Identification of Oestrone Sulphatase Inhibitors in Breast Cyst Fluid

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Introduction

Oestrogens play an important role in the development of breast cancer. Oestrone sulphate is a major source of active oestrogens in the breast, being converted to oestrone by oestrone sulphatase. Oestrone is then converted to oestradiol, a potent oestrogen strongly implicated in the development of breast cancer, by the enzyme oestradiol-17β hydroxysteroid dehydrogenase. *In vitro* oestrone sulphatase activity in breast tissue has been shown to be at least 1,000 times as high as aromatase activity, which converts androgens to oestrogens. Hence, local production of active oestrogens from oestrone sulphate is a very important source of oestrogens in breast tumours. We have previously found that breast cyst fluid is a potent inhibitor of oestrone sulphatase activity in certain breast cancer cell lines. Oestrone sulphatase inhibition is potentially of great importance in the treatment of women with hormone-dependent breast cancers. The aim of this study is to identify the substances present in breast cyst fluid, which are responsible for the oestrone sulphatase inhibitory property of breast cyst fluid.

Materials and Methods

Breast cyst fluid was obtained by needle aspiration of palpable breast cysts from women attending the Breast Clinic at University Hospital, Kuala Lumpur, under the care of Professor Dr. Yip Cheng Har. The samples were centrifuged and the supernatant stored at –20°C until assayed. High performance liquid chromatography was performed to separate substances according to molecular weight. The fractions collected were tested on the oestrogen-receptor positive MCF-7 and oestrogen-receptor negative MDA-MB-231 human breast cancer cell lines to see which fractions inhibited oestrone sulphatase activity. The procedure for MCF-7 and MDA-MB-231 cell culture as well as the oestrone sulphatase assay has been described (Erbas et al. 1996).

Results and Discussion

In the present study 10 of 19 breast cyst fluid samples inhibited oestrone sulphatase activity in the oestrogen-receptor positive MCF-7 breast cancer cell line whereas 19 of 21 samples inhibited oestrone sulphatase activity in the orstrogen-receptor negative MDA-MB-231 breast cancer cell line. All samples of breast cyst fluid inhibited growth of both MCF-7 and MDA-MB-231 breast cancer cell lines. This is in contrast to the study which we previously conducted on British women where the majority of breast cyst fluid samples inhibited oestrone sulphatase activity in the MCF-7 breast cancer cell line but stimulated oestrone sulphatase activity in the MDA-MB-231 breast cancer cell line. This observed difference may be due to the differences in the hormonal constituents of breast cyst fluid between British women and Malaysian women. Malaysian women tended to have much smaller breast cysts than their British counterparts. Fractionation of breast cyst fluid by high performance liquid chromatography using a column which separates proteins according to molecular weight yielded 2 fractions corresponding to molecular weights of 28 kDa and >158 kDa which inhibited both MDA-MB-231 breast cancer cell growth and oestrone sulphatase activity. These two substances are of particular interest since they may have roles to play in the prevention and treatment of breast cancer.

Conclusions

The 28 kDa and >158 kDa substances present in breast cyst fluid which inhibited oestrone sulphatase activity in the MDA-MB-231 human breast cancer cell line may have roles to play in preventing breast cancer development as well as the treatment of breast cancer. Further work is required to fully characterize both of these substances.

Benefits from the study

The study accounted for publications in national and international peer-reviewed scientific journals and presentations at national and international congresses. It also provided collaborative linkages between Universiti Putra Malaysia, Universiti Malaya and University of Newcastle upon Tyne, UK.

The study also made it possible to learn new techniques such as cell culture and protein separation techniques.

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