Genetic polymorphisms in DNA repair and damage response genes and late normal tissue complications of radiotherapy for breast cancer

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Breast-conserving surgery followed by radiotherapy is effective in reducing recurrence; however, telangiectasia and fibrosis can occur as late skin side effects. As radiotherapy acts through producing DNA damage, we investigated whether genetic variation in DNA repair and damage response confers increased susceptibility to develop late normal skin complications. Breast cancer patients who received radiotherapy after breast-conserving surgery were examined for late complications of radiotherapy after a median follow-up time of 51 months. Polymorphisms in genes involved in DNA repair (APEX1, XRCC1, XRCC2, XRCC3, XPD, P21, TP53) and damage response (TP53, P21) were determined. Associations between telangiectasia and genotypes were assessed among 409 patients, using multivariate logistic regression. A total of 131 patients presented with telangiectasia and 28 patients with fibrosis. Patients with variant TP53 genotypes either for the Arg72Pro or the PIN3 polymorphism were at increased risk of telangiectasia. The odds ratios (OR) were 1.66 (95% confidence interval (CI): 1.02–2.72) for 72Pro carriers and 1.95 (95% CI: 1.13–3.35) for PIN3 A2 allele carriers compared with non-carriers. The TP53 haplotype containing both variant alleles was associated with almost a two-fold increase in risk (OR 1.97, 95% CI: 1.11–3.52) for telangiectasia. Variants in the TP53 gene may therefore modify the risk of late skin toxicity after radiotherapy.

Keywords: TP53; cosmesis; late side effects; radiotherapy; telangiectasia; fibrosis

Radiotherapy is commonly applied after breast-conserving surgery to reduce the risk of locoregional recurrence of breast cancer and has been shown to be as effective as radical mastectomy (Fisher et al, 2002). Although standard radiation therapy is well tolerated by the majority of patients, late normal tissue complications arising from the intrinsic sensitivity of normal tissue, and correlated poor cosmetic results, remain as health concerns of treated breast cancer patients over time (Getinas et al, 2002; Deutsch and Flickinger, 2003; Smith and Ross, 2004). The process of endothelium reconstruction is radiation dose-dependent, progresses over months and years and leads to increases in the severity of both telangiectasia and fibrosis (Bentzen et al, 1989; Archambeau et al, 1995; Chen et al, 2006). Telangiectasias are small dilated blood vessels near the surface of the skin and fibrosis is the development of excess fibrous connective tissue leading to induration. There is, however, considerable inter-individual variability in the development of adverse reactions in normal tissue of irradiated patients. Besides duration, radiation dose and schedule (Turesson et al, 1996; Hill et al, 2001), patient-related factors, such as age, acute skin reaction and lifestyle factors (Bentzen and Overgaard, 1991; Bentzen et al., 1996; Turesson et al, 1996; Johansen et al, 2002; Deutsch and Flickinger, 2003; Chen et al, 2006; Lilla et al, 2007), as well as genetic susceptibility (Bentzen and Overgaard, 1994; Chang-Claude et al, 2005; Andreassen et al, 2006; Popanda et al, 2008) have been implicated. Sensitivity to radiation exposure is suggested to be a complex, polygenic trait, which results from the interaction of a number of genes in different cellular pathways (Travis, 2007).

As radiation therapy exerts its cytotoxic effects through damage to cells, proteins and DNA, the individual capacity to repair damaged DNA may modify the response of the normal tissue. Radiation-induced DNA damage is diverse and therefore nearly all DNA repair pathways might be involved in its removal, especially repair of double-strand breaks through mechanisms such as homologous recombination and non-homologous end joining (Jeggo and Lobrich, 2006). In addition, nucleotide and base-excision repair play an important role, mainly in the repair of oxidative DNA damage (Hoeijmakers, 2001).
Furthermore, the complex response to ionising radiation requires the expression and activity of the p53 pathway (Gudkov and Komarova, 2003). The p53 protein is activated through phosphorylation by radiation DNA damage-induced kinases, including ataxia telangiectasia-mutated and the DNA-dependent protein kinase (Banin et al, 1998; Fei and El-Deiry, 2003; Schwartz, 2007). Activated p53 protein has various downstream targets, including genes involved in cell-cycle regulation, apoptosis, and DNA repair. Regulation of these processes by p53 controls the cellular response to ionising radiation-induced damage. p21 is a critical cell-cycle checkpoint gene, regulated tightly by p53. As soon as DNA is damaged by radiation, binding of p53 protein induces transcription of the downstream gene p21, which stops cells from entering into the S phase (Robles et al, 2002). p21, together with p53, is directly involved in G1/S checkpoint control in response to ionising radiation (Dotto, 2000).

We therefore evaluated the association between several putative functional polymorphisms in six genes involved in DNA repair and two damage response genes and development of late normal tissue complications in a prospective study of breast cancer patients who received radiotherapy after breast-conserving surgery.

MATERIAL AND METHODS

Patient population and data collection

The methods of this study have been described earlier (Twardella et al, 2003; Chang-Claude et al, 2005; Lilla et al, 2007). Briefly, women diagnosed with breast cancer who received radiotherapy after breast-conserving surgery were enrolled between June 1998 and March 2001 from four radiotherapy units in Germany (Women’s Clinic at the University of Heidelberg, St Vincentius Clinic in Karlsruhe, City Hospital in Karlsruhe and University Hospital of Mannheim). Patients who received chemotherapy before or during radiation were not eligible for the study. Information on demographic factors, medical history, and lifestyle factors was obtained through self-administered questionnaires. Details on clinical tumor characteristics and treatment regimen were abstracted from patient records. Informed consent was obtained from all participants, and the study was approved by the ethics committee of the University of Heidelberg, the Institutional Review Board for Roswell Park Cancer Institute, and the US Army Medical Research and Materiel Command Human Subjects Research Review Board.

Breast irradiation

Details on the radiotherapy regimen (total dose, dose per fraction, treatment time, boost dose) were abstracted from the irradiation protocols. As described earlier (Twardella et al, 2003), all patients received a common breast irradiation treatment with conformal tangential irradiation with lateral and medial wedge fields, including CT-based planning, simulation, verification, and quality assurance. At three hospitals, the standard regimen included irradiation of the whole breast, either 50 Gy given in 5 2.0 Gy fractions or 50.4 Gy in 5 1.8 Gy fractions per week, followed by a photon or electron boost with doses ranging from 5 to 20 Gy. Three patients were treated with brachytherapy (20 or 25 Gy). In the fourth radiation department, patients received 56 Gy of whole breast irradiation in 5 2.0 Gy fractions without boost. The biologically effective dose (BED) of radiotherapy relative to an irradiation with a fraction dose of 2.0 Gy, that is the normalised total dose (NTD), was calculated to account for differences in fractionation according to the following formula:

\[
\text{NTD} = \frac{\text{BED}}{1 + 2\text{GY} / (\alpha/\beta)} = \text{n.d.} - (1 + \frac{d}{(\alpha/\beta)}) \left(1 + \frac{d}{(2\text{GY} / (\alpha/\beta))}\right)
\]

given the number of fractions \( n \), the fraction size of \( d \), and an \( \alpha/\beta \) ratio of 3 Gy for telangiectasia and 2 Gy for fibrosis.

Follow-up and evaluation of toxicities

The occurrence of acute side effects of radiotherapy was monitored and documented by physicians several times during the study. We have earlier reported on acute radiation-induced toxicity, defined as grade 2c and above (at least one moist desquamation or interruption of radiotherapy due to toxicity), in this patient cohort (Twardella et al, 2003; Chang-Claude et al, 2005; Ambrosone et al, 2006; Popanda et al, 2006; Tan et al, 2006). Patients were recontacted between June 2003 and July 2005 to assess the occurrence of late adverse effects of radiotherapy and course of disease (relapse, metastases, secondary carcinoma, and death). A self-administered questionnaire similar to that applied at baseline was used to collect information on demographic and epidemiological risk factors, and to record behavior changes that may have occurred after radiotherapy. Patients were examined by the study physician or the treating physician to assess the occurrence of late adverse effects of radiotherapy.

The late side effects were classified according to the RTOG/EORTC late radiation morbidity scoring schema (Seegenschmiedt, 1998) supplemented by LENT-SOMA scores. Patients’ general condition, weight changes, nausea and development of lymphatic edema (arm or breast), and adverse reactions of the skin (telangiectasia), subcutaneous tissue (fibrosis) and other organ tissues (heart, lung, lymph nodes) were recorded. The severity of late effects was scored from 0 to 4, whereby the development of side effects of scores \( \geq 2 \) was considered to indicate late normal tissue complications.

Genotyping assays

Most polymorphisms (see Table 2) were detected by amplification with real-time PCR followed by melting-curve analysis with fluorescence-labeled hybridisation probes in a LightCycler (Roche Diagnostics, Mannheim, Germany) as described earlier (Chang-Claude et al, 2005; Popanda et al, 2006; Tan et al, 2006). The oligonucleotides for analysis of the XRCCI -77 polymorphism (rs3213243) were the PCR primers (sense) 5'-cttacagcgccagctg-3'OH and (antisense) 5'-cccatgcaggtccctcac-3'OH, sensor 5'-ccgcccctccttac-3'FL and anchor 5'-LC Red640-ccctgcccctcggaccccatactcac-3'PF. The sense primer included a mismatch to avoid stem loops in the amplicon because of the high and repetitive G/C content of the target sequence. PCR primers and probes were designed with the help of Tib Molbiol (Berlin, Germany). Annealing temperature of the primers was 60°C. The PCR was performed for all polymorphisms with Qiagen reagents (Qiagen, Hilden, Germany) in a volume of 10 µL using 10 ng of DNA. Overall, 10% randomly selected samples were analysed by conventional PCR-RFLP to verify the LightCycler results; 100% concordance was found. The insertion of the TP53 PIN3 polymorphism was identified by standard PCR and electrophoresis (Tan et al, 2006). A negative control containing all the reagents but with water instead of the DNA template was included in every amplification set. All genotyping assays were carried out blinded to the clinical diagnosis. For each polymorphism, PCR fragments of the homozygous wild-type allele, the homoygous variant allele, and one heterozygous sample were sequenced.

Statistical analysis

Significant differences in distribution of genotypes by presence of late skin toxicities (scores \( \geq 2 \)) were tested by the \( \chi^2 \) and Fischer’s exact tests. Each polymorphism was tested for deviation from Hardy–Weinberg equilibrium by comparing the observed and expected genotype frequencies using the \( \chi^2 \)-test with one degree of freedom. Multivariate unconditional logistic regression analysis was used to assess the association of genotypes with occurrence of late complications of radiotherapy. Odds ratios (OR) and 95%
Results

Data on late effects of radiotherapy as well as information on demographic and epidemiological factors were available for 421 breast cancer patients, as reported earlier (Lilla et al., 2007; Kuptsova et al., 2008). After a median follow-up time of 51 months (range 36–77 months), the most common symptoms of scores ≥2, which were observed included telangiectasia (32.1%), impairment of the general condition (15.9%), fibrosis (7.1%), lymphatic edema in the arm and breast (6.2%), and pain (5.5%). Of 416 patients (after excluding 3 patients treated with interstitial boost and 2 patients with missing information on fibrosis), 131 patients presented with telangiectasia and 28 with fibrosis of grades ≥2, whereby 21 patients presented with both adverse reactions. Characteristics of the 409 breast cancer patients who also had genotype data and were included in this analysis (excluding the seven patients presenting with fibrosis only) are shown in Table 1.

We found a significant association between genetic polymorphisms in the TP53 gene and risk for telangiectasia (Table 2). Compared with non-carriers, patients carrying the variant TP53 72Pro allele had an increased risk of adverse effects (OR of 1.66, 95% CI: 1.02–2.72). Carriers of the TP53 PIN3 A2 allele were also at increased risk of telangiectasia (OR 1.95, 95% CI: 1.13–3.35). None of the other genetic polymorphisms studied showed significant associations with occurrence of telangiectasia.

Strong association (linkage disequilibrium) was found between the TP53 Arg72Pro and TP53 PIN3 polymorphisms (P < 0.001). We therefore investigated haplotype effects of the two TP53 polymorphisms. Compared with the common ArgA1 haplotype, the ProA2 haplotype containing both variant alleles was associated with a significantly increased OR of 1.97 (95% CI: 1.11–3.52) for telangiectasia (Table 3). Haplotype association analysis for the XRCC1 and XPD genes with data for at least two genetic polymorphisms did not reveal further significant findings.

Further analysis for effect modification yielded differences in the effect of TP53 on risk for telangiectasia, according to occurrence of acute skin toxicity (moist desquamation). Thirty women (22.9%) had presented with acute skin toxicity during radiotherapy in patients with telangiectasia, and 45 women (16.2%) in those without telangiectasia. The elevated risk of telangiectasia associated with the TP53 ProA2 haplotype was found only in patients who did not present with acute toxicity during radiotherapy (OR 2.78, 95% CI: 1.44–5.35) and not in those who experienced acute skin toxicity during radiotherapy (P heterogeneity = 0.06) (Table 3).

DISCUSSION

In this study of breast cancer patients treated with radiotherapy after breast-conserving surgery, we found that variants of TP53 were associated with an increased risk for developing telangiectasia after radiation therapy. Although both variants, TP53 72Pro and PIN3 A2, were associated with elevated risk, the haplotype results suggested that cis effects of the two variants may be most relevant. Two of the many p53 functions may be important in modulating radiosensitivity. Growth arrest mediated by p53 is relevant.
### Table 2: Association between polymorphisms in DNA repair and cell-cycle genes and risk of developing late skin toxicity (telangiectasia) with score $\geq 2$ after radiotherapy

| Gene polymorphism | Genotype | $N = 278$ | % | $N = 131$ | % | OR* | 95% CI |
|-------------------|----------|----------|---|----------|---|-----|-------|
| APEX1             | TT       | 71       | 25.9 | 39       | 30.7 | 1.00 |        |
|                  | TG       | 134      | 48.9 | 65       | 51.2 | 1.03 | 0.58–1.83 |
|                  | GG       | 69       | 25.2 | 23       | 18.1 | 0.66 | 0.33–1.32 |
|                  | TG+GG    | 203      | 74.1 | 88       | 69.3 | 0.90 | 0.53–1.54 |
| XRCC1             | TT       | 94       | 34.2 | 43       | 33.9 | 1.00 |        |
|                  | TC       | 136      | 49.5 | 54       | 42.5 | 0.97 | 0.56–1.67 |
|                  | CC       | 45       | 16.4 | 30       | 23.6 | 1.87 | 0.94–3.70 |
|                  | TC+CC    | 181      | 65.8 | 84       | 66.1 | 1.17 | 0.71–1.95 |
| XRCC1             | CC       | 242      | 88.3 | 117      | 92.1 | 1.00 |        |
|                  | CT       | 30       | 11.7 | 10       | 7.9  | 0.58 | 0.24–1.40 |
|                  | TT       | 2        | 0.7  | 0        | 0    | 1.00 |        |
|                  | CT+TT    | 32       | 11.7 | 10       | 8    | 0.57 | 0.24–1.38 |
| XRCC1             | GG       | 244      | 88.4 | 118      | 92.9 | 1.00 |        |
|                  | GA       | 30       | 10.9 | 9        | 7.1  | 0.49 | 0.19–1.24 |
|                  | AA       | 2        | 0.7  | 0        | 0    | 1.00 |        |
|                  | GA+AA    | 32       | 11.6 | 9        | 7.1  | 0.43 | 0.17–1.09 |
| XRCC1             | GG       | 112      | 40.6 | 50       | 39.4 | 1.00 |        |
|                  | GA       | 120      | 43.5 | 63       | 49.6 | 1.09 | 0.65–1.82 |
|                  | AA       | 44       | 15.9 | 14       | 11   | 0.63 | 0.29–1.37 |
|                  | GA+AA    | 164      | 59.4 | 77       | 60.6 | 0.96 | 0.59–1.57 |
| XRCC2             | GG       | 236      | 85.5 | 113      | 89   | 1.00 |        |
|                  | GA       | 38       | 13.8 | 13       | 10.2 | 0.83 | 0.39–1.76 |
|                  | AA       | 2        | 0.7  | 1        | 0.8  | 1.05 | 0.08–13.93 |
|                  | GA+AA    | 40       | 14.5 | 14       | 11   | 0.84 | 0.41–1.74 |
| XRCC3             | CC       | 104      | 38    | 45       | 35.4 | 1.00 |        |
|                  | CT       | 126      | 46    | 63       | 49.6 | 1.05 | 0.62–1.79 |
|                  | TT       | 44       | 16.1 | 19       | 15   | 1.12 | 0.53–2.40 |
|                  | CT+TT    | 170      | 62    | 82       | 64.6 | 1.07 | 0.65–1.77 |
| NBS1              | GG       | 120      | 43.5 | 53       | 41.7 | 1.00 |        |
|                  | GC       | 137      | 49.6 | 58       | 45.7 | 0.92 | 0.55–1.54 |
|                  | CC       | 19       | 6.9  | 16       | 12.6 | 2.14 | 0.88–5.19 |
|                  | GC+CC    | 156      | 56.5 | 74       | 58.3 | 1.06 | 0.65–1.72 |
| XPD               | GG       | 120      | 43.8 | 42       | 33.3 | 1.00 |        |
|                  | GA       | 117      | 42.7 | 69       | 54.8 | 1.51 | 0.89–2.55 |
|                  | AA       | 37       | 13.5 | 15       | 11.9 | 0.91 | 0.41–2.01 |
|                  | GA+AA    | 154      | 56.2 | 84       | 66.6 | 1.36 | 0.82–2.24 |
| XPD               | AA       | 109      | 39.6 | 42       | 33.3 | 1.00 |        |
|                  | AC       | 133      | 48.4 | 65       | 51.6 | 1.15 | 0.68–1.95 |
|                  | CC       | 33       | 12    | 19       | 15.1 | 1.21 | 0.57–2.58 |
|                  | AC+CC    | 166      | 60.4 | 84       | 66.6 | 1.16 | 0.70–1.92 |
| P21               | CC       | 242      | 87.7 | 110      | 86.6 | 1.00 |        |
|                  | CA       | 31       | 11.2 | 17       | 13.4 | 1.54 | 0.71–3.32 |
|                  | AA       | 3        | 1.1  | 0        | 0    | 0    |        |
|                  | CA+AA    | 34       | 12.3 | 17       | 13.4 | 1.27 | 0.60–2.68 |
| TP53              | GG       | 160      | 58.0 | 64       | 50.4 | 1.00 |        |
|                  | GC       | 96       | 34.8 | 49       | 38.6 | 1.67 | 0.98–2.83 |
|                  | CC       | 20       | 7.3  | 14       | 11   | 1.62 | 0.71–3.70 |
|                  | GC+CC    | 116      | 40    | 63       | 49.6 | 1.66 | 1.02–2.71 |
| TP53              | AA       | 214      | 77.6 | 87       | 68.5 | 1.00 |        |
|                  | A1A1     | 56       | 20.3 | 40       | 31.5 | 2.14 | 1.23–3.71 |
|                  | A1A2     | 6        | 2.2  | 0        | 0    | 1.00 |        |
|                  | A1A2+A2A2| 62       | 22.4 | 40       | 31.5 | 1.95 | 1.13–3.37 |

CI = confidence interval; NTD = normalised total dose; OR = odds ratio. *Adjusted for NTD, age at the time of late toxicities evaluation, time since radiotherapy (months), clinic, acute skin toxicity, high blood pressure, allergy, pack-years (never, $<20$, $\geq 20$), skin type (always/moderate/seldom sunburn), clinic, marital status (single/divorced/widowed, married/partner).
Genetic factors for late radiation-related side effects

Table 3 Reconstructed haplotypes and the association with risk of developing late skin toxicity (telangiectasia) with score $\geq 2$ after radiotherapy

| Gene   | Haplotype | Frequency | OR* | 95% CI  |
|--------|-----------|-----------|-----|---------|
| **All patients** |
| TP53   |
| GA1    | 0.71      | 1         |     |         |
| GA2    | 0.03      | 1.02      | 0.29–3.63 | |
| CA1    | 0.16      | 1.20      | 0.79–1.82 | |
| CA2    | 0.11      | 1.97      | 1.11–3.52 | |
| **Patients without acute toxicity during radiotherapy** |
| TP53   |
| GA1    | 0.71      | 1         |     |         |
| GA2    | 0.02      | 0.68      | 0.13–3.60 | |
| CA1    | 0.16      | 1.15      | 0.71–1.85 | |
| CA2    | 0.12      | 2.78      | 1.44–5.37 | |
| **Patients with acute toxicity during radiotherapy** |
| TP53   |
| GA1    | 0.71      | 1         |     |         |
| GA2    | 0.04      | 1.10      | 0.09–13.41 | |
| CA1    | 0.17      | 1.47      | 0.52–4.17 | |
| CA2    | 0.09      | 0.52      | 0.11–2.53 | |
| **All patients** |
| XRCC1  |
| CCGG   | 0.41      | 1         |     |         |
| TCGG   | 0.12      | 1.15      | 0.66–2.00 | |
| TCGA   | 0.36      | 0.78      | 0.53–1.15 | |
| TTGG   | 0.05      | 0.51      | 0.21–1.24 | |
| Rare*  | 0.07      | 0.36      | 0.26–1.20 | |
| XPD    |
| GA     | 0.56      | 1         |     |         |
| GC     | 0.09      | 1.25      | 0.64–2.46 | |
| AA     | 0.06      | 1.14      | 0.55–2.38 | |
| AC     | 0.29      | 1.09      | 0.74–1.62 | |

CI = confidence interval; NTD = normalised total dose; OR = odds ratio. *Adjusted for NTD, age the time of late toxicities evaluation, time since radiotherapy (months), clinic, acute skin toxicity, high blood pressure, allergy, pack-years (never, <20, $\geq 20$), skin type (always/moderate/seldom sunburn), clinic, marital status (single/divorced/widowed, married/partner). **A2 allele carries a duplication of 16 bp in intron 3.

In all, ten polymorphisms causing an amino acid change in six different DNA repair genes were investigated for associations with telangiectasia, but no significant effects were detected. The TP53 Arg399Gln polymorphism has been reported to be associated with telangiectasia but not with fibrosis, particularly in patients who did not receive a boost, albeit based on 167 patients in whom 39 presented with telangiectasia (Giovanella et al., 2007). This polymorphism was also not found to be associated with severe grade 3 fibrosis after irradiation of the breast (Andreassen et al., 2006). A further study, which did not differentiate between early and late adverse reaction to radiotherapy, reported an elevated risk in women carrying both the variant alleles of the Arg194Trp and the Arg399Gln polymorphisms (Brem et al., 2003) and a protective effect for the T-C-G-G haplotype determined by all four XRCC1 genetic polymorphisms, -77T$>$$>$$>$$>0$, Arg194Trp, Arg280His, and Arg399Gln polymorphisms (Brem et al., 2006). Although the results appear divergent, the studies differ in the specific type(s) of adverse reactions being studied, the length of follow-up for side effects, and adjustment for patient-related factors; therefore, comparison of the findings is problematic. Polymorphisms in XRCC1 and APEX1 were studied in breast cancer patients receiving radiotherapy (summarised in Chistiakov et al., 2008; Popanda et al., 2008). Consistent with our null results, all of these studies failed to show a contribution of these SNPs to the risk of adverse reactions after radiotherapy, implying that they may not be promising candidates for predicting late radiosensitivity.

To our knowledge, this is the first epidemiological study on the two TP53 genetic variants as predictors of late tissue reactions to radiotherapy. However, both the TP53 codon 72 and intron 3 variants have been found to be associated with poorer prognosis of non-small cell lung cancer (Baldini et al., 2008). Patients receiving chemoradiotherapy for advanced head and neck cancer were found to have higher response rates and survival when their tumors expressed the proapoptotic 72 Arg allele (Sullivan et al., 2004).

This study has a number of strengths. Breast cancer patients from this cohort were treated similarly, with radiation dosage carefully assessed, and patients were followed prospectively. Improved radiation techniques at the time of patient recruitment, as well as retrieval of individual irradiation dose methods and records, allowed for proper calculations of BED. The phenotype was precisely defined using the standardised scoring system for late toxicity. In addition, we accounted for patient- and treatment-related factors that influenced risk for telangiectasia when assessing the effect of the genetic variants.

Both telangiectasia and subcutaneous fibrosis are among the most common long-term skin side effects of radiotherapy.
Owing to differences in physiological response to radiation of the various skin layers involved and thereby possible differing genetic susceptibility, we opted to restrict the present analysis to telangiectasia because of the limited occurrence of fibrosis and therefore restricted power. Progressive nature of these complications, together with longer time to follow-up, may permit later analyses of late normal tissue complications in this cohort in the future.

In conclusion, this prospective study showed that variants in the TP53 gene are associated with risk of late skin toxicity after accounting for patient-related factors and treatment modalities. As this is the first report on the involvement of p53 in late skin adverse effects, replication of these findings in other studies is encouraged. Advances in the search for biomarkers of radiation-induced late skin side effects may lead to improved treatment choices for breast cancer patients, and improve cosmetic outcome as well as quality of life after surviving breast cancer.

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