Peer Review File

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Reviewer A

Comment 1: The study addresses very interesting issue of the significance of NSAT in stroke (CIS) patient. Authors showed that the patients with NSAT and embolic pattern more commonly present clinical characteristics closer to that of patients with AF. This is however not supported by any comparison of studied group to non-NSAT patients. In fact Authors limited their report to present a clinical characteristic of NSAT pts divided into 2 subgroups based on the presence of embolic pattern in imaging studies. **To make the conclusions more justifiable; my suggestion would be to add age/sex-matched group of non NSAT and then compare the proportions of embolic/non-embolic patterns and other characteristics.** Considering the large number of cases you have in your database (>1700), even a propensity score matching seems be feasible and this would grossly affect the quality of data. The other option would be to add a confirmed AF group and compare their characteristics to NSAT, but I don't think the Authors have to go both directions in one manuscript;

Reply 1: First of all, thank you for giving great interest in our research and an opportunity to publish for *Annals of Translational Medicine*. We appreciate your comment on comparing the proportions of embolic/non-embolic patterns and other characteristics between the group of NSAT with non-NSAT patients.

<1>

To justify the hypothesis that patients with NSAT showed more embolic patterns and clinical characteristics closer to that of patients with AF compared with non-NSAT patients, we slightly modified the patient category as shown below.

Inclusion criteria:

(1) Acute ischemic stroke with
(2) 24-hour Holter monitoring and detected with NSAT
(3) MRI with MRA or CTA
(4) TTE with and without TEE

Change of inclusion criteria

(1), (3), (4): no change

(2): 24-hour Holter monitoring; detection of NSAT was deleted to include patients without NSAT

Exclusion criteria

(1) A history of AF
(2) Documented AF on EKG, 24-hour Holter monitoring or echocardiography

Change of exclusion criteria

(1) Any potential cardiac source of embolism including PFO, aortic atheroma, and other causes in addition to initial exclusion criteria (1), (2).

- To truly compare embolic patterns between patients with and without NSAT, other cardiac embolic sources would potentially disturb the outcome. Therefore, we exclude all patients who can be classified as cardiac embolism with or without other causes of stroke.

<2>
We collected all the imaging characteristics as well as clinical characteristics of patients with and without NSAT. Before any age/sex-matching or propensity score matching, we found that patients with NSAT were statistically older and had a higher proportion of females compared with patients without NSAT. Also, the ratio of patients with and without NSAT was approximately 1:2. We have done propensity score matching (PSM) using age and sex (Table shown at the end). Out of 243 of NSAT and 438 of non-NSAT patients, 179 of patients were matched 1:1 respectively. The data shows that after PSM, the difference in proportion of patients with embolic images were still statistically significant while other risk factors did not show any significant differences. Because a major portion of patients was discarded from the study and that higher age and higher proportion of females seemed to be the characteristic of patients with NSAT, we decided to compare patients with and without NSAT without propensity score matching. To see whether age/sex has any association with embolic patterns in imaging, we have conducted univariable and multivariable logistic regression analysis, including age and sex. The result shows that there was no association between age/sex and embolic patterns.

<3>
Since direct comparison of embolic patterns between patients with and without NSAT was performed, comparison of subgroups based on the presence of embolic patterns in imaging studies in NSAT patients became less significant. Moreover, because patients with a potential cardiac embolic source such as PFO, aortic atheroma were excluded, changes in data were inevitable. Therefore, specific data were modified, and the table was moved to the supplementary material.

<4>
Now that the data related to TOAST classification and imaging characteristics of patients without NSAT is described, a direct comparison of the proportion of specific embolic subtypes depending on the classification of TOAST can be made. Therefore, we added a supplementary table comparing the proportion of specific embolic subtypes upon TOAST classification and discussed the results.

<5>
Since significant changes in the results were made, discussions regarding the results were modified. We have added discussion of the main result about the higher portion of embolic pattern in patients with NSAT and that NSAT was found to be an independent risk factor of embolic stroke. Also, modifications in the discussion on the clinical characteristics of NSAT patients were made.

Changes in the text:
<1> The text has been modified to includes the changes in inclusion/exclusion criteria. (See Page 6-7, line 97-110)
<2> The clinical characteristics compared between patient with and without NSAT were added. (See Page 7, line 110-114) The baseline characteristic as well as comparison of imaging and clinical characteristic between NSAT and non-NSAT patient were modified and added. (See Page 9-10, line 155-174) (see Table 1)
<2> Method and results of logistic analysis were added. (See Page 8, line 140-145) (see Page 10, line 175-187) (see Table 2)
<3> The text in the method of comparison of subgroups based on the presence of embolic pattern in imaging studies in NSAT patients were modified. (See Page 8, line 129-135)
<3> The result of comparison of subgroups based on the presence of embolic pattern in imaging studies in NSAT patients were modified. (See Page 10-11, line 188-198) (See Table S2)
<4> The results on further analysis of imaging features by TOAST classification was added. (See Page 10, line 184-187) (See Table S1)
<4> The earlier result describing the imaging characteristic upon TOAST classification was deleted. (See Page 11, line 201-213)
<5> The text has been modified to include the discussion regarding the changed result due to the added data of patients without NSAT. (See Page 12-14, line 216-262)

Comment 2: The study is a retrospective analysis and does not allow to draw cause-effect conclusions; one can say the findings are related, but not a cause-effect.

Reply 2: We agree with the point that this study was done retrospectively, and it is inappropriate to draw cause-effect conclusions. We have modified the conclusion to reflect the comment made.
Changes in the text: The text has been modified from "In conclusion, an embolic pattern of acute ischemic stroke frequently occurred in patients with NSAT." to "In conclusion, NSAT was significantly associated with embolic pattern in acute ischemic stroke patients. Moreover, NSAT was an independent risk factor of embolic stroke." (See Page 17, line 331-333)

Comment 3: NSAT are recently thoroughly studied in parents with cryptogenic stroke, but it is doubtful they are responsible for thromboembolism; they are more likely to be the markers of PAF which was not found in this particular Holter, but if there are NSATS, it is more likely to catch AF on extended monitoring [1,2,3]; please discuss this issue. Briefly - Short runs are more likely to precede or coincide with AF, but it's probably AF that causes hemodynamic compromise of left atrium which leads to clot formation in LAA; the mechanism where short SV runs would be able to generate clots and cause stoke is unlikely to exist.

1. Gladstone, et al. EMBRACE Investigators and Coordinators. Atrial premature beats predict atrial fibrillation in cryptogenic stroke: Results from the EMBRACE trial. Stroke 2015, 46, 936–941. [CrossRef] [PubMed]
2. Kamel, H.Paroxysmal Supraventricular Tachycardia and the Risk of Ischemic Stroke. Stroke 2013, 44, 1550–1554. [CrossRef] [PubMed]
3. Kulach A et al, Supraventricular Runs in 7-Day Holter Monitoring Are Related to Increased Incidence of Atrial Fibrillation in a 3-Year Follow-Up of Cryptogenic Stroke Patients Free from Arrhythmia in a 24 h-Holter, J. Cardiovasc. Dev. Dis. 2021, 8, 81

Reply 3: We further reviewed the articles, including the 3-references attached in the comment, regarding the relationship between short runs, AF, and thromboembolism. The opinion that short runs are more likely a precursor of AF rather than directly causing hemodynamic clot formation was more plausible. Moreover, our results show that patients with NSAT had characteristics more similar to AF, and the subgroup analysis with NSAT patients with and without an embolic feature showed no significant differences in the total APB or longest run of APB between the two groups. Therefore, we have modified the discussion accounting for the mechanism of embolic stroke in patients with NSAT.

Changes in the text: Discussion on clinical characteristics and laboratory findings comparing subgroups based on the presence of embolic patterns in imaging studies in NSAT patients was deleted. (See Page 14, line 263-278) Discussion regarding the mechanism of embolic stroke in patients with NSAT was modified to reflect the changes in the result. (See Page 14-16, line 279-315) New references were added. (See Page 21, line 424-429)

Comment 4: Results: authors claim the difference in EF 64 vs 67%; I am aware the difference is statistically significant, but this is also irrelevant from the clinical point of view. Then they state in the first sentence of the discussion that lower EF is a risk factor for AF. Low EF is a RF for AF, but when it is low; 64% is certainly not.

Reply 4: Although the difference in EF was statistically significant, we agree that the difference of 3% is irrelevant from the clinical perspective. However, after modification of the exclusion criteria of the study, the difference in EF became insignificant. Therefore, the results and discussion regarding the difference in ER were deleted.

Changes in the text: The text on the result and discussion on the difference in EF was deleted. (See Page 10, line 194) (See Page 14, line 263-265)

Reviewer B

Comment 1: Interesting paper on non sustained atrial tachycardia and the risk of ischemic stroke. There is a growing body of evidence showing atrial tachycardia as a

potential risk factors for ischemic stroke. Some studies have suggested a score similar to the chadvasc score for atrial fibrillation to be developed in patients with atrial tachycardia to ascertain theses risk and the need for anticoagulation.

Reply 1: We appreciate the interest in our paper discussing the relationship between NSAT as a potential risk factor for embolic type ischemic stroke. It would be a great idea to develop a scoring system that can anticipate the risk of embolism and the need for anticoagulation. We will definitely consider it in the near future with accumulated data.

Changes in the text: none

Table. Clinical Characteristics and Imaging Findings Between Patients with or without Nonsustained Atrial Tachycardia in Acute ischemic Stroke Patients Before and After Propensity Score Matching of Age and Sex

	Before matching				After matching			
Variables	**NSAT (n=243)**	**non-NSAT (n=438)**	**Standardized mean difference**	***P***	**NSAT (n=179)**	**non-NSAT (n=179)**	**Standardized mean difference**	***P***
Age, year	71.247±10.734	58.984±13.984	-0.9837	<.0001	68.480±10.564	68.461±10.720	-0.0019	0.986
Female sex	103 (42.39%)	137 (31.28%)	-0.2318	0.0036	74(41.34%)	79(44.13%)	-0.0565	0.593
Hypertension	164 (67.49%)	236 (53.88%)	-0.2813	0.0005	116(64.80%)	112(62.57%)	0.0465	0.66
Diabetes mellitus	84 (34.57%)	124 (28.31%)	-0.1351	0.0894	63(35.20%)	64(35.75%)	-0.0117	0.912
Coronary artery disease	22 (9.05%)	32 (7.31%)	-0.0638	0.4188	16(8.94%)	18(10.06%)	-0.0381	0.718
Dyslipidemia	136 (55.97%)	215 (49.09%)	-0.1381	0.0852	90(50.28%)	103(57.54%)	-0.1461	0.168
Prior history of TIA/stroke	35 (14.40%)	42 (9.59%)	-0.1486	0.0574	22(12.29%)	26(14.53%)	-0.0656	0.535
Smoker	91 (37.45%)	234 (53.42%)	0.3251	<.0001	71(39.66%)	69(38.55%)	0.0229	0.829
Initial NIHSS score	2.000(1.000, 5.000)	2.000(1.000, 4.000)	-0.1184	0.1356	2.000(1.000, 4.000)	2.000(1.000, 4.000)	0.0441	0.4416
CHA_2DS_2-VASc	3.000(2.000, 4.000)	2.000(1.000, 3.000)	-0.6072	<.0001	3.000(2.000, 4.000)	3.000(1.000, 4.000)	0.0452	0.9692
LV EF, %	67.000(62.500, 71.000)	67.000(62.000, 71.000)	-0.1439	0.1214	68.000(62.500, 72.000)	67.000(62.000, 71.000)	-0.1387	0.3494
LA index, ml/m^2	26.800(22.200, 33.900)	24.455(20.105, 29.845)	-0.3299	<.0001	26.300(21.400, 33.700)	25.670(21.130, 30.775)	-0.1506	0.3636
TOAST								
LAA	90 (37.04%)	169 (38.58%)	0.0319	0.6903	63(35.20%)	66(36.87%)	0.0349	0.741
SVO	73 (30.04%)	182 (41.55%)	0.2419	0.0029	60(33.52%)	80(44.69%)	0.2305	0.03
Cryptogenic	76 (31.28%)	78 (17.81%)	-0.3169	<.0001	52(29.05%)	32(17.88%)	-0.266	0.013
NSAT	4 (1.65%)	9 (2.05%)	0.0303	0.7799	4(2.23%)	1(0.56%)	-0.1432	0.371
Any embolic feature	66 (27.16%)	63 (14.38%)	-0.3189	<.0001	46(25.70%)	25(13.97%)	-0.2975	0.005
Simultaneous acute lesions in multiple arterial territories	29 (11.93%)	26 (5.94%)	-0.2114	0.0059	20(11.17%)	12(6.70%)	-0.1571	0.138
Isolated acute cortical lesions	13 (5.35%)	10 (2.28%)	-0.1606	0.0338	8(4.47%)	2(1.12%)	-0.2045	0.054
Angiographic evidence of cutoff or recanalization	24 (9.88%)	27 (6.16%)	-0.137	0.0779	18(10.06%)	11(6.15%)	-0.1437	0.175