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Safety and technical success of methylene blue dye for lymphatic mapping in breast cancer

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Abstract

BACKGROUND: We aimed to evaluate the safety and efficacy of methylene blue dye (MBD) for sentinel lymph node (SLN) mapping in breast cancer.

METHODS: A total of 398 patients undergoing 401 SLN biopsy procedures with MBD were retrospectively reviewed and divided into 3 groups based on dye concentration and location of dye injection. Technical success and complication rates were compared.

RESULTS: Overall technical success of SLN biopsy was 99.7%. Technical success of SLN mapping with MBD was lowest in the group receiving full strength dye in a peritumoral location (74%) and highest in the group that received very dilute MBD (1.25 mg/mL) in the subareolar location (92%, $P = .004$). The complication rate was highest in the first group (21%) and lowest in the latter (2%, $P = .0003$).

CONCLUSIONS: Dilute MBD (1.25 mg/mL) successfully maps SLNs with very few complications. © 2008 Elsevier Inc. All rights reserved.

Sentinel lymph node (SLN) biopsy has become an established standard of care for staging clinically node-negative patients with breast cancer. The American Society of Breast Surgeons recommends using both blue dye and radioisotope in their consensus statement of December 2005.¹ A dual-tracer approach for SLN biopsy affords the highest technical success and lowest false negative rates.^{2–5} The dye commonly chosen for lymphatic mapping is isosulfan blue dye (IBD; Lymphazurin; United States Surgical, North Haven, CT, USA). Unfortunately, IBD is associated with allergic reactions in up to 3% of patients^{6–10} and can be life-threatening in .1% to 1% of patients.^{6–9,11} In addition, chronic blue staining is reported in up to 41% of patients.¹²

A national shortage of IBD led to a search for alternative dyes for SLN mapping in breast cancer. Methylene blue dye (MBD) is one such alternative, with success rates reported

in several series.^{13–19} These reports on MBD use did not provide information on safety and complications related to use of the dye, despite a publication indicating risk of tissue necrosis after MBD injection in the breast. The objective of our study was to determine the safety profile and technical success of MBD for lymphatic mapping in breast cancer. We also aimed to identify factors related to technical success and morbidity of SLN mapping with MBD.

Materials and Methods

An institutional review board–approved retrospective chart review was performed on breast cancer patients who underwent SLN biopsy with MBD by a single surgeon, from August 2003 to June 2006. All patients received preoperative intradermal radioisotope injection. All patients received local anesthetic at the planned incision site except those with documented allergy. Operative technique, other than

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Table 1 Concentrations of MBD used for SLN mapping

Concentration	mg/mL	n	%
Full strength	10	96	24
Diluted		305	
1:1	5	48	12
1:2	3.3	49	12
1:3	2.5	91	23
1:7	1.25	103	26
Diluted, concentration unknown		14	3

specified details of dye injection, was the same for the entire study population as all procedures were performed by a single surgeon.

Definitions

Overall technical success was defined as identification of any SLN—either blue or radioactive or clinically suspicious. Technical success of blue dye refers to identification of SLN(s) demonstrating dye uptake in the node or its afferent lymphatic channel. Volume of MBD is noted in milliliters, concentration in milligrams per milliliter, and site of injection of blue dye refers to the anatomic location of MBD injection for SLN mapping. Complications (either local or systemic) occurring within 30 days of the SLN biopsy procedure were recorded.

Groups for analysis

The MBD technique for SLN mapping evolved over time, with some aspects of technique related to dilution and injection location changing simultaneously. Patients were therefore grouped based on their combination of dye concentration and the location of injection of MBD for the purposes of analysis. For 20 of 401 cases, dye concentration and/or injection location were unspecified. The remaining 381 cases were divided into 4 groups. Group A ($n = 43$) spanned August 2003 to April 2004 and received full-strength MBD injected in a peritumoral location. Group C ($n = 102$) consisted of patients treated from August 2005 to June 2006 who were injected with very dilute dye (1.25 mg/mL) in the subareolar location. Group B spanned August 2003 to April 2006 and comprised the remaining 236 individuals who had intermediate dilutions of MBD; almost all (97%) were injected in the subareolar location.

Data collection and statistical analysis

Multiple data points were collected, including clinico-pathologic factors, volume and concentration of MBD used, site of injection of MBD, dye uptake in SLNs, and complications. Mapping success and frequency of complications were determined. Technical success and complication rates

among the 3 groups were compared using chi-square or Fisher exact tests, as appropriate. Data are presented as median (range) or frequency (percentage). A 2-sided P value of less than .05 was considered statistically significant. Statistical analysis was performed with JMP statistical software 6.0.0 (SAS Institute Inc, Cary, NC).

Results

During the study period, 398 patients underwent 401 SLN biopsy procedures using MBD and radioisotope. Median age was 61 years (range 27–90) and median tumor size was 1.4 cm (range .1–14 cm). The volume of MBD injected was less than 3 mL in 42 of 401 cases (10%), between 3 and 4 mL in 341 (85%), and unknown in 18 (4%). Full-strength dye (10 mg/mL) was injected in the first 96 of 401 (24%) cases, and the remaining 305 (76%) received increasing dilutions over the study period (Table 1). Overall, the following dye dilutions were used: 5 mg/mL (1:1 dilution) in 48 of 401 patients (12%), 3.3 mg/mL (1:2 dilution) in 49 (12%), 2.5 mg/mL (1:3 dilution) in 91 (23%), 1.25 mg/mL (1:7 dilution) in 103 (26%), and diluted of unknown concentration in 14 (3%) cases. Site of injection of blue dye was subareolar in 336 of 401 (84%) cases, peritumoral in 50 (12%), and unknown in 15 (4%).

Technical success

Overall technical success of SLN biopsy using both MBD and radioisotope for mapping was 99.7%. The identification rate of any blue SLN with MBD was 88% for the entire study period, increasing over time from 81% in 2003 to 94% by 2006, the last 6 months of the study period (Fig. 1), during which 63 of 66 (95%) cases the dilution and injection route of group C. The observed improvement in technical success with the use of MBD was statistically significant ($P = .03$) when comparing 2003 to 2006. Tech-

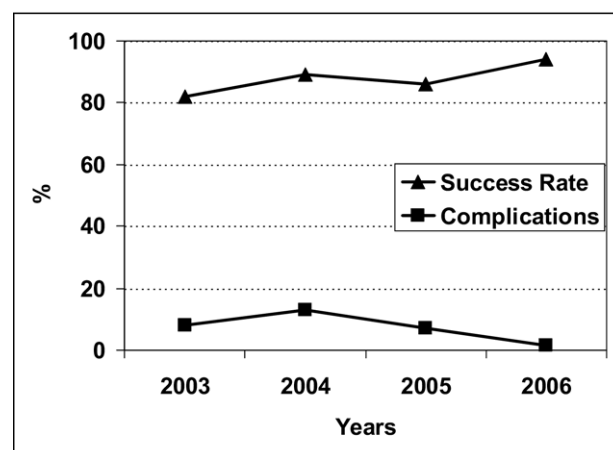


Figure 1 Technical success of mapping with MBD and complication rate of SLN mapping with MBD over time.

Table 2 Technical success of SLN mapping with MBD among the study groups

	Dye concentration	Injection location	n	%	Technical success	
					n	%
Group A	Full strength (10 mg/mL)	PT	43	11	32	74
Group B	Intermediate	SA	236	59	208	88
Group C	Most dilute (1.25 mg/mL)	SA	102	25	94	92*
Unknown			20	5	17	85

PT = peritumoral; SA = subareolar.
*P = .004 compared to group A.

nical success of MBD mapping also varied among the 3 patient groups based on dye concentration and injection location (Table 2). A blue node was identified in 74% of group A patients, 88% of group B, and 92% of group C. A statistically significant difference in mapping success with MBD was observed between patients in group A compared to group C ($P = .004$).

Complication rate

Information on complications was available in 399 cases (Table 3). Median follow-up for the study population was 27 days (range 1–421), with at least 14 days of follow-up for 314 of 401 patients (78%). Of 399 cases with information on complications, there were no cases of anaphylaxis. Immediate hypersensitivity reactions in the form of wheal and flare response occurred in two of 399 patients (.5%) and resolved with antihistamines in both. In terms of local complications to MBD injection, a local inflammatory reaction was the most common problem observed, seen in 21 of 399 cases (5%). These local inflammatory reactions were characterized by focal erythema and/or palpable induration at the injection site. Most were apparent several days postoperatively and resolved over several weeks’ time. Superficial tissue toxicity manifest as skin necrosis occurred in 5 of 399

patients (1.25%). One of these patients received 3 mL of half-strength dye (5 mg/mL) and developed full-thickness skin necrosis that required excision. In the other 4 cases, areas of skin necrosis were less than 2 mm, superficial, and healed without intervention. Blue staining was observed in 5 cases (1.25%); this resolved within 3 weeks in 4 cases and further follow-up was not available for the other patient. The complication rates of lymphatic mapping with MBD differed among the 3 study groups at 21% in group A, 9% in group B, and 2% in group C (Table 3). A statistically significant difference in complication rates was confirmed between groups A and C ($P = .0003$) and between groups B and C ($P = .02$). The lower complication rate in group C appears mostly attributable to a reduction in the frequency of local inflammatory reactions. The 1 other complication in group C consisted of focal (<2 mm) necrosis of the lumpectomy incision edge that was self-limited and possibly unrelated to the dye injection. The complication rate also varied over time (8% overall but only 1.5% by the last 6 months of the study period) (Fig. 1), again consistent with the fact that group C technique was applied to the vast majority of cases in the final 6 months.

Comments

SLN biopsy is a well-established, minimally invasive method of axillary staging in clinically node-negative patients with breast cancer. Current literature supports the use of a dual-tracer technique for lymphatic mapping, using both radioisotope and blue dye, as this results in the highest identification rates and lowest false-negative rates for the procedure.^{2–5} The most commonly used tracer agents are technetium-99 sulfur colloid and 1% IBD. Mapping success rates using a dual-tracer technique that incorporates IBD are 90% to 98%.^{2,17,20,21} The published success rates for mapping with IBD alone are slightly lower—between 83% and 93%^{2,17,20,22} (Table 4). Despite an established track record of mapping success with IBD, its safety record is not optimal. Allergic or adverse reactions to IBD have been reported in .1 to 3% of patients undergoing SLN biopsy.^{6–11} Three

Table 3 Complication rates of SLN mapping with MBD among the study groups

	N	Complication info unknown	Any complication	Hypersensitivity reactions		Local reactions		
				Anaphylaxis	Wheal and flare	Local inflammation	Skin necrosis	Blue staining
Group A	43	1	9 (21%)	0	0	7	1	1
Group B	236	1	21 (9%)	0	2	12	3	4
Group C	102	0	2 (2%)*	0	0	1	1	0
Group unknown	20	0	1 (5%)	0	0	1	0	0
Total	401	2	33 (8%)	0	2 (.5%)	21 (5%)	5 (1%)	5 (1%)

*P = .0003 compared to group A, P = .02 compared to group B.

Table 4 Technical success of SLN mapping with IBD and MBD

	Year	n	Dye alone success rate (%)	Dual technique success rate (%)
IBD				
Guiliano ¹²	1997	107	93	NA
McMasters ²⁰	2000	244	86	90
Cody ²	2001	2000	83	97
Grant ²¹	2003	675	NR	98
Vargas ¹⁷	2004	83	88	98
MBD				
Simmons ¹³	2001	30	90	NR
Blessing ¹⁴	2002	112	NR	99
Simmons ¹⁵	2003	112	92	96
Nour ¹⁶	2004	54	83	NR
Vargas ¹⁷	2004	81	93	98
Golshan ¹⁸	2006	141	96.5	NA
Degnim ⁵	2007	398	88	99.7

NA = not applicable; NR = not reported.

distinct patterns of allergic reactions to isosulfan blue dye are described: grade 1 (urticaria or blue hives, pruritus, or a generalized rash), grade 2 (hypotension not requiring vasopressors), or grade 3 (hypotension requiring pressor support).²³ Life-threatening anaphylaxis with cardiovascular collapse is rare but is reported in up to 1% of patients receiving IBD.¹⁰ In order to reduce the risk of anaphylaxis associated with use of IBD, corticosteroids may be administered prior to injecting IBD. However, this approach results in an increase in surgical site infections and possible delays for adjuvant treatment.¹⁰ Reducing the volume of IBD decreases the severity but not the frequency of allergic reactions.²⁴ Chronic blue staining is also a common occurrence after IBD injection, reported in up to 41% of patients 12 months after the SLN surgery.¹² In addition, a false desaturation in the pulse oximetric reading mimicking true hypoxia may occur with IBD.²⁵

Because of a national shortage of IBD in the late 1990s, MBD was reported as an alternative dye for lymphatic mapping in breast cancer with a success rate of 90%.¹³ Other reports followed on the use of MBD for SLN mapping with overall identification rates of up to 99% with a dual technique and dye-alone success rates of 83% to 93%^{13–19} (Table 4). Although MBD appears to be equally effective and is less costly than IBD, it has not gained widespread use. Potential reasons are the lack of published experience with this dye in large cohorts as well as concern for local tissue necrosis. One of the first reports on MBD for SLN mapping described technical success but did not provide information on complication rates.¹³ However, a follow-up report by the same group noted no cases of anaphylaxis or allergic reactions in the 112 patients included in the study. Two other studies of 54¹⁶ and 173 patients¹⁹ each provided excellent identification rates with MBD but did not review the safety profile of MBD. Other similar stud-

ies^{14,17,18} of 141, 112, and 81 patients each reported no allergic reactions or anaphylaxis with MBD but did not address local complications associated with the use of this dye.

Development of adverse skin lesions in 5 of 24 patients (21%) after MBD injection for SLN biopsy was first reported by Stradling et al in 2002.²⁶ Patients received 3 to 5 mL of full-strength (1% = 10 mg/mL) MBD injected into both the parenchyma and the skin. Five patients developed a variety of local inflammatory presentations including intense erythematous macular lesions, superficial ulcers, and necrotic ulceration at the injection site. The skin lesions of all 5 patients improved with topical silver sulfadiazine cream application; none required surgical debridement. The authors recommended avoiding intradermal injections and favored the use of deep parenchymal injection of MBD for lymphatic mapping. Although deep parenchymal injection may reduce superficial tissue necrosis, it is not the preferred location to optimize the technical success of lymphatic mapping (which seems to be best when tracers are injected superficially).²⁷ A recent single case report of skin and fat necrosis of the breast following MBD injection did not state the size of necrotic area, and no specifics on the volume, concentration, or site of injection of MBD were mentioned in the report.²⁸

MBD is a cationic thiazine dye with characteristic deep blue color in its oxidized state. Several properties of this dye are responsible for its toxic effects on tissues. Once in contact with tissues, MBD forms oxidation products and free radicals that are toxic to tissues and may cause a local inflammatory lesion.²⁹ MBD may also cause local tissue ischemia via its vasoconstrictive effects.³⁰ Injection of this dye in the colon has also caused local toxicity, including submucosal ulceration and necrosis.³¹ Despite its potential local toxicity, anaphylaxis is a very rare event after MBD administration, with only a single reported case we could identify in the literature.³² In fact, because of its strong vasoconstrictive properties, MBD has been proposed as a lifesaving drug to restore arterial blood pressure after contrast medium-induced anaphylaxis.³³ We did not observe any cases of anaphylaxis in patients given MBD. The immediate hypersensitivity reactions we observed manifested as wheal and flare reactions at the injection site and resolved with antihistamines. In addition to its low risk of allergic reactions, MBD interferes much less than IBD with the peripheral monitoring of oxygen saturation.²⁵

In our early experience with MBD for lymphatic mapping, local complications at the injection site were concerning. In addition to the single patient who developed frank tissue necrosis, several individuals also experienced painful local inflammatory reactions at the dye injection site. Furthermore, residual palpable areas of induration created concern among both patients and other providers involved in their care. This experience, along with published reports on optimal injection site²⁷ and expert recommendation to use dilute MBD,⁵ prompted us to modify our mapping tech-

nique in order to reduce the risk of adverse effects while maintaining technical success. Therefore, we shifted to a subareolar injection site and proceeded to use increasing dilutions of MBD for lymphatic mapping. We found that at a concentration of 1.25 mg/mL, the MBD was still easily visible in lymphatics and sentinel nodes. Our clinical impression was that dilute dye results in fewer complications without reducing mapping success, and we reviewed our patient outcomes to determine if our clinical impressions were accurate.

Our study suggests that MBD performs as well as IBD for lymphatic mapping in breast cancer. Furthermore, when used at a dilute concentration, we observed very little tissue toxicity directly attributable to the dye. Over time and with evolving technique, technical success increased and the complication rate decreased. The retrospective nature of this study does present certain limitations regarding our findings and the possible conclusions. Since more than one aspect of the mapping technique changed simultaneously, it is not possible to determine the relative impact of the various factors (as well as accumulated surgical experience) on technical success. However, other groups have published retrospective results on sentinel node mapping techniques where changes in technique occurred over time.² As a result of this limitation of our study, we cannot be certain which aspect of technique accounts for the lower complication rate in the most recent patients. However, it is logical that the dilution of the MBD is most likely responsible for the decreased complication rate seen, especially since the lowest complication rate occurred in the group that had MBD dye injected superficially, where MBD injection is known to cause skin necrosis.²⁶ Diluting the dye appears to reduce local complications, likely via less chemical toxicity to the local tissues and less local ischemia.

Another limitation of retrospective review is possible underestimation of the complication rates due to inadequate documentation in the medical record. Although underestimation is a weakness of this study due to its retrospective nature, underestimation of complications should have been uniform across the study groups. Of note, median follow-up for the study population was 27 days, and most wound complications occur within this time period. Overall, complications are uncommon after use of blue dyes for lymphatic mapping, and therefore the sample size required to evaluate significant differences in a prospective trial would be quite large. Until then, this study provides useful information regarding the use of MBD for lymphatic mapping and its safety profile.

We conclude that MBD demonstrates technical mapping success similar to published success rates for IBD. Anaphylaxis is exceedingly rare after MBD injection and was not observed in any patient in this study. Furthermore, when MBD is sufficiently diluted, the rate of local complications is very low. We believe that dilute MBD is the preferred dye for lymphatic mapping in breast cancer due to its comparable technical success and its low risk profile.

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