MRI-guided optimisation of neoadjuvant chemotherapy duration in stage II–III HER2-positive breast cancer (TRAIN-3): a multicentre, single-arm, phase 2 study

*Anna van der Voort, Fleur M Louis, Mette S van Ramshorst, Rob Kessels, Ingrid A Mandjes, Inge Kemper, Mariette J Agterof, Wim A van der Steeg, Joan B Heijns, Marlies L van Bekkum, Ester J Siemerink, Philomeen M Kuijer, Astrid Scholten, Jelle Wesseling, Marie-Jeanne T F D Vrancken Peeters, Ritse M Mann, Gabe S Sonke, on behalf of the Dutch Breast Cancer Research Group**

Summary

Background Patients with stage II–III HER2-positive breast cancer have good outcomes with the combination of neoadjuvant chemotherapy and HER2-targeted agents. Although increasing the number of chemotherapy cycles improves pathological complete response rates, early complete responses are common. We investigated whether the duration of chemotherapy could be tailored on the basis of radiological response.

Methods TRAIN-3 is a single-arm, phase 2 study in 43 hospitals in the Netherlands. Patients with stage II–III HER2-positive breast cancer aged 18 years or older and a WHO performance status of 0 or 1 were enrolled. Patients received neoadjuvant chemotherapy consisting of paclitaxel (80 mg/m² of body surface area on day 1 and 8 of each 21 day cycle), trastuzumab (loading dose on day 1 of cycle 1 of 8 mg/kg bodyweight, and then 6 mg/kg on day 1 on all subsequent cycles), and carboplatin (area under the concentration time curve 6 mg/mL per min on day 1 of each 3 week cycle) and pertuzumab (loading dose on day 1 of cycle 1 of 840 mg, and then 420 mg on day 1 of each subsequent cycle), all given intravenously. The response was monitored by breast MRI every three cycles and lymph node biopsy. Patients underwent surgery when a complete radiological response was observed or after a maximum of nine cycles of treatment. The primary endpoint was event-free survival at 3 years; however, follow-up for the primary endpoint is ongoing. Here, we present the radiological and pathological response rates (secondary endpoints) of all patients who underwent surgery and the toxicity data for all patients who received at least one cycle of treatment. Analyses were done in hormone receptor-positive and hormone receptor-negative patients separately. This trial is registered with ClinicalTrials.gov, number NCT03820063, recruitment is closed, and the follow-up for the primary endpoint is ongoing.

Findings Between April 1, 2019, and May 12, 2021, 235 patients with hormone receptor-negative cancer and 232 with hormone receptor-positive cancer were enrolled. Median follow-up was 26·4 months (IQR 22·9–32·9) for patients who were hormone receptor-negative and 31·6 months (25·6–35·7) for patients who were hormone receptor-positive. Overall, the median age was 51 years (IQR 43–59). In 233 patients with hormone receptor-negative tumours, radiological complete response was seen in 84 (36%; 95% CI 30–43) patients after one to three cycles, 140 (60%; 53–66) patients after one to six cycles, and 169 (73%; 66–78) patients after one to nine cycles. In 232 patients with hormone receptor-positive tumours, radiological complete response was seen in 68 (29%; 24–36) patients after one to three cycles, 118 (51%; 44–57) patients after one to six cycles, and 138 (59%; 53–66) patients after one to nine cycles. Among patients with a radiological complete response after one to nine cycles, a pathological complete response was seen in 147 (87%; 95% CI 81–92) of 169 patients with hormone receptor-negative tumours and was seen in 73 (53%; 44–61) of 138 patients with hormone receptor-positive tumours. The most common grade 3–4 adverse events were neutropenia (175 [37%] of 467), anaemia (75 [16%]), and diarrhoea (57 [12%]). No treatment-related deaths were reported.

Interpretation In our study, a third of patients with stage II–III hormone receptor-negative and HER2-positive breast cancer had a complete pathological response after only three cycles of neoadjuvant systemic therapy. A complete response on breast MRI could help identify early complete responders in patients who had hormone receptor negative tumours. An imaging-based strategy might limit the duration of chemotherapy in these patients, reduce side-effects, and maintain quality of life if confirmed by the analysis of the 3-year event-free survival primary endpoint. Better monitoring tools are needed for patients with hormone receptor-positive and HER2-positive breast cancer.

Funding Roche Netherlands.

Lancet Oncol 2024; 25: 603–13

Published **Online** April 5, 2024 https://doi.org/10.1016/S1470-2045(24)00104-9

*The members of the Dutch Breast Cancer Research Group are listed in the appendix (p 2)

Department of Medical Oncology (A van der Voort MD, F M Louis MD, M S van Ramshorst MD, I Kemper MANP, Prof G S Sonke MD), **Department of Biometrics** (R Kessels PhD, I A Mandjes MSc), **Department of Radiation** (A Scholten MD), **Division of Molecular Pathology and Department of Pathology** (Prof J Wesseling MD), **Department of Surgery** (Prof M-J T F D Vrancken Peeters MD), **Department of Radiology** (R M Mann MD), **Netherlands Cancer Institute, Amsterdam, Netherlands; Department of Medical Oncology, St Antonius Hospital, Nieuwegein, Netherlands** (M J Agterof MD); **Department of Medical Oncology, Isala, Zwolle, Netherlands** (W A van der Steeg MD); **Department of Medical Oncology, Amphia, Breda, Netherlands** (J B Heijns MD); **Department of Medical Oncology, Reinier de Graaf Gasthuis, Delft, Netherlands** (M L van Bekkum MD); **Department of Medical Oncology, Ziekenhuisgroep Twente, Hengelo, Netherlands**; (E J Siemering MD); **Department of Medical Oncology, Spaarne Gasthuis Hoofdorp, Netherlands** (P M Kuijer MD); **Department of Medical Oncology** (Prof G S Sonke) **and Department of Surgery** (Prof M-J T F D Vrancken Peeters), **Amsterdam University Medical Centre, Amsterdam, Netherlands; Department of**

Medical Imaging, Radboud University Medical Center, Amsterdam, Netherlands (R M Mann); **Department of Pathology, University Medical Centre, Leiden, Netherlands** (Prof J Wessling)

Correspondence to: Prof Gabe S Sonke, Department of Medical Oncology, Netherlands Cancer Institute, Amsterdam 1066 CX, Netherlands
g.sonke@nki.nl

See Online for appendix

Research in context

Evidence before this study

We performed a PubMed search using the terms "HER2 positive breast cancer", "neoadjuvant", "trastuzumab", AND "pertuzumab" for articles of clinical trials published in English up to Dec 31, 2017, from database inception assessing the response to a neoadjuvant chemotherapy regimen of different durations in patients with early stage HER2-positive breast cancer. Neoadjuvant treatment of HER2-positive breast cancer mostly consisted of six cycles of chemotherapy plus HER2-targeted agents. However, pathological complete responses have been reported after just one cycle of treatment, whereas the highest pathological complete response rates were reported in patients who receive neoadjuvant chemotherapy for nine cycles. Baseline characteristics of this patient population were not sufficiently accurate to tailor treatment duration. Therefore, we searched PubMed for articles published up to Dec 31, 2017, from database inception in English using the terms "response", "HER2 positive breast cancer", "neoadjuvant", AND "MRI" OR "FDG-PET" OR "PET-CT" OR "ultrasound". Breast MRI has been shown to accurately identify complete responders after neoadjuvant chemotherapy.

Added value of this study

TRAIN-3 is the first trial to our knowledge to use breast MRI to tailor neoadjuvant chemotherapy duration in stage II–III HER2-positive breast cancer and provides detailed information on response percentages after three, six, and nine cycles. Our study showed that approximately a third of patients with stage II–III hormone receptor-negative HER2-positive breast cancer had a complete pathological response after only three cycles of neoadjuvant systemic therapy. This study provides estimates of the negative predictive value of MRI breast-based response evaluation in a multicentre study using a predefined definition of radiological complete response. This study also provides new information about dose-related toxicity.

Implications of all the available evidence

There are now five trials showing that a substantial percentage of patients with early-stage HER2-positive breast cancer can have a complete pathological response with fewer cycles of chemotherapy: NEOSPHERE, WSG-TP-II, WSG-ADAPT-HER2+/HR-, PHERGAIN, and TRAIN-3. If the current results are confirmed in the analysis of the primary endpoint of event-free survival, treatment duration can be tailored using breast MRI to better select complete responders in patients with hormone receptor-negative HER2-positive breast cancer. However, there is still a need to improve the selection of patients upfront and to optimise response evaluation methods in hormone receptor-positive disease. In addition, TRAIN-3 confirms previous findings from TRAIN-2 that more patients have a pathological complete response when the duration of neoadjuvant chemotherapy is extended up to nine cycles, which could reduce the need for adjuvant trastuzumab emtansine.

Introduction

The introduction of HER2-directed agents in the treatment of HER2-positive breast cancer has improved outcomes for patients.[1] A pathological complete response after neoadjuvant systemic therapy predicts excellent long-term outcome and can be used to tailor treatment.[2–4] Pathological complete response rates up to 55% in hormone receptor-positive and HER2-positive tumours and 90% in hormone receptor-negative and HER2-positive tumours have been described.[5,6] Most neoadjuvant treatment regimens consist of six cycles of chemotherapy combined with dual HER2 blockade consisting of trastuzumab and pertuzumab. Although longer duration of chemotherapy, up to nine cycles, is associated with the highest pathological complete response rates, a subset of tumours show a complete response sometimes as early as 14 days after starting treatment.[5,7] Because each additional treatment cycle increases toxicity,[5,8–11] monitoring the treatment response to identify when patients are likely to have had a complete response can optimise therapy.

MRI of the breast after neoadjuvant treatment correctly identifies patients with a pathological complete response in approximately three quarters of cases.[12] However, the negative predictive values differ between hormone receptor subgroups, with a negative predictive value of approximately 84% in hormone receptor-negative breast cancer and 52% in hormone receptor-positive breast cancer.[12–14] MRI might also indicate a complete response in the axillary lymph nodes. Dedicated axillary protocols in a primary setting have shown a negative predictive value of more than 95%,[15] but detecting residual disease in the lymph nodes is less accurate in a post-systemic therapy setting. In a small study, axillary response evaluation in lymph node-positive disease using an ultrasound and fine needle aspiration correctly identified a nodal complete response in 47% of patients.[16] A targeted biopsy of a marked lymph node could further improve response evaluation if limited to patients showing nodal involvement at baseline and a radiological complete response in the breast.

We hypothesised that monitoring for a complete MRI response during neoadjuvant treatment could be used to establish the optimal duration of chemotherapy in patients with stage II–III HER2-positive breast cancer. Here, we report data on radiological complete response, pathological complete response, and toxicity from the TRAIN-3 study.

Methods

Study design and participants

The TRAIN-3 study is a multicentre, non-comparative, single-arm, phase 2 study performed in 43 hospitals in

the Netherlands. We included patients with stage II–III HER2-positive breast cancer. HER2 status was established in invasive breast cancer cells at a baseline core biopsy and defined as overexpression or amplification, or both, of HER2 according to the American Society of Clinical Oncology and College of American Pathologists 2013 guideline (which was not centrally reviewed).[17] Hormone receptor negativity was defined as an oestrogen and progesterone receptor expression of less than 10%. Hormone receptor positvity was defined as an oestrogen or progesterone receptor, or both, expression of 10% or more. Other eligibility criteria were an age of 18 years or older, WHO performance status of 0 or 1, left ventricular ejection fraction of 50% or higher, adequate organ function as established by laboratory assessment, and radiological evaluability per protocol. Patients were excluded if they were pregnant or had a history of cancer less than 5 years ago or having previously been administered chemotherapy. Before enrolment, all patients underwent [18F]fluorodeoxyglucose (FDG)-PET to check for distant metastases. A full list of the eligibility criteria can be found in the study protocol (appendix). All patients underwent breast MRI (minimal quality requirements are in the study protocol). Staging included an ultrasound of the axilla and peri-clavicular region in the case of suspicious axillary lymph nodes (10 mm or more in the short axis or with thickening of the cortex). Lymph node-positive disease was confirmed with fine needle aspiration and a marker was placed in a confirmed positive node. Before the initiation of chemotherapy, a marker was also placed at the site of the primary breast tumour. The medical ethics committee of the Netherlands Cancer Institute approved the study protocol, including all amendments. All patients provided written informed consent before enrolment.

Procedures

All patients received neoadjuvant treatment once every 3 weeks consisting of paclitaxel (80 mg/m² on day 1 and 8 of each 3 week cycle), trastuzumab (loading dose on day 1 of cycle 1 of 8 mg/kg, and then 6 mg/kg on day 1 of each subsequent cycle), and carboplatin (area under the concentration time curve 6 mg per min per mL on day 1 of each 3 week cycle), plus pertuzumab (loading dose on day 1 of cycle 1 of 840 mg, and then 420 mg on day 1 of each subsequent cycle) intravenously, with a maximum of nine cycles (figure 1). Subcutaneous administration of trastuzumab (600 mg) instead of the intravenous administration detailed earlier and dividing carboplatin as 3 mg/mL per min on days 1 and 8 was allowed per hospital preference. Assessment of laboratory results was performed before each cycle. Left ventricular ejection fraction was evaluated every 3 months during anti-HER2 treatment with the same method (echocardiography, multigated acquisition scan, or MRI) used for the baseline assessment. Grade 3 and worse adverse events were registered using the Common Terminology Criteria for Adverse Events (version 5.0) at each cycle and study visit. Dose adjustment criteria are described in the study protocol and consisted of, but were not limited to, treatment delay and initiation of growth factor support with G-CSF in the case of isolated grade 3 neutropenia. When grade 3 neutropenia occurs during G-CSF treatment or is combined with thrombocytopenia grade 2, a dose reduction of 25% is warranted. Pertuzumab was permanently stopped in the case of grade 4 diarrhoea, and both HER2 agents (pertuzumab and trastuzumab) were withheld in the case of a left ventricular ejection fraction decrease of more than 10% to less than 50% and permanently in the case of symptomatic cardiac toxicity. We did not routinely collect sex, gender, race, or ethnicity data.

Responses were monitored every three cycles using multiparametric breast MRI. The MRI scan protocol

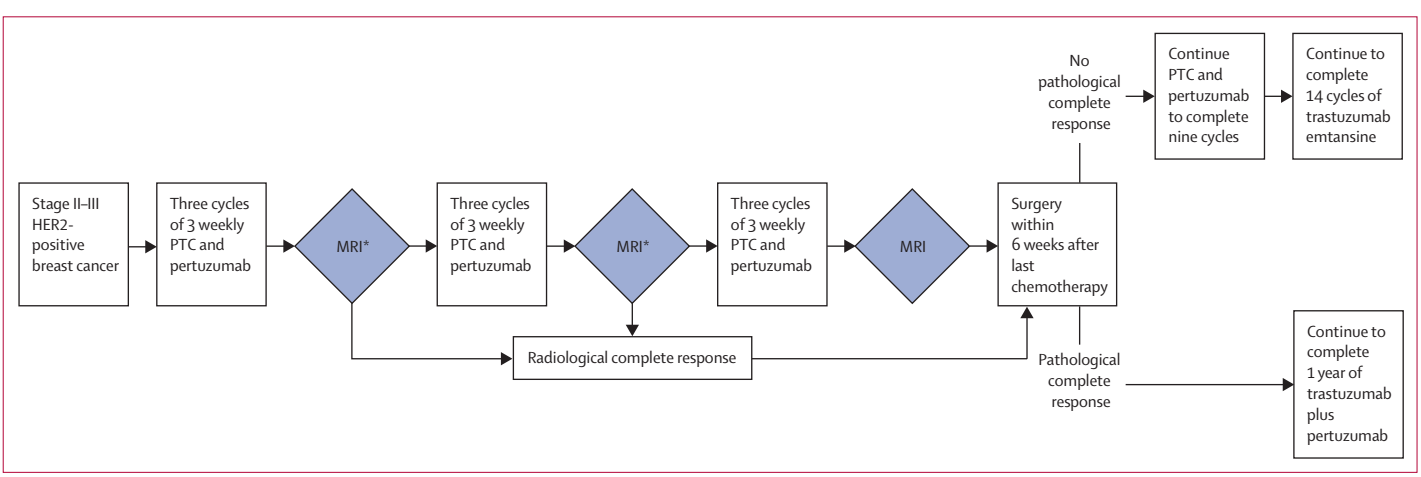

Figure 1: **Study procedure**
HER2=human epidermal growth factor 2. PTC=paclitaxel, trastuzumab, and carboplatin. *If a radiological complete remission on breast MRI was observed in patients with clinical lymph node-positive disease, a targeted biopsy of the at-baseline marked lymph node was warranted—ie, an ultrasound guided targeted biopsy (fine needle aspiration or core biopsy). In hormone receptor-positive disease, a non-PCR could also be detected using vacuum-assisted core biopsies.

included a T1 dynamic sequence with an isotropic spatial resolution of 1 mm or less and lasted at least 7 min with a minimum of four timepoints after contrast. A complete remission was defined as the absence of pathological contrast enhancement in the original tumour region. Other minimal contrast enhancement in the tumour bed similar to (or less than) surrounding or contralateral normal breast tissue was considered physiological. Small foci in the original tumour bed with residual pathological enhancement did not qualify as complete remission. Complete resolution of the mass was not required. Response evaluation was performed locally by trained radiologists and followed a specific radiology protocol with features including enhancement and response patterns. In the case of a radiological complete remission on breast MRI in patients who were lymph node positive, fine needle aspiration or 18-G core biopsy of the lymph node that was marked at baseline was performed. At least two attempts were required in case of inconclusive results. After registration and reimbursement of adjuvant trastuzumab emtansine in the absence of pathological complete response, the fourth amendment of the protocol (in May 2020) added vacuum-assisted core biopsies in patients with hormone receptor-positive tumours to confirm radiological complete response in the breast. Nine to 12 core biopsies (9 gauge) were obtained, and the initially placed marker in the tumour needed to be removed as per the protocol. This method was included in the study protocol because the early cessation of neoadjuvant therapy could result in potential overtreatment with trastuzumab emtansine in patients with residual invasive disease at the time of surgery, who would have had a complete pathological response if they had completed all neoadjuvant cycles.

We referred patients for surgery in the case of radiological complete response after three or six cycles or after completing the maximum of nine cycles. Patients underwent breast-conserving surgery or mastectomy within 6 weeks of the last chemotherapy. Axillary staging consisted of a sentinel node procedure, selective removal of the initially positive and marked lymph node, or an axillary lymph node dissection, or a combination.

In the case of a pathological complete response, adjuvant therapy consisted of completing 1 full year of trastuzumab and pertuzumab, without chemotherapy. In the case of residual invasive disease after three or six neoadjuvant treatment cycles, patients completed the nine cycles of adjuvant paclitaxel, trastuzumab, and carboplatin plus pertuzumab. Patients with residual invasive disease received 14 cycles of trastuzumab emtansine (intravenously, 3·6 kg/mg, once every 3 weeks), which was added by amendment in May, 2020, after market authorisation and reimbursement. Patients without a pathological complete response who were included before this amendment could still opt to replace trastuzumab and pertuzumab with trastuzumab emtansine. Targeted therapy was given concomitantly with radiotherapy if indicated.

Outcomes

The primary endpoint of TRAIN-3 was 3-year event-free survival defined as the interval from registration to the earliest occurrence of disease progression resulting in inoperability, invasive locoregional recurrence, distant metastases, or death from any cause, whichever came first. The results for the primary endpoint will be reported after 700 patient-years of follow-up. The key secondary endpoints were a pathological complete response defined as the absence of invasive tumour cells in the breast and axilla at surgery or vacuum-assisted core biopsies, irrespective of the presence of in-situ lesions; radiological complete response defined as the absence of pathological enhancement in the original tumour region on breast MRI with negative fine needle aspiration or core biopsy of a marked lymph node in the case of node positive disease; number of neoadjuvant chemotherapy cycles administered; number of radical resections; and overall survival defined as the time from registration to death from any cause. Safety outcome measures include adverse events grade 3 or higher (Common Terminology Criteria for Adverse Events version 5.0), neuropathy, cardiotoxicity grade 2 or worse, incidence of symptomatic and asymptomatic left ventricular systolic dysfunction, decline in left ventricular ejection fraction requiring treatment or leading to the discontinuation of HER2-directed agents, or a decrease of 10 percentage points or more from baseline to a left ventricular ejection fraction of less than 50%. Patient-reported outcome measures include quality of life scored using the European Organisation For Research And Treatment of Cancer quality of life core questionnaire and quality of life breast cancer module. Finally, the exploratory endpoints of TRAIN-3 included mechanisms of resistance to treatment established on the basis of immunohistochemistry; whole-exome sequencing plus proteomics on pretreatment biopsies, on-treatment biopsies, and residual tumours after treatment; sequential circulating free DNA; tumour-educated platelets; and the effect of in vivo biotransformation of trastuzumab and pertuzumab on the treatment response. The analysis for the primary endpoint for each hormone receptor subgroup is planned after a follow-up of 700 patient-years. Analysis of the other secondary outcome measures are ongoing and will be reported separately, along with updated safety data. Other exploratory endpoints will be reported after publication of the primary endpoint.

Statistical analysis

All analyses were performed for hormone receptor negative and hormone receptor positive subgroups separately. Subgroup analyses were also performed for patients who were lymph node positive (post hoc) versus

lymph node negative and immunohistochemistry subgroups (2+ *vs* 3+; prespecified). All patients who underwent surgery were included in the response analysis. All patients with at least one cycle of study treatment were included in the safety analysis. We hypothesised that chemotherapy duration could be safely tailored in patients with a pathological complete response while maintaining similar rates of event-free survival. The maximum number of events were calculated using the exact conditional Poisson distribution on the basis of 700 patient-years of follow-up per hormone receptor subgroup. We used a 3-year event-free survival of 88% as the reference for patients who were hormone receptor-negative and HER2-positive, and 90% for patients who were hormone receptor-positive and HER2-positive.[1,18,19] Therefore, the study would be declared successful if no more than 38 events occur at 700 patient-years of follow-up in patients who are hormone receptor-negative with a power of 93% probability. For hormone receptor-positive disease there was a 96% probability that the study was correctly declared successful if no more than 34 events were observed at 700 patient-years. The minimum required sample size was 220 patients per hormone receptor subgroup, based on the expected accrual over 4 years, and it was anticipated that 5% of patients would be not evaluable for the primary endpoint. We performed one protocol-defined interim analysis per hormone receptor subgroup for the primary efficacy endpoint after the follow-up of 150 patient-years, which did not lead to a premature closure of the trial (appendix). The report of secondary outcomes was of a descriptive nature and included the following secondary outcome measures: pathological complete response, radiological complete response, radical resections, grade 3–4 adverse events during neoadjuvant treatment, neuropathy and ejection fraction decreases of grade 2 or higher (<50% or a decrease of ≥10 percentage points of lower ventricular ejection fraction), and symptomatic heart failure. Post-hoc secondary outcome measures included radiological complete responses on breast MRI, not taking axillary biopsies into account. The statistical analyses were done using SAS (version 9.4) and R (version 4.2.0). This study is registered with ClinicalTrials.gov, number NCT03820063.

Role of the funding source

The funder of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report.

Results

Between April 1, 2019, and May 12, 2021, 493 patients were assessed for eligibility and 472 were registered for study participation (figure 2). Five protocol violations occurred directly after study registration, leading to non-protocol treatment and therefore exclusion. 235 patients with hormone receptor-negative cancer and 232 patients with hormone receptor-positive tumours were enrolled and treated across 43 hospitals (appendix p 2). At the time of data cutoff (May 24, 2023) median follow-up was 26·4 months (IQR 22·9–32·9) for patients who were hormone receptor-negative and 31·6 months (25·6–35·7) for patients who were hormone receptor-positive. Within the hormone receptor-negative subgroup, two patients had a pathological complete response of the breast but incomplete axillary staging at surgery and were therefore excluded from the pathological complete response analysis. Table 1 shows the baseline patient and tumour characteristics per hormone receptor stratum. Baseline MRI characteristics are shown in the appendix (p 4). Overall, the median age was 51 years (IQR 43–59) and 212 (45%) of 467 patients were post-menopausal. Most patients had a clinical tumour stage 2–3 (405 [87%] of 467), had lymph node metastasis (279 [60%]), had tumour grade III (247 [53%]), were unifocal on baseline MRI (254 [54%]), and most had a HER2 immunohistochemistry score of 3+ (385 [82%]). Higher numbers of patients with hormone receptor-negative tumours had a HER2 immunohistochemistry score of 3+ (207 [88%] of 235 *vs* 178 [77%] of 232) and tumour grade three (148 [63%] 235 *vs* 99

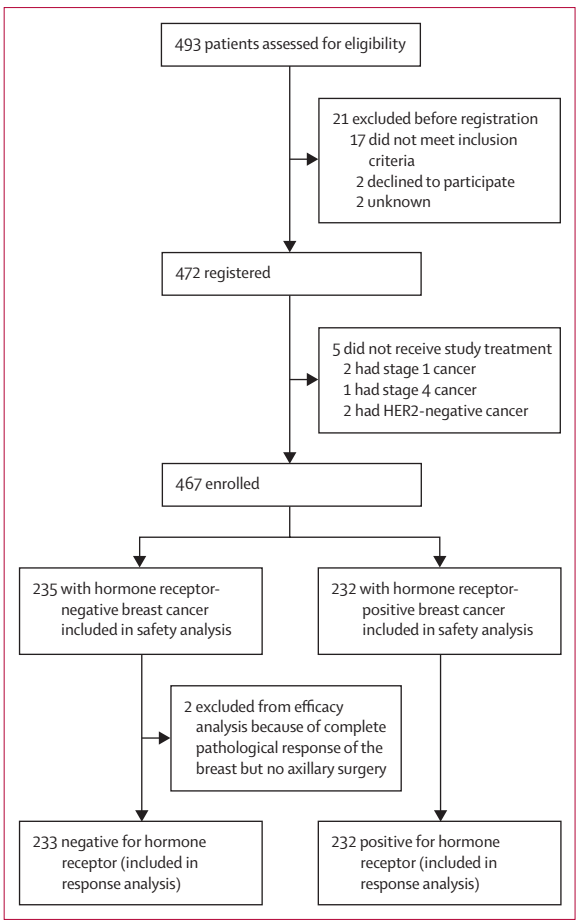

Figure 2: **Trial profile**

	Total (N=467)	Oestrogen or progesterone receptor, or both, <10% (N=235)	Oestrogen or progesterone receptor, or both, ≥10% (N=232)
Age, years	51 (43–59)	52 (43–59)	50 (43–58)
Menopausal status			
Premenopausal or perimenopausal	240 (51%)	112 (48%)	128 (55%)
Postmenopausal	212 (45%)	116 (49%)	96 (41%)
Unknown	15 (3%)	7 (3%)	8 (3%)
Clinical tumour stage			
cTx, cT0, or cT1*	49 (10%)	25 (11%)	24 (10%)
cT2 to T3	405 (87%)	205 (87%)	200 (86%)
cT4	13 (3%)	5 (2%)	8 (3%)
Clinical nodal status			
Negative	187 (40%)	93 (40%)	94 (41%)
Positive	279 (60%)	142 (60%)	137 (59%)
Unknown (cNx)	1 (<1%)	0	1 (<1%)
Clinical stage of breast cancer			
II	322 (69%)	156 (66%)	166 (72%)
III	145 (31%)	79 (34%)	66 (28%)
HER2 immunohistochemistry score			
2+ and in-situ hybridisation positive	57 (12%)	14 (6%)	43 (19%)
3+	385 (82%)	207 (88%)	178 (77%)
Unknown and in-situ hybridisation positive	25 (5%)	14 (6%)	11 (5%)
Tumour grade			
I	8 (2%)	0	8 (3%)
II	205 (44%)	83 (35%)	122 (53%)
III	247 (53%)	148 (63%)	99 (43%)
Unknown	7 (1%)	4 (2%)	3 (1%)
Histology			
No special type†	446 (96%)	225 (96%)	221 (95%)
Lobular	11 (2%)	4 (2%)	7 (3%)
Other	10 (2%)	6 (3%)	4 (2%)

Data are n (IQR) or n (%). For clinical tumour stage: Tx means primary tumour cannot be assessed; T0 means no evidence of primary tumour; T1 means tumour is ≤20 mm at its greatest dimension; T2 means tumour is >20 mm to ≤50 mm at its greatest dimension; T3 means tumour is >50 mm at its greatest dimension; T4 means tumour of any size with direct extension to the chest wall or to the skin (ulceration or macroscopic nodules), or both (note: invasion of the dermis alone does not qualify as T4). *Including three patients with occult (T0 or Tx) breast cancer with an oestrogen or progesterone receptor, or both, of less than 10%. †Previously known as invasive ductal breast cancer. Percentages might not add up due to rounding.

Table 1: **Baseline characteristics**

[43%] of 232) compared with hormone receptor-positive tumours.

160 (34%) of 467 patients received up to three cycles of neoadjuvant chemotherapy, 147 (31%) patients received four to six cycles, and 160 (34%) patients received seven to nine cycles (appendix p 5). Reasons for early neoadjuvant chemotherapy discontinuation were a radiological complete response in 260 (56%) patients, toxicity in 48 (10%) patients, patient wish in 14 (3%) patients, and other non-protocol reasons in four (1%) patients. One (<1%) patient stopped neoadjuvant chemotherapy early because of disease progression and underwent early surgery (appendix p 5). Of the patients who received less than three cycles, one (<1%) patient received only one cycle of carboplatin and paclitaxel and stopped due to toxicity, and although this patient showed a radiological complete response, they did not reach a pathological complete response. Pertuzumab was stopped prematurely because of toxicity in 20 (4%) patients and trastuzumab in seven (1%) patients. After completing neoadjuvant systemic treatment, 325 (70%) patients underwent breast-conserving surgery. Surgical resection was radical in 231 (98%) of 235 patients with hormone receptor-negative tumours and 217 (94%) of 232 patients with hormone receptor-positive tumours.

84 (36% [95% CI 30–43]) of 233 patients with hormone receptor-negative and HER2-positive tumours had a radiological complete response after one to three cycles, 140 (60% [53–66]) patients had a complete response after one to six cycles, and 169 (73% [66–78]) patients had a complete response after one to nine cycles. 68 (29% [24–36]) of 232 patients with hormone receptor-positive and HER2-positive tumours had a radiological complete response after one to three cycles, 118 (51% [44–57]) patients had a complete response after one to six cycles, and 138 (59% [53–66]) patients had a complete response after one to nine cycles (figure 3). Post-hoc analyses of response rates using breast MRI only and targeted lymph node biopsies are presented in the appendix (pp 6–7).

Among 169 patients with hormone receptor-negative tumours who showed a radiological complete response after one to nine treatment cycles, 147 (87% [81–92]) had a pathological complete response. 74 (88% [79–94]) of 84 patients with an early radiological complete response after three cycles also had a pathological complete response (figure 3). Among 138 patients with hormone receptor-positive tumours that had a radiological complete response, 73 (53% [44–61]) patients had a pathological complete response. Cumulative pathological complete response rates including patients undergoing surgery in the absence of a radiological complete response were 80% (187 of 233) in patients with hormone receptor-negative tumours and 43% (100 of 232) in patients with hormone receptor-positive tumours. Post-hoc explorative subgroup analyses including pathological complete response rates in lymph-node-positive disease and in immunohistochemistry subgroups are reported in the appendix (pp 8–9). Pathological complete response rates were higher in patients with a HER2 immunohistochemistry score of 3+ compared with a score of 2+ in the hormone receptor-positive subgroup.

270 (58%) of 467 participants had one or more grade 3 adverse events and 37 (8%) patients had one or more grade 4 adverse event (table 2). The frequency of grade 3–4 adverse events increased with the increasing number of chemotherapy cycles. After one to three cycles, 69 (43%) of 160 patients had grade 3–4 adverse events; in patients who received four to six cycles, 109 (74%) of 147 patients had grade 3–4 adverse events; and after seven to nine cycles, 129 (81%) of 160 patients had grade 3–4

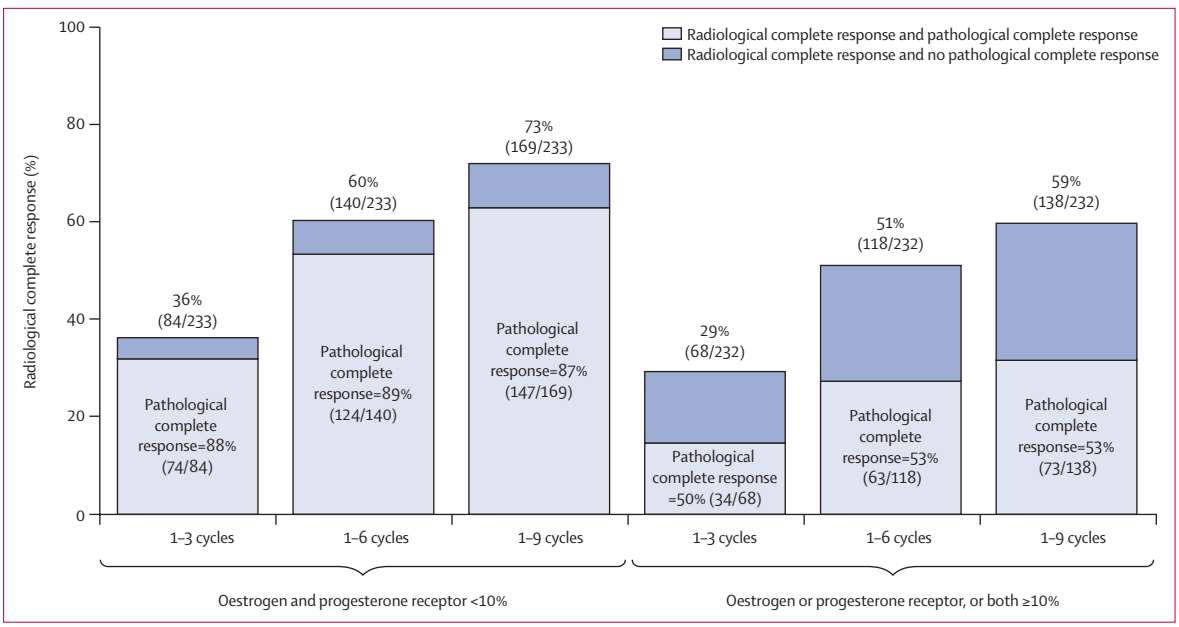

Figure 3: **Cumulative response rates according to hormone receptor status**
Radiological complete response on MRI breast scan as concluded by local radiologist and targeted lymph node biopsy in the case of lymph node positive disease at baseline. Pathological complete response was defined as the absence of invasive tumour cells in the breast and axilla irrespective of the presence of in-situ lesions.

adverse events. The most common grade 3–4 adverse events were neutropenia (175/467 [37%]), anaemia (75/467 [16%]), diarrhoea (57/467 [12%]), thrombocytopenia (40/467 [9%]), and hypokalaemia (34/467 [7%]; table 2). Grade 2 peripheral sensory neuropathy was observed in 85 (18%) of 467 patients, grade 3 in 27 (6%) patients, and grade 4 in one (<1%) patient. Grade 2 or worse decreased ejection fraction was seen in 45 (10%) patients. In 209 (45%) patients at least one dose reduction of paclitaxel or carboplatin, or both, was performed. The most common dose-limiting adverse events were peripheral neuropathy (193 [35%] of 551 events), neutropenia (82 [15%] of 551), thrombocytopenia (80 [15%] of 551) and diarrhoea (58 [11%] of 551). No treatment-related deaths were reported.

Discussion

TRAIN-3 showed that 87% of patients with hormone receptor-negative and HER2-positive breast cancer with a radiological complete response on MRI-based response evaluation had a pathological complete response. If confirmed in the event-free survival analysis, one in three patients could be treated with only three cycles of chemotherapy using MRI-guided treatment optimisation. We showed that patients treated with fewer cycles of neoadjuvant chemotherapy also had fewer grade 3–4 adverse events, although the overall toxicity of this strategy needs to be confirmed, including the use of adjuvant treatment. In addition, continuing neoadjuvant chemotherapy up to nine cycles in the case of no radiological complete response increased pathological complete response rates. Notably, in hormone receptor-positive and HER2-positive breast cancer, only 53% of patients had a pathological complete response out of those who had a radiological complete response, and better monitoring tools are required for these patients.

Patients with breast cancer with a pathological complete response after neoadjuvant treatment have excellent event-free survival compared with patients with residual invasive disease.[2–4] Therefore, treatment tailoring strategies using imaging-based response evaluation need high negative predictive values. PHERGAIN is a chemotherapy de-escalation trial using [^{18}F]FDG-PET-CT. A response was defined as an at least 40% maximum standardised uptake value decrease after two cycles of docetaxel, carboplatin, and dual HER2 blockade, or dual HER2 blockade only. In the group not given chemotherapy, a pathological complete response rate of 38% in responders was observed. This study found a 98·8% 3-year invasive-disease-free survival rate in patients treated completely without chemotherapy, paving the way for further image-guided treatment optimisation.[20] However, 62% of the patients who responded to [^{18}F]FDG-PET-CT did not have a pathological complete response, and chemotherapy was delayed in these patients. Using an imaging method with a higher negative predictive value could prevent this delay in treatment with chemotherapy. Awaiting the comparison data of MRI versus [^{18}F]FDG-PET-CT of the PHERGAIN trial, the TRAIN-3 study shows promising results for patients with hormone receptor-negative breast cancer in early complete responders by breast MRI. The predictive value of MRI could potentially be further increased by using volume-based response

	1–3 cycles (N=160)			4–6 cycles (N=147)			7–9 cycles (N=160)		
	Grade 2	Grade 3	Grade 4	Grade 2	Grade 3	Grade 4	Grade 2	Grade 3	Grade 4
Any	..	64 (40%)	5 (3%)	..	96 (65%)	13 (9%)	..	110 (69%)	19 (12%)
Neutropenia	..	27 (17%)	3 (2%)	..	49 (33%)	5 (3%)	..	79 (49%)	12 (8%)
Anaemia	..	5 (3%)	0	..	33 (22%)	0	..	37 (23%)	0
Diarrhoea	..	20 (13%)	1 (1%)	..	22 (15%)	1 (1%)	..	13 (8%)	0
Thrombocytopenia	..	2 (1%)	0	..	17 (12%)	1 (1%)	..	15 (9%)	5 (3%)
Hypokalaemia	..	9 (6%)	0	..	9 (6%)	1 (1%)	..	11 (7%)	4 (3%)
Peripheral sensory neuropathy*	3 (2%)	4 (3%)	0	30 (20%)	9 (6%)	1 (1%)	52 (33%)	14 (9%)	0
Hypomagnesemia	..	2 (1%)	0	..	5 (3%)	2 (1%)	..	10 (6%)	0
Leukopenia	..	3 (2%)	0	..	3 (2%)	0	..	9 (6%)	0
Peripheral motor neuropathy*	1 (1%)	0	0	4 (3%)	0	0	8 (5%)	0	0
Fatigue	..	0	0	..	9 (6%)	0	..	2 (1%)	0
Hypertension	..	3 (2%)	0	..	4 (3%)	0	..	3 (2%)	0
Alanine aminotransferase increased	..	3 (2%)	0	..	4 (3%)	0	..	2 (1%)	0
Stomatitis	..	2 (1%)	0	..	3 (2%)	0	..	4 (3%)	0
Nausea	..	2 (1%)	0	..	3 (2%)	0	..	4 (3%)	0
γ-glutamyl transferase increased	..	3 (2%)	0	..	1 (1%)	2 (1%)	..	2 (1%)	0
Allergic reaction	..	3 (2%)	0	..	3 (2%)	0	..	1 (1%)	0
Dehydration	..	2 (1%)	1 (1%)	..	4 (3%)	0	..	0	0
Ejection fraction decreased†	10 (6%)	2 (1%)	0	10 (7%)	3 (2%)	0	19 (12%)	1 (1%)	0
Gastrointestinal disorders (other)	..	1 (1%)	0	..	2 (1%)	0	..	3 (2%)	0
Malaise	..	2 (1%)	0	..	2 (1%)	0	..	2 (1%)	0
Vomiting	..	2 (1%)	0	..	2 (1%)	0	..	2 (1%)	0
Febrile neutropenia	..	2 (1%)	0	..	1 (1%)	0	..	2 (1%)	0
Fever	..	1 (1%)	0	..	1 (1%)	0	..	3 (2%)	0
Infection	..	0	0	..	2 (1%)	0	..	3 (2%)	0
Surgical and medical procedures	..	3 (2%)	0	..	1 (1%)	0	..	1 (1%)	0
Syncope	..	0	0	..	4 (3%)	0	..	1 (1%)	0
Sepsis	..	0	0	..	0	2 (1%)	..	2 (1%)	0
Wound infection	..	1 (1%)	0	..	0	0	..	3 (2%)	0
Infusion-related reaction	..	1 (1%)	0	..	2 (1%)	0	..	0	0
Acute kidney injury	..	1 (1%)	0	..	1 (1%)	0	..	0	0
COVID-19 positive	..	1 (1%)	0	..	0	0	..	1 (1%)	0
Dyspnoea	..	1 (1%)	0	..	1 (1%)	0	..	0	0
Hypocalcaemia	..	0	0	..	0	0	..	2 (1%)	0
Postoperative haemorrhage	..	0	0	..	1 (1%)	0	..	1 (1%)	0
Abdominal pain	..	0	0	..	0	0	..	1 (1%)	0
Alkaline phosphatase increased	..	1 (1%)	0	..	0	0	..	0	0
Anaphylaxis	..	0	0	..	1 (1%)	0	..	0	0
Aspartate aminotransferase increased	..	0	0	..	0	0	..	1 (1%)	0
Bilirubin increased	..	1 (1%)	0	..	0	0	..	0	0
Catheter related infection	..	1 (1%)	0	..	0	0	..	0	0
Cardiac chest pain	..	0	1 (1%)	..	0	0	..	0	0
Colitis	..	0	0	..	1 (1%)	0	..	1 (1%)	0
Creatinine increased	..	0	0	..	0	0	..	1 (1%)	0
Desaturation (oxygen)	..	0	0	..	0	0	..	1 (1%)	0
Dizziness	..	0	0	..	0	0	..	1 (1%)	0
Duodenal ulcer	..	0	0	..	0	0	..	1 (1%)	0
Epistaxis	..	0	0	..	1 (1%)	0	..	0	0
Fracture	..	0	0	..	1 (1%)	0	..	0	0
Reflux (gastroesophageal reflux disease)	..	1 (1%)	0	..	0	0	..	0	0

(Table 2 continues on next page)

	1–3 cycles (N=160)			4–6 cycles (N=147)			7–9 cycles (N=160)		
	Grade 2	Grade 3	Grade 4	Grade 2	Grade 3	Grade 4	Grade 2	Grade 3	Grade 4
(Continued from previous page)									
Generalised muscle weakness	..	0	0	..	0	0	..	1 (1%)	0
Headache	..	0	0	..	1 (1%)	0	..	0	0
Haematoma	..	1 (1%)	0	..	0	0	..	0	0
Haemorrhoids	..	0	0	..	1 (1%)	0	..	0	0
Hyperkalaemia	..	1 (1%)	0	..	0	0	..	0	0
Hyponatraemia	..	1 (1%)	0	..	0	0	..	0	0
Hypophosphataemia	..	1 (1%)	0	..	0	0	..	0	0
Immune systems disorders	..	0	0	..	1 (1%)	0	..	0	0
Mucositis	..	1 (1%)	0	..	0	0	..	0	0
Myositis	..	0	0	..	0	0	..	1 (1%)	0
Other neoplasms	..	1 (1%)	0	..	0	0	..	0	0
Obesity	..	1 (1%)	0	..	0	0	..	0	0
Pain in extremity	..	0	0	..	1 (1%)	0	..	0	0
Pruritus	..	0	0	..	1 (1%)	0	..	0	0
Psychological overload	..	0	0	..	1 (1%)	0	..	0	0
Radiculitis	..	0	0	..	0	0	..	1 (1%)	0
Renal calculi	..	0	0	..	0	0	..	1 (1%)	0
Seroma	..	0	0	..	0	0	..	1 (1%)	0
Skin infections	..	0	0	..	0	0	..	1 (1%)	0
Spinal cord haemorrhage	..	0	0	..	1 (1%)	0	..	0	0
Thromboembolic event	..	1 (1%)	0	..	0	0	..	1 (1%)	0
Tinnitus	..	0	0	..	1 (1%)	0	..	0	0
Urinary tract infection	..	0	0	..	0	0	..	1 (1%)	0
Weight loss	..	0	0	..	0	0	..	1 (1%)	0

Data are n (%). Grade 1–2 adverse events were not recorded, except for sensory and motor peripheral neuropathy and ejection fraction decrease. No grade 5 events were reported. Data are all grade 3–4 adverse events between registration and 30 days after surgery. *Grade 2 sensory and motor peripheral neuropathy were recorded per protocol. †Grade 2 ejection fraction decrease (to less than 50% or a more than 10% drop from baseline, or both) was recorded per protocol. One symptomatic (grade 2) restrictive cardiomyopathy was reported without signs of heart failure.

Table 2: **Adverse events grade per total number of neoadjuvant chemotherapy cycles**

measurements as shown by the I-SPY consortium.[21,22] Additionally, for [18F]FDG-PET-CT, the cutoff percentage maximum standardised uptake value decrease in response evaluation or the timing of the scan could potentially still be optimised to select complete responders. Nevertheless, both techniques do not have sufficient accuracy in hormone receptor-positive breast cancer.

Potentially, combining [18F]FDG-PET-CT and MRI-based response evaluation, or the targeted imaging of HER2 expression, could better choose patients who have a pathological complete response.[23] The targeted imaging of HER2 expression is currently primarily investigated in the metastatic setting for HER2 status evaluation, but in the future could potentially be developed for more specific heterogeneity identification and therefore rule out non-responders or to see in heterogeneous tumours whether the cells with HER2 expression fully respond or not.

In the near future, residual cancer burden instead of pathological complete response will probably be used to tailor treatment. Residual cancer burden is a better prognostic marker and patients with minimal residual disease could potentially be spared cytotoxic adjuvant treatment.[4,24] However, since the TRAIN-3 study aimed to select patients with the best prognosis (ie, residual cancer burden score of 0, which is a pathological complete response) to safely de-escalate chemotherapy, a pathological complete response was chosen as the endpoint. Additionally, the TRAIN-3 study is a multicentre study and residual cancer burden was not yet incorporated in the Dutch breast cancer guidelines at the time the study protocol was developed.

De-escalating chemotherapy strategies in HER2-positive breast cancer have the highest chance of success in patients with biologically HER2-driven disease. Selecting those patients upfront might therefore be a useful strategy. Tumours with a HER2-enriched intrinsic molecular subtype identified using gene signatures (eg, PAM-50 or BluePrint) are more likely to respond to HER2-directed therapy.[25,26] Molecular subtyping, however, is not universally available. Two German trials within the ADAPT umbrella showed the successful de-escalation of chemotherapy in both hormone receptor-negative and

hormone receptor-positive HER2-positive breast cancer.[27,28] Notably, they showed that selecting those with a HER2 immunohistochemistry score of 3+ only was an effective alternative method for selecting HER2-driven tumours.[27,28] This finding is consistent with the complete response rates we found for tumours with HER2 immunohistochemistry score of 3 or higher in the TRAIN-3 study. Another potential marker for an early response could be circulating tumour DNA response, which will be looked at in the translational work within the TRAIN-3 study.[29] Additionally, adding different gene signatures to the molecular subtypes combined with other clinical predictors might increase the predictive value of responses to different targeted therapies with or without chemotherapy, especially in patients with oestrogen receptor-positive and HER2-positive breast cancer.[30,31] Furthermore, biomarker research is currently ongoing in the TRAIN-3 study to help tailor treatment for HER2-positive breast cancer in future trials.

The TRAIN-3 study has some limitations. Firstly, patients were treated with paclitaxel on day 1 and 8 instead of the more commonly used regimen of docetaxel once every 3 weeks. However, both taxanes showed excellent results and once per week paclitaxel seems to be better tolerated.[32,33] Secondly, patients with oestrogen or progesterone receptor, or both, expression between 1% and 10% were considered hormone receptor negative. This cutoff differs from international guidelines (eg, American Society of Clinical Oncology and European Society of Medical Oncology), which recommend considering these tumours as hormone receptor positive. However, the TRAIN-3 protocol followed the Dutch National Guidelines and evidence of adjuvant endocrine treatment benefit is low in these patients. Therefore, there will not be much of an effect of these cutoffs on 3-year event-free survival. Thirdly, masked pathology and radiology review were not done and all analyses were based on local assessment, not accounting for inter-reader variation because in clinical practice double reading is uncommon. However, response monitoring was standardised using a specific radiology protocol, and the breast MRI had to meet protocol quality requirements. Lastly, patients who were referred for early surgery and did not reach a pathological complete response were potentially overtreated with adjuvant trastuzumab emtansine, and could potentially be overtreated with other targeted agents when they become available in the future. This stresses the need to aim for a high pathological complete response rate in patients who undergo early surgery. The toxicity of a few additional cycles of neoadjuvant chemotherapy might need to be balanced against the toxicity and burden of long-term adjuvant treatment approaches.

In conclusion, one in three patients with stage II–III hormone receptor-negative and HER2-positive breast cancer could potentially be treated with only three cycles of chemotherapy. This should be confirmed by the analysis of event-free survival, overall survival, and health-related quality of life. However, breast MRI is not sufficient in hormone receptor-positive tumours, for which accurate predictive biomarkers are highly needed.

Contributors

Conception and design: GSS and MSvR. Administrative support: AvdV, RK, and IAM. Provision of study materials or patients: all authors. Collection and assembly of data: AvdV, IAM, and RK. Data analysis and interpretation: AvdV, RK, and GSS. Manuscript writing: all authors. Final approval of manuscript: all authors. AvdV, RK, and GSS had full access to and verified the data in this article. All authors had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Declaration of interests

GSS reports receiving institutional research funding from Agendia, AstraZeneca, Merck, Novartis, Roche, and Seagen; and consulting fees from Biovica and Seagen. RMM reports receiving institutional research funding from Siemens Healthineers, Bayer Healthcare, Beckton & Dickinson, Screenpoint Medical, Koning, Lunit, PA Imaging, the Dutch Research Council, European Research Council, Dutch Cancer Society, European Union, and EFRO/OP-Oost; consulting fees from Siemens Healthineers, Bayer Healthcare, Beckton & Dickinson, Guerbet, Bracco, and Screenpoint Medical; is an advisory editorial board member of European Radiology; is an associate editor for breast imaging of the Radiology journal; is a member of the scientific committee of European Society of Radiology and Dutch Breast Cancer Research Group (chair working group for diagnostics); and is a member of the executive board of the European Society of Breast Imaging. All other authors declare no competing interests.

Data sharing

The anonymised data can be made available to other researchers after the primary and secondary endpoints have been published and in the presence of a data transfer agreement. Requests for data sharing can be made to the corresponding author (g.sonke@nki.nl).

Acknowledgments

Unrestricted financial support and pertuzumab and trastuzumab emtansine were provided by Roche Netherlands. We thank the patients and their families for participating in this study and all staff of the participating hospitals for their effort and commitment. We also thank the research department of the Netherlands Cancer Institute, in particular Kenneth Pengel, for central data management and monitoring. We thank the Dutch Breast Cancer Research Group as the study sponsor and Roche Netherlands for funding the trial and providing study drugs.

References

1 Gianni L, Pienkowski T, Im YH, et al. 5-year analysis of neoadjuvant pertuzumab and trastuzumab in patients with locally advanced, inflammatory, or early-stage HER2-positive breast cancer (NeoSphere): a multicentre, open-label, phase 2 randomised trial. *Lancet Oncol* 2016; **17:** 791–800.

2 Broglio KR, Quintana M, Foster M, et al. Association of pathologic complete response to neoadjuvant therapy in HER2-positive breast cancer with long-term outcomes: a meta-analysis. *JAMA Oncol* 2016; **2:** 751–60.

3 Cortazar P, Zhang L, Untch M, et al. Pathological complete response and long-term clinical benefit in breast cancer: the CTNeoBC pooled analysis. *Lancet* 2014; **384:** 164–72.

4 Yau C, Osdoit M, van der Noordaa M, et al. Residual cancer burden after neoadjuvant chemotherapy and long-term survival outcomes in breast cancer: a multicentre pooled analysis of 5161 patients. *Lancet Oncol* 2022; **23:** 149–60.

5 van Ramshorst MS, van der Voort A, van Werkhoven ED, et al. Neoadjuvant chemotherapy with or without anthracyclines in the presence of dual HER2 blockade for HER2-positive breast cancer (TRAIN-2): a multicentre, open-label, randomised, phase 3 trial. *Lancet Oncol* 2018; **19:** 1630–40.

6 Nitz UA, Gluz O, Christgen M, et al. De-escalation strategies in HER2-positive early breast cancer (EBC): final analysis of the WSG-ADAPT HER2+/HR- phase II trial: efficacy, safety, and predictive markers for 12 weeks of neoadjuvant dual blockade with trastuzumab and pertuzumab ± weekly paclitaxel. *Ann Oncol* 2017; **28:** 2768–72.

7 Bundred N, Porta N, Brunt AM, et al. Combined perioperative lapatinib and trastuzumab in early HER2-positive breast cancer identifies early responders: randomized UK EPHOS-B trial long-term results. *Clin Cancer Res* 2022; **28:** 1323–34.

8 Gianni L, Pienkowski T, Im YH, et al. Efficacy and safety of neoadjuvant pertuzumab and trastuzumab in women with locally advanced, inflammatory, or early HER2-positive breast cancer (NeoSphere): a randomised multicentre, open-label, phase 2 trial. *Lancet Oncol* 2012; **13:** 25–32.

9 Schneeweiss A, Chia S, Hickish T, et al. Pertuzumab plus trastuzumab in combination with standard neoadjuvant anthracycline-containing and anthracycline-free chemotherapy regimens in patients with HER2-positive early breast cancer: a randomized phase II cardiac safety study (TRYPHAENA). *Ann Oncol* 2013; **24:** 2278–84.

10 Baselga J, Bradbury I, Eidtmann H, et al. Lapatinib with trastuzumab for HER2-positive early breast cancer (NeoALTTO): a randomised, open-label, multicentre, phase 3 trial. *Lancet* 2012; **379:** 633–40.

11 Hurvitz SA, Martin M, Symmans WF, et al. Neoadjuvant trastuzumab, pertuzumab, and chemotherapy versus trastuzumab emtansine plus pertuzumab in patients with HER2-positive breast cancer (KRISTINE): a randomised, open-label, multicentre, phase 3 trial. *Lancet Oncol* 2018; **19:** 115–26.

12 van Ramshorst MS, Loo CE, Groen EJ, et al. MRI predicts pathologic complete response in HER2-positive breast cancer after neoadjuvant chemotherapy. *Breast Cancer Res Treat* 2017; **164:** 99–106.

13 De Los Santos JF, Cantor A, Amos KD, et al. Magnetic resonance imaging as a predictor of pathologic response in patients treated with neoadjuvant systemic treatment for operable breast cancer. Translational Breast Cancer Research Consortium trial 017. *Cancer* 2013; **119:** 1776–83.

14 Hayashi Y, Takei H, Nozu S, et al. Analysis of complete response by MRI following neoadjuvant chemotherapy predicts pathological tumor responses differently for molecular subtypes of breast cancer. *Oncol Lett* 2013; **5:** 83–89.

15 Kuijs VJ, Moossdorff M, Schipper RJ, et al. The role of MRI in axillary lymph node imaging in breast cancer patients: a systematic review. *Insights Imaging* 2015; **6:** 203–15.

16 Caudle AS, Kuerer HM, Krishnamurthy S, et al. Feasibility of fine-needle aspiration for assessing responses to chemotherapy in metastatic nodes marked with clips in breast cancer: a prospective registry study. *Cancer* 2019; **125:** 365–73.

17 Wolff AC, Hammond ME, Hicks DG, et al. Recommendations for human epidermal growth factor receptor 2 testing in breast cancer: American Society of Clinical Oncology/College of American Pathologists clinical practice guideline update. *Arch Pathol Lab Med* 2014; **138:** 241–56.

18 Schneeweiss A, Chia S, Hickish T, et al. Long-term efficacy analysis of the randomised, phase II TRYPHAENA cardiac safety study: evaluating pertuzumab and trastuzumab plus standard neoadjuvant anthracycline-containing and anthracycline-free chemotherapy regimens in patients with HER2-positive early breast cancer. *Eur J Cancer* 2018; **89:** 27–35.

19 de Azambuja E, Holmes AP, Piccart-Gebhart M, et al. Lapatinib with trastuzumab for HER2-positive early breast cancer (NeoALTTO): survival outcomes of a randomised, open-label, multicentre, phase 3 trial and their association with pathological complete response. *Lancet Oncol* 2014; **15:** 1137–46.

20 Cortes J, Pérez-García JM, Ruiz-Borrego M, et al. 3-year invasive disease-free survival (iDFS) of the strategy-based, randomized phase II PHERGain trial evaluating chemotherapy (CT) de-escalation in human epidermal growth factor receptor 2-positive (HER2[+]) early breast cancer (EBC). *Proc Am Soc Clin Oncol* 2023; **41** (suppl)**:** LBA506.

21 Newitt DC, Aliu SO, Witcomb N, et al. Real-time measurement of functional tumor volume by MRI to assess treatment response in breast cancer neoadjuvant clinical trials: validation of the Aegis SER software platform. *Transl Oncol* 2014; **7:** 94–100.

22 Hylton NM, Gatsonis CA, Rosen MA, et al. Neoadjuvant chemotherapy for breast cancer: functional tumor volume by MR imaging predicts recurrence-free survival-results from the ACRIN 6657/CALGB 150007 I-SPY 1 trial. *Radiology* 2016; **279:** 44–55.

23 Schmitz AMT, Teixeira SC, Pengel KE, et al. Monitoring tumor response to neoadjuvant chemotherapy using MRI and 18F-FDG PET/CT in breast cancer subtypes. *PLoS One* 2017; **12:** e0176782.

24 Symmans WF, Wei C, Gould R, et al. Long-term prognostic risk after neoadjuvant chemotherapy associated with residual cancer burden and breast cancer subtype. *J Clin Oncol* 2017; **35:** 1049–60.

25 Schettini F, Pascual T, Conte B, et al. HER2-enriched subtype and pathological complete response in HER2-positive breast cancer: a systematic review and meta-analysis. *Cancer Treat Rev* 2020; **84:** 101965.

26 Liefaard MC, van der Voort A, van Ramshorst MS, et al. BluePrint molecular subtypes predict response to neoadjuvant pertuzumab in HER2-positive breast cancer. *Breast Cancer Res* 2023; **25:** 71.

27 Nitz U, Gluz O, Graeser M, et al. De-escalated neoadjuvant pertuzumab plus trastuzumab therapy with or without weekly paclitaxel in HER2-positive, hormone receptor-negative, early breast cancer (WSG-ADAPT-HER2+/HR-): survival outcomes from a multicentre, open-label, randomised, phase 2 trial. *Lancet Oncol* 2022; **23:** 625–35.

28 Gluz O, Nitz U, Christgen M, et al. De-escalated chemotherapy versus endocrine therapy plus pertuzumab+ trastuzumab for HR+/HER2+ early breast cancer (BC): First efficacy results from the neoadjuvant WSG-TP-II study. *Proc Am Soc Clin Oncol* 2020; **38** (suppl)**:** 515.

29 Magbanua MJM, Swigart LB, Wu HT, et al. Circulating tumor DNA in neoadjuvant-treated breast cancer reflects response and survival. *Ann Oncol* 2021; **32:** 229–39.

30 Wolf DM, Yau C, Wulfkuhle J, et al. Redefining breast cancer subtypes to guide treatment prioritization and maximize response: predictive biomarkers across 10 cancer therapies. *Cancer Cell* 2022; **40:** 609–23.e6.

31 Waks AG, Ogayo ER, Paré L, et al. Assessment of the HER2DX assay in patients with ERBB2-positive breast cancer treated with neoadjuvant paclitaxel, trastuzumab, and pertuzumab. *JAMA Oncol* 2023; **9:** 835–40.

32 Carbognin L, Sperduti I, Nortilli R, et al. Balancing activity and tolerability of neoadjuvant paclitaxel- and docetaxel-based chemotherapy for HER2-positive early stage breast cancer: sensitivity analysis of randomized trials. *Cancer Treat Rev* 2015; **41:** 262–70.

33 van der Voort A, van Ramshorst MS, van Werkhoven ED, et al. Three-year follow-up of neoadjuvant chemotherapy with or without anthracyclines in the presence of dual ERBB2 blockade in patients with ERBB2-positive breast cancer: a secondary analysis of the TRAIN-2 randomized, phase 3 trial. *JAMA Oncol* 2021; **7:** 978–84.