



Combined near-infrared autofluorescence and preresection indocyanine green angiography for parathyroid gland preservation during thyroid surgery: A clinical controlled trial

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ABSTRACT

Background: This was a single-institution controlled trial examining whether the intraoperative combination of near-infrared-induced autofluorescence of the parathyroid glands and preresection indocyanine green near-infrared angiograms of the parathyroid feeding vessels can reduce the incidence of postoperative hypoparathyroidism 1 year after surgery in patients undergoing total or completion thyroidectomy.

Methods: Included patients underwent total or completion thyroidectomy. The intervention group was prospective and underwent surgery with bimodal near-infrared-induced autofluorescence of the parathyroid glands and preresection indocyanine green near-infrared angiograms. The control group was retrospective and underwent standard surgery. Patients were matched with a 1:3 ratio for intervention/control. Minimal matching criteria were procedure (completion/total thyroidectomy), surgical indication (compressive symptoms/hyperthyroidism/suspicion or diagnosis of cancer), substernal goiter, and lymph node resection/neck dissection. The primary outcome was hypoparathyroidism 1 year after surgery, defined as treatment with an active vitamin D analogue.

Results: A total of 154 intervention patients were included and matched with 462 control patients. The incidence of hypoparathyroidism 1 year after surgery was 6.5% in the intervention group and 12.3% in the control group. The risk of hypoparathyroidism 1 year after surgery was significantly lower in the intervention group compared with control patients (odds ratio, 0.50; 95% confidence interval, 0.248–0.996). The difference was even more pronounced after adjustment for potential confounders (sex, age, body mass index, perioperative blood loss, weight of both thyroid lobes, former central neck surgery, operating surgeon) (odds ratio, 0.16; 95% confidence interval, 0.048–0.560).

Conclusion: The bimodal application of near-infrared-induced autofluorescence of the parathyroid glands and pre-resection indocyanine green near-infrared angiograms during total and completion thyroidectomy significantly reduces the risk of hypoparathyroidism 1 year after surgery.

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Introduction

The parathyroid glands (PGs) can be difficult to identify and therefore difficult to preserve during thyroid surgery.¹ Despite protective measures like capsular dissection, loupe magnification,

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and autotransplantation of inadvertently resected PGs, a considerable number of patients are affected by postoperative hypoparathyroidism.¹ This condition carries significant morbidity and is characterized by hypocalcemia because of insufficient levels of circulating parathyroid hormone (PTH).^{1–3} The rate of long-term postoperative hypoparathyroidism after bilateral thyroidectomy may be as high as 12.5–16.6%.^{4,5} Consequently, innovative techniques with the potential to prevent intraoperative parathyroid injury are warranted.⁶

In recent years, 2 near-infrared (NIR) fluorescence techniques have emerged that may help in this regard: near-infrared-induced autofluorescence (NIRAF) and indocyanine green near-infrared angiography (ICGA).⁶ NIRAF relies on the fact that the PGs emit a fluorescent signal when exposed to NIR light.⁷ The technique helps to identify the PGs but does not assess their viability because a completely devascularized PG also emits a fluorescent signal.⁸ The second technique, ICGA, uses the fluorophore indocyanine green (ICG) to create real-time angiograms during surgery.⁹ ICGA can be applied before thyroid resection to create mapping angiograms of the parathyroid-feeding vessels, and/or after resection to guide the decision to autotransplant devascularized PGs.^{10,11} Recently, 3 meta-analyses of randomized controlled trials (RCTs) have demonstrated a significant decrease in transient hypocalcemia in patients operated with NIRAF compared with standard care, whereas the impact on long-term hypoparathyroidism remains unclear.^{12–14} Fewer studies have combined NIRAF with ICGA, and most only apply ICGA after thyroid resection.^{10,15,16}

In this study, we evaluated the effect of the bimodal application of NIRAF and pre-resection ICGA on the incidence of postoperative hypoparathyroidism 12 months after total (TT) or completion thyroidectomy (CT). We hypothesized that this approach would decrease the incidence of postoperative hypoparathyroidism by two-thirds compared with a matched retrospective control group.

Methods

Study design

This was a single-institution clinical controlled trial comparing a prospective intervention group with a matched retrospective control group. The intervention group comprised consecutive patients who underwent TT or CT with NIRAF + ICGA from October 2021 to October 2023 at the Department of Otorhinolaryngology, Head & Neck Surgery at Odense University Hospital, Denmark. Participants signed an informed consent form. The matched control group was randomly selected among 1,041 adults from a database of patients who underwent TT or CT according to standard care during the period January 2011 to December 2020 at the same department.¹⁷ Thyroid surgeons in the database were categorized as low volume if their yearly average of total/completion procedures over the 10-year period was <10.¹⁸ No individual surgeon changed status from low volume/not low volume between the intervention and the control period, and the annual incidence of hypoparathyroidism remained similar throughout the decade. Standard care from 2011 to 2020 was comparable with today's standard practice without fluorescent aides: autotransplantation of accidentally excised parathyroid glands was routine in benign surgery but rare if malignancy was suspected (frozen section was not readily available), and autotransplantation of seemingly devascularized parathyroid glands remaining in situ was performed at the discretion of the surgeon.

The study was registered at both [ClinicalTrials.gov](https://clinicaltrials.gov) (NCT05130476) and the Region of Southern Denmark (21/28,899) and approved by the Region of Southern Denmark (20/50,700) and the Regional Committees on Health Research Ethics for Southern

Denmark (S-20200154). Expanded access to indocyanine green was granted by the Danish Medicines Agency (case number 2020091021). The Strengthening the Reporting of Observational Studies in Epidemiology guideline was used.

Intervention group

Patients ≥ 18 years scheduled to undergo TT or CT were eligible for inclusion. Exclusion criteria were no bilateral thyroidectomy, allergy to iodides/ICG, estimated glomerular filtration rate <45 mL/min/1.73 m², radioactive iodine uptake study scheduled <7 days after surgery, primary/tertiary hyperparathyroidism, preoperative hypoparathyroidism/treatment with an activated vitamin D analogue (AVDA; Anatomical Therapeutic Chemical classification system A11CC02, A11CC03, or A11CC04), or postoperative AVDA treatment for reasons other than iatrogenic parathyroid injury. Deviations from the strict intraoperative fluorescence protocol ([Figure 1](#)) were not an exclusion criterion.

Matching criteria

We used 1:3 matching for intervention/control. Minimal matching criteria were surgical indication, surgical procedure (TT/CT), substernal goiter (yes/no), and lymph node resection/neck dissection (yes/no). When possible, the operating surgeon was matched as well.

Matching algorithm

The matching algorithm “calipmatch” was installed to Stata from Boston College Statistical Software Components (SCC) archive. This algorithm performs one-to-many matching without replacement on pre-specified exact criteria as well as on a mandatory caliper variable. The mandatory caliper variable was bypassed by matching body mass index within a caliper so big (± 60) that any body mass index was in fact accepted. Consequently, matching was only performed on the exact criteria mentioned above. The initial value of the random-number seed was set to “12345” and the seed for randomly breaking ties was set to “67890”.

Demographic and clinical data

Collected variables consisted of blood tests and the variables listed in [Tables I–III](#). Surgical indication was divided into (1) compressive symptoms, (2) hyperthyroidism (Graves disease, toxic nodular goiter, or hyperfunctioning thyroid nodules), and (3) a diagnosis/suspicion of cancer. Patients from the latter category adhered to a standardized national protocol, the “cancer patient pathway.”¹⁹

Primary outcome

The primary outcome was the incidence of hypoparathyroidism 12 months after surgery. In compliance with the Danish National Thyroid Database and the recent recommendations from a European expert panel, only treatment with an AVDA served as a marker of hypoparathyroidism.^{20,21} Ordinary over-the-counter vitamin D and calcium supplements, in contrast, may be taken for various reasons and were not considered markers of hypoparathyroidism. For instance, the Danish Health Authority recommends these supplements for all citizens at risk of osteoporosis and/or age ≥ 70 years.²² Remission was defined as discontinued treatment with an AVDA. A cross-link with the Danish National

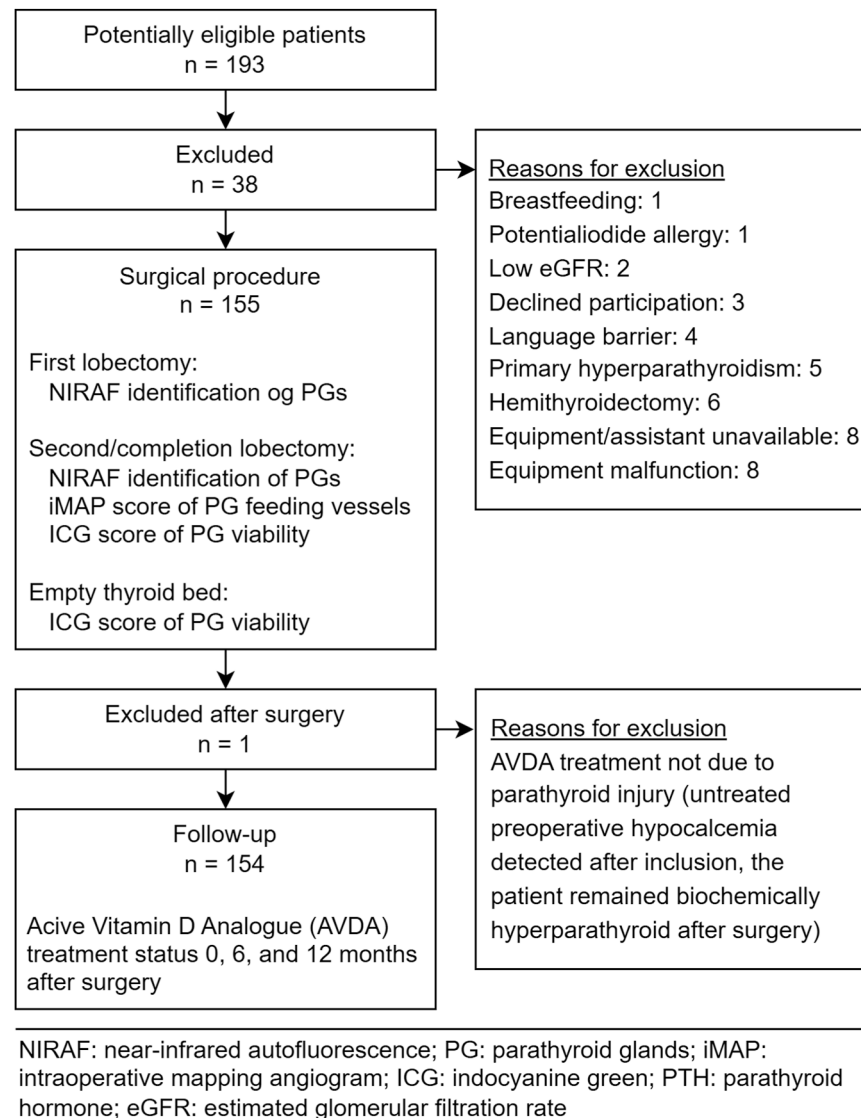


Figure 1. Flowchart for the prospective intervention group. AVDA, activated vitamin D analogue; eGFR, estimated glomerular filtration rate; ICG, indocyanine green; iMAP, intraoperative mapping angiogram; NIRAF, near-infrared autofluorescence; PG, parathyroid gland; RLN, recurrent laryngeal nerve.

Prescription Registry regarding filled prescriptions for AVDAs ensured complete follow-up regarding the primary outcome.¹⁷

Secondary outcomes

Secondary outcomes were (1) The PG identification rate, assuming 2 PGs at risk per thyroid lobe; (2) the duration of NIRAF/ICGA, including time trends; (3) PG/vessel perfusion scores; (4) whether the presence of one well-vascularized PG could rule out long-term hypoparathyroidism²³; (5) postoperative over-night stays in hospital; (6) recurrent laryngeal nerve (RLN) palsies; and (7) whether the surgeon considered NIRAF/ICGA to have benefitted the procedure.

Intraoperative protocol (intervention group)

Neural monitoring, bipolar coagulation, and loupe magnification was used. Lymph node exploration of level VI was performed if malignancy was diagnosed/suspected. If metastases were suspected, the involved and adjacent levels were dissected. Frozen section to confirm parathyroid tissue was not standard practice. A

dedicated assistant familiar with the equipment was present for most surgeries. To maximize external validity, any thyroid surgeon on staff could perform fluorescence procedures.

NIRAF and ICGA was performed using an EleVision IR H&N System (Medtronic, Minneapolis, MN), which has a 785-nm laser and captures emitted light at wavelengths of 800–850 nm.²⁴ The system automatically recorded each NIR laser session, thereby documenting the duration of the fluorescence procedures. The designated distance between the camera and the tissue was 20 cm, but could be modified by the surgeon since the device was handheld.²⁴

First lobectomy (only TT)

Once the thyroid lobe was sufficiently mobilized to expose its backside, the surgical field was scrutinized for PGs by the surgeon's unaided eye and by NIRAF. Fluorescent structures detected by NIRAF were reinspected by the surgeon to rule out false positive results. ICGA was not utilized during the first lobectomy, since the signal from NIRAF can no longer be detected once ICG has been injected.²⁵

Table I
Patient characteristics

Preoperative variables	Control <i>n</i> = 462	Intervention <i>n</i> = 154	<i>P</i> value
Matched variables			
Indication			.999
Compressive symptoms	159 (34.4%)	53 (34.4%)	
Hyperthyroidism	66 (14.3%)	22 (14.3%)	
Diagnosed/suspected cancer	237 (51.3%)	79 (51.3%)	
Procedure			.999
Completion thyroidectomy	198 (42.9%)	66 (42.9%)	
Total thyroidectomy	264 (57.1%)	88 (57.1%)	
Lymph node resection/neck dissection	84 (18.2%)	28 (18.2%)	.999
Substernal goiter	84 (18.2%)	28 (18.2%)	.999
Nonmatched variables			
Sex ratio (M/F), %M	127:335 (27.5%)	45:109 (29.2%)	.679
Age, yr	53.0 (42.0–66.0)	56.0 (42.0–67.0)	.705
Body mass index, kg/m ²	27.1 (23.7–30.8)	27.7 (24.5–32.3)	.073
Former central neck surgery	22 (4.8%)	12 (7.8%)	.158
Operating surgeon [*]	<i>n</i> = 8	<i>n</i> = 10	<.001
Peri- and postoperative variables			
Perioperative blood loss, mL [†]	38.0 (18.0–83.0)	35.0 (18.5–90.0)	.205
Weight of both thyroid lobes, g [‡]	55.5 (27.0–120.0)	34.5 (18.0–84.0)	.037
Duration without NIRAF/ICGA, min [§]	75.0 (57.0–102.0)	81.0 (57.6–108.2)	.073
Malignant histopathology	233 (50.4%)	70 (45.5%)	.307
Postoperative nights in hospital	2.0 (2.0–3.0)	2.0 (1.0–2.0)	.002
Detected PGs at risk	768 (52.9%)	384 (79.3%)	<.001
≥1 PG on histopathology	63 (13.6%)	10 (6.5%)	.021
Autotransplantation	41 (8.9%)	17 (11.0%)	.489
Reoperation for bleeding ≤14 d	10 (2.2%)	5 (3.2%)	.545
RLN palsy at discharge	28 (6.1%)	7 (4.5%)	.553
RLN palsy at 12 mo	13 (2.8%)	2 (1.3%)	.378

Data are presented as median (interquartile range) for continuous measures and number (%) for categorical measures.

F, female; ICGA, indocyanine green angiography; M, male; NIRAF, near-infrared autofluorescence; PG, parathyroid gland; RLN, recurrent laryngeal nerve.

^{*} Surgeons differ because of new additions to the surgical team + supervised residents are grouped as “other surgeon.”

[†] Missing data from 53 controls + 6 intervention.

[‡] Missing data from 80 controls + 12 intervention.

[§] Missing data from 5 controls.

^{||} Diagnosed by fiberoptic/indirect laryngoscopy.

Table II
Postoperative hypoparathyroidism over time in intervention and control group

Hypoparathyroidism (AVDA treatment)	Intervention, <i>n</i> (%)	Control, <i>n</i> (%)	Unadjusted OR [95% CI]	Adjusted [*] OR [95% CI]
At discharge				
Yes	17 (11.0%)	42 (9.1%)	1.24 [0.68–2.26]	0.67 [0.29–1.53]
No	137 (89.0%)	420 (90.9%)	<i>P</i> value .476	<i>P</i> value .345
6 mo after surgery				
Yes	14 (9.1%)	56 (12.1%)	0.74 [0.40–1.34]	0.44 [0.18–1.06]
No	140 (90.9%)	406 (87.9%)	<i>P</i> value .319	<i>P</i> value .067
12 mo after surgery				
Yes	10 (6.5%)	57 (12.3%)	0.50 [0.25–0.996]	0.16 [0.05–0.56]
No	144 (93.5%)	405 (87.7%)	<i>P</i> value .049	<i>P</i> value .004

AVDA, active vitamin D agonist; CI, confidence interval; OR, odds ratio for fluorescence group.

^{*} Adjusted for sex, age, body mass index (kg/m²), perioperative blood loss (mL), weight of both thyroid lobes (g), former central neck surgery, operating surgeon.

Second/completion lobectomy (TT and CT)

The PGs were identified as described previously. Subsequently, preresection ICGA was performed by injecting 0.1 mg/kg ICG intravenously, followed by 5–10 mL of isotonic saline. The contrast arrived at the surgical field after approximately 30 seconds, and the surgeon then adjusted the planned path of resection to spare the parathyroid feeding vessels whenever possible. The ability of the angiogram to guide dissection was evaluated with “intraoperative mapping angiogram” (iMAP) scores¹⁰: iMAP-0 (not

informative), iMAP-1 (no clear vascular pedicle to the PG, but general vascular pattern visible), iMAP-2 (clear vascular pedicle to the PG). PG perfusion was evaluated with ICG scores²⁶: ICG-0 (black, not vascularized), ICG-1 (gray/heterogeneous, partially vascularized), ICG-2 (white, well vascularized).

After resection (TT and CT)

In the empty thyroid bed, previously detected PGs were rediscovered with the surgeon's unaided eye (if possible).

Table III
Minutes spent on NIRAF and ICGA according to procedure

Modality	Completion thyroidectomy	Total thyroidectomy
	<i>n</i> = 66 Median (IQR)	<i>n</i> = 88 Median (IQR)
NIRAF	2.99 (2.15–3.65)	4.18 (3.38–5.19)
ICGA*	2.87 (2.06–3.67)	4.40 (3.40–5.90)
NIRAF + ICGA*	6.07 (4.95–7.24)	8.77 (7.27–11.05)

ICGA, indocyanine green angiography; IQR, interquartile range; NIRAF, near-infrared autofluorescence.

* Excluding 6 completion and 2 total thyroidectomies without ICGA.

Subsequently, ICG was reinjected to score PG perfusion. At this point, the initial bolus of ICG had been cleared exponentially from the system (half-life: 3–4 minutes).⁹ Autotransplantation was guided—but not dictated—by ICG scores, and ultimately performed at the discretion of the surgeon.²⁷ Finally, the surgeon subjectively indicated whether NIRAF/ICGA had benefitted the surgery.

Postoperative management

In accordance with the national treatment algorithm, the PTH level did not factor into the decision to start AVDA treatment.²⁸ For research purposes, however, plasma PTH was measured on the first postoperative morning in the intervention group. In both groups, plasma ionized calcium was measured twice daily until stable or increasing values were reached, and treatment with an AVDA was initiated if ionized calcium levels declined despite calcium supplementation.²⁸ All patients were examined for recurrent laryngeal nerve dysfunction by fiberoptic or indirect laryngoscopy before hospital discharge. Follow-up took place at the department of oncology for patients with malignant histopathology and (also) at the department of endocrinology for patients treated with an AVDA and patients with benign thyroid disease.

Sample size

The pooled results of 4 previous Danish studies indicated that the probability of hypoparathyroidism among controls was 0.124.^{29–32} On the basis of an anticipated odds ratio (OR) of one-third for the intervention group, a correlation coefficient of 0.2, a power of 80%, and a significance of 5%, a sample size of at least 139 intervention patients and 417 matched controls was required.^{33,34}

Statistics

Because no variables were normally distributed (Shapiro-Wilk test), continuous variables were compared with simple linear regression using standard errors that allow for intragroup correlation (proportion of cases to controls 1:3). Categorical variables were compared with stratified ORs. The primary outcome was assessed with conditional logistic regression for 1:3 matching. Time trends were examined with linear regression.

Statistical calculations were carried out in StataNow/BE: release 18.5 (StataCorp LLC, College Station, TX) and Excel 2016 (Microsoft, Redmond, WA). REDCap electronic data capture tools hosted at Odense University Hospital were used for data collection and management.^{35,36} Confidence intervals (CIs) were 2-sided.

Results

A total of 193 consecutive patients were assessed for eligibility. Thirty-eight were excluded before inclusion, and 1 patient with hyperparathyroidism was excluded after surgery due to starting treatment with an AVDA for reasons other than iatrogenic parathyroid injury (Figure 1). The remaining 154 patients were each matched with 3 controls, resulting in 462 control patients. The operating surgeon could only be matched in 78% of cases (*n* = 120/154) and constituted the only significant difference between groups at baseline (Table I). In the 34 cases in which the operating surgeon could not be completely matched, there was an overweight of low volume surgeons (new additions to the surgical team). Any theoretical effect of this discrepancy on the results would favor the control group, however, because intervention patients who underwent surgery by a low volume surgeon (*n* = 19) were matched 1:3 with patients who underwent surgery by a high-volume surgeon in the majority of cases (*n* = 53/57, 93%). In contrast, intervention patients who underwent surgery by a high volume surgeon (*n* = 15) were matched with patients who underwent surgery by a low-volume surgeon in only a minority of cases (*n* = 6/45, 13%).

Preoperative biochemistry from the intervention group showed that 31.8% (*n* = 49/154) of patients had subnormal vitamin D levels, 23.4% (*n* = 36/154) had secondary hyperparathyroidism, and 5.2% (*n* = 8/154) were hypocalcemic. Complete preoperative biochemistry was not available for the control group.

Primary outcome

One year after surgery (median, 367 days; interquartile range, 362–379), 10 (6.5%) patients in the intervention group received treatment with an AVDA. Of these, 2 (20%) had normoparathyroid normocalcemia, 2 (20%) had normoparathyroid hypocalcemia, 5 (50%) had hypoparathyroid normocalcemia, and 1 patient (10%) had hypoparathyroid hypocalcemia. A measurable level of PTH was detected in all patients at some point during follow-up.³⁷

In the control group, 57 (12.3%) patients were treated with an AVDA 1 year after surgery (data from the Danish National Prescription Registry 365 days after surgery). Complete biochemistry was not available, but during the subsequent 2–12 years of follow-up, 9 additional control patients started treatment with an AVDA for hypoparathyroidism (median 18 months after surgery), whereas 5 patients went into remission (median 16 months after surgery).

The risk of hypoparathyroidism 1 year postoperatively was significantly lower in the intervention group compared with the control group (OR, 0.50; 95% CI, 0.25–0.996) (Table II). The difference was even more pronounced after adjustment for all potential confounders (OR, 0.16; 95% CI, 0.05–0.56) (Table II).

Secondary outcomes

The identification of PGs at risk was significantly greater in the intervention group (*P* < .001, Table I). Furthermore, 36.5% of identified PGs in the intervention group were detected by NIRAF before they were seen by the surgeon (*n* = 140/384). Twenty of these (5.2% of 384) were located under a thin layer of tissue with no further attempt at visual confirmation by the surgeon.³⁸

The duration of NIRAF/ICGA is presented in Table III. The time spent on ICGA during TT procedures decreased significantly over the 2 year duration of the study (*P* < .001) (Figure 2).

Intraoperative mapping angiogram scores are presented in Table IV. Only 78.5% of PGs identified preresection during the second/completion lobectomy were evaluated with preresection

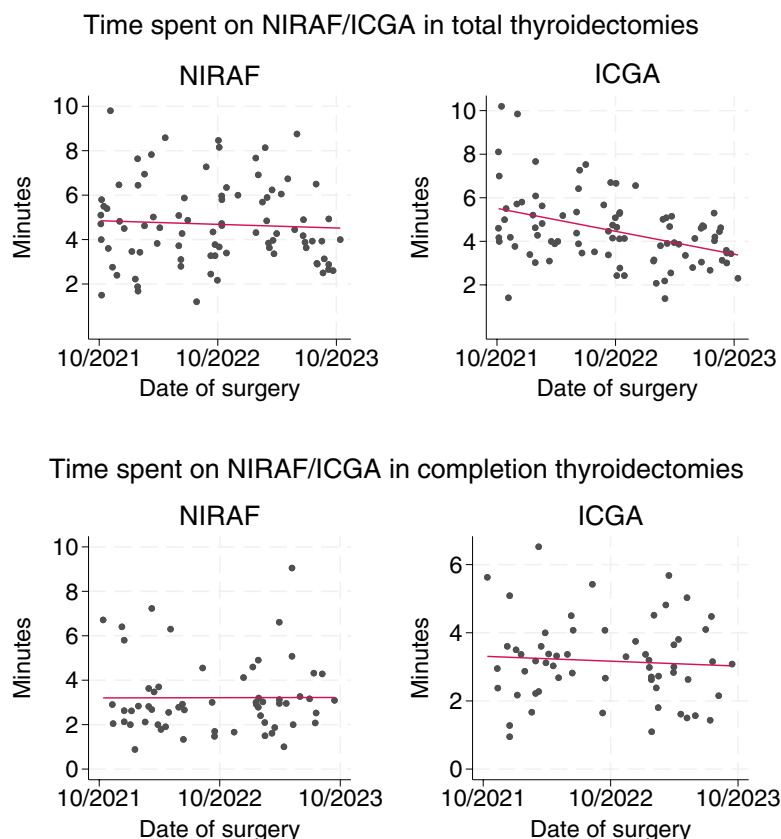


Figure 2. Duration of fluorescence procedures over time. ICGA, indocyanine green angiography; NIRAF, near-infrared autofluorescence.

Table IV
Detected parathyroid glands and iMAP scores in the intervention group

	<i>n</i> (%)
Parathyroid glands at risk	484
Number of detected parathyroid glands	384/484 (79.1%)
Parathyroid glands identified preresection	213
Parathyroid glands evaluated with ICGA preresection	168/213 (78.9%)
iMAP scores	
iMAP-2: Clear vascular pedicle to the parathyroid gland	44/168 (26.2%)
iMAP-1: No clear vascular pedicle, but general vascular pattern discernible	100/168 (59.5%)
iMAP-0: Not informative	24/168 (14.3%)

ICGA, indocyanine green angiography; iMAP, intraoperative mapping angiogram.

ICGA ($n = 168/213$). The most common reason for the omission of the procedure was the wish to use NIRAF to locate a yet-unidentified PG. The preresection angiogram showed a clear vascular pedicle going into the PG in 26.2% of cases ($n = 44/168$), could guide resection in the absence of a clear vascular pedicle in 59.5% of cases ($n = 100/168$), and was noninformative in 14.3% of cases ($n = 24/168$). Some parathyroid glands had already been devascularized in the process of mobilizing the thyroid lobe to expose its backside. Consequently, by the time preresection ICGA was performed, 13.7% of PGs were already devascularized (ICG-0, $n = 23/168$). Of the remaining 145 perfused glands, 99.3% ($n = 144/145$) could be spared during the subsequent angiogram-guided dissection.

The presence of at least 1 well-perfused PG (ICG-2) at the conclusion of surgery did not rule out postoperative hypoparathyroidism, seeing as this applied to 4 of 10 patients with hypoparathyroidism 1 year after surgery. The number of postoperative overnight stays in hospital was significantly lower in the intervention group, whereas there was no significant difference in the rate of laryngeal nerve palsy (Table 1). Finally, surgeons subjectively judged that either NIRAF or ICGA had benefitted the surgery in 81.8% of cases ($n = 126/154$). NIRAF was of benefit in 73.4% ($n = 113/154$) of cases, and ICGA in 61.0% ($n = 94/154$) of cases.

Discussion

To our knowledge, this is the largest study combining NIRAF and pre-resection ICGA to date.^{14,39} We made 4 principal findings: (1) The combination of NIRAF and preresection ICGA significantly reduced the risk of hypoparathyroidism 12 months after surgery compared with a matched retrospective control group; (2) significantly more PGs were detected in the fluorescence group; (3) the duration of ICGA decreased significantly over time in TTs; and (4) the hospital stay was significantly shorter in the fluorescence group.

Previously, prospective studies from high-volume institutions have not been able to demonstrate a significant reduction in long-term hypoparathyroidism by using NIRAF and/or ICGA.^{6,13,40} However, very few studies have combined NIRAF with preresection ICGA, as done in the present trial, and this combination has recently been shown to reduce postoperative

hypoparathyroidism compared with NIRAF or postresection ICGA alone.^{41,42} Furthermore, previous studies were often performed by high-volume “expert surgeons,” which may underestimate the true incidence of long-term hypoparathyroidism among controls and make it difficult to detect significant differences in the target group.⁴³ In comparison, the incidence of long-term hypoparathyroidism in our control group reflects current clinical practice and matches the incidence of nationwide studies.^{4,5}

There are conflicting results in the literature regarding whether NIRAF can identify more PGs than the unaided eye.¹³ However, the gold standard for identification of PGs is visual confirmation by the surgeon, which may bias outcomes.⁶ We believe that the low detection rate of PGs in our control group was partly due to inconsistent PG notation in the retrospective surgical notes. Nevertheless, the increased detection rate in the intervention group was corroborated by significantly fewer inadvertently excised PGs (Table 1) as well as a lower rate of long-term hypoparathyroidism.

Concerns about increased operating time may be an obstacle to the implementation of intraoperative fluorescence procedures. In our study, the median duration of NIRAF + ICGA in TTs was less than 9 minutes, and we detected a decreasing time trend for ICGA over the course of the study. If intraoperative fluorescence techniques can significantly reduce the incidence of long-term hypoparathyroidism, small increases in operating time may be an acceptable trade-off.

There have been contradictory reports regarding the effect of NIRAF/ICGA on the length of the postoperative hospital stay.^{12,44} In our study, the hospital stay was significantly shorter in the intervention group than in the control group. Notably, the rates of AVDA treatment upon hospital discharge were similar in the 2 groups, despite a higher rate of long-term hypoparathyroidism among control patients (Table II). This probably reflects that a significant fraction of control patients encountering postsurgical hypocalcemia were monitored in-hospital for a longer time while being treated with calcium supplements only instead of AVDA. In the intervention group, in contrast, the increased awareness of postoperative hypoparathyroidism among attending surgeons, combined with routine PTH measurements, resulted in a lower threshold for initiating AVDA in case of hypoparathyroid hypocalcemia.

Although NIRAF/ICGA may be a helpful tool, our results do not support that postoperative measurements of calcium and PTH can be omitted if at least one well-perfused parathyroid gland is preserved, as indicated by a previous study.²³ On the contrary, it is well known—and in line with our data—that patients with postoperative parathyroid insufficiency may require medical treatment despite detectable, or even normal, PTH levels.^{1,37}

Finally, the increased focus on the PGs in the intervention group did not come at the expense of more RLN palsies. Nevertheless, surgeons should remain aware of the risk of stretching the RLN when manipulating the thyroid gland to improve exposure to NIR light.¹⁰

Our study has some limitations. It was a single-center study, which limits external validity. However, we attempted to maximize external validity by not excluding results from surgical procedures in which the intraoperative protocol was not strictly followed, and by neither precluding junior surgeons nor patients with preoperative vitamin D deficiency from participating. Despite our encouraging results with NIRAF + ICGA, the incidence of long-term hypoparathyroidism in the intervention group was greater than hypothesized. Because our study was embedded in a routine clinical setting, the results reflect the very first experience with fluorescence equipment among 10 surgeons with mixed attitudes toward the technology. It is therefore likely that outcomes could improve with further practice.

The main limitation and a relevant point of criticism is our use of a historical control group. We thereby risk overlooking a Hawthorne effect, in which results improve simply because surgeons come under scrutiny. However, there is data to support that the impact hereof on our results is limited. For instance, the prospective—but otherwise comparable—control arm in a fluorescence RCT from a neighboring hospital, showed levels of postoperative hypoparathyroidism which were similar to ours, despite their surgeons coming under scrutiny (11.8% 3 months postoperatively vs. our 12.1% 6 months postoperatively).⁴⁵ Furthermore, when Benmiloud et al controlled for the Hawthorne effect in a before-and-after study, they reported an improved PG identification rate in the “Hawthorne arm,” but no resulting decrease in the rate of hypocalcemia or inadvertent PG resection.⁴⁶ This underlines that an artificially increased attention to the PGs, however welcome, does not necessarily result in an increased preservation of parathyroid function.

The limitations of the NIRAF/ICGA techniques per se have been described in previous papers.^{6,10,47} In our experience, a potential pitfall, which can lead to false negative results for both NIRAF and ICGA, is when a deep or narrow surgical field creates obstructions between the NIR laser beam and the tissue of interest. Consequently, sound surgical judgment is still required.

In future research on NIRAF/ICGA, investigators should consider scaling studies to be able to detect 50% reductions in the rate of long-term hypoparathyroidism, instead of anticipating 70–75% reductions.^{48,49} This would be in line with our results and a recent meta-analysis of RCTs on the use of NIRAF.¹⁴ Such considerations, however, will probably warrant cost-effectiveness analyses that take quality of life issues and the economic burden on the healthcare system into account.⁵⁰

In conclusion, this study has shown that NIRAF combined with pre-resection ICGA can significantly reduce long-term postoperative hypoparathyroidism, even when implemented in an everyday clinical setting and including a broad range of patients with benign and malignant thyroid diseases.

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Conflict of Interest/Disclosure

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CRediT authorship contribution statement

Sanne H. Michaelsen: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Resources,

Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Helle Døssing:** Writing – review & editing, Data curation. **Oke Gerke:** Writing – review & editing, Writing – original draft, Methodology, Formal analysis. **Kristine Bjørndal:** Writing – review & editing, Data curation. **Anders R. Madsen:** Writing – review & editing, Data curation. **John Jakobsen:** Writing – review & editing, Data curation. **Mette Bay:** Writing – review & editing, Data curation. **Max Rohde:** Writing – review & editing, Data curation. **Marie Westergaard-Nielsen:** Writing – review & editing, Data curation. **Christian Godballe:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Funding acquisition, Data curation, Conceptualization. **Steen J. Bonnema:** Writing – review & editing, Writing – original draft, Supervision, Methodology. **Viveque E. Nielsen:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Data curation, Conceptualization.

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