Effects of Continuous Combined Oral Contraceptives on Mouse Mammary Gland Structure and Tumor Progression

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Abstract

The use of cyclical oral contraceptives (OC) is associated with an overall increased risk of ER-positive breast cancer in particular, in young women, and women who start OC early in life. The effect of estrogen (E2) in cyclical OC regimens is an important factor in the risk of breast cancer in recent breast cancer risk. Treatment with estrogen alone, however, does not increase the risk of breast cancer. The addition of progestins to cyclical OC regimens is expected to reduce the risk of breast cancer. However, the results of previous studies have been inconsistent. The present study aimed to investigate the effects of estrogen and progestins on breast cancer risk in mice.

Materials and Methods

Animals

BALB/c mice were purchased from Jackson Laboratory at 35 days of age. Mice were tail-veined for 4 weeks to ensure that they were estrous. The mice were housed and kept on a 12-hr light-dark cycle and were treated with 20 mg of tamoxifen daily. The mice were then divided into three groups: control (C), continuous OC (COC), or cyclic OC (COC). The mice were treated with a single oral dose of tamoxifen to prevent the development of mammary gland tumors. The mice were then divided into three groups: control (C), continuous OC (COC), or cyclic OC (COC). The mice were treated with a single oral dose of tamoxifen to prevent the development of mammary gland tumors.

Dietary model of OC exposure

Mice were grouped in two sets of 7 days, where they were single housed for acclimation to liquid Lucifer/Carb (Dyets, Inc., Cat #870072). Weights and dietary consumption were measured and calculated to be 10 g per day. Mice were fed a commercial diet (Dyets, Inc., Cat #990012) to ensure that the mice were receiving the same amount of food. Mice were then divided into three groups: control (C), continuous OC (COC), or cyclic OC (COC). The mice were treated with a single oral dose of tamoxifen to prevent the development of mammary gland tumors.

Results

Figure 1: Effect of OC on epithelial gland morphometry. Top: Whole mount mammary gland images showing terminal structures in mammary epithelial/white adipose tissue. Control (C), continuous OC (COC), cyclic OC (COC). Bottom: Continuous OC, but not cyclic OC, demonstrates increases in epithelial density on H&E-stained paraffin sections. Bars represent mean ± SEM, n=10 (control and continuous OC) or n=11 (cyclic OC).

Figure 2: Effect of OC on mammary gland structure. Top: Representative histology images showing terminal structures in mammary white adipose tissue. Control (C), continuous OC (COC) and cyclic OC (COC). Bottom: Continuous OC, but not cyclic OC, demonstrates increases in epithelial density on H&E-stained paraffin sections. Bars represent mean ± SEM, n=10 (control and continuous OC) or n=11 (cyclic OC).

Figure 3: Continuous OC increases proliferation in the mammary gland. Top: Representative histology images showing increases in BrdU incorporation in situ. Mice were treated with BrdU and killed on day 16 of the treatment period. Bars represent mean ± SEM, n=10 (control and continuous OC) or n=11 (cyclic OC).

Figure 4: Both continuous and cyclic OC increase the cellular infiltration in the white adipose tissue. Top: Representative images from H&E-stained paraffin sections from mammary glands in mice of the same size, age, and weight. Control (C), continuous OC (COC), cyclic OC (COC). Bars represent mean ± SEM, n=10 (control and continuous OC) or n=11 (cyclic OC).

Figure 5: Effects of OC regimens on palpable tumor burden and multiplicity. Left: Both continuous and cyclic OC decreased palpable tumor burden. Right: Despite injection into a single site in the mammary fat pad, multiple lesions arose at the site of injection. Both continuous and cyclic OC regimens decreased palpable tumor burden. Bars represent mean ± SEM, n=10 (control and continuous OC) or n=11 (cyclic OC).

Summary

- Both continuous and cyclic OC regimens have been shown to reduce the risk of breast cancer.
- Continuous OC regimens have been shown to reduce the risk of breast cancer.
- Cyclic OC significantly increased epithelial proliferation.
- Continuous OC significantly increased epithelial proliferation.
- Only continuous OC significantly increased tumor burden.
- Both OC regimens increased tumor burden.
- Once tumors were established, OC did not affect tumor growth rate.

References

