Proteomic Evaluation of Mammary Glands of Rats Exposed Prepubertally to Bisphenol A

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Exposure to environmental chemicals that produce alterations in hormonal regulation during important periods of development is important in analyzing susceptibility to cancer. One such chemical, bisphenol A (BPA), is commonly used in the production of polycarbonate plastic, epoxy resins, and dental sealants. BPA has been shown to advance puberty and disrupt estrous cyclicity, identifying it as an endocrine disruptor. Our goal is to identify differentially expressed proteins in mammary glands of rats that are treated early in postnatal life with BPA. Lactating dams were gavaged with 250 µg BPA/kg body weight or an equivalent volume of sesame oil on days 1-20. On days 21, 50 and 100, female offspring were euthanized and the 4th abdominal mammary glands were dissected and frozen in liquid nitrogen for later use. Mammary glands were homogenized and subjected to 2-D gel electrophoresis. Gels were analyzed using *Progenesis Discovery*. Protein spots that were common to all gels were statistically evaluated to identify significantly regulated spots. Using MALDI-TOF mass spectrometry, we have identified 21-day differential regulation of crystalline beta-B2, heat shock 70kD protein 5 (GRP78) and RAD51 homolog 2 in mammary glands of rats exposed to BPA as compared to control treated rats. Immuno-blot analysis confirmed the down-regulation of GRP78 and crystalline beta-B2, and the up-regulation of RAD51. RAD51, a protein of specific interest in carcinogenesis, functions in DNA maintenance and participates in a common DNA damage response pathway associated with double-strand DNA repair. Immuno-blot analysis has confirmed down-regulation of RAD51 in 50-day old treated rats, an observation that emphasizes BPA's potential to alter the cell's ability to effectively respond to DNA damage. Proteomic analysis of 50-day glands revealed differential regulation of A-kinase anchor protein 4, alpha-1-antitrypsin, ATP synthase beta subunit, and apolipoprotein A-I. Future directions include immuno-blot confirmation of 50-day protein identifications, as well as immuno-blot analysis of proteins commonly linked to RAD51 in the DNA damage response pathway. A comparison of differentially regulated biomarkers from 21 and 50 day animals can yield useful information on developmental changes and protein-protein interaction in detailed biochemical pathways related to BPA exposure. Specifically, we should be able to construct a virtual pathway outlining BPA's possible mechanism of action in the mammary gland. Supported by NIEHS 1U01 ES012771-03