

## Original Article

# Therapeutic effects of cytoprotective agent on breast reconstruction after breast cancer surgery

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**Abstract:** Most patients will choose breast reconstruction after breast cancer surgery, while radiotherapy will damage skin and soft tissue so that will have adverse effect on reconstruction. In this study, we assume that the usage of Amifostine can reduce the incidence of complications after breast reconstruction so that provides more choices of reconstruction operation. Dividing SD rats into surgical placement expansion material group (include 15 ml normal saline) and simple operation group. Then further divide the former into non intervention group, radiation group and Radiation therapy combined with Amie amifostine treatment group. The decubation is 45 days after operation. Macroscopic evaluate the complications of skin and soft tissue by ImageJ. There is no obvious complications of skin and soft tissue for control group, radiotherapy alone group and radiotherapy with application of Amifostine group by macroscopic evaluation. The animals that are in expanded object group, damage probability of skin and soft tissue when use Amifostine is lower than that of radiotherapy alone group (30% vs. 69%,  $P=0.041$ ). ImageJ shows the necrosis probability of skin and soft tissue when use Amifostine is obvious lower than radiotherapy alone group (6.96% vs. 12.94%,  $P=0.019$ ). In conclusion, prevention and treatment of Amifostine can significantly reduce the complications of skin and soft tissue which is helpful to breast reconstruction after breast cancer surgery.

**Keywords:** Mammary cancer, breast reconstruction, cytoprotective agent, Amifostine

## Introduction

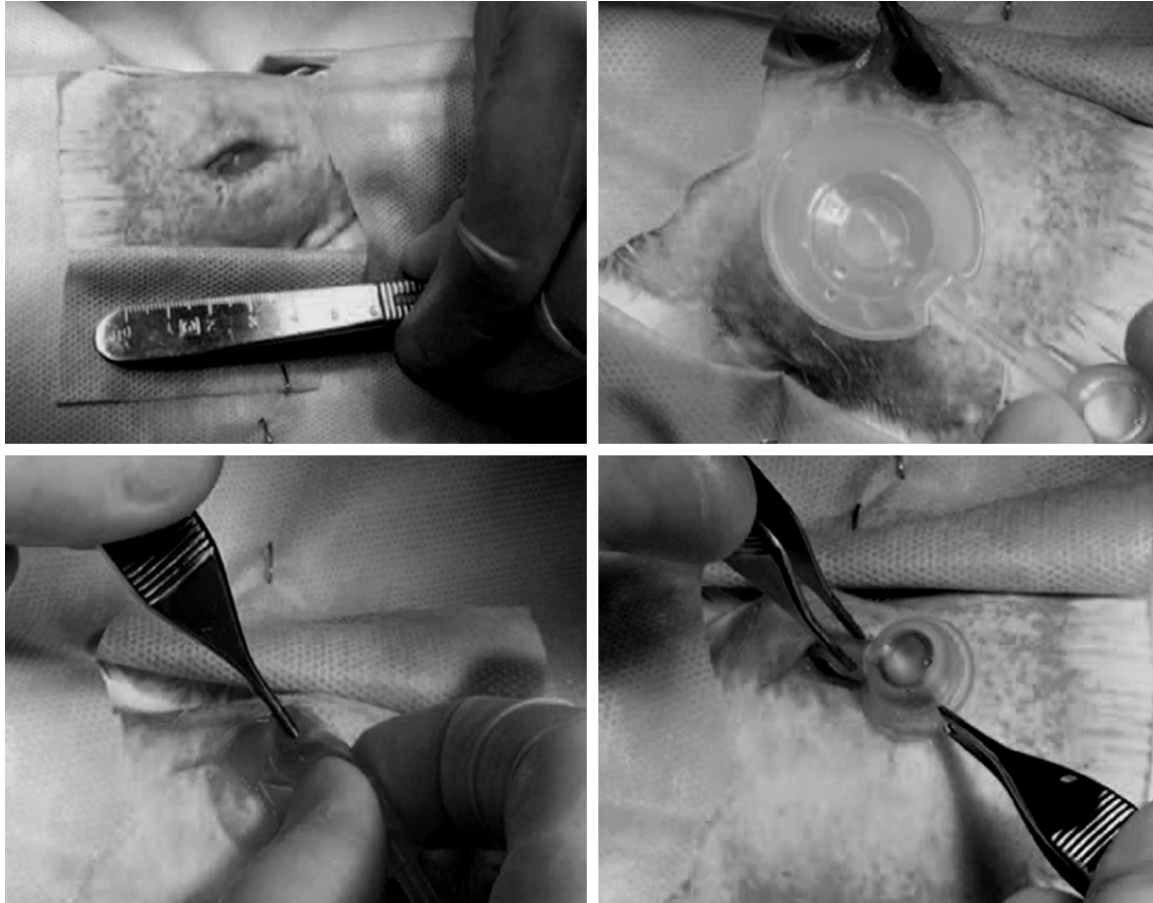
Mammary cancer is one of the malignant tumors which can cause female death. It is reported that the risk of having mammary cancer is 12.5%. With the development of social cognition and clinical examination technology, the diagnoses of mammary cancer are in the early stage at present. Therefore, we can adopt less invasive treatment method to get better outcomes [1, 2]. Besides excising lesions by operation, postoperation radiotherapy is always the necessary steps. Although it can reduce the palindromia of tumor in some degree, at the same time, it can also cause collagen synthesis disorders, prolonged inflammation, poor wound healing and other adverse effects which can directly increase the risk of breast reconstruction. Some study show the incidence rate of complications reaches 60%, include infection of incisional wound, expansion material shift, Tissue contracture and etc, even can bring serious physiological and psychological damage to

patient [3-5]. Amifostine belongs to cytoprotective agent which are always used in xerostomia and mucositis that are caused by head and neck tumors postoperative radiotherapy. It can repair the damaged DNA by cleaning the regulation of cell cycle checkpoint free radical and gene expression. At present, study shows Amifostine will not influence the effect of radiotherapy so that it will not increase the recurrence risk of tumor and have no influence on prognosis of patient [6, 7]. This study is aimed at exploring the influence of prophylactic usage of Amifostine on skin and soft tissue after radiotherapy, which is propitious to breast reconstruction after breast cancer surgery. It provides some experiences for clinical diagnosis and treatment.

## Materials and methods

### *Animal grouping and operative procedure*

Dividing 60 male SD rats into two groups: operation with expansion object (15 rats will only



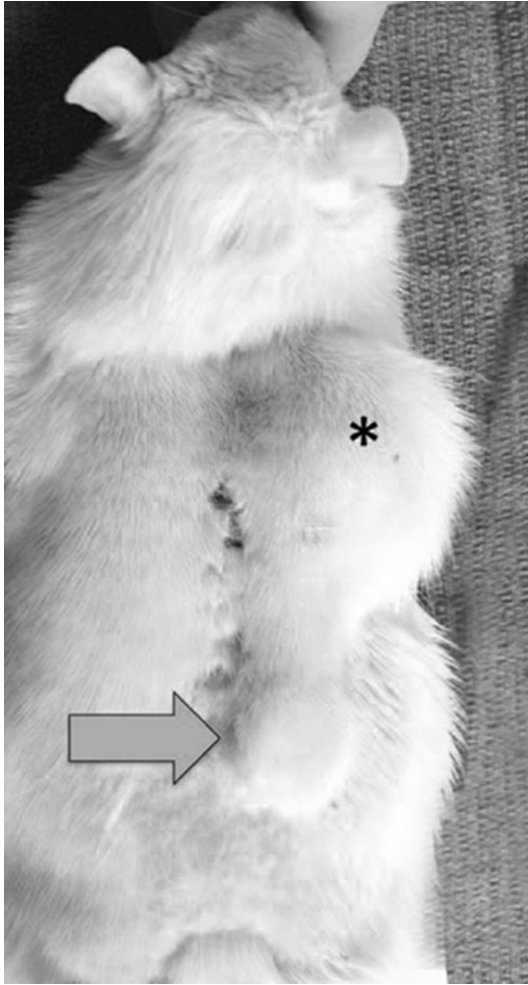
**Figure 1.** Picture of operation, median back incision , the placement direction and position of expansion object and the placement of caudal marker.

receive radiotherapy, 15 rats will receive radiotherapy and Amifostine treatment) and simple operation group (10 rats for each group: control group, radiotherapy alone group and Amifostine group). Adopt blank control group to exclude the adverse influence of simple operation on the postoperative complications. Put the rats in the low-flow isoflurane and oxygen chamber to finish anesthesia induction. After that, put them on temperature suitable plates and use silksuture to fix isoflurane and oxygen inhalation device. Use eye lubricants to protect bilateral eyes. Subcutaneous injection of cefazolin (60 mg/kg) in 45 min before operation and add to the original amount for 2 times every 12 h to prevent infection. Give them labour pains by subcutaneous injection of 0.04 ml buprenorphine and at the same time, adopt 25 ml/kg Lactated Ringer's Solution as operation fluid. Do the depilatory preparation in a 8×6 cm area of the right back and the middle back part. Put

the animals in prone position, on a sterile towel. Cut 3 cm long longitudinal incision in the lower corner of the scapula position along the right dorsal midline, at the same time, cut 2 cm long incision in the latissimus dorsi to form a latissimus dorsi pocket. Put asepsis smooth texture of mini extender (diameter is 3 cm, Allergan, Inc., Santa Barbara, Calif.) in laboratory. Put the other side in the caudal superficial subcutaneous space as marker (**Figure 1**). It is obvious that its platysma expands, then sew up the incision, put the rats in suitable environment to observe continuously. For the control group, all the operative procedures are the same and implant nothing.

## Postoperative recovery

The decubation of all the animals is 2 weeks after operation. During this period, check whether there is any wound infection, hematoma, tissue necrosis and breakdown, displace-



**Figure 2.** Twenty-one days after operation, inject 15 ml normal saline into extended object (marked by '\*'), arrowhead shows the distal end of the marking position.

ment of expansion object and etc. Measure the weight to evaluate nutriture every day. After operation, give 5 ml 0.9% normal saline for 3 times at 15 d, 18 d and 21 d to form a 15 ml expansion volume (**Figure 2**).

#### *The application of Amifostine and radiotherapy*

The patients always need to receive the radiotherapy of 50-60 Gy dosage for 5-6 weeks after breast cancer surgery. So in this study, we adopt the positive voltage unit of Philips RT250 (250-kV x-rays, 15 mA; Kimtron Medical, Oxford, Conn). There are 5 phases in the radiotherapy for rats, 5.6 Gy for each phase (total amount is 28 Gy), the dose rate is 147.7 cGy/min which is close to the radiation dose for human. Inject 100 mg/kg Amifostine to the

dorsal subcutaneous of rats in 45 min before radiotherapy. Put the rats in a sealed container filled with isoflurane to anesthesia induct and then transfer the rats to a radiation chamber where only exposes a circular area with the coverage of a expanded object which diameter is 4 cm. Other areas will be covered by lead shielding materials to protect. During the process of radiotherapy, keep the supply of low-flow isoflurane and oxygen. Put the rats in the cage alone and then wake and observe them in a suitable environment when the treatment finish.

#### *Postoperative observation and ImageJ analyze*

For a period of 45 days, check the position of operation and radiotherapy to evaluate whether there is any erythema, necrosis, lipotrichia or tissue thinning, displacement of expanded object, any damage of skin or soft tissue. Take photos to compare and record the situations of related complications. All the animals will be executed after 45 days and do the depilatory preparation on the back, choose a 6×4 cm area to analyze. All the rats that are in the subzone will get radiotherapy and expanded object at the same time. Use ImageJ to choose the damaged area of skin and soft tissue and calculate the corresponding pixel, and the ratio of its pixel and the pixel of the whole area will be used as the quantization scale of the damage of skin and soft tissue (**Figure 3**).

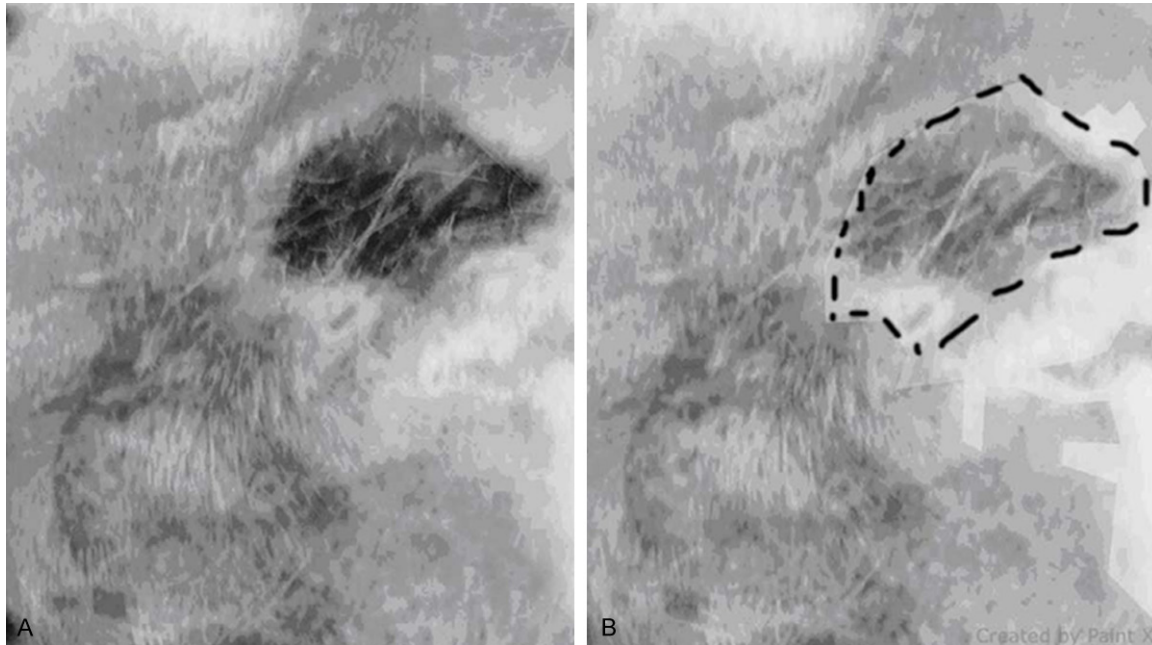
#### *Statistical analysis*

ImageJ data use T-test to check and calculate the percentage of the affected skin and soft tissue. Use two-sample Mann-Whitney U test to analyze all other variables.  $P < 0.05$  is defined as the results have statistical significance.

## **Results**

#### *Blank control group*

During the period of 45 d after radiotherapy, we find no complications of skin, soft tissue injury, wound breakdown and etc that are related with radiation by eyes for blank control group, radiotherapy alone group and radiotherapy with application of Amifostine group. And point out that the potential decisive factor of affecting operation site complications are the expansion operation itself, radiation and the usage of



**Figure 3.** Use ImageJ to choose a 6×4 cm area to compare necrosis status of skin and soft zone. A. The selected area; B. The area that is selected and analyzed by ImageJ.

**Table 1.** Comparison of the complication rate between radiotherapy alone group and radiotherapy with application of Amifostine group

	Radiotherapy alone group (%)	Radiotherapy with application of Amifostine group (%)
Case number	14	14
Necrosis	6 (43)	4 (27)
Erythema	7 (50)	4 (27)
Gression prominent	2 (14)	0 (0)
Depilation/tissue thinning	8 (57)	1 (7)
Hematoncus	2 (14)	0 (0)
Total	9/13 (69)	4/13 (31)

Amifostine. For that reason, we won't do the related quantitative comparison by ImageJ.

#### *Simulation group of expanded object breast reconstruction*

There are 9 cases and 14 cases have apparent skin and soft tissue changes in radiotherapy alone group, include the form of erythema, necrosis and hematoncus. Two cases have the displacement of expanded object, 1 case appears saline volume reduction (judged by the separation of expanded object and marker position change). The related complications include erythema, necrosis and depilation/tis-

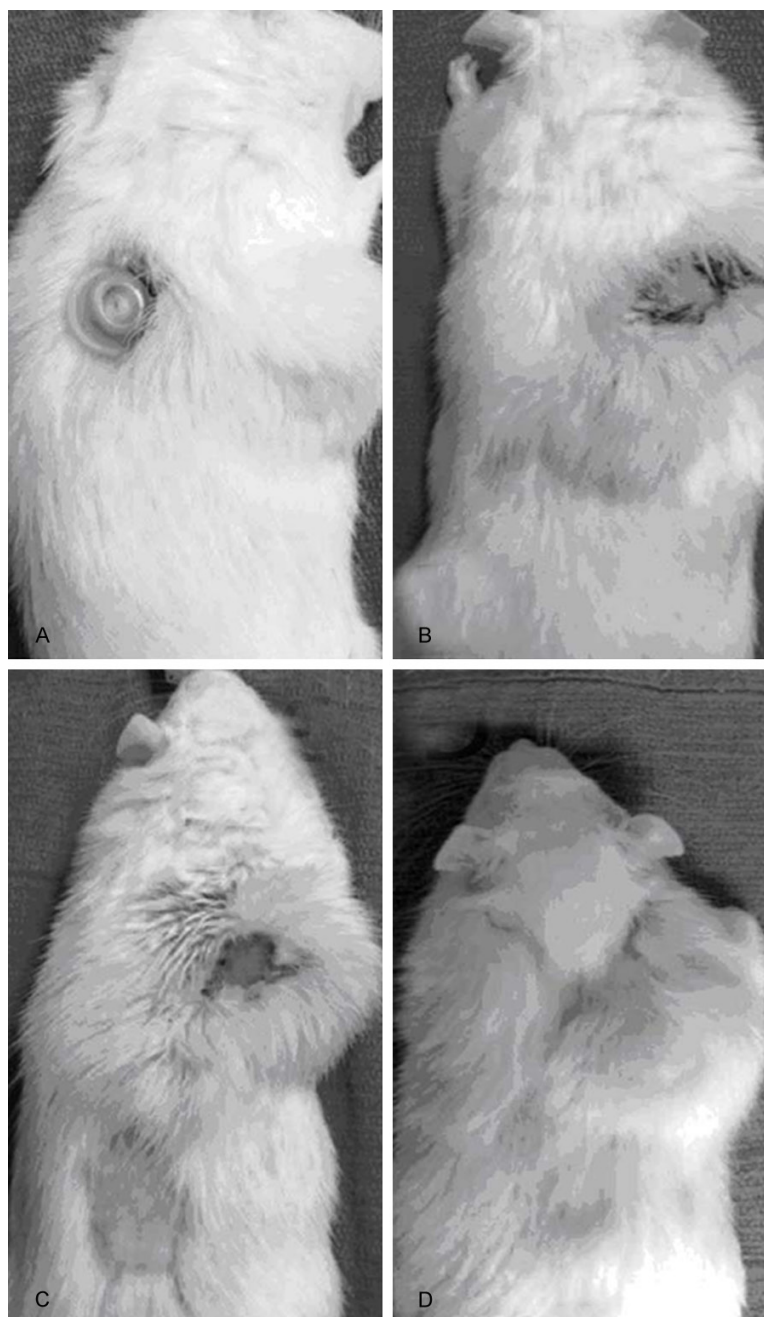
sue thinning in radiotherapy with application of Amifostine group. There are 2 cases appear saline volume reduction which is similar with the radiotherapy alone group, but it is not included in the analysis of results. **Table 1** shows the morbidity of the damage of skin and soft tissue of the 2 groups are 69% and 31% respectively, Amifostine group is obviously lower than radiotherapy alone group, the results have statistical significance ( $P=0.041$ ). **Figure 4** shows the related complications which are observed in the tset. Besides using eyes to evaluate the complications of the operation sites, we also use ImageJ to analyze

the damage ratio of skin and soft tissue in the target area of 6×4 cm. **Figure 5** is the result which shows the average damage area of Amifostine group is lower than that of radiotherapy alone group (6.96% vs. 12.94,  $P=0.019$ ).

#### **Discussion**

Radiotherapy is the main method of adjunctive therapy after tumorectomy for breast cancer patients. This method can reduce danger coefficient of tumor recurrence and increase survival rate, but at the same time, the negative effects which are caused by radiation will seriously affect breast reconstruction [8, 9].





**Figure 4.** Related complications of skin and soft tissue. A. The displacement of expanded object is obvious; B, C. Erythema and necrosis; C, D. Depilation/tissue thinning, the incidence rate of complications of Amifostine group is obvious lower than that of radiotherapy alone group.

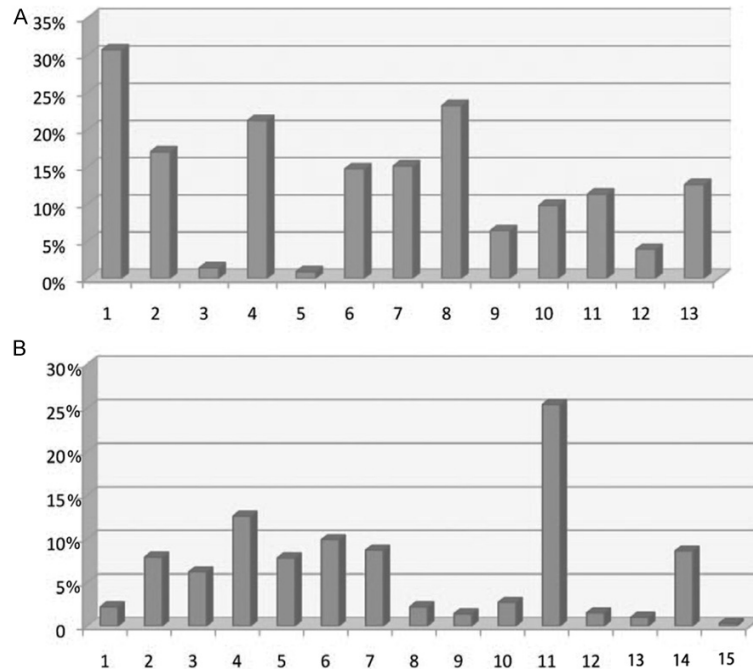
Breast reconstruction after mastectomy is very important for breast cancer patients to recover their physiological and psychological rehabilitation. After the resection, the immediate breast reconstruction usually put the prosthesis in mastectomy site, which possesses the advantages of shorter operative time, improvement

of appearance, the tissues will not influenced by radiation and etc. But the sustained high-dose radiotherapy will cause wound rupture, cutaneous necrosis, prosthesis exposure and etc which cause the failure rate reaches 21%-60%. So nearly 1/3 patients need to accept revision surgery [10-12]. Prosthesis implantation of postponed breast reconstruction always requires autologous flap, but the injury of skin and soft tissues which are caused by postoperation radiotherapy will cause the failure rate of reconstruction reaches 50% [13]. Although the operative timing and surgical approaches are different, postoperative radiotherapy will increase the risk of complications after breast reconstruction and seriously affect the life quality and rehabilitation of patients [13].

In this study, we imagine that the usage of Amifostine can reduce the incidence of complications after breast reconstruction so that provides more choices of reconstruction. We adopt rat model and imitate the breast reconstruction which is based on tissue expansion. Blank control group shows no obvious injury of skin and tissues and points out that risk factors of tissue necrosis could be the expansion operation itself, postoperation radiotherapy and the usage of Amifostine. During the experiment, only accept

the obvious differences between radiotherapy alone group and radiotherapy with application of Amifostine group in the aspect of animal lip-sotrichia, tissue thinning, erythema, necrosis and hematocrit. Macroscopic observation shows the damage probability of skin and soft tissues which are caused by the latter is lower

## Effect of cytoprotective agent on breast reconstruction



**Figure 5.** The percentage of skin necrosis and soft tissue in quantifying regional. A is radiotherapy alone group, B is radiotherapy with application of Amifostine group.

than that are caused by the former (31% vs. 69%,  $P=0.014$ ). Use ImageJ to quantitative evaluate radiation area, the result shows that the average value of necrotic area for the group which only receive the radiotherapy is 2 times than the group which receive the radiotherapy and Amifostine treatment (12.94% vs. 6.96%,  $P=0.019$ ). Therefore, amifostine possesses the advantages of reducing the radiation complications which are caused by radiotherapy. And it can reduce the difficulty of reconstruction or revision surgery and improve the treatment satisfaction.

The limitation of this study is using naked eye to macroscopic evaluate operative site and tissue necrosis. And the assessors knows specific situation of grouping so that there is a certain bias when evaluate. In the late period, we compare and analyze the pictures by using ImageJ, but it is necessary to train the assessor. If the assessor knows nothing about the grouping, it will increase the accuracy of result. Some studies confirm that necrosis of skin and tissues which are caused by radiotherapy radiation is related with vascular damage and dysfunction [14-16]. In the next step, it is necessary to microscopic evaluate vascular density param-

ter of local tissues and the improvement of tissue morphology after Amifostine treatment to further confirm the importance for it to reconstruction after breast cancer operation.

In conclusion, Amifostine has significant clinical application value for shaping operation after radiotherapy for breast cancer. Amifostine can also reduce the related biomechanical load of tumor and side reaction of skin and soft tissues. Amifostine provides more choices and reduces related risk and difficulty, so it is worthy of clinical reference and promotion.

### Disclosure of conflict of interest

None.

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