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Alternative treatment strategies for bacterial vaginosis: the role of lactic acid in combating antibiotic resistance

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ABSTRACT

BACKGROUND: Bacterial vaginosis is a common infectious non-inflammatory vaginal condition that increases susceptibility to sexually transmitted diseases, negatively impacts perinatal outcomes, and reduces overall quality of life. Considering the low long-term effectiveness of antibiotic therapy, the high recurrence rates, and the frequent side effects associated with its use, there is a growing need to explore alternative approaches for bacterial vaginosis treatment.

AIM: To evaluate the efficacy and tolerability of a two-step treatment approach for bacterial vaginosis, which includes clindamycin or dequalinium chloride and lactic acid, in women of reproductive age.

MATERIALS AND METHODS: An open-label randomized clinical trial was conducted, including 93 women aged 18–45 years diagnosed with bacterial vaginosis according to Amsel's criteria. Participants were randomly assigned to three groups: 31 women in the first group received lactic acid monotherapy, 31 in the second group received a combination of clindamycin and lactic acid, and 31 in the third group were treated with dequalinium chloride and lactic acid. Treatment efficacy was assessed after 14 days using Amsel's criteria. Three months post-treatment, patient-reported symptoms and vaginal pH levels were evaluated.

RESULTS: Two weeks post-treatment, bacterial vaginosis symptoms persisted in 3 (9.7%) patients from the first group and in 1 (3.2%) patient from the second group. A positive trend in vaginal pH normalization was observed in all groups both at the two-week and three-month follow-ups. At the three-month follow-up, vaginal discharge complaints persisted in one patient from the first group, one from the second, and two from the third. The efficacy of lactic acid monotherapy at day 14 was 90.3%, increasing to 96.4% at three months. The two-step therapy combining clindamycin and lactic acid demonstrated an efficacy of 96.8% and 96.7%, respectively. The two-step therapy with dequalinium chloride and lactic acid demonstrated 100% efficacy after 14 days and 93.3% at the three-month follow-up. A case of vulvovaginal candidiasis was reported in the second group three months after treatment.

CONCLUSION: This study demonstrated the sustained high efficacy of the two-step treatment approach in both short-term and long-term perspectives. Whereas monotherapy initially showed lower efficacy, its long-term outcomes became comparable, highlighting the importance of lactic acid in combination therapy.

Keywords: bacterial vaginosis; lactic acid; clindamycin; dequalinium chloride; two-step treatment.

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Альтернативные стратегии лечения бактериального вагиноза, включающие использование молочной кислоты, как потенциальное решение проблемы антибиотикорезистентности

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АННОТАЦИЯ

Обоснование. Бактериальный вагиноз является распространённым инфекционным невоспалительным заболеванием влагалища, которое повышает вероятность заражения инфекциями, передающимися половым путём, оказывая негативное влияние на перинатальные исходы и снижая качество жизни в целом. С учётом низкой эффективности антибактериальной терапии в долгосрочной перспективе, а также высокой частоты рецидивов и побочных эффектов, связанных с применением антибиотиков, возникает необходимость в поиске альтернативных подходов к лечению бактериального вагиноза.

Цель. Оценить эффективность и переносимость комплексного двухэтапного метода лечения бактериального вагиноза, включающего клиндамицин или деквалиния хлорид и молочную кислоту, у женщин репродуктивного возраста.

Материалы и методы. Проведено открытое рандомизированное клиническое исследование, в котором приняли участие 93 женщины 18–45 лет с диагнозом «бактериальный вагиноз», подтверждённым по критериям Амселя. Пациентки были случайным образом распределены на три группы: у 31 женщины из первой группы использовали молочную кислоту; у 31 женщины из второй группы — комбинацию клиндамицина и молочной кислоты; у 31 женщины из третьей группы — деквалиния хлорид с молочной кислотой. Эффективность лечения оценивали через 14 дней с использованием критериев Амселя. Спустя три месяца после завершения лечения оценивали жалобы и измеряли pH влагалищного отделяемого.

Результаты. В ходе исследования отмечено, что через две недели после завершения лечения симптомы бактериального вагиноза продолжали сохраняться у 3 (9,7%) пациенток в первой группе, у 1 (3,2%) — во второй группе. Положительная динамика изменения pH вагинального отделяемого наблюдалась во всех группах как через две недели, так и через три месяца после окончания лечения. Спустя три месяца после окончания лечения жалобы на выделения из половых путей сохранились у одной женщины из первой группы, у одной — из второй, у двух — из третьей. Эффективность лечения с использованием молочной кислоты на 14-й день составила 90,3%, через три месяца — 96,4%. Двухэтапная терапия с применением клиндамицина и молочной кислоты показала эффективность 96,8% и 96,7% соответственно. Лечение с использованием деквалиния хлорида и молочной кислоты продемонстрировало 100,0% эффективность через 14 дней и 93,3% через три месяца. Во второй группе был зафиксирован случай вульвовагинального кандидоза через три месяца.

Заключение. Проведённое исследование подтвердило высокую эффективность двухэтапного метода лечения как в краткосрочной, так и в долгосрочной перспективе. Несмотря на то что монотерапия изначально показала меньшую эффективность, её результаты в дальнейшем стали сравнимыми, что подчёркивает важность использования молочной кислоты в комплексной терапии.

Ключевые слова: бактериальный вагиноз; молочная кислота; клиндамицин; деквалиния хлорид; двухэтапное лечение.

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包括乳酸在内的细菌性阴道病替代治疗策略：抗生素耐药性问题的潜在解决方案

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摘要

背景。细菌性阴道病（BV, Bacterial Vaginosis）是一种常见的非炎症性阴道感染，可增加性传播感染的风险，对围产期结局产生不良影响，并降低整体生活质量。由于抗菌治疗在长期管理中的效果有限，并且抗生素治疗相关的复发率和不良反应较高，因此迫切需要寻找BV的替代治疗方法。

目的。 评估采用克林霉素或地喹氯铵联合乳酸的两阶段综合治疗方案在育龄期女性中的疗效和耐受性。

材料与方法。本研究为一项开放性随机临床试验，共纳入93名18-45岁、符合Amsel标准确诊的BV女性患者。患者被随机分为三组：第一组（n=31）接受乳酸治疗；第二组（n=31）接受克林霉素联合乳酸治疗；第三组（n=31）接受地喹氯铵联合乳酸治疗。在治疗结束后14天，依据Amsel标准评估治疗效果。在治疗后三个月评估患者症状并测量阴道分泌物pH值。

结果。研究显示，在治疗结束两周后，细菌性阴道病症状仍持续存在于第一组3例（9.7%）患者，第二组1例（3.2%）患者。阴道分泌物pH值在所有研究组中均呈现积极变化，且该变化在治疗结束两周及三个月后仍保持稳定。三个月后，第一组1例患者、第二组1例患者、第三组2例患者仍报告阴道分泌物异常。乳酸单一治疗的有效率在治疗后14天为90.3%，三个月后为96.4%。克林霉素联合乳酸的两阶段治疗有效率分别为96.8%（14天）和96.7%（三个月）。地喹氯铵联合乳酸治疗的有效率在14天时为100.0%，三个月后为93.3%。此外，在第二组随访中发现1例治疗后三个月出现外阴阴道念珠菌病的病例。

结论。本研究证实了两阶段治疗方案在短期及长期均具有较高的疗效。尽管乳酸单一治疗的初始效果较低，但其远期疗效与其他方案相当，突出了乳酸在细菌性阴道病综合治疗中的重要性。

关键词：细菌性阴道病；乳酸；克林霉素；地喹氯铵；两阶段治疗。

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BACKGROUND

Bacterial vaginosis (BV) is a clinical and laboratory non-inflammatory condition of the vagina in which the dominant lactobacilli lose their predominant position in the microbiome. In women of reproductive age, the prevalence of BV ranges from 23% to 29%, exhibiting variations based on ethnicity and geographical location. For instance, in North America, 33% of non-Hispanic black women and 31% of Hispanic women are affected by BV, whereas the rates for Asian and Caucasian women are 11% and 23%, respectively [1].

BV contributes to an increased likelihood of contracting sexually transmitted infections (STIs) such as human immunodeficiency virus, herpes simplex virus, chlamydia, gonorrhea, trichomoniasis, and mycoplasma infection and increases the risk of pelvic inflammatory disease [2]. In addition, this condition is associated with adverse pregnancy and fetal outcomes, including intrauterine infections, premature rupture of the fetal membranes, late spontaneous miscarriage, preterm labor, and postpartum and postabortion purulent-septic complications [3, 4]. Currently, the first-line treatment of BV is antibacterial medications. These include oral metronidazole 500 mg twice daily for seven days, intravaginal metronidazole 5 g once daily for five days, and intravaginal clindamycin 5 g once daily for seven days [5]. However, BV recurrence after standard antibiotic therapy occurs in 43% of women within 3 months and in 58% within 12 months. This may be due to resistance to antimicrobial therapy, biofilm formation, subsequent persistence of microorganisms, and reinfection [6]. The most promising alternative method for the treatment and prevention of BV is the use of the antiseptic dequalinium chloride and lactic acid, an agent that lowers the vaginal pH.

Aim

The study aimed to evaluate the efficacy and tolerability of a comprehensive two-step treatment regimen for BV, including clindamycin or dequalinium chloride and lactic acid, in women of reproductive age.

METHODS

An open-label, randomized, clinical trial was conducted involving 93 women aged 18–45 years with BV diagnosed according to the Amsel criteria. The study was conducted as part of A.D. Minakova's dissertation research and approved by the Local Ethics Committee of the Sechenov First Moscow State Medical University (Protocol No. 01-22 dated January 20, 2022). After obtaining voluntary informed consent to participate in the study, the women were randomly divided into three groups. Group 1 (n = 31) was treated vaginally with 100 mg of lactic acid for ten days. Group 2 (n = 31) received 100 mg of vaginal clindamycin for three days, followed by 100 mg of lactic acid for ten days. Group 3 (n = 31) received 10 mg of vaginal dequalinium chloride for six days, followed by 100 mg of lactic acid for ten days.

Inclusion criteria: complaints of malodorous genital discharge and patient age between 18 and 45 years. Exclusion criteria: pregnant and lactating women; acute or chronic active pelvic inflammatory disease; STIs (infections caused by *N. gonorrhoeae*, *C. trachomatis*, *T. vaginalis*, and *M. genitalium*), vulvovaginal candidiasis, aerobic vaginitis; and menopause before the age of 45 years.

Upon enrollment, patients underwent a gynecological examination. Vaginal pH was measured using Kolpo-Test pH indicator strips (Russia), and vaginal secretions were collected from the upper third of the lateral vaginal vault for Gram-staining microscopy. STI screening by polymerase chain reaction was performed if indicated (less than 25 years of age, new sexual partner in the past 12 weeks, two or more sexual partners, sexual contacts without barrier contraception, history of STIs in the woman or her partner, and procedures that interrupted the cervical barrier in the past six weeks).

The patients were followed up for three months. Two weeks after the end of treatment, complaints were assessed; a gynecological examination was performed; the pH of the vaginal contents was measured, and vaginal fluid was collected from the upper third of the lateral vaginal vault for Gram-staining microscopy. Three months after the end of treatment, the women who complained of genital discharge and/or odor underwent were re-examined; vaginal pH was measured, and Gram microscopy was performed. The evaluation of treatment efficacy was based on the following parameters: the absence of a homogeneous, malodorous, white-gray discharge; a pH of the vaginal fluid below 4.5, and the absence of clue cells on microscopy.

The analysis of the study's results was conducted by employing Python statistical libraries, namely Numpy and Pandas.

RESULTS

The mean age of the women was 29.70 ± 2.70 years in group 1, 31.40 ± 2.71 years in group 2, and 33.50 ± 2.53 years in group 3. Group 1 did not include any smokers, whereas 3.2% (n = 1) of the women in group 2 used tobacco. This number increased significantly to 32.3% (n = 10) in group 3.

The results showed that no infections were detected in any of the examined women: 38.7% (12/31) in group 1, 29.0% (9/31) in group 2, and 38.7% (12/31) in group 3.

There were no significant differences between the groups in the presence of somatic diseases. The analysis of the history of urinary tract infections in women in each group showed that chronic cystitis was diagnosed in 19.4% (n = 6) of the women in group 1, 9.7% (n = 3) of those in group 2, and 3.2% (n = 1) of those in group 3. Postcoital cystitis was identified in 3.2% (n = 1) of the participants in group 2. In group 3, chronic pyelonephritis was diagnosed in 12.9% of the patients (n = 4). These findings are consistent with prior reports of statistically significant associations between urinary tract infections and BV.

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Most women chose barrier methods of contraception, including 28 (90.3%) participants in group 1, 22 (71.0%) in group 2, and 23 (74.2%) in group 3. Combined oral contraceptives were used by one (3.2%) woman in group 2 and the copper intrauterine device by one (3.2%) woman in group 3. Interrupted coitus as a contraceptive method was used in all groups, including three (9.7%) women in group 1, eight (25.8%) in group 2, and six (19.4%) in group 3. In addition, one (3.2%) woman in group 3 did not use any contraception because she was planning a pregnancy.

A preliminary examination with a Cusco speculum revealed that patients in all three groups exhibited substantial amounts of grayish-white vaginal discharge accompanied by an amine malodor.

The mean pH of the vaginal discharge before treatment was 5.3 ± 0.1 in group 1, 5.6 ± 0.2 in group 2 and 5.5 ± 0.2 in group 3.

In all three groups, clue cells were found in 32.3% (*n* = 10), 54.8% (*n* = 17) and 48.4% (*n* = 15) of the women, respectively.

Two weeks after the end of treatment, all participants in group 3 had mucus discharge without a characteristic odor. Three patients (9.7%) in group 1 and one patient (3.2%) in group 2 continued to have odorous vaginal discharge, although in smaller amounts.

The mean pH of vaginal discharge 14 days after therapy in patients of all three groups was 4.4 ± 0.1, 4.4 ± 0.1 and 4.5 ± 0.1, respectively.

Two weeks after treatment, clue cells were detected in only 3.2% (*n* = 1) of the women in group 1, whereas none were detected in other participants.

As a result of clinical and laboratory examination using Amsel criteria, three patients in group 1 and one patient in group 2 were rediagnosed with BV, indicating the lack of treatment efficacy, and the patients were therefore excluded from further follow-up.

When lactic acid was used as a second step of treatment, one patient in group 2 and one in group 3 experienced adverse effects in the form of vaginal burning. One woman in group 3 stopped taking the product after the fifth suppository and withdrew from the study. After discontinuing the lactic acid, this discomfort resolved. The participant in group 2 continued to use the product despite the burning sensation, which ceased after the end of therapy. The group that received lactic acid alone did not exhibit any adverse effects.

Three months after therapy, one (3.6%) woman in group 1, two (6.7%) in group 2, and two (6.7%) in group 3 complained of genital discharge.

The mean pH of the vaginal discharge was 4.3 ± 0.1, 4.5 ± 0.1, and 4.5 ± 0.1 in all three groups, respectively.

Gram microscopy was performed to detect clue cells three months after treatment in cases where patients complained of discharge and/or odor from the genital tract. Clue cells were detected in 3.3% (*n* = 1) of the participants in group 2 and in an equal proportion (*n* = 1) in group 3. Conversely, no such cells were identified in group 1.

Three months after the end of treatment, recurrence of BV (3 or 4 positive Amsel criteria) was diagnosed. One case (3.6%) was recorded in group 1, one case (3.3%) in group 2, and two cases (6.7%) in group 3. In addition, one case of vulvovaginal candidiasis (3.3%) was recorded in group 2 after three months.

The efficacy of lactic acid treatment after 14 days was 90.3% (28/31). After three months, this figure increased to 96.4% (27/28), thereby indicating the durability of achieved results. A two-step treatment regimen, including clindamycin and lactic acid, demonstrated 96.8% (30/31) efficacy after two weeks and 96.7% (29/30) efficacy after three months. Treatment with dequalinium chloride and lactic acid showed 100.0% (30/30) efficacy after 14 days and 93.3% (28/30) efficacy after three months.

DISCUSSION

The problems associated with the use of standard antibiotic therapy in the treatment of BV, such as low long-term efficacy and high recurrence rates, as well as adverse side effects of antibiotics, emphasize the need to find alternative therapies. Lactic acid produced by lactobacilli has been shown to help maintain an acidic pH in the vagina, creating favorable conditions for lactobacilli growth and reproduction while preventing the development of anaerobic microorganisms [7]. A study by O'Hanlon et al. [8] demonstrated that physiological concentrations of lactic acid under anaerobic conditions inactivated BV-associated bacteria without affecting lactobacilli.

A study by Kira et al. [9] included 116 women diagnosed with BV according to the Amsel criteria. The aim of the study was to evaluate the efficacy and safety of vaginal suppositories containing lactic acid on days 2–3 and 32 after the end of therapy. The participants were divided into two groups: 64 women received lactic acid and 52 received placebo. The results showed a significant decrease in the incidence of pathological discharge (from 100.00% to 4.69% and 10.94%), pH (from 6.84 to 4.28 and 4.34), and the number of clue cells (from 100.00% to 7.81% and 10.94%) in the group receiving lactic acid. The absence of adverse effects suggests the safety of this method.

Another study by Ross et al. [10] found that metronidazole was more effective than lactic acid-based gel in women with recurrent BV (70% vs. 47% at day 14). After six months, however, recurrence rates were similar (71% vs. 70%), raising concerns about the long-term benefits of metronidazole. Nevertheless, many patients preferred the lactic acid gel, probably because of fewer adverse effects.

A study conducted by Paavonen et al. [11] found that oral administration of metronidazole showed similar efficacy to intravaginal clindamycin, but with lower tolerability among patients.

Thus, the results of this study are consistent with the data presented by Kira et al. [9] and confirm the efficacy and good tolerability of lactic acid monotherapy in BV. The study by Ross et al. [10] suggests a lower efficacy of lactic acid gel in BV recurrences compared with metronidazole. However, the long-term recurrence rate was comparable, suggesting that lactic acid monotherapy may not be effective enough in such cases, but may remain effective in sporadic manifestations of the disease. In the present study, a two-step treatment consisting of dequalinium chloride and lactic acid or clindamycin and lactic acid showed better short-term efficacy than treatment with lactic acid alone. However, the recurrence rate at three months was similar to that reported in the study by Ross et al.

When considering alternative antiseptic treatments for BV, special attention should be paid to dequalinium chloride.

Dequalinium chloride, a quaternary ammonium salt, exhibits both anti-inflammatory and antibacterial properties. The anti-inflammatory effect is attributed to the inhibition of protein kinase C, whereas the antibacterial activity is the result of multiple mechanisms, including adsorption on the surface of microorganisms, interaction with the cytoplasmic membrane, changes in cell wall permeability, denaturation of proteins, inhibition of glucose metabolism and adenosine triphosphate synthesis in bacterial mitochondria, disruption of ribosomal protein synthesis and its precipitation, and precipitation of nucleic acids. These are the factors that explain the lack of resistance to dequalinium chloride. Furthermore, dequalinium chloride exhibits minimal systemic absorption, resulting in negligible systemic effects [12].

An *in vitro* study by Gaspar et al. [13] showed that dequalinium chloride effectively destroys biofilms dominated by *Gardnerella* spp. by affecting their biomass and metabolic activity.

A retrospective, multicenter, observational study by Vives et al. [14] demonstrated the efficacy of treatment of BV with 10 mg of dequalinium chloride vaginally for six days in 573 women. Four to six weeks after the end of therapy, 84.8% of participants reported complete resolution of symptoms. A multicenter, blinded, randomized trial by Weissenbacher et al. [15] compared the efficacy of clindamycin with that of dequalinium chloride in women with BV. The clinical cure rate seven days after therapy was 81.5% in the dequalinium chloride group and 78.4% in the clindamycin group. Twenty-five days after therapy, clinical cure rates remained high at 79.5% and 77.6% in the dequalinium chloride and clindamycin groups, respectively.

According to our study, combination therapy with clindamycin and lactic acid showed higher clinical efficacy than clindamycin monotherapy, as reviewed in the study by Weissenbacher et al. [15]. Nevertheless, both the combination therapy during the 3-month follow-up and the clindamycin monotherapy during the 1-month follow-up in the previous study showed high efficacy. In addition, both studies showed good tolerability.

Raba et al. [16] conducted a multicenter, triple-blind, randomized clinical trial to evaluate the efficacy of 10 mg of intravaginal dequalinium chloride used once daily for six days versus 500 mg of oral metronidazole used twice daily for seven days. The clinical cure rate was achieved within 7–11 days after the start of therapy in 92.8% (n = 64/69) of the women receiving dequalinium chloride and 93.2% (n = 69/74) of those receiving metronidazole. However, 20–40 days after treatment, the clinical cure rate decreased in both groups to 79.7% in the dequalinium chloride group and 87.3% in the metronidazole group. Tolerability was rated as very good by 60.0% of patients in the dequalinium chloride group and 38.9% in the metronidazole group. The difference in tolerability between the groups was statistically significant.

The present study, utilizing a two-step treatment with dequalinium chloride and lactic acid, exhibited higher clinical efficacy compared with the studies conducted by Weissenbacher et al. [15] and Raba et al. [16], which evaluated dequalinium chloride monotherapy. In contrast to patients receiving dequalinium chloride monotherapy, patients in our study demonstrated sustained efficacy over a period of three months, a finding that surpasses the outcomes observed in previous studies of dequalinium chloride monotherapy. Furthermore, the tolerability of dequalinium chloride in the two-step therapy in our study was superior to that of monotherapy in previous studies.

The short-term efficacy of two-step therapy, comprising clindamycin and lactic acid, as well as dequalinium chloride and lactic acid, is consistent with the efficacy of clindamycin and dequalinium chloride monotherapy observed in previous studies. Nonetheless, the long-term results of the two-step treatment in this study demonstrated greater efficacy compared with the use of monotherapy in previous studies.

Due to the low sensitivity of this diagnostic criterion, the microscopic examination of the Gram-stained vaginal smear revealed clue cells in a few patients. However, the presence of the other three Amsel criteria (homogeneous grayish-white discharge, specific amine odor, and pH of vaginal discharge >4.5) in the absence of clue cells in the smear allows for the diagnosis of BV [17].

CONCLUSION

The present study demonstrated the high efficacy of a two-step regimen that included both clindamycin and lactic acid and dequalinium chloride and lactic acid. The efficacy of both treatment regimens was demonstrated by their positive impact on both short-term and long-term outcomes. In contrast, lactic acid monotherapy exhibited less pronounced results on day 14 (90.3% vs. 96.8% and 100.0% for the two-step regimens), yet the rates were more comparable after three months (96.4% vs. 96.7% and 93.3%). Importantly, two-step therapy with dequalinium chloride and lactic acid showed high efficacy comparable to two-step therapy with clindamycin and lactic acid. Taking into account the increasing

antibiotic resistance, this strategy is particularly relevant in clinical practice.

The study was conducted as part of A.D. Minakova's dissertation research and was approved by the Local Ethics Committee of First Moscow State Medical University (Protocol No. 01-22, dated January 20, 2022).

ADDITIONAL INFORMATION

Authors' contribution. A.D. Minakova: concept and design of the study, collection and processing of material, statistical data processing, writing the text; T.A. Dzhibladze writing the text; V.M. Zuev, I.D. Khokhlova editing the article. All authors confirm that their authorship meets the international ICMJE criteria (all authors made a substantial contribution to the conception of the work, acquisition, analysis, interpretation of data for the work, drafting and revising the work, final approval of the version to be published and agree to be accountable for all aspects of the work).

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Вклад авторов. А.Д. Минакова — концепция и дизайн исследования, сбор и обработка материала, статистическая обработка данных, написание текста; Т.А. Джибладзе — написание текста; В.М. Зуев, И.Д. Хохлова — редактирование статьи. Все авторы подтверждают соответствие своего авторства международным критериям ICMJE (все авторы внесли существенный вклад в разработку концепции, проведение исследования и подготовку статьи, прочли и одобрили финальную версию перед публикацией).

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