

Research Article**EFFICACY OF METHYLENE BLUE DYE FOR INTRAOPERATIVE IDENTIFICATION OF PARATHYROID GLANDS AND RECURRENT LARYNGEAL NERVE DURING THYROIDECTOMY: A PROSPECTIVE STUDY****Dr Chaitra D Y^{1*}, Dr Subhash Nadagouda², Dr Nishan Bhoopathi Shetty³**¹Fellow, Department of Surgical Gastroenterology, IGOT Hospital, Bengaluru.²Assistant Professor, Dept of General Surgery, YIMS Yadgiri.³Senior Resident, Dept of Paediatric Surgery, IGICH Bengaluru.

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Abstract

Background: Thyroidectomy carries risks of injury to parathyroid glands and the recurrent laryngeal nerve (RLN). Methylene blue dye has emerged as a potential aid for the intraoperative identification of these structures.

Aim: To evaluate the efficacy of methylene blue dye for intraoperative identification of parathyroid glands and the RLN during thyroidectomy.

Methods: A prospective study was conducted on 34 patients undergoing thyroidectomy. Methylene blue dye was applied intraoperatively, and identification of the parathyroid glands and RLN was recorded. Postoperative outcomes, including hypocalcemia and RLN injury, were assessed.

Results: Methylene blue successfully identified parathyroid glands in 64.7% of cases, with a sensitivity of 74.1% and a specificity of 71.4%. The positive predictive value was 90.9% and the negative predictive

value was 41.7%. No cases of postoperative RLN injury were observed. Transient hypocalcemia occurred in 23.5% of patients. The most common final diagnosis was multinodular goitre (61.8%), followed by papillary carcinoma (23.5%).

Conclusion: Methylene blue dye shows promise as an adjunct for identifying parathyroid glands during thyroidectomy, with a high positive predictive value. The absence of RLN injuries is encouraging, although its relationship to dye use requires further investigation. While it did not eliminate postoperative hypocalcemia, the technique may contribute to improved surgical outcomes. Larger randomized controlled trials are needed to fully establish the role of methylene blue in thyroid surgery.

Keywords: Thyroidectomy, Methylene blue, Parathyroid glands, Recurrent laryngeal nerve, Surgical complications, Hypocalcemia

INTRODUCTION

Thyroid disorders represent a significant and growing health concern in India, second only to diabetes mellitus in the spectrum of endocrine diseases¹. This increasing disease burden is attributed to various factors, including improved diagnostic capabilities, increased awareness, and environmental influences. Approximately 42 million people in India are affected by thyroid disorders, underscoring the critical importance of effective management strategies². As the prevalence of thyroid disorders continues to rise, so does the need for effective management strategies, with thyroidectomy remaining the surgical treatment of choice for thyroid neoplasms and many benign thyroid conditions³.

While a standard procedure, thyroidectomy carries inherent risks that can significantly impact patient outcomes. The two most critical and potentially debilitating complications following this procedure are hypocalcemia and recurrent laryngeal nerve (RLN) injury. These complications not only affect the immediate postoperative course but can also have long-lasting effects on patients' quality of life^{4,5}.

Post-thyroidectomy hypocalcemia, resulting from inadvertent damage to the parathyroid glands or their blood supply, is a frequent complication, with reported incidence rates ranging from 1.6% to 50%⁶. Its manifestations range from mild symptoms, such as perioral numbness and paresthesias of the fingertips, to more severe presentations, including muscle spasms, tetany, seizures, and cardiac arrhythmias. The severity and duration of hypocalcemia can vary, with some patients requiring

lifelong calcium and vitamin D supplementation. These complications affect patients' physical and psychological well-being and prolong hospital stays, increasing healthcare costs and resource utilization⁷.

The recurrent laryngeal nerve, crucial for vocal cord function, is another structure at risk during thyroidectomy. RLN injury incidence ranges from 0.4% to 7.2% for temporary paresis and 0% to 5.2% for permanent paralysis. Injury to this nerve can result in voice changes, breathing difficulties, and, in severe cases, airway compromise. Bilateral RLN injury, although rare, can be life-threatening and may necessitate tracheostomy. The potential for these complications emphasizes the critical need for precise intraoperative identification and preservation of the parathyroid glands and the RLN⁸.

Traditionally, surgeons have relied on anatomical landmarks and visual inspection to identify these structures during thyroidectomy. However, the small size of the parathyroid glands (typically 3-7 mm in diameter) and their similarity to surrounding fat and lymph nodes make this approach challenging^{9,10}. The parathyroid glands' colour can vary from yellow-brown to reddish-brown, further complicating visual identification. Similarly, anatomical variations and alterations due to pathological conditions can complicate identification of the RLN based on its conventional relationships with the inferior thyroid artery, Berry's ligament, and the tracheoesophageal groove¹¹.

Methylene blue dye (methyl-thioninium chloride) has emerged as a promising approach. This thiazine dye, discovered by

Heinrich Caro in 1876, has a long history of medical applications, including its use as an antidote for certain types of poisoning and as a diagnostic aid in various surgical procedures¹². It is safe, readily available, and has shown potential to enhance parathyroid gland visibility during surgery. A previous case series reported that when sprayed on the surgical field, methylene blue was absorbed more rapidly by the parathyroid glands than by the surrounding perithyroidal tissues, attributed to the glands' rich lymphovascular supply⁹.

The mechanism of action of methylene blue in this context is believed to be related to its rapid uptake by tissues with high metabolic activity. Despite their small size, the parathyroid glands have a rich blood supply and high metabolic rate, which may explain their preferential dye uptake. This property could provide surgeons with a simple, effective tool for intraoperative identification of these critical structures if consistently demonstrable¹³.

The present study aims to evaluate the efficacy of methylene blue spray in the intraoperative identification of both the parathyroid glands and the recurrent laryngeal nerve during thyroidectomy, with the objectives of (1) evaluating methylene blue spray for identifying the parathyroid gland and RLN intraoperatively during thyroidectomy, and (2) minimizing postoperative complications from inadvertent injury to the parathyroid gland and RLN.

MATERIALS AND METHODS

This prospective study was conducted at M.S. Ramaiah Hospitals, Bengaluru,

between August 2022 and November 2023, on patients undergoing thyroidectomy.

Inclusion criteria

- Patients aged 18-80 years
- Patients who provided written informed consent
- Patients with thyroid swelling requiring thyroidectomy

Exclusion criteria

- Patients who had undergone previous thyroid surgery
- Pregnancy
- Patients with pre-operative unilateral or bilateral vocal cord palsy

Surgical technique

Eligible patients presenting to M.S. Ramaiah Hospitals, Bengaluru were enrolled after obtaining written informed consent. All participants underwent open thyroidectomy under general anaesthesia. Recurrent laryngeal nerve function, thyroid hormone levels, and calcium levels were evaluated pre- and post-operatively.

During thyroidectomy, after ligation of the superior pole and before ligation of the inferior pole, the thyroid lobe was deviated medially and 0.5 mL of methylene blue diluted with 5 mL normal saline was sprayed over the thyroid lobe and perilobar area, where the recurrent laryngeal nerve and parathyroid gland are expected to be present. The recurrent laryngeal nerve was expected not to take up the dye and hence to appear as a pale white structure, whereas the surrounding tissues stained blue, permitting its intraoperative identification. The parathyroid gland was expected to absorb the dye and regain its original yellow colour within five minutes, while the surrounding tissue remained stained.

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Five minutes after methylene blue spray, an incisional biopsy sample of 2 mm diameter was taken from the tissue suspected to be the parathyroid gland, identified by its pale yellow colour relative to the surrounding methylene-blue-stained tissue. This sample was sent for histopathological examination, and patients received standard postoperative care.

Sample size

In a study on the sensitivity and specificity of methylene blue spray for identifying the parathyroid gland during thyroidectomy, the sensitivity was reported as 92.31% and specificity as 56.79%, with a PPV of 25.53% and NPV of 97.87%¹⁵. Expecting a similar result, and to achieve a precision of 95% confidence level and 10% absolute precision, the study required a minimum of 34 subjects.

Statistical analysis

Data were entered into Microsoft Excel 2019 and analysed using SPSS version 26. Descriptive statistics (frequencies and percentages) were computed for demographic and clinical characteristics, including age distribution, gender, comorbidities, final diagnosis, parathyroid gland detection, and postoperative outcomes. The diagnostic performance of methylene blue in detecting the parathyroid gland was assessed by calculating sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) from the cross-tabulation of detected versus observed parathyroid glands. A p-value <0.05 was considered statistically significant.

RESULTS

Table 1. Age distribution of patients undergoing thyroidectomy

Age (years)	N	%
25-30	1	2.9%
31-40	9	26.5%
41-50	10	29.4%
51-60	9	26.5%
61-70	4	11.8%
71-80	1	2.9%

The highest proportion of patients was in the 41-50-year age group (29.4%), followed by the 31-40 and 51-60-year age groups (26.5% each). Patients aged 61-70 years accounted for 11.8%, while the 25-30 and 71-80-year age groups each comprised 2.9%.

Table 2. Gender distribution of patients undergoing thyroidectomy

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Gender	N	%
Male	13	38.2%
Female	21	61.8%

A higher proportion of patients were female (61.8%), while males comprised 38.2% of the study population.

Table 3. Distribution of comorbidities among patients undergoing thyroidectomy

Comorbidities	N	%
Hypertension	5	14.7%
Type 2 diabetes mellitus	8	23.5%
IHD + Hypertension	2	5.9%
None	19	55.9%

A majority of patients (55.9%) had no comorbid conditions. Among those with comorbidities, 23.5% had type 2 diabetes mellitus, 14.7% had hypertension, and 5.9% had both ischemic heart disease and hypertension.

Table 4. Detection of parathyroid gland using methylene blue

Detected using Methylene Blue	N	%
Yes	22	64.7%
No	12	35.3%

The parathyroid gland was successfully identified using methylene blue in 64.7% of cases, and was not detected in 35.3% of cases.

Table 5. Parathyroid gland observed during thyroidectomy

Observed Parathyroid Gland	N	%
Yes	27	79.4%
No	7	20.6%

The parathyroid gland was visually confirmed intraoperatively in 79.4% of cases and was not observed in 20.6% of cases.

Table 6. Final diagnosis of patients undergoing thyroidectomy

Final Diagnosis	N	%
Multinodular goitre	21	61.8%
Follicular adenoma	4	11.8%
Papillary carcinoma	8	23.5%
Adenomatous hyperplasia	1	2.9%

Multinodular goitre was the most common diagnosis (61.8%), followed by papillary carcinoma (23.5%), follicular adenoma (11.8%), and adenomatous hyperplasia (2.9%).

Table 7. Post-operative transient hypocalcaemia

Post-op Transient Hypocalcaemia	N	%
Yes	8	23.5%
No	26	76.5%

Transient hypocalcaemia occurred in 23.5% of patients following thyroidectomy.

Table 8. Post-operative signs of recurrent laryngeal nerve injury

Post-op Signs of RLN Injury	N	%
Absent	34	100.0%

No recurrent laryngeal nerve injuries were observed in the study population.

Table 9. Diagnostic performance of methylene blue for detection of the parathyroid gland

Detected using Methylene Blue	Observed PT gland - Yes	Observed PT gland - No	Total
Yes (N, % within observed)	20 (74.1%)	2 (28.6%)	22 (64.7%)
No (N, % within observed)	7 (25.9%)	5 (71.4%)	12 (35.3%)
Total	27 (100.0%)	7 (100.0%)	34 (100.0%)

True positives = 20, false negatives = 7, true negatives = 5, false positives = 2. Sensitivity = $20/27 = 74.1\%$; Specificity = $5/7 = 71.4\%$; Positive predictive value = $20/22 = 90.9\%$; Negative predictive value = $5/12 = 41.7\%$. The high positive predictive value indicates that when methylene blue identified a structure as the parathyroid gland, this was very likely to be correct. The comparatively low negative predictive value indicates that absence of staining could not reliably exclude the presence of a parathyroid gland.

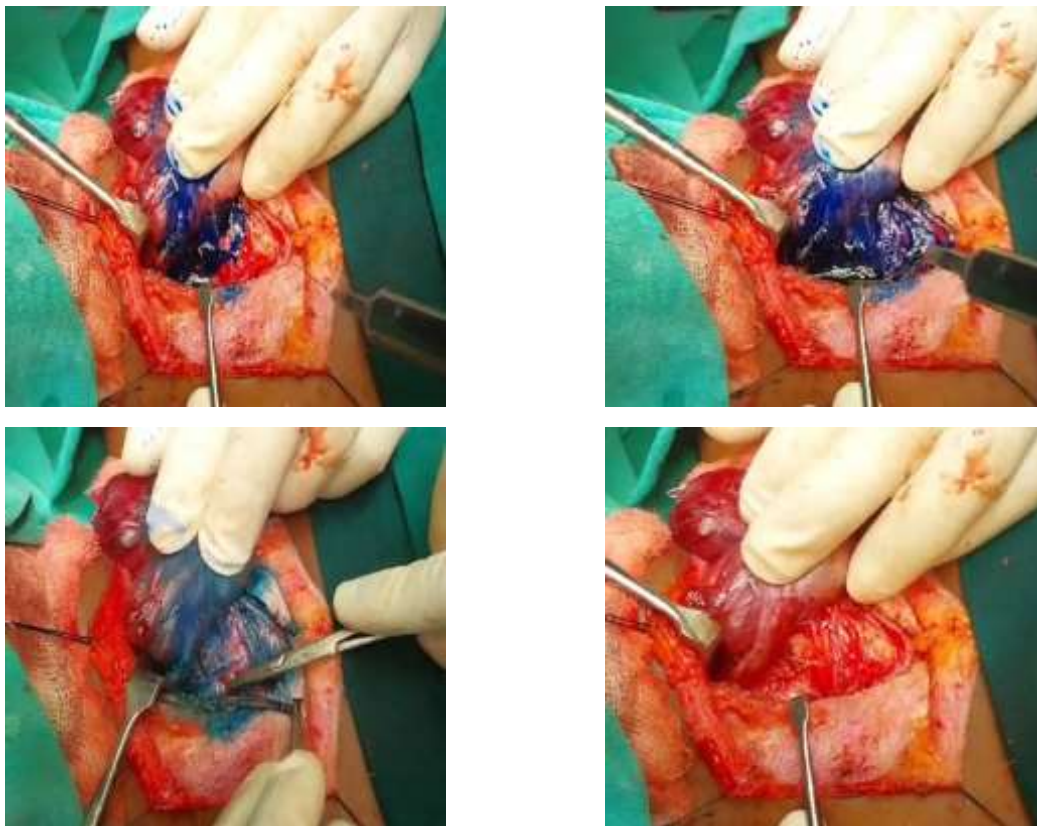


Figure 1. Intraoperative photographs showing methylene blue staining of the perithyroidal field, with unstained (pale) parathyroid tissue and the recurrent laryngeal nerve distinguishable from the surrounding stained tissue.

DISCUSSION

Utilizing methylene blue dye during thyroidectomy has been proposed as a means of improving surgical accuracy and minimizing complications by facilitating detection of the parathyroid glands and recurrent laryngeal nerve. This study assessed the effectiveness of this approach in 34 patients undergoing thyroidectomy.

Most study participants were middle-aged, with the largest proportion in the 41-50-year age range. This age distribution corresponds to the usual occurrence of thyroid disease, as documented by Menon et al.² and supported

by Leso et al.³ The gender distribution showed a higher proportion of females, consistent with the greater occurrence of thyroid disease in women reported in numerous epidemiological studies.

A substantial proportion of patients had comorbidities, with type 2 diabetes mellitus being the most common. This is notable given the established association between thyroid dysfunction and diabetes mellitus described by Biondi et al.¹ Bansal et al.⁴ stressed the importance of considering comorbidities in perioperative management, since they can affect surgical outcomes.

Methylene blue identified the parathyroid gland in approximately 66% of cases in our study. This detection rate, although encouraging, is lower than the 100% identification rate reported by Sari et al.¹⁴ using intraoperative methylene blue spraying, but aligns closely with the 78.7% sensitivity reported by Piromchai et al.¹⁵.

In our study, the sensitivity and specificity of methylene blue for detecting the parathyroid gland were approximately 74% and 71% respectively. The high positive predictive value suggests that when methylene blue identified a structure as a parathyroid gland, it was very likely to be correct; however, the lower negative predictive value indicates that absence of staining could not reliably exclude the presence of a parathyroid gland. These results are similar to findings reported by Orban et al.¹⁶ and Kamal et al.¹⁷, who also concluded that methylene blue is a valuable tool for identifying the parathyroid gland. Nevertheless, our findings highlight its limitations: given the reduced negative predictive value, surgeons should not rely on methylene blue staining alone and should employ additional identification techniques, as underlined by Stefanou et al.¹⁸.

No postoperative recurrent laryngeal nerve injury was observed in our study. This favourable result compares well with the RLN injury rates reported by Zakaria et al.⁵ and Alqahtani et al.⁸ Although our study does not provide precise data on RLN identification using methylene blue, the absence of injury suggests that the surgical technique, which included methylene blue use, successfully preserved RLN function. This is particularly relevant given that RLN injury is one of the most severe consequences

of thyroid surgery, as emphasized by Culp and Patel¹⁹. Our findings are consistent with those of Orban et al.¹⁶ and Kamal et al.¹⁷, who reported enhanced identification and preservation of the RLN with the use of methylene blue.

The occurrence of transient postoperative hypocalcaemia in our study is consistent with the findings of Alqahtani et al.⁶ and Privitera et al.²⁰ These findings indicate that although methylene blue may assist in identifying the parathyroid gland, it does not entirely prevent postoperative hypocalcaemia, underscoring the need for meticulous parathyroid preservation and careful postoperative calcium management, as highlighted by Schafer and Shoback⁷.

The distribution of final diagnoses in our study, with multinodular goitre being most frequent, followed by papillary carcinoma, aligns with the patterns of thyroid disease described by Allen and Fingeret²¹ and corroborated by Efremidou et al.²² The high prevalence of malignancy (approximately 25% of cases) underscores the importance of precise surgical technique and comprehensive intraoperative assessment during thyroidectomy.

CONCLUSION

This study assessed the efficacy of methylene blue dye for intraoperative identification of the parathyroid gland and recurrent laryngeal nerve during thyroidectomy. Methylene blue demonstrated a sensitivity of 74.1% and specificity of 71.4% for identifying the parathyroid gland, with a notably high positive predictive value (90.9%), indicating that surgeons can have high confidence in a positive result. The relatively low negative predictive value (41.7%), however, indicates

that absence of staining cannot reliably exclude the presence of a parathyroid gland. No recurrent laryngeal nerve injury occurred in this cohort, a promising finding, although it cannot be attributed to methylene blue use alone. Transient postoperative hypocalcaemia occurred in 23.5% of patients, consistent with rates reported in the literature, indicating that methylene blue does not entirely eliminate this common complication. Overall, methylene blue appears to be a useful adjunct in thyroid surgery, particularly for identifying the parathyroid gland, but should be incorporated into a comprehensive surgical approach rather than relied upon in isolation. Larger, randomized controlled trials with long-term follow-up are needed to more definitively establish its role, in comparison with alternative techniques such as intraoperative neuromonitoring and autofluorescence, in routine thyroid surgery practice.

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