Deletion of Chromosome 11q Predicts Response to Anthracycline-Based Chemotherapy in Early Breast Cancer

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Abstract

Despite the recent consensus on the eligibility of adjuvant systemic therapy in patients with lymph node–negative breast cancer (NNBC) based on clinicopathologic criteria, specific biological markers are needed to predict sensitivity to the different available therapeutic options. We examined the feasibility of developing a genomic predictor of chemotherapy response and recurrence risk in 185 patients with NNBC using assembled arrays containing 2,460 bacterial artificial chromosome clones for scanning the genome for DNA copy number changes. After surgery, 90 patients received anthracycline-based chemotherapy, whereas 95 did not. Tamoxifen was administered to patients with hormone receptor–positive tumors. The association of genomic and clinicopathologic data and outcome was computed using Cox proportional hazard models and multiple testing adjustment procedures. Analysis of NNBC genomes revealed a common genomic signature. Specific DNA copy number aberrations were associated with hormonal receptor status, but not with other clinicopathologic variables. In patients treated with chemotherapy, none of the genomic changes were significantly correlated with recurrence. In patients not receiving chemotherapy, deletion of eight bacterial artificial chromosome clones clustered to chromosome 11q was independently associated with relapse (disease-free survival at 10 years ± SE, 40% ± 14% versus 86% ± 6%; P < 0.0001). The 54 patients with deletion of 11q (29%) did not present more aggressive clinicopathologic features than those without 11q loss. The adverse influence of 11q deletion on clinical outcome was confirmed in an independent validation series of 88 patients with NNBC. Our data suggests that patients with NNBC with the 11q deletion might benefit from anthracycline-based chemotherapy despite other clinical, pathologic, or genetic features. However, these initial findings should be evaluated in randomized clinical trials. [Cancer Res 2007;67(2):818–26]

Introduction

The use of adjuvant systemic chemotherapy and/or endocrine therapy in the treatment of lymph node–negative breast cancer (NNBC) has increased greatly in the last decade (1–5). Despite recent consensus, the number of patients with NNBC that will obtain clinical benefit from the use of adjuvant chemotherapy is still a problematic debate. In young patients with large sized or high pathologic grade tumors, or with hormone receptor–negative tumors, systemic chemotherapy improves the odds of disease-free and overall survival (1, 2, 5, 6). Among the different chemotherapy regimens, those containing anthracyclines are, on average, more effective (5, 6). Therapy with trastuzumab seems to be effective in the subset of HER2-positive NNBC (7). However, despite obvious therapeutic advances, approximately one fourth of the patients with NNBC will have tumor recurrence that is potentially treatable but ultimately fatal. These data highlight the need for more sensitive and specific therapy-predictive indicators to refine the use of multiple treatment options.

Using gene expression profiling, several investigators have reported gene expression signatures of breast tumor cells that were more powerful predictors of disease outcome than standard clinical and histologic criteria (8–12). However, the apparent variability and lack of reproducibility observed among these previous transcriptional analyses and the requirements for high-quality RNA obtained from fresh tissues have limited their application to the clinical setting. A different type of microarray technology, termed comparative genomic hybridization (CGH) to microarrays (array CGH), allows a quantitative detection of DNA copy number changes in tumor genomes with high resolution (13, 14). This method enables the identification of precise areas in which genetic changes occur, including loss of genomic material (deletion) and genomic gain (amplification). These genomic alterations usually result in the damage of specific genes involved in cancer development and progression. In breast cancer, the definition of recurrent genomic aberrations has revealed loci encoding genes involved in the pathogenesis of the disease (15–20), some of which have been correlated with different pathologic subtypes (21). As the most prominent example, amplification of chromosome 17q12 targets the HER2 gene, which results in deregulation at the RNA and protein levels (22). Recently, array CGH has also proven its value for predicting clinical outcome in prostate carcinoma, lymphoma, and gastric carcinoma (23–25). However, to date, no similar studies have been attempted in breast cancer.

In the present study, we examined the feasibility of developing a predictor of recurrence risk and therapeutic response for 185 patients with NNBC, using array CGH for scanning the entire genome for DNA copy number changes.
Materials and Methods

Study Design and Selection of Patients

From September 1979 to June 2000, >3,100 new breast cancer patients were diagnosed at the University of Valencia. From these patients, 1,482 tumors were cryopreserved at −80°C. For this study, biopsies were randomly selected based on the following criteria for inclusion: (a) diagnosis of primary invasive breast carcinoma of any size; (b) treatment by modified radical mastectomy or breast-conserving surgery, including dissection of axillary lymph nodes, followed by radiotherapy if indicated; (c) the apical axillary lymph nodes were tumor-negative (pathologic examination, pN0); and (d) complete clinical data were available. More than 363 samples fulfilled the criteria. Previous to DNA extraction, these frozen tumor sections stained by H&E and reviewed for tumor infiltration: only samples that showed clearly >50% tumor cells were selected. A cohort of 185 patients fulfilled these final criteria. Clinicopathologic variables including tumor size, histologic grade and subtype, and estrogen receptor (ER) and progesterone receptor (PR) status were determined following standard methods previously reported (26). Human investigations were done after approval by an institutional review board on scientific and ethical affairs.

Array CGH

DNA extraction, hybridization, and imaging. Frozen tumors were included in ornithine carbamyl transferase compound. Prior to DNA extraction, H&E-stained tumor sections were examined to select samples with >50% tumor cells. Approximately 20 to 30 sections of 25 µm were used for DNA extraction. After removing the ornithine carbamyl transferase with PBS washes, DNA was extracted as described previously (26). Genome-wide analyses of DNA copy number changes were done using array CGH on a microchip with ~2,460 bacterial artificial chromosome (BAC) and P1 clones printed in triplicate (UCSF Hum Array 2.0) with a resolution of 1.4 Mb across the genome (27). Methods and analytic procedures have been described in detail elsewhere (refs. 27–29; Fig. 1). For visualization of

Figure 1. Representation and description of genome-wide array CGH technique. Genome-wide analysis of DNA copy number changes were done using array CGH on a microchip with ~2,460 BAC and P1 clones printed in triplicate. Briefly, 0.5 µg of test (tumor) and reference genomic DNAs were labeled by random priming using Cy3 and Cy5, respectively. After 48 h of hybridization, slides were washed and mounted with 4',6-diamidino-2-phenylindole. The images of the arrays were captured using a CCD camera, and the "UCSF SPOT" 2.0 software (available at http://www.jainlab.org/downloads.html) was used to analyze the images and measure tumoral versus control fluorescence intensity ratios that were converted to the log 2 scale. A second program, the "UCSF SPROC," was used to associate clones with each spot and to create a mapping information file that allows the data to be plotted relative to the position of the BACs on the draft human genome sequence (http://genome.cse.ucsc.edu; May 2004 freeze). A formal data-filtering procedure was then done, and a SPROC output file consisting of the averaged ratios of the triplicate spots for each clone, SDs of the replicates, and plotting positions for each clone on the array, was obtained.
genomic data, the TreeView program 1.60 (Stanford, CA) was used. To confirm array CGH data, CGH to chromosomes was done in 44 of the biopsies included in this study.

Interphase fluorescence in situ hybridization analysis. To confirm specific gains and losses of BAC clones observed in the array CGH analyses, fluorescence in situ hybridization (FISH) studies using individual BAC clones as probes on isolated nuclei from frozen tumor sections were done using a previously reported technique (30). The gene loci examined corresponded to five overrepresented and four deleted BAC clones, using appropriate centromeric probes as controls. A total of 100 cells were examined on each of the 22 tumors examined. These clones were obtained from RZPD German Resource Center (Berlin, Germany) or purchased from Vysis (Downers Grove, IL).

Statistical Data Analysis
To process the genomic data obtained with array CGH, we used a previously described analytic model (19, 31). Arrays were normalized by subtracting the median of each array from the average log 2 ratio for every clone. The array CGH data were then analyzed using a hidden Markov model using the Bioconductor package aCGH. Log 2 ratios, as ordered in the genome, were segmented into regions of constant copy number. In addition, the hidden Markov model was employed to impute missing values by using the estimated copy number ratio for the segment containing the clone(s) with missing values. Clones with missing values located between segmented regions were assigned the mean value of the segment that was closer in genomic distance. Thus, each clone was assigned a segment value referred to as its “smoothed” value. The median absolute deviation (MAD) of the difference between the observed and smoothed values was used to estimate the tumor-specific experimental variation. Clones for each array were assigned into three groups: gained (smoothed log 2 ratio more than thrice the MAD), lost (smoothed log 2 ratio less than thrice the MAD), and not changed (the log 2 ratio could not be assigned to lost or gained groups). Smoothed data was used to study the association with the following phenotypes: age, tumor size, histologic grade and subtype, stage, ER and PR status, and recurrence/survival. Finally, we used Fisher’s two-sided exact test to compare genomic

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NOTE: Differences between patients treated with chemotherapy (CHEMO group) and patients who did not receive chemotherapy after surgery (non-CHEMO group).
events or clinical variables among both groups of treatment. To evaluate differences in disease-free survival (DFS), Kaplan-Maier survival curves for the sets of patients were examined.

**Gene ontology classification.** Gene ontology (GO) was used to construct a unified and structured vocabulary for the description of genes and their products in any organism. A GO term is descriptive of the molecular function, biological process, or cellular component. We then did a statistical analysis to check if there were any GO categories that were enriched in the genes located in the region of deletion in chromosome 11q23-q24 with respect to the whole genome.

**Clinical Series for Validation of Array CGH Results**

To validate the possible association of chromosome 11q deletion with increased relapse rate, this was tested in a validation group of 88 tumor biopsy samples from an independent cohort of patients with NNBC: 18 Spanish patients, treated in different institutions within the Valencia area, whose genomes were analyzed with array CGH as described above. In addition, data from 70 patients were obtained from a recently published series of American patients with breast cancer who were analyzed using similar whole-genome array CGH techniques. All patients fulfilled the inclusion criteria reported for this study. Kaplan-Maier survival curves for the two sets of patients were evaluated. Clinicopathologic characteristics of the validation series are shown in Supplemental Table S1.

**Results**

**Characteristics of the patients.** The clinicopathologic characteristics of the 185 patients and tumors are summarized in Table 1. Based on the clinicopathologic features, 90 women received anthracycline-based chemotherapy (CHEMO group) whereas 95 patients did not (non-CHEMO group). In both groups, women with ER/PR-positive tumors were treated with tamoxifen: 42 in the CHEMO group (47%) and 56 in the non-CHEMO group (59%; Supplemental Table S2). With a median duration of follow-up time of 82 months (range, 9-218 months), 45 of the 185 patients (24%) had relapsed. Median duration of follow-up time for patients who were free of disease was 96 months in both CHEMO and non-CHEMO groups. Death from the disease was assessed in 16 of 185 patients (9%). We initially determined differences in clinicopathologic features and outcome between patients in the CHEMO group versus those in the non-CHEMO group. Women in the CHEMO group were younger (mean, 51 versus 67 years; \(P = 0.003\)) and had a more frequent premenopausal status (43% versus 17%; \(P = 0.001\)). We did not observe statistically significant differences in any other histopathologic feature (Table 1; Supplemental Fig. S1A). In addition, no statistically significant differences in DFS and overall survival were found between CHEMO and non-CHEMO groups (Supplemental Fig. S1B and C).

**Genomic profiling of NNBC.** Array CGH analysis was done in the 185 tumor biopsies, and allowed the identification of specific regions of gain and loss throughout the genome with high resolution in all of the biopsies. All tumors showed genomic changes (Fig. 2). A total of 112 clones that were mapped to 40 different chromosome loci in 9 different chromosome arms were found. These corresponded to 23 genomic gains and 17 genomic losses involving regions known to be commonly involved in breast cancer as well as uncharacterized genomic aberrations. The most common gains corresponded to chromosomes 1q21 and 20q12 (91 of 185, 49%), 8q24.2 (40%), 17q21 (39%), 1q32, 8q23.1 and 20q13.1 (35%), 1q23 (34%), and 8q24.1 at the MYC gene locus (32%). In addition, high-level amplification (defined as log 2 ratio >1 observed in at least 10 different samples) was identified in five different regions of chromosomes 11q13-q14 at the CCND1 gene (17 of 185 tumors, 9%), HER2 (13 tumors, 7%), 1q31 and 8p12 at the FGFR1 gene (11 tumors, 6%), and 8q21-q24.1 including the MYC gene (10 tumors, 5%). The most frequently deleted regions were observed at chromosomes 13q14-q22 (66 of 185 tumors, 36%), 17p12-p13 including the P53 gene locus (34%), 16q21-q22 including the CDH1 gene (30%), and 11q23-q24 (29%), 16q24 and 16p12-p13.1 (26%), 11q12 (25%), 8p21.3-p22 (25%), and 22q11.2. A total of 18 homozygous deletions (defined as log 2 ratio <−1.4) were identified, being the loss of 13q21.3-q22 at KLFI2 gene observed in two different tumors. The full array CGH data has been deposited in the GEO database (Platform GPL3632).

To initially validate the array CGH results, a subset of 44 samples was also analyzed with CGH to chromosomes, and the two techniques showed concordant values (Supplemental Fig. S2). To further validate our data, the analysis of nine individual BAC clones in 22 frozen tumor sections using FISH also showed a high concordance with array CGH results (Supplemental Table S3). The array CGH data defined a common genomic signature of NNBC (Fig. 2; detailed information in Supplemental Table S4).

**Correlation of genomic alterations and clinicopathologic features.** We tested the association of clinical and pathologic variables with each of the BAC clones in the 185 patients. After adjustment for multiple testing, these analyses showed that the only variables correlated with genomic changes were ER and PR status (Fig. 3). Tumors that showed expression of ER (ER+) presented with frequent gain of chromosomes 1q21-q43 (35% versus 14%; \(P < 0.05\)) and 16p12 (17% versus 1%; \(P < 0.01\)) and...
losses of chromosome 16q21-q24 (25% versus 7%; \( P < 0.01 \)). Tumors negative for PR (PR−) also presented with frequent deletion of chromosomes 4p13-p16 (19% versus 5%; \( P < 0.001 \)) and 5q11.2-q31 (16% versus 3%; \( P < 0.001 \); Fig. 3B; Supplemental Table S5). In addition, tumors negative for ER displayed a similar genomic profile than PR− tumors, including deletions of 4p and 5q; however, after statistical adjustments, these changes were not statistically significant (Fig. 3C). We also determined the correlation of the genomic status of the 17q12 locus at the HER2 gene (determined by array CGH analysis) with other clinical variables. Amplification or gain of HER2 gene was observed in 29 tumors (16%) and was correlated with negativity for PR expression (\( P = 0.007 \)), but not with other clinicopathologic features.

**Association of genomic abnormalities with clinical outcome: 11q loss may predict response to chemotherapy.** We compared the genomic profiles of tumors in the CHEMO and non-CHEMO groups. None of the abnormal BAC clones showed a significantly different distribution between the two cohorts, indicating that both groups were comparable at the genomic level (Fig. 2). To develop a genomic predictor of clinical outcome, we examined the association of the genomic aberrations with disease recurrence in the two differently treated cohorts. In the CHEMO group, after adjustment for multiple testing, none of the abnormal BAC clones were associated with tumor relapse (Fig. 4A). In the non-CHEMO group, however, there were statistically significant differences (\( P < 0.05 \)) in eight BAC clones that showed more common deletions in tumor recurrences with respect to nonrecurrences (Fig. 4B). Notably, these eight clones clustered to the long arm of chromosome 11 from 11q23.1 to 11q24.1, spanning ~9 Mb in size (Fig. 4C). Therefore, deletion of chromosome 11q was associated with decreased DFS in patients with NNBC in the non-CHEMO group (DFS ± SE at 10 years, 40% ± 14% versus 86% ± 6%; \( P < 0.0001 \)) but not in the CHEMO group (DFS at 10 years ± SE, 92% ± 21% versus 65% ± 9%; \( P = 0.13 \)). Analyses of the association of the genomic changes with overall survival in the two treatment cohorts did not reveal any significant correlation, probably due to the low number of patients who have thus far died of the disease. Finally, we did a statistical analysis to check if there were any GO categories that were enriched in the genes located in the region of deletion in chromosome 11q23.1-q24.1. Among them, DNA repair genes and meiotic-related genes were significantly enriched (hypergeometric test, \( P < 0.00092 \)). Four genes belonged to this category (CHK1, H2A, ATM, and ZW10). These data may suggest that deletion of chromosome 11q targets the gene(s) involved in DNA repair; thus, possibly contributing to the increased sensitivity to anthracyclines identified in our study.

**Characteristics of patients with 11q deletions.** To determine whether the negative effect of 11q deletion on DFS was dependent

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**Figure 3.** Correlation of ER/PR expression status with genomic data. A, the genomic gain of BAC clones mapped to 1q21-q43 and to 16p12 chromosome regions and the genomic loss of clones mapped to 16q21-q24 were associated with positive ER (arrowheads). B, the PR status was not significantly associated with abnormalities of the 1q, 16p, or 16q arms, but a borderline significance in chromosome 16 was observed. Instead, the genomic loss of clones mapped to 4p13-16 and 5q11.2-q31 was observed associated statistically with negative PR (arrowheads). C, frequency plot of all BAC clones in chromosome 16 comparing ER-positive (C2) versus ER negative (C1) tumors. D, log 2 ratios from clones in chromosome 16 in one ER-positive breast tumor.
Figure 4. Association of genomic results with clinical outcome in patients with NNBC. A, in the CHEMO group, after adjustment for multiple testing, none of the 2,460 BAC clones were associated with tumor relapse. B, in the non-CHEMO group, however, there were statistically significant differences ($P < 0.05$) in eight BAC clones, clustered to the long arm of chromosome 11, which showed more common deletion in the group of tumor recurrences. Kaplan-Meier curves showed differences in DFS for 11q-deleted tumors versus those without deletion in the non-CHEMO group (DFS = 35 at 10 yrs, 40% ± 14% versus 66% ± 6%: $P < 0.0001$) but not in the non-CHEMO group. Considering the patients harboring deletion of chromosome 11q, 5 of 31 patients in the CHEMO group (16%) had recurrence of the disease, whereas the relapse rate was much higher in the non-CHEMO group: 14 of 23 with 11q deletion (62%) had a relapse ($P < 0.0001$). Among the 59 patients in the CHEMO group who did not show deletion of 11q, 19 patients (30%) presented recurrence of the disease, whereas only 8 of 72 patients without the 11q deletion (11%) in the non-CHEMO group relapsed. C, region of deletion in chromosome 11q. The eight clones clustered to chromosome 11 from bands 11q23.1 to 11q24.1 (yellow). When the adjusted value for statistical significance was of <0.1 instead of <0.05, the number of BAC clones correlated with relapse increased to 24, all of them mapped to 11q21-q25.
on other clinical and biological features, we compared the clinical and biological characteristics of the 54 patients with an 11q deletion versus those 131 patients without a deletion of 11q. In the whole group of 185 patients, there were no statistically significant differences for age, clinical stage, hormonal status, tumor size and grade, and expression of ER/PR for 11q-deleted versus nondeleted tumors (Supplemental Table S6). These data indicate that the influence of 11q deletions in the relapse rate of the patients in the non-CHEMO group is independent of other known clinical and pathologic features. We also analyzed possible differences in patients with and without 11q deletion for genetic alterations reported in correlation with aggressive breast cancer (HER2, CCND1, MYC, and FGFR1 amplifications and P53/P16 deletions). We did not observe changes in the distribution of these genomic alterations, with the exception of CCND1 amplification that was more common in tumors harboring deletion of chromosome 11q: among 17 cases with CCND1 amplification, 12 (70%) presented deletion of 11q, whereas only 42 cases (25%) showed 11q deletions among the 168 nonamplified CCND1 cases ($P < 0.001$). This association could probably be explained by the proximity of the
CCND1 gene (which maps to the 11q13 band) to the 11q23.1-q24.1 deletion (Supplemental Fig. S4B). Notably, genomic amplification of CCND1 was not associated with decreased DFS in both the non-CHEMO and CHEMO groups. In summary, tumors with 11q deletion do not show a more aggressive phenotype or genotype that can distinguish them from those without this chromosome deletion.

**Validation of 11q deletion as a therapy-predictive indicator.**

To validate the association of chromosome 11q deletion with worse outcome in patients not receiving anthracycline-based chemotherapy, we analyzed a second series (validation group) of 88 NNBC biopsies (Supplemental Table S1). After surgery, 27 of the patients received chemotherapy, whereas the 61 remaining did not. Sixty-two patients with ER/PR-positive expression received hormonal therapy based on tamoxifen. In the group treated with chemotherapy, 6 of 15 patients (40%) without an 11q deletion relapsed, whereas 3 of 12 patients (25%) with an 11q deletion had a recurrence (P = 0.23). However, in the group not receiving chemotherapy, tumor recurrence was observed in 4 of 33 patients (12%) without an 11q deletion and in 10 of 28 patients (35%) with an 11q deletion (P = 0.02; Fig. 5). Kaplan-Meier curves also showed that deletion of chromosome 11q was associated with inferior DFS in patients not treated with chemotherapy (DFS ± SE at 10 years, 65% ± 13% versus 88% ± 8%; P < 0.1). Notably, in the CHEMO group, patients with an 11q deletion had a tendency to show a superior DFS compared with those without 11q loss (73% ± 18% versus 50% ± 18%; P < 0.7). These differences, however, did not reach statistically significant values, probably because of the limited number of patients and the relatively short median follow-up time (Fig. SD). In summary, the results observed in this validation set were coincident with those obtained in the training set of 185 patients, and confirm that deletion of 11q was associated with relapse in patients with NNBC who did not receive anthracycline-based chemotherapy.

**Discussion**

An alteration in the DNA copy number is one of the many ways in which gene expression and function may be modified. In cancer, these gains and losses of genomic material frequently disrupt genes implicated in the development and progression of the disease. Array CGH enables the detection of these genomic aberrations with high-resolution (13, 15, 19, 20, 25, 29). In this exploratory study, we used genome-wide array CGH to study a series of patients with breast cancer. Our initial data suggest that patients with NNBC with the 11q deletion may benefit from anthracycline-based chemotherapy despite other clinical, pathologic, or genetic features. Thus, loss of chromosome 11q may make tumor cells more responsive to anthracycline-based chemotherapy through a currently unknown mechanism. Theoretically, adjuvant systemic chemotherapy is administered to the patients to kill any remaining malignant cells after surgical removal of the tumor. Cellular DNA is the primary target for anthracyclines, by binding and inserting between DNA bases, leading to chromatin unfolding and aggregation. These chromatin structural changes primarily interfere with DNA replication and transcription, thus leading to the apoptosis undergone by the cells treated with anthracyclines (32). The selectivity of these drugs for targeting cancer cells, but not nontumoral cells, may reside in the lower ability of cancer cells to repair the damage induced by the drugs (32). We may speculate that deletion of chromosome 11q in breast tumor cells leads to functional impairment of the gene(s) involved in DNA repair, thus, contributing to the increased sensitivity to anthracyclines. Indeed, we have observed that genes involved in DNA repair were enriched in the deleted 11q region with respect to the rest of the genome. Two of the most prominent candidate targets for 11q deletion in breast cancer are (a) the ataxia-telangiectasia mutated gene ATM which codifies for a protein involved in DNA repair and cell cycle control (33, 34), and (b) the cell cycle checkpoint kinase CHK1 which acts downstream of ATM in response to DNA damage (35). Other candidate targets include the gene encoding for the H2AFX histone, which is critical for facilitating the assembly of specific DNA-repair complexes on damaged DNA (36); and the gene encoding for the mitotic checkpoint protein Zw10 (37). In this theoretical scenario, 11q-deleted tumor cells that become deficient for these DNA repair genes cannot detect and/or repair DNA damage induced by anthracyclines, whereas tumor cells with intact chromosome 11q and functional DNA repair proteins are able to repair DNA efficiently, evade apoptosis, and ultimately metastasize. We may also suggest that deletion of 11q targets the noncoding micro-RNA gene miR125b-1, which is specifically down-regulated in breast cancer and may potentially regulate oncogenes such as ETS1, which plays a role in cell growth and has been shown to be overexpressed in breast cancer (38). Ongoing genetic and functional studies will try to define the damaged genes of chromosome 11q that are responsible for the anthracycline-resistant tumor phenotype.

Finally, our study could be valuable in the clinical management of patients with NNBC, by adding the 11q deletion status to the currently accepted prognostic and therapy-predictive markers (4). According to our data, tumors should be screened for the presence or absence of 11q deletion at diagnosis using rapid quantitative PCR, FISH, and/or mini-array CGH devices using a reduced set of BAC clones. These diagnostic tests should allow clinicians to prospectively identify patients who are candidates to receive anthracycline-based chemotherapy, such as standard AC/FAC, which are widely used as front-line therapies in NNBC, irrespective of other clinicopathologic features. We may propose that in patients with NNBC presenting factors that imply a good prognosis, such as age >35, clinical stage I, low-grade tumors sized ≤1 cm, and ER/PR positivity, systemic chemotherapy could be avoided only if 11q deletion is not identified. Although our study requires further validation and refinement in prospective clinical trials, future application of 11q deletion measurement as a novel therapeutic indicator in NNBC is clearly challenged.

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