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ORIGINAL RESEARCH

Analysis of the Relationship Between Cervical Cancer Progression and the Microbiome

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Background: Cervical cancer remains a leading malignancy among women worldwide. Emerging evidence suggests that alterations in the cervical microbiota may influence its development and progression.

Objective: To compare the cervical microbiota composition and diversity between cervical cancer patients and healthy women using 16S rRNA gene sequencing.

Methods: Between January and June 2024, cervical tissue samples were collected from 40 cervical cancer patients and 40 healthy women. Microbial DNA was extracted and the V3–V4 region of the 16S rRNA gene was sequenced using the Illumina Novaseq 6000 platform. QIIME2 and R software were used for microbial classification and diversity analysis. LEfSe was applied to identify differentially abundant taxa between groups.

Results: At the phylum level, *Firmicutes* dominated the control group (67.91%), while dropped to 31.03% in cervical cancer patients. *Actinobacteria* (26.22% vs 14.37%) and *Proteobacteria* (27.61% vs 0.72%) were significantly elevated in the cancer group. At the genus level, *Lactobacillus* was predominant in controls (46.27%), while reduced in patients, and Rhodococcus and Klebsiella were notably enriched. Alpha diversity (Shannon index) exhibited no significant difference between groups, whereas richness indices (Chao1 and ACE) were significantly higher in the cancer group ($p < 0.05$). Beta diversity analysis revealed a clear distinction in community structure ($t = 10.225$, $P = 0.001$). LEfSe identified Rhodococcus (LDA = 5.24), Klebsiella (LDA = 5.17), and Ralstonia as significantly more abundant in the cancer group, while *Firmicutes* (LDA = 5.31) was characteristic of the control group.

Conclusion: Cervical cancer patients exhibited distinct cervical microbiota profiles, with reduced Firmicutes and elevated Actinobacteria and Proteobacteria. Increased levels of Rhodococcus and Klebsiella suggested a potential association between microbial dysbiosis and cervical cancer. These findings highlight the microbiome's relevance to tumor biology and support further investigation into its diagnostic and therapeutic potential.

Keywords: cervical cancer, microbiome, high-throughput sequencing, alpha diversity, beta diversity, LEfSe analysis

Introduction

Cervical cancer remains one of the most prevalent malignancies affecting women globally, ranking as the fourth most common cancer among women according to recent global cancer statistics.[1] Its high incidence poses a serious threat to women's health and represents a significant public health challenge. The etiology of cervical cancer is multifactorial, involving early marriage, multiple pregnancies, frequent abortions, genetic susceptibility, sexually transmitted infections, and, most notably, persistent infection with high-risk human papillomavirus (HPV).[2,3] HPV, a circular double-stranded DNA virus with a tropism for epithelial cells, is typically cleared within two years; however, in a subset of individuals, persistent infection may lead to malignant transformation, with approximately 5–10% progressing to cervical cancer.[4]

Although persistent HPV infection is the primary cause of cervical cancer, emerging evidence highlights the role of the cervical microbiome in its pathogenesis.[5,6] Advances in microbiome research have provided novel insights into the interactions between microbial communities and cancer development. These communities may contribute to tumorigenesis by modulating the host immune system and promoting chronic inflammation.[7,8] Furthermore, microbial dysbiosis

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may influence the persistence of high-risk HPV infection and modulate local immune responses, creating a tumor-promoting microenvironment. Certain bacteria may produce metabolites or toxins that contribute to epithelial damage or alter host gene expression, thereby facilitating oncogenic transformation.

Recent studies have demonstrated that the cervical microbiome in healthy women is predominantly composed of Lactobacillus species, playing a vital role in maintaining microbial homeostasis in both the vagina and cervix. These bacteria help inhibit the proliferation of pathogenic microorganisms. In contrast, cervical cancer patients exhibit a notable increase in microbial diversity, characterized by a reduction in Lactobacillus abundance and a significant rise in the relative abundance of certain pathogenic bacteria. Such dysbiosis may disrupt microbial equilibrium and contribute to the development of cervical lesions.[9] Despite these observations, research on the cervical microbiome in the context of cervical cancer remains limited, and the specific microbial alterations and their functional implications have not yet been fully explored.[10]

Although several studies have suggested links between cervical microbiota composition and HPV persistence or cervical neoplasia, there is still limited understanding of the specific microbial taxa involved and how they vary between cervical cancer patients and healthy individuals. Most existing studies have concentrated on vaginal samples, while fewer studies have directly analyzed cervical tissue microbiota. In addition, regional differences, host genetic factors, and environmental influences may result in distinct microbial profiles that require population-specific investigation.

This study utilized high-throughput 16S rRNA gene sequencing to investigate differences in the cervical microbiota between cervical cancer patients and healthy women. Through a comprehensive analysis of microbial composition and diversity, it aimed to characterize the structural features of microbial communities in cervical cancer and to identify disease-associated taxa. The findings provide new insights into the microbial mechanisms involved in cervical cancer and may help identify potential biomarkers for early diagnosis and personalized treatment strategies.

Study Subjects and Methods

Study Subjects and Grouping

This study aimed to investigate the differences in cervical microbiota composition and diversity between cervical cancer patients and healthy women using 16S rRNA gene sequencing. By characterizing the microbial shifts associated with cervical carcinogenesis, the research could identify specific bacterial taxa that may serve as biomarkers or potential contributors to the pathogenesis of cervical cancer. Understanding these microbial signatures could not only enhance early diagnostic strategies, but also provide novel insights for preventive and therapeutic interventions. Despite the emerging interest in the cervical microbiome, its role in cancer development remains underexplored, particularly in non-Western populations, making this study both timely and valuable for the broader scientific and clinical communities. This study included cervical tissue samples from 40 cervical cancer patients who underwent cervical biopsy or surgical treatment at our hospital and affiliated clinics between January and June 2024. These samples formed the experimental group. At the same time, 40 normal cervical tissue samples were obtained from women undergoing gynecologic surgery (eg, hysterectomy for benign uterine conditions) or cervical conization during routine health check-ups. These women were classified as the control group after confirmation of normal cervical histology. In the experimental group, clinical staging of cervical cancer patients included 9 cases in stage I, 17 in stage II, and 14 in stage III. The average age of the experimental group was 56.17 ± 3.16 years, with 9 smokers and 13 alcohol consumers. In the control group, the average age was 56.33 ± 3.19 years, with 10 smokers and 13 alcohol consumers. There were no statistically significant differences in age, smoking history, or alcohol consumption between the two groups ($P > 0.05$), making the groups comparable. All participants provided informed consent, understanding the purpose, content, and potential risks and benefits of the study. The study was approved by the Ethics Committee of The First Affiliated Hospital of Hebei North University (Zhangjiakou, China; Approval No. 15927299). All the methods were carried out in accordance with the Declaration of Helsinki.

Inclusion Criteria

All specimens were confirmed by pathology, and none of the patients had received radiation therapy, chemotherapy, or immunotherapy. Additionally, participants had not taken antibiotics within the past month and had no metabolic disorders

or other conditions that could affect the cervical microbiota. None had liver or kidney dysfunction, and none had undergone recent hormone therapy.

Exclusion Criteria

Patients with a history of pelvic radiation therapy or chemotherapy, those currently menstruating, pregnant, or breast-feeding, as well as those with immune or endocrine disorders, or a history of hormone use, were excluded from the study.

Other factors known to influence the cervical microbiota, such as recent sexual activity, vaginal douching, intrauterine device (IUD) use, recent gynecological infections, or use of topical vaginal medications, were also considered. Participants with any of these factors in the preceding four weeks were excluded to minimize confounding variables influencing microbial composition. For the control group, inclusion criteria additionally required a negative HPV test result, no cytological abnormalities on recent Pap smears, and no history of cervical pathology. All controls were undergoing cervical conization or hysterectomy for benign, non-infectious gynecologic conditions (eg, uterine fibroids, uterine prolapse), and their cervical tissues were confirmed as histologically normal by pathology.

Methods

DNA Extraction and Library Construction

Microbial DNA was extracted from approximately 20 mg of cervical tissue samples (stored at –80°C) using a commercial extraction kit (OMEGA Bio-Tek, Norcross, GA, USA), following the manufacturer's protocol with minor modifications. Briefly, tissue samples were washed with sterile phosphate-buffered saline (PBS) and lysed using SL3 reagent. After incubation and centrifugation, the supernatant was filtered to remove potential inhibitors. DNA binding and washing were performed using buffers SB, SW1, and SW2, and DNA was eluted with 80 μL of preheated elution buffer (Buffer SE, 70°C). The eluted DNA was stored at –20°C until further analysis. For library preparation, the V3–V4 hypervariable region of the bacterial 16S rRNA gene was amplified using specific primers. PCR products were purified using a commercial DNA purification column (OMEGA Bio-Tek) and assessed via agarose gel electrophoresis. DNA fragments passing quality control were used for downstream analysis.

Sequencing and Quality Control

High-throughput sequencing was performed on an Illumina Novaseq 6000 platform. Raw image data were converted into sequencing reads and stored in FASTQ format. The preprocessing of raw data included quality filtering using Trimmomatic v0.33 and primer removal using cutadapt 1.9.1 to obtain clean reads. These clean reads were then stitched together using Usearch v10, followed by the removal of chimeric sequences using UCHIME v4.2 to obtain valid data.

Species Distribution Heatmap and Phylogenetic Tree Construction

Valid reads were clustered into operational taxonomic units (OTUs) at a 97% similarity threshold using Usearch. Taxonomic annotation of OTUs was performed using the SILVA database and QIIME2 software, providing species abundance information at various taxonomic levels (phylum, class, order, family, genus, species). A phylogenetic tree was constructed by aligning representative sequences of the most abundant species, and the resulting tree was visualized using Python.

Alpha Diversity and Statistical Analysis

Alpha diversity, which reflects species richness and evenness within a sample, was assessed using the Chao1 richness estimator and ACE richness estimator to measure species abundance. Shannon-Wiener and Simpson indices were used to evaluate species diversity. A higher Shannon or Simpson index indicates greater species diversity. Alpha diversity indices were calculated using the QIIME2 platform, and statistical differences between groups were evaluated using *t*-tests.

Beta Diversity and Statistical Analysis

Beta diversity was analyzed using QIIME to compare the differences in microbial composition between samples. Principal Component Analysis (PCA) and Principal Coordinates Analysis (PCoA) were employed to visualize the diversity between samples, focusing on the top contributing factors to observe any significant differences.

Linear Discriminant Analysis (LDA) and LEfSe Analysis

Linear discriminant analysis effect size (LEfSe) was conducted using R software to identify biomarkers with a LDA score greater than 4.0. LEfSe analysis was used to determine the species with significant differences between groups and assess their relative abundance's contribution to these differences.

Results

Clinical Characteristics and Risk Factors of Cervical Cancer Patients

Among the 40 cervical cancer patients included in the study, the mean age was 56.17 ± 3.16 years. In terms of disease staging, 9 patients were classified as stage I, 17 as stage II, and 14 as stage III according to the FIGO (International Federation of Gynecology and Obstetrics) staging system. Clinically, common symptoms included abnormal vaginal bleeding, pelvic pain, and increased vaginal discharge. High-risk factors identified in the patient group included early onset of sexual activity (before age 18) in 14 patients, multiple sexual partners in 17, and a history of persistent high-risk HPV infection in 28 cases. Additionally, 23 patients had a history of at least three full-term pregnancies, and 19 had a record of long-term oral contraceptive use. Smoking was reported by 9 patients, and 13 reported regular alcohol consumption. These clinical and behavioral characteristics are consistent with established epidemiological risk factors for cervical cancer and provide further context for interpreting the observed microbiota alterations.

Microbial Composition of the Two Groups

Phylum-Level Composition

At the phylum level, operational taxonomic units (OTUs) were assigned to several bacterial phyla, including *Firmicutes, Actinobacteria, Proteobacteria, Bacteroidetes, Fusobacteria*, and *Tenericutes*. In the control group, *Firmicutes* dominated the microbiota, comprising 67.91% of the total composition, while its relative abundance significantly declined to 31.03% in the experimental group. *Actinobacteria* was the second most prevalent phylum in both groups, increasing from 14.37% in the control group to 26.22% in the experimental group. *Proteobacteria* exhibited a marked rise in abundance in the experimental group, from 0.72% to 27.61%. *Fusobacteria* also showed a slight increase, from 4.72% to 6.81%. In contrast, *Bacteroidetes* and *Tenericutes* decreased from 8.91% to 6.74% and from 2.53% to 0.79%, respectively. The relative abundance of other phyla remained stable, accounting for 0.84% in the control group and 0.80% in the experimental group (Figure 1). These results indicate a substantial shift in microbial composition, characterized by a decrease in *Firmicutes* and an increase in *Proteobacteria* and *Actinobacteria* in the experimental group.

Genus-Level Composition

At the genus level, the *Lactobacillus* genus was dominant in the control group, representing 46.27% of the total microbiota. However, in the experimental group, the proportion of *Lactobacillus* and Corynebacterium significantly decreased, losing their dominance. In contrast, genera, such as *Rhodococcus, Klebsiella*, and *Aerococcus* exhibited a marked increase in the experimental group. Additionally, the experimental group displayed a greater variety of genera compared to the control group (Figure 2).

Alpha Diversity of Cervical Microbiota

Alpha diversity analysis showed no significant difference in overall diversity between the two groups. The Shannon index was 4.986 ± 0.991 in the control group and 5.196 ± 1.006 in the experimental group (P = 0.537). Similarly, the Simpson index was 0.872 ± 0.087 in the control group and 0.881 ± 0.011 in the experimental group, with no significant difference (P = 0.119). However, species richness was significantly higher in the experimental group. The Chao1 index for the experimental group was 724.470 ± 306.348, compared to 619.868 ± 256.652 in the control group (P = 0.024). Likewise, the ACE index was 739.886 ± 308.73 in the experimental group, significantly higher than the control group's 629.151 ± 260.328 (P = 0.031). While there was no significant difference in overall microbial diversity (Shannon and Simpson indices), the experimental group demonstrated significantly greater species richness (Chao1 and ACE indices) compared to the control group (Figure 3).

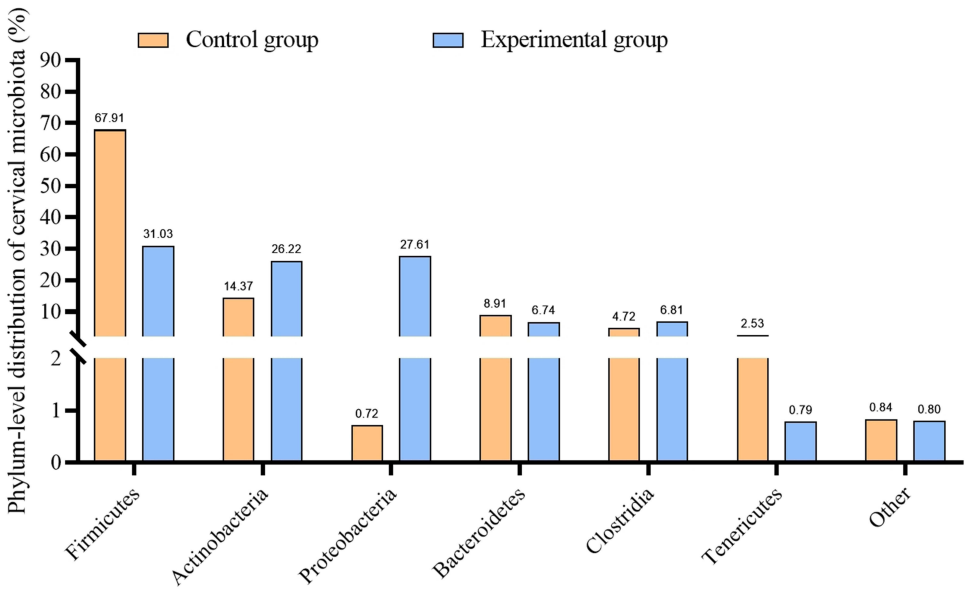

Figure 1 Phylum-Level Distribution of Cervical Microbiota (%).
Notes: The experimental group consists of 40 cervical cancer patients, while the control group consists of 40 healthy women. High-throughput sequencing was used to amplify the V3-V4 region of the bacterial 16S rRNA gene, and relative abundance represents the percentage of each phylum in the total microbiota. The control group was selected from healthy women undergoing routine physical exams during the same period.

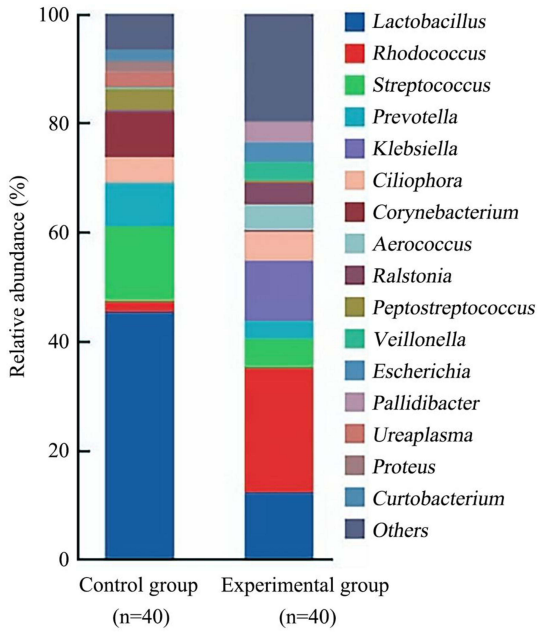

Figure 2 Genus-Level Distribution of Cervical Microbiota in Cervical Cancer Patients and Healthy Controls.
Notes: The experimental group consists of 40 cervical cancer patients, while the control group includes 40 healthy women. High-throughput sequencing was used to amplify the V3-V4 region of the bacterial 16S rRNA gene. Relative abundance represents the percentage of each genus in the total microbiota.

Beta Diversity of Cervical Microbiota

Principal Coordinates Analysis (PCoA) revealed distinct separation between the experimental and control groups, indicating significant differences in the overall microbial community structure (Figure 4). Statistical analysis confirmed that the differences in microbial community structure between the two groups were significant ($t = 10.225$, $P = 0.001$).

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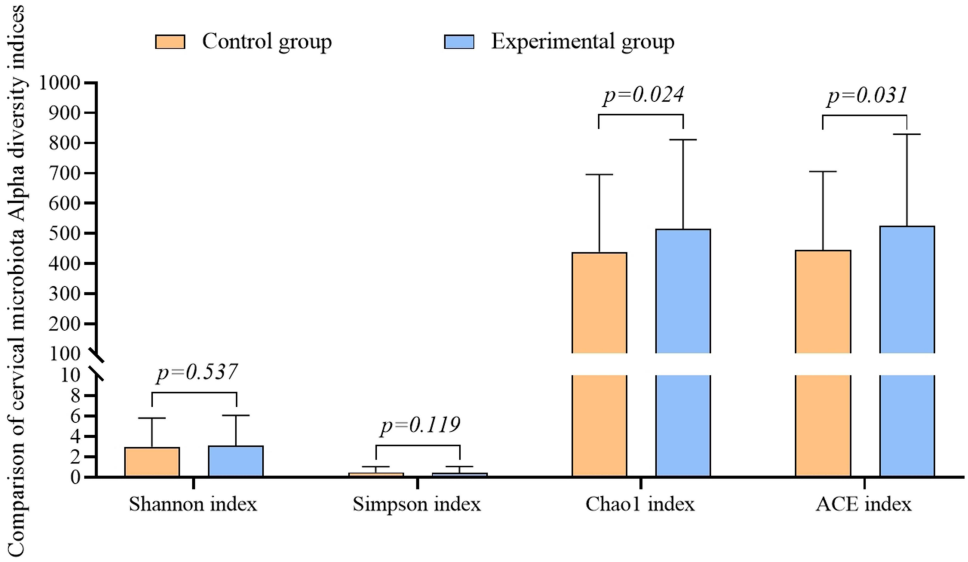

Figure 3 Comparison of Alpha Diversity Indices Between Cervical Cancer Patients and Healthy Controls.

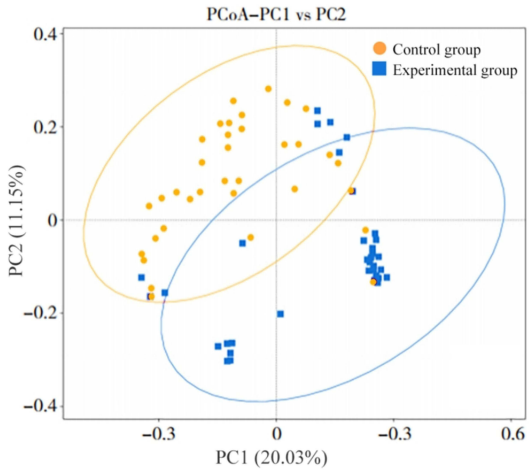

Figure 4 Beta Diversity PCoA Analysis of Cervical Microbiota in Cervical Cancer Patients and Healthy Controls.
Abbreviation: PCoA, Principal Coordinates Analysis.

Differential Species Analysis of Cervical Microbiota

LDA identified bacterial taxa with a LDA score > 4.0 (Figure 5). In the experimental group, *Rhodococcus* (LDA = 5.24), *Klebsiella* (LDA = 5.17), *Enterobacter* (LDA = 5.11), *Ralstonia* (LDA = 5.16), *Pseudomonas* (LDA = 4.66), and *Veillonella* (LDA = 4.71) were identified as key genera. At the phylum level, *Firmicutes* (LDA = 5.31) was a characteristic phylum in the control group, although no representative genera were identified at the genus level. Further genus-level analysis (Figure 6) revealed that *Rhodococcus* (P = 0.028), *Ralstonia* (P = 0.016), *Veillonella* (P = 0.042), *Burkholderia* (P = 0.036), and *Pseudomonas* (P = 0.023) were significantly more abundant in the experimental group compared with the control group.

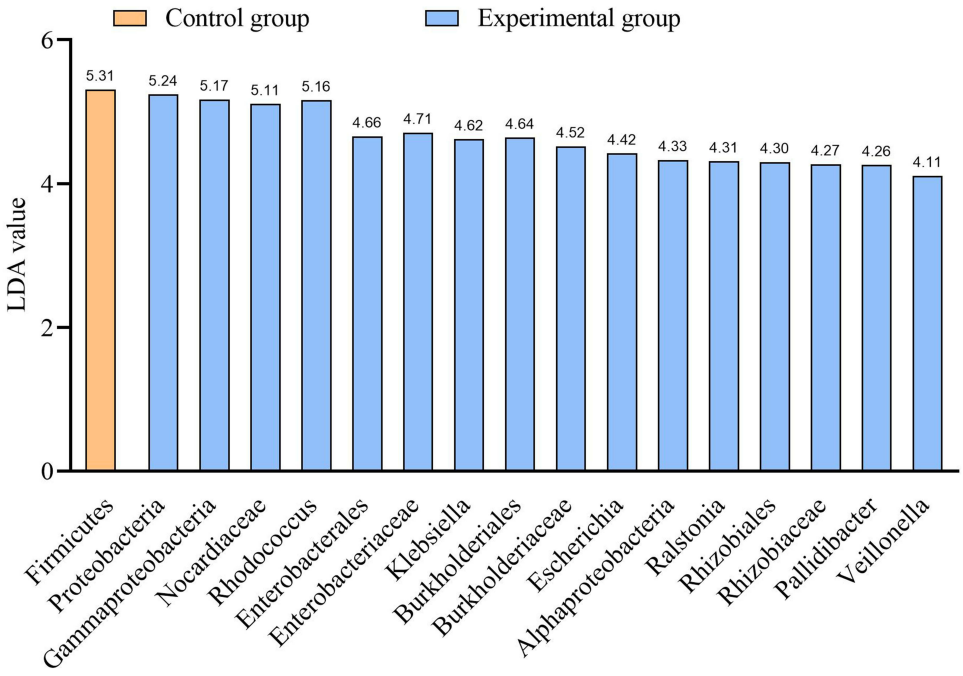

Figure 5 LDA > 4.0: Taxonomic Distribution of Cervical Microbiota in Cervical Cancer Patients and Healthy Controls.
Notes: The experimental group includes 40 cervical cancer patients, and the control group includes 40 healthy women. High-throughput sequencing was used to amplify the V3-V4 region of the bacterial 16S rRNA gene. Relative abundance represents the percentage of each genus in the total microbiota.

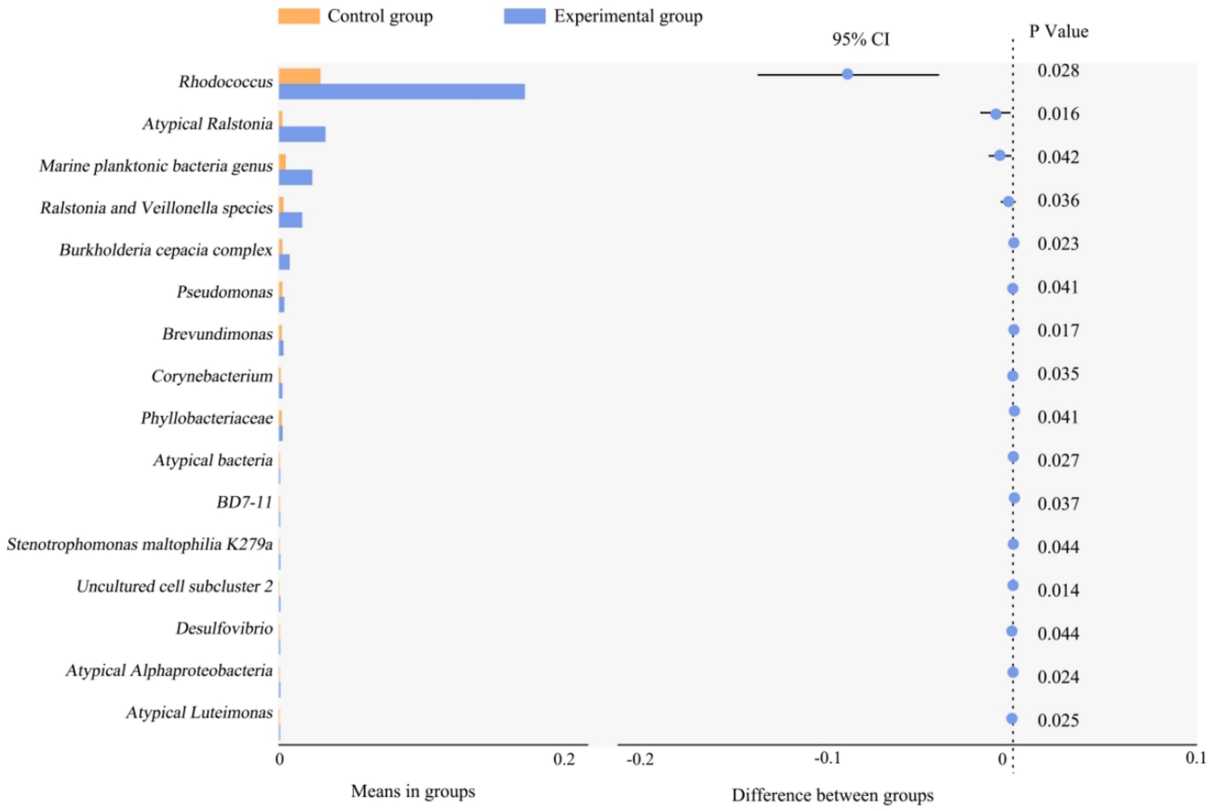

Figure 6 Genus-Level Differences in Cervical Microbiota Between Cervical Cancer Patients and Healthy Controls.

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Discussion

The primary aim of this study was to explore how microbial community composition varies between cervical cancer patients and healthy controls, and to identify microbial signatures potentially linked to carcinogenesis. While prior research has primarily concentrated on the vaginal microbiome, the cervical microbiota and its possible roles in cervical cancer remain poorly understood. This study addressed this knowledge gap by employing high-throughput sequencing to provide a more comprehensive overview. The findings may provide a basis for microbiota-based screening or therapeutic strategies, supporting the growing recognition of the microbiome as an influential factor in cancer biology.

Historically, the upper female reproductive tract was considered sterile, leading most studies to concentrate primarily on the vaginal microbiota. However, in 2017, Chinese researcher Wu Ruifang was the first to demonstrate the presence of diverse microbial communities in the uterus and upper reproductive tract. Furthermore, microbial composition has shown to vary progressively along the reproductive tract, with *Firmicutes* predominating in the lower tract and *Proteobacteria, Actinobacteria*, and *Bacteroidetes* becoming increasingly prevalent in the upper regions.[11] Compared with the vaginal microbiota, the cervical microbiota exhibits reduced *Lactobacillus* abundance and increased microbial diversity.[12] In the present study, high-throughput 16S rRNA gene sequencing was employed to investigate the composition and diversity of the cervical microbiota in patients with cervical cancer and in healthy controls.

Previous research has demonstrated that treatment with *Lactobacillus* can upregulate E-cadherin expression in tumor tissues, thereby inhibiting the migration of cervical cancer cells, suggesting a potential role for *Lactobacillus* in preventing HR-HPV infection and cervical carcinogenesis.[13] In the present study, significant alterations in the cervical microenvironment were found in patients with cervical cancer. At the phylum level, the relative abundance of *Firmicutes* was markedly reduced, whereas *Actinobacteria* and *Proteobacteria* were notably enriched. In healthy women, the cervical microbiota is predominantly composed of *Lactobacillus*, playing a crucial role in maintaining microbial homeostasis in the vagina and cervix by suppressing the proliferation of pathogenic microorganisms. Conversely, in cervical cancer patients, *Lactobacillus* was significantly depleted, accompanied by a marked increase in genera such as *Rhodococcus* and *Klebsiella*. These findings suggest a strong association between cervical microbiota dysbiosis and the pathogenesis of cervical cancer. Specifically, the disruption of the vaginal microenvironment may facilitate persistent HR-HPV infection and promote the progression of cervical intraepithelial neoplasia (CIN) and cervical cancer. This highlights the importance of preserving a healthy vaginal microbiota to reduce the risk of HPV persistence and malignant transformation.[14] The compositional shifts may reflect a disrupted cervical mucosal barrier and altered immune signaling. For instance, species, such as *Rhodococcus* and *Klebsiella* are known to produce pro-inflammatory factors and biofilms, which can impair epithelial integrity and immune surveillance, thereby facilitating persistent HPV infection and cervical epithelial neoplasia. Increased abundance of *Proteobacteria* has been associated with inflammatory microenvironments in other cancer types, such as colorectal cancer and gastric cancer, suggesting a potentially conserved oncogenic microbial signature across tissues.

Other studies have shown that frequent vaginal douching may disrupt the balance of the normal vaginal microbiota, affecting both its abundance and diversity, thereby increasing the risk of CIN and carcinogenesis. Under normal conditions, Lactobacillus maintains the vaginal acidic environment by producing lactic acid, which inhibits the growth of pathogenic microorganisms. Disrupting this balance can lead to vaginal dysbiosis and disease.[15]

Similar trends have been found in other malignancies. In colorectal cancer, for instance, increased microbial diversity and the overrepresentation of genera, such as *Fusobacterium* have been implicated in tumor progression and immune evasion. Breast and pancreatic cancer studies also report distinct microbial shifts that are being explored as early diagnostic markers.[16] These cross-cancer comparisons highlight the emerging role of the microbiome not only as a bystander, but also as an active participant in tumor microenvironment modulation. This study adds to this growing body of evidence by identifying cervical cancer–specific microbial signatures, particularly the depletion of *Lactobacillus* and enrichment of *Klebsiella* and *Rhodococcus*, which may have similar roles in cervical carcinogenesis.

Additionally, a cohort study in China found that HPV infection increases the richness and diversity of vaginal bacteria, a phenomenon that is more pronounced in patients with cervical cancer.[16] However, a few studies present conflicting views, leaving the relationship between HPV and the vaginal microbiome uncertain and in need of further

investigation. Some research suggests that a relatively high viral load may be critical, though the mechanism remains to be explored.[14] In HPV-positive women, the growth of Lactobacillus is further suppressed, altering the distribution of microorganisms and ultimately leading to an imbalance in the vaginal microenvironment. This points to a complex interaction between HPV infection and vaginal microbiota dysbiosis.[17] On the one hand, HPV infection may lead to changes in the vaginal microenvironment, increasing bacterial richness and diversity; on the other hand, an imbalanced vaginal microbiota may further increase the risk of cervical carcinogenesis. Therefore, monitoring and intervening in the vaginal microenvironment could be an effective strategy to prevent persistent HPV infection and the progression of cervical cancer.[18] The findings of the present study have potential implications for precision medicine. Microbial signatures can aid in stratifying patients based on their risk profiles for persistent HPV infection and progression to high-grade cervical lesions or cancer. Such profiles can also inform individualized interventions, including microbiome modulation through probiotics, antibiotics, or personalized immunotherapy. As in colorectal and lung cancer research, where gut microbiota composition has been linked to immunotherapy response, similar strategies may emerge in cervical cancer management based on cervicovaginal microbiome profiling.

Alpha diversity analysis showed that the Chao1 and ACE indices in the experimental group were significantly higher than those in the control group, indicating greater species richness in cervical cancer patients. Although the Shannon and Simpson indices did not exhibit significant differences, the findings suggest that cervical cancer patients harbor a microbiota with greater species richness. However, increased diversity does not necessarily reflect a healthy ecological balance. This finding aligns with previous studies, which have shown that cancer patients often exhibit higher microbial diversity than healthy individuals, possibly due to the increased presence of pathogenic bacteria.[19,20] Beta diversity analysis revealed a significant difference in microbial community structure between cervical cancer patients and healthy women. PCoA analysis further confirmed this marked difference in community structure between the two groups, consistent with the identification of specific microorganisms.[21] In the present study, 28 out of 40 cervical cancer patients were confirmed to be positive for high-risk HPV (HR-HPV), consistent with the established role of HPV infection in cervical carcinogenesis. Notably, patients with HR-HPV infection showed reduced relative abundance of *Lactobacillus* and increased prevalence of genera such as *Rhodococcus* and *Veillonella*, which have previously been associated with chronic inflammation and epithelial barrier disruption. These findings support the hypothesis that HPV infection and microbiota dysbiosis may act synergistically to promote cervical cancer development. However, as HPV testing was not conducted for the control group, these relationships should be interpreted with caution and confirmed in future studies with matched HPV status.

Numerous studies have reported that cervical cancer is associated with increased microbial diversity and decreased dominance of Lactobacillus. For instance, Mitra et al[22] found that women with high-grade cervical lesions had a significant shift from Lactobacillus-dominated microbiota to diverse anaerobic bacteria. Another study by Brusselaers et al[23] highlighted consistent patterns of vaginal dysbiosis in HPV-positive and cervical cancer patients, suggesting that specific microbial profiles could influence persistence of HPV and carcinogenic progression. These findings are in line with our results and underscore the potential role of the cervicovaginal microbiome in modulating immune responses and epithelial homeostasis during cervical carcinogenesis. In the present study, in the cervical cancer group, characteristic bacteria, such as *Rhodococcus* and *Klebsiella* were significantly increased, which may be closely related to the onset and development of cervical cancer. Additionally, LEfSe analysis showed a significant increase in the abundance of pathogenic bacteria, such as *Rhodococcus, Ralstonia*, and *Veillonella* in cervical cancer patients. These bacteria may promote cancer development by inducing chronic inflammation or interfering with the host immune response. These findings support current theories about the interaction between the microbiome and cancer, suggesting that changes in the abundance of specific microorganisms may influence the host's immune status and microbial balance, thereby contributing to tumor development and progression.[24]

Recent advances in microbiome research, powered by 16S rRNA sequencing and AI-assisted analytics, hold significant promise for advancing personalized medicine in cervical cancer care. Stratifying patients based on microbiota profiles may enable the development of individualized prevention strategies or microbiome-targeted therapies.[19] However, such innovation also raises important ethical questions. The collection and analysis of highly sensitive biological data require robust informed consent procedures and safeguards for privacy and data use. Policy frameworks

in precision medicine should ensure that patient autonomy is protected while maximizing the translational potential of microbiome-based diagnostics. As evidence accumulates, regulatory guidelines should evolve to support the clinical integration of microbiome data, balancing innovation with ethical responsibility.

Medical ethics has largely evolved in response to past socio-scientific calamities. Personalized medicine, with its heightened ability to address the individuality of illness, requires a continuously evolving feedback mechanism, serving as its ethical foundation.[25] The predictive, diagnostic, and therapeutic strengths of personalized medicine depend on high-dimensional data generated through genomics and related technologies. To address the growing challenges and unique complexities of its future mainstream application, legal and regulatory frameworks must be continuously updated and refined to keep pace with the evolving technological landscape.[26] Artificial intelligence is poised to revolutionize reproductive medicine and healthcare by enhancing treatment options for infertility, thereby optimizing procedural planning. It provides a powerful tool for predicting clinical outcomes based on initial parameters. However, integrating artificial intelligence into routine practice will require time and coordinated efforts to fully utilize risk-assessment systems and reshape care delivery models.[27]

The observed alterations in cervical microbiota, especially the depletion of *Lactobacillus* and enrichment of genera, such as *Rhodococcus, Klebsiella*, and *Ralstonia*, may provide novel directions for the development of diagnostic and prognostic biomarkers in the framework of precision medicine. In cancer research more broadly, microbial signatures have already been implicated in stratifying patients for immunotherapy response and treatment outcomes, as evidenced in colorectal and lung cancers. Similarly, the cervical microbiome may serve not only as a marker for disease progression, but also as a potential modulator of treatment efficacy. Integrating microbiome profiling into routine clinical assessments can lead to individualized therapeutic strategies, such as microbiota-targeted interventions, probiotics, or microbiome transplantation, tailored to specific microbial imbalances. Moreover, identifying microbial biomarkers associated with early stages of cervical carcinogenesis holds promise for non-invasive screening tools and risk stratification, particularly in HPV-positive populations. These personalized approaches may significantly enhance current preventive and therapeutic measures by accounting for patient-specific microbiome compositions.

While these findings highlight promising associations between cervical microbiota alterations and cervical cancer, caution must be exercised in interpreting their clinical applicability. This study establishes correlations, but not causality. The observed microbial changes could be a consequence rather than a cause of cervical cancer. Moreover, environmental, behavioral, and treatment-related factors may confound microbiota profiles. Therefore, further longitudinal and mechanistic studies are necessary before considering microbiota-based screening, prevention, or therapeutic strategies.

This study is limited by its cross-sectional design, precluding causal inference between microbial changes and cancer development. Furthermore, 16S rRNA sequencing lacks the resolution to capture functional microbial dynamics and strain-level variation. Future studies integrating metagenomics, transcriptomics, and host immune profiling are essential to elucidate the mechanisms by which specific bacteria influence cervical carcinogenesis. Longitudinal studies may also help determine whether microbial shifts precede, accompany, or result from tumor development, which is crucial for evaluating their biomarker potential.

Conclusion

In conclusion, this study revealed significant differences in the composition and diversity of cervical microbiota between cervical cancer patients and healthy women. These differences suggest a potential association between microbiome alterations and cervical cancer. However, due to the observational nature of this study and the limited sample size, these findings should be interpreted with caution. Further studies with larger cohorts and longitudinal designs are needed to validate these associations and explore the underlying mechanisms. While the results provide valuable insights, additional functional and mechanistic research is essential to determine whether specific microbial changes contribute to cervical carcinogenesis. Future investigations should also assess the feasibility of using microbiome profiles as biomarkers for early diagnosis or as targets for personalized interventions in cervical cancer.

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Disclosure

The authors declare that they have no competing interests in this work.

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