PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Rapid rEcognition of COrticosteRoiD resistant or sensitive Sepsis (RECORDS): study protocol for a Multicenter, placebo-controlled, biomarker-guided, adaptive Bayesian design Basket trial.
AUTHORS	Fleuriet, Jérôme; heming, nicholas; MEZIANI, Ferhat; Reignier, Jean; Declerq, Pierre-Louis; Mercier, Emmanuelle; Muller, Grégoire; Colin, Gwenhaël; Monnet, Xavier; Robine, Adrien; Siami, Shidasp; Uhel, Fabrice; Quenot, Jean-Pierre; Plantefeve, Gaetan; Badie, Julio; Schneider, Francis; Cerf, Charles; Troché, Gilles; Monchi, Mehran; Mira, Jean-Paul; Francois, Bruno; Chevret, Sylvie; Annane, Djillali

VERSION 1 – REVIEW

REVIEWER	Sun, Tong-Wen The first affiliated hospital of Zhengzhou University, General ICU
REVIEW RETURNED	08-Sep-2022

GENERAL COMMENTS	In this study, the heterogeneity of corticosteroid treatment in sepsis patients was studied. I note that the value of biomarkers, including series levels of proteins, cellular markers, and multiomics, and of intelligent algorithms to identify the best sepsis population to be selected for corticotherapy and those who should not be treated with cortical steroids are proposed to be provided in this multicenter randomized controlled study. If this complex large-scale clinical trial can be carried out smoothly, the results obtained may screen out endotypes associated with adults with sepsis responsiveness to corticosteroids. The study protocol lists the inclusion and exclusion criteria of the trial, and the study measurements and procedures are very detailed and have clinical value.

REVIEWER	Walkey, , Allan J Boston University Medical Campus, The Pulmonary Center
REVIEW RETURNED	12-Jan-2023

GENERAL COMMENTS	Annane and colleagues submit a study protocol for an ongoing trial to identify biomarkers of responsiveness to combined glucocorticoid (hydrocortisone, or if SARS-CoV-2, dexamethasone) and mineralocorticoid (fludrocortisone) therapy among adults with sepsis. The study is in two stages – first a prospective observational study to collect biomarkers and then an RCT. Overall the research question of finding subgroups particularly responsive to steroids in sepsis is important and use of a basket trial with adaptive design is innovative. However, the description of how participants will be allocated to biomarker strata, how the subgroups within each biomarker strata will be analyzed, and how decisions will be made to drop futile biomarkers or preferentially enroll in promising biomarker

	strata is currently confusing and under-developed. Additional areas that require clarification are listed below. 1) Has the observational study to identify biomarkers to use stratification been completed? Is there a publication to describe the observational biomarker selection phase? a. It appears 12 different strata are listed, though the manuscript (p13, lines 10-12) states 13 strata have been identified. 1COVID, 2flu, 3other respiratory viruses, 4CIRCI, 5endocan, 6monocyte distribution width, 7lymphocyte count, 8transcriptome sepsis response signatures 1 and 2, 9adaptive immunity endotypes, 10expression of glucocorticoid induced leucine zipper and dual specificity phospatatase-1, 11cutaneous vasoconstrictor to glucocorticoids, and 12 a machine learning algorithm (unclear to what outcome ML was intended to predict). 2) Randomization: it is unclear how the biomarker strata will be used in randomization and how patients will be assigned to strata. Is it that patients are first randomized to a strata category, then assigned to a strata level based on their baseline data and strata cutoff, then randomized to the intervention group? This is what figure 1 seems to show. But statements later on in the Data Analysis section seem to introduce more confusion around the way biomarker strata will be approached. For example, the statement that patients are randomized regardless of biomarker status seems to contradict the statement in lines 8-13 page 18 that “Within the randomly selected biomarker strata, the randomization algorithm then determines the assigned treatment arm: hydrocortisone and fludrocortisone versus their respective placebos”. Are patients randomized to hydorcort/fludrocort vs placebo within 1 biomarker strata or without regard to strata placement? 3) Inclusion/exclusion: Inclusion criteria are also somewhat confusing and require more detailed descriptions. a. What criteria will define “proven or suspected infection”, is this the same as Sepsis-3 suspected infection criteria (cultures drawn, antibiotics started)? b. Do patients with COVID or “proven or suspected infection” other than CAP need to have shock or vasopressor dependence or ARDS to be included? c. How is “Community Acquired Pneumonia” defined, and what will constitute “CAP-related sepsis”? d. What organ dysfunction criteria are used to define sepsis? e. Is ARDS defined by Berlin definition, and is only infection-triggered ARDs considered? f. In summary, is it correct (and potentially more clear) to state that the inclusion criteria is: either CAP+organ dysfunction, or non-CAP infection+shock or ARDS? 4) Enrollment: Testing for trial biomarkers seems like it should be a post-inclusion criteria and post-consent event – that is, patients should not be tested for study biomarkers before consent and patients who meet all inclusion criteria, but miss biomarker testing (perhaps death before biomarker testing) should be tracked and reported as study deviation and shown in CONSORT diagram, as their characteristics may differ from other study participants. In fact, collection of biomarkers is listed as a post-consent event under ‘screening visit’. Please clarify. 5) Ethics: The trial is approved by local ethics board with either consent by the patient (unlikely feasible during sepsis), or by a legally authorized representative. Please clarify that if patients are enrolled and randomized without prospective consent, then that there is approval for consent to participate in the trial to be waived for patients without a LAR, as one cannot defer consent for

	randomization and enrollment (once you are enrolled, randomized, and given study drug, without providing consent then consent is waived). It appears that deferred consent is only for subsequent data use, not randomization and receipt of the intervention. 6) Outcomes: How is competing risk of death going to be factored into the long-term HRQoL outcome assessment? 7) Sample size: why is vasopressor free days at 28 used to calculate sample size and adaptation (and shown to be primary outcome in Figure 2) when the "composite of death or persistent organ dysfunction" at 90 days is stated to be the primary outcome of the trial on page 15 lines 13-18? Also please also explain why an absolute risk difference in mortality of 10% was chosen in power calculations when it is well-known that ARR of 10% are likely overestimated and infeasible in sepsis trials (PMID: 30158216, PMID: 24786714) and the APPORACHES trial found only ARR 6% for the same interventions tested. 8) Biomarker subgroups and adaptation: How will decisions be made about whether a biomarker subgroup represents a promising or futile stratification method or when to adapt enrollment to different strata based on prior results? How will the strata subgroups be handled in the statistical analysis and what are the power calculations for the subgroup analysis? It seems likely that analysis of 12 subgroups will be underpowered with total a sample size of 1800 – will all patients be analyzed for each biomarker or just patients selected to be analyzed within each strata and randomized within the strata? 9) Study treatments: The methods state that fludrocortisone will be administered via NG tube daily. In that case, should presence of an NG tube be an inclusion criterion for the trial? Or is placement of an NG tube part of the trial procedure and discussed in the consent process? I imagine some patients will be able to take PO and some might have gastric tubes, thus route of fludrocortisone may be likely expanded in the methods. Please clarify. a. How will other common, evidence-supported treatments for severe COVID-19 be handled such as IL-6 inhibitors, JAK inhibitors and higher dose dexamethasone? Minor comment: Lines 24-27: "So far, there are no data from randomized trial evaluating endotypes-guided corticotherapy in patients with sepsis." There are data from sepsis RCTs retrospectively analyzed for corticosteroid endotypes (e.g., PMID 30365341), just no prospective RCT data.

VERSION 1 – AUTHOR RESPONSE

Comment	Response	Line number of change
Reviewer 1: Prof. Tong-Wen Sun, The first affiliated hospital of Zhengzhou University		
In this study, the heterogeneity of corticosteroid treatment in sepsis patients	Thank you for your comments.	No change.

was studied. I note that the value of biomarkers, including series levels of proteins, cellular markers, and multiomics, and of intelligent algorithms to identify the best sepsis population to be selected for corticotherapy and those who should not be treated with cortical steroids are proposed to be provided in this multicenter randomized controlled study. If this complex large-scale clinical trial can be carried out smoothly, the results obtained may screen out endotypes associated with adults with sepsis responsiveness to corticosteroids. The study protocol lists the inclusion and exclusion criteria of the trial, and the study measurements and procedures are very detailed and have clinical value.		
Reviewer 2: Dr. Allan J Walkey, , Boston University Medical Campus		
Annane and colleagues submit a study protocol for an ongoing trial to identify biomarkers of responsiveness to combined glucocorticoid (hydrocortisone, or if SARS-CoV-2, dexamethasone) and mineralocorticoid (fludrocortisone) therapy among adults with sepsis. The study is in two stages – first a prospective observational study to collect biomarkers and then an RCT. Overall the research question of finding subgroups particularly responsive to steroids in sepsis is important and use of a basket trial with adaptive	Thank you for your comments. We agree that this is the main issue of such a study that focuses on the biomarker-by-treatment interactions. Randomization is balanced, 1:1, centralized using a website, to ensure allocation concealment. Stratification of the randomization first concerns the SARS-CoV-2 status. Patients tested positive for SARS-CoV-2 patients are directly randomized to receive either dexamethasone and fludrocortisone or dexamethasone and fludrocortisone placebo. Patients tested negative for SARS-CoV-2 patients are then tested for influenza. Influenza positive patients are subsequently randomized for treatment	Pages 18-19 (Allocation Stratification), 22-23 (Data Analysis).

design is innovative. However, the description of how participants will be allocated to biomarker strata, how the subgroups within each biomarker strata will be analyzed, and how decisions will be made to drop futile biomarkers or preferentially enroll in promising biomarker strata is currently confusing and under-developed. Additional areas that require clarification are listed below.	(hydrocortisone (HC) and fludrocortisone (FC) or their respective placebos). This one-step randomization for Influenza is an amendment to the first protocol version submitted to BMJ Open and justified because of the seasonal variation of influenza related sepsis. Patients tested negative for SARS-CoV-2 and Influenza are randomly assigned to one biomarker/signature stratum among all biomarkers available for the patient, and then randomized to hydrocortisone (HC) and fludrocortisone (FC) or their respective placebos according to the list of the biomarker/signature result. This was first briefly described in the "Allocation of stratification" section. The treatment-by-biomarker interaction will be assessed using Bayesian interaction measures, either based on the Gail and Simon's statistic and the Millen's criteria derived from the ratio of the treatment effect in each stratum (Vinnat 2022), iteratively computed when the biomarker has been measured on at least 100 patients. Decision thresholds are computed based on a grid search to optimize the rate of false positive and false negative findings based on large simulated trials. All these points, accurately described in the statistical analysis plan (in the way to be published), have been more clearly reported in the revised manuscript.	
Has the observational study to identify biomarkers to use stratification been completed? Is there a publication to describe the observational biomarker selection phase?	The last patient was recruited in the observational period of the study on June 10, 2021 and his/her last follow-up occurred on December 10, 2021. Measurements of the various biomarkers (proteins, hormones, metabolome, genome, transcriptome) from blood, serum, exhaled air, circulating cells, were completed by September 2022. The subsequent months were required for completion of data entry, data review, data cleaning, database lock, generation and review of TLFs, and communication within the sponsor's organization. Statistical analysis is to be completed by March 2023 and manuscript to be submitted by June	Page 8.

	2023.	
It appears 12 different strata are listed, though the manuscript (p13, lines 10-12) states 13 strata have been identified. 1COVID, 2flu, 3other respiratory viruses, 4CIRCI, 5endocan, 6monocyte distribution width, 7lymphocyte count, 8transcriptome sepsis response signatures 1 and 2, 9adaptive immunity endotypes, 10expression of glucocorticoid induced leucine zipper and dual specificity phospatatase-1, 11cutaneous vasoconstrictor to glucocorticoids, and 12 a machine learning algorithm (unclear to what outcome ML was intended to predict).	There are 14 different strata (11 biomarker strata in addition to the COVID-19, Influenza and Other repiratory virus strata), as now corrected in the manuscript The machine learning algorithms have been developed to predict the clinical response to corticosteroids.	Page 11.
Randomization: it is unclear how the biomarker strata will be used in randomization and how patients will be assigned to strata. Is it that patients are first randomized to a strata category, then assigned to a strata level based on their baseline data and strata cutoff, then randomized to the intervention group? This is what figure 1 seems to show. But statements later on in the Data Analysis section seem to introduce more confusion around the way biomarker strata will be approached. For example, the statement that patients are randomized regardless of biomarker status seems to contradict the statement in lines 8-13 page 18 that "Within the randomly selected biomarker strata, the randomization algorithm then determines the assigned treatment arm: hydrocortisone and fludrocortisone versus	You are right, except that such a random allocation of randomization strata does not concern COVID19 patients who all have to receive open label dexamethasone as per current evidence-based recommendations, and are randomized to receive either fludrocortisone or placebo. Patients tested negative for SARS-CoV-2 and Influenza are first randomized to a strata category, then assigned to a strata level based on their baseline data and strata cutoff, then randomized to the intervention group based on this stratification list. See answer to the previous point above.	Pages 7-8, 18-19.

their respective placebos". Are patients randomized to hydrorcort/fludrocort vs placebo within 1 biomarker strata or without regard to strata placement?		
Inclusion/exclusion: Inclusion criteria are also somewhat confusing and require more detailed descriptions. a. What criteria will define "proven or suspected infection", is this the same as Sepsis-3 suspected infection criteria (cultures drawn, antibiotics started)?	It is indeed the same as Sepsis-3. We specify now that it refers to Sepsis-3 definition.	Page 9.
b. Do patients with COVID or "proven or suspected infection" other than CAP need to have shock or vasopressor dependence or ARDS to be included?	Yes, they do.	
c. How is "Community Acquired Pneumonia" defined, and what will constitute "CAP-related sepsis"?	CAP is defined as by the IDSA/ATS CAP severity criteria (table 1 of Metlay et al., 2019) CAP related sepsis is defined as CAP plus SOFA>2	Page 9.
d. What organ dysfunction criteria are used to define sepsis?	It is the SOFA score superior or equal to 2.	
e. Is ARDS defined by Berlin definition, and is only infection-triggered ARDs considered?	Yes, it is.	Page 9.
f. In summary, is it correct (and potentially more clear) to state that the inclusion criteria is: either CAP+organ dysfunction, or non-CAP infection+shock or ARDS?	It is actually CAP and organ dysfunction (SOFA >2), non-CAP sepsis (non CAP infection + SOFA>2), or sepsis and vasopressors and lactate <2 mmol/L or septic shock (Sepsis 3 definition) or sepsis- ARDS.	
Enrollment: Testing for trial biomarkers seems like it should be a post-inclusion criteria and post-consent event – that is, patients should	We have clarified the title and the text of this subsection. Biomarker testing is indeed a post-inclusion and post-consent event and prior to randomization. It is part of the initial study visit and not the	Page 10.

not be tested for study biomarkers before consent and patients who meet all inclusion criteria, but miss biomarker testing (perhaps death before biomarker testing) should be tracked and reported as study deviation and shown in CONSORT diagram, as their characteristics may differ from other study participants. In fact, collection of biomarkers is listed as a post-consent event under 'screening visit'. Please clarify.	screening visit.	
Ethics: The trial is approved by local ethics board with either consent by the patient (unlikely feasible during sepsis), or by a legally authorized representative. Please clarify that if patients are enrolled and randomized without prospective consent, then that there is approval for consent to participate in the trial to be waived for patients without a LAR, as one cannot defer consent for randomization and 1nrolment (once you are enrolled, randomized, and given study drug, without providing consent then consent is waived). It appears that deferred consent is only for subsequent data use, not randomization and receipt of the intervention.	Thank you.	Page 8.
Outcomes: How is competing risk of death going to be factored into the long-term HRQoL outcome assessment?	HRQoL will be analysed by a joint mixed model for longitudinal and survival data, that is, a shared parameter model where the HRQoL and survival models share common random effect(s). This has been reported in the revised manuscript.	Page 23.
Sample size: why is vasopressor free days at 28 used to calculate sample size and adaptation (and shown to	The sample size used the primary outcome measures of the sequential analyses, while the composite of death or persistent organ dysfunction at day 90 will	No change.

be primary outcome in Figure 2) when the "composite of death or persistent organ dysfunction" at 90 days is stated to be the primary outcome of the trial on page 15 lines 13-18? Also please also explain why an absolute risk difference in mortality of 10% was chosen in power calculations when it is well-known that ARR of 10% are likely overestimated and infeasible in sepsis trials (PMID: 30158216, PMID: 24786714) and the APPORACHES trial found only ARR 6% for the same interventions tested.	be used at the terminal analysis only. APROCCHS primary outcome was 90-day all cause-mortality and the absolute risk reduction was 6%. In this trial the primary outcome is a composite of death and persistent organ dysfunction. Thus, the ARR of 10% does not refer only to mortality reduction. In addition, the 6%ARR in APROCCHS was observed in the whole heterogenous population. In this trial, we expect endotyping guided corticotherapy to provide indeed a much greater ARR than for the non-selected population.	
Biomarker subgroups and adaptation: How will decisions be made about whether a biomarker subgroup represents a promising or futile stratification method or when to adapt enrollment to different strata based on prior results? How will the strata subgroups be handled in the statistical analysis and what are the power calculations for the subgroup analysis? It seems likely that analysis of 12 subgroups will be underpowered with total a sample size of 1800 – will all patients be analyzed for each biomarker or just patients selected to be analyzed within each strata and randomized within the strata?	We fully agree with the Reviewer regarding the lowered power of interaction tests, which is a main issue given the large number of strata in this trial. However, as reported in the manuscript, we computed that a sample of 352 patients with a measured biomarker/signature achieves 80% power to detect a minimal effect size of 0.3, with a two-sided alpha level of 0.05. We also scheduled to assess the treatment-by-covariate interaction only for biomarker/signature strata with at least 100 observations. Main interaction measures will consider only patients randomly allocated to the specific strata under study. As sensitivity analyses, we will also consider all the patients with the available biomarker, possibly handling imbalances across treatment arms using propensity score based approaches.	Pages 22-23.
Study treatments: The methods state that fludrocortisone will be administered via NG tube daily. In that case, should presence of an NG tube be an inclusion criterion for the trial? Or is placement of an NG tube part of the trial procedure and discussed in the consent	Fludrocortisone as to be given orally in patients not requiring a NG tube, and via the NG tube.	Page 19.

process? I imagine some patients will be able to take PO and some might have gastric tubes, thus route of fludrocortisone may be likely expanded in the methods. Please clarify.		
How will other common, evidence-supported treatments for severe COVID-19 be handled such as IL-6 inhibitors, JAK inhibitors and higher dose dexamethasone?	All recommended treatments in severe COVID-19 are authorized to be used in this trial. As far as we know, higher dose of dexamethasone is not recommended for the management of severe COVID-19.	Page 20.
Minor comment: Lines 24-27: "So far, there are no data from randomized trial evaluating endotypes-guided corticotherapy in patients with sepsis." There are data from sepsis RCTs retrospectively analyzed for corticosteroid endotypes (e.g., PMID 30365341), just no prospective RCT data.	Bullet point modified on demand from the editor.	Page 5.

VERSION 2 – REVIEW

REVIEWER	Walkey, , Allan J Boston University Medical Campus, The Pulmonary Center
REVIEW RETURNED	16-Feb-2023

GENERAL COMMENTS	The authors have appropriately addressed my comments.