Impact of age, tumor characteristics, and treatment on local control and disease outcome in early stage breat cancer : an EORTC translational research project

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Part III

CHAPTER 8

Impact of established prognostic factors in early stage breast cancer in very young breast cancer patients; a translational research project using pooled datasets derived from 4 EORTC Breast Group Trials

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Abstract

Young age at time of diagnosis of breast cancer is an independent prognostic factor associated with unfavorable outcome in terms of survival and locoregional control. This has led to the general recommendation to administer adjuvant systemic chemotherapy to patients aged 35 years or less at time of diagnosis regardless of other tumor characteristics like tumor size and axillary lymph node status.
However, since breast cancer at a very young age, i.e. < 41 years is a relative rare event, evidence concerning prognostic factors within this subgroup of patients is lacking. Therefore the data of four EORTC Breast Group Trials concerning primary operable breast cancer were combined to study prognostic factors on long term outcome in very young breast cancer patients. The total dataset consisted of 9938 early breast cancer patients. Tumor material was collected from 549 patients aged under 41 years of age at time of diagnosis. In the multivariate analyses, only histological grade remained a significant prognostic factor for both overall survival (Grade II HR 2.67; 95% CI 0.91 to 7.80; P = 0.07, Grade III HR 3.92; 95%CI 1.38 to 11.16; P = 0.01) and distant metastasis free survival (Grade II HR 2.04; 95% CI 1.07 to 3.88; P = 0.03, Grade III HR 2.38; 95%CI 1.29 to 4.39; P < 0.01). However, large tumor size remained an independent unfavorable prognostic factor on outcome in terms of distant metastasis free survival as well (HR 1.64 (1.17-2.31) P < 0.01). In the subgroup of node negative very young breast cancer patients, histological grade remained an independent prognostic factor for both overall survival (Grade III HR 8.92; 95%CI 1.17 to 68.20; P = 0.04) and distant disease-free survival respectively (Grade III HR 4.12; 95%CI 1.42 to 11.98; P < 0.001). Histological grade is a strong independent prognostic factor, even in young breast cancer patients. These findings support the fact that histological grade is an excellent diagnostic tool to assess disease outcome in this specific subset of very young breast cancer patients.

Introduction

The incidence of early stage breast cancer in very young women is increasing. At present breast cancer at young age, i.e. under age 35, does account for approximately 5% of the total number of cases diagnosed each year in the US.
Based upon multiple retrospective analyses demonstrating the independent unfavorable prognostic impact of young age on prognosis in breast cancer, current consensus guidelines have included young age (< 35) as an absolute indication for adjuvant systemic chemotherapy after primary removal of the tumor irrespective of other tumor characteristics [1-4]. Such guidelines imply that young patients with favorable tumor features such as small tumor size and a negative axillary nodal status will receive chemotherapy as well although absolute treatment benefits for these patients are not well known which is the result of the fact that breast cancer at very young age remains a relatively infrequent event.
Retrospective analyses have demonstrated breast cancer at a very young age to be associated with higher grade, ER negative tumors and later stage disease at time of diagnosis [5,6].
However, other yet unknown factors may be responsible for the poorer outcome in this subset of patients and this hypothesis is emphasized by the fact that BRCA I and II mutation carriers only account for 10% in this population [7-9].

Therefore, two questions remain still very much open for discussion to date. First, do all very young breast cancer patients require adjuvant systemic chemotherapy, and second, by which means can subsets of patients within this group of very young women be identified who have an excellent or poor prognosis.
To study these questions we pooled the data of four randomized trials conducted by the EORTC Breast Cancer Group and the EORTC radiotherapy Group and collected tumor material of patients under age 41 who participated in one of these trials.

Patients and Methods

The data used in this study was obtained from 4 randomized phase III EORTC trials that included patients with early stage breast cancer. Two trials randomized between two types of locoregional therapy whereas two trials randomized between different timing of the same type of systemic therapy. The detailed features of these trials have been described in detail previously (ref). In summary, the trial protocols are listed below:

EORTC trial 10801 (1980-1986, median follow up 13.4 years) was conducted in order to assess the safety of breast conserving treatment. In this trial, patients were randomized between breast conserving surgery combined with radiotherapy and radical mastectomy. Six cycles of adjuvant chemotherapy with cyclophosphamide 100 mg/m^2 given orally on days 1-14, methotrexate 40mg/m^2 given intravenously on days 1 and 8, and 5-fluorouracil 600 mg/m^2 given intravenously on days 1 and 8, were indicated for all patients under the age of 55 with positive nodes. No information was collected on hormonal therapy. In this study, 902 patients were randomized [10].

EORTC trial 10854 (1986-1991, median follow up 10.8 years) studied the question whether one course of peri-operative chemotherapy given directly after surgery yields better results in terms of treatment outcome than surgery alone. Peri-operative chemotherapy consisted of one single course of doxorubicin 50 mg/m^2, 5-fluorouracil 600 mg/m^2, and cyclophosphamide 600 mg/m^2 (FAC), administered intravenously within 36 hours after surgery. Axillary lymph node-positive premenopausal patients in the peri-operative chemotherapy group were recommended to receive an extra 5 cycles of cyclophosphamide, methotrexate, and 5-fluorouracil (CMF). Node-positive patients, younger than 50 years, who did not receive peri-operative chemotherapy, were advised to give one conventional course of FAC followed by five cycles of CMF after surgery. Patients were stratified for breast conserving therapy and modified radical mastectomy. Prolonged adjuvant systemic treatment was left to the discretion of the local investigators. 2795 patients were included in this trial [11].

EORTC trial 10902 (1991-1999, median follow up 6.1 years) was set up to determine the value of pre-operative chemotherapy. Patients were randomized to receive four cycles of chemotherapy either before or after surgery. Chemotherapy consisted of four cycles of 5-fluorouracil 600 mg/m^2, epirubicin 60 mg/m^2, and cyclophosphamide 600 mg/m^2 (FEC) administered intravenously, at 3-weekly intervals. In the pre-operative chemotherapy group, surgical therapy followed within four weeks of the fourth course of chemotherapy. In the postoperative chemotherapy group, the first

	Patient characteristics (N = 9938)									
	10801		10854		10902		22881		Total	
	No.	%	No.	%	No.	%	No.	%	No.	%
Age (median 53.6 years)										
≤ 40 years	113	13	396	14	125	18	558	10	1192	12
≤ 30 years	14	2	34	1	18	3	44	1	*110*	*1*
31-35 years	26	3	101	4	33	5	158	3	*318*	*3*
35-40 years	73	8	261	9	74	10	356	6	*763*	*8*
> 40 years	789	87	2399	86	573	82	4985	90	8746	88
Tumor size*										
T1	175	19	823	30	96	14	2868	52	3962	40
T2	725	81	1759	64	403	59	2662	48	5549	56
T3	-		166	6	188	27	13	0.2	367	4
Missing	2		47		11				60	
Nodal status**										
N-	514	59	1467	53	253	39	4351	79	6585	67
N+	361	41	1327	47	401	61	1192	21	3281	33
Missing	27		1		44				72	
ER status***										
Positive	-		1823	73	337	70	2930	73	5090	73
Negative	-		673	27	141	30	1092	27	1906	27
Missing	902		299		220		1521		2942	
Surgery										
Breast conserving	466	52	1544	56	198	30	5543	100	7751	78
Mastectomy	436	48	1235	44	475	70	-		2128	22
Missing			16		43				59	
Adjuvant CT****										
No	753	83	2227	82	-		4792	87	7772	79
Yes	149	17	502	18	698	100	699	13	2028	21
Missing			66				52		118	

* Clinical tumor size
** Pathological nodal status
*** EORTC trial 10801 did not report estrogen receptor status, all patients (N=902 have been deemed missing)
**** EORTC trial 10854 randomized between 1 course of peri-operative chemotherapy. One course of peri-operative chemotherapy was not deemed as prolonged chemotherapy

Table 1. All patients

cycle was given within 36 hours after surgery. Stratification was performed for planned type of surgery instead of performed type of surgery. This was done because of the expected effect of pre-operative chemotherapy on downstaging of the tumor. A total number of 698 patients were randomized [12].

EORTC trial 22881 (1989 – 1996, median follow up 5.1 years) studied the value of a boost dose after primary breast conserving surgery. Patients with breast cancer of clinical stage T1–2,N0–1,M0 were eligible for the trial. Patients with stage I or II breast cancer who had undergone macroscopically complete surgical removal of the tumor and axillary dissection were randomly assigned to undergo 50-Gy irradiation of the whole breast with or without an additional dose of 16 Gy to the tumor bed. Patients with a microscopically incomplete excision were assigned to receive booster doses of 10 or 26 Gy. Patients with axillary lymph-node involvement received adjuvant systemic therapy: premenopausal patients received chemotherapy, and postmenopausal patients received tamoxifen. Patients not given adjuvant chemotherapy began radiotherapy within nine weeks after lumpectomy. For patients who received adjuvant chemotherapy, a delay of up to six months before irradiation was allowed. This study enrolled 5569 patients [13].
In all trials if adjuvant chemotherapy was indicated, patients either received CMF or an anthracyclin-based regimen (FAC or FEC). Adjuvant hormonal therapy for premenopausal ER or PgR positive patients was not yet recommended at the time when these trials were conducted. No information concerning estrogen receptor status and tamoxifen use was available for patients who participated in EORTC trial 10801. In the trials where tamoxifen use was recorded, less than 5% of patients ≤ 41 years received tamoxifen.

Collection of tumor material and immunohistochemistry
A questionnaire was send to participating institutions to collect paraffin tumor specimens from all patients aged under 41 at time of diagnosis except for those who had participated in EORTC trial 10902 and received neoadjuvant chemotherapy. Tumor tissue was collected and processed for immunohistochemistry using a tissue microarray. Three core biopsies were taken from every tumor specimen and put in a so-called donor block. On average, one tissue array donor block consisted of three biopsies from sixty tumor specimens. This procedure has been described in debt by others previously [14-17].

Histological grading, scoring of the extent of intraductal carcinoma and lymph vessel invasion was performed on H&E colored slides according to Bloom and Richardson [18,19]. ER, PgR, Her2 and P53 expression levels were estimated by immuno-histochemistry. Detailed procedures have been described previously [20-22]. In summary, a tissue microarray slide was stained and scored counting the percentage of positive nuclei and taking the mean value of the three tumor biopsies. For estrogen- and progesterone receptor expression, Tumors with >10% of the tumor cells showing nuclear staining were considered positive. Tumor were deemed p53 positive if there was > 50% nuclear staining. Her2 expression was scored estimating the level of membranous staining. Strong membranous staining in > 10% of tumor cells was considered positive. Estimation of tumor grade and protein expression levels were scored by two investigators (MJ vd V & JA vd H) simultaneously who had to come to an agreement in case of different views.

Selection of endpoints
Since this study was set up to study the impact of potential prognostic factors in very young breast cancer patients on long term outcome, endpoints studied were overall survival and distant metastasis free survival. Survival time was defined as the time between randomization and death from any cause. Distant metastasis free survival time was defined as time to distant metastasis or death if the latter event occurred before a distant metastasis was diagnosed. Breast cancer specific survival was not included since exact information concerning the cause of death was lacking in three out of four trials.

Statistical analyses
All analyses were performed for overall survival and distant metastasis free survival. Apart from patient age, covariates included consisted of tumor-, and treatment related characteristics. Tumor characteristics were tumor size, nodal status, tumor grade, hormone receptor status, Her2 overexpression, p53 overexpression, and lymphangio invasion. Treatment characteristics consisted of type of surgery and the administration of chemotherapy. Tamoxifen use was not included because of the high rate of missing data for this covariant. Cox proportional-hazard regression models [23] were used to estimate hazard ratios with 95% confidence intervals. A 5 % significance level was used and all tests are two-sided. Survival analyses were performed using the Kaplan Meyer method [24].

Results

Patient characteristics
A total of 9938 early stage breast cancer patients participated in one of four trials. The majority of these patients, i.e. approximately 67%, were node negative. In addition, approximately 70% of the patients whose hormone receptor status was available had estrogen receptor positive breast cancer. Further patient characteristics are listed in Table I. 1192 patients were aged under 41 years at time of diagnosis. Paraffin embedded tumor material was successfully obtained and processed into a tissue micro array for 549 patients younger than 41 years. This subgroup of patients had

	Patient characteristics (N= 549)	
	Age (median 36.7 years)	
	No.	%
Tumor type		
Ductal	497	96
Lobular	17	3
Other	5	1
Missing	30	
Clinical Tumor size		
T1	219	40
T2	308	56
T3	20	4
Missing	2	
Pathological tumor size		
T1	333	67
T2 / T3	164	33
Missing	52	
Nodal status*		
N-	341	63
N+	204	37
Missing	4	
Tumor grade		
I	76	15
II	165	32
III	276	53
Missing	32	
Lymfangio invasion		
None	357	69
2-5 vessels	86	17
> 5 vessels	76	14
Missing	30	
ER status		
Positive	288	61
Negative	180	39
Missing	81	
PgR status		
Positive	223	48
Negative	241	52
Missing	85	
HER2 status		
Negative	349	74
Positive	121	26
Missing	79	
P53 status		
Negative	331	71
Positive	133	29
Missing	85	
Surgery		
Breast conserving therapy	446	81
Mastectomy	102	19
Missing	1	
Prolonged adjuvant chemotherapy		
No		
Yes	326	60
Missing	221	40
	2	

*Pathological nodal status

Table 2. Patients < 41 years

similar characteristics in terms of nodal status and hormone receptor status as compared to the whole group of patients. Patient characteristics of this subgroup are listed in Table 2. At time of the analysis, 1837 patients have died and 603 patients developed distant disease and were still alive. The median follow up period was 7 years.

Young patients versus older patients

Univariate prognostic factor analyses were performed using age as a covariant to determine whether age had significant prognostic impact on disease outcome in this patient population. First, patients aged under 41 years of age were compared with patients older than 41 years and secondly this group of patients was further divided into three subgroups; < 30 years, 31-35 years and 36-40 years.

In the univariate analysis, age under 41 was a significant prognostic factor for overall survival (HR 1.34 (1.18-1.52) P < 0.01) (Figure 1) and distant metastasis free survival (HR 1.48 (1.33-1.65) P < 0 .01) associated with unfavorable prognosis. The unfavorable prognostic impact was most profound in patients aged under 31 for overall survival (HR 1.77 (1.25-2.51) P < 0.01) (Figure 2) and distant metastasis free survival (HR 2.16 (1.63-2.86) P < 0.01).

To test whether the observed prognostic impact of young would remain significant when other tumor characteristics are taken into account, we first performed univariate analyses for overall and distant metastasis free survival including tumor size, nodal status, estrogen receptor status, type of surgery, and the administration of adjuvant chemotherapy. To prevent potential confounding due to selection bias as a result of the different trials in which patients participated; we also inserted trial as a covariant. Trial 22881 was defined as reference trial. All the above mentioned covariates were significantly associated with outcome for overall survival and distant metastasis free survival (Table 3).

Next, we included all covariates, including patient age, into a multivariate analysis to test the independent effect of age on outcome. Estrogen receptor status was not included in the multivariate analysis since no information was available for 2942 patients including all patients who participated in EORTC trial 10801 and therefore

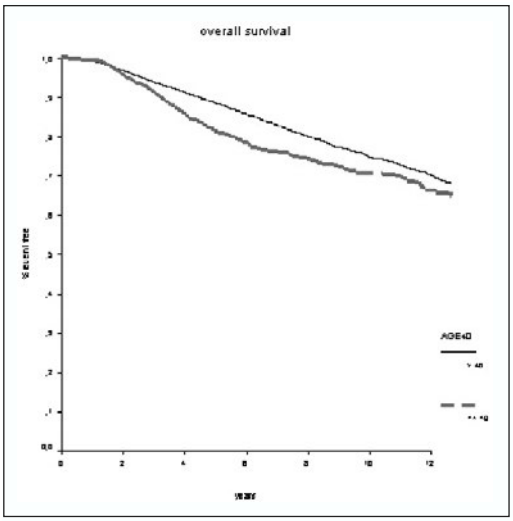

Figure 1. Overall survival

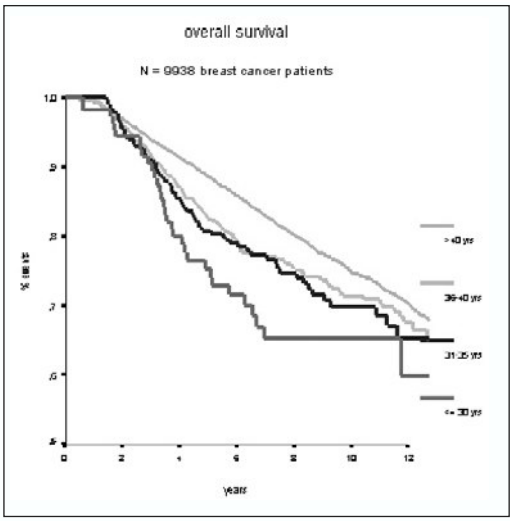

Figure 2. Overall survival

these data would be lost. The prognostic impact of all covariates except type of surgery remained significant (See table 3). Tumor size > 5cm and positive axillary lymph nodes were strong prognostic factors for poor prognosis, risk ratio's being 2.28 and 2.37 for overall survival and 2.25 and 1.97 for distant metastasis free survival respectively. In addition, young age remained an independent prognostic factor for overall (RR 1.43 (1.25-1.63) P < 0.01) and distant metastasis free survival (RR 1.58 (1.41-1.77 P < 0.01).

Prognostic factor analyses within the young age group

Next, we studied the prognostic impact of several different tumor characteristics in the subset of 549 patients aged under 41 of which tumor material was collected. Patient characteristics and immunohistochemistry results are listed in Table 2. To test whether these covariant had significant impact on prognosis in young breast cancer patients, univariate analyses for overall survival and distant metastasis free survival were performed. Large tumor size, positive nodal status, poorly differentiated histological grade, extensive lymphangio invasion and negative hormone receptor status were all associated with poor survival (Table 4). In addition, adjuvant chemo-therapy was associated with poor outcome (HR 1.90 (1.34-2.71) P < 0.01). Her2 over expression (HR 1.09 (0.70-1.69) P = 0.71) and P53 overexpression (HR1.53 (0.90-2.04) P = 0.15) were not significantly associated with poor overall survival in this group of patients.

For distant metastasis free survival, large tumor size, nodal status, poorly differentiated histological grade, and adjuvant chemotherapy were associated with poor outcome (Table 4). Positive ER status (HR 0.90 (0.65-1.24) P = 0.51) did not have a significant impact on distant metastasis free survival. Similar results were found for progesterone receptor status. In addition, Her2 and P53 overexpression did not have a significant impact on distant metastasis free survival.

Subsequently, we tested the independent significant covariates in the univariate

	Overall survival			Distant disease-free survival		
	HR	95% CI	P	HR	95% CI	P
≤ 40 years	1.34	1.18-1.52	<0.01	1.48	1.33-1.65	<0.01
≤ 30 years	*1.77*	*1.25-2.51*	*<0.01*	*2.16*	*1.63-2.86*	*<0.01*
31-35 years	*1.34*	*1.07-1.69*	*0.01*	*1.54*	*1.27-1.87*	*<0.01*
36-40 years	*1.29*	*1.10-1.50*	*<0.01*	*1.38*	*1.20-1.57*	*<0.01*
cT2 (vs cT1)	1.77	1.59-1.97	<0.01	1.66	1.52-1.81	<0.01
cT3 (vs cT1)	3.70	3.06-4.48	<0.01	3.18	2.68-3.78	<0.01
pN+	2.40	2.18-2.63	<0.01	1.99	1.84-2.16	<0.01
ER+	0.65	0.57-0.73	<0.01	0.78	0.71-0.87	<0.01
BCT	0.61	0.55-0.67	<0.01	0.66	0.60-0.72	<0.01
Adj CT*	1.46	1.31-1.62	<0.01	1.34	1.23-1.47	<0.01
Trial (ref trial: 22881)						
10801	2.13	1.85-2.46	<0.01	1.59	1.40-1.81	<0.01
10854	1.58	1.41-1.78	<0.01	1.48	1.34-1.62	<0.01
10902	2.51	2.11-3.0	<0.01	1.92	1.65-2.23	<0.01
	Multivariate analyses all patients					
	Overall survival			Distant disease-free survival		
	RR	95% CI	P	RR	95% CI	P
≤ 40 years	**1.43**	**1.25-1.63**	**<0.01**	**1.58**	**1.41-1.77**	**<0.01**
cT2 (vs cT1)	1.46	1.30-1.64	<0.01	1.47	1.34-1.62	<0.01
cT3 (vs cT1)	2.28	1.82-2.86	<0.01	2.25	1.84-2.75	<0.01
pN+	2.37	2.12-2.64	<0.01	1.97	1.80-2.17	<0.01
BCT	**0.93**	**0.82-1.04**	**0.19**	**0.97**	**0.87-1.08**	**0.51**
Adj CT*	0.69	0.60-0.79	<0.01	0.71	0.62-0.80	<0.01
Trial (ref trial: 22881)						
10801	1.55	1.32-1.81	<0.01	1.25	1.09-1.45	<0.01
10854	1.03	0.90-1.18	0.68	1.07	0.95-1.19	0.26
10902	1.62	1.28-2.06	<0.01	1.45	1.18-1.77	<0.01

* Prolonged chemotherapy

Table 3. Univariate and multivariate analyses all patients

	Overall survival			Distant disease-free survival		
	HR	95% CI	P	HR	95% CI	P
cT2	2.15	1.42-3.24	< 0.01	1.92	1.38-2.67	<0.01
cT3	4.11	1.95-8.64	<0.01	5.10	2.82-9.28	<0.01
pT2/3	2.00	1.37-2.93	<0.01	2.01	1.47-2.74	<0.01
pN+	2.13	1.50-3.03	<0.01	1.91	1.43-2.55	<0.01
Gr II	2.65	1.10-6.39	0.03	2.59	1.39-4.85	<0.01
Gr III	4.69	2.04-10.76	<0.01	3.04	1.67-5.54	<0.01
Lymfangio invasion	1.29	0.88-1.89	0.19	1.33	0.97-1.83	0.07
> 5 vessels	*1.81*	*1.17-2.80*	*<0.01*	*1.82*	*1.25-2.64*	*<0.01*
ER +	0.63	0.43-0.93	0.02	0.90	0.65-1.24	0.51
PgR +	0.59	0.40-0.88	0.01	0.78	0.57-1.08	0.14
HER2 +	1.09	0.70-1.69	0.71	1.00	0.69-1.45	0.99
P53 +	1.35	0.90-2.04	0.15	1.03	0.72-1.46	0.89
BCT	0.67	0.45-0.99	0.04	0.71	0.51-1.00	0.05
Adj CT*	1.90	1.34-2.71	<0.01	1.58	1.18-2.12	<0.01

*Prolonged chemotherapy

Table 4. Univariate analyses young patients

analyses. Therefore, we performed a multivariate survival analysis for the endpoints overall survival and distant metastasis free survival. In the multivariate analysis we selected tumor size as assessed by pathological examination and discarded clinical tumor size.
In the multivariate analyses, only histological grade remained a significant prognostic factor for both overall survival and distant metastasis free survival (Table 5). However, large tumor size remained an independent unfavorable prognostic factor on outcome in terms of distant metastasis free survival as well (HR 1.64 (1.17-2.31) P < 0.01).

Node negative patients who did not receive chemotherapy

Young versus Old
To detect whether differences in prognosis between young and older patients would still exist in node negative patients, we selected all axillary node negative patients who had not received adjuvant-prolonged chemotherapy. This subgroup consisted of 6060 patients of whom characteristics are listed in Table 6. Except for estrogen receptor status, patients characteristics were not significantly different between both groups. Young age remained of significant prognostic impact for patients bearing cT2 or cT3 tumors in terms of distant metastasis free survival (Table 7). In terms of overall survival, young age still showed a trend significant effect on outcome for smaller tumors but not for larger tumors (Table 7).

	Overall survival			Distant disease-free survival		
	HR	95% CI	P	HR	95% CI	P
pT2/3	1.43	0.92-2.23	0.11	1.64	1.17-2.31	<0.01
pN+	1.70	0.81-3.56	0.16	1.67	0.92-3.01	0.09
Gr II	**2.67**	**0.91-7.80**	**0.07**	2.04	1.07-3.88	0.03
Gr III	**3.92**	**1.38-11.16**	**0.01**	2.38	1.29-4.39	<0.01
Lymfangio invasion				1.06	0.74-1.51	0.77
ER +	0.83	0.48-1.45	0.51			
PgR +	0.90	0.52-1.58	0.72			
BCT	0.82	0.49-1.36	0.44	0.90	0.59-1.36	0.61
Adj CT*	0.98	0.47-2.05	0.96	0.81	0.45-1.43	0.46

*Prolonged chemotherapy

Table 5. Multivariate analyses young patients

Patient characteristics	≤ 40 years	> 40 years
	No / %	No / %
Clinical Tumor size		
T1	312 / 49	2580 / 48
T2	320 / 50	2772 / 51
T3	6 / 1	53 / 1
ER status* **		
Positive	**260 / 61**	**2785 / 75**
Negative	**167 / 39**	**938 / 25**
Surgery		
Breast conserving therapy	573 / 89	4773 / 88
Mastectomy	67 / 11	646 / 12

*Missing data not shown, **significant difference between both groups

Table 6. Node negative patients who did not receive prolonged CT (N= 6060)

	Overall survival			Distant disease-free survival		
	HR	95%CI	P	HR	95%CI	P
≤ 40 years vs. > 40 years						
cT1 (312 pts vs. 2579 pts)	1.38	0.99-1.92	0.06	1.50	1.16-1.94	<0.01
cT2 (319 pts vs. 2765 pts)	1.13	0.85-1.50	0.39	1.44	1.17-1.79	<0.01

Table 7. Multivariate analyses node negative patients who did not receive prolonged CT

Prognostic factors within young node negative patients

The subgroup of young node negative patients of whom tumor material was collected consisted of 341 women. Patient characteristics are listed in Table 8. In this subgroup, univariate analyses were performed, including tumor size, histological grade, vessel invasion, hormone receptor status, Her2 status, P53 status, and type of surgery and chemotherapy. In the univariate analyses, tumor size, grade and hormone receptor status demonstrated to be significant prognostic factors on overall and distant metastasis-free survival (See Table 9).

Next, in the multivariate analyses, histological grade remained an independent prognostic factor for both overall survival (Gr II vs Gr I NS, Gr III vs Gr I HR 8.92 (1.17-68.20) P 0.04) and distant disease-free survival respectively (Gr II vs Gr I NS, Gr III vs Gr I HR 4.12 (1.42-11.98) P <0.001). Further results are listed in Table 10 and univariate Kaplan Meyer curves for overall survival and distant disease free survival concerning histological grade are depicted in Figures 3 and 4.

Discussion

In this study we performed a retrospective analysis to gain further insight in tumor characteristics of young breast cancer patients. Young age at onset of breast cancer is a well-known independent prognostic factor but a genotypical explanation for this phenomenon is still lacking. Part of the more aggressive behavior of breast cancer at a young age may be attributable to hereditary

	Patient characteristics, (N = 341)*	
	No.	%
Clinical Tumor size		
T1	167	49
T2	169	50
T3	4	1
Pathological tumor size		
T1	231	73
T2 / T3	84	27
Tumor grade		
I	54	17
II	93	29
III	172	54
Lymfangio invasion		
None	243	76
2-5 vessels	49	15
> 5 vessels	27	9
ER status		
Positive	165	59
Negative	115	41
PgR status		
Positive	136	49
Negative	141	51
HER2 status		
Negative	216	77
Positive	64	23
P53 status		
Negative	198	72
Positive	78	28
Surgery		
Breast conserving therapy	229	88
Mastectomy	42	12
Prolonged adjuvant chemotherapy		
No	304	89
Yes	37	11

* Missing data not shown

Table 8. Node-negative patients aged < 41

	Overall survival			Distant disease-free survival		
	HR	95% CI	P	HR	95% CI	P
cT2	2.47	1.39-4.38	< 0.01	1.84	1.19-2.84	<0.01
cT3	6.44	1.48-27.97	<0.01	15.5	4.57-52.59	<0.01
pT2/3	2.38	1.37-4.13	<0.01	1.81	1.15-2.83	0.01
Gr II	1.80	0.48-6.81	0.38	2.35	0.95-5.84	0.07
Gr III	5.64	1.74-18.23	<0.01	3.89	1.67-9.05	<0.01
Lymfangio invasion	1.13	0.61-2.13	0.69	1.41	0.68-2.26	0.16
> 5 vessels	*1.12*	*0.44-2.83*	*0.82*	*1.71*	*0.87-3.34*	*0.12*
ER +	0.43	0.24-0.78	<0.01	0.61	0.39-0.96	0.03
PgR +	0.44	0.24-0.82	<0.01	0.64	0.40-1.02	0.06
HER2 +	0.75	0.35-1.60	0.45	0.90	0.51-1.59	0.71
P53 +	1.54	0.82-2.87	0.18	1.15	0.69-1.9	0.59
BCT	1.14	0.53-2.43	0.74	1.44	0.72-2.88	0.30
Adj CT*	2.73	1.36-5.46	<0.01	1.19	0.60-2.38	0.62

*37 pts in this subset received prolonged adjuvant chemotherapy

Table 9. Univariate analyses node-negative patients aged < 41

factors. However, at present only approximately 10% of young breast cancer cases have a documented BRCA I or BRCA II mutations or have a strong positive family history of breast cancer [7-9, 25].

We demonstrated in approximately 10000 early stage breast cancer patients that age > 41 years is a strong prognostic factor on disease outcome independent of other covariates. This is in accordance with previous data, which have led to the recommendation that all patients aged ≤ 35 years at time of diagnosis should receive adjuvant chemotherapy irrespective of other tumor characteristics. In this study the effect was most profound for patients aged under 31. However, the finding that patients aged between 35 and 41 still had a poor prognosis compared to older patients as well could raise the question whether or not these patients should also receive adjuvant chemotherapy.

In the subgroup of node negative patients who did not receive prolonged adjuvant chemotherapy the prognostic effect of young age was less clear. In terms of overall survival, young age as a prognostic factor failed to reach statistical significance. However, there was a significant effect on distant disease free survival. Hazard ratios varied between 1.13 and 1.50 in these analyses which could be roughly converted in NNT's (numbers needed to treat) varying between 11 and 38 hypothesizing an expected 30% event rate at 10 years. In addition, in this study young node negative patients bearing grade I tumors had excellent 10 years survival and distant disease-free survival rates of approximately 90% for both endpoints.

This raises the discussion whether or not all young node negative patients should

	Overall survival			Distant disease-free survival		
	HR	95% CI	P	HR	95% CI	P
pT2/3*	1.50	0.80-2.81	0.20	1.39	0.84-2.30	0.21
Gr II	4.11	0.49-34.57	0.19	2.49	0.81-7.66	0.11
Gr III	**8.92**	**1.17-68.20**	**0.04**	**4.12**	**1.42-11.98**	**<0.01**
ER +	0.73	0.32-1.70	0.47	0.90	0.46-1.76	0.76
PgR +	0.97	0.40-2.34	0.95	1.08	0.55-2.09	0.83
Adj CT	1.87	0.77-4.57	0.17			

*Pathological tumor size was included in the multivariate analysis and clinical tumor size was left out

Table 10. Multivariate analyses node-negative patients aged < 41

receive chemotherapy. Probably two subgroups can be defined comprising young patients who do not require adjuvant chemotherapy. First, young early breast cancer patients who have an excellent prognosis and second patients with chemotherapy-resistant tumors who do not benefit from chemotherapy anyway. Current research using microarray based prognostic and predictive risk models [26-28] may further elucidate this challenge of so-called treatment tailoring. In this study, histological grade was the strongest prognostic factor of the covariates studied, distinguishing young patients with a favorable prognosis from young patients with an unfavorable prognosis. The majority of young patients had grade III tumors (53%). In addition, large tumor size remained an independent risk factor for distant disease free survival as well. Axillary nodal status was a prognostic factor in the univariate analyses but did not remain significant in the multivariate analyses. Her 2 overexpression and p53 overexpression failed to be of prognostic significance in this subset of young patients. This is not in accordance with previous reports [29, 30]. Maru et reported a positive p53 status in 22 of 44 patients (50%), and a positive HER-2/*neu* status in 18 of 41 patients (44%) scored by FISH. In our study, the p53 and Her 2 positive rates were 29% and 26% respectively estimated by immunohistochemistry. Although Her2 overexpression is a well-known risk factor associated with poor prognosis, we were not able to demonstrate a significant effect. This could be due to insufficient sample size since only 121 patients had Her2 overexpressing tumors

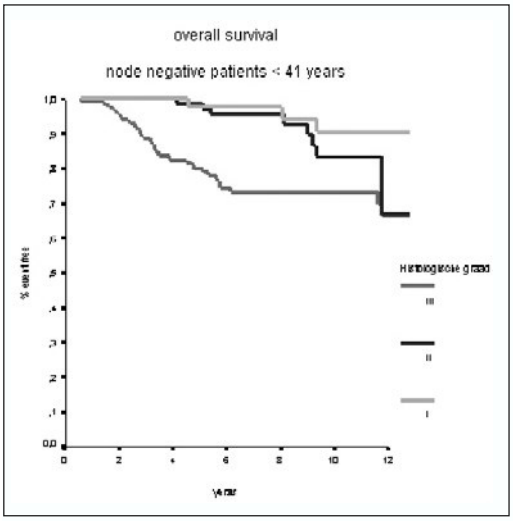

Figure 3. Overall survival and grade

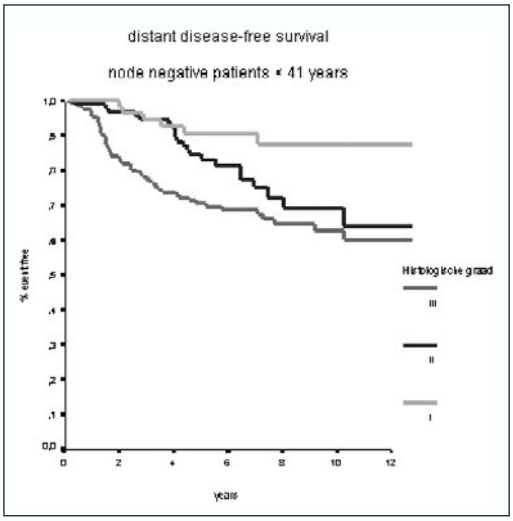

Figure 4. Distant disease-free survival and grade

estimated by immunohistochemistry. It may also be due to other unknown factors in young breast cancer patients, which result in a more aggressive genotype, which is much less influenced by Her2 expression. These plausible unknown factors yet have to be discovered [31].
In conclusion, well known established prognostic factors as tumor size and histologic grade still remain independent prognostic factors on disease outcome in young breast cancer patients and therefore can be a valuable tool in patient information and education. Treatment guidelines concerning young breast cancer patients should be refined in the future based on tumor characteristics, probably derived from microarray driven translational research projects, and not based upon age alone.

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