European Journal of Cancer 102 (2018) 1–9

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.ejcancer.com

Original Research

Adjuvant dose-dense doxorubicin-cyclophosphamide versus docetaxel-doxorubicin-cyclophosphamide for high-risk breast cancer: First results of the randomised MATADOR trial (BOOG 2004-04)

A.G.J. van Rossum [a], M. Kok [b], E. van Werkhoven [c], M. Opdam [a], I.A.M. Mandjes [d], A.E. van Leeuwen − Stok [e], H. van Tinteren [c], A.L.T. Imholz [f], J.E.A. Portielje [g], M.M.E.M. Bos [h,2], A. van Bochove [i], J. Wesseling [a,j], E.J. Rutgers [k], S.C. Linn [a,b,l,*,1], H.M. Oosterkamp [m,1] on behalf of the MATADOR Trialists' Group

[a] *Department of Molecular Pathology, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[b] *Department of Medical Oncology, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[c] *Biometrics Department, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[d] *Data Centre, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[e] *Dutch Breast Cancer Research Group, BOOG Study Centre, IJsbaanpad 9-11, 1076 CV, Amsterdam, The Netherlands*
[f] *Department of Medical Oncology, Deventer Ziekenhuis, Nico Bolkesteinlaan 75, 7416 SE, Deventer, The Netherlands*
[g] *Department of Medical Oncology, HagaZiekenhuis, Els Borst-Eilersplein 275, 2545 AA, The Hague, The Netherlands*
[h] *Department of Internal Oncology, Reinier de Graaf Gasthuis, Reinier de Graafweg 5, 2625 AD, Delft, The Netherlands*
[i] *Department of Medical Oncology, Zaans Medisch Centrum, Koningin Julianaplein 58, 1502 DV, Zaandam, The Netherlands*
[j] *Department of Pathology, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[k] *Department of Surgical Oncology, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX, Amsterdam, The Netherlands*
[l] *Department of Pathology, University Medical Centre, Heidelberglaan 100, 3584 CX, Utrecht, The Netherlands*
[m] *Department of Medical Oncology, Haaglanden Medisch Centrum, The Hague, The Netherlands*

Received 6 July 2018; accepted 9 July 2018
Available online ■ ■ ■

* *Corresponding author:* Department of Molecular Pathology, Netherlands Cancer Institute, Plesmanlaan 121, 1066 CX Amsterdam, The Netherlands.
E-mail address: s.linn@nki.nl (S.C. Linn).
[1] S.C. Linn and H.M. Oosterkamp contributed equally to this work. [2] Present address: Erasmus Medical Centre, Rotterdam, The Netherlands.

https://doi.org/10.1016/j.ejca.2018.07.013

KEYWORDS
Chemotherapy;
Breast cancer;
Dose-dense;
Taxane;
Efficacy;
Biomarker discovery

Abstract ***Background:*** Dose-dense administration of chemotherapy and the addition of taxanes to anthracycline-based adjuvant chemotherapy have improved breast cancer survival substantially. However, clinical trials directly comparing the additive value of taxanes with dose-dense anthracycline-based chemotherapy are lacking.

Patients and methods: In the multicentre, randomised, biomarker discovery Microarray Analysis in breast cancer to Tailor Adjuvant Drugs Or Regimens (MATADOR) trial, patients with pT1-3, pN0-3 breast cancer were randomised (1:1) between six adjuvant cycles of doxorubicin 60 mg/m^2 and cyclophosphamide 600 mg/m^2 every 2 weeks (ddAC) and six cycles of docetaxel 75 mg/m^2, doxorubicin 50 mg/m^2 and cyclophosphamide 500 mg/m^2 every 3 weeks (TAC). The primary objective was to discover a predictive gene expression profile for ddAC and TAC benefit. Here we report the preplanned secondary end-point recurrence-free survival (RFS) and overall survival (OS).

Results: Between 2004 and 2012, 664 patients were randomised. At 5 years, RFS was 87% (95% confidence interval [CI] 83%–91%) in the ddAC-treated patients and 88% (84–92%) in the TAC-treated subgroup (hazard ratio [HR] 0.89, 95% CI 0.62–1.28, $P = 0.53$). OS at 5 years was 93% (90%–96%) in the ddAC-treated and 94% (91%–97%) in the TAC-treated patients (HR 0.89, 95% CI 0.57–1.39, $P = 0.61$). Anaemia was more frequent in ddAC-treated patients (62/327 patients [18.9%] versus 15/319 patients [4.7%], $P < 0.001$) and diarrhoea (21 [6.4%] versus 53 [16.6%], $P<0.001$) and peripheral neuropathy (15 [4.6%] versus 46 [14.4%], $P < 0.001$) were observed more often in TAC-treated patients.

Conclusions: With a median follow-up of 7 years, no significant differences in RFS and OS were observed between six adjuvant cycles of ddAC and TAC in high-risk breast cancer patients.

Trial registration numbers: ISRCTN61893718 and BOOG 2004-04.

1. Introduction

Adjuvant chemotherapy for early breast cancer aims to eradicate micrometastases to improve survival. Anthracycline-containing regimens have increased breast cancer survival substantially [1].

Incorporation of taxanes into anthracycline-based schedules has further improved the efficacy of adjuvant chemotherapy. Compared with six cycles of 5-fluorouracil-doxorubicin-cyclophosphamide, six cycles of adjuvant docetaxel-doxorubicin-cyclophosphamide (TAC) significantly improved overall survival (OS) from 81% to 87% in node-positive breast cancer [2]. The addition of four cycles of a taxane to a fixed anthracycline-based regimen, thereby extending treatment duration, also improved breast cancer–specific survival (BCSS) [1].

Dose-dense scheduling of chemotherapeutic agents accounted for another important step forward. Dose densification is defined as the shortening of the interval between cycles, giving the tumour less time to regrow between treatment cycles. Three meta-analyses showed that adjuvant dose-dense chemotherapy improves disease-free survival (DFS) and OS of breast cancer patients compared with conventionally scheduled chemotherapy regimens [3–5].

Knowing that both the addition of a taxane and dose-dense scheduling increase efficacy of adjuvant chemotherapy, it is unclear which of these strategies gives the largest benefit for an individual patient. Two studies compared a taxane-based, dose-dense regimen directly with conventional dosed anthracycline-based treatment, resulting in a minor survival advantage for dose-dense–treated patients compared with conventionally treated patients [6,7]. However, to date, no randomised trial has directly compared a taxane-containing, conventionally scheduled treatment with a non-taxane–containing, dose-dense regimen. Here, we report the results of the preplanned secondary analyses of a randomised, biomarker discovery trial comparing six cycles of dose-dense–administered AC (ddAC) with six cycles of adjuvant TAC. The primary objective of this trial was to investigate whether a gene expression profile could be identified that could predict who should receive ddAC and who should receive TAC for the best outcome. Application of such a classifier would then lead to a better outcome for the whole group, than when all patients would have received one of these regimens that would have turned out best for the average patients.

2. Materials and methods

2.1. Study design and patients

The Microarray Analysis in breast cancer to Tailor Adjuvant Drugs Or Regimens (MATADOR, ISRCTN61893718) study is a multicentre, randomised, open-label, phase III trial primarily designed to identify a gene expression profile that can predict survival benefit

of ddAC or TAC. Women with a pathologically confirmed T1-T3, N0-3b adenocarcinoma of the breast without signs of distant metastases were considered eligible. The study was amended to also include N0 patients from June 2008 onwards (Amendment 2). Adequate bone marrow, liver and renal functions were required. Main exclusion criteria were prior systemic treatment for cancer, history of breast cancer and other cancers (except for curatively treated non-melanoma skin cancer, in situ carcinoma of the cervix and ipsilateral ductal carcinoma in situ) and significant cardiac, neurological or psychiatric disorders. With trastuzumab not being part of the study treatment and accumulating evidence showing that concurrent trastuzumab and chemotherapy appeared superior compared with sequential scheduling, patients with human epidermal growth factor receptor (HER2)–positive disease were considered ineligible after 2007 (Amendment 2).

The study protocol and amendments were approved by the ethical committee of the Netherlands Cancer Institute and the institutional review boards of the participating centres. The study was performed in accordance with Good Clinical Practice guidelines and with the Declaration of Helsinki (version 17C). All patients provided written informed consent.

2.2. Randomisation and treatment

Patients were initially randomised among four treatments: four or six cycles of ddAC or four or six cycles of TAC. With emerging evidence that six cycles of fluorouracil-doxorubicin-cyclophosphamide (FAC) resulted in better outcomes than six cycles of cyclophosphamide-methotrexate-fluorouracil (CMF) [8], with six cycles of CMF being equally effective as four cycles of AC [9], randomisation was limited to the six cycle regimen (Amendment 1). By then, five patients had received four cycles of ddAC and five patients received four cycles of TAC. Randomisation (1:1) was performed centrally at the Netherlands Cancer Institute using the automated ALEA system (FormsVision BV, the Netherlands).

Patient received either six cycles of doxorubicin 60 mg/m^2 plus cyclophosphamide 600 mg/m^2 every 2 weeks or six cycles of docetaxel 75 mg/m^2, doxorubicin 50 mg/m^2 and cyclophosphamide 500 mg/m^2 every 3 weeks. Granulocyte colony–stimulating factor (pegfilgrastim 6 mg) was given to all patients the day after chemotherapy administration. Prophylactic antibiotics were not standard of care in the study.

Randomisation was stratified by the menopausal status, type of surgery, sequence of adjuvant therapy, tumour size and lymph node status according to AJCC staging, hormone receptor status, HER2 status and treatment centre using Pocock's minimisation technique.

Dose reductions and interruptions were allowed in case of adverse events grade III or higher according to common toxicity criteria for adverse events (CTCAE), version 3.0, except for peripheral neuropathy that required dose reduction of docetaxel at grade II. Adjuvant radiotherapy and/or endocrine therapy were initiated according to the Dutch guidelines on breast cancer treatment (www.oncoline.nl).

2.3. Assessments

Patients were assessed for relapse of disease at regular intervals for 10 years. Evaluation included physical examination and yearly mammography. Adverse events grade II and higher were reported using the CTCAE, v3.0.

Histological grade according to the modified Bloom-Richardson classification [10] and morphology were assessed locally. Tissue microarrays (3 cores of 0.6 mm per patient) were constructed and stained for oestrogen receptor (ER), progesterone receptor (PR) and HER2. According to the Dutch guidelines, ER and PR staining of 10% or more and HER2 score of 3 + or more were scored as positive. In case of a 2 + HER2 score, an in situ hybridisation assay was performed. Central assessment of ER, PR and HER2 was used. If tumour tissue was unavailable, local assessment was used. Breast cancer subtype was defined as (1) ER and/or PR positive and HER2 negative; (2) HER2 positive, regardless of ER and PR status or (3) triple negative.

2.4. Objectives and end-points

The primary objective of the trial was to generate a gene expression profile predictive of DFS benefit of either dose-dense chemotherapy or a docetaxel-containing schedule. DFS was defined as the interval between randomisation and locoregional or distant relapse, second primary cancer, or death by any cause. Because a second primary cancer could not directly be attributed to failure of eradicating micrometastases with systemic treatment, the study protocol was amended (Amendment 3) to change the primary end-point to recurrence-free survival (RFS). RFS was defined as the interval between randomisation and locoregional or distant relapse or death by any cause [11].

The secondary objective was to compare the efficacy of TAC and ddAC. End-points included RFS, distant recurrence-free interval (DRFI), defined as the time from randomisation until distant relapse or breast cancer–related death, OS and BCSS. Also, we evaluated the patients who received at least one cycle of the allocated treatment for toxicity during follow-up.

2.5. Statistics

The primary end-point of the trial was the gain in RFS attributed to the genetic profile. This gain was defined as the improvement of RFS at 5 years with the treatment strategy using the profile, over the strategy in which all patients would get the same treatment (either ddAC or

TAC), whichever would appear better from the direct comparison (which was the secondary objective). It was calculated that if the profile would be developed using data from 400 patients, the standard error of the estimate of the gain would be less than 2.5%. The sample size of the study was set at 660 so that 1/3 of the data could be used as a validation cohort, allowing for 10% early dropout. For the direct comparison of the arms (the secondary objective), 192 RFS events were required to obtain 80% power to detect a difference of a hazard ratio (HR) of 0.67. During the course of the study, it became clear that the event rate was lower than expected. Therefore an amendment was made to the protocol. At the time of this amendment, RFS 87 events were observed, and it was calculated that with a two-sided significance level of $\alpha = 0.025$ (to account for a final analysis after 10 years of follow-up), the smallest difference that could be detected with 80% power was an HR of approximately 0.50. Results from the Early Breast Cancer Trialists' Collaborative Group (EBCTCG) overview [1] suggested that the benefit of taxanes diminishes after 5 years; so waiting for more events would not provide much more information about sensitivity to treatment with taxanes. Therefore, the analysis after 5-year follow-up was added to the amendment (Amendment 3). In addition, it was decided to use a cross-validation method instead of separation in a development and a validation cohort as this may result in a better profile and more precise estimates of its predictive accuracy.

The database was closed on 14 November 2017. We compared the categorical clinicopathological characteristics of the two treatment groups using a Chi-square or Fisher's exact test.

Efficacy analyses were performed in the intention-to-treat (ITT) population, including all patients who were allocated to one of the two treatment arms. RFS, DRFI, OS and BCSS of the two treatments were estimated using the Kaplan-Meier method and compared with a log-rank test. Multivariable Cox proportional hazards models were generated to correct for known prognostic factors. Exploratory subgroup analyses on RFS and OS, including interactions, were performed using Cox regression models.

Additionally, efficacy analyses were performed in the per-protocol treated (PPT) subgroup. The PPT population consisted of patients who received at least one treatment of ddAC or TAC. Patients were excluded if they were randomised to and received four cycles of chemotherapy, if they randomised for ddAC and were treated with an adjuvant taxane outside the scope of this study or if they had HER2-positive disease.

Observed toxicity was evaluated in all patients who received at least one cycle of the allocated treatment and was compared using a Chi-square or Fisher's exact test.

All p-values were two sided, and values below 0.05 were considered significant, except for the comparison of ddAC with TAC for the RFS efficacy end-point, where the threshold was set at 0.025 (two sided). Statistical analyses were performed using SPSS 22 and R 3.3.1.

3. Results

Between 2004 and 2012, 664 patients were enrolled and randomised in 29 centres throughout the Netherlands (ITT population). Toxicity analysis was performed in 646 patients. The PPT population consisted of 614 patients (Fig. 1).

The treatment groups were well balanced regarding prognostic clinicopathologic characteristics (Table 1). Mean age was 51.1 years (standard deviation, 8.0). Five hundred thirty-one of 664 patients (80%) had lymph node–positive disease and 108 patients (16.3%) had triple-negative breast cancer. Twenty-one patients with HER2-positive disease were included of whom 14 were treated with trastuzumab.

3.1. Efficacy

At the time of the analyses, the ITT population had a median follow-up of 7 years. Two hundred eighty (84.3%) of 332 patients completed six cycles ddAC at the planned dose; 271 (81.6%) of 332 patients received six full cycles of TAC treatment ($P=0.41$).

The estimated 5-year RFS rate was 86.9% (95% CI 83.3–90.6) in the ddAC-treated patients and 87.9% (84.4–91.5) in the TAC-treated subgroup, which was not significantly different (HR 0.89, 95% CI 0.62–1.28, $P=0.53$; Fig. 2a), neither after adjustment for known prognostic factors (Supplementary Table S1). The same holds true for DRFI (Supplementary Fig. S1 and Table S2). Of note, although not shown here, similar results were obtained using DFS as primary end-point.

The 5-year OS did not significantly differ between the two treatment arms: 92.6% (95% CI 89.8–95.5) in the ddAC-treated subgroup and 93.8% (91.1–96.5) in the TAC-treated patients (HR 0.89, 95% CI 0.57–1.39, $P=0.61$; Fig. 2b), neither when adjusted for known prognostic factors (Supplementary Table S3). No difference was observed for BCSS between ddAC and TAC (Supplementary Fig. S2 and Table S4).

In the exploratory subgroup analyses, the interaction between age as a dichotomous variable and treatment showed a trend for OS ($P_{\text{interaction}} = 0.040$; Fig. 3) with a numerical survival benefit for patients younger than 50 years when treated with ddAC (HR 1.72, 95% CI 0.79–3.73) and for patients who were 50 years or older when treated with TAC (HR 0.62, 95% CI 0.35–1.11). The interaction was not significant for RFS ($P_{\text{interaction}} = 0.084$; Supplementary Fig. S3).

Fifty patients were excluded from the PPT analyses (Fig. 1). Similar to the ITT population, RFS and OS were not significantly different between the ddAC- and the TAC-treated patients (Supplementary Fig. S4a-b).

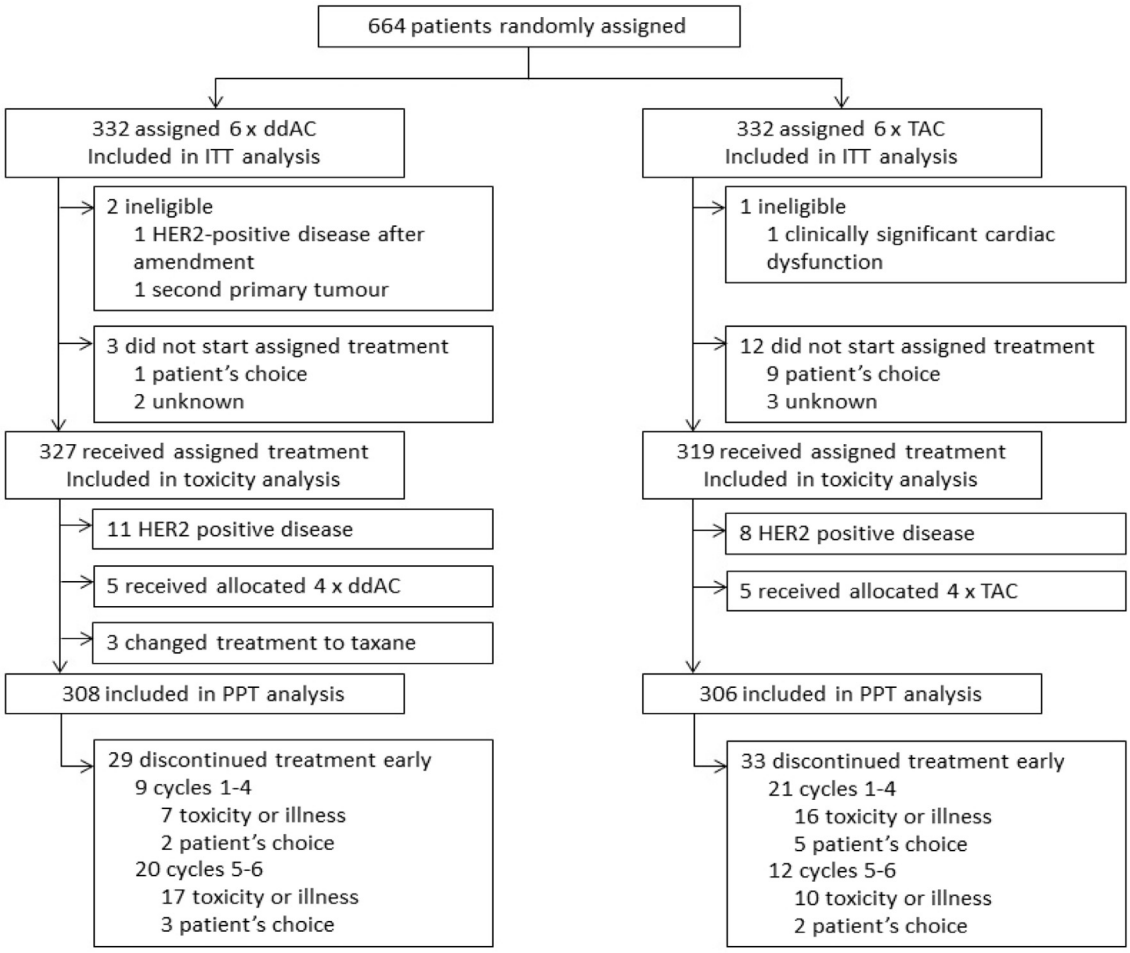

Fig. 1. CONSORT diagram. A, doxorubicine; C, cyclophosphamide; T, docetaxel; dd, dose-dense; HER2, human epidermal growth factor receptor 2; ITT, intention-to-treat; PPT, per-protocol treated.

3.2. Toxicity

The observed adverse events (grade II and higher) of the two treatments are distinct (Table 2). Importantly, anaemia was more frequent in ddAC-treated patients (62 [18.9%] of 327 patients versus 15 [4.7%] of 319 patients, $P<0.001$) and diarrhoea (21 [6.4%] versus 53 [16.6%], $P<0.001$) and peripheral neuropathy (15 [4.6%] versus 46 [14.4%], $P<0.001$) were observed more often in TAC-treated patients. Regarding severe adverse events, acute myeloid leukaemia (AML) occurred twice in both treatment groups. One ddAC-treated patient developed myelodysplastic syndrome (MDS). Cardiac failure grade III or IV was observed in one ddAC-treated patient and in two TAC-treated patients. Toxicity of ddAC and TAC treatment in the context of drug metabolism–related polymorphisms was reported elsewhere [12].

4. Discussion

Here we present the first direct comparison of efficacy of six cycles of ddAC and six cycles of TAC as adjuvant treatment for breast cancer as a secondary analysis of a randomised biomarker discovery trial. With a median follow-up of 7 years, ddAC and TAC were not significantly different regarding the survival end-points in our study. This is in line with the Oxford Overview meta-analysis [1] that contains more than 14,000 patients for the specific comparison between taxanes given concurrently with anthracyclines versus a non-taxane–containing regimen with a less than two times increased dose of non-taxane chemotherapy and with the CALGB40101 trial [13]. Interestingly when compared with the previously mentioned meta-analysis data, the survival rates in our cohort were remarkably high, particularly in this high-risk patient population in which 80.0% of the patients had lymph node–positive disease.

Several factors might have contributed to the relatively high survival rates of our cohort compared with previously reported outcomes in older studies. First, patients with HER2-positive disease were excluded after the introduction of trastuzumab. In older cohorts that included the HER2-positive tumours that were not treated with anti-HER2–based therapy, the survival

Table 1
Baseline characteristics of intention-to-treat population.

Clinicopathologic characteristic		6× ddAC, $N = 332$	6× TAC, $N = 332$	p-value
Age groups (%)	<50 years	143 (43.1)	154 (46.4)	0.435
	≥50 years	189 (56.9)	178 (53.6)	
Surgery (%)	Breast-conserving surgery	180 (54.2)	169 (50.9)	0.538
	Mastectomy	151 (45.5)	158 (47.6)	
	Missing	1 (0.3)	5 (1.5)	
Endocrine treatment (%)	No	54 (16.3)	59 (17.8)	0.641
	Yes	278 (83.7)	268 (80.7)	
	Missing	0 (0)	5 (1.5)	
T stage[b] (%)	T1	158 (47.6)	155 (46.7)	0.654[a]
	T2	156 (47.0)	152 (45.8)	
	T3	16 (4.8)	19 (5.7)	
	T4	2 (0.6)	0 (0)	
	Missing	0 (0)	6 (1.8)	
N stage[b] (%)	N0	65 (19.6)	63 (19.0)	0.889
	N1	208 (62.7)	200 (60.2)	
	N2	44 (13.3)	45 (13.6)	
	N3	15 (4.5)	19 (5.7)	
	Missing	0 (0)	5 (1.5)	
Grade[c] (%)	Good	32 (9.6)	35 (10.5)	0.796
	Intermediate	151 (45.5)	138 (41.6)	
	Poor	139 (41.9)	137 (41.3)	
	Missing	10 (3.0)	22 (6.6)	
Histology (%)	Ductal	270 (81.3)	257 (77.4)	0.507
	Lobular	47 (14.2)	46 (13.9)	
	Other	13 (3.9)	19 (5.7)	
	Missing	2 (0.6)	10 (3.0)	
Subtype[d] (%)	ER and/or PR positive, HER2 negative	266 (80.1)	269 (81.0)	0.800
	HER2 positive	12 (3.6)	9 (2.7)	
	Triple negative	54 (16.3)	54 (16.3)	

A, doxorubicin; C, cyclophosphamide; T, docetaxel; dd, dose-dense.
[a] Pearson Chi-square test or Fisher's exact test (two sided), missing values excluded.
[b] According to AJCC staging 6th edition.
[c] Grading according to the modified Bloom-Richardson grading system.
[d] ER and PR nucleic staining of 10% staining or more was scored as positive and HER2 score of 3 + was considered positive; in case of a 2 + HER2 score, an in situ hybridisation assay was performed; subtypes were defined as (1) oestrogen receptor (ER) and/or progesterone receptor (PR) positive, human epidermal growth factor receptor 2 (HER2) negative; (2) HER2 positive, regardless of ER or PR status; (3) triple (ER, PR, HER2) negative.

was less favourable [14,15]. Also stage migration, also known as the Will Rogers phenomenon, might play a role. Improved diagnostics and new technologies, as shown previously for ^{18}F-fluorodeoxyglucose positron emission tomography combined with computed tomography [16], lead to more accurate identification of (distant) metastases. Patients who would have been diagnosed with stage III disease in the past and treated with adjuvant systemic therapy are nowadays diagnosed with stage IV disease [17]. The taxane plus anthracycline trials reported in the Oxford Overview meta-analysis enrolled patients between 1994 and 2005, almost a decade earlier than inclusion of patients in the current trial (2004–2012). Interestingly, the MINDACT trial (2007–2011) was executed in the same time period in Europe, and our relatively favourable survival data resemble the survival data of the high-risk patients included in MINDACT who received adjuvant chemotherapy [18].

The primary objective of this trial is to generate a predictive gene expression profile, which is currently being explored. Because the sample size was calculated for the primary end-point, the study may be underpowered for the secondary objective, particularly with the unexpected low number of events observed. However, because chemotherapy displays the largest survival effect in the first years after diagnosis and the carry-over effect diminishes after 7 years for taxanes and even earlier for anthracycline-based regimens [1], it seems relevant to report these results now.

The enrolment period from 2004 until 2012 was relatively long. The novel design of a biomarker study required some adjustments of daily clinical practice. To ensure sufficient quality of the RNA, the ability to freeze tumours was a requirement for hospitals to participate in the trial. At the start of this trial, only a few hospitals had the logistics in place to freeze tumours after surgery. Given the speedy accrual of other

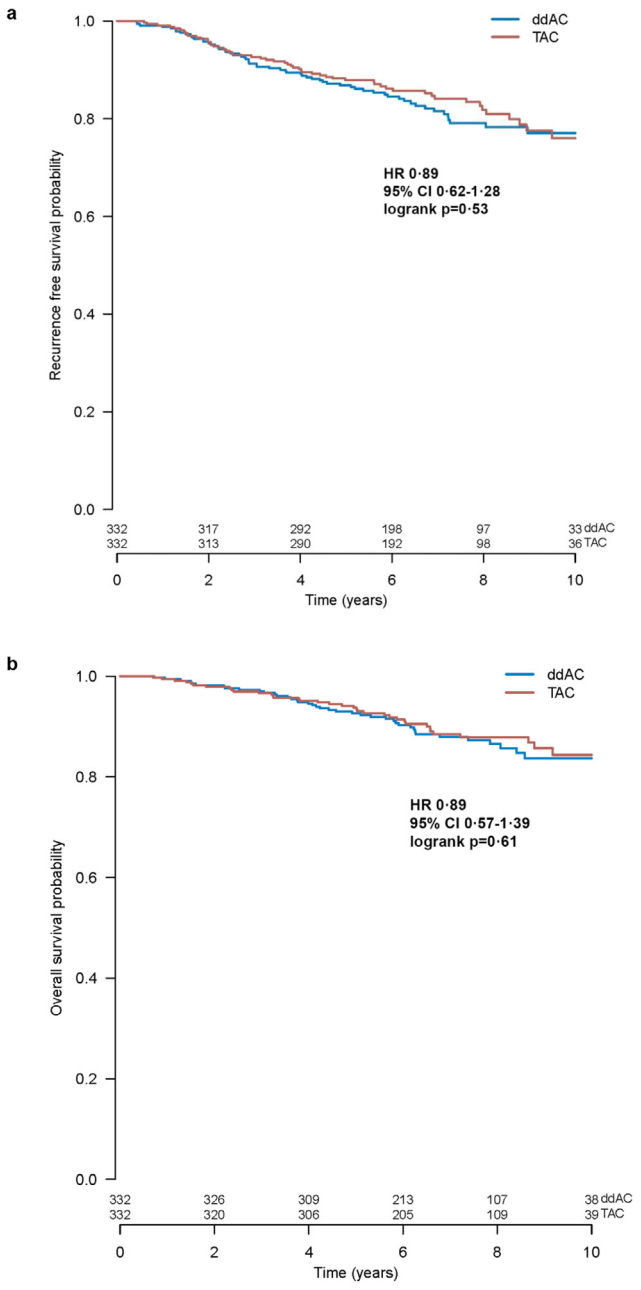

Fig. 2. Recurrence-free survival (a) and overall survival (b) of the intention-to-treat population. A, doxorubicine; C, cyclophosphamide; T, docetaxel; dd, dose-dense; HR, hazard ratio; CI, confidence interval.

biomarker-based trials that started a couple of years later, such as but not limited to the MINDACT trial, developments in molecular diagnostics have resulted in logistics for frozen tumours in the majority of hospitals nowadays. Also, emerging evidence caused a shifting landscape of potential adjuvant systemic treatment regimens, compromising the accrual. Nevertheless, the primary objective of this trial is still a valid and clinically relevant aim.

In this trial, we evaluated three variables: (1) the time between cycles (2 weeks versus 3 weeks), (2) the different dosages of doxorubicin (60 mg/m^2 versus 50 mg/m^2) and cyclophosphamide (600 mg/m^2 versus 500 mg/m^2) and (3) the taxane addition. The number of variables makes it difficult to assess to what extent a specific factor contributes to the efficacy of these regimens. The lack of superiority of TAC over ddAC could be due to the somewhat higher dosed doxorubicin and cyclophosphamide in the ddAC arm compared with TAC, thereby increasing the dose intensity defined as mg/m^2 per time interval. The dose-dense schedule further increases the dose intensity without increasing the toxicity [19]. Dose intensification of doxorubicin and cyclophosphamide seems, therefore, equally effective as the addition of docetaxel to these agents after a median follow-up of 7 years in our cohort.

The unplanned subgroup analysis provided some evidence of an interaction between age and treatment, with a numerical OS benefit for younger patients (<50 years) when treated with ddAC compared with TAC and for older patients (≥50 years) when treated with TAC compared with ddAC. These results are in line with a previous report on improved survival after dose-dense chemotherapy compared with standard-interval chemotherapy in young breast cancer patients [20]. Also, higher survival rates are observed in older patients treated with taxane-containing regimens compared with patients of the same age treated with non-taxane–based regimens [1,21]. Although one might expect ddAC to be more efficacious in relative aggressive tumours that are more prevalent in younger patients [19,22], we did not observe an association between the grade and age in our population, nor did we find a significant interaction between the grade and treatment effect. Currently ongoing gene expression analyses might provide hints on the biology that could be driving this.

The regimens used in our cohort displayed distinct toxicity profiles, which are in line with previous studies on dose-dense chemotherapy [4,13] and reports on taxane-based treatments [23,24]. AML and MDS were observed in 2 (0.6%) of 327 ddAC-treated patients and 2 (0.6%) of 319 TAC-treated patients. Previous anthracycline-based studies have shown a similar probability of AML and MDS of 0.55% at 8 years of follow-up [25]. Compared with the BCIRG 001 trial [24], cardiac failure was uncommon in our study population (1 ddAC-treated patient [0.3%], 2 TAC-treated patients [0.6%]). However, longer follow-up is needed to assess the long-term toxicity of these regimens. Because these toxicities are associated with anthracyclines in a dose-dependent manner, four courses of anthracycline-based chemotherapy, followed by taxanes may be the preferred regimen in the absence of predictive biomarkers for regimen-specific efficacy. Predicting sensitivity for toxicity, for instance by screening for genetic polymorphisms, may help to tailor treatment [12,26]. In addition, treatment duration might be important for some patients. For these patients, a 12-week during schedule might be more attractive than an 18-week during schedule.

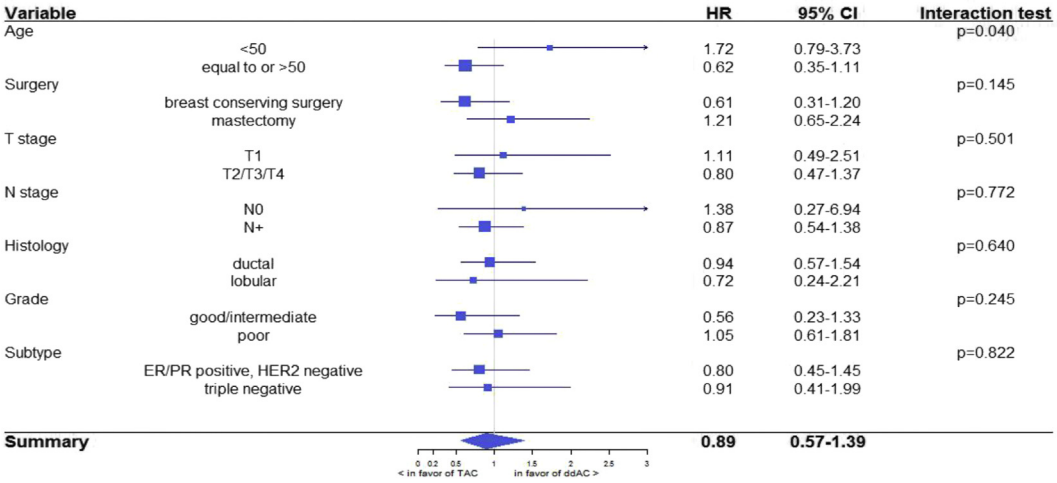

Fig. 3. Forest plot of treatment effect on overall survival in subgroups. T stage and N stage are based on the TNM classification 2002. A, doxorubicin; C, cyclophosphamide; T, docetaxel; dd, dose-dense; HR, hazard ratio; CI, confidence interval; ER, oestrogen receptor; PR, progesterone receptor; HER2, human epidermal growth factor receptor 2; TNM, . Subtypes were defined as (1) ER and/or PR positive, HER2 negative and (2) triple (ER, PR, HER2) negative.

Table 2
Most frequent toxicities (grade II or higher) for ddAC-treated patients and TAC-treated subgroup.

Side-effects	ddAC, n = 327 (%)	TAC, n = 319	p-value[a]
Anaemia	62 (18.9)	15 (4.7)	**<0.001**
Leukocytopenia	30 (9.2)	20 (6.3)	0.167
Fatigue	117 (35.8)	109 (34.2)	0.668
Diarrhoea	21 (6.4)	53 (16.6)	**<0.001**
Nausea	65 (20.0)	52 (16.3)	0.238
Vomiting	35 (10.7)	21 (6.6)	0.063
Febrile neutropenia	36 (11.0)	40 (12.5)	0.546
Peripheral neuropathy	15 (4.6)	46 (14.4)	**<0.001**

A, doxorubicin; C, cyclophosphamide; T, docetaxel dd, dose-dense. The p-values printed in bold are below a significance level of 0.05.
[a] Pearson Chi-square test (two-sided).

5. Conclusions

Our data show that the 5-year survival of high-risk breast cancer patients is excellent after adjuvant treatment with six cycles of TAC or six cycles of ddAC and that distinct toxicity profiles and treatment durations characterise these schedules. Although the preferred adjuvant schedule may shift towards dose-dense sequential chemotherapy [5], knowledge about 'second best' schedules with their own characteristics may help to search for alternative regimens if required. In addition, predictive biomarkers are warranted to further improve well-informed treatment decisions. Therefore, we aim to develop a gene expression profile predictive for treatment efficacy of either ddAC or TAC.

Role of the funding sources

This work was supported by the Dutch Cancer Society and by unrestricted institutional research grants from Sanofi and Amgen to S.C.L. and H.M.O. The funding sources had no role in the design and conduct of the study; collection, management, analysis and interpretation of the data; preparation or approval of the manuscript and decision to submit the manuscript for publication.

Conflict of interest statement

S.C.L. and H.M.O. received an unrestricted institutional research grant from Sanofi and Amgen to conduct the MATADOR study (ISRCTN61893718). M.K. is an advisory board member for BMS and received institutional research support funding from Roche and BMS. S.C.L. is an advisory board member for AstraZeneca, Cergentis, Novartis, Pfizer, Roche and Sanofi and received institutional research support funding from Amgen, AstraZeneca, Genentech, Roche, Sanofi and TESARO. H.M.O. is an advisory board member for Roche, Pfizer and Novartis and received research support funding from Roche. Remaining authors have declared no conflicts of interest.

Acknowledgements

The authors would like to thank all patients and their families. Also, they would like to express their gratitude to the study teams of the participating centres, the Data Centre of the Netherlands Cancer Institute for collecting the data, the Core Facility—Molecular Pathology and Biobanking of the Netherlands Cancer Institute for their help with the immunohistochemistry stainings and storage of the patients' tissue and the Dutch Breast Cancer Research Group (BOOG) for their role in coordinating the study.

Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.ejca.2018.07.013.

References

[1] Early Breast Cancer Trialists' Collaborative G, Peto R, Davies C, Godwin J, Gray R, Pan HC, et al. Comparisons between different polychemotherapy regimens for early breast cancer: meta-analyses of long-term outcome among 100,000 women in 123 randomised trials. Lancet 2012;379(9814):432–44.

[2] Martin M, Pienkowski T, Mackey J, Pawlicki M, Guastalla JP, Weaver C, et al. Adjuvant docetaxel for node-positive breast cancer. N Engl J Med 2005;352(22):2302–13.

[3] Bonilla L, Ben-Aharon I, Vidal L, Gafter-Gvili A, Leibovici L, Stemmer SM. Dose-dense chemotherapy in nonmetastatic breast cancer: a systematic review and meta-analysis of randomized controlled trials. J Natl Cancer Inst 2010;102(24):1845–54.

[4] Petrelli F, Cabiddu M, Coinu A, Borgonovo K, Ghilardi M, Lonati V, et al. Adjuvant dose-dense chemotherapy in breast cancer: a systematic review and meta-analysis of randomized trials. Breast Cancer Res Treat 2015;151(2):251–9.

[5] Gray RB, Bradley R, Braybrooke J, Davies C, Pan H, Peto R, et al. Increasing the dose density of adjuvant chemotherapy by shortening intervals between courses or by sequential drug administration significantly reduces both disease recurrence and breast cancer mortality: an EBCTCG meta-analysis of 21,000 women in 16 randomised trials. In: San Antonio Breast Cancer Symposium; 2017. San Antonio.

[6] Kummel S, Krocker J, Kohls A, Breitbach GP, Morack G, Budner M, et al. Randomised trial: survival benefit and safety of adjuvant dose-dense chemotherapy for node-positive breast cancer. Br J Cancer 2006;94(9):1237–44.

[7] Burnell M, Levine MN, Chapman JA, Bramwell V, Gelmon K, Walley B, et al. Cyclophosphamide, epirubicin, and fluorouracil versus dose-dense epirubicin and cyclophosphamide followed by paclitaxel versus doxorubicin and cyclophosphamide followed by paclitaxel in node-positive or high-risk node-negative breast cancer. J Clin Oncol 2010;28(1):77–82.

[8] Early Breast Cancer Trialists' Collaborative G. Effects of chemotherapy and hormonal therapy for early breast cancer on recurrence and 15-year survival: an overview of the randomised trials. Lancet 2005;365(9472):1687–717.

[9] Fisher B, Brown AM, Dimitrov NV, Poisson R, Redmond C, Margolese RG, et al. Two months of doxorubicin-cyclophosphamide with and without interval reinduction therapy compared with 6 months of cyclophosphamide, methotrexate, and fluorouracil in positive-node breast cancer patients with tamoxifen-nonresponsive tumors: results from the National Surgical Adjuvant Breast and Bowel Project B-15. J Clin Oncol 1990;8(9):1483–96.

[10] Elston CW, Ellis IO. Pathological prognostic factors in breast cancer. I. The value of histological grade in breast cancer: experience from a large study with long-term follow-up. Histopathology 1991;19(5):403–10.

[11] Hudis CA, Barlow WE, Costantino JP, Gray RJ, Pritchard KI, Chapman JA, et al. Proposal for standardized definitions for efficacy end points in adjuvant breast cancer trials: the STEEP system. J Clin Oncol 2007;25(15):2127–32.

[12] van Rossum AGJ, Kok M, McCool D, Opdam M, Miltenburg NC, Mandjes IAM, et al. Independent replication of polymorphisms predicting toxicity in breast cancer patients randomized between dose-dense and docetaxel-containing adjuvant chemotherapy. Oncotarget 2017;8(69):113531–42.

[13] Shulman LN, Berry DA, Cirrincione CT, Becker HP, Perez EA, O'Regan R, et al. Comparison of doxorubicin and cyclophosphamide versus single-agent paclitaxel as adjuvant therapy for breast cancer in women with 0 to 3 positive axillary nodes: CALGB 40101 (Alliance). J Clin Oncol 2014;32(22):2311–7.

[14] Ohnstad HO, Borgen E, Falk RS, Lien TG, Aaserud M, Sveli MAT, et al. Prognostic value of PAM50 and risk of recurrence score in patients with early-stage breast cancer with long-term follow-up. Breast Cancer Res 2017;19(1):120.

[15] Liu S, Chapman JA, Burnell MJ, Levine MN, Pritchard KI, Whelan TJ, et al. Prognostic and predictive investigation of PAM50 intrinsic subtypes in the NCIC CTG MA.21 phase III chemotherapy trial. Breast Cancer Res Treat 2015;149(2):439–48.

[16] Garg PK, Deo SV, Kumar R, Shukla NK, Thulkar S, Gogia A, et al. Staging PET-CT scanning provides superior detection of lymph nodes and distant metastases than traditional imaging in locally advanced breast cancer. World J Surg 2016;40(8):2036–42.

[17] Koolen BB, Vrancken Peeters MJ, Aukema TS, Vogel WV, Oldenburg HS, van der Hage JA, et al. 18F-FDG PET/CT as a staging procedure in primary stage II and III breast cancer: comparison with conventional imaging techniques. Breast Cancer Res Treat 2012;131(1):117–26.

[18] Cardoso F, van't Veer LJ, Bogaerts J, Slaets L, Viale G, Delaloge S, et al. 70-gene signature as an aid to treatment decisions in early-stage breast cancer. N Engl J Med 2016;375(8):717–29.

[19] Simon R, Norton L. The Norton-Simon hypothesis: designing more effective and less toxic chemotherapeutic regimens. Nat Clin Pract Oncol 2006;3(8):406–7.

[20] Lambertini M, Ceppi M, Cognetti F, Cavazzini G, De Laurentiis M, De Placido S, et al. Dose-dense adjuvant chemotherapy in premenopausal breast cancer patients: a pooled analysis of the MIG1 and GIM2 phase III studies. Eur J Cancer 2017;71:34–42.

[21] Roche H, Fumoleau P, Spielmann M, Canon JL, Delozier T, Serin D, et al. Sequential adjuvant epirubicin-based and docetaxel chemotherapy for node-positive breast cancer patients: the FNCLCC PACS 01 Trial. J Clin Oncol 2006;24(36):5664–71.

[22] Anders CK, Hsu DS, Broadwater G, Acharya CR, Foekens JA, Zhang Y, et al. Young age at diagnosis correlates with worse prognosis and defines a subset of breast cancers with shared patterns of gene expression. J Clin Oncol 2008;26(20):3324–30.

[23] Nowak AK, Wilcken NR, Stockler MR, Hamilton A, Ghersi D. Systematic review of taxane-containing versus non-taxane-containing regimens for adjuvant and neoadjuvant treatment of early breast cancer. Lancet Oncol 2004;5(6):372–80.

[24] Mackey JR, Martin M, Pienkowski T, Rolski J, Guastalla JP, Sami A, et al. Adjuvant docetaxel, doxorubicin, and cyclophosphamide in node-positive breast cancer: 10-year follow-up of the phase 3 randomised BCIRG 001 trial. Lancet Oncol 2013;14(1):72–80.

[25] Azim Jr HA, de Azambuja E, Colozza M, Bines J, Piccart MJ. Long-term toxic effects of adjuvant chemotherapy in breast cancer. Ann Oncol 2011;22(9):1939–47.

[26] Eckhoff L, Feddersen S, Knoop AS, Ewertz M, Bergmann TK. Docetaxel-induced neuropathy: a pharmacogenetic case-control study of 150 women with early-stage breast cancer. Acta Oncol 2015;54(4):530–7.