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Results of a phase II randomized, double-blind, placebo-controlled trial of Polyphenon E in women with persistent high-risk HPV infection and low-grade cervical intraepithelial neoplasia☆

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HIGHLIGHTS

- We conducted the largest randomized trial of a green tea extract for HPV cervical disease.
- Results show that Polyphenon E is acceptable, safe and well tolerated.
- Polyphenon E was not shown to promote the resolution of persistent high-risk HPV.

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ABSTRACT

Objective. *In vitro* data and pilot data suggest that green tea catechins may possess chemopreventive activity for cervical cancer and precursor lesions. We conducted a randomized, double-blind, placebo-controlled trial of Polyphenon E (decaffeinated and enriched green tea catechin extract) in women with persistent human papillomavirus (HPV) infection and low-grade cervical intraepithelial neoplasia (CIN1) to evaluate the potential of Polyphenon E for cervical cancer prevention.

Methods. Ninety-eight eligible women were randomized to receive either Polyphenon E (containing 800 mg epigallocatechin gallate) or placebo once daily for 4 months. The primary study outcome was oncogenic HPV clearance and clearance of CIN1.

Results. Polyphenon E was shown to be acceptable, safe and well tolerated. There was no difference in the response rate by treatment allocation. Complete response, defined as negative for high-risk HPV and normal histopathology, was noted in 7 (17.1%) and 6 (14.6%) women in the Polyphenon E and placebo arms, respectively. Progression, defined as persistent oncogenic HPV with histopathologic evidence of progression, was more common in the Polyphenon E group than in the placebo group [6 (14.6%) vs. 3 (7.7%)].

Conclusion. Based on the largest randomized placebo-controlled trial of a green tea extract for HPV related cervical disease, we conclude that 4 months of Polyphenon E intervention did not promote the clearance of persistent high-risk HPV and related CIN1. Further studies may be necessary to better delineate the risk factors for persistent HPV infection and biology of the disease to facilitate the evaluation of chemopreventive strategies.

1. Introduction

Cervical cancer is the most common gynecologic malignancy in women worldwide with an estimated annual incidence of 470,000 cases [1]. Persistent high-risk human papillomavirus (HPV) infection is a requisite precursor to the development of nearly all (>99%) of pre-malignant (high-grade cervical intraepithelial neoplasia) and invasive carcinomas of the cervix. The recent introduction of virus-like particle prophylactic HPV vaccination provides an effective primary prevention strategy for squamous and adenomatous malignancies of the cervix.

Abbreviations: HPV, human papillomavirus; CIN1, low-grade cervical intraepithelial neoplasia; EGCG, epigallocatechin gallate.

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Prophylactic vaccines however are type-specific (16/18 and 16/18/6/11, respectively) and relatively expensive making them inaccessible to many domestic and developing world underserved populations. Although recent encouraging results from the National Health and Nutrition Examination Survey (NHANES) indicate that within 4 years of HPV vaccination introduction there has been a 56% decrease in vaccine-type HPV prevalence among females aged 14–19 years[2], there has not been a decrease in HPV prevalence among older age groups, and current bivalent and quadrivalent prophylactic type restricted vaccines leave nearly a third of high-risk oncogenic strains without a viable primary prevention strategy. A range of non-surgical treatments have been evaluated for the treatment of cervical cancer precursor lesions and prevention of cervical cancer, but the results of these studies have been clinically inconclusive and/or have been associated with safety concerns.

In vitro studies have suggested that green tea catechins may exert chemopreventive activity for cervical cancer. We have shown that epigallocatechin gallate (EGCG, a major green tea catechin) and Polyphenon E (a decaffeinated, enriched, and defined mixture of green tea catechins) inhibited the growth of HPV-positive cervical cancer cells and HPV-immortalized cervical epithelial cells in a dose dependent fashion [3]. Induction of apoptosis and cell cycle arrest was observed in EGCG and Polyphenon E treated cells. Apoptosis-related proteins, p53 and p21 showed dose dependent increase while p27 was not affected. HPV-E7 protein expression was decreased with green tea catechin treatment. In addition, green tea catechins or EGCG has been shown to inhibit epidermal growth factor receptor activation [4], telomerase activity [5], phosphatidylinositol 3-kinase/Akt and extracellular signal-regulated kinase 1/2 signaling pathways [6] in HR HPV immortalized cervical epithelial cells or cervical cancer cells which lead to growth cession.

Preliminary clinical efficacy of green tea extracts (Polyphenon E or EGCG) in patients with HPV infected cervical lesions has been reported [7]. Fifty-one patients with various degrees of cervical dysplasia (mild, moderate, and severe) were divided to receive oral Polyphenon E capsules (containing 200 mg EGCG), EGCG capsules (200 mg), vaginal Polyphenon E ointment, or a combination of oral and vaginal Polyphenon E. Following 8 to 12 weeks of intervention, 23 of 33 subjects receiving either oral or vaginal Polyphenon E demonstrated regression of cervical dysplasia, with a trend toward clearance of HPV. Clinical efficacy of topical sinecatechins, a defined green tea extract, in the treatment of external genital and perianal warts has been documented and topical green tea therapy is now commercially available for treatment of condyloma.[8]

Based on these preliminary findings, we conducted a randomized, double-blind, placebo-controlled clinical trial of oral Polyphenon E in women with persistent high-risk HPV (HR HPV) infection and concomitant low-grade cervical intraepithelial neoplasia (CIN1) to evaluate the potentials of Polyphenon E for cervical cancer prevention. We hypothesized that oral Polyphenon E intervention would induce the histologic regression of CIN1 by promoting the clearance on oncogenic HPV.

2. Materials and methods

2.1. Study drugs

Polyphenon E contains 85%–95% total catechins, with 56%–72% as EGCG. The agent was provided to the National Cancer Institute (IND number 58,367), Division of Cancer Prevention (NCI, DCP) by Mitsui Norin. This study used Polyphenon E oral capsules, standardized to contain 200 mg EGCG per capsule, and matched placebo capsules, supplied by NCI, DCP. Study capsules were stored at room temperature and protected from environmental extremes.

2.2. Study population

The study was conducted primarily at the University of Arizona (Tucson, Arizona), with additional accrual at Maricopa Integrated Health System (Phoenix, Arizona), and Southern Pines Women's Health Center (Southern Pines, North Carolina). All potential participants were identified by participating community clinicians, who provided documentation of HPV, and/or cytology. In cases where oncogenic HPV status was unknown, specimens were collected to establish baseline HPV status during the pre-screening phase of the trial. Participants were required to be at least 18 years of age, had normal liver and kidney function, and good performance status. To be eligible subjects were required to be positive for HR HPV by DNA hybrid capture and have at least one biopsy with histologically documented low-grade cervical intraepithelial neoplasia (CIN) at the time of enrollment. To document persistence prior to enrollment, eligibility also required (6 to 12 months prior) a positive HR HPV on DNA hybrid capture and either LSIL cytology or histologically documented CIN1 on cervical biopsy.

Participants were excluded if they were pregnant or breast feeding, consumed tea regularly within 1 month of enrollment, had a history of allergic reaction to tea or related dietary projects, had been treated for genital condyloma within 30 days of enrollment, were receiving other investigational agents, had prior pelvic irradiation, were HIV positive, had uncontrolled inter-current illness, had invasive or high-grade intraepithelial neoplasia, or had a history of cancer except non-melanoma skin cancer. The study was approved by the institutional review boards of the University of Arizona as well as the collaborating institutions. Written informed consent in English or Spanish was obtained from all participants.

2.3. Study procedures

During the initial visit, participants underwent eligibility evaluation. Baseline HPV, cytology and histopathology data were reviewed as part of eligibility determination. Each participant underwent an interview and a brief physical exam to document medical history, performance status, height, weight, blood pressure, pulse, and temperature measurements. Blood samples were collected for complete blood count with differentials and blood chemistry panel. A urine sample was collected for urine pregnancy test. Participants also underwent baseline colposcopy to confirm the presence of an evaluable low-grade CIN lesion occupying at least 1 quadrant and to exclude the presence of a concurrent high-grade CIN. Colposcopic biopsy with endocervical curettage was obtained if not performed within 90 days prior to enrollment. Specimens were collected for cervical cytology and HR HPV testing.

Upon determination of eligibility, participants were randomized to receive Polyphenon E or placebo. An adaptive allocation randomization procedure was implemented [9] to balance the two groups on the basis of age. Participants were instructed to take 4 study capsules each morning with food for 4 months. Participants returned to the clinic every month for the following procedures: urine collection for pregnancy test, assessment of adverse events, compliance verification with review of drug diary and pill count, and blood collection for hepatic panel.

At the end of the 4-month intervention, blood samples were collected for complete blood count with differentials and blood chemistry panel. A urine sample was collected for urine pregnancy test. All participants underwent a standardized post-intervention colposcopic evaluation with collection of endocervical curettage and collection of specimens for cervical cytology and HR HPV testing. This included biopsy of all clinically visible lesions as well as random biopsies of three of four quadrants when no lesions were visible. All histopathology specimens were reviewed in a blinded fashion by an experienced gynecologic pathologist and were subjected to a second quality control review.

The primary study outcome was HR HPV clearance and resolution of the low-grade CIN at month 4. HR HPV status was determined using DNA hybrid capture testing using the high-risk oncogenic probe (HC2, Hologic, Lexington Massachusetts) and performed by a commercial laboratory. Resolution of low-grade CIN was based on evaluation of cervical cytology and biopsy/endocervical curettage obtained at month 4. This