moxifen treatment significantly improves disease-free survival. However, the improvements are small in absolute terms and the effects on breast cancer and overall mortality, and the long-term toxicity of AI treatment remain unclear. We undertook a meta-analysis of the studies of AIs to clarify the risks and benefits.

Results: AIs reduced the absolute risk of breast cancer recurrence by 2.5% (NNT 40; confidence interval 33–100; $p < 0.0001$), breast cancer death by 1.0% (NNT 110; 65–200; $p < 0.001$) and all-cause mortality by 0.7% (NNT 140; 70–700; $p < 0.05$) compared to tamoxifen. Gynaecological symptoms were fewer with AIs but osteoporosis, hypercholesterolaemia, arthralgia and diarrhoea were increased.

Interpretation: Using AIs instead of tamoxifen improves survival of post-menopausal women with ER-positive breast cancer over the first few years of treatment. However, the life years gained from AI use depend on whether the short-term benefits persist and are less for lower risk and older women. Cost/QALY appears high and switching from tamoxifen to aromatase inhibitors as first line treatment (which would cost about £70 million per year in the UK and about £1.5 billion per year worldwide) does not seem justified on current evidence. Uncertainty remains about the relative benefits and risks of aromatase inhibitors compared to tamoxifen, how the relative benefits vary over time and by background risk, whether a combination of tamoxifen and AIs is better than either treatment alone and on how long AI treatment should continue.

O-113. Aromatase inhibitors upfront: the switch of 2–5 years or extended adjuvant – how do we choose?

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Background: The aim of the study was to identify post-menopausal patients with ER+ disease who have relapsed while taking tamoxifen in the first 2–3 years and over 5 years to identify groups of patients who should be treated either with up front AIs between 2 and 5 years, switched after 2–3 years or who should have extended adjuvant hormonal treatment.

Patients: 670 post-menopausal women with ER+ disease who were given 5 years of adjuvant tamoxifen were identified. Of these 121 have relapsed. An analysis looking for risk factors for relapse <2.5 years, 2.5–5 years and 5 years was performed.

Results: Women >70 years of age, those with ER poor tumours and those which were grade 3 or had 4 or more nodes involved were at the highest risk of relapse in the first $2\frac{1}{2}$ years. These patients should be considered for immediate treatment with AIs. From 2–5 years the rate of relapse was still high for patients with ER poor turnouts, women with grade 3 tumours and multiple node involvement. The only group who did not have a significant relapse rate in the first 5 years was patients with grade 1 cancers. Beyond 5 years, only grade and number of lymph nodes involved predicted for recurrence, such that patients with grade 2 tumours had a higher rate of recurrence than grade 1 or grade 3, and risk increased as number of involved nodes increased.

Conclusion: The study identified groups of high risk post-menopausal women with ER+ breast cancers who should be considered for immediate AIs. Thereafter analysis of relapse indicates that all other patients should be switched to an AI after 2–3 years of tamoxifen. We have also identified which women benefit from extended adjuvant therapy.

O-114. Letrozole and Anastrozole: a pre-operative study of their effects on ER positive breast cancers in postmenopausal women

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Background: Letrozole appears to be a more potent inhibitor of oestrogen synthesis than anastrozole. Biological changes occur within 14 days of starting treatment and it is the aim of this study to investigate the changes within the first 14 days of treatment with anastrozole or letrozole.

Patients and Methods: 206 patients with 209 ER positive breast cancers (3 bilateral) were randomly allocated to receive either 2.5 mg of letrozole or 1 mg of anastrozole daily for 14 days prior to surgery. Proliferation ER, PR and Her2 were measured

Results: ER and PR: After letrozole and anastrozole treatment, there was a significant but small fall in ER (0.32, 0.20–0.44) $p < 0.0001$, and a much larger fall in PR 2.54 (2.20–2.89) $p < 0.0001$. More cases showed a reduction in PgR expression following letrozole than with anastrozole.

Proliferation: Both letrozole and anastrozole significantly reduced proliferation. Reductions in proliferation were higher in ER rich cancers, Allred 6–8 than Allred poor cancers 2–5 $p = 0.009$. There are no significant differences between the 2 drugs. Her2+ cancers had a higher initial proliferation than Her2− cancers $p < 0.003$. Both letrozole and anastrozole produced significant falls in proliferation with no quantitive differences between Her2+ and Her2− cancers. Change in PgR expression after treatment was also similar in Her2+ and Her2− groups.

Conclusion: 14 days of letrozole or anastrozole produced significant falls in proliferation and PR expression. Her2+ cancers had a higher rate of proliferation greater than Her2− cancers. Both letrozole and anastrozole produced a similar magnitude of reduction and proliferation in both Her2+ and Her2− cancers. Reduction in proliferation was greater in ER low tumours.

O-115. Is there an optimal duration of neoadjuvant letrozole therapy?

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Background: Randomised studies of neoadjuvant aromatase inhibitors have treated patients for 3–4 months. The aim of this review was to assess whether tumours continue to respond to neoadjuvant letrozole for periods longer than 3–4 months.

Patients and Methods: 142 postmenopausal women with large operable or locally advanced ER rich (ER Allred score

6 or more) breast cancer were enrolled into a prospective audit assessing response to neoadjuvant letrozole 2.5 mg per day. Clinical response was assessed at 3 months; non responders and patients whose tumours had become operable or had responded sufficiently to allow breast conserving surgery proceeded to surgery. The remaining 42 patients who were either unfit for surgery, refused surgery, had responded but still required mastectomy or were inoperable, continued letrozole for a further 3 months. 22 patients continued letrozole for a total of 12 months. Reductions in tumour volume over the first 3 months were compared with 3–6 and a period of between 6 and 12 months were calculated.

Results: Median % reduction in the tumour volumes from 0–3 months, 3–6 months and 6–12 months are shown in the table.

	Number of Patients	Median	95% CI
% reduction from 0–3 months	42	52	37–62
% reduction from 3–6 months	42	57	26–100
% reduction from 6–12 months	22	66	22–100

Tumours continued to reduce in volume during the 12 months study period.

Complete responses: At 3 months there were 4/42 (9.5%) complete responses, by 6 months there were 12/42 (29%) and by 12 months 8/22 (36%). One patient who was responding at 3 months had disease progression at 12 months. Conclusion: Neoadjuvant letrozole produces ongoing tumour shrinkage in postmenopausal women over 12 months in large operable or locally advanced ER+ breast cancers. Patients whose tumours are responding to letrozole at 3 months can expect further reduction in tumour volume with continued treatment. There is no optimum duration for use of neoadjuvant letrozole; it can be used safely for up to 12 months.

O-116. A molecular analysis of the relationship between ERα and ERβ in primary breast cancer

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Endocrine status plays a crucial role in the diagnosis, treatment and prognostication of breast cancer patients. Our primary aim was to quantitate and compare the levels of ERα and ERβ mRNA expression in malignant and benign breast specimens and to ascertain any association between mRNA levels, prognostic indicators and patient outcome. We also analysed the mRNA expression of mammaglobin, a putative breast cell-specific marker, and determined if any relationship lay between its expression and that of the breast cancer associated hormone receptors.

2% of tumours did not express ERα mRNA and 11% did not express ERβ mRNA. (6% of tumours proved to be mammaglobin negative). No significant difference was evident between the benign and tumour mRNA levels for any gene. Spearman's correlation tests showed that in the tumour group ERα mRNA levels positively correlated with ERβ mRNA levels for both pre ($p = 0.002$) and post ($p = 0.001$) menopausal patients. It was also demonstrated that elevated ERα mRNA levels were associated with high mammaglobin mRNA levels in both benign ($p = 0.025$) and tumour tissue ($p = 0.035$). Kaplan-Meier survival analysis did not report any significant association between ER mRNA levels and disease free survival. However, both 5 and 10 year overall survival was reduced in premenopausal patients expressing below median levels of ERβ.

mRNA quantitation is a more sensitive approach to identifying the hormonal mechanisms of breast cells than nuclear immunostaining. Demonstrating an association between ERα and ERβ could be of relevance in that high levels of ERα are accompanied by high levels of ERβ, suggesting the existence of molecular crosstalk between these two markers potentially resulting in either an enhanced or diminished response to therapy. The correlation observed between ERα and mammaglobin may serve to further characterize breast cancer cells, identifying them by mammaglobin expression and determining their hormonal status by ERα analysis.

O-117. The influence of tumour grade on DCIS stem cell growth

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A model has been suggested in which transformation of stem cells or early progenitor cells may result in carcinogenesis and recent studies have described cancer stem cells in breast cancer. To determine the factors controlling DCIS stem cell (mammosphere-MS) growth we have used a non adherent culture system producing stem and progenitor cells in an undifferentiated state to generate cultures with self renewal ability and also 3D culture in matrigel to recapitulate DCIS *in vitro*.

Single cell suspensions of mechanically and enzymatically dissociated DCIS samples were seeded at 10,000 cells/ml in non-adherent plates. Twelve out of 16 DCIS samples from surgery produced DCIS MS measuring >60 μm within 3 days, whereas normal mammary epithelial MS formed after 5–10 days, indicating greater proliferation rate. A 5 fold greater MS formation with DCIS was seen compared to normal tissue. MS immunostained for differentiated luminal (CK18) myoepithelial (CK14) markers and ErbB2 which corresponded to the original DCIS tissue.

Percentage MS formation was significantly greater in the high grade (1.9±0.2, %±SEM) MS than low grade (1.2±0.1%, $p = 0.045$). Removal of EGF significantly decreased low grade ($p = 0.002$) and but not high grade MS, although Iressa, an EGFR tyrosine inhibitor significantly ($p = 0.01$) inhibited high grade DCIS MS formation. Single cells from dissociated DCIS MS have been grown as 3D cultures in matrigel which recapitulate DCIS with solid acini structures containing both myo and luminal epithelial cell lineages.

This is the first report of DCIS stem cell culture and the technique allows investigation of stem cell signalling pathways which are critical to growth of DCIS *in vivo*.

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