Detecting estrogenic activity in water samples with estrogen-sensitive yeast cells using spectrophotometry and fluorescence microscopy

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Poster Abstract

Environmental estrogens, are environmental contaminants that can mimic the biological activities of the female hormone estrogen in the endocrine system, i.e. they act as endocrine disrupters. Several substances are reported to have estrogen-like activity or estrogenic activity. These include steroid hormones, synthetic estrogens (xenoestrogens), environmental pollutants and phytoestrogens (plant estrogens).

Using the chromogenic substrate ortho-nitrophenyl-beta-D-galactopyranoside (ONPG) we show that an estrogen-sensitive yeast strain RMY/ER-ERE, with human estrogen receptor (hER alpha) gene and the *lacZ* gene which encodes the enzyme beta-galactosidase, is able to detect estrogenic activity in water samples over a wide range of spiked concentrations of the hormonal estrogen 17 beta-estradiol (E2). Orthonitrophenol, the yellow product of this assay can be detected using spectrophotometry but requires cell lysis to release the enzyme and allow product formation.

We improved this aspect in a fluorogenic assay by using fluorescein di-beta-D-galactopyranoside (FDG) as a substrate. The product was visualized using fluorescence microscopy without the need to kill, fix or lyse the cells. We show that in live yeast cells, the uptake of E2 and the subsequent production of beta-galactosidase enzyme occur quite rapidly, with maximum enzyme-catalyzed fluorescent product formation evident after about 30 minutes of exposure to E2. These findings are promising and may shorten the duration of environmental water screening and monitoring regimes using yeast-based estrogen assays, and the development of biosensors for environmental estrogens designed to complement quantification methods.

Reference:

Wozei, E., S. W. Hermanowicz, and H-Y. N. Holman. Developing a biosensor for estrogens in water samples: Study of the real-time response of live cells of the estrogen-sensitive yeast strain RMY/ER-ERE using fluorescence microscopy. Biosensors & Bioelectronics. In press.