rate, and blood pressure decreased after inhalation; but no significant variation of pain scores was observed after inhalation in both groups.

Conclusion: We concluded that LV decreased preoperative anxiety in this recent study, therefore, aromatherapy with LV essential oil as a preemptive adjuvant to relieve anxiety before cesarean section is recommended by us; although more studies are required to endorse the results.

KEYWORDS

Aloysia citriodora Paláu, anxiety, aromatherapy, cesarean delivery, essence, Spielberger (STAI-S)

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1 | BACKGROUND

Cesarean section (C/S) is one of the most common surgeries in the world.[1] It has been shown that the prevalence of C/S in Iran is 45% which is higher than the global average.[2,3] On the other hand, anxiety is a common problem before surgeries including C/S. Anxiety causes psychologic and physiologic effects both in mother and fetus.

High anxiety levels increase the need for anesthetics and require additional treatments to stop developing complications throughout the preoperative period. These physiologic changes can alter the duration of the surgical procedure.[4,5] To decrease complications, preoperative anxiety can be reduced by different medications, methods, and techniques.[6,7] It has been shown that pharmacologic agents such as benzodiazepines[8] as well as nonpharmacologic methods such as preoperative information, music, aromatherapy, and acupuncture can reduce preoperative anxiety.[9–12] Aromatherapy which uses essential oil derived from plants, is a complementary and alternative medicine.[13] There has been a lot of research on anxiolytic properties of *Lavandula angustifolia*, *Rosa damascene*, and *Osmanthus fragrans*.[14,15] Lemon verbena (LV) belongs to the lemon verbenaceae family so its leaves have lemon odor and flavor. This plant was initially cultivated in Central and South America and was transferred to Europe in the 17th century.[16–18] Moreover, it has received the approval of the American Food and Drug Administration.

LV is a safe plant[19] and its ingredients have antioxidant, analgesic, antimicrobial, and anti-inflammatory properties.[16,19–24] Likewise, the remarkable anxiolytic effect of LV has been reported by several studies.[25–27] But there have been no studies about the relationship between its anxiolytic effect and C/S. Given the fact that the inhalation method is safe and cost-effective, we decided to design this randomized controlled trial to assess the effects of inhaled essential oil of lemon verbenaceae on preoperative anxiety and hemodynamic changes before C/S. Hopefully, confirmation of this method will bring more comfort with decreased periods of recovery and hospitalization as well as reduced costs for patients and hospitals.

2 | MATERIALS & METHODS

2.1 | Trial design

This study was a randomized single-blind clinical trial that was registered in the Iranian Clinical Trials Registration Center (IRCT2016073116325N4). Its design was a parallel randomized clinical trial and the allocation ratio was usually 1:1.

2.2 | Participants and sampling methods

The study was performed on term pregnant women who were candidates for C/S. They were referred to Al-Zahra hospital, Rasht, Iran from September 2017 to September 2021. The essential oil aroma of LV was extracted by the Institute of Medicinal Plants, Alborz province, Karaj, Iran.

This essential aroma is made of phytochemical extracts of LV which has an excessive amount of phenolic substances such as phenylpropanoids, flavonoids, lignans, tannins, and a variety of other nonphenolic compounds.[28] For the placebo group (group P), distilled water was used. The chemical ingredients of the LV essential oil were explored using gas chromatography–mass spectrometry. The chemical composition of the essential oil was displayed with retention times, distribution constant (KI), and relative area percentage of compounds identified in the essential oil. A total of 29 compounds were detected within the essential oil of LV constituting 92.01% of it and α-Cadinol (44.26%) was the major compound detected in the essential oil (Table 1).

Inclusion criteria included 18 to 40-year-old term pregnant women who were candidates for nonemergency C/S, American Society of Anesthesiology Class I or II.

Exclusion criteria included acute or chronic respiratory failures (chronic obstructive pulmonary disease, asthma, bronchitis, seasonal allergy, etc.), smoking, cognitive disorders, any allergic reaction to LV, remarkable laboratory abnormalities, history of previous surgeries (affecting the preoperative anxiety), and history of using anxiolytics.

Four blocks were randomly put in sealed envelopes. Random sequences were generated using computer software. After generating the list, each person was assigned a unique code so that during the study, she could be identified with this code. None of the participants were aware of the randomization list. Registration and random allocation sequence was done by an educated nurse. Sealed envelopes were numbered to conceal the randomization process and each envelope was opened only after the eligibility criteria were approved and each individual had signed the consent form. Finally, subjects were allocated to either the intervention or the placebo group.

Based on the randomized grouping, all the subjects participating in the study were given pieces of neat cotton smeared with three drops of essential oil or distilled water. They were inhaled at a distance of 7–10 cm from the nose, 30 min before the surgery. Subsequently, these pieces of neat cotton were packed in opaque sealed cans by an educated nurse while patients were unaware of the type of intervention.

In the morning of the surgery, participants were asked to complete the State-Trait (Overt- Covert) Anxiety Inventory-State of Spielberger (STAI-S) questionnaire before and 30 min after the intervention. Vital signs including heart rate (HR) (bpm), respiratory rate (RR) (bpm), and blood pressure (BP) (mm Hg) were recorded during this time.

In our randomized study, the Persian version of the STAI-S questionnaire was used after measuring its validity and reliability.[29] In Iran, the validity of the Persian version of this questionnaire has been reported by Abdoli and colleagues.[30] Randomization flowchart is mentioned in Figure 1.

STAI-S questionnaire includes 20 items that measure the anxiety level. State and trait are subclasses of STAI. To determine anxiety,

TABLE 1 *Aloysia citriodora* essential oil composition (%).

GC–MS analysis					
Nu	**RT**	**%**		**KI**	**Type**
1	11.39	0.70	α-Pinene	935	MH
2	13.46	1.41	Sabinen	977	MH
3	14.03	0.45	1-Octen-3-ol	988	Other
4	14.32	3.53	6-Methyl-5-hepten-2-one	994	Other
5	16.46	15.03	Limonen	1035	MH
6	16.63	6.24	Eucalyptol	1038	MO
7	19.74	0.28	Rosefuran	1098	MO
8	20.19	0.37	Linalool	1107	MO
9	25.17	2.02	α-Terpineol	1209	MO
10	26.39	0.78	Nerol	1234	MO
11	27.21	7.24	Z-Citral	1252	MO
12	27.63	1.16	Carvone	1261	MO
13	28.06	0.30	Piperitone	1270	MO
14	28.63	8.28	Geranial	1282	MO
15	33.14	1.66	α-Copaene	1382	SH
16	33.31	1.10	Geranyl acetate	1386	MO
17	33.51	1.28	β-Bourbonene	1390	SH
18	35.10	2.94	Caryophyllen	1428	SH
19	36.85	1.03	Alloaromadendrene	1470	SH
20	37.16	0.68	Geranyl propionate	1477	SO
21	37.44	0.32	γ-Muurolene	1484	SH
22	37.69	9.48	α -Curcumene	1490	SH
23	39.07	0.36	γ-Cdinene	1524	SH
24	39.19	1.54	δ-Cadinene	1527	SH
25	40.87	1.77	Nerolidol	1569	SO
26	41.80	6.38	β -Spathulenol	1593	SO
27	41.99	12.83	Caryophyllen oxide	1598	SO
28	43.07	1.37	α-Humulene epoxide II	1627	SO
29	44.26	1.49	α-Cadinol	1658	SO
		92.01	Totsl identified		

Abbreviations: GC–MS, gas chromatography–mass spectrometry; KI, distribution constant; MH, monoterpene Hydrocarbons; MO, oxygenated monoterpene; RT, retention time; SH, sesquiterpene hydrocarbons; SO, sesquiterpene oxygenated.

20 questions per subscale consisting of 10 negative and 10 positive points were used based on STAI-S of Spielberger. Each question was scored from one to four. A score of one means anxiety has never been experienced and conversely, a score of four means a high level of anxiety. The minimum and maximum scores of anxiety levels are 20 and 80, respectively. These scores are computed by adding a standard number to distinguish between positive and negative scores. Scores of 21–40, 41–60, and 61–80 are supposed to be mild, moderate, and severe anxiety, respectively.[29,30]

Afterward, all patients in both groups were prepared for C/S and all were given spinal anesthesia with Bupivacaine (13.5 mg/0.75%) and Fentanyl (25 μg). They were not given Midazolam to decrease anxiety. The primary outcome of the present study was a decrease in the mean score of anxiety and an improved hemodynamic profile before C/S.

2.3 | Statistical analysis

The sample size was found by G-power statistical software. The mean difference between the two groups was considered to be one. For each group, 42 samples were allocated based on 80% power and 0.05 error. The data were analyzed by SPSS software version 21. A *t*-test was used to compare the groups after examining the normal distribution of quantitative data with the Kolmogorov–Smirnov test. χ^2 tests were used to evaluate the qualitative variables and paired *t*-test was used to compare before and after the test. $p < 0.05$ were considered significant.

Ethical considerations: The study was approved by the ethics committee of Guilan University of Medical Sciences (IR. GUMS. REC.1395.104). All stages of this research have been performed according to the principles of the Helsinki declaration. All procedures of the study were explained clearly to those participants who had the eligible inclusion criteria. Moreover, all participants filled out the written informed consent form before joining the study voluntarily and were free to decide to attend or withdraw at any time, for any reason without changing their medical care.

3 | RESULTS

In the present study, 84 cases participated in two groups.

Of the 117 patients screened for eligibility, 33 patients were excluded from the study (Not inclusion passed criteria [$n = 21$], declined to join into the study [$n = 8$] and un forseen exclusion [$n = 4$]). The remaining 84 cases allocated to one of the two groups randomaly, then basline data were compared. There were no statistical differences between the groups in terms of maternal age, gravidity, pariety, and body mass index.

The percetage of LV essentional oil is showed in Table 1 and characteristic of two groups is mentioned in Table 2.

There were significance statistical differences between the groups in terms of gestational age, neonatal score, and birth weight (<0.001, 0.036 respectively), but this difference was very partial and there was no clinical significant difference in term of these three items.

A: There was not significant statistical difference in term of pain score before and after intervention ($p = 0.29$).

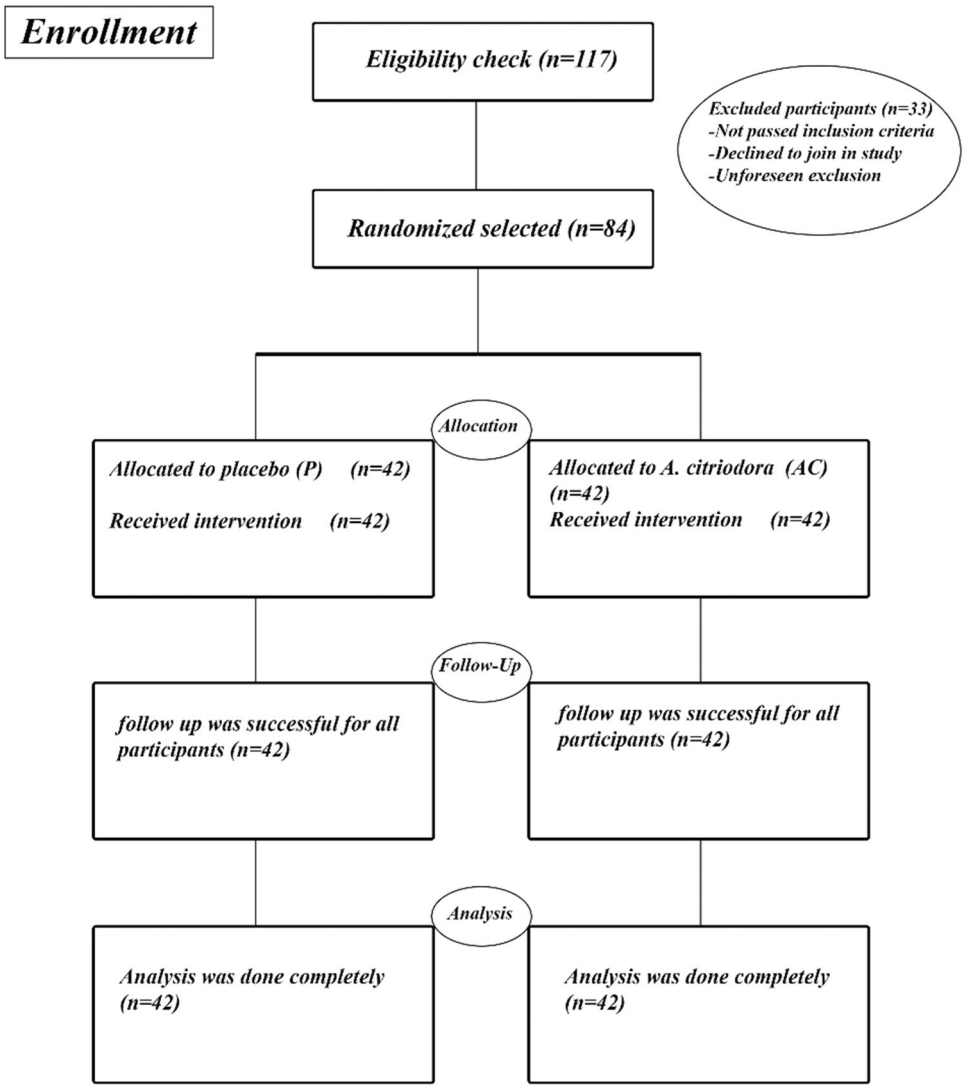

FIGURE 1 Study flowchart.

TABLE 2 Characteristic of two groups.

Group variable	LV mean ± SD	P mean ± SD	*p* Value[a]
Maternal age (y)	30.23 ± 0.61	30.19 ± 0.74	0.96
BMI (kg/m^2)	22.79 ± 3.11	23.13 ± 2.63	0.72
Gravidity (*n*)	2.84 ± 1.48	2.46 ± 1.30	0.28
Pariety (*n*)	1.41 ± 1.09	1.00 ± 0.89	0.131
Gestational age (week)	39.06 ± 1.04	38.57 ± 0.96	<0.001
Birth weight	3377.91 ± 386.5	3294.14 ± 357.2	0.036
APGAR score	9.3 ± 1.7	8.3 ± 2.6	<0.001

Note: Using *t*-test. There was no significant statistically difference in the maternal age between two groups. $p = 0.96$.

Abbreviation: Apgar, Appearance/Pulse/Grimace/Activity/Respiration; BMI, body mass index.

[a]Unpaired *t*-test.

B: There was decreased systolic BP after intervention in LV group ($p = 0.0001$), and there was not statistical significant difference before and after in Placebo group ($p = 0.85$).

Diastolic BP in LV group had significant statistical difference before and after intervention ($p = 0.001$), but this item had no significant statistical difference in placebo group before and after that ($p = 0.91$).

In LV group, RR had significant statistical difference before and after intervention ($p = 0.001$), but there was not significant statistical difference before and after intervention in placebo group ($p = 0.05$).

In LV group, HR had significant statistical difference before and after intervention ($p = 0.001$), but there was not significant statistical difference before and after intervention in placebo group ($p = 0.31$).

Pain score had not significant statistical difference before and after intervention in both two groups (LV $p = 0.14$, Placebo $p = 0.35$).

In LV group, covert anxiety decreased after intervention and there was significant statistical difference before and after

TABLE 3 Comparison of the variables between two groups.

Variable	Groups (*N* = 84)	BI (Mean ± SD)	AI (Mean ± SD)	*p* value intra-group[a]	*p* Value[a]
1. State anxiety	LV	52.07 ± 0.09	39.92 ± 0.34	0.0001	0.0001
	P	47.33 ± 0.21	44.97 ± 0.5	0.002	
P-Value-Inter group[b]		0.004	0.0001		
2. Trait anxiety	LV	51.4 ± 5.92	40.28 ± 0.01	0.0001	0.005
	P	47.09 ± 0.22	47.73 ± 0.03	0.49	
P-Value-Inter group[b]		0.007	0.0001		
3. Pain score	LV	1.88 ± 0.59	1.64 ± 0.07	0.14	0.85
	P	2.32 ± 0.11	2.11 ± 0.64	0.35	
P-Value-Inter group[b]		0.29	0.12		
4. Heart rate	LV	80.85 ± 0.59	72.16 ± 0.06	0.0001	0.0001
	P	86.95 ± 0.57	88.19 ± 0.73	0.31	
P-Value-Inter group[b]		$p = 0.08$	0.0001		

Abbreviation: AI, after Intervention; BI, before Intervention; LV, lemon verbena.

[a]*t*-test.

[b]Paired *t*-test.

intervention (0.001), but there was not significant statistical difference in placebo group before and after intervention ($p = 0.49$).

There was statistical significance difference before and after intervention between the groups in term of overt anxiety (LV $p = 0.0001$, placebo $p = 0.002$) (Table 3).

4 | DISCUSSION

Anxiety is an annoying and unpleasant feeling which must be controlled or even avoided.[31] It has been shown that persistent postsurgical anxiety leads to prolonged recovery period, physiologic changes and deficiencies as well as sleep disorders.[32] Aromatherapy has already been used as a noninvasive intervention to restrict anxiety. It acts through passing fragrance molecules from olfactory receptors which are transmitted to the limbic portion of the brain.[33] One of the most important aromatic compounds is LV. The recent study has been designed as a clinical trial to determine the effect of this essence on the level of pre-C/S anxiety.

The latest trials have focused on the antianxiety effects of an edible extract of LV in animal models.[34] The results of an animal model study conducted by Razavi et al. has demonstrated that intraperitoneal ethanolic and aqueous extracts of LV leaves have antianxiety effects which are probably due to the interaction between GABA receptors and LV ingredients.[35] We obtained similar results in our study but with a difference in the method of usage in which we used the fragrance type of LV instead of the intraperitoneal method. Another difference was that our subjects were humans.

Igarashi T. designed a study on pregnant subjects and concluded that aromatherapy with the essential oil of Linalyl acetate content, improved postsurgical tension anxiety because of its relaxing effects.[36] The results were the same as ours but the type of fragrance was different between the two studies.

Razavi et al. investigated about the effect of intraperitoneal LV on mice and concluded that it has a hypnotic, anxiolytic and muscle relaxant effect.[35] Their results were the same as ours but they used the intraperitoneal method and selected different subjects (mice).

Jaruzel and colleagues evaluated lavender skin patch on women who went through breast surgery. They concluded that this type of remedy can decrease postsurgical anxiety.[32] The results of their evaluation were the same as ours.

Bahramsoltani and colleagues in a review demonstrated the anxiolytic activity of LV. Their conclusion was the same as ours.[20]

Dagli R. et al. evaluated the preoperative antianxiety effects of rose oil inhaler on rhinoplasty and concluded that this method reduced preoperative anxiety.[15] Their results were the same as ours but the agent was different.

Bonyani A. and colleagues evaluated the leaf extract essence of LV on mouse models in terms of its sedative and anxiolytic activity. They concluded that the essence of the leaf acts as an antianxiety agent like diazepam through the GABAergic system without any significant reduction of locomotor activity.[34] The results were the same as ours with the exception that we performed the study on humans instead of mice.

Bahramsoltani R. et al. studied the published pharmacologic properties of LV and concluded that LV has not been fully investigated regarding its safety and effects on humans. They presented perfect outcomes and asked research to acknowledge the remedial benefits of this plant.[20] Their results are the same as ours.

Motti R. et al. suggested that LV could be consumed as an alternate therapy for anxiety. They claimed that obviously more studies are required to affirm the results.[37] These results are the same as ours.

Following a study on antioxidant properties of LV, Catula N. et al. concluded that LV could be consumed as a supplement to reduce anxiety but they believed more studies are needed to confirm that.[38] Their results were the same as ours.

Martinez-Rodriguez A. and Martinez-Olcina M. found out that the extract of LV could improve anxiety and sleep quality.[38] The results were the same as ours.

Ebrahimi M. et al. studied on Iranian herbal therapeutic plants and concluded that *Melissa Officinalis* which is an endemic of Iran has antianxiety properties. Moreover, *Avena Sativa* can be consumed as part of an antianxiety remedy. *Passiflora Incarnata* has antianxiety properties. Flavonoid Krayzn is an extract of *Passiflora Incarnata* that has antianxiety effects. *Valeriana Officinalis*, *Satureja hortensis* L and Menta Piperita all have antianxiety activities. *Crocus Sativus* has a golden color, a pleasant odor, and beneficial antianxiety effects.[39] These plants act like LV to reduce anxiety.

Agatonovic-Kustrin S. et al. concluded that terpenoids and aromatherapy have antianxiety properties.[40] We reached the same conclusion with LV.

It seems that clinical aromatherapy of essential oil of LV has not been fully studied in relation with preoperative anxiety and there are very few articles on this topic. In summary, the aroma of LV seems to reduce preoperative anxiety.

Regarding its safety to use, cost efficiency, and comfort, LV can be consumed as an adjuvant medication before C/S but more studies are needed to confirm the results.

5 | CONCLUSION

The results of this recent study illustrated that inhaling the aroma of LV reduces preoperative anxiety, breathing rate, pulse rate as well as systolic and diastolic BP before C/S. As LV is safe, cheap, and simple to use, its inhalation can be recommended as an adjunct medication to alleviate anxiety before C/S. But, it seems more clinical trials are required to confirm our findings. This research suggests that LV oil could be used as an adjuvant remedy before operations because not only it reduces preoperative anxiety scores but also is cost-effective and easy to use.

AUTHOR CONTRIBUTIONS

Katayoun Haryalchi: Writing—original draft. **Soudabeh Kazemi Aski**: Conceptualization. **Mandana Mansour Ghanaie**: Writing—review and editing. **Masoumeh Fotouhi**: Data curation; writing—original draft. **Roghayeh Mansoori**: Data curation; writing—review and editing. **Seyed Mahdi Sadraei**: Data curation; writing—review and editing. **Yasaman Yaghobi**: Conceptualization; methodology; writing—review and editing. **Sepehr Olangian-Tehrani**: Formal analysis.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

Related data of this project are available on request. Supporting data are available in Reproductive Health Research Center, Department of Obstetrics & Gynecology, Al-zahra Hospital, School of Medicine, Guilan University of Medical Sciences, Rasht, Iran. In this study, all the data presented will be available on request from the corresponding author by this journal's representative at any time during submission or after publication.

ETHICS STATEMENT

Ethical approval was obtained from the Ethics Committee of the Vice-Chancellery for Research at Guilan University of Medical Sciences (Code: IR. GUMS. REC.1395.104) and was registered at the Iranian Registry of Clinical Trials with code number: IRCT2016073116325N4. Informed consent was obtained from all participants before commencing the study.

DATA REPRODUCIBILITY

In this study, all the data presented will be available on request from the corresponding author by this journal's representative at any time during submission or after publication.

TRANSPARENCY STATEMENT

The lead author Mandana Mansour Ghanaie affirms that this manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned (and, if relevant, registered) have been explained.

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