

PEER REVIEW HISTORY

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This paper was submitted to the HEART but declined for publication following peer review. The authors addressed the reviewers' comments and submitted the revised paper to BMJ Open. The paper was subsequently accepted for publication at BMJ Open.

ARTICLE DETAILS

TITLE (PROVISIONAL)	Effects of myocardial fibrosis assessed by magnetic resonance imaging on dynamic left ventricular outflow tract obstruction in patients with hypertrophic cardiomyopathy
AUTHORS	Rapezzi, Claudio ; BIAGINI, ELENA; Lorenzini, Massimiliano; Olivotto, Iacopo; Rocchi, Guido; Lovato, Luigi; Lai, Francesco; Rosmini, Stefania; Pazzi, Chiara; Pasquale, Ferdinando; Bacchi Reggiani, M. Letizia; Fattori, Rossella

VERSION 1 - REVIEW

REVIEWER	Ommen, Steve Mayo Clinic
REVIEW RETURNED	20-Feb-2012

GENERAL COMMENTS	<p>The manuscript by Biagini, et al, describes findings in 76 patients with hypertrophic cardiomyopathy to examine the association between late gadolinium enhancement on cardiac magnetic resonance imaging with left ventricular outflow tract obstruction. The authors found that the larger the amount of myocardial fibrosis detected, the smaller the increase in left ventricular outflow tract obstruction. The authors conclude that larger amounts of fibrosis are the cause for a decreased exacerbation of dynamic outflow tract obstruction observed during exercise.</p> <p>Specific Comments</p> <ol style="list-style-type: none">1. Page 5: The authors lay out on Page 5 four points for study to examine the association between fibrosis and gradient provocation. Other factors that would be important to consider are whether the severity and duration of exposure to excessive ventricular load could cause the fibrosis detected by gadolinium enhancement. This effect could also potentially be at least partially diminished in some of the patients by early exposure to gradient suppressing agents (e.g., beta-blockade, verapamil, disopyramide, et cetera). Additionally, the load experienced by the myocardium would be proportional to the gradient plus the peak systolic blood pressure. Finally, the severity of left ventricular wall thickness may also play a role in determining the presence of both fibrosis formation and gradient formation.2. Page 6, Results: Can the authors define the difference between diffuse and confluent gadolinium enhancement patterns? Is this reproducible by others?3. Page 7: The authors describe quartiles of gradient increase as groups in which they assessed extent of fibrosis. These results do
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	<p>not show a clear dose response in terms of fibrosis dose leading to lower gradient response.</p> <p>4. Page 7, last paragraph: The authors state that fibrosis “can influence the development of LVOT gradient”. What the authors have shown is an association between the two entities and not a cause and effect relationship.</p> <p>5. Page 8: Again, the authors have stated they have investigated the “role” of fibrosis in gradient development. Again, they have shown a statistical association between the two entities.</p> <p>6. Figure 1B: The linear regression plot of myocardial fibrosis versus delta LVOT gradient is instructive in that it clearly shows that three patients are the primary determinants of this regression curve (those three patients with nearly 15% of the myocardium replaced by fibrosis). Similarly, these three patients have a delta LVOT gradient that overlaps with patients who have almost no change in LVOT gradient with exercise.</p> <p>7. Was the delta gradient in any way related to the resting gradient? In other words, were patients with resting gradient more likely to have a higher or lower change in gradient with exercise?</p>
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REVIEWER	Carr-White, Gerry Guy's and St. Thomas' Foundation Trust, Department of Cardiology
REVIEW RETURNED	12-Feb-2012

GENERAL COMMENTS	<p>Dr Biagini and colleagues describe the relationship between fibrosis on CMR and exercise induced outflow tract gradients in 91 patients with hypertrophic cardiomyopathy. They show that patients with fibrosis are less likely to generate a significant outflow tract gradient even with preserved LV volumes and systolic function on exercise. The paper is well written and the echo and CMR methods are robust. The statistical methods are sound though some caution has to be given to the multiple subgroups used to find statistically significant relationships.</p> <p>My main question for the authors is what do the results mean to readers of quite a general cardiology journal. When you look at the raw scatter plots and R values the relationship is really quite weak so I am unsure how this will influence clinical practice or guide further studies and I think this really needs to be elaborated on in the discussion.</p> <p>The authors describe reproducibility measurements in the methods but need to give the results of these as it may be the errors here are larger than those between the different groups.</p>
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VERSION 1 – AUTHOR RESPONSE

Reviewer Comments:

Reviewer: 1

The paper is well written and the echo and CMR methods are robust. The statistical methods are sound though some caution has to be given to the multiple subgroups used to find statistically significant relationships. My main question for the authors is what do the results mean to readers of quite a general cardiology journal. When you look at the raw scatter plots and

R values the relationship is really quite weak so I am unsure how this will influence clinical practice or guide further studies and I think this really needs to be elaborated on in the discussion.

Thank you for the observations.

Our study was designed to be small and hypothesis generating and we do not think that our results should influence current clinical practice. However, myocardial fibrosis in HCM is currently the object of a considerable amount of research regarding its pathophysiology and meaning. Our results should be read along with these other papers as a basis for expanding the knowledge of HCM and designing larger studies combining cardiac MRI and exercise echocardiography.

The authors describe reproducibility measurements in the methods but need to give the results of these as it may be the errors here are larger than those between the different groups.

Thank you for the observation, we have now added this data (page 6, lines 15-20).

Reviewer: 2

1. Page 5: The authors lay out on Page 5 four points for study to examine the association between fibrosis and gradient provocation. Other factors that would be important to consider are whether the severity and duration of exposure to excessive ventricular load could cause the fibrosis detected by gadolinium enhancement. This effect could also potentially be at least partially diminished in some of the patients by early exposure to gradient suppressing agents (e.g., beta-blockade, verapamil, disopyramide, etcetera). Additionally, the load experienced by the myocardium would be proportional to the gradient plus the peak systolic blood pressure. Finally, the severity of left ventricular wall thickness may also play a role in determining the presence of both fibrosis formation and gradient formation.

Thank you for the observations. We agree that the relationship between fibrosis and outflow gradient is complex and includes many interrelated factors. We have therefore modified the discussion section of our paper in order to include these concepts. The revised version reads as follows: "The exact mechanism leading to fibrosis remains unknown but it has been hypothesized that the main triggers for the fibrotic process include molecular factors at cellular level (induced by sarcomeric mutations), hemodynamic factors (overall ventricular afterload resulting from the sum of LV outflow obstruction and systolic blood pressure), and ischemia (mainly related to small intramural coronary vessel disease) [8]."

2. Page 6, Results: Can the authors define the difference between diffuse and confluent gadolinium enhancement patterns? Is this reproducible by others?

The categorization was carried out subjectively by experienced cardiac MRI readers, as in most published papers on this topic. In order to clarify our definition however, we have modified the manuscript as follows: "Fibrosis consisted of small, diffuse areas in 32 patients (59%) and was confluent into a smaller number of larger areas in 22 patients (41%)". The two pattern used to describe late Gd enhancement were included solely for descriptive purposes as they were not used in any of our analyses.

3. Page 7: The authors describe quartiles of gradient increase as groups in which they assessed extent of fibrosis. These results do not show a clear dose response in terms of fibrosis dose leading to lower gradient response.

The small number of patients included in our study probably underpowers it statistically and is probably responsible for the absence of a clear dose response in the analysis of the population divided into quartiles according to gradient increase. However, the fact that the extent of fibrosis was significantly lower in the highest quartile of LVOT gradient increase and higher in the lowest quartile of LVOT gradient increase indicates a trend and is in line with our other findings. As stated in the limitations section, this was conceived as a hypothesis generating study and larger studies are necessary to confirm our hypotheses.

4. Page 7, last paragraph: The authors state that fibrosis “can influence the development of LVOT gradient”. What the authors have shown is an association between the two entities and not a cause and effect relationship.

Thank you for the observation. You are correct in saying that we have only shown an association between to entities and we hypothesize that this is the result of a cause and effect relationship. We have therefore modified our manuscript as follows: “This study shows that myocardial fibrosis (detected as LGE on MRI) may influence the development of LVOT gradient during exercise in patients with HCM and normal EF: patients with higher exercise-induced gradients show a lesser degree of myocardial fibrosis and vice versa (Figure 3).”

5. Page 8: Again, the authors have stated they have investigated the “role” of fibrosis in gradient development. Again, they have shown a statistical association between the two entities.

As above you are correct, thank you. We have therefore modified the manuscript that now reads as follows: “The present study confirmed the association between myocardial fibrosis and contractility, assuming that LV systolic function is one of the major determinants of the LVOT gradient increase during effort.”

6. Figure 1B: The linear regression plot of myocardial fibrosis versus delta LVOT gradient is instructive in that it clearly shows that three patients are the primary determinants of this regression curve (those three patients with nearly 15% of the myocardium replaced by fibrosis). Similarly, these three patients have a delta LVOT gradient that overlaps with patients who have almost no change in LVOT gradient with exercise.

We agree with this observation. Given the small number of patients the statistical significance emerges due to patients with an extreme behaviour. However, this does not undermine the statistical significance of the study or its potential hypothesis generating role.

7. Was the delta gradient in any way related to the resting gradient? In other words, were patients with resting gradient more likely to have a higher or lower change in gradient with exercise?

Thank you for the observation. The data was not included in our paper but patients with a significant obstruction at rest do tend to develop a greater gradient during exercise. In order to not complicate the paper we would prefer to not include this data in the manuscript.