signaling [5]. Also, it further supports that increased *ESR1* to centromere 6 copy number ratios (indicating focal amplifications), determined by use of FISH, might be predictive for response to antiestrogen treatment with tamoxifen in early breast cancer [6].

Analysis of large genomic data by TCGA, functional cell line studies, as well as data from massive parallel sequencing with high coverage, suggest the significance of *ESR1* as a target of gene amplification. Nevertheless, there are other mechanisms that drive ERα overexpression [12], and the deregulated metabolism of tumors might promote increased expression of ERα for various reasons.

In conclusion, data from copy number analysis and the functional evidence of this genetic alteration beg the question of whether *ESR1* amplification might be a marker for the addiction of a tumor to the activity of *ESR1* in ERα-positive tumors, independent of the significance to predict increased ERα-protein expression in general. As such, *ESR1* amplification might be considered a marker for the Achilles' heel of the tumor; the dependence on ERα signaling. This could be important for tumor response to antiestrogen therapy and might have therapeutic implications.

Conflict of interest

Frederik Holst has royalty interest associated with intellectual property of ZytoVision GmbH concerning patent US8101352B2 "Detection of ESR1 Amplification in Breast Cancer" and according EU patent application.

Resources

[i] http://cancergenome.nih.gov/

[ii] www.broadinstitute.org/tcga/gistic/browseGisticByGene

[iii] https://gdac.broadinstitute.org

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http://dx.doi.org/10.1016/j.tem.2016.08.002

References

1. Thomas, C. and Gustafsson, J.A. (2015) Estrogen receptor mutations and functional consequences for breast cancer. *Trends Endocrinol. Metab.* 26, 467–476
2. Mermel, C.H. *et al.* (2011) GISTIC2.0 facilitates sensitive and confident localization of the targets of focal somatic copy-number alteration in human cancers. *Genome Biol.* 12, R41
3. Holst, F. (2016) Estrogen receptor alpha gene amplification in breast cancer: 25 years of debate. *World J. Clin. Oncol.* 7, 160–173
4. Beroukhim, R. *et al.* (2010) The landscape of somatic copy-number alteration across human cancers. *Nature* 463, 899–905
5. Desmedt, C. *et al.* (2016) Genomic characterization of primary invasive lobular breast cancer. *J. Clin. Oncol.* 34, 1872–1881
6. Holst, F. *et al.* (2007) Estrogen receptor alpha (ESR1) gene amplification is frequent in breast cancer. *Nat. Genet.* 39, 655–660
7. Aguilar, H. *et al.* (2010) Biological reprogramming in acquired resistance to endocrine therapy of breast cancer. *Oncogene* 29, 6071–6083
8. Li, S. *et al.* (2013) Endocrine-therapy-resistant ESR1 variants revealed by genomic characterization of breast-cancer-derived xenografts. *Cell Rep.* 4, 1116–1130
9. Li, J. *et al.* (2013) TCPA: a resource for cancer functional proteomics data. *Nat. Methods* 10, 1046–1047
10. Pruitt, K.D. *et al.* (2007) NCBI reference sequences (RefSeq): a curated non-redundant sequence database of genomes, transcripts and proteins. *Nucleic. Acids Res.* 35, D61–D65
11. Kok, M. *et al.* (2009) Estrogen receptor-alpha phosphorylation at serine-118 and tamoxifen response in breast cancer. *J. Natl. Cancer Inst.* 101, 1725–1729
12. Thomas, C. and Gustafsson, J.A. (2011) The different roles of ER subtypes in cancer biology and therapy. *Nat. Rev. Cancer* 11, 597–608

Forum

Estrogen and Microbiota Crosstalk: Should We Pay Attention?

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Recent advances have suggested that steroid hormones such as estrogens, and gut microbiota might synergize to influence obesity, diabetes, and cancer. We discuss recent knowledge of the interactions between estrogens and gut microbiota, and new insights that might offer new approaches to influence this crosstalk and improve metabolic outcomes.

Introduction

Estrogens are steroid hormones that bind to and activate their cognate receptors, ERα and ERβ. In addition to their roles in development and maintenance of reproductive tissues, estrogens have many beneficial effects in diverse, nonreproductive target tissues, such as liver, adipose, skeletal muscle, bone, and vasculature. Sedentary lifestyle, restricted mobility, and western-style, fat- and sugar-rich, diets, promote obesity, and when combined with low estrogen levels in postmenopausal women, make postmenopausal women more susceptible to weight gain, fat redistribution to abdominal areas, dyslipidemia, hypertension, and insulin resistance; the major hallmarks of metabolic syndrome [1].

The microbiota consists of a large population of microorganisms including fungi, viruses, and bacteria, present in and on the body. One of the largest collections of these microorganisms is found in the gastrointestinal tract where they act symbiotically with the human body to ferment dietary fibers, aid in digestion and absorption, and protect against pathogens [2]. The diversity of this host–microbe interaction allows it to be involved in a wide range of pathways that affect human health, disease, and aging [2]. Emerging evidence suggests an interaction between estrogens and the microbiome that might affect metabolic rate, body weight, and adiposity. In this article, we discuss current research linking obesity, estrogens, and the microbiome with a focus on gut bacterial communities. Furthermore, we propose prospective and future research to improve the metabolic health in the face of

reduced estrogen, as seen in postmenopausal women.

Interaction of Microbiota and Estrogens

Certain microbiota compositions have been suggested to ameliorate symptoms associated with obesity. For example, insulin sensitivity in humans was improved by transferring gut microbiota from lean donors to those with metabolic syndrome [3]. Manipulating the gut microbiota composition with probiotics and prebiotics may also improve weight gain and fat deposition. Experiments using mice fed a high-fat diet (HFD) showed improved glucose–insulin homeostasis and attenuation of weight gain when mice were given *Lactobacillus* and *Bifidobacterium* probiotics, compared to mice fed a normal diet [4].

Estrogens and the microbiota play major roles in obesity independent of each other. However, recent evidence suggests that estrogens and the microbiota may act together to regulate weight gain and lipid deposition. In a reciprocal relationship, microbiota can metabolize estrogen-like compounds we consume to biologically active forms, and estrogen-like compounds may promote the proliferation and growth of certain types of bacteria [5]. For example, daidzein, an isoflavonoid enriched in soy foods is metabolized by gut bacteria to O-desmethylangolensin (ODMA) and S-equol [5], which are structurally similar to estrogen, and which can activate ERα or ERβ [6,7]. Additionally soy isoflavones such as, genistein, and glycitin can significantly alter the structure and the composition of the fecal bacterial community in postmenopausal women by increasing the concentration of the beneficial Gram-positive *Bifidobacterium*, while suppressing Clostridiaceae, which can be the cause of several human diseases [5,6]. Diets that contain more phytoestrogens have been shown to improve weight gain, and high levels of gut microbial metabolites of estrogenic plant lignans in the blood are associated with a lower rate of being overweight and obese [7]. Individuals who do not harbor bacteria that can produce ODMA, for example, are more likely to be obese than those who are producers [5]. Furthermore, > 50% of obese individuals are equol non-producers [6]. Equol may aid in glycemic control and lower low-density lipoprotein levels, demonstrating the crucial role bacteria have on the activation of soy phytoestrogens, in relation to obesity [7]. Thus, phytoestrogen metabolites may have a role in preventing aspects of metabolic syndrome. Recent studies have shown that supplementation of the chalconoid isoliquiritigenin, a low-affinity ER ligand found in licorice root, improves ovariectomy-associated weight gain and metabolic syndrome in mice, without stimulating reproductive tissues [8]. It is currently unclear how supplementation of isoliquiritigenin or any other low-affinity estrogens would alter gut microbiota and blood metabolites (Figure 1).

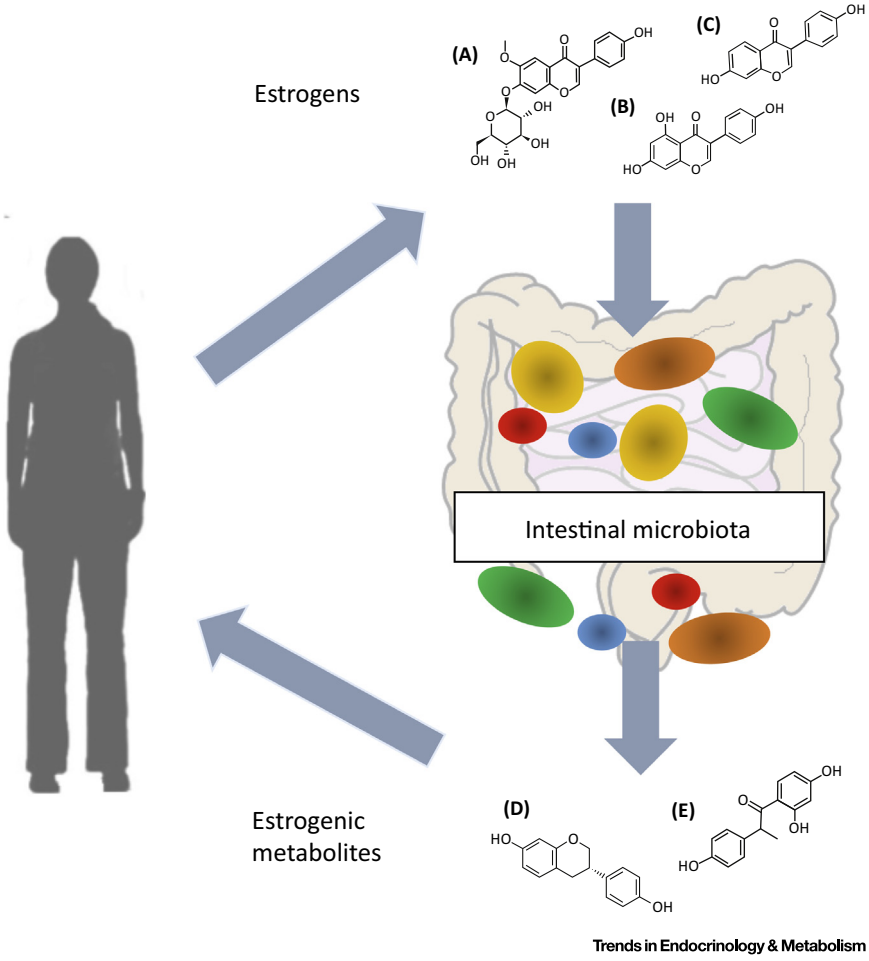

Figure 1. Potential Microbiome and Estrogen Interaction. Estrogens inherent in the body or from the diet may be metabolized by gut microbes and produce estrogenic metabolites that affect host metabolism and reduce disease risk. (A) Glycitin, (B) genistein, (C) daidzein, (D) equol, (E) O-desmethylangolensin.

More recently, intestinal microbiota genes that are capable of producing estrogen-metabolizing enzymes have been identified. These sets of genes, or the 'estrobolome', implies a potential for biomarker applications in several metabolic diseases [9]. Estrogens in the body are absorbed by the liver through first-pass metabolism, and biotransformed through methylation, hydroxylation, and conjugation into metabolites of varying potency for ERs.