

Peer Review File

Article information: <https://dx.doi.org/10.21037/gs-22-35>

Reviewer A Comment 1:	<p>whether it should really combine the description of three cases with a literature review rather than focus only on the above-mentioned description and refer to data from the literature while expanding the discussion,</p> <ul style="list-style-type: none"> • If we include a literature review in the proposed article, in my opinion, it should refer to the following points: <ul style="list-style-type: none"> <input type="checkbox"/> Comparison of therapeutic effects, <input type="checkbox"/> Comparison of possible complications in the analyzed groups, <input type="checkbox"/> Comparison of possible complications in the analyzed groups,
Reply	We appreciate reviewer suggestion. We edited the manuscript to highlight the points recommended by the reviewer.
Changes in the text	Page 11, Paragraph 2, Line 223-227. “Thyroidectomy allows definitive cure, however it by default carries with it the risk a relatively large risk of permanent hypothyroidism, recurrent laryngeal nerve injury, and hypoparathyroidism. While effective, RAI however goes the patient to exogenous radioactive elements which, according to a recent meta-analysis, display a linear-dose response relationship with solid cancer mortality. Resultantly, the obvious benefits of RFA, including minimal risk of hypothyroidism and a lack of scar, may be an especially attractive therapy for patients with severe hyperthyroidism, previous malignancy, or cosmetic concern.
Reviewer A Comment 2:	<input type="checkbox"/> Treatment cost,
Reply	We appreciate the author comment pinpointing this question, especially given the higher cost of care in the United States. This concern was discussed in the discussion.
Changes in the text	Page 11-12, Paragraph 2, Line 234-237. “With respect to quality of life and cost, however, a Chinese study found that RFA patients had significantly better general health and mental health than open surgery for benign thyroid nodules, though it was more costly. No studies from the US investigate the const effectiveness of RFA of toxic thyroid nodules.”
Reviewer A Comment 3:	<ul style="list-style-type: none"> <input type="checkbox"/> Comparison of the current guidelines and recommendations • (e.g., Thyroid Radiofrequency Ablation Guideline: Korean Society of Thyroid Radiology, AACE / ACE / AME Task Force on Thyroid Nodules, • Italian Minimally Invasive Treatment of the Thyroid (MITT), and, finally: • European Thyroid Association Clinical Practice Guideline for the use of image-guided ablation in benign thyroid nodules. • European Thyroid Association and Cardiovascular and Interventional Radiological Society of Europe 2021: Clinical Practice Guideline for the use of minimally invasive treatments in malignant thyroid lesions.
Reply	Addressed in the manuscript.
Changes in the text	Page 4, paragraph 1, line 60-62. “International guidelines, including those of Italy and Korea, allow RFA of AFTN’s if a patient refuses first-line treatments (which include radioactive iodine ablation and surgery).”
Reviewer A Comment 4:	• Is only one FNAB biopsy sufficient to evaluate a nodule and qualify for RFA treatment on TIRADS 4 and 5?

Reply	We appreciate the reviewer comment. The 2015 ATA guidelines recommend two FNA biopsies prior to RFA for typical nodules, so as to ensure non-malignant lesions. All our patient has at least one fine needle aspiration biopsy performed by their endocrinologist and was benign. Prior to RFA, a second FNA was performed at our clinic. Second FNA was benign as well.
Changes in the text	NA
Reviewer A Comment 5:	Is it advisable to test TRAb prior to therapy?
Reply	Establishing a clear diagnosis in patient with hyperthyroidism is critical for treatment planning. Ultrasonography, nuclear medicine thyroid scan and laboratory work up including TG antibodies play a significant role to differentiate Graves' disease from toxic thyroid nodule. Although some studies included the TG antibodies data (see "Percutaneous microwave ablation of thyroid nodules: effects on thyroid function and antibodies" by Heck et al.), the 2016 ATA guidelines did not list or discuss a routine preprocedural evaluation of TG antibodies.
Changes in the text	NA
Reviewer A Comment 6:	• Does the size of the nodular lesion affect the therapeutic effects in AFTN?
Reply	The relationship between treatment response and nodular size and volume has been previously studied. We highlighted this association as reported in the manuscript.
Changes in the text	Page 11, paragraph 1, line 215-218. "A recent meta-analysis published by Cesareo et al. investigated the association between the baseline toxic nodules volume and rate of post ablation TSH normalization. The authors reported that smaller nodules had a higher chance to be treated with TSH normalization than larger ones."
Reviewer A Comment 7:	• What group of patients can RFA be recommended to (can the criteria of age, comorbidities, cosmetic effects? affect the recommendations?)
Reply	The Korean guidelines recommended RFA only as an alternative treatment option to surgery and radioactive iodine ablation especially in those who are poor surgical candidates or those who refuse primary treatment options. Similar recommendation was previously adopted by the ATA guidelines. Despite gaining attention and gaining popularity, there is a little data in the literature highlighting which patients categories can benefit the most from RFA (except from few studies that concluded better response rate with smaller nodules with lower volumes.
Changes in the text	Page 4, paragraph 1, line 60-62
Reviewer B Comment 1:	Abstract: Line 47: several case reports from Asia and Europe.. It is not just case reports. Italian and Korean groups have already published multicenter study regarding the efficacy of RFA for AFTN. Besides, the meta-analysis about efficacy of RFA in AFTN can also be found in the literatures (Your reference 18).
Reply	We appreciate the reviewer comments improving our manuscript. We edited the introduction as recommended.
Changes in the text	Page 1, paragraph 1, line 45-64 "However, several reports from Asia and Europe have described the resolution of hyperthyroidism secondary to AFTN's with RFA."
Reviewer B Comment 2:	Introduction: Line 58: Small scale studies of euthyroid patients... It is not just small-scale studies.

	Multiple studies with large patient numbers have shown that RFA does not typically affect thyroid function.
Reply	Addressed.
Changes in the text	Page 3, paragraph 2, line 56-58. “Several studies of euthyroid patients receiving RFA for benign nodules have concluded that RFA does not typically affect thyroid function (8–10).”
Reviewer B Comment 3:	Literature Review First paragraph: Line 73-88: The role of RFA to treat benign, non-functional thyroid nodules has already been well-established and accepted worldwide. Brief review it and mainly focus on the efficacy of RFA to treat AFTN.
Reply	We thank the reviewer for their suggestion. Section was addressed.
Changes in the text	Page 3, paragraph 2, line 54-55. Removed 3 sentences and summarized them as: “Since its introduction, RFA has been shown to achieve volume reduction rates (VRRs) as impressive as 80 and 90%.”
Reviewer B Comment 4:	Line 89: There are small scale studies... It is not just small-scale studies, please modify the descriptions.
Reply	Addressed.
Changes in the text	
Reviewer B Comment 5:	Line 118: making our study the first of kind in the USA... In your reference 3, a study from Mayo Clinic has mentioned using RFA to treat one patient with subclinical hyperthyroidism due to toxic adenoma. Besides, a study from Iram Hussain et al. (published in Journal of the Endocrine Society, 2021, Vol. 5, No. 8, 1–12), from University of Texas Southwestern Medical Center, Dallas, USA, has included 24 AFTNs assessed after RFA.
Reply	Addressed.
Changes in the text	Page 10-11, paragraph 2, line 210-214. “Within the U.S, there are currently two studies which have reported the efficacy and safety of RFA treatment on AFTN’s. Hussain et al. reported the largest American study including 24 AFTN’s which were ablated without major complication, achieving mean TSH and free T4 levels within normal range and a 71.1% VRR after a median of 87 days (27). One study of a single patient with an AFTN was found to have normal thyroid function 4 months post-ablation (9).”
Reviewer B Comment 6:	Line 89-104 The three cited references (5,6,7) provided similar information (no significant relationship between nodule characteristics and TSH remission after RFA). However, other studies have found some nodule characteristics might be predictive factors of RFA in treating AFTN. Please cite these references and discuss the possible reasons.
Reply	Thank you for bringing this to our attention. Addressed.
Changes in the text	Addressed this now in 2 places: “A recent meta-analysis published by Cesareo et al. investigated the association between the baseline toxic nodules volume and rate of post ablation TSH normalization. The authors reported that smaller nodules had a higher chance to be treated with TSH normalization than larger ones (29).” Also “In contrast the previously mentioned studies, a study of 24 AFTN’s found small nodules (<12 mL) respond better to RFA than do medium-sized nodules (>12 mL).” Page 10, paragraph 1, line 193-195.
Reviewer B	Case series:

Comment 7:	<p>What kinds of RFA electrodes do you use? What kinds of anesthesia do you use? Local, intravenous or general anesthesia? What kinds of techniques do you use for RFA? Trans-isthmus or lateral to medial approach? Moving shot or fixed needle techniques? What is the final total energy for each target nodule? Please describe in each cases.</p>
Reply	The operative details have been included in the article.
Changes in the text	Please see page 4, Paragraph 2, Line 70-74. "RFA was performed as an outpatient procedure. The neck was prepared with povidone-iodine swabs and draped in a standard surgical fashion to avoid the uncommon risk of infection or abscess. We utilized internally-cooled 18G STARmed (Seoul, Korea) catheters with a 7 mm tip size to thermally ablate each nodule. In each of the following cases we utilized the moving-shot technique along with a long-axis approach (13)."
Reviewer B Comment 8:	All of the three patients still presented low TSH level before RFA. Although all of your patients did not have special discomforts after RFA, is it still possible to aggravate the symptoms of hyperthyroidism after RFA?
Reply	Although RFA may theoretically aggravate the symptoms of hyperthyroidism in the immediate post ablation period, none of our patients reported any worsening symptoms. All patients were under follow-up with their endocrinologist and our team. Furthermore, TSH was either within normal or elevated at the 1-month interval. Future studies with narrower interval follow up might be needed to address such potential impact.
Changes in the text	NA
Reviewer B Comment 9:	In some studies, they request patients to restore euthyroid stage before RFA procedures, while others do not. Please discuss this issue in the discussion section.
Reply	Addressed.
Changes in the text	Page 11, paragraph 3, line 233-235. "Additionally, both RAI ablation and thyroidectomy require starting medical treatment to control TSH levels preoperatively with methimazole or propylthiouracil, which are contra-indicated in pregnancy (33). The notion of achieving a euthyroid state prior to RFA for AFTN's is currently not discussed by recent guidelines (34)."
Reviewer B Comment 10:	Case 1 had TSH of 13.6 after RFA and case 2 presented with transient high TSH after RFA, what is the possible reason to cause this situation? Please discuss in the discussion section.
Reply	Addressed.
Changes in the text	Page 12, paragraph 3, line 254-258. "These two patients had a long history of uncontrolled hyperthyroidism. The disease was poorly controlled even while on antithyroid medications. The temporary hypothyroidism can be explained by the successful ablation of the autonomous functioning tissue, with potentially delayed response of the normal thyroid tissue."
Reviewer B Comment 11:	For case 3, why did the patient stop taking PTU two days before RFA? Did the patient keep taking PTU after RFA?
Reply	Addressed. This patient discontinued his PTU per his endocrinologist recommendations prior to surgery. He did not restart his PTU following ablation.
Changes in the text	NA.
Reviewer B Comment 12:	For figure1,2,3: following RFA, how long? One month later? Please describe in the legends.

	In the figure legends, please also describe the size change before and after RFA and also mention the volume reduction ratio.
Reply	Addressed. Ultrasound pictures at 1 month following ablation.
Changes in the text	Figures edited.
Reviewer B Comment 13:	Discussion: Line 222-225: This paragraph discusses the role of RFA in non-functional thyroid nodules. The main point might focus on the comparison of RFA with surgery to treat AFTN. Please cite references to discuss the efficacy of RFA or surgery to treat AFTN.
Reply	Addressed.
Changes in the text	Paragraph edited to read “Thyroidectomy allows definitive cure, however it by default carries with it the risk a relatively large risk of permanent hypothyroidism, recurrent laryngeal nerve injury, and hypoparathyroidism. While effective, RAI however goes the patient to exogenous radioactive elements which, according to a recent meta-analysis, display a linear-dose response relationship with solid cancer mortality. Resultantly, the obvious benefits of RFA, including minimal risk of hypothyroidism and a lack of scar, may be an especially attractive therapy for patients with severe hyperthyroidism, previous malignancy, or cosmetic concern”
Reviewer B Comment 14:	Please discuss if the patients should restore euthyroid status before RFA.
Reply	Addressed.
Changes in the text	Page 11, paragraph 3, line 232-235. “Additionally, both RAI ablation and thyroidectomy require starting medical treatment to control TSH levels preoperatively with methimazole or propylthiouracil, which are contra-indicated in pregnancy (33). The notion of achieving a euthyroid state prior to RFA for AFTN’s is currently not discussed by recent guidelines (34).
Reviewer B Comment 15:	Also discuss the possible reason to cause hypothyroidism after RFA.
Reply	Addressed.
Changes in the text	Page 12, paragraph 3, line 254-258. “Additionally, both RAI ablation and thyroidectomy require starting medical treatment to control TSH levels preoperatively with methimazole or propylthiouracil, which are contra-indicated in pregnancy (33). The notion of achieving a euthyroid state prior to RFA for AFTN’s is currently not discussed by recent guidelines (34)”
Reviewer C Comment 1:	First the lay out of the article is not correct. As it is a case report the article should start with a description of the cases. Then in the discussion the literature should be mentioned.
Reply	Addressed
Changes in the text	NA
Reviewer C Comment 2:	Introduction: It is a good initiative to address RFA treatment in thyroid disease in the USA as it is underdeveloped in North America. However the literature is ample and consists of more than "several" case reports. The author states that their cases have been cured biochemically however one case is not.
Reply	Thank you for your comment. All patients responded effectively to the RFA with TSH significantly increasing post-ablation. TSH levels were normal or elevated in all patients – suggesting biochemical cure.

Changes in the text	Manuscript edited to read “In our study we found that RFA successfully reversed the thyroid function in one month for all three AFTN’s. However, only a single patient had complete normalization of their thyroid function tests.”
Reviewer C Comment 3:	The case studies are poorly described concerning the RFA procedure. For example we are not informed about the total amount of energy which has been applied.
Reply	Addressed. We added a section named procedural details. We also detailed data about the energy and Impedance in the case description.
Changes in the text	Manuscript edited to read “RFA was performed as an outpatient procedure. The neck was prepared with povidone-iodine swabs and draped in a standard surgical fashion to avoid the uncommon risk of infection or abscess. We utilized internally-cooled 18G STARmed (Seoul, Korea) catheters with a 7 mm tip size to thermally ablate each nodule. In each of the following cases we utilized the moving-shot technique along with a long-axis approach (13).”
Reviewer C Comment 4:	All cases are multinodular and not the typical single autonomic toxic nodule. This raises the question of the short follow up. Are these patients definitely cured in the end? Definite proof of a normal scintigraphy or a longer follow up is absent.
Reply	Although patients may have had multiple nodules on the ultrasound evaluation, there was only a single toxic nodule demonstrated on nuclear medicine scintigraphy scan. The current study reports the short-term outcomes. A follow-up study with larger sample size and a longer follow-up interval to follow.
Changes in the text	NA
Reviewer C Comment 5:	In the discussion the authors mention the chance of hypothyroidism. The greatest fear of the treatment however is vocal cord paralysis. The authors do not mention the results of the literature in that respect.
Reply	Addressed
Changes in the text	Page 12, paragraph 2, line 148. Manuscript edited to read “In his large study, Beak et al. established that the rate of recurrent laryngeal nerve injury following RFA is 1.02% (39)”
Reviewer D Comment 1:	- The term AFTN is explained in the abstract but not in the running text.
Reply	Addressed
Changes in the text	Page 4, paragraph 1, line 60
Reviewer D Comment 2:	- line 62 'hyperthyroid nodules'. Hyperthyroidism is the biochemical status of the patient; a nodule isn't hyperthyroid, it causes hyperthyroidism.
Reply	Addressed
Changes in the text	Edited in the manuscript
Reviewer D Comment 3:	- The introduction ends with stating that three case reports will be presented but is followed by the literature review which in term starts with a repetition of the introduction.
Reply	Addressed and reformatted.
Changes in the text	
Reviewer D Comment 4:	- The authors do not mention the systematic review and meta-analysis by Kim and Cho (Kim, H.J., Cho, S.J., Baek, J.H. et al. Efficacy and safety of thermal ablation for autonomously functioning thyroid nodules: a systematic review and meta-analysis. Eur Radiol 31, 605–615 (2021). https://doi.org/10.1007/s00330-020-07166-0); this review includes the same plus more recent articles on RFA for AFTN.
Reply	Addressed
Changes in the text	Page11, paragraph 1, line 215-219. “Overall, this data highlighted the ability of RFA to restore euthyroid status for a significant number of hyperthyroid patients (28,29). A

	recent meta-analysis published by Cesareo et al. investigated the association between the baseline toxic nodules volume and rate of post ablation TSH normalization. The authors reported that smaller nodules had a higher chance to be treated with TSH normalization than larger ones (29)”
Reviewer D Comment 5:	- Please explain your centres' experience with RFA, ablative device used, which approach and technique was used (e.g. moving-shot), which way the patient was sedated (locally, generally?). Case series:
Reply	Addressed
Changes in the text	Page 4, paragraph 2, line 71-76. “Procedural Details RFA was performed as an outpatient procedure. The neck was prepared with povidone-iodine swabs and draped in a standard surgical fashion to avoid the uncommon risk of infection or abscess. We utilized internally cooled 18G STARmed (Seoul, Korea) catheters with a 7 mm tip size to thermally ablate each nodule. In each of the following cases we utilized the moving-shot technique along with a long-axis approach (13).”
Reviewer D Comment 6:	- In general, every case lacks detailed information on patient history, RFA parameters, sufficient follow-up time (each about 1 month?); follow-up should be at least 6 months and preferably 24 months to monitor recurrence.
Reply	We addressed the reviewer comments. Detailed data about the operative detailed and Impedence were included. We agree with the reviewer concern regarding the duration of the follow up. The current study reports the short-term outcomes to address. A follow-up study with larger sample size and a longer follow-up interval to follow.
Reviewer D Comment 7:	- Case 1; what kind of thyroid scan? Was a TSH antibody test performed? Please report volume and not only nodule sizes.
Reply	NM thyroid uptake scan. Antibodies were tested by the referring endocrinologist and were undetectable for Thyroglobulin and thyroid peroxidase antibodies. Volume have been reported and included in the figures.
Reviewer D Comment 8:	- Case 2: Same concerns. What do you mean with 'multiple shots were delivered'? Please describe if the patient was symptomatic of the subclinical hypothyroidism at one month, please show the results of the normalized thyroid function test.
Reply	The patient did not exhibit symptoms of hypothyroidism. The case was discussed with his endocrinologist and the patient was counselled about the symptoms of hypothyroidism. Treatment was applied in a systematic way starting deep and moving superficially.
Changes in the text	Page 6, Line 119-134.
Reviewer D Comment 9:	- Case 3; mostly the same concerns. line 204 ' T3 wnl'?. (were normal?). Line 208 'change in thyroid volume calculator'? This is the first mentioning of this calculator in you last case.
Reply	Addressed.
Changes in the text	
Reviewer D Comment 10:	- Discussion: Part of your discussion is a review of literature on a novel (for this article) part of RFA (comparison to RAI and complications),I feel that should belong to the review part and not the discussion.
Reply	Addressed
Changes in the text	