PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Multi-Centre Cross-Sectional Observational Registry to Monitor the Safety of Early Discharge after Rule-Out of Acute Myocardial Infarction by Copeptin and Troponin: The Pro-Core registry
AUTHORS	Giannitsis, Evangelos; Clifford, Piers; Slagman, Anna; Ruedelstein, Ralph; Liebetrau, Christoph; Hamm, Christian; Honnart, Didier; Huber, Kurt; Vollert, Jörn; Simonelli, Carlo; Schröder, Malte; Wiemer, Jan; Mueller-Hennessen, Matthias; Schroer, Hinrich; Kastner, Kim; Möckel, Martin

VERSION 1 – REVIEW

REVIEWER	Philip Haaf University Hospital Basel, Switzerland
REVIEW RETURNED	16-Jan-2019

GENERAL COMMENTS	Comments to the authors Multi-Centre Registry to Monitor the Safety of Copeptin and Troponin for the Early Rule-Out of Acute Myocardial Infarction in Patients with Suspected Acute Coronary Syndrome: The Pro-Core registry General remark The article is well written, interesting and addresses an important clinical issue. Major: Please outline separately for all patients with conventional vs. high-sensitivity cardiac troponin assays that copeptin really had an incremental prognostic benefit. The incremental benefit of copeptin is probably much lower for hs-cTn assays than conventional assays. Major: New biomarkers should always prove their incremental benefit on top of already established biomarkers. Did you measure natriuretic peptides in this patient cohort? It would be really interesting to show the incremental benefit of copeptin/(hs-)cTN vs. BNP/NT-proBNP/(hs-)cTn. Minor: The official (Clinical Chemistry) abbreviation of high-sensitivity cardiac Troponin is “hs-cTnT”. So, the authors might use this abbreviation instead of Hs TnT Abstract Please verify whether the word count (319) complies with the journal requirements. Study cohort size is very good, also the high proportion of female patiens and high proportion of “younger” patients. Strengths and limitations of the study “This is the first study of its kind to examine the clinical use of a fast rule-out and early discharge concept in suspected acute myocardial infarction using

	cardiac troponin and copeptin in a large European registry" In my opinion, this is not fully true. There have been several other large European studies analysing fast-rule out of suspected AMI with this dual-marker approach. If you underline the safety (prognostic) of such an approach (30-day mortality) this statement might be correct. • Incremental value of copeptin for rapid rule out of acute myocardial infarction J Am Coll Cardiol. 2009 Jun 30;54(1):60-8. doi: 10.1016/j.jacc.2009.01.076. • Incremental value of copeptin to highly sensitive cardiac Troponin I for rapid rule-out of myocardial infarction. Int J Cardiol. 2015;190:170-6. doi: 10.1016/j.ijcard.2015.04.133. • Comparison of the performances of cardiac troponins, including sensitive assays, and copeptin in the diagnostic of acute myocardial infarction and long-term prognosis between women and men. Am Heart J. 2013 Jul;166(1):30-7. doi: 10.1016/j.ahj.2013.03.014. • Diagnostic accuracy of adding copeptin to cardiac troponin for non-ST-elevation myocardial infarction: A systematic review and meta-analysis PLoS One. 2018 Jul 6;13(7):e0200379. doi: 10.1371/journal.pone.0200379. eCollection 2018. Statistical evaluation "Enrolment was restricted to a maximum number of 300 patients per center to ensure generality by avoiding the dominance of single centers." This is a very good approach. Results The proportion of patients who were allocated to the conventional care pathway at the discretion of the local investigator although they were categorized into the primary discharge after fast rule-out pathway is substantial: 503 patients (21.9%). Altogether 503+151=654 patients switched groups (28,5%). This, I think, is a real bias makes the main conclusions less generalizable. This should mentioned in both discussion and limitations. Figure 1 • Minor comment: The categories should be modified from (<109, 109-140, ≥140) to (<109, 109-140, >140). Otherwise Grace Score of 140 could belong to both group "109-140" and "≥140" • "ECG not diagnostic" might imply to the reader that the ECG was not readable/of poor quality. Probably you mean that the ECG was not helpful in the distinction of NSTEMI Figure 1S • CVK and UKB should be explained in the figure legend Discussion One of the big advantages of using a combined copeptin/troponin single blood draw instead of serial troponin measurements is the broader exclusion (or at least making less likely) of other differential diagnoses such as acute heart failure, acute pulmonary embolism and non-cardiac causes as you mentioned at the end of your discussion. Nevertheless, I believe, that other diagnostic biomarkers are superior to copeptin in the diagnosis of heart failure (natriuretic peptides), pulmonary embolism (d-dimes and hs-cTnT). You should mention this in the discussion. Discuss the problem of the term "troponinn-negative chest pain". "Troponin-negative chest pain" should only be a working diagnosis and is often an evasion for identifying the underlying cause of acute

	chest pain. All serious and life-threatening diseases (according to their pre-test probabilities) should be excluded in the emergency department and not only acute coronary syndrome. Therefore, a combined measurement of copeptin and hs-cTnT might be (at least prognostically) more helpful. Nevertheless, I think, you should really make it clear that in the diagnosis of AMI there is no incremental benefit of copeptin when used with (serial) hs-cTnT. The combination of copeptin and conventional troponin (which in less and less used) might still be helpful with regard to diagnosis

REVIEWER	Edd Carlton North Bristol NHS Trust, UK I undertake research in this field and have received industry funding from various biomarker manufacturers
REVIEW RETURNED	28-Jan-2019

GENERAL COMMENTS	Many thanks for asking me to review this interesting manuscript. The authors should be congratulated for putting together an excellent dataset analysing the real world performance of a combined copetin/troponin algorithm for rule-out of MI. I do however have some comments for the authors to consider as follows: MAJOR 1. The authors need to decide whether this is a registry study examining clinical effectiveness of their approach or an observational diagnostic accuracy study. This is confused throughout the manuscript and both terms are mentioned. Similarly very limited diagnostic accuracy statistics are quoted (NPV only and no confidence intervals). If it is a true registry study then the primary outcomes should focus on clinical effectiveness ie the proportion discharged early/at what time point, rather than 30 day all cause mortality. 2. I am left wondering at the end of the manuscript what the added value of copeptin is. It would be worthwhile for the authors to consider a secondary analysis around this in their early discharge patients and may add more weight as to why they feel a combined biomarker strategy is required. 3. The comparative analysis of the early discharge cohort vs conventional care is problematic. This is not a randomised controlled trial and by definition those who have conventional care will be those with elevated presentation troponin/copeptin results. Demonstrating a difference in length of stay difference between these two groups is therefore not surprising and lending much of the results section to the conventional care pathway is misleading and outside of the primary aim of this manuscript. 4. I find the exclusion of high GRACE score patients a priori problematic. The authors will be aware that the GRACE score includes biomarker results. The authors argue their population is unselected yet they have excluded high GRACE scores. This effects the applicability of the study findings since very few centres (certainly in the UK) use GRACE scoring for risk stratification. 5. Adjudication of outcomes is again problematic. MI was defined by the treating ED clinician which will lead to adjudication bias. I note all patients were contacted at 30 days, did you collect information on 30 day MI/revascularistaion in a robust manner? 6. Why was all cause mortality at 30 days chosen as the primary outcome? I would be more interested in emergency revascularistion/prevalent MI as an outcome that reflects diagnostic

	performance (although this returns me to point 1). 7. It is stated that patients were screened in a consecutive manner. Is this really the case? Were all sites recruiting over the same time period? I note one site only recruited 2 participants. Again this leads me to have concerns that the population is not really as unselected as the authors suggest. Minor points: 1. The introduction and discussion are both too long and would benefit from trimming down to really focus on what the added scientific value of this manuscript is. 2. The analysis would really benefit from further analysis investigating early presenters since this (in my understanding) is where copeptin may add value. 3. I would suggest toning down the conclusion that this is the only strategy that supports the safe discharge of patients with chest pain under routine conditions. 4. Is there data available from this analysis as to how long it takes for copeptin results to become availaible. I note even in the fast discharge pathway that the majority of patients stayed 3-6 hours even though they had a single test at presentation. This is a point worthy of discussion.

REVIEWER	Eiichiro Nishi Shiga University of Medical Science, Japan
REVIEW RETURNED	31-Jan-2019

GENERAL COMMENTS	In this prospective multi center cohort study, Giannitsis et.al. describe the results of dual marker measurement (copeptin and troponin) and clinical outcome for patients suggestive of ACS at low-to-intermediate risk. Although the large registration study addresses a topic of considerable importance, there are several concerns. The following specific points are worth consideration. 1. For the primary comparison, the authors compare the group in which patients were early discharged with combined negative testing of copeptin and troponin (defined as “primary discharge group”) and all other patients. First of all, the name “primary discharge group” is confusing because 151 patients with positive testing of copeptin and/or troponin were also primarily discharged. The authors should define 4 groups shown in Box Line 5 in Figure 2 as A: discharge/negative (n=974), B: admission/negative (n=503), C: admission/positive (n=666) and D: discharge/positive (n=151). Second, the comparison of group A and all others (B, C and D) does not make sense. Simply, the authors should compare all 4 groups in new Table 2 and 3. 2. Although diagnostic performance of copeptin/copeptin+troponin for ACS has been already reported in several manuscripts, ROC curve analysis with AUC determination should be performed in this cohort and compared with the previous studies. Moreover, the sensitivity, specificity, PPV, and NPV value of copeptin/copeptin+troponin should be demonstrated. 3. Diagnostic criteria of unstable angina (UA) should be clearly described. 4. In this study, the attending physician made a decision for primary discharge or hospital admission according to conventional clinical assessment and the results of dual marker measurement. Since

	copeptin and troponin were measured in all participants, diagnostic value of the measurement on top of the conventional clinical assessment cannot be assessed in this study. Accordingly, the authors should rephrase the Conclusion in Abstract.

VERSION 1 – AUTHOR RESPONSE

Reviewer: 1
Major: Please outline separately for all patients with conventional vs. high-sensitivity cardiac troponin assays that copeptin really had an incremental prognostic benefit. The incremental benefit of copeptin is probably much lower for hs-cTn assays than conventional assays.

The reviewer is correct that the incremental diagnostic and/or prognostic benefit is presumably larger with conventional or contemporary sensitive troponin assays. Indeed, only few studies have addressed the incremental prognostic benefit of Copeptin added to cTn or hsTn. However, this study sought to validate prospectively the safety of discharge in the real world, and thus to confirm the beneficial findings of the randomized BIC-8 trial. The concept proved to be so effective that a comparison between different cTn assay generations is not meaningful since only 1 fatal event occurred in the experimental arm.

This issue has now been added to the statistics.

Major: New biomarkers should always prove their incremental benefit on top of already established biomarkers. Did you measure natriuretic peptides in this patient cohort? It would be really interesting to show the incremental benefit of copeptin/(hs-)cTN vs. BNP/NT-proBNP/(hs-)cTn.

The aim of this study was not to test the incremental diagnostic/prognostic performance of the dual biomarker strategy but to confirm the safety of primary discharge from ED that has been already demonstrated in the randomized BIC-8 trial (Biomarkers in Cardiology-8). The decision to admit or discharge is complex usually requiring clinical judgement or use of a validated clinical score such as the GRACE score. Indeed, the GRACE score was used as a filter to preclude the use of the dual marker strategy (DMS) for patients at high risk, i.e. a GRACE score > 140 points. Use of natriuretic peptides might be useful to refine clinical judgement in patients presenting primarily with acute dyspnoea. However, measurement of natriuretic peptides is not recommended routinely for low risk patients for the decision to admit or discharge.

We clarified in the discussion and limitation section that the DMS was restricted to patients at low or intermediate risk for death or MI at 30 days and 6 months based on the GRACE score.

Minor: The official (Clinical Chemistry) abbreviation of high-sensitivity cardiac Troponin is “hs-cTnT”. So, the authors might use this abbreviation instead of Hs TnT
There is no consensus on the correct abbreviation of hs-cTnT. The consensus group on biomarkers (two of the authors, i.e. MM and EG belong to this consensus group) never made a specific comment on the exact wording although the most widely used abbreviation is hs-cTnT or hsTnT. In addition, the manufacturer still uses the term TnThs or cTnThs, an abbreviation that we dislike.
In order to comply with the reviewer´s suggestion, we exchanged hsTnT by hs-cTnT throughout the manuscript.

Abstract
Please verify whether the word count (319) complies with the journal requirements.
Study cohort size is very good, also the high proportion of female patients and high proportion of "younger" patients.

The abstract was shortened to 294 words (300 words limit).

Strengths and limitations of the study
"This is the first study of its kind to examine the clinical use of a fast rule-out and early discharge concept in suspected acute myocardial infarction using cardiac troponin and copeptin in a large European registry"
In my opinion, this is not fully true. There have been several other large European studies analysing fast-rule out of suspected AMI with this dual-marker approach. If you underline the safety (prognostic) of such an approach (30-day mortality) this statement might be correct.
J Am Coll Cardiol. 2009 Jun 30;54(1):60-8. doi: 10.1016/j.jacc.2009.01.076.

- Incremental value of copeptin for rapid rule out of acute myocardial infarction.. J Am Coll Cardiol. 2009 Jun 30;54(1):60-8. doi: 10.1016/j.jacc.2009.01.076.

Incremental value of copeptin to highly sensitive cardiac Troponin I for rapid rule-out of myocardial infarction. Int J Cardiol. 2015;190:170-6. doi: 10.1016/j.ijcard.2015.04.133.

- Comparison of the performances of cardiac troponins, including sensitive assays, and copeptin in the diagnostic of acute myocardial infarction and long-term prognosis between women and men.

Am Heart J. 2013 Jul;166(1):30-7. doi: 10.1016/j.ahj.2013.03.014.

- Diagnostic accuracy of adding copeptin to cardiac troponin for non-ST-elevation myocardial infarction: A systematic review and meta-analysis PLoS One. 2018 Jul 6;13(7):e0200379. doi: 10.1371/journal.pone.0200379. eCollection 2018.

There are numerous publications and even two meta-analyses on the diagnostic or prognostic value of a DMS for rule-out of MI that ultimately led to the ESC guideline recommendation for DMS as an alternative option for instant rule-out of MI. The present ProCore registry is distinct to these studies as it does not study the diagnostic performance, but was planned to confirm the findings of the randomized BIC-8 trial on the safety of early discharge (which is not identical to "rule-out") in low-to intermediate risk patients with suspected ACS. In contrast to the BIC-8 trial this registry reflects clinical routine conditions evidence and as such included patients with a broader spectrum of symptoms, a broad range of troponin assays and different assay generations, and different level of care provision, i.e. academic and non-academic hospitals.

We rephrased the introduction and discussion to improve the understanding of the study goal and the unmet need that was addressed.

Results
The proportion of patients who were allocated to the conventional care pathway at the discretion of the local investigator although they were categorized into the primary discharge after fast rule-out pathway is substantial: 503 patients (21.9%). Altogether 503+151=654 patients switched groups (28,5%). This, I think, is a real bias makes the main conclusions less generalizable. This should mentioned in both discussion and limitations.
Again we would like to highlight that this registry describes routine clinical care. That means, no patients were "allocated". Thus, the current findings do not limit the generalizability, they show additional potential once the attending physicians get more confident with the concept. Of note, there are no data published about routine discharge rates and timing with respect to hs troponin only based protocols (e.g. 1hour-protocol)!

Figure 1

- Minor comment: The categories should be modified from (<109, 109-140, ≥140) to (<109, 109-140,
>140). Otherwise Grace Score of 140 could belong to both group “109-140” and “≥140”

We agree with reviewer and corrected the intervals. Correction for this overlap was not associated with a reclassification.

- “ECG not diagnostic” might imply to the reader that the ECG was not readable/of poor quality. Probably you mean that the ECG was not helpful in the distinction of NSTEMI

We prefer to keep this wording which is very common in the literature and simply indicates that the ECG was either normal or unspecific, i.e. without significant ST-segment depression, or presence of left or right bundle branch block or pacemaker stimulation.

Figure 1S

- CVK and UKB should be explained in the figure legend

We agree with the reviewer and corrected this accordingly.

Discussion

One of the big advantages of using a combined copeptin/troponin single blood draw instead of serial troponin measurements is the broader exclusion (or at least making less likely) of other differential diagnoses such as acute heart failure, acute pulmonary embolism and non-cardiac causes as you mentioned at the end of your discussion.

Nevertheless, I believe, that other diagnostic biomarkers are superior to copeptin in the diagnosis of heart failure (natriuretic peptides), pulmonary embolism (d-dimes and hs-cTnT). You should mention this in the discussion.

Discuss the problem of the term “troponinn-negative chest pain”. “Troponin-negative chest pain” should only be a working diagnosis and is often an evasion for identifying the underlying cause of acute chest pain. All serious and life-threatening diseases (according to their pre-test probabilities) should be excluded in the emergency department and not only acute coronary syndrome. Therefore, a combined measurement of copeptin and hs-cTnT might be (at least prognostically) more helpful.

Nevertheless, I think, you should really make it clear that in the diagnosis of AMI there is no incremental benefit of copeptin when used with (serial) hs-cTnT. The combination of copeptin and conventional troponin (which in less and less used) might still be helpful with regard to diagnosis

First, we agree with reviewer that the term troponin-negative chest pain is incorrect and particularly for the usefulness of DMS which provides most benefit among patients with detectable but still normal hsTnT and no incremental benefit beyond a single very low hsTn value (Ref #29: Vafaie M; et al. Am J Med. 2016;129:274-82), provided the patients presents more than 3 hours after onset of symptoms.

Regarding the performance of other biomarkers, we disagree because correct diagnosis is not the scope of this registry but rather safety of discharge in low or intermediate risk patients, regardless the exact underlying reason.

Accordingly, we rephrased the critical sentence and added text to the limitations regarding the appropriate use of Copeptin. Reviewer: 2 Major:

The authors need to decide whether this is a registry study examining clinical effectiveness of their approach or an observational diagnostic accuracy study. This is confused throughout the manuscript and both terms are mentioned. Similarly very limited diagnostic accuracy statistics are quoted (NPV only and no confidence intervals). If it is a true registry study then the primary outcomes should focus

on clinical effectiveness ie the proportion discharged early/at what time point, rather than 30 day all cause mortality.

We regret the confusion that our wording created. The study is neither a diagnostic study nor a purely observational registry but rather a prospective validation study of a ESC recommended diagnostic strategy to confirm the efficacy and safety of primary discharge from the ED after instant rule out of an MI among patients with low to intermediate risk for future death or MI. As such we did not focus on the diagnostic performance. Nevertheless, we appreciate the reviewer´s comment on the need to provide confidence intervals. In our manuscript, data on efficacy in terms of how many patients qualify for the instant rule-out protocol are provided in the result section. In addition, length of ED stay was investigated to demonstrate the clinical relevance.
Action: We clarified the scope of the study by rephrasing the introduction and added 95% CI.

2. I am left wondering at the end of the manuscript what the added value of copeptin is. It would be worthwhile for the authors to consider a secondary analysis around this in their early discharge patients and may add more weight as to why they feel a combined biomarker strategy is required.

The study does not test the added diagnostic or prognostic benefits of adding Copeptin to a normal troponin but is purely a prospective validation study of the BIC-8 trial, a randomized validation study on 910 patients that tested the safety of early discharge in patients qualifying for instant rule-out. This prospective validation was thought to add confidence for clinicians who have concerns that this concept may not work in the real world, i.e. consecutive patients, early presenters, range of troponin and across different hospital settings.
We rephrased to clarify the need for such a prospective validation study

3. The comparative analysis of the early discharge cohort vs conventional care is problematic. This is not a randomised controlled trial and by definition those who have conventional care will be those with elevated presentation troponin/copeptin results. Demonstrating a difference in length of stay difference between these two groups is therefore not surprising and lending much of the results section to the conventional care pathway is misleading and outside of the primary aim of this manuscript.
We agree with the reviewer. The effects of reducing the length of stay were already demonstrated in the BIC-8 trial and were successfully confirmed in the ProCore registry.

4. I find the exclusion of high GRACE score patients a priori problematic. The authors will be aware that the GRACE score includes biomarker results. The authors argue their population is unselected yet they have excluded high GRACE scores. This effects the applicability of the study findings since very few centres (certainly in the UK) use GRACE scoring for risk stratification.

We agree with the reviewer and will exchange unselected for consecutive as patients were preselected by low or intermediate risk. However, this registry sought to evaluate the safety of discharge. In this context, use of clinical judgement or a clinical score is recommended. In clinical practice, the score is used after diagnostic classification. In our cohort, patients with a high risk GRACE score underwent the same diagnostic process but it was not intended to include them since admission is highly likely. Nevertheless, as the GRACE-score is sometimes only available after primary stratifications, some patients with higher scores are in the registry.

5. Adjudication of outcomes is again problematic. MI was defined by the treating ED clinician which will lead to adjudication bias. I note all patients were contacted at 30 days, did you collect information on 30 day MI/revascularisation in a robust manner?

We agree with the reviewer regarding the lack of an independently adjudicated MI. However, this is usual care in clinical routine where a physician does not receive a retrospectively confirmed diagnosis. Nevertheless, the adjudication and clinical judgement of the ED physician (different level of experience) as well as the resulting discharge did not result in an excess of fatality within 30 days.
Action: we expanded the limitations to address the reviewer´s comment on lack of an independent (retrospective) adjudication.

6. Why was all cause mortality at 30 days chosen as the primary outcome? I would be more interested in emergency revascularisation/prevalent MI as an outcome that reflects diagnostic performance (although this returns me to point 1).

This study is not a diagnostic study and rates of missed NSTEMI are reported below 0.5% using the dual biomarker strategy. In addition, revascularization therapies in patients at low risk may increase procedure related complications such as procedure related myocardial injury or major bleedings. We chose all-cause mortality because this event is unequivocal and more convenient to collect by phone than cardiovascular death.
We added text to illustrate the rationale to use all-cause death.

7. It is stated that patients were screened in a consecutive manner. Is this really the case? Were all sites recruiting over the same time period? I note one site only recruited 2 participants. Again this leads me to have concerns that the population is not really as unselected as the authors suggest.

The recruitment was heterogeneous between sites. In order to avoid dominance from leading centers diluting the real world evidence, enrolment was restricted to a maximum of 300 patients per site. The numbers of enrolled patients is transparently displayed in supplemental Figure 1S.

We expanded the limitations to indicate that enrolment was heterogeneous and that we therefore restricted enrolment numbers per site to avoid a site dominance on results.

Minor points:

1.The introduction and discussion are both too long and would benefit from trimming down to really focus on what the added scientific value of this manuscript is.

We rephrased the introduction and discussion

2. The analysis would really benefit from further analysis investigating early presenters since this (in my understanding) is where copeptin may add value.

We agree with the reviewer that an analysis on the diagnostic performance strongly depends on time to presentation since Copeptin shows a rapid reverse release kinetic and as such is not helpful in late presenters (Karakas M, et al.), showing particular benefits in very early presenters (Stengaard). However, this is outside the scope of this study that aims to test safety of early discharge from ED. We expanded this issue in the limitations indicating scrutiny in the interpretation of Copeptin in late presenters due to the rapid return of Copeptin levels to normal.

3. I would suggest toning down the conclusion that this is the only strategy that supports the safe discharge of patients with chest pain under routine conditions.
We agree with the reviewer and decided to soften the conclusion. The new wording is that there a only few randomized trials that provide evidence for a safe discharge after rule-out in low risk patients.

The present registry confirms findings from the randomized BIC-8 trial in an independent real world registry.

4. Is there data available from this analysis as to how long it takes for copeptin results to become available. I note even in the fast discharge pathway that the majority of patients stayed 3-6 hours even though they had a single test at presentation. This is a point worthy of discussion.

The measuring time for Copeptin is below 15 minutes on a Kryptor analyser. However, we agree with the reviewer on disproportionally longer lengths of stay in ED. Therefore, we added text to the limitations stating that patients could have been discharged earlier. However, obviously there are delays in processes in the management of patients that are independent of the biomarker algorithm such as waiting times for diagnostic workup, time for report drafting etc.

Reviewer 3
Giannitsis et.al. describe the results of dual marker measurement (copeptin and troponin) and clinical outcome for patients suggestive of ACS at low-to-intermediate risk. Although the large registration study addresses a topic of considerable importance, there are several concerns. The following specific points are worth consideration.

1. For the primary comparison, the authors compare the group in which patients were early discharged with combined negative testing of copeptin and troponin (defined as “primary discharge group”) and all other patients.
First of all, the name “primary discharge group” is confusing because 151 patients with positive testing of copeptin and/or troponin were also primarily discharged. The authors should define 4 groups shown in Box Line 5 in Figure 2 as A: discharge/negative (n=974), B: admission/negative (n=503), C: admission/positive (n=666) and D: discharge/positive (n=151). Second, the comparison of group A and all others (B, C and D) does not make sense. Simply, the authors should compare all 4 groups in new Table 2 and 3.

We disagree with the reviewer regarding the term primary discharge group. There are two major pathways per protocol. Not unexpectedly, there were protocol violations in both directions based on the clinical judgement of treating physicians. We provided sufficient data to demonstrate that a) violations were justified as they characterised a patient group with different risk profile than the protocol adherent group and b) that the number of patients potentially qualifying for this strategy could be higher (efficacy).

2. Although diagnostic performance of copeptin/copeptin + troponin for ACS has been already reported in several manuscripts, ROC curve analysis with AUC determination should be performed in this cohort and compared with the previous studies. Moreover, the sensitivity, specificity, PPV, and NPV value of copeptin/copeptin + troponin should be demonstrated.
The reviewer´s comment raise the suspicion that we did not clearly state the purpose of the present prospective interventional registry. We rephrased the introduction to state the purpose of this registry. THIS IS NOT A DIAGNOSTIC STUDY BUT A MANAGEMENT STUDY TESTING MAINLY SAFETY OF DISCHARGE AFTER RULE-OUT USING THE DMS PROTOCOL

3. Diagnostic criteria of unstable angina (UA) should be clearly described.

We added the definition criteria of unstable angina in the method section.

4. In this study, the attending physician made a decision for primary discharge or hospital admission according to conventional clinical assessment and the results of dual marker measurement. Since copeptin and troponin were measured in all participants, diagnostic value of the

measurement on top of the conventional clinical assessment cannot be assessed in this study. Accordingly, the authors should rephrase the Conclusion in Abstract.

The reviewer would be correct only if this study would test the added diagnostic value of copeptin on top of troponin. However, this is NOT a diagnostic study and the purpose was mainly to test safety of discharge after instant rule-out in low to intermediate risk patients with suspected ACS (along with efficacy and some performance measures). Therefore, we rephrased the introduction to clearly state the purpose of this study.

VERSION 2 – REVIEW

REVIEWER	Edward Carlton North Bristol NHS Trust, UK I have undertaken research in this area, including an NIHR funded randomised controlled trial
REVIEW RETURNED	28-Mar-2019

GENERAL COMMENTS	The authors have in the main addressed my concerns. There are a few grammatical errors which will hopefully be picked up during the editing process. I still have one minor comment regarding the conclusion as follows: "We believe that the present findings will have enormous implications on health care resources by shortening observation times, hospitalization rates, reducing diagnostic resources, and avoid unnecessary coronary angiographies." This statement is still a little over stated and assumes widespread adoption of this strategy, which of course is not a given. Perhaps it would be wort qualifying this by stating it has potential should barriers to adoption be overcome and cost-effectiveness is demonstrated?

REVIEWER	Eiichiro Nishi Shiga University of Medical Science, Japan
REVIEW RETURNED	28-Mar-2019

GENERAL COMMENTS	In the author's response, the authors repeatedly stated that the aim of this study is to validate the results of BIC-8 trial. In that trial, they demonstrated the safety of DMS for ruling out ACS, and suggested that DMS has the potential to shorten LOS in the ED. The design of the current study, the comparative analysis of the "primary discharge group" and "conventional work-up group", however, cannot validate the previous trial and cannot support their conclusion. The reviewer concluded that the manuscript in the current form cannot be accepted in BMJ Open.

VERSION 2 – AUTHOR RESPONSE

Reviewer: 2

The authors have in the main addressed my concerns. There are a few grammatical errors which will hopefully be picked up during the editing process. I still have one minor comment regarding the conclusion as follows:

"We believe that the present findings will have enormous implications on health care resources by shortening observation times, hospitalization rates, reducing diagnostic resources, and avoid unnecessary coronary angiographies." This statement is still a little over stated and assumes widespread adoption of this strategy, which of course is not a given. Perhaps it would be worth qualifying this by stating it has potential should barriers to adoption be overcome and cost-effectiveness is demonstrated?

Answer: We thank the reviewer for this balanced comment and re-worded the conclusion as follows:

“We believe that the present findings have potential impact on health care resources by shortening observation times, hospitalization rates, reducing diagnostic resources, and avoid unnecessary coronary angiographies should barriers to adoption be overcome.”

We also added a sentence and a reference in the first paragraph of the discussion related to the recently published economic sub-analysis of the BIC-8-study.

Following the randomized BIC-8 study, which proofed safe discharge after instant rule-out of AMI by the use of troponin and Copeptin from a single blood draw (22) and also showed cost-effectiveness in a health economic sub-study (50), we could confirm in a large European registry that this is also true in clinical routine.

Reviewer: 3

In the author’s response, the authors repeatedly stated that the aim of this study is to validate the results of BIC-8 trial. In that trial, they demonstrated the safety of DMS for ruling out ACS, and suggested that DMS has the potential to shorten LOS in the ED. The design of the current study, the comparative analysis of the “primary discharge group” and “conventional work-up group”, however, cannot validate the previous trial and cannot support their conclusion. The reviewer concluded that the manuscript in the current form cannot be accepted in BMJ Open.

Answer: As outlined above in the response to reviewer 2, we have revised the conclusion.