ORIGINAL ARTICLE

INOCA: Non-Invasive Assessment of the Pathophysiological Mechanisms Using CZT-SPECT

INOCA: Evaluación no invasiva de los mecanismos fisiopatológicos mediante CZT-SPECT

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ABSTRACT

Background: One of the causes of INOCA (Ischemia with Non- Obstructive Coronary Arteries) is microvascular dysfunction (MVD), which can be noninvasively assessed through the quantification of myocardial blood flow (MBF) and myocardial flow reserve (MFR). Dynamic myocardial perfusion imaging (MPI) by CZT-SPECT at rest, with dipyridamole stress test and cold pressor test (CPT) can establish the presence of two different pathophysiological mechanisms of MVD: endothelium-independent or endothelium-dependent, respectively.
Objectives: The aim of this study was to evaluate the usefulness of CZT-SPECT for the diagnosis of MVD and the different mechanisms involved in patients with INOCA.
Materials and Methods: A total of 93 consecutive INOCA patients were prospectively included and underwent dynamic MPI with CZT-SPECT at rest and with dipyridamole stress test and CPT. THe MBF was quantified using 4DM® software. A MFR response to dipyridamole <2, and changes in MBF (ΔMBF) <1.5 with CPT were considered abnormal responses. MVD was defined in the presence of one abnormal response or both.
Results: CZT-SPECT detected MVD in 85% (n = 79) of the patients with INOCA. Forty-two percent had an abnormal response to both stressors while 43% presented an abnormal response of MBF only with CPT.
Conclusion: The use of CZT-SPECT with both stress tests allowed the evaluation of different possible pathophysiological mechanisms of MVD present in most patients with INOCA.

Key Words: Microvascular blood flow- Vascular endothelium-Vascular smooth muscle-SPECT

RESUMEN

Introducción: Una de las causas propuestas del síndrome INOCA (por sus siglas en inglés: *Ischemia with Non-Obstructive Coronary Arteries*) es la disfunción microvascular (DMV), la cual puede evaluarse en forma no invasiva, mediante la cuantificación del flujo sanguíneo miocárdico (FSM) y la reserva de flujo miocárdica (RFM).
Las imágenes de perfusión miocárdica (IPM) y dinámicas con CZT-SPECT en reposo- dipiridamol - y prueba de frio (PF), permiten establecer la presencia de DMV evaluando diferentes mecanismos fisiopatológicos: endotelio independiente o dependiente, respectivamente.
Objetivos: Evaluar la utilidad de CZT-SPECT en el diagnóstico de DMV y los diferentes mecanismos patológicos involucrados, en pacientes con diagnóstico de INOCA.
Material y métodos: Se incluyeron en forma prospectiva 93 pacientes consecutivos con diagnóstico de INOCA, a los que se les realizó IPM e imágenes dinámicas con CZT-SPECT en reposo-dipiridamol-PF. El FSM se cuantificó con el software 4DM. Se consideró respuesta anormal al dipiridamol una RFM <2 y a la variación del FSM (ΔFSM) <1,5 con PF. Se definió DMV a la presencia de una o ambas respuestas anormales.
Resultados: El CZT-SPECT detectó DMV en un 85% (n=79) de los pacientes con INOCA. El 42% tuvo respuesta anormal con ambos apremios mientras que el 43% restante, mostró una respuesta alterada del FSM sólo con PF.
Conclusiones: El uso de CZT-SPECT empleando ambos apremios,
permitió evaluar diferentes mecanismos fisiopatológicos que causan DMV presente en la mayoría de los pacientes con INOCA.

Palabras Clave: Flujo sanguíneo microvascular - Endotelio vascular - Músculo liso vascular - SPECT

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INTRODUCTION

Ischemia with Non- Obstructive Coronary Arteries (INOCA), a syndrome defined by the presence of signs or symptoms of myocardial ischemia without obstruction of the epicardial coronary arteries, is an increasingly common finding, particularly in women. (1-3)

The overall prevalence is estimated to be close to 39%; however, it varies considerably according to sex, with a prevalence of 33% in men and up to 65% in symptomatic women undergoing elective coronary angiography. (1-3)

Among the different pathophysiological causes explaining these syndromes, vasospasm and microvascular dysfunction (MVD) are the two main mechanisms proposed. (4,5)

Coronary artery vasoreactivity can be invasively assessed by coronary angiography. Microvascular function can also be evaluated by noninvasive imaging methods; among these, cardiac positron emission tomography (PET) imaging is currently the gold standard and most validated method, as it allows quantification of myocardial blood flow (MBF) and myocardial flow reserve (MFR). (6-10)

The novel CZT cameras have higher sensitivity and energy resolution than the conventional SPECT cameras with sodium iodide crystal scintillation detectors, and allow dynamic quantification of MBF and estimation of MFR comparable to PET. (11-13)

Myocardial blood flow and MFR can be quantified after inducing maximal hyperemia which can be attained by different stimuli evaluating different physiological mechanisms. Dipyridamole inhibits endogenous adenosine reuptake, thereby causing microvascular vasodilation by inhibiting calcium influx into the smooth muscle cells, while cold pressor test (CPT) is a powerful sympathetic nervous system stressor that, like acetylcholine, leads to the release of nitric oxide and endothelium-derived hyperpolarizing factors. (7, 14-17)

Noninvasive imaging tests using both stressors to measure MBF and MFR could be useful to establish the diagnosis of MVD and differentiate the mechanisms involved.

The aim of this investigation was to determine the prevalence of MVD in INOCA patients using noninvasive imaging tests with estimation of MFR after dipyridamole stress and changes in MBF (ΔMBF) to CPT.

METHODS

Study design: we conducted a prospective, single-center cohort study.

Population: the study population was made up of 93 consecutive patients with INOCA in the absence of a ≥ 50% diameter stenosis documented by elective conventional coronary angiography (n = 83) or computed tomography coronary angiography (n = 10). Patients with evidence of myocardial infarction, cardiomyopathies, left ventricular dysfunction and valvular heart disease were excluded from the study.

Method for image acquisition and processing

All the patients underwent CZT-SPECT myocardial perfusion scintigraphy using a 2-day protocol without discontinuing their usual medication.

On the first day, 7mCi of Tc^{99m}-MIBI were injected at rest, and dynamic images were obtained for determining baseline MBF, followed by conventional myocardial perfusion imaging (MPI) protocol. At 60 minutes, 0.56 mg/kg of dipyridamole were administered intravenously over 4 minutes; thereafter, 21mCi of Tc^{99m}-MIBI were injected. The hemodynamic values and dynamic images were obtained again to determine MBF after dipyridamole stress and MPI.

On the second day, the baseline hemodynamic values were obtained again and compared with those measured on the previous day. As there were no significant variations (see appendix) and, in agreement with the institutional review board to follow the standard regulations of administering the lowest possible radiation dose, Tc^{99m}-MIBI was not re-injected at rest, and the protocol was directly initiated with the second stressor.

For the CPT, each patient immersed his/her hand into a cold water container with a temperature of 4 °C over 2 minutes. Then, 21 mCi of Tc^{99m}-MIBI were injected and dynamic images were obtained to determine MBF, followed by conventional MPI protocol.

The 4DM® software was used for image processing and MBF quantification at rest and after both stressors. The MBF was expressed in mL/min/g. The MFR was calculated as the ratio of MBF during dipyridamole stress test to resting MBF, and the ΔMBF response to CPT as the ratio of MBF during CPT to resting MBF.

A value of MFR <2 and a ΔMBF to CPT <1.5 were considered abnormal. (14-17) Microvascular dysfunction was defined in the presence of one abnormal response or both. Four groups were obtained after combining the results obtained in our sample of patients: 1) normal MFR and abnormal ΔMBF; 2) abnormal MFR and abnormal ΔMBF; 3) abnormal MFR and normal ΔMBF, and) normal MFR and normal ΔMBF.

Statistical analysis

Quantitative variables were expressed as median and interquartile range (IQR), according to their distribution, and were compared using the Kruskal-Wallis test. Qualitative variables were expressed as percentage and compared using multiple chi-square test. The Bonferroni test was used for comparing groups.

A p value <0.05 was considered statistically significant. All the calculations were performed using StatsDirect 3.3.5 software package.

Ethical considerations

The study was approved by the institutional review board and all the subjects signed and informed consent form.

RESULTS

A total of 93 patients were analyzed. There were no significant differences in patients' baseline characteristics or medications between the different groups, except for the use of statins (Table 1). There were no patients in group 3.

Eighty-eight patients were symptomatic. MVD was evaluated after the first episode of precordial pain in 32, while in the remaining 56 the evaluation was carried out after several symptomatic episodes. Even 18 of them underwent diagnostic angiography on more than one occasion. All cases had the last episode

Table 1. Baseline characteristics of the patients

	Group 1 (n = 40)	Group 2 (n = 39)	Group 4 (n = 14)	p
Age (yrs), mean±SD	59±11	58±12	56±10	ns
Sex				
Male	12 (30%)	16 (41%)	6 (43%)	ns
Female	28 (70%)	23 (59%)	8 (57%)	ns
Symptoms or ECG changes				
Atypical angina	34 (74%)	28 (72%)	10 (71%)	ns
Typical angina	3 (8%)	9 (23%)	4 (29%)	ns
STD >3 mm	3 (8%)	2 (5%)	0	ns
CVRF present				
Diabetes	5 (12%)	5 (13%)	1 (7%)	ns
Hypertension	19 (48%)	19 (48%)	6 (43%)	ns
Smoking habit	14 (35%)	15 (38%)	5 (36%)	ns
Dyslipidemia	24 (60%)	24 (62%)	7 (50%)	ns
4 CVRF	1 (2%)	3 (8%)	0	ns
3 CVRF	5 (12%)	6 (16%)	3 (21%)	ns
2 CVRF	17 (43%)	10 (25%)	3 (21%)	ns
1 CVRF	9 (23%)	13 (33%)	4 (29%)	ns
No CVRF	8 (20%)	7 (18%)	4 (29%)	ns
Menopause	25 (89%)	18 (78%)	8 (100%)	ns
Usual medication				
Aspirin	16 (40%)	17 (44%)	7 (50%)	ns
Beta blockers	11(28%)	15 (38%)	9 (64%)	ns
ACEI	11(28%)	9 (23%)	9 (64%)	ns
ARB	6 (15%)	5 (13%)	0	ns
Clopidogrel	2 (5%)	2 (5%)	2 (5%)	ns
Calcium channel blockers	6 (15%)	12 (31%)	2 (5%)	ns
Trimetazidine	4 (10%)	6 (15%)	2 (5%)	ns
Isosorbide dinitrate	2 (5%)	7 (18%)	0	ns
Statins	25 (63%)	20 (51%)	14 (100%)	0.005

SD: standard deviation ECG: electrocardiogram; STD: ST-segment depression; CVRF: cardiovascular risk factor. ACEI: angiotensin-converting enzyme inhibitors; ARB: Angiotensin II receptor blockers

of precordial pain, at least 30 days prior to the study. Five patients were considered INOCA due to a history of asymptomatic ST segment depression (STD) in stress tests (3 patients in group 1 and 2 patients in group 2).

Myocardial perfusion images were normal at rest and after both stress tests in all the cases. Normal MPI was defined as absence of segmental uptake defects of Tc^{99m}-MIBI, assessed quantitatively by a sum score of zero after each stress and at rest, and qualitatively by comparing the polar maps of each patient with those of the software program. In addition, the normal MPI definition included absence of regional or global wall motion abnormalities or a left ventricular ejection fraction < 55%.

Five patients reported chest discomfort during dipyridamole stress test and none of them presented electrocardiographic changes with both stressors.

Microvascular dysfunction occurred in 79 patients (85%). There were 40 patients (43%) in group 1, 39 (42%) in group 2, no patients in group 3, and 14 patients (15%) in group 4.

Hemodynamic parameters at baseline and after stress tests did not show statistically significant differences between groups, except for MBF at rest between groups 2 and 4 ($p<0.05$), after dipyridamole between group 1 vs 2 ($p< 0.0001$) and vs group 4 ($p<0.05$), as well as between groups 2 and 4 ($p<0.05$), and in CPT between groups 1 and 2 vs group 4 ($p<0.0001$).

We found a significant difference in the MFR between group 1 and 2 ($p < 0.0001$) and between group 2 and 4 ($p < 0.0001$), as well as in the CPT ΔMBF between group 1 vs. group 2 ($p = 0.0013$), group 1 vs. group 4 ($p < 0.0001$) and between group 2 vs. group 4 ($p < 0.0001$). ΔMBF values less than 1 also showed a significant difference between groups 1 and 2 ($p<0.0001$) (Table 2)

DISCUSSION

In our study, most INOCA patients (n=79) evaluated with CZT-SPECT had an abnormal vasodilator response with CPT, which demonstrates an endothelium-dependent MVD. In addition, patients in group 2 had decreased MFR after dipyridamole stress test,

showing a dual pathophysiological component in this subgroup (Figure 1).

The use of both stressors in our patients allowed the evaluation of different possible pathophysiological mechanisms of MVD: endothelium-dependent or smooth muscle-dependent. The CPT was useful to detect MVD in most cases, even in patients with normal vasodilator response to dipyridamole, as patients in group 1.

The MBF response to CPT was abnormal not only because it did not increase by 50% as expected, but also because the MBF decreased compared with the baseline value in 38 patients, which could be interpreted as an endothelium-dependent microvascular vasospasm (Figure 2). Twelve of these 38 patients (32%) belonged to group 1 and 26 patients (68%) to group 2 ($p < 0.0001$), which could reflect that the microvascular involvement was greater in the latter group of patients, with both functional and structural impairment.

We failed to establish the clinical and methodological characteristics of the 14 patients with normal ΔMBF with CPT and normal MFR with dipyridamole stress test to explain a difference with the rest of the patients. However, it could be related with the presence of vasospastic angina, not detectable by this method, or to other causes of chest pain, such as neuropathic pain. We can only mention as a distinctive finding that this group of patients were all medicated with statins.

We did not perform intracoronary injection of acetylcholine or ergonovine during the index invasive coronary angiography in any of our patients; therefore, we cannot affirm that MVD is the only pathophysiological mechanism involved, since a small percentage of patients with this syndrome may present vasospastic angina associated with microvascular angina. (8,9)

The current evidence demonstrates that MVD is present in the early stages of atherogenesis due to structural and functional changes that occur in the walls of arterioles and intramural capillaries, often related with the presence of cardiovascular risk factors (CVRF). (7)

CVRF increase reactive oxygen species production, leading to endothelial dysfunction. In consequence, the release of vasodilator substances such as nitric oxide is reduced, resulting in a reduction in smooth muscle cell relaxation. Thus, the assessment of the endothelium-dependent vasodilator response with CPT could detect earlier stages of MVD even if smooth

Table 2. Hemodynamic values and results obtained by groups.

	Group 1 (n = 40)	Group 2 (n = 39)	Group 4 (n = 14)
Rest:			
HR (bpm)	65 (58-75)	64 (59-70)	62 (59-71)
SBP (mm Hg)	130 (120-135)	130 (120-130)	130 (120-140)
DBP (mm Hg)	80 (80-80)	80 (80-80)	80 (70-90)
RPP	7975 (7450-10010)	8160 (7200-9100)	8305 (7080-9230)
LVEF (%)	75 (68-83)	71 (66-71)	70 (67-77)
MBF (mL/min/g)	1,05 (0,81-1,29)	1,18 (0,93-1,32) *vs G4	0,71 (0,66-1,15)
Dipyridamole:			
HR (bpm)	80 (68-93)	74 (68-80)	75 (64-86)
SBP (mm Hg)	120 (117-130)	130 (120-140)	125 (110-140)
DBP (mm Hg)	80 (75-80)	80 (80-80)	80 (70-80)
RPP	9615 (8425-11850)	9000 (8360-10200)	9030 (7920- 10360)
LVEF (%)	73 (70- 82%)	74 (70-80%)	75 (70-81)
MBF (mL/min/g)	2.69 (2.15-3.32) †vs G2 *vs G4	1.84 (1.49-2.07) *vs G4	2.09 (1.72-2.84)
CPT:			
HR (bpm)	70 (61-80)	72 (67-78)	70 (66-80)
SBP (mm Hg)	120 (110-130)	120 (120-130)	125 (120-140)
DBP (mm Hg)	80 (75-80)	80 (70-80)	80 (80-80)
RPP	8400 (7250-9610)	8760 (8040-9620)	9045 (8260-9940)
LVEF (%)	74 (69-81)	72 (67-77)	73 (69-80)
MBF (mL/min/g)	1.04 (0.83-1.59) †vs G4	0.99 (0.77-1.23) †vs G4	1.70 (1.19-1.96)
MFR	2.58 (2.13-3.26)	1.56 (1.41-1.69) †vs G1 and G4	2.53 (2.25-3.18)
ΔMBF	1.08 (0.95-1.26) * vs G2, †vs G4	0.87 (0.72-1.11) †vs G4	1.79 (1.54-2.02)
ΔMBF <1	0.87 (0.71-0.94)	0.79 (0.69-0.87) †	-

*p<0.05 †p < 0.0001
All the results expressed as median and interquartile range.
HR: heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure
RPP: rate pressure product (maximum SBP x maximum HR); LVEF: left ventricular ejection fraction; MBF: myocardial blood flow; MFR: myocardial flow reserve; ΔMBF: changes in myocardial blood flow ; CPT: cold pressor test.

muscle-dependent vasodilator response is normal. Moreover, vascular impairment will be greater as the exposure time to these CVRF increases. (7)

The population evaluated in our study had one or more CVRF. Of the 19 patients without any traditional CVRF, 9 were postmenopausal women. Several studies with PET demonstrated that the reduction in estrogen levels in postmenopausal women predisposes to a reduction in coronary artery vasodilation similar to that observed in premenopausal women with diabetes. (19-21)

The clinical manifestations of MVD include typical exercise-induced angina, angina in the immediate recovery after exercise or even at rest, atypical chest pain or angina equivalents such as exercise-induced dyspnea. Because of this wide variety of symptoms, different criteria were established for the diagnosis of microvascular angina, considering not only the clinical aspects, but also the abnormal values of coronary vasoreactivity according to different methods, as MBF and MFR. (9, 21,22)

Although MVD can worsen the prognosis in patients with or without obstructive coronary artery disease, particularly in the presence of symptoms, there is currently no specific treatment. This leads to multiple combinations of different drugs, some of them with no clear evidence of benefit, as in our patients. (23,24)

The importance of knowing the pathophysiological mechanisms involved in INOCA syndromes lies in tailoring the treatment to each particular case. Several studies have demonstrated that the quality of life of these patients improves when the different mechanisms are evaluated and treatment is based on the pathophysiological cause involved. The CormicA trial evaluated invasive coronary function testing at time of

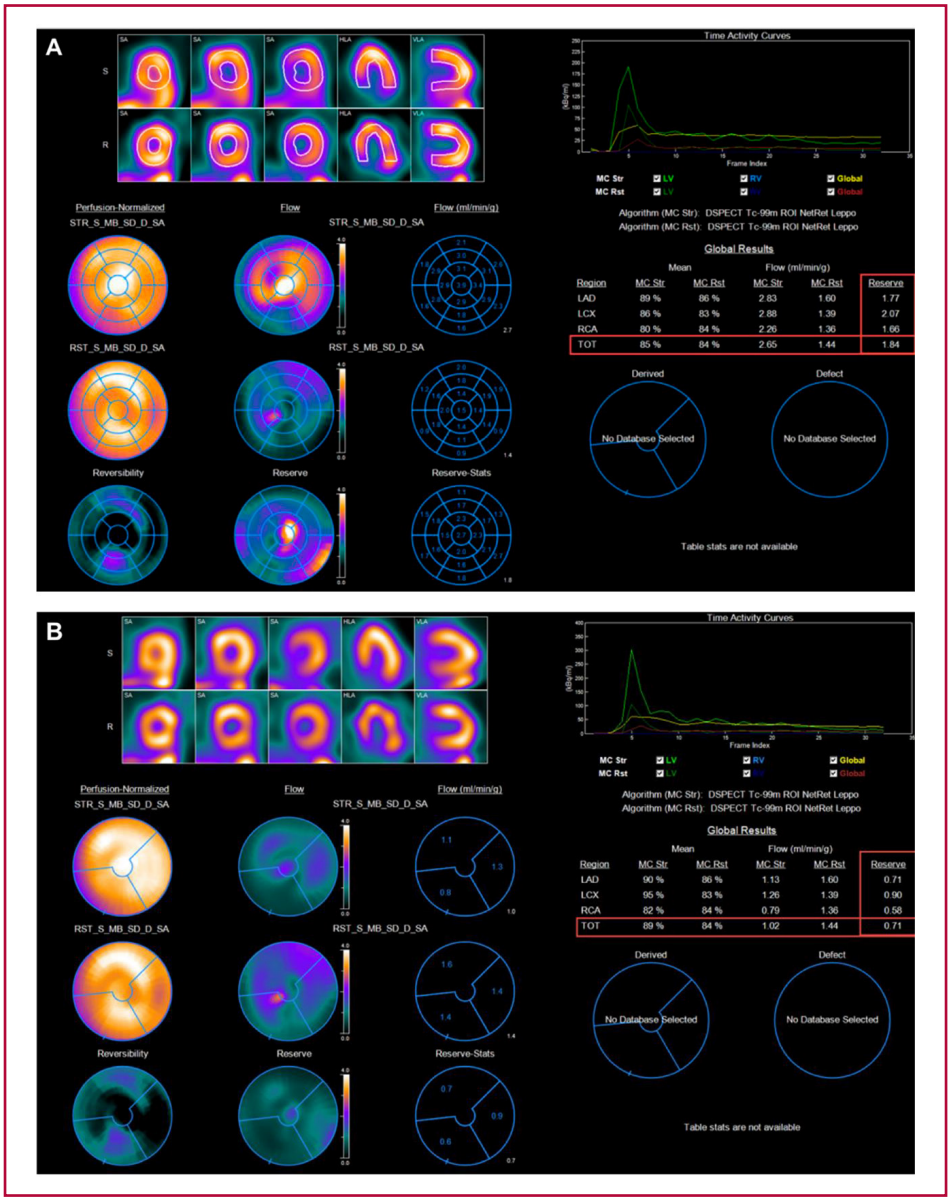

Fig. 1. Patient with smooth-muscle dependent and endothelium-dependent MVD. **A.** Dipyridamole (MFR: 1.84). **B.** Cold pressor test (ΔMBF: 0.71)

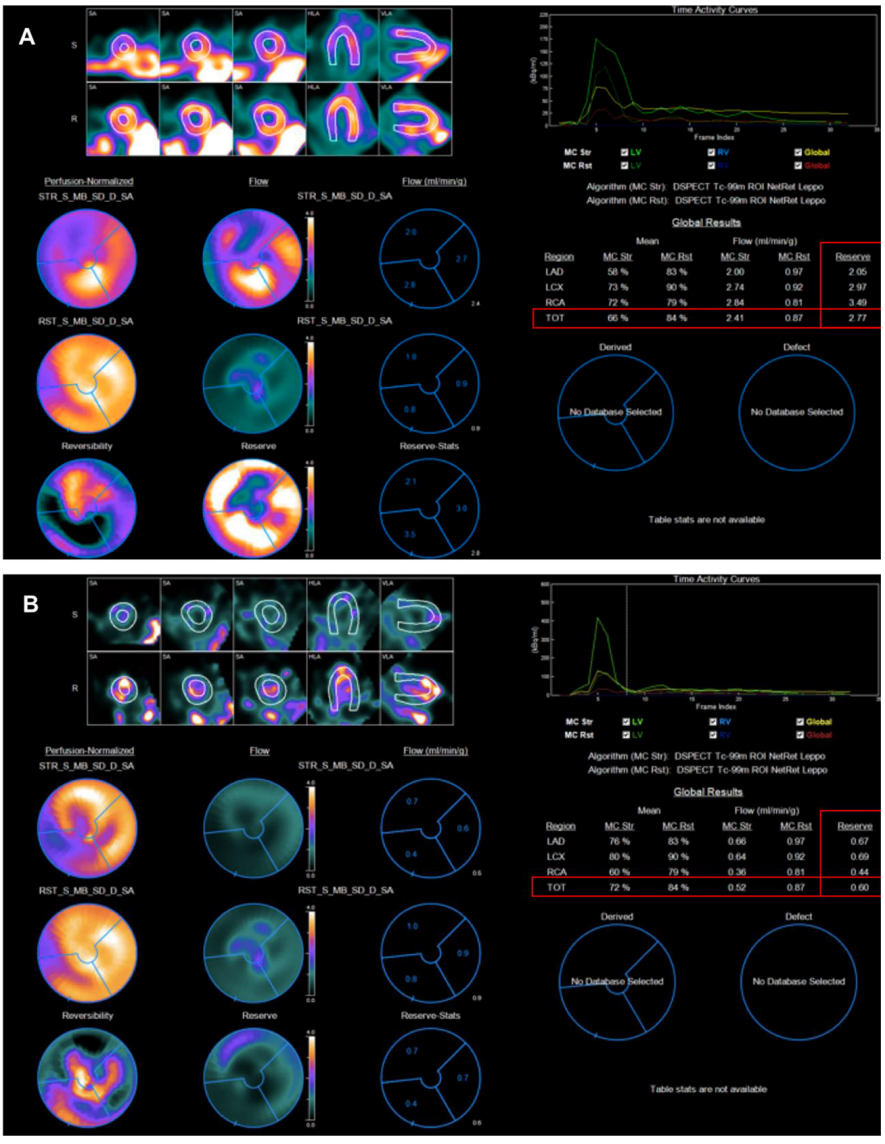

Fig. 2. Patient with endothelium-dependent microvascular vasospasm. **A.** Dipyridamole (MFR: 2.77) **B.** Cold pressor test (ΔMBF: 0.60)

the index diagnostic angiography in 151 patients with INOCA randomized to medical therapy guided by an interventional diagnostic procedure (group 1) versus control group (group 2). Quality of life and clinical events at 1 year were compared in both groups. Group 1 showed marked and sustained clinical improvement and better quality of life compared with the control group. (2,25).

We present a simple, noninvasive diagnostic algorithm, which includes the evaluation of the two possible mechanisms of coronary vasoreactivity that produce MVD in patients with INOCA (Figure 3).

Study limitations:

The sample size is small and the method used is relatively new, although its validation and reproducibility are currently accepted. Future studies in larger populations will be necessary to evaluate the usefulness of this approach in patients with INOCA.

Although invasive functional coronary testing is not routinely performed in our country in patients with INOCA, we consider that such evaluation should be performed during diagnostic coronary angiography to rule out epicardial coronary vasospasm as a probable or concomitant cause.

CONCLUSIONS

In our experience, the use of CZT-SPECT devices detected MVD in 85% of patients with INOCA.

The use of both stress tests allowed the evaluation of two different pathophysiological mechanisms of MVD: endothelium-dependent or smooth muscle-dependent.

Evaluation with CPT should be included in the

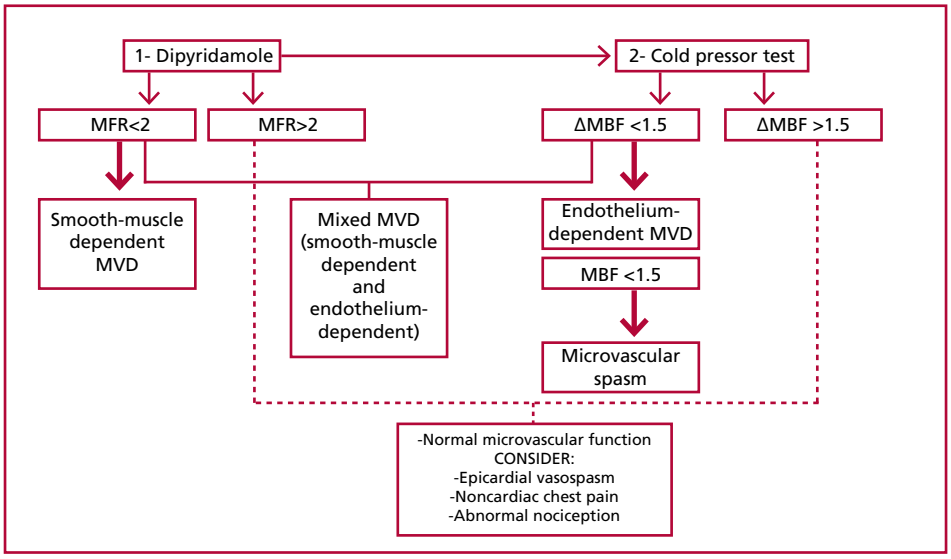

Fig. 3. Diagnostic algorithm proposed to for non-invasive assessment of microvascular function in patients with INOCA.

MFR: myocardial flow reserve. Δ MBF: myocardial blood flow variation. MVD: microvascular dysfunction

noninvasive assessment of INOCA patients, as MVD may be present even with normal MFR with dipyridamole stress test.

Conflicts of interest

None declared.

(See authors' conflict of interests forms on the web/Additional material).

REFERENCES

1. Jespersen L, Abildstrom SZ, Hvelplund A, Madsen JK, Galatius S, Pedersen F, et al. Burden of hospital admission and repeat angiography in angina pectoris patients with and without coronary artery disease: a registry-based cohort study. PLoS One. 2014;9:e93170. https://doi.org/10.1371/journal.pone.0093170

2. Ford TJ, Standley B, Good R, Rocchiccioli P, McEntegart M, Watkin S, et al. Stratified Medical Therapy Using Invasive Coronary Function Testing in Angina: The CorMicA Trial. J Am Coll Cardiol 2018;72(23 Pt A):2841-55. https://doi.org/10.1016/j.jacc.2018.09.006

3. Pasupathy S, Air T, Dreyer RP, Tavella R, Beltrame JF. Systematic review of patients presenting with suspected myocardial infarction and nonobstructive coronary arteries. Circulation. 2015; 131:861–70. https://doi.org/10.1161/CIRCULATIONAHA.114.011201

4. Arrebola-Moreno A, Laclaustra M, Kaski JC. Noninvasive assessment of endothelial function in clinical practice. Rev Esp Cardiol 2012;65:80-90. https://doi.org/10.1016/j.rec.2011.10.004

5. Pepine CJ, Anderson RD, Sharaf BL, Reis SE, Smith KM, Handberg EM, et al. Coronary microvascular reactivity to adenosine predicts adverse outcome in women evaluated for suspected ischemia results from the National Heart, Lung and Blood Institute WISE (Women's Ischemia Syndrome Evaluation) study. J Am Coll Cardiol 2010;55:2825–32. https://doi.org/10.1016/j.jacc.2010.01.054

6. Schelbert H. Anatomy and physiology of coronary blood flow. J Nucl Cardiol 2010;17: 545-54. https://doi.org/10.1007/s12350-010-9255-x

7. Taqueti V, Di Carli M. Coronary microvascular disease pathogenic mechanism and therapeutic options: JACC state-of the-Art Review. J Am Coll Cardiol 2018;72:2625-41. https://doi.org/10.1016/j.jacc.2018.09.042

8. Ford T, Ong P, Sechem U, Beltrame J, Camici P, Crea F, et al. Assesment of Vascular Dysfunction in Patients Without Obstructive Coronary Artery Disease. Why, How and When. JACC Cardiovasc Interv 2020;13:1847-64. https://doi.org/10.1016/j.jcin.2020.05.052

9. Kunadian V, Chieffo A, Camici P, Berry C, Escaned J, Maas A, et al. An EAPCI Expert Consensus Document on Ischaemia with Non-Obstructive Coronary arteries in collaboration with European Society of Cardiology working group on Coronary Pathophysiology and Microcirculation Endorsed by Coronary Vasomotor Disorders International Study Group. Eur Heart J 2020;41:3504-20. https://doi.org/10.1093/eurheartj/ehaa503

10. Camici P, Mamati G, Rimoldi O. Coronary microvascular dysfunction: mechanisms and assessment. Nat Rev Cardiol 2015;12:48-62. https://doi.org/10.1038/nrcardio.2014.160

11. Agostini D, Roule V, Nganoa C, Roth N, Baavour R, Parienti J, et al. First validation of myocardial flow reserve assessed by dynamic 99mTc- sestamibi CZT-SPECT camera: head to head comparison with 15O-water PET and fractional flow reserve in patients with suspected coronary artery disease. The WATERDAY study. Eur J Nucl Med Mol Imaging 2018;45:1079-90. https://doi.org/10.1007/s00259-018-3958-7

12. Acampa W, Zampella E, Assante R, Genova A, De Simini G, Mannarino T, et al. Quantification of myocardial perfusion reserve by CZT-SPECT: A head to head comparison with 82Rubidium PET imaging. J Nucl Cardiol 2021;28:2827-39. https://doi.org/10.1007/s12350-020-02129-w

13. Wells G, Marvin B. Measuring SPECT myocardial blood flow at the University of Ottawa Heart Institute. J Nucl Cardiol. 2021;28:1298-303. https://doi.org/10.1007/s12350-020-02102-7

14. Mathew R, Bourque J, Salerno M, Kramer C. Cardiovascular Imaging Techniques to Assess Microvascular dysfunction. JACC Cardiovasc Imaging 2020;7:1577-90. https://doi.org/10.1016/j.jcmg.2019.09.006

15. Camici PG, Crea F. Coronary microvascular dysfunction. N Engl J Med 2007;356: 830-40. https://doi.org/10.1056/NEJMra061889

16. Prior JO, Schindler TH, Facta AD, Hernandez-Pampaloni M, Campisi R, Dahlbom M, et al. Determinants of myocardial blood flow response to cold pressor testing and pharmacologic vasodilation in healthy humans. Eur J Nucl Med Mol Imaging 2007;4:20-7. https://doi.org/10.1007/s00259-006-0193-4

17. Schindler TH, Nitzsche EU, Olschewski M, Brink I, Mix M, Prior J. PET-measured responses of MBF to cold pressor testing correlate with indices of coronary vasomotion on quantitative coronary angiography. J Nucl Med 2004;45:419-28.

18. Levy BI, Heusch G, Camici PG. The many faces of myocardial ischemia and angina. Cardiovasc Res 2019;115:1460-70. https://doi.org/10.1093/cvr/cvz160

19. Rahman H, Demir O, Khan F, Ryan M, Ellis H, Mills M, et al. Physiological Stratification of Patients with Angina due to Coro-

nary Microvascular Dysfunction. J Am Coll Cardiol 2020;75:2538-49. https://doi.org/10.1016/j.jacc.2020.03.051
20. Murthy Vl, Naya M, Taqueti VR, Foster CR, Gaber M, Hainer J, et al. Effects of sex on coronary microvascular dysfunction and cardiac outcomes. Circulation 2014;129:2518-27. https://doi.org/10.1161/CIRCULATIONAHA.113.008507
21. Campisi R, Marengo F. Coronary microvascular dysfunction in women with nonobstructive ischemic heart disease as assessed by positron emission tomography. Cardiovasc Diagn Ther. 2017;7:196-205. https://doi.org/10.21037/cdt.2017.04.08
22. Schindler T, Dilsizian V. Coronary microvascular Dysfunction. Clinical considerations and noninvasive diagnosis. JACC Cardiovasc Imaging 2020;13:140-55. https://doi.org/10.1016/j.jcmg.2018.11.036
23. Ong P, Camici P, Beltrame J, Crea F, Shimokawa H, Sechtem U, Kaski J, Merz C, Coronary Vasomotion Disorders International Study Group (COVADIS). International standardization of diagnostic criteria for microvascular angina. Int J Cardiol 2018;250:16-20. https://doi.org/10.1016/j.ijcard.2017.08.068
24. Kenkre TS, Malhotra P, Johnson BD, Handberg EM, Thompson DV, Marroquin OC. Ten-Year Mortality in the WISE Study (Women's Ischemia Syndrome Evaluation). Circ Cardiovasc Qual Outcomes 2017;10:e003863. https://doi.org/10.1161/CIRCOUTCOMES.116.003863
25. Ford T, Stanley B, Sidik N, Good R, Rocchiccioli P, McEngart M, et al. 1-Year Outcomes of Angina Management Guided by Invasive Coronary Function Testing (CorMicA). JACC Cardiovasc Interv 2020;13:33-45. https://doi.org/10.1016/j.jcin.2019.11.001

Appendix

Comparison between the resting hemodynamic values (day 1 vs. day 2) across the different groups

	Rest Day 1	Rest Day 2	p
Group 1			
HR (bpm)	65 (58.5-75.5)	67 (58.5-76)	ns
SBP (mm Hg)	130 (120-135)	130 (120-130)	ns
DBP (mm Hg)	80 (80-80)	80 (75-80)	ns
RPP	7975 (7450-10010)	8140 (7285-9755)	ns
Group 2			
HR (bpm)	64 (59-70)	64 (59-68)	ns
SBP (mm Hg)	130 (120-130)	130 (120-140)	ns
DBP (mm Hg)	80 (80-80)	80 (80-80)	ns
RPP	8160 (7200-9100)	8260 (7370-9100)	ns
Group 4			
HR (bpm)	62 (59-71)	68 (62-77)	ns
SBP (mm Hg)	130 (120-140)	125 (120-140)	ns
DBP (mm Hg)	80 (70-90)	80 (80-80)	ns
RPP	8305 (7080-9230)	8400 (7680-10140)	ns

HR: heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure; RPP: rate pressure product