PEER REVIEW HISTORY

ARTICLE DETAILS

TITLE (PROVISIONAL)	Multicentre, open label, randomised, controlled clinical trial comparing 2% chlorhexidine – 70% isopropanol and 5% povidone iodine – 69% ethanol for skin antisepsis in reducing surgical-site infection after cardiac surgery: the CLEAN 2 study protocol
AUTHORS	Boisson, Matthieu; CORBI, Pierre; KERFORNE, Thomas; CAMILLERI, Lionel; DEBAUCHEZ, Mathieu; DEMONDION, Pierre; ELJEZI, Vedat; FLECHER, Erwan; LABROUSSE, Louis; lepelletier, didier; Leprince, Pascal; NESSELER, Nicolas; NIZOU, Jacques Yves; OUATTARA, Alexandre; ROUSSEL, Jean Christian; Rozec, Bertrand; RUCKLY, Stéphane; Lucet, Jean-Christophe; Timsit, Jean-François; MIMOZ, Olivier

VERSION 1 – REVIEW

REVIEWER	Gaetano Privitera Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa, Italy
REVIEW RETURNED	31-Oct-2018

GENERAL COMMENTS	The study appears to be rather well planned and the protocol is accurately described. As regards the methods however some objections can be raised. The participating centres remain free to choose to apply a single-step or double step (with or without previous scrubbing) procedure for the antiseptic application and also to repeat or not the application ; this should not be necessary, since according to the literature there is no difference between the two methods. This fact should eventualy been taken into account when performing the statistical analysis of the results. It should also be advisable to restrict the choice of the participating centres as regards the antibiotic and the duration of the surgical prophylaxis (perioperative alone or postoperative admitted? which duration?). The protocol should also clearly state if the use of antimicrobial-coated sutures and antimicrobial dressings is allowed or not. If this is the case, these factors will also need to be collected and taken into account in the statistical analysis of the results since they may represent relevant confounding factors.

REVIEWER	Melissa Rochon Royal Brompton & Harefield NHS Foundation Trust, United Kingdom
REVIEW RETURNED	10-Nov-2018

GENERAL COMMENTS	A very well-written study protocol. Please review and revise:

	The primary end-point of 'any re-sternotomy' is not specific. The primary endpoint should be any occurrence SSI at 30 days (superficial incisional, sternal and donor) and 90 days (deep incisional sternal and organ/space, eg/mediastinitis), as per CDC protocol. If IE is excluded this should be stated. Will all the patients have open vein harvest or all patients have endoscopic (EVH), or all via bridge/tunnel approach? The technique will have an impact on the incidence of SSI. This point needs to be clarified. Re-sternotomy for SSI - do any of hospitals /surgeons included in the study have SOP or recommend/ practice early return to theatre for exploration for deep incisional sternal SSI? This may affect the rate of re-sternotomy - non-infected 'late' re-operation' (>72 hours), any SSI arising may not be related to the first operation - how will the authors address this? - re-operation for SSI - will subgroups be included? (dethronement, re-suturing, rewiring, corrective surgery eg/muscle flap or stratos)

REVIEWER	DT Ubbink Amsterdam University Medical Centers, The Netherlands
REVIEW RETURNED	20-Nov-2018

GENERAL COMMENTS	Being the statistics reviewer, I have some methodological comments: Relevant outcomes to be considered are: the occurrence of a documented superficial or deep infection <30 days after surgery, time to wound infection, usage of antibiotics, costs and adverse effects of the antiseptics used, and environmental consequences of their use (especially because relatively high concentrations are to be used). The 'need for re-sternotomy' may have other reasons beside infection, while not every SSI will need reoperation. The trial should yield results that enable weighing the benefits against the harms involved. When exactly will the randomisation take place? Any opportunity for the surgeon to deviate from this for whatever reason? Will the authors use decontamination of the nasopharynx and oropharynx with chlorhexidine gluconate? (Segers P et al., JAMA 2006) On which evidence is the anticipated 33% reduction in reoperation rate based? This is really a large treatment effect. Is it realistic? The authors may want to follow the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) statement.

REVIEWER	Koji Maeda Department of surgery, Jikei university school of medicine, JAPAN
REVIEW RETURNED	03-Dec-2018

GENERAL COMMENTS	This trial will be enrolled in multicenter and involves 4100 patients undergoing the cardiac surgery. This manuscript is well-written including the protocols and limitation of study. I have a few questions for authors.

	1. Please address the range of disinfection. How far is the proximal and the distal disinfection? For instance, the proximal disinfection may be performed until lower jaw when cardiac surgery. Additionally, please explain the range of disinfection when collecting the saphenous vein. I think that the range of disinfection must be the same. 2. Use of antibiotics is one of the factors affecting the occurrence of postoperative SSI. Please address in detail the use of intraoperative antibiotics. Will you administer every 3 hours during operation, and have you determined dosage by checking the eGFR? Additionally, will you change the gloves during the operation? 3. Please address the period antibiotics administration after surgery.

REVIEWER	Hui Nian Department of Biostatistics, Vanderbilt University Medical Center
REVIEW RETURNED	05-Dec-2018

GENERAL COMMENTS	This is a multicenter, randomized study to compare the antiseptic effect of 2% CHG-70% isopropanol versus 5% PVI-69% ethanol for perioperative skin preparation. The primary endpoint is the incidence of any reoperation at both surgical sites, which is clearly defined. There are a few statistical issues in the protocol. 1. Please clarify that the sample size calculation is based on the two-sided test. 2. I assume the primary analysis is the chi-square test to compare the reoperation rate between the two groups, and a marginal Cox model is the secondary analysis. Please make it clear in the analysis plan. 3. Generally speaking, it is not a good idea to adjust for the covariates that are shown imbalanced between groups. It is better to prespecify the critical potential confounders for the multivariable analysis in the analysis plan.

VERSION 1 – AUTHOR RESPONSE

Reviewer 1's comments
1- The participating centres remain free to choose to apply a single-step or double step (with or without previous scrubbing) procedure for the antiseptic application and also to repeat or not the application; this should not be necessary, since according to the literature there is no difference between the two methods. This fact should eventualy been taken into account when performing the statistical analysis of the results.

We agree with the uselessness of the double step. Nevertheless, to facilitate the recruitment and to be more in line with the real life, each centre is free to choose its procedure for the antiseptic application but should apply it for every patient and throughout the duration of the study. This fact will be taken into account.

2- It should also be advisable to restrict the choice of the participating centres as regards the antibiotic and the duration of the surgical prophylaxis (perioperative alone or postoperative admitted? which duration?)

Thanks for this comment. Actually, all participating centres must respect the French recommendation as regards as the surgical antibiotic prophylaxis (perioperative alone). We added this clarification in the revised manuscript (Page 9, lines 247-249).

3- The protocol should also clearly state if the use of antimicrobial-coated sutures and antimicrobial dressings is allowed or not. If this is the case, these factors will also need to be collected and taken into account in the statistical analysis of the results since they may represent relevant confounding factors.

Thanks for this comment. We agree with the lack of precision regarding the applied care for SSI prevention. We have modified the manuscript accordingly. (Page 10, lines 258-261).

Reviewer 2's comments
1- The primary end-point of 'any re-sternotomy' is not specific. The primary endpoint should be any occurrence SSI at 30 days (superficial incisional, sternal and donor) and 90 days (deep incisional sternal and organ/space, eg/mediastinitis), as per CDC protocol. If IE is excluded this should be stated.

Despite reoperation is not a specific end-point for SSI, we made this choice because it is a predefined strong unquestionable criterion, avoiding any evaluation bias in an open study. Furthermore, every case of reoperation will be reviewed by two independent and blinded assessors who will classify the case report as SWI, deep or superficial SSI or no SSI according to CDC criteria (Pages 10-11, Lines 287-295).

2- Will all the patients have open vein harvest or all patients have endoscopic (EVH), or all via bridge/tunnel approach? The technique will have an impact on the incidence of SSI. This point needs to be clarified.

Among participating centres, none realize endoscopic vein harvest. Finally, open vein harvest is quite rare and the majority of patients will do not have any harvest. Most of the centres use the two mammary arteries. For the peripheral harvest, radial artery is the most common. We clarified the manuscript according to this data (page 11, lines 317-318)

3- Re-sternotomy for SSI:
- do any of hospitals /surgeons included in the study have SOP or recommend/ practice early return to theatre for exploration for deep incisional sternal SSI? This may affect the rate of re-sternotomy
- non-infected 'late' re-operation' (>72 hours), any SSI arising may not be related to the first operation
- how will the authors address this?
- re-operation for SSI - will subgroups be included? (dethronement, re-suturing, rewiring, corrective surgery eg/muscle flap or stratos)

- To date, no participating centre practice early return to the theatre for exploration of wound sternal infection.
- Patients will stay in the study up to three months after surgery. Any SSI arising during this period with be related to the antiseptic group.
- Unfortunately, we did not include these types of subgroups.

Reviewer 3's comments

1- Relevant outcomes to be considered are: the occurrence of a documented superficial or deep infection <30 days after surgery, time to wound infection, usage of antibiotics, costs and adverse effects of the antiseptics used, and environmental consequences of their use (especially because relatively high concentrations are to be used). The 'need for re-sternotomy' may have other reasons beside infection, while not every SSI will need reoperation. The trial should yield results that enable weighing the benefits against the harms involved.

Thanks for this comment. Reoperation is a predefined strong unquestionable criterion and it avoids any evaluation bias in an open study. We agree that a part of reoperation will not be link with SSI, but with randomization, “no-SSI reoperation” will be equally in the two groups. Furthermore, every case of reoperation will be reviewed by two independent and blinded assessors who will classify the case report as SWI, deep or superficial SSI or no SSI according to CDC criteria (Pages 10-11, Lines 287-295). As regard as SSI without reoperation they will be detected by the monitoring of wound aspect, use of antibiotic and bacteriological sampling.

2- When exactly will the randomisation take place? Any opportunity for the surgeon to deviate from this for whatever reason?

The randomization will take place very close to the day of surgery (few days before, the day before or the day of surgery depending on local organization) and we added this information in the manuscript (page 9, lines 226-227). There is no opportunity for the surgeon to deviate from the assigned intervention and in order to ensure respect of treatment group, individual boxes containing all disinfecting products required for disinfecting the skin before surgery and during patients’ care will be supplied and will follow him from the operating room to hospital discharge.

3- Will the authors use decontamination of the nasopharynx and oropharynx with chlorhexidine gluconate? (Segers P et al., JAMA 2006) ?

Each participating centre is free to provide others preventive cares. Before the beginning of inclusion a list of applied care for prevention of SSI (Staphylococcus aureus decontamination, antimicrobial-coated sutures, adhesive incises drapes…) will be established and will not be modified throughout the duration of the study. We added this information in the manuscript (page 10, lines 258-261). To date, no participating centre use decontamination of the nasopharynx and oropharynx with chlorhexidine gluconate. But according to the WHO’s recommendations, the majority of them use nasal decontamination with mupiricin.

4- On which evidence is the anticipated 33% reduction in reoperation rate based? This is really a large treatment effect. Is it realistic?

According to our expertise and literature, reoperation rate after cardiac surgery is around 6% with more than half of them due to SSI (Lemaignen et al. Clin Microbiol. Infect 2015).The anticipated 33% reduction in reoperation rate is based on previous studies which had shown 41 to 45% reduction in SSI with chlorehexidine-alcohol (Darouiche et al. NEJM 2010, Tuuli et al. NEJM 2016).

5- The authors may want to follow the Standard Protocol Items: Recommendations for Interventional Trials (SPIRIT) statement.

We completed the SPIRIT checklist and we have joined it with the revision. The missing items have now been added to the manuscript

Reviewer 4’s comments

1- Please address the range of disinfection. How far is the proximal and the distal disinfection? For instance, the proximal disinfection may be performed until lower jaw when cardiac surgery. Additionally, please explain the range of disinfection when collecting the saphenous vein. I think that the range of disinfection must be the same.

The surgical field extends from the jaw to the shoulders and down to the tip of both feet in case of surgery with harvesting of the saphenous vein. In the event of surgery without saphenous vein harvesting, the field stops at the knees. We added this information in the manuscript (page 9, lines 251-254).

2- Use of antibiotics is one of the factors affecting the occurrence of postoperative SSI. Please address in detail the use of intraoperative antibiotics. Will you administer every 3 hours during operation, and have you determined dosage by checking the eGFR? Additionally, will you change the gloves during the operation?

According to the French recommendations (https://sfar.org/antibioprophylaxie-en-chirurgie-et-medecine-interventionnelle-patients-adultes-maj2018/), cefazolin (2g 30 min prior to incision + 1g at the priming) or cefuroxime/cefamandol (1.5g 30 min prior to incision + 0.75g at the priming) will be administered intravenously to patients. No dosage modification will be done according to eGFR but dose will have to be doubled for patients with BMI over 35 Kg/m2. Reinjection will have to be performed every four hours during surgery. Antibiotic will be not prolonged after the end of surgery. For patients with beta-lactam allergy or with MRSA carriage, vancomycin (30 mg/kg 120 min prior to incision) will be preferred. Reference to the French recommendations on antibiotic prophylaxis during surgery has been added to the revised manuscript (Page 9, line 247).

3- Please address the period antibiotics administration after surgery

According to the French recommendations (https://sfar.org/antibioprophylaxie-en-chirurgie-et-medecine-interventionnelle-patients-adultes-maj2018/). Antibiotic will be stopped at the end of surgery. (Page 9, line 249)

Reviewer 5's comments
1- Please clarify that the sample size calculation is based on the two-sided test.

Thanks for this suggestion. It's done (page 13, line 359)

2- I assume the primary analysis is the chi-square test to compare the reoperation rate between the two groups, and a marginal Cox model is the secondary analysis. Please make it clear in the analysis plan.

It's true. We now make it clear in the analysis plan (page 13, lines 364-365).
3- Generally speaking, it is not a good idea to adjust for the covariates that are shown imbalanced between groups. It is better to prespecify the critical potential confounders for the multivariable analysis in the analysis plan.

Thank you with this comment. We agree with the reviewer's comment and we modified the analysis plan in according with his suggestion. (pages 13-14, lines 370-380)

VERSION 2 – REVIEW

REVIEWER	Gaetano Privitera Department of Translational Research and New Technologies in Medicine and Surgery, University of Pisa and Pisa University Hospital, Italy
REVIEW RETURNED	04-Feb-2019

GENERAL COMMENTS	The revised version submitted responds adequately to the comments by the reviewers and may be published in the present form.

REVIEWER	D Ubbink Amsterdam University Medical Centers, location AMC, Amsterdam, The Netherlands
REVIEW RETURNED	24-Jan-2019

GENERAL COMMENTS	Thank you for your revisions. It has substantially improved the manuscript. I still have concerns about achieving the expected 33% reduction in reoperation rate of 6%, of which half are due to SSI. Darouche et al. found a much higher absolute SSI risk (9.5-16%) than the expected 6% here, which the authors want to further reduce to 4%. If this difference is not reached (which is quite possible, particularly because the authors accept a 20% risk of a type-II error), which 'secondary' endpoint will then be chosen to decide upon which antiseptic to choose? Or just based on the likely cost difference? The authors might want to address this in the manuscript. Detail: 'saphen venous' should read 'saphenous vein'; 'COMPETITING' should read 'COMPETING'.

REVIEWER	KOJI MAEDA Jikei university school of medicine, Tokyo, JAPAN
REVIEW RETURNED	04-Feb-2019

GENERAL COMMENTS	The authors have improved their manuscript. All of my questions have been resolved.

REVIEWER	Hui Nian Department of Biostatistics, Vanderbilt University Medical Center, USA
REVIEW RETURNED	28-Jan-2019

GENERAL COMMENTS	Concerns have been addressed, and I have no further comments.

VERSION 2 – AUTHOR RESPONSE

In order to address the reviewer's concerns, we propose to add the following paragraph in the discussion: "We assumed a 33% reduction in reoperation with the use of alcoholic chlorhexidine in our study. This choice may appear too ambitious. However, it is based on the existence of several surgical sites in the majority of patients, the major role of SSI in reoperation and the expected effect of antiseptic choice on SSI prevention. In clean contaminated surgery, a 50% reduction in SSI with

alcoholic chlorhexidine use has been reported in digestive[7] or obstetrical[9] surgery. In these types of surgery, a significant fraction of pathogens involved comes from the digestive or gynaecological flora not accessible to the action of antiseptics. In intensive care, an 85% reduction in infections related to short-term central venous and arterial catheters has been reported with alcoholic chlorhexidine use.[19] As in clean surgery, the skin flora is the main reservoir of pathogens involved in these infections, and the effectiveness of skin disinfection is essential to prevent them. In total, if we consider that among the 6% of reoperation in the povidone iodine group, half are related to an SSI (which is probably underestimated), we can expect an incidence of reoperation in the chlorhexidine group between 3.5% (hypothesis very favourable to alcoholic chlorhexidine use) and 4.5% (hypothesis not very favourable to alcoholic chlorhexidine use). In the event of negative results, the choice of the antiseptic strategy could be based on the incidence of secondary endpoints in both arms of our study, and finally, on the cost of antiseptic strategies, even if it is insignificant compared to that of SSI." (Lines 451-468)

We also corrected "saphen venous" throughout the manuscript. We did not find "competiting".

Finally we up dated the number of patients included.