



# Evaluation of frozen-section analysis of surgical margins in the treatment of breast cancer

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## Summary

**Objective:** To evaluate surgical margins in cases of ductal carcinoma through a histopathological exam using frozen sections. **Materials and Methods:** Retrospective study encompassing 242 conservative surgeries, 179 of which included intraoperative frozen-section histopathology and 63 intraoperative nonfreezing techniques (macroscopy/gross examination and cytology). The results of such analyses were compared with those of the histology processing following paraffin embedment and hematoxylin and eosin (H & E) staining. A margin was deemed free when the distance between the tumor and the surgical border was equal to or greater than two millimeters. The factors given consideration for possibly affecting the results were: age, surgical aspects (skin removal and widening of surgical margins), histopathological findings (size, affected lymph nodes, and angiolymphatic invasion), and extensive intraductal and immunohistochemical components (estrogen, progesterone, Ki-67, and HER-2 receptors). In the statistical analyses, the chi-square test was used and negative predictive values were calculated. **Results:** The negative predictive values were 87.1% and 79.3% for frozen and nonfrozen sections, respectively. There was no significant difference between the two groups ( $p = 0.14$ ). The factors under consideration had no influence on the results of the intraoperative exam of the margins. **Conclusion:** The present study allowed to conclude that the intraoperative exam of the surgical margins by frozen section is not superior to a macroscopy and / or cytology exam.

**Key words:** Breast cancer; Conservative surgery; Intraoperative analysis of margins; Freezing of margins.

## Introduction

The major issue regarding conservative surgical treatment of breast cancer is local remission (LR), which requires reintervention [1] and may produce negative psychic disturbances in women [2, 3].

Recent data indicate a decrease in local remission rates and give evidence of factors associated with greater or lesser LR in oncology follow-up [4-11]. The most relevant of such factors is the absence of compromised surgical margins. Clinical essays have shown that the LR rate ranges from 3% to 13% when margins are free. In compromised cases, these rates are much higher (21% - 31%) [4-11]. Intraoperative investigation allows immediate on-the-spot evaluation of surgical margins enabling reintervention during the same surgery, thus lowering the rates of new operations [12].

There are several ways of examining resection margins intraoperatively, such as: a macroscopic study or gross exam (visual measurement of the distance between the tumor and the resection margins) [12], a microscopic study (imprint and / or cancer cytology [13], and frozen section of the margins close to the tumor [12]. All techniques yield variable degrees of false-negative results when they are compared with the anatomopathological exam in paraffin [14-16]. However, there are not enough comparative data in the literature concerning the different techniques for analyzing resection margins in the intraoperative exam.

## Materials and Methods

A retrospective study was conducted based on the reports of intraoperative anatomopathological exams of 242 patients with a preoperative diagnosis of invasive ductal carcinoma (IDC). The women underwent conservative surgery with an intraoperative exam of the margins. All 242 patients were given a diagnosis of free margins at the same evaluation. The study protocol was approved by the institutional ethics committee.

The patients were divided into two groups according to the histopathological technique employed in the intraoperative exam. In group A ( $n = 179$ ) a freezing technique was applied, and in group B ( $n = 63$ ), a nonfreezing procedure (macroscopy and/or cytology) was applied. The intraoperative exam results were compared with those of the definitive postoperative exam (paraffin-embedded sections and H & E staining). The final anatomopathological report stated that the resection margins were insufficient (presence of invasive or intraductal carcinoma on the resection border or less than two mm away from it), or free (absence of tumor at least two mm away from the nearest margin) [17].

All patients with definitive positive or close margins were sent to additional extended surgery and they were recorded as false-negative results of the intraoperative exam.

The method of intraoperative evaluation of the margins was described by Pinotti and Carvalho in 2002 [17]. The method for evaluating hormone (estrogen and progesterone) and HER-2 receptors was described by Fernandes *et al.* in 2009 [18]. Histological classification followed the criteria laid down by the World Health Organization (WHO) [19]. The parameters evaluated in this study were tumor size, lymph node stage [20], extensive intraductal component, angiolymphatic invasion [19], need for extended surgery [21], and hormone and HER-2 receptors [18]. Other analyses were carried out classifying patients according to their clinical stage following the system proposed

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by the seventh edition of the American Joint Committee on Cancer (AJCC) [20].

The immunohistochemical subdivision of breast cancer by analogy with molecular subtypes met the criteria proposed by Nielsen *et al.* in 2004 [22], and used the following classification: luminal (ER+ and / or PR+ and HER2-), HER2 (ER-, PR-, and HER2+) and triple negative (ER-, PR-, and HER2-).

#### Statistical analysis

To compare the medians from each group, the Mann-Whitney test was applied, and to compare proportions, the chi-square and the FISH tests were applied. A 5% significance level was adopted [23, 24]. A sample size requiring a minimum of 60 patients per group for 90% power was used based on the anatomopathological exam.

## Results

Of the 242 patients in this study, 85.1% had free margins in the histopathological exam. In group A, 87.1% in the frozen-section exam, and in group B, 79.3% in the nonfreezing tests (macroscopy / gross exam and cytology). There was no significant difference between the two groups ( $p = 0.135$ ).

The parameters that were evaluated and that could have influenced the results concerning free margins did not significantly differentiate the two groups (Table 1). With respect to freezing, the patients were grouped according to final stage, i.e., compromised margins or free margins. No significant differences were found among the parameters shown in Table 2.

Table 1. — Characteristics of total number of patients and of subgroups with and without intraoperative frozen-section exam.

		Frozen-section (Group A)	Non-frozen-section (Group B)
Number of patients		n = 179	n = 63
Mean age (years)		58.14	55.59
Tumor size (pT)	pT0 - pTis	2.3%	2.1%
	pT1	63.4%	59.5%
	pT2	30.9%	27.6%
	pT3	0.6%	0%
	pT4	2.9%	10.6%
Lymph node stage (pN)	pN0	69%	65.9%
	pN1	23.4%	25%
	pN2	5.8%	2.2%
	pN3	1.8%	6.8%
Ki-67	< 25%	62.5%	65.7%
	> 25%	37.5%	34.3%
ALI	Absent	67.7%	77.4%
	Present	32.3%	22.6%
EIC	Absent	40.5%	41.4%
	Present	59.5%	58.6%
Surgical cavity shaving?	Yes	59.8%	50%
	No	40.2%	50%
Estrogen and progesterone receptors	Positive	88.2%	88.8%
	Negative	11.8%	11.2%
	Luminal	72.2%	55.3%
Immunohistochemical subtypes	Her-2	19.5%	19.1%
	Triple negative (ER, PR and Her-2)	8.3%	6.3%

EIC = Extensive intraductal component; ALI = Angiolymphatic invasion; ER = estrogen receptor; PR: progesterone receptor.

Table 2. — Analysis of the influence of variables of surgical margins in patients from group A subjected to the frozen-section exam.

Variable		Positive margins (n = 59)	Free margins (n = 120)	p
Age	< 50 years	21.74%	26.97%	0.59
	> 50 years	78.26%	73.03%	
Immunohistochemical subtype	Luminal	58.82%	73.91%	0.41
	Her-2	29.41%	18.26%	
	Triple negative	11.76%	7.83%	
Ki-67	< 25%	66.67%	61.45%	1.00
	> 25%	33.33%	38.55%	
IAL	Absent	66.67%	67.63%	0.94
	Present	33.33%	32.37%	
Surgical cavity shaving?	No	34.78%	41.29%	0.55
	Yes	65.22%	58.71%	
Size	< 1 cm	23.81%	27.45%	0.73
	> 1 cm	76.19%	72.55%	
Tumor stage (pT)	pT 0-1	76.19%	64.71%	0.29
	pT 2-3-4	23.81%	35.29%	
Positive lymph nodes/ Resected lymph nodes	0	61.9%	69.33%	0.67
	< 10%	14.29%	10%	
	> 10%	23.81%	20.67%	
Lymph node staging (pN)	pN0	60%	70%	0.36
	pN+	40%	30%	
EIC	Absent	25%	41.98%	0.19
	Present	75%	58.02%	

The chi-square test was employed to compare variables.

## Discussion

The intraoperative analysis of surgical margins is a crucial stage in the conservative treatment of breast cancer, for it allows the detection of compromised margins at the time of surgery itself, thus rendering further interventions unnecessary [25] and keeping costs and morbidity from rising. The present study makes a contribution as it analyzed only IDC cases (except by lobular histology) and compared the group subjected with the other study patients who were also operated, but did not undergo such a procedure.

There is a great deal of controversy in the literature respecting the width of the ideal margin [26]. The studies from major world centers consider a margin to be free when there are no tumor cells in the resection margin regardless of the distance of the tumor [3, 4]. However, for other authors, a margin is free when it is 1-10 mm away from the tumor [9, 27]. In this study, a safety (free) margin was defined as a two mm margin in accordance with Pinotti and Carvalho in 2002 [17].

The intraoperative exam with cryoablated margins is criticized by some authors for increasing the costs and the complexity of the intraoperative exam without improving the end results [12, 16, 28]. However, there are other authors who claim such a procedure allows for greater diagnostic precision. The results in the present study, though, do not bear out their allegation.

The medical literature has shown that the intraoperative investigation of the margins effectively lowers the reoperation rate [1]. However, the frozen-section exam has produced less emphatic results. Olson *et al.* in 2007, estimated that it raises the cost and lengthens the time of





surgery considerably. Besides, there is a hypothetical risk of loss of material during the exam [28, 29].

Unlike Weber *et al.* in 2008 [30], who found that was superior to the other procedures, the present study showed that the frozen-section exam did not make a significant difference. However, the number of women in this study was greater than that in the study of the aforementioned authors. Also, all of the participants were operated and evaluated by the same surgery and pathology teams. One should keep in mind, though, that this study was retrospective. Ideally, a prospective and randomized study should be conducted to reach definitive conclusions as to the value of intraoperative diagnosis.

In contrast to a study published by Riedl *et al.* in 2009 [15] who also analyzed variables that might influence the frozen-section exam, this study did not find it difficult to evaluate the margins of smaller tumors. However, this study did not make a preoperative distinction between tumors previously subjected to chemotherapy and those without a previous diagnosis.

The results obtained by Moore *et al.* in 2007, were dissimilar to those included in that in this study did not find predictors pointing to higher failure rates in the frozen-section exam. Moreover, the cases of lobular histology were previously excluded from analysis in this study [31].

Contrary to other studies [21], the authors did not observe important differences among the intraoperative techniques that were analyzed. In fact, the results suggest that the intraoperative evaluation of cryoablated surgical margins does not improve the intraoperative diagnosis.

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