

ORIGINAL ARTICLE

Minimally Invasive Cardiac Surgery versus Sternotomy - Pain Investigation

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Abstract

Background: Treatment of postoperative (PO) pain is essential after surgery, as it contributes to a faster rehabilitation. Assessment of PO pain after minimally invasive (MI) surgery has not been regularly addressed, especially when compared with median sternotomy (MS).

Objective: This study aims to evaluate the intensity of thoracic pain in the PO period in patients subjected to MI surgery and MS.

Methods: This study compared the intensity of thoracic pain in 34 patients subjected to minimally invasive (MI; n = 17) and median sternotomy (MS; n = 17) from June 2015 to June 2016. The intensity and sites of pain in the PO period, assessed using the visual numeric pain scale, and the need for pain medications were analyzed using the Student's t-test and the z test, with confidence level of 95% ($p < 0.05$).

Results: Almost all patients reported pain on the third PO day (MS = 94.1% and MI = 88.2%; $p = 0.5410$). On the seventh PO day, there were significantly more patients free of pain in the group of patients subjected to the MI procedure (MS = 94.1% and MI = 64.7%; $p = 0.0341$). also, these patients reported fewer pain sites (3rd PO day: MS = 3.2 ± 1.5 ; MI = 1.5 ± 1.2 ; $p = 0.001$; 7th PO day: MS = 3.1 ± 1.4 ; MI = 0.9 ± 0.9 ; $p = 0.000$). Patients undergoing MS reported higher pain intensity and longer lasting pain (3rd PO: MS = 4.8 ± 2.2 ; MI = 3.0 ± 1.6 ; 7th PO: MS = 5.3 ± 2.0 ; MI = 1.2 ± 1.3 ; $p = 0.001$), with no difference in pain intensity between the third and the seventh PO days ($p = 0.4931$). In addition, patients subjected to MI procedure had a significant decrease in pain intensity from the third to the seventh PO days ($p = 0.001$).

Conclusion: According to these results, we concluded that a MI procedure leads to lower intensity of pain in the PO period (from the third PO day on) when compared to a MS; also, patients undergoing MI patients reported fewer pain sites. (Int J Cardiovasc Sci. 2020;33(1):24-33)

Keywords: Minimally Invasive Surgical Procedures; Cardiovascular Surgical Procedures; Sternotomy; Postoperative Care.

Introduction

Minimally invasive (MI) cardiac surgery is a safe procedure with similar mortality and morbidity, but better surgical outcomes compared with conventional sternotomy (MS) in some groups of patients.¹⁻⁴ The potential benefits of MI procedures include better stability of the sternum in the postoperative period, with implications on deep infection prevention, improvement of respiratory function, mobility and

bleeding.⁵ The MI approach was introduced to reduce surgical trauma with better cosmetic results; this approach is currently applied to procedures including valve and septal defect surgeries.^{6,7}

Pain has been shown to be one of the primary sources of concern in surgical patients,⁸ even though it is expected in the postoperative (PO) period. Inadequate management of pain can have profound clinical (deep vein thrombosis, pulmonary embolism, coronary ischemia, myocardial infarction, pneumonia, and poor wound healing) and

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DOI: 10.5935/2359-4802.20190048

Manuscript received on March 20, 2018, revised manuscript on December 02, 2018, accepted on February 17, 2019.

psychological (insomnia) implications that increase morbidity and mortality.^{8,9}

In the last few years, there has been a significant increase in knowledge about PO pain and tissue trauma that influence the choice for MI surgical procedures.^{1,9} Although a median sternotomy (MS) remains the main access in cardiac surgeries, the intercostal access combined with a MI approach has been progressively used.^{2,3,5,6,10} Systematic assessment and treatment of pain is essential after cardiac surgery and can contribute to a faster recovery and rehabilitation of patients.¹¹ Some studies have reported less PO pain and shorter in-hospital stays after a MI procedure,^{1,3,12–14} but the literature lacks a comparative study between MS and MI procedures regarding pain intensity (PI). This prospective observational study aims to evaluate the PO thoracic PI by comparing patients subjected to a MI and MS procedures. We evaluated if patients with valve and septal defects subjected to MI procedure have less PO pain compared with those undergoing MS.

Materials and methods

Study population

We evaluated PO thoracic pain in 34 patients who were subjected to MS or MI procedure between June 2015 and June 2016. The Institutional Review Board of the university approved this prospective observational comparative study (approval number 1104.606). We included symptomatic patients presenting with mitral valve (MV) disease or an atrial septal defect (ASD). Exclusion criteria were patients older than 60 years old, patients with body mass index (BMI) (for the MI group only) greater than 32 kg/m², chronic obstructive lung disease, previous heart or thoracic interventions, renal failure, interstitial or inflammatory lung disease, thoracic deformities, mitral valve or aortic calcifications, systolic pulmonary pressure greater than 80 mmHg, coronary artery disease, severe tricuspid valve insufficiency, femoral vessel calcification, femoral artery smaller than 5 mm, moderate or severe aortic valve insufficiency, requirement for re-intervention for any cause after the end of surgical procedure, communication impairments, pain syndrome before the procedure, and patients who withdrew consent at any moment throughout the study. The surgical access technique was chosen according to the pathoanatomical characteristics of each patient and the recommended surgical protocol. The patients were divided in two groups: MS (n = 17) and MI (n = 17).

Written informed consent was obtained from all patients before treatment.

Surgical technique

The surgical procedures were all carried out by the same surgeon (JVC). The patients underwent surgical interventions under general anesthesia and a cardiopulmonary bypass with moderate hypothermia and cold crystalloid cardioplegic arrest.

Description of the MS procedure: a main incision of twenty centimeters was performed followed by bone division from the manubrium to xiphoid. A sternal retraction was made to provide a 12-centimeter working space, with full vision of the heart and vessels. The second incision was made two centimeters below main incision for placement of a chest tube drain. Arterial perfusion was achieved by direct cannulation of the ascending aorta. Systemic venous return was achieved with two individual caval cannulas. The aortic occlusion was made by direct clamping of the aorta. Usual techniques and instruments were applied for the procedure.

Minimally invasive procedure: main surgical access was a right minithoracotomy (5 centimeters) into the fourth intercostal space. A periareolar incision and a submammary incision were made for men and women, respectively, between the midclavicular and the anterior axillary line for valve surgery and on the midclavicular line for atrial septal defect closure. The main incision was enlarged with a wound protector and soft tissue retractor (ALEXIS™), and a steel retractor was used as necessary during the procedure. Three auxiliary 5 mm ports were placed in the anterior axillary line. In the second space, the port was used for placement of a transthoracic aortic cross-clamp to obtain an aortic occlusion. In the fourth space, a 30-degree high definition camera was placed. In the seventh space, the port was used for atrial venting and CO₂ flow in the operative field at 2 L/min. This port was also used for placement of a 24 F Blake™ drain at the end of the procedure. An atrial lift retractor system was positioned at the fourth intercostal space near the sternum, when required. Femoral arterial perfusion was performed using a cannula adjusted for patient's body surface and internal diameter of the femoral artery. The cannula was inserted by direct puncture using the Seldinger's technique. The vacuum assisted venous return with a single right femoral venous cannula was associated or not with a right jugular venous cannula. Patients

were monitored by a transesophageal echocardiogram. Specific instruments for minimally invasive surgery were used.

Management of postoperative thoracic pain

The same protocol for induction and maintenance of anesthesia was followed in all patients. All of them were intubated with a single lumen endotracheal tube that remained until extubation criteria were met. In the first 48 hours, the patients received 1 g of dipyron every 6 hours and 100 mg of tramadol every 8 hours for pain relief. According to each patient's needs, 2 mg of morphine was administered. After this initial management, 500 mg of dipyron were administered orally every 6 hours with or without 50 mg of tramadol, if necessary.

Thoracic pain evaluation

Thoracic pain was assessed using the Visual Numeric Scale (VNS).¹⁵ Patients were instructed on the use of the VNS before the surgery. In the VNS, pain is rated from zero to 10, where zero indicates no pain and 10 the maximum pain level tolerated by the patient. Pain data were obtained on the third and seventh PO days. All patients were free of chest tubes at the moment of data collection. Patients were asked about the presence of pain, and for positive responses, pain location was registered, and PI rated on the scale. This was repeated for each pain site, if there was more than one. The pain drugs were then verified and registered.

Statistical analysis

Statistical analyses were performed using the Statistical Package for Social Science (SPSS) software for WindowsTM, version 17.0 or the Statistica 12. A power of 80% was calculated for 34 patients (17 in each group). Normality of the data was determined using the Kolmogorov-Smirnov test, and homogeneity of the variance was assured by the Levene's test. Results were expressed as mean and standard deviation (mean \pm SD) for continuous variables; categorical data were summarized by frequencies and percentages and compared by a z test for two proportions. Data were parametric and compared using paired (within group comparisons) and unpaired (between group comparison) Student's t test, with a confidence level of 95% ($p < 0.05$).

Results

Demographic and clinical data, including the presence of cardiovascular risks factors in the study groups are described in Table 1. Most patients were women, and age was not different between the groups. Weight and BMI were different between the groups, probably due to the non-inclusion of patients with BMI > 32 kg/m² in the MI group. The prevalence of comorbidities was not relevant, except for systemic arterial hypertension. Mitral valve insufficiency was the main diagnosis, followed by *ostium secundum* atrial septal defects, mitral valve stenosis, and *ostium primum* atrial septal defects.

Patients were subjected to mitral valve surgical repair or replacement (MS $n = 11$ and MI $n = 9$) or surgical closure of an atrial septal defect (MS- $n = 6$ and MI- $n = 8$). Procedures for mitral correction included bioprosthetic replacement (7 in the MS group and 8 in the MI group) and mitral valve repair (4 in the MS group and 1 in the MI group). Valve resection, valve reconstruction, and semi-rigid ring annuloplasty were the main procedure for mitral repair. In the MS group, one patient required a Neochord placement, and no patient was subjected to pulmonary vein isolation. Regarding the procedures for atrial septal defect closure, there were one suture closure and five patch closures in the MS group and three suture closures and five patch closures in the MI group.

Table 2 lists the PO data for the MS and MI groups. The MS procedure time was shorter than MI procedures. No difference was observed in mean aortic cross-clamping time or cardiopulmonary bypass time between the groups. Despite longer procedural times, MI patients needed less intensive care unit time for recovery in comparison with MS patients (Table 2). Mean hospital stay after the procedure was longer in the MS than in MI group. None of the patients had major complications, stroke or death after the surgery.

The PO pain evaluation indicated that most of the patients reported pain on the third PO day (MS = 94.1% and MI = 88.2%; $p = 0.5410$). On the seventh PO day, significantly more patients were free of pain in the MI group compared with the MS group (MS = 94.1% and MI = 64.7%; $p = 0.0341$). The patients in the MI group reported fewer pain sites than the patients in the MS group (Figure 1) on the third (MS = 3.2 ± 1.5 ; MI = 1.5 ± 1.2 ; $p = 0.001$) and seventh (MS = 3.1 ± 1.4 ; MI = 0.9 ± 0.9 ; $p = 0.000$) PO day.

Table 1 - Demographic and clinical characteristics of patients enrolled in the study

Characteristics	MS group (n = 17)	MI group (n = 17)	p-value
Age (years)	47.60 ± 15.10	40.10 ± 13.90	0.100
Gender (n/%)			
Male	6/35.30%	3/17.60%	0.200
Female	11/64.70%	14/82.4%	
Weight (kg)	71.90 ± 13.40	60.70 ± 10.1	0.010*
Hight (cm)	1.62 ± 0.09	1.61 ± 0.06	0.600
BMI (kg/m ²)	27.10 ± 3.70	23.30 ± 3.90	0.007*
Diagnosis (n/%)			
Mitral insufficiency	10/58.80%	6/35.30%	0.400
OSASD	4/23.50%	6/35.30%	
OPASD	1/5.90%	1/5.90%	
SVASD	1/5.90%	0/0.00%	
PFO	0/0.00%	1/5.90%	
Mitral stenosis	1/5.90%	3/17.60%	
Comorbidities (n/%)			
Smoking	1/5.90%	1/5.90%	1.000
Drinking	0/0.00%	2/11.80%	0.300
Systemic arterial hypertension	10/58.80%	3/17.60%	0.010*
Diabetes mellitus	1/5.90%	1/5.90%	1.000
Stroke	1/5.90%	0/0.00%	1.000
Transient ischemic attack	0/0.00%	1/5.90%	1.000
COPD	0/0.00%	0/0.00%	
Dyslipidemia	1/5.90%	0/0.00%	1.000
Thrombolysis	1/5.90%	0/0.00%	1.000

MS: median sternotomy; MI: minimally invasive; n: number of patients; %: frequency; OSASD: ostium secundum atrial septal defect; OPASD: ostium primum atrial septal defect; SVASD: sinus venosus atrial septal defect; PFO: patent foramen ovale; COPD: chronic obstructive pulmonary disease. * $p < 0.05$ between groups by unpaired Student's *t*-test for continuous variables and *z* test for categorical variables.

The main sites of pain were those related to the surgical incision site. The upper area of the sternum (around the manubrium) was more painful for the MS group and the right submammary region was more painful for the MI group (Table 3). PI was different between the groups. The MS group had more intense pain than the MI group on the third PO day and on the seventh PO day. The MS group showed a mean of 5.3 ± 2.0 of maximal PO thoracic PI three days after the procedure, which was

not significantly decreased on the seventh day after the surgical procedure (4.8 ± 2.2 ; $p = 0.4931$). The maximum PI reported by MI patients was significantly ($p = 0.001$) decreased from the third (3.0 ± 1.6) to the seventh PO day (1.2 ± 1.3). The comparison between groups demonstrated that the postoperative PI was higher and lasted longer in the MS group than in the MI group ($p = 0.001$) (Figure 2).

These data were in accordance with the medical necessity to control the symptoms. On the 3rd PO day, all

Table 2 - Postoperative data of the patients

	MS group (n = 17)	MI group (n = 17)	p-value
Surgery (n/%)			
Mitral valve replacement	7/41.2	8/47.1	
Atrial septal defect closure	6/35.3	8/47.1	0.300
Mitral reconstruction	4/23.5	1/5.9	
Procedure time (minutes)	194.7 ± 60	251.0 ± 52.3	0.006*
Extracorporeal circulation time (minutes)	101.8 ± 35.4	118.4 ± 26.1	0.100
Cross-clamp time (minutes)	78.2 ± 33.5	88.5 ± 21.9	0.300
ICU time (hours)	52.2 ± 16.8	33.7 ± 12.7	0.001*
Hospital length of stay (days)	6 ± 1.4	3.5 ± 0.9	0.001*

MS: median sternotomy; MI: minithoracotomy; n: number of patients; %: frequency; ICU: intensive care unit. * $p < 0.05$ between groups, unpaired Student's *t*-test for continuous variables and *z* test for categorical variables; 95% confidence level.

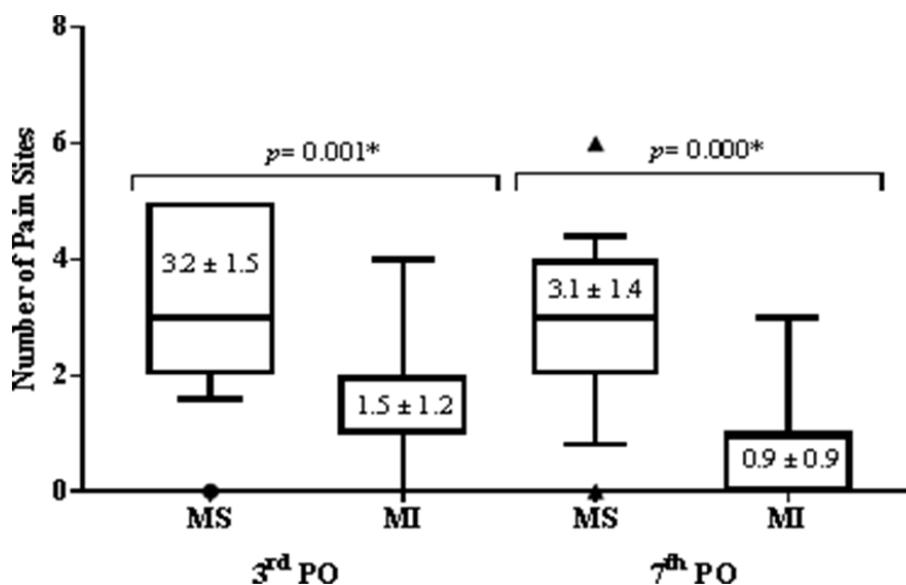


Figure 1 - Number of pain sites in patients undergoing minimally invasive cardiac surgery and median sternotomy on the third and seventh postoperative days. Box plot of the number of pain sites reported by 34 patients (17 patients subjected to minimally invasive cardiac surgery, MI, and 17 to median sternotomy, MS) on the third (3rd) and seventh (7th) postoperative days. Circles and triangles represent outliers.

* $p < 0.05$ between groups, unpaired Student's *t*-test at 95% confidence level.

patients in the MS group and 16 patients in the MI group were receiving pain medications ($p = 0.3052$). On the 7th PO day, 16 patients in the MS group and only six in the MI group were receiving pain medications ($p = 0.0003$).

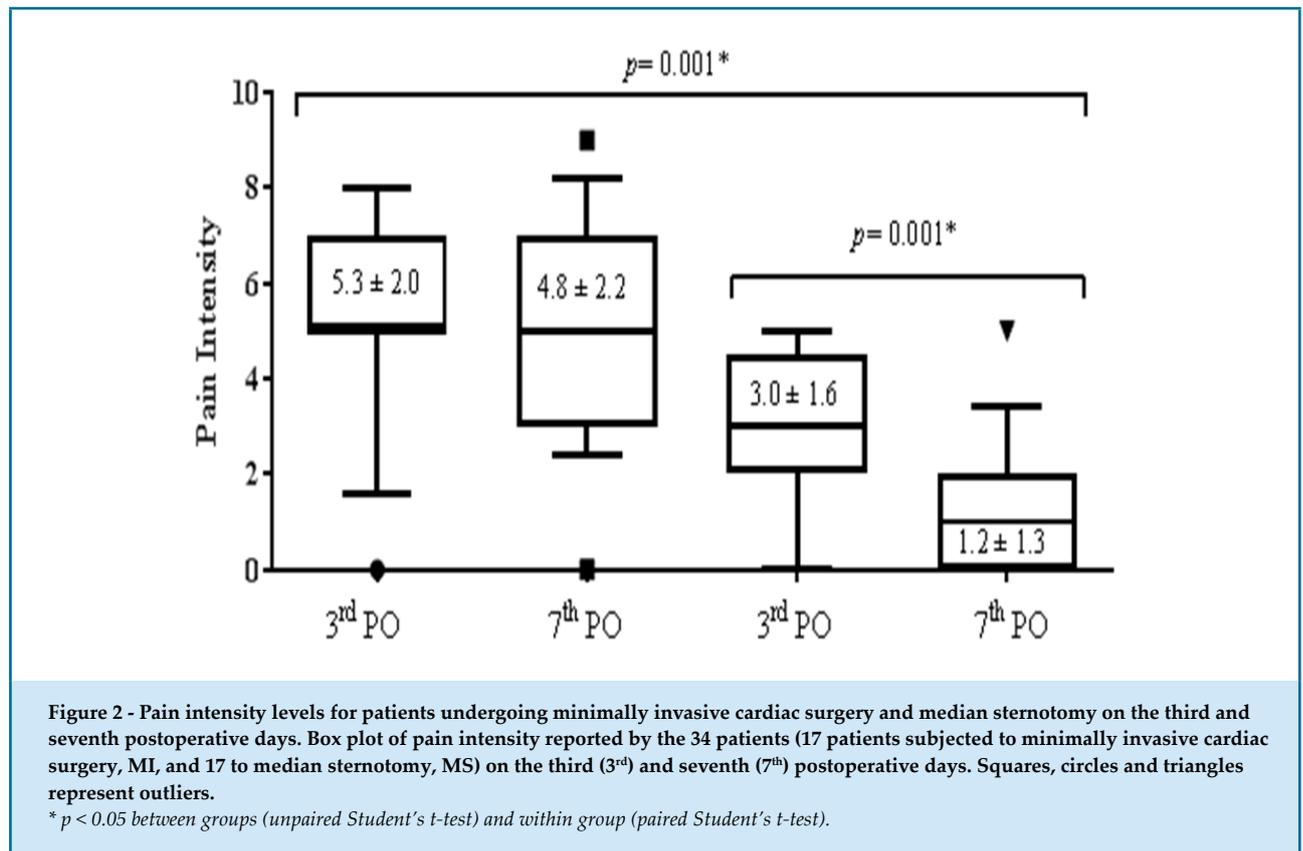
Discussion

The present study showed that, in patients undergoing surgical treatment for mitral valve and septal defects,

Table 3 - Pain intensity according to patients' body area

	MS group (n = 17)		p-value*	MI group (n = 17)		p-value*
	3 rd PO	7 th PO		3 rd PO	7 th PO	
Body region						
Right periclavicular	0.7 ± 2.0	0.7 ± 2.0	1.000	0.0	0.0	1.000
Left periclavicular	0.4 ± 1.6	0.2 ± 1.2	0.300	0.0	0.0	1.000
Intersection between 2 nd right intercostal space and right anterior axillary line	0.0	0.0	1.000	0.4 ± 1.0	0.0	0.100
Intersection between 4 th right intercostal space and right anterior axillary line	0.0	0.0	1.000	0.0	0.0	1.000
Intersection between 7 th right intercostal space and right anterior axillary line	0.0	0.0	1.000	0.2 ± 1.2	0.0	0.300
Periareolar	0.4 ± 1.6	0.2 ± 1.2	0.300	0.5 ± 1.2	0.1 ± 0.5	0.300
Submammary	0.4 ± 1.6	0.2 ± 1.2	0.700	1.3 ± 1.4	0.5 ± 0.9	0.070
Upper sternal	4.0 ± 2.6	2.8 ± 2.0	0.070	0.1 ± 0.7	0.0	0.300
Lower sternal	3.5 ± 2.7	3.0 ± 1.9	0.400	0.1 ± 0.4	0.1 ± 0.7	0.700
Subxiphoid	1.4 ± 2.2	0.2 ± 8.0	0.060	0.0	0.0	1.000
Intersection between 2 nd left intercostal space and left anterior axillary line	0.0	0.0	1.000	0.0	0.0	1.000
Intersection between 4 th left intercostal space and left anterior axillary line	0.0	0.0	1.000	0.0	0.0	1.000
Intersection between 7 th left intercostal space and left anterior axillary line	0.0	0.0	1.000	0.0	0.0	1.000
Right inguinal	0.3 ± 1.4	0.0	0.300	0.0	0.0	1.000
Left inguinal	0.0	0.0	1.000	0.0	0.0	1.000
Posterior cervical	1.0 ± 2.2	0.6 ± 1.9	0.600	0.0	0.0	1.000
Right scapular	2.5 ± 3.1	1.7 ± 3.0	0.500	0.2 ± 0.9	0.0	0.300 [†]
Left scapular	1.4 ± 2.4	2.5 ± 3.3	0.100	0.8 ± 1.6	0.2 ± 0.6	0.100 [†]
Vertebral	0.8 ± 1.9	1.2 ± 2.5	0.600	0.0	0.0	1.000
Right infrascapular	0.0	0.0	1.000	0.2 ± 1.2	0.4 ± 1.1	0.500
Left infrascapular	0.4 ± 1.6	0.0	0.300	0.2 ± 1.2	0.4 ± 1.3	0.300

*Caption: MS: median sternotomy; MI: minithoracotomy; n: number of patients; %: frequency; PO: postoperative; *within group by paired Student's t-test for continuous variables and z test for categorical variables; †p < 0.05 between groups by unpaired Student's t-test for continuous variables and z test for categorical variables; 95% confidence level.*



those subjected to MI procedure had less pain from the third PO day on and fewer sites of pain than the patients who underwent a sternal procedure. Our findings showed that a reduction in PI can lead to better recovery, indicated by shorter ICU and hospital stays as well as a diminished need for pain relief medications.

In the 1990s, MI techniques were initially used in cardiac surgeries.^{7,16-18} Meanwhile, Carpentier et al.,¹⁹ Chitwood et al.,²⁰ Vanermen et al.,²¹ and Mohr et al.,²² established the MI approach for mitral valve surgery, and numerous studies started to report the feasibility, safety and efficacy of these procedures.¹⁻⁵ However, although many studies have evaluated the advantages of MI cardiac procedures, including pain sites and PI, in addition to hospital stay duration,^{1-3,5,6,10,23} none of them performed a systematic comparison between MI and MS procedures regarding PO pain.

In the current investigation, we studied patients with mitral valve disease and patients with septal defects, since these are among the most prevalent cardiovascular diseases among Brazilian adults²⁴ that can be addressed by either MI or MS procedure. The main surgical procedure was valve replacement followed by valve

reconstruction, and the most common cause of valve dysfunction was inflammatory in both groups.

The procedure time was longer in the MI group in comparison to the MS group, as reported in many other studies.^{6,7,10,18,22,25-29} This difference was due to intrinsic characteristics of the MI procedure, which demands a femoral incision for echo-guided cannulation before the insertion of chest ports. Although a longer cardiopulmonary bypass and cross-clamp times may lead to higher mortality and morbidity,³⁰ Raja et al.,⁷ demonstrated that these adverse outcomes were not evident in the MI group. Differently from other studies on MI procedures,^{28,31-33} we observed similar circulatory support and clamp times between the groups. These surgical variables are related to the complexity of the surgical procedure (mainly valve replacement) as well as the surgeon's experience. In addition, the time for cardiopulmonary bypass could be reduced by percutaneous insertion of the cannula. Of note, the higher weight and body surface in the MS group was due to the exclusion criterion of a BMI greater than 32 kg/m² in the MI group. Despite this, we do not believe that BMI is directly related to PI in the PO period, as we do not report any complication related to higher BMIs in this period.

Table 4 - Assessment of pain medications in the postoperative of patients undergoing minimally invasive cardiac surgery and median sternotomy

	MS group (n = 17)		p-value*	MI group (n = 17)		p-value*
	3 rd PO day	7 th PO day		3 rd PO day	7 th PO day	
Pain drugs (n/%)	17.0/100.0	15.0/88.3	0.600	16.0/94.1	6.0/35.3†	0.010*
Daily prescriptions	1.1 ± 0.3	1.0 ± 0.8	0.600	0.9 ± 0.2	0.3 ± 0.4	0.010*
Daily frequency	4.4 ± 1.8	3.0 ± 1.6	0.040*	3.1 ± 1.2	0.8 ± 1.4†	0.001*

MS: median sternotomy; MI: minithoracotomy; n: number of patients; %: frequency; PO: postoperative; *: $p < 0.05$ within group by paired Student's *t*-test for continuous variables and *z* test for categorical variables; † $p < 0.05$ between groups by unpaired Student's *t*-test for continuous variables and *z* test for categorical variables; 95% confidence level.

In addition, the number and sites of the incisions differed between the groups. The MS group had two thoracic incisions (main and chest tube incisions). The MI group had two incisions (thoracic and inguinal) and at least three right thoracic punctures. For the main incision, we used a wound protector for soft tissue³⁴ to diminish intercostal retraction that could lead to nerve stimulation^{11,35} during the procedure (e.g. valve replacement), which could be a cause of pain. We observed moderate to intense pain after both surgical approaches (MS and intercostal). According to the literature, at least 60% of the patients who underwent MS and MI procedures report moderate to severe pain in the early PO days.¹ As expected, the main sites of pain were directly related to the surgical incision, i.e., the sternal wound for MS patients and the inframammary area for patients who underwent MI procedure, although the MS group also reported extra-wound pain sites (posterior thoracic area). Although MI procedure involves a higher number of incision/punctures, these patients did not report more pain sites as compared with those undergoing MS procedure.

Regarding PI, we observed that the most remarkable differences between the groups occurred on the seventh PO day. There were no significant differences in PI between the third and the seventh PO days in the MS group. This data agrees with the study by Mueller et al.,²⁷ that indicated a slow reduction of thoracic pain following a sternal based procedure. On the other hand, there was a statistically significant reduction in PI from the third to the seventh PO days in the MI group. The presence of moderate to severe pain on the third PO day for both groups was in accordance with previous studies.^{1,11,23,35,36} Landreanu et al.,³⁷ and Nagahiro et al.,³⁸

compared conventional posterolateral thoracotomy with MI (video-assisted) thoracotomy procedures and verified that the MI approach promoted less postoperative PI. The advantages regarding PI and length of hospital stay for the MI procedures are well established¹ and were corroborated in our study. In patients subjected to MI procedure, there was lower PO pain and need for pain medication, and shorter ICU and hospital stay (19 hours shorter and two and a half days shorter, respectively). It is worth mentioning that although the group differed in the presence of comorbidities, no PO complications related to these conditions were found in neither of the groups, such as hypertensive crisis or pulmonary complications.

This work resulted in important findings regarding PO pain, a symptom often overlooked by healthcare professionals dealing with cardiac surgery. Wildgaard et al.,¹¹ noted that the strategies for PO pain control after thoracic procedures has evolved in the last years. However, inadequate pain management still affects the quality of life and postoperative outcomes of cardiac surgery patients,^{8,9} whereas adequate pain control results in better rehabilitation after a cardiovascular surgical procedure. Thus, pain management is a challenging task, as it demands attention by health professionals in making correct decisions towards medications and PI ratings, considering patients' pain tolerance and differences between protocols.

Despite all advances in the diagnosis and management of PO pain, accurate evaluation of this symptom is still very difficult, since the perception of pain and the response to pain medications vary widely between individuals. Also, the experience of the surgical team on MI procedures reduces possible complications of this type of surgery. A multicenter study involving larger

number of patients should be performed to confirm the differences in PI reported in our study and to overcome the limitations of this study.

Conclusion

Patients subjected to a MI cardiac procedure reported lower PI and fewer pain sites from the third PO day on, and showed lower need for pain medication and shorter ICU stay when compared to those subjected to a MS procedure. Based on these results, our study reinforces the advantages of a MI procedure for valve surgery.

Author contributions

Conception and design of the research: Silva JF, Castro JV, Montenegro RB, Lira R and Melo EC. Acquisition of data: Silva JF and Cavalcante, MP. Analysis and interpretation of the data: Silva JF, Castro JV and Melo EC. Writing of the manuscript: Silva JF, Cavalcante MP, Montenegro RB, Lira R, Melo EC, Castro JV. Critical revision of the manuscript for intellectual content: Silva JF, Cavalcante MP, Montenegro RB, Lira R, Melo EC, Castro JV.

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Potential Conflict of Interest

No potential conflict of interest relevant to this article was reported.

Sources of Funding

There were no external funding sources for this study.

Study Association

This article is part of the thesis of master submitted by Juliana Fernandes Silva, from *Universidade de Fortaleza* (UNIFOR).

Ethics approval and consent to participate

This study was approved by the Human Research Ethics Committee of *Universidade de Fortaleza* (Coética) under the protocol number 1104.606. All the procedures in this study were in accordance with the 1975 Helsinki Declaration, updated in 2013. Informed consent was obtained from all participants included in the study.

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