Genetic Alteration Mapping on Chromosome 7 in Primary Breast Cancer

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ABSTRACT
Alterations of chromosome 7 are among the most frequent cytogenetic abnormalities found in human breast carcinoma. We examined genetic changes on chromosome 7 in 113 primary human breast tumors, using both microsatellite and restriction fragment length polymorphism-variable number of tandem repeats polymorphism markers mapping to the long arm (15 markers) and the short arm (8 markers). Allelic imbalance at 1 or more loci was observed in 50 (44%) of 113 tumors on the long arm of chromosome 7 and in 41 (36%) tumors on the short arm. Genetic changes of one arm were significantly associated with alterations of the other arm. The 50 7q-altered tumor DNAs exclusively showed a loss of heterozygosity (LOH), 23 (46%) at all informative loci tested on 7q and 27 (54%) at some loci (interstitial and/or telomeric deletions on 7q). The pattern of LOH of these 27 tumors enabled us to identify 3 distinct consensus regions of deletions on 7q, only 1 of which (7q31 region) has already been described in breast cancer. Among the 41 7p-altered tumor DNAs, 32 had a gain and/or loss of the entire short arm of chromosome 7. Fourteen tumor DNAs showed an allelic gain, and 18 tumor DNAs showed a LOH at each locus on the short arm. The other 9 7p-altered tumors showing partial random alterations of chromosome 7p revealed no common altered regions. This is the first report of an association between alterations of DNA sequences on chromosome 7p and breast cancer. The results suggest that tumor suppressor genes are present on the long arm of chromosome 7 and are associated with breast tumorigenesis. Moreover, the frequent loss or gain of a whole copy of chromosome 7p suggests the involvement of a gene dosage effect of this chromosomal arm in the pathogenesis of breast cancer.

INTRODUCTION
Carcinogenesis is now considered to be a highly complicated process in which an accumulation of genetic mutations is required to transform a normal cell into a malignant cell. Any one of several mechanisms, such as point mutations, translocations, gene amplifications, and loss or gain of all or part of a chromosome, can activate oncogenes or inactivate tumor suppressor genes, thereby allowing a cell to escape from normal growth control.

One of the first indications that a cancer gene was present on a specific chromosome consisted of structural abnormalities and the gain or loss of a specific chromosome detected by karyotypic analysis of tumor cells. Using molecular analysis of the allelic pattern of tumor DNAs with polymorphic probes mapping to the suspected chromosome, it is possible to locate a common region for allelic alterations, which could lead to the identification of cancer genes (1).

Cytogenetic analyses and comparative genomic hybridization have indicated that alterations to chromosome 7 are one of the most frequent abnormalities associated with breast cancer. There are several karyotypes with numerical alterations (polysomy or monosomy 7 and 7p) and structural abnormalities on both arms of chromosome 7 (2-5).

Microcell fusion experiments have indicated the presence of one or more genes on chromosome 7 with the ability to suppress the tumorigenicity of transformed cells (6). Furthermore, Ogata et al. (7) demonstrated that the insertion of an intact human chromosome 7 into a immortalized human fibroblast cell lines with LOH3 in segment 7q31–q32 restored senescence properties.

All these observations suggest that chromosome 7 harbors unidentified cancer genes whose activation (oncogenes) or inactivation (tumor suppressor genes) may be involved in human breast cancer tumorigenesis.

Only the long arm of chromosome 7, especially the 7q31 region, has been studied by means of molecular analysis in breast tumors (8–11). We have previously shown that a high frequency (41%) of LOH occurs at the MET locus mapping to 7q31 and that this alteration is associated with shorter survival (8). Later, using polymorphic marker mapping to the 7q31 chromosomal region, we identified a common region of LOH distributed around the D7S522 probe (10). However, this study involved a small tumor series (only 11 tumors were informative with the D7S522 marker) and had to be confirmed in a larger series. Deletion of 7q31 was also frequently observed in other tumors, including human primary squamous cell, colon, ovarian, and prostate carcinomas (12–16), suggesting the presence of...
either a cluster of tumor suppressor-type genes, each involved in one tumor type, or a single suppressor gene with a single function involved in the etiology of all these tumor types. Deng et al. (9) have detected a strong association between one tumor type, or a single suppressor gene with a single TP53 genetic markers located at chromosomal segment 7q31.2 (LOH at 7q31.2) by analyzing a number of closely spaced markers and also RFLP/VNTR markers to distinguish unequivocally 7p, and 15 mapping to 7q) to produce an alteration map of the chromosome 7, using a panel of 23 polymorphic markers (8 mapping in breast cancer (17) and prostate carcinoma (15).

In the present study, we examined 113 human breast tumors for evidence of gain or loss of DNA sequences on chromosome 7, using a panel of 23 polymorphic markers (8 mapping to 7p, and 15 mapping to 7q) to produce an alteration map of the two arms and to locate genes of interest. We used microsatellite markers and also RFLP/VNTR markers to distinguish unequivocally between allelic gains and losses (usually difficult using only microsatellite markers). We sought to confirm and further define the first identified molecular alteration on chromosome 7 (LOH at 7q31.2) by analyzing a number of closely spaced genetic markers located at chromosomal segment 7q31.1–7q31.2 in a larger series of tumors than in our first study (113 versus 31 cases). The aim was to determine whether other regions bearing DNA rearrangement or allelic imbalance were linked or independent.

Table 1 Polymorphic probes used and genetic change results

<table>
<thead>
<tr>
<th>Chromosomal location</th>
<th>Locus</th>
<th>Probe</th>
<th>Polymorphism type</th>
<th>Enzyme</th>
<th>No. of screened patients</th>
<th>No. of informative patients (%)</th>
<th>Tumor DNAs with LOH</th>
<th>Allelic gain or allelic imbalance</th>
<th>Altered tumor DNAs (%)</th>
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<td>7p22 D7S531</td>
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<td>CA repeat</td>
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<td>RFLP</td>
<td>HindIII</td>
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<td></td>
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<td>HindIII</td>
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<td>0</td>
<td>20 (37.0)</td>
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<td>VNTR</td>
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<td>69 (81)</td>
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<td>0</td>
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PATIENTS AND METHODS

Patients and Samples. Samples were obtained from 113 primary breast tumors surgically removed from patients at the Center René Huguenin (St-Cloud, France); none of the patients had undergone previous radiation therapy or chemotherapy. Immediately after surgery, the tumor samples were stored in liquid nitrogen until extraction of high molecular weight DNA. A blood sample was also taken from each patient.

Seven of these 113 tumors were included in a previous study of 31 cases (10). DNA was extracted from the tumor tissue and blood leukocytes of each patient using standard methods (18).

DNA Probes. We used 10 DNA probes for RFLP/VNTR polymorphism and 13 microsatellite markers on chromosome 7 (15 on 7q and 8 on 7p) to screen the 113 samples. Table 1 gives details of the loci investigated and their corresponding chromosomal location (19–21).

RFLP/VNTR Marker Analysis. Ten μg of DNA from each sample were digested with the appropriate restriction endonuclease (Table 1). The resulting fragments were separated by electrophoresis in agarose gel (leukocyte and tumor DNA samples) and on 7% and 8% on 7p) to screen the 113 samples. Table 1 gives details of the loci investigated and their corresponding chromosomal location (19–21).

Microsatellite Marker Analysis. PCR was run in a total volume of 50 μl, with 50 ng of genomic DNA, 20 mM each primer, 1.5 mM MgCl₂, 0.1 mM each deoxynucleotide triphosphate, and 1 unit of Taq DNA polymerase. Microsatellite markers were assayed by PCR amplification of genomic DNA. The annealing temperature, number of amplification cycles, and extension time were adapted to each primer set. One μl of product was mixed with 3 μl of denaturing loading buffer and heat-denatured, then 1.5-μl aliquots of each sample were loaded on 6% acrylamide gels containing 7.5 mM urea. DNA was then transferred to nylon membrane filters. The CA repeat probe was labeled with [32P]dCTP by using terminal deoxynucleotidyl

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transf erase. The membrane filters were hybridized overnight at 42°C with the labeled probe, washed, and autoradiographed at −80°C for an appropriate period.

**Determination of Allelic Dosage.** Normal DNA samples that were polymorphic at a given locus were considered to be informative, whereas homozygotes were considered to be uninformative. Only cases of constitutional heterozygosity were used in the evaluation of allelic dosage. The signal intensity of the polymorphic alleles was determined by visual examination (three observers) and confirmed by means of densitometry. The results of all of the scanned samples were in direct agreement with the initial visual scoring. The nature of the imbalance (allelic gain or LOH) was only determined with the RFLP/VNTR markers. For each tumor, we used a specific control probe that demonstrated retention of heterozygosity for the tumor. The following control probes were used, mapping to chromosomes other than chromosome 7: D1S57 (pYNZ2); D1S61 (pMIAJ1); D17S5 (pYNZ22); and D17S4 (THH59); all had been previously studied on this series of tumors.

The loading of the paired normal and tumor DNA from each patient was taken into account in judging allele gain or loss in tumor DNA (Fig. 1). The loading of DNA in each lane was evaluated by rehybridization of the same membranes with control probes.

Allelic gain or LOH was considered to occur when the intensity (peak area corresponding to the hybridizing signal) of one allele in tumor DNA was more or less than 50% of that in corresponding leukocyte DNA, respectively.

LOH was partial in most cases, with the band being fainter than the conserved allele but still visible. Such partial losses are due to either normal tissue contamination or tumor heterogeneity.

Allelic gain was quantified by serial dilution of the tumor DNA samples to obtain a Southern hybridization signal similar to that obtained with leukocyte DNA.

**Statistical Analysis.** Differences were analyzed for statistical significance by using the $\chi^2$ test with Yates’ correction to adjust for the continuity of the $\chi^2$ distribution when appropriate. Differences between the two populations were judged significant at a confidence level greater than 95% ($P < 0.05$).

**RESULTS**

We analyzed normal DNA (peripheral blood leukocytes) and autologous tumor DNA from 113 breast cancer patients, using 15 polymorphic DNA markers spanning the entire long arm of chromosome 7 and 8 polymorphic markers mapping to the short arm, to obtain an overall picture of chromosome 7.

All patients were informative for two or more loci on 7p and five or more loci on 7q. The frequencies of allelic gain and/or LOH for each of these loci are summarized in Table 1. The highest frequencies of allelic imbalance (allelic gain and/or LOH) were observed at the D7S13 and D7S650 loci (37.0%) on 7q and at the D7S135 locus (43.7%) on 7p. Allelic imbalance occurred in at least 1 locus on chromosome 7 in 61 of 113 tumors (54%). Fig. 1 shows examples of allelic gain and LOH. Allelic imbalance affecting 7q was found in 44% of the samples (50 of 113), whereas allelic imbalance for 7p was detected in 36% (41 of 113).

**LOH on Chromosome 7q.** The 50 7q-altered tumors showed only LOH. We observed no allelic gain at any informative loci tested on 7q. Of these 50 deleted tumors, 23 (46%) showed LOH at all informative loci tested on 7q, indicating deletion of the entire long arm of chromosome 7, whereas the other 27 (54%) showed partial (interstitial and/or telomeric) deletions.

The 27 tumors showing partial deletions on chromosome 7q proved to be of greater interest than those showing total deletion. Figure 2 shows examples of the most common patterns of genetic changes at 7q. Tumors 1156, 1288, and 1362 showed interstitial deletions on chromosome 7q. Tumor 1156 showed LOH for D7S480 and D7S650 and a retention for D7S440 and D7S22, whereas D7S13 was uninformative. Tumor 1288 had LOH for D7S13 and retention for D7S440, D7S480, D7S650,
and D7S22. Tumor 1362 showed LOH for D7S650 and retention for D7S13, D7S480, and D7S22, whereas D7S440 was uninformative. Tumor 345 showed a telomeric deletion, LOH for D7S22, and retention for D7S440, D7S13, D7S480, and D7S650. Fig. 3 (Groups A and B) summarizes 7q LOH data in the form of deletion maps. In spite of a high background of alterations throughout chromosome 7q, three independent SCDRs (SCDR1, SCDR2, and SCDR3) were clearly identified by studying the peak frequencies of LOH for the markers tested (Table 1), and the specific deletion map for certain tumors in Fig. 3 (e.g., T1353 and T1362 for SCDR2). The first proximal region was at 7q11.23-q22, defined by D7S440 and D7S13. The second region of 7q LOH was central and pointed to the presence of a target gene located in the thin 7q31.2 region (smaller than 1 cM) lying between D7S480 and D7S650. Lastly, these data suggested that a third potential target gene could be located in the distal region between D7S495 and D7S396. The proximal and distal regions identified are still quite large, being an estimated 40 and 41 cM apart (19, 20). Note that a large proportion of these tumor deletions encompassed more than one common deleted region on 7q. Moreover, some tumors showed noncontiguous LOHs affecting different SCDRs, the most extreme example being tumor T220, with losses at three distinct regions on 7q.

Two tumors (T1303 and T1373) showed a DNA fragment breakpoint within the anonymous DNA markers D7S13 and D7S23, respectively. Structural alterations were evident from the appearance of an extra band of a different size in tumor DNA (a band not present in leukocyte DNA). These DNA rearrangements were confirmed by other restriction digests.

Allelic Imbalance on Chromosome 7p. Of 41 7p-altered tumors, a majority (32, 78%) showed allelic imbalance at all informative loci tested on 7p, whereas only 9 (22%) showed partial (interstitial and/or telomeric) alterations on 7p. The 32 tumors in which the whole short arm was altered comprised 14 tumors with allelic gain and 18 tumors with LOH at all informative loci tested on 7p, findings highly suggestive of polysomy and/or deletion of the entire short arm of chromosome 7. Denitometric quantification of the polysomies showed that one allele was multiplied 2–7-fold in tumor DNA.

The partial alterations (deletions or polysomy) on chromosome 7p observed in the other nine tumors could have been not
specific, because these tumors did not display a consensus for a small common altered region. Fig. 3 (Groups B and C) summarizes imbalance data in the form of an alteration map. One tumor (T1355) showed a DNA structural alteration within the intranormal DNA marker D7S21; this abnormal case had a telomeric alteration also affecting the telomeric D7S531 locus. A second tumor (T311) showed insertion of four nucleotides from the dinucleotide repeat D7S507. This alteration did not seem to be the result of traditional microsatellite instability, because no instability was observed in any of the other 12 microsatellite markers tested on this tumor or in 112 other tumors.

The other seven tumors showed partial alterations, including partial deletions (T54 and T383) and partial polysomy (T212, T274, and T1298), whereas it was not possible to determine the nature of the partial imbalances in tumors T1316 and T1305, because they bore alterations in areas studied with microsatellite markers only.

**Simultaneous Alterations of the Long and Short Arms of Chromosome 7.** We observed a strong association between alterations at 7q and 7p ($\chi^2 = 21.82; P < 0.01$; Table 2). Thirty cases showed alterations on both arms.

**Table 2 Relationship between alterations at 7p and 7q in breast tumor tissues**

<table>
<thead>
<tr>
<th>7q DNA status</th>
<th>Altered</th>
<th>Normal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Altered</td>
<td>30</td>
<td>20</td>
</tr>
<tr>
<td>Normal</td>
<td>11</td>
<td>52</td>
</tr>
</tbody>
</table>

With regard to the type of alterations (partial alteration, whole-arm deletion, or whole-arm polysomy), several points of interest emerged (Table 3): (a) among 23 of 113 (20%) tumors showing loss of the whole long arm, 8 were associated with loss of the entire short arm, and another 8 were associated with a gain of 7p, suggesting monosomy 7 (all informative loci on chromosome 7 had lost heterozygosity) or isochromosomy 7p (all informative loci had lost heterozygosity on 7q and gained on 7p), respectively. Moreover, we did not observe entirely 7q-deleted tumors with partial alteration at 7p (Table 3); (b) most of...
Alteration Mapping of Chromosome 7 in Breast Cancer

The tumors in which the short arm was gained were deleted partially (5 cases) or globally (8 cases) at 7q (13 of 14, 93%), whereas the frequency of 7q-deleted tumors was only 37% (37 of 99) among the cases without entire 7p polysomy; and (c) we also observed a high frequency of tumors that showed partial alterations of both 7p and 7q; five (56%) of the nine partially 7p-altered tumors were also partially deleted at 7q.

**Correlation between Allelic Imbalance and Clinical and Pathological Parameters.** We sought links between entire 7p LOH, entire 7p allelic gain, and each region of 7q LOH (7q11.23–q22, 7q31.2, and 7q33–q35) in 113 tumors and macroscopic tumor size, histopathological grade, and lymph node or steroid receptor status. The only statistically significant correlation was between 7q33–q35 LOH and high histopathological grade (P < 0.05).

**DISCUSSION**

Various approaches can be used to detect genes that may be tumorigenic when altered. Cytogenetic analyses have been used to locate distinct chromosomal regions that might harbor genes contributing to tumorigenesis. Fine-scale molecular mapping of altered regions is needed to locate such genes precisely, and this can be done by studying the imbalance of heterozygosity. We thus carried out a molecular description of chromosome 7 alterations previously associated with human breast cancer in cytogenetic studies. We found allelic imbalance of chromosome 7 in 61 of 113 human breast tumors studied (54%) using chromosome 7-specific DNA probes. This rate of allelic imbalances on chromosome 7 is one of the highest, compared with other frequently altered chromosomes in human breast tumors, including chromosomes 1, 3, 11, 16, and 17 (22).

We used both microsatellite markers and RFLP/VNTR markers, which both have advantages and drawbacks. Southern hybridization with appropriate controls can be used to assign deletions and polysomy events unambiguously in particular tumor DNAs (Fig. 1). However, RFLP/VNTR markers are poorly informative and require a large amount of tumor DNA. The use of microsatellite repeats assayed by PCR solves some of these problems but creates others. Indeed, microsatellites enable highly informative markers to be analyzed on smaller amounts of tumor DNA, but these markers cannot be used to distinguish between allelic gain and LOH. The ability to determine whether alleles are gained or lost is important in evaluating the possible nature of the target sequence(s); proto-oncogenes may be activated by amplification, whereas tumor suppressor genes are inactivated by other mechanisms as deletion.

The judicious choice of RFLP/VNTR/microsatellite markers regularly mapping to chromosome 7 allowed us to determine that the two arms are subjected to different kinds of alteration. The long arm seems to be affected by LOH exclusively (often partial deletions), whereas the short arm showed both deletions and polysomy, especially of the entire arm.

Most of the tumors with 7p alterations also showed LOH on the long arm (Table 2), a different situation from that observed on chromosome 1, where the alterations observed on each arm are independent (23). In tumors with both entire 7p and 7q alterations, we observed a similar signal intensity ratio between the two alleles with markers located on the two chromosomal arms, suggesting that these alterations arose from the same tumor cells, perhaps simultaneously though the same mechanism. Conversely, in the case of tumor DNAs with partial alterations, we observed differences in signal intensity in some, thus confirming intratumoral polymyclonality.

These findings are consistent with the results of previous cytogenetic studies: the frequency of chromosomal alterations was similar and involved mostly monosomy 7 and 7p, polysomy 7p, isochromosomy 7p, and structural rearrangements of both 7p and 7q (2–5).

Our fine analysis of chromosome 7p alterations in human breast tumors mainly identified DNAs showing loss (18 tumors) or gain (14 tumors) of the whole copy of chromosome 7p. The rearrangements seemed to be associated with a breakpoint in the constitutive heterochromatin, but not in the euchromatin, because the most centromeric marker used (D7S228) located in the euchromatin (7p12) but very near the heterochromatin was altered in all but two (T54 and T212) of these tumors. Partial alterations of 7p (deletions or polysomy), observed in only nine tumors, did not display a consensus for a SCDR or a smaller common multiplied region (Fig. 3). Such tumors may represent either a specific uncommon suppression (or activation) pathway involved in breast tumorigenesis or a background allelic imbalance that is unrelated to gene mutations.

The high rate of gain and loss of the whole chromosome 7p in our panel of tumors suggests that minor changes in gene dosage brought on by either the loss of one copy or the gain of one or several extra copies of the entire short arm of chromosome 7 (affecting hundreds of genes) may be important in human breast cancer. It is noteworthy that multiplication of 7p, where low-level amplification is observed, is due to a genetic mechanism different from that involved in the high levels of gene amplification that is cytogenetically described as homogeneously staining regions and double-minute chromosomes. The latter type of chromosomal alteration, frequently observed in solid tumors, results in 10–100-fold amplification of a small

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**Table 3** Relationship between the different types of alterations at 7p and 7q in breast tumor tissues

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<th>Entire deletion</th>
<th>Entire polysomy</th>
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<td>(isochromosome 7p)</td>
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</tr>
<tr>
<td>(normal)</td>
<td></td>
<td>(monosomy 7q)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Table 2**

<table>
<thead>
<tr>
<th>Tumor DNA status</th>
<th>Entire deletion</th>
<th>Partial deletion</th>
<th>Partial alteration</th>
<th>Entire polysomy</th>
<th>Normal</th>
</tr>
</thead>
<tbody>
<tr>
<td>(monosomy 7)</td>
<td>0</td>
<td>8</td>
<td>4</td>
<td>5</td>
<td>13</td>
</tr>
<tr>
<td>(isochromosome 7p)</td>
<td></td>
<td></td>
<td></td>
<td>8</td>
<td>7</td>
</tr>
<tr>
<td>(monosomy 7q)</td>
<td></td>
<td></td>
<td></td>
<td>1</td>
<td>52</td>
</tr>
<tr>
<td>(normal)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
The number of genes, few of which are thought to contribute in a
dominant manner to the malignant phenotype (genes MYC and
ERBB2 and the 1q13 region in human breast cancer; Ref. 22).

Studies of the expression of several putative cancer genes
on chromosome 7p are needed to confirm the possible gene
dosage effect. Platelet-derived growth factor α (PDGFA), inter-
leukin 6 (IL6), and epidermal growth factor receptor (EGF)
are good candidates (24).

This group of human breast 7p-altered tumors points to an
interesting dosage effect in breast tumor development involving
a large number of genes, rather than to the role of a single or
a few putative tumor gene(s). Conversely, the study of 7q-deleted
tumor DNAs, especially tumors that showed partial deletion of
the long arm of chromosome 7, provided more information on
the location of such putative tumor genes, especially tumor
suppressor genes.

Alteration mapping revealed that breakpoints on chromo-
some 7q differed between tumors, although 13 tumors had
breakpoints between the D7S440 and D7S13 loci (Fig. 3). The
hypothesis that a chromosome 7q breakpoint itself plays an
important role in the development of human breast tumors
cannot thus be ruled out, although these breakpoints are within
a large region of 41 cM.

LOH data for the 27 breast tumors with partial 7q deletion
identified 3 distinct deleted regions on chromosome 7q. It is
noteworthy that two and even three of these SCDRs could be
simultaneously involved in an individual tumor (Fig. 3). We
have previously described one of these deleted regions (7q31.2;
Refs. 8 and 10), whereas the other two regions in the proximal
(7q11.23–q22) and distal (7q33–q35) parts of chromosome 7q
are new.

The 7q31.2-deleted region identified in our first study (8)
has been further investigated to detect LOHs distributed around
the D7S522 locus but in a small series of tumors (10). Our
present findings show that the potential target gene could be
located slightly more telomeric than D7S522, in the small region
between D7S480 and D7S650 that is estimated to be smaller
than 1 cM long (21). In fact, because the markers D7S522,
D7S480, and D7S650 are located very close to one another,
and because the precise order of these markers in this region is
not clearly established, these results obtained on a large tumor series
strongly support the existence of a putative tumor suppressor
gene at 7q31.2 involved in breast cancer.

Deletions in 7q31 have been detected by the use of both
molecular and cytogenetic approaches in a wide variety of
tumors (12–16, 25, 26). The striking concordance between
the central deletions in these different cancers suggests that func-
tional inactivation of the same suppressor gene might play a
common etiological role.

Zenklusen et al. (12–14) have identified D7S522 as the
center of the SCDR at 7q31.2 in all types of cancer studied in
their laboratory, including primary squamous cell, colon, pros-
tate, and ovarian carcinomas. Latil et al. (15) and Takahashi et
al. (16), studying prostate tumors, suggested that the ubiquitous
suppressor gene could be located around D7S480 and D7S486,
respectively. The use of a large number of closely spaced
genetic markers in these different cancers, together with
the construction of a physical map of the 7q31.2 region, could
ultimately lead to the critical gene.

The other two regions of LOH on 7q spanned the loci
D7S440 and D7S13 (7q11.23–q22) and D7S495 and D7S396
(7q33–q35). These distal and proximal regions of LOH on 7q,
described in this study, have not previously been reported in
breast cancer. No common deletions have been reported in the
7q33–q35 region in any other cancer, and no strong candidate
tumor suppressor gene has been located. Inactivation of this
unknown suppressor gene could be a late event in breast tumor
progression, because a significantly higher rate of 7q33–q35
LOH was found in histopathological grade III tumors. On the
other hand, a putative tumor suppressor gene has been located at
7q22. This gene, named CUTL1, encodes a transcription factor
that represses MYC gene expression (27). This 7q22-deleted
region is not specific to breast cancer but is also frequently
observed in uterine leiomyoma (28).

These two new regions are still far apart (about 40 cM), but
there are a number of microsatellite markers that map within
these intervals and can be used to narrow down the target
regions.

In conclusion, this study shows a high frequency (54%) of
complex and multiple genetic changes on both the long and
short arm of chromosome 7 in human breast cancer, suggesting
that genes on this chromosome are involved in human breast
cancer tumorigenesis. In general, 7q was characterized by par-
tial (interstitial and/or telomeric) deletions, whereas gross alter-
ations (both deletion and polysomy) occurred on 7p.

This study both points to a possible gene dosage effect of
chromosome 7p in breast tumor development, reflecting quanti-
tative alteration of expression involving many genes. We also
clearly defined the boundaries of three distinct deleted regions
on chromosome 7q. The availability of high-density polymor-
phic microsatellite loci on chromosome 7q and the construction
of a physical map of these deleted regions should help to locate
and ultimately clone the putative target tumor suppressor
gene(s).

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