

Katharina Buck  
Dr. sc. hum.

## **Influence of phytoestrogens on prognosis of postmenopausal breast cancer**

Promotionsfach: DKFZ (Deutsches Krebsforschungszentrum)  
Doktormutter: Frau Prof. Dr. sc. hum. Jenny Chang-Claude

Breast cancer is the most frequent cancer and the primary cause of cancer death in German women. Incidence has increased yet mortality has decreased since the mid 1990's, attributable to early detection and better treatment options, resulting in a growing number of breast cancer survivors. Albeit a number of diagnostic and clinical factors are known to influence recurrence and survival after a primary breast cancer diagnosis, even in women with similar presenting diagnoses and conventional therapy, there is considerable variation in recurrence and survivorship. Thus, there may be important factors that influence these differing prognoses, such as lifestyle and genetic factors. Only few studies have considered these aspects comprehensively. Phytoestrogens are plant-derived estrogenic substances present in various foods. In Western populations with a low intake of isoflavones (present in soy foods), phytoestrogen intake is predominantly derived from intake of dietary plant lignans (present in plant foods such as vegetables, grains and fruits). After consumption, these plant compounds are metabolized to enterolignans (enterolactone and enterodiols) and become bioavailable.

Due to their structural similarities with human estrogens, phytoestrogens may bind to estrogen receptors and thereby exert competitive effects to endogenous estrogen. Also, they may exert hormone-independent anti-carcinogenic actions such as stimulation of apoptosis, inhibition of angiogenesis and tumor metastasis.

A literature search and meta-analysis was performed, to summarize the current knowledge on lignans and breast cancer. These assessments focused primarily on breast cancer risk studies because only a limited number of prognostic studies are available. In summary, lignans were not significantly inversely associated with overall breast cancer risk. However, high lignan exposure, particularly high plant lignan intake, was associated with a significant risk reduction in postmenopausal women (pooled estimate=0.86, 95% confidence interval (CI)=0.78-0.94). It remains unclear whether the effect of lignans on breast cancer risk differs by ER status of the tumor. However, the results propose an effect of lignans on breast cancer that might also influence postmenopausal breast cancer prognosis in Western populations, which may depend on ER status of the tumor.

Therefore, we assessed the association of dietary phytoestrogen intake and serum levels of phytoestrogens with postmenopausal breast cancer prognosis. Differential effects by tumor characteristics, adjuvant endocrine therapy use, menopausal hormone therapy use, and genetic variants in estrogen pathway and apoptosis genes were evaluated for an effect modification of phytoestrogens on breast cancer prognosis.

Data from a multi-center prospective breast cancer patient cohort study was used to assess the association of phytoestrogens (derived from dietary intake and measured in serum) with prognosis of postmenopausal breast cancer. Patients aged 50-74 years and diagnosed with a primary breast cancer were recruited between 2001 and 2005 in two German study regions Hamburg and Rhine-Neckar-Karlsruhe (RNK). For all patients, vital status through the end of 2009 was ascertained via local population registries and deaths were verified by death

certificates. Information on recurrences and secondary tumors was verified by clinical records/treating physicians.

Pre-diagnostic dietary lignan intake was assessed using a database assembled from literature searches, whereas the estimation of enterolignans was conducted using data on in vitro incubation of various foods with human feces. Furthermore, information on lignan- and isoflavone-rich food intake was collected. Complete data was available for 2,653 postmenopausal breast cancer patients from both study regions.

Post-diagnostic serum enterolactone (a major lignan metabolite) and genistein (a major isoflavone) levels were measured using time-resolved fluoroimmunoassays for 1,140 and 1,132 patients from the RNK study region, respectively.

Hazard ratios (HR) and 95% CIs for dietary lignans, estimated enterolignans, phytoestrogen-rich foods, and dietary fiber as well as measured enterolactone and genistein levels in relation to overall survival (OS), breast cancer-specific survival (BCSS), and/or distant disease-free survival (DDFS) were calculated using Cox proportional hazards models stratified by age at diagnosis and adjusted for prognostic/confounding factors.

For the dietary intake assessments, median follow-up time of patients was 6.4 years, and 321 patients died, 235 with breast cancer. High levels of enterolactone and enterodiol were associated with significantly reduced overall mortality (for the highest quintile, HR=0.58, 95% CI=0.39-0.86,  $p_{Trend}=0.01$  and HR=0.64, 95% CI=0.43-0.96,  $p_{Trend}=0.02$ , respectively), which was stronger for patients with low fiber intake (HR=0.32, 95% CI=0.10-1.07,  $p_{Trend}=0.03$  and HR=0.48, 95% CI=0.21-1.07,  $p_{Trend}=0.01$ , respectively). High dietary fiber intake was also associated with a significantly reduced mortality. Dietary lignans and phytoestrogen-rich foods were not associated with OS and BCSS. There was no effect modification by estrogen receptor (ER) status, adjuvant hormone therapy use, tumor size and grade, as well as menopausal hormone therapy use.

During a median of 6.1 years follow-up after diagnosis, 162 total deaths were confirmed in the patients with serum enterolactone measurements. Higher serum enterolactone levels were associated with significantly reduced HRs for overall mortality (HR for the highest quartile=0.58, 95% CI=0.34-0.99,  $p_{Trend}=0.04$ ). Serum enterolactone was also associated with a similarly reduced yet non-significant HR for distant disease (HR=0.62 for the highest quartile, 95% CI=0.35-1.09,  $p_{Trend}=0.08$ ). Although there was no significant heterogeneity by ER status ( $p=0.09$ ), the highest quartile of serum enterolactone was associated with a significantly better OS only for ER-negative tumors (HR=0.27, 95% CI=0.08-0.87). No association was observed for serum genistein.

A meta-analysis of four studies (including this thesis' results) substantiates that lignans and enterolignans may be associated with a better prognosis of postmenopausal breast cancer patients (pooled estimate=0.56, 95% CI=0.39-0.74 for high compared to low lignan exposure).

No effect modification by genetic variants in estrogen pathway and apoptosis genes was observed for the association between serum enterolactone levels and survival after breast cancer. The sample size of this study was small and thus the findings need replication in sufficiently powered studies.

In summary, the present findings provide evidence for an association of lignans but not isoflavones with prognosis of postmenopausal breast cancer in Western populations. Although no significant effect heterogeneity was observed, enterolignans were more strongly associated with a better survival in patients with estrogen receptor-negative tumors. Further studies are needed to confirm our results and to elucidate the mechanisms involved in hormone-dependent and independent cancer. However, if these results are confirmed by other studies, they have potential implications for clinical application in the prevention of breast cancer and in dietary recommendations for postmenopausal breast cancer patients.