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Supplement to Manuscript Submission JAMA Surgery

“Intraoperative wound irrigation for the prevention of surgical site infection after laparotomy

The multicenter, double-blind, randomized controlled IOWISI trial (DRKS00012251) of the Study Centre of the German Surgical Society (SDGC CHIR-Net)”

This supplement contains the following items:

1. Clinical study protocol (CSP)

- 1.1 CSP original version 2.0 (06.06.2017) Page 1-41
- 1.2 CSP final version 3.0 (02.03.2021) Page 42-82
- 1.3 CSP summary of changes Page 83

2. Statistical analysis plan (SAP)

- 2.1 SAP original version 1.0 (28.04.2021) Page 84-95
- 2.2 SAP final version 1.1 (08.11.2022) Page 96-107
- 2.3 SAP summary of changes Page 108

25 **1. Study Protocol**

26 **1.1 CSP original version 2.0 (06.06.2017)**

27

TECHNISCHE UNIVERSITÄT MÜNCHEN KLINIKUM RECHTS DER ISAR

STUDY PROTOCOL

IOWISI

Intraoperative wound irrigation to prevent surgical site infection after laparotomy

Sponsor:

Technische Universität München (TUM)
School of Medicine
Represented by the Dean
Ismaninger Str. 22
81675 München, Germany

Coordinating Investigator (LKP):

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EudraCT Number: 2017-000152-26

Study Code: IOW-1755-REI-0540-I

DRKS Number: DRKS00012251

Version: Final 2.0

Date 06.06.2017

! Confidential !

This document is confidential and should serve as a source of information for Investigators and other personnel involved in this clinical study, consultants and ethics committees and regulatory authorities. The contents of this document shall only be disclosed to others in agreement with the coordinating investigator and/or sponsor.

28

29 Responsibility

30

Protocol development	Sponsor	Technische Universität München School of Medicine Represented by the Dean Ismaninger Str. 22 81675 München Tel.: 089 4140-4020 Fax: 089-4140-4935 E-Mail: dekanat.medizin@tum.de
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	Project management Monitoring Data management Safety management	Münchner Studienzentrum (MSZ) Ismaninger Str. 22 81675 München Tel: 089 4140-6321 Fax: 089 4140-6322 Email: Beate.schossow@mri.tum.de

31

32 **1. DECLARATION OF INVESTIGATOR**

33

34 I have read the trial protocol and I confirm that it contains all information to accordingly conduct
35 the clinical trial. I pledge the clinical trial will be conducted at my trial center according to the
36 protocol.

37

38 The first patient will be enrolled only after all ethical and regulatory requirements are fulfilled. I
39 pledge that written informed consent for trial participation will be obtained from all patients.

40

41 I know the requirements for accurate notification of serious adverse events and I pledge to
42 document and notify such events as described in the protocol.

43

44 I pledge to retain all trial-related documents and source data as described. All necessary
45 documents will be provided before trial start. I agree that these documents will be submitted to
46 the responsible regulatory authorities and ethics committees.

47

2. SYNOPSIS

Sponsor	Technische Universität München, School of Medicine
Name of the trial	Intraoperative wound irrigation to prevent surgical site infection after laparotomy - IOWISI
Trial design	Prospective, randomized, controlled, observer and patient-blinded, multicenter, surgical trial according to German drug law (AMG) phase IIIb, with three parallel comparison groups
Objectives	To investigate whether the use of intraoperative, epifascial wound irrigation with polyhexanide (PHX) solution can reduce surgical site infections after laparotomy for visceral surgery compared to saline irrigation or no irrigation.
Interventions	<p><u>Experimental intervention/index test:</u></p> <ul style="list-style-type: none"> Intervention 1: Irrigation of the subcutaneous tissue after closure of the abdominal fascia with 1000ml PHX solution (0.04%) Intervention 2: Irrigation of the subcutaneous tissue after closure of the abdominal fascia with 1000ml saline solution (NaCl 0.9%) <p><u>Control intervention/reference test:</u> No epifascial wound irrigation</p> <p><u>Follow-up per patient:</u> Postoperative day 30 (+6 at the latest)</p> <p><u>Duration of intervention per patient:</u> One intraoperative application</p> <p><u>Experimental and/or control off-label or on-label in Germany:</u> All interventions are on-label in Germany</p>
Key inclusion and exclusion criteria	<p><u>Key inclusion criteria:</u></p> <ul style="list-style-type: none"> Clean-contaminated, contaminated or dirty surgery (class II-IV) according to Centre for Disease Control (CDC) classification; Abdominal surgery by midline or transverse laparotomy; elective and emergency procedures; Age \geq 18 years; American Society of Anesthesiologists (ASA) score \leq 3; Ability to understand the nature and extent of the trial and to give written informed consent; <p><u>Key exclusion criteria:</u></p> <ul style="list-style-type: none"> Pregnancy or breast feeding; Known hypersensitivity/allergy to PHX; Inability to give/understand informed consent; Critical medical condition of emergency patients, precluding informed consent or sufficient time to reflect on the decision to participate in the trial; ASA $>$3; Inability to attend follow-up visits; Clean procedures according to the CDC classification or surgery without opening of the abdominal cavity; Laparoscopic surgery; Revision-surgery (previous abdominal surgery within the last 30 days); Planned re-laparotomy within 30 days; Severe immunosuppression; Concurrent abdominal wall infections; Pre-operative systemic antibiotic therapy within 5 days prior to surgery (except emergency pre-operative antibiotic treatment due to septic peritonitis after admission to the hospital); Participation in another clinical trial that interferes with the primary or secondary outcomes of this trial.
Outcomes	<p><u>Primary efficacy endpoint:</u> SSI according to CDC criteria within 30 days postoperatively</p> <p><u>Key secondary endpoint(s):</u></p> <ul style="list-style-type: none"> Non-infectious wound complications (e.g. seroma, hematoma, delayed healing) within 30 days postoperatively

	<ul style="list-style-type: none"> • Duration of hospital stay • Mortality and morbidity within 30 days postoperatively • Incidence of reoperation within 30 days postoperatively • Incidence of AE/SAE within 30 days postoperatively <p>Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space), NNSI risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of wound-edge protectors and changing of gloves, presence of an enterostomy.</p> <p><u>Safety:</u> Adverse events (AE) and serious adverse events (SAE) are documented for all groups. Surgical complications will be additionally evaluated according to the Clavien-Dindo classification</p>
Study registry	German CTR (DRKS): DRKS00012251 / EudraCT: 2017-000152-26
Statistical analysis	<p><u>Efficacy:</u> The incidence of SSI within 30 days after surgery will be compared between three study groups in two ways: Test 1: PHX irrigation vs. saline irrigation Test 2: PHX irrigation vs. no intervention</p> <p><u>Description of the primary efficacy analysis and population:</u> The incidence of SSI within 30 days of surgery will be compared in test 1 and test 2 using the Fisher Exact test. Both tests will be performed on the ITT set, consisting of all patients included in the study in the treatment arm they were randomized to. First analysis will be based on all patients with complete follow-up. For sensitivity, multiple imputations for missing primary endpoint data will be used. The tests will be performed two-sided and with a global significance level of 5%. Using the Bonferroni-Holm adjustment, the local significance level for test 1 will be 2.5% and for Test 2 it will be 5%.</p> <p><u>Safety:</u> The assessment of safety will be based on the frequency of AE/SAE other than SSI within the safety population (according to CTCAE Version 4.03), consisting of all patients randomized into the study.</p> <p><u>Secondary endpoint(s):</u> Secondary endpoints will be analyzed on the ITT set using appropriate descriptive statistics. Any explorative statistical testing will be performed two-sided using a significance level of 5%.</p>
Sample size	<p><u>To be assessed for eligibility (n):</u> approximately 1500 <u>To be assigned to the trial (n):</u> 540 <u>To be analyzed (n):</u> 540</p> <p>The sample size was calculated assuming 30-day SSI rates of 2.2% in the PHX group, 8.7% in the saline group, and 16.2% in the control group. If 230 patients are recruited in the PHX group, 230 patients in the saline group and 80 patients in the no irrigation group (a total of 540 patients), the two-sided Fisher exact test with a global significance level of 5% will have a power of 94% for test 1 ($\alpha=2.5\%$) and a power of 85% for test 2 ($\alpha=5\%$) to detect differences between the treatment groups.</p>
Trial duration subject	<p><u>Intervention:</u> Single intraoperative intervention <u>Follow-up:</u> max. 36 days</p>
Trial duration project	<p><u>First patient in to last patient out (months):</u> 28 <u>Recruitment period (months):</u> 27 <u>Duration of the entire trial (months):</u> 42</p>
Participating centers	Planned: n= 10
Financing	Deutsche Forschungsgemeinschaft (DFG) grant number: MU 3928/1-1

3. ABBREVIATIONS

51	AE	Adverse Event
52	ALT/ALAT	Alanine Aminotransferase
53	AMG	Arzneimittelgesetz
54	aPTT	Activated partial Thromboplastin time
55	ASA	American Society of Anesthesiologists
56	AST/ASAT	Aspartate Aminotransferase
57	BfArM	Bundesinstitut für Arzneimittel und Medizinprodukte
58	BMI	Body-Mass Index
59	CDC	Centre for Disease Control and Prevention
60	CI	Confidence Interval
61	Cr	Creatinine
62	CTCAE	Common Terminology Criteria for Adverse Events
63	DFG	Deutsche Forschungsgemeinschaft
64	DRKS	Deutsches Register Klinischer Studien
65	dsUR	Development Safety Update Report
66	eCRF	electronic Case Report Form
67	EDTA	Ethylene-diamineteraacetic acid
68	GCP	Good Clinical Practice
69	Glu	Glucose
70	ICF	Informed consent form
71	ICH	International Conference on Harmonization
72	ICMJE	International Committee of Medical Journal Editors
73	IMP	Investigational Medicinal Product
74	IMSE	Institut für Medizinische Statistik und Epidemiologie
75	INR	International normalized ratio
76	IOWI	Intraoperative wound irrigation
77	ISF	Investigator site file
78	ITT	Intention-To-Treat
79	K	Potassium
80	MeSH	Medical Subject Heading
81	MRI	Klinikum München rechts der Isar
82	MSZ	Münchener Studienzentrum
83	Na	Sodium
84	NaCl	Sodium chloride
85	NICE	National Institute for Health and Clinical Excellence
86	NNIS	National Nosocomial Infections Surveillance
87	PHX	Polyhexanide
88	PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
89	PT	Prothrombin time
90	PVP	Polyvinylpyrrolidone, Povidone
91	RCT	Randomized Controlled Trial
92	RDE	Remote Data Entry
93	SAE	Serious Adverse Event
94	SAR	Serious Adverse Reaction
95	SAS	Statistical analysis system
96	SGOT	Serum glutamic oxaloacetic transaminase
97	SGPT	Serum glutamic pyruvic transaminase
98	SMB	Safety Monitoring Board
99	SmPC	Summary of product characteristics
100	SOP	Standard operating procedure
101	SSI	Surgical site infection
102	SUSAR	Suspected Unexpected Serious Adverse events
103	TUM	Technical University of Munich
104	WHO	World Health Organization
105		

106 4. INTRODUCTION

107 4.1 The medical problem

108 Postoperative surgical site infection (SSI) represents the third most common hospital infection.
109 According to the CDC's classification [1], SSI can be subdivided into infections of the
110 subcutaneous tissue (superficial SSI), deep soft tissues such as fascial and muscle layers
111 (deep SSI) and infections of organs or spaces (organ/space SSI) that occur within 30 days after
112 surgery (attachment 1). In abdominal surgery, SSI rates are especially high. Recent high-level
113 randomized controlled trials (RCTs) with standardized SSI definitions found rates between
114 14.5% (BaFO trial) [2], 15.4% (PROUD trial) [3] and 25.0% (ROSSINI trial) [4] following
115 laparotomy. Therefore, measures to prevent SSI in this field are urgently needed. Prophylactic
116 intraoperative wound irrigation (IOWI) of the subcutaneous and deep soft tissue before skin
117 closure with saline or antiseptic solutions hypothetically represents an easy and economical
118 option to reduce SSI rates and is already frequently used in clinical practice, even though there
119 are currently no definite recommendations on this practice [5]. The latest official guideline for the
120 prevention of SSI by the World Health Organization (WHO) published in 2016, states that IOWI
121 with saline is not efficient, but IOWI with diluted Polyvinylpyrrolidone (PVP)-iodine solutions has
122 a potential benefit in preventing SSI, however, due to the low level of underlying evidence these
123 recommendations are conditional and not limited to abdominal surgery [6]. In contrast, the
124 clinical guidelines of the British National Institute for Health and Clinical Excellence (NICE) from
125 2008 state that IOWI's efficacy is unproven and its use should be avoided at all. However, this
126 recommendation too, is based on a small number of unstandardized RCTs evaluating different
127 types of surgery and irrigation solutions [7]. Antiseptic PHX-based solutions are approved for
128 intraoperative soft-tissue wound irrigation in surgery, and have been shown to be tissue
129 tolerable and even promote wound healing. To our knowledge prophylactic PHX wound
130 irrigation has not yet been evaluated in RCTs in abdominal, visceral surgery [8, 9].

132 4.2 Evidence

133 Even though the literature concerning prevention of SSI is substantial, high-level evidence to
134 guide decisions on the use of IOWI with saline or antiseptics remains scarce. Clinical trials
135 investigating the efficacy of IOWI have been conducted mainly in the 1980-90's and their results
136 are inconclusive and heterogeneous patient inclusion and outcome criteria were used. A few
137 authors conducted systematic reviews and meta-analyses investigating specific irrigation
138 solutions such as PVP-iodine or antibiotic solutions [10-13]. However, none of these reviews
139 resulted in a definite conclusion, although they all observed a positive trend in the reduction of
140 SSI rates through IOWI. Furthermore, more recent clinical trials have been conducted in the
141 meantime. Therefore, we performed a large-scale meta-analysis in accordance with the
142 Cochrane guidelines of the existing evidence on IOWI with saline, PVP-iodine or antibiotic

143 irrigation solutions. Pubmed/MEDLINE, EMBASE, and the Cochrane Central Register of
144 Controlled Trials (CENTRAL) were searched in May 2013. The following search terms were
145 used in various combinations: prevention of surgical site infection, abdominal surgery, surgical
146 wound infection/prevention and control [MeSH Terms], wound irrigation, wound lavage,
147 incisional surgical site infection, intra operative irrigation, intra operative lavage, antibiotic
148 irrigation, antibiotic irrigation solutions, iodine irrigation, povidone iodine irrigation, saline
149 irrigation, and topical anti-infective agents [MeSH Terms]. The abstract and title search was
150 limited to clinical trials published in English or German between January 1, 1970 and May 1,
151 2013. In addition, all articles within the reference list of retrieved studies and reviews were
152 hand-searched. The search was performed by two independent reviewers and followed the
153 published protocol corresponding to the PRISMA statement and the Cochrane Handbook of
154 systematic reviews of interventions. Prospective RCTs investigating the primary outcome of
155 postoperative SSI after IOWI of the surgical incision after closure of the fascia or peritoneum
156 and before skin closure were eligible for inclusion. Eligible irrigation solutions were saline, PVP-
157 iodine, or topical antibiotics in different forms and concentrations (dry powder sprays or wound
158 powder were also acceptable), irrespective of the closure and irrigation technique. Acceptable
159 comparators were 'no irrigation' or irrigation with saline. All types of open abdominal surgeries
160 were eligible, including visceral, gynecological, urological, or vascular procedures irrespective of
161 the urgency of operation (elective or emergency). All trials reporting clinical SSI were included
162 irrespective of the SSI definition used. Trials in which only one of the compared treatment arms
163 received systemic prophylactic antibiotics were excluded, as this would have caused substantial
164 bias. Methodological quality of individual clinical trials was assessed by examination of the
165 allocation sequence, allocation concealment and double blinding using the Cochrane tool for
166 assessing the risk of bias [21]. The risk of bias was graded as low, unclear, or high. In addition,
167 the risk of publication bias was investigated by means of a funnel plot. Due to the naturally
168 expected heterogeneity in performance of surgical procedures between different types of
169 surgery, grade of contamination, and hence trials, random effect models with Mantel-Haenszel
170 weights were used to estimate the average treatment effect and a corresponding 95 % CI.
171 Forest plots were shown to illustrate treatment effects estimated for each trial and the estimated
172 average treatment effect for all investigated subgroups. A two-sided level of significance of less
173 than 5.0 % was considered for all tests. The results of this analysis show a risk reduction of 46
174 % in the treatment group (IOWI with any irrigation solution). Incidence of SSI was 9% in the
175 irrigation group compared to 16% in the untreated group [14]. However, the majority of included
176 trials have been published from 1970 to 1990, and the quality assessment revealed that most of
177 them were at a high risk of bias, mainly because of insufficient data reporting and
178 methodological flaws. Methods of sequence generation, allocation concealment, and blinding
179 were often inadequate or not reported. In addition, interventions, follow-up times, and definitions

180 of SSI varied widely between studies, which might explain the large variance in overall SSI rates
181 between 3.0 and 58.2%. Most studies used a non-standardized definition of SSI. The current
182 internationally accepted CDC definition was not published until 1999. The funnel plot showed an
183 asymmetry, which indicates a possible publication bias, as all included trials with a high
184 standard error for the log odds ratio show a large benefit for the experimental group.
185 Furthermore, PVP-I and antibiotic solutions are currently not recommended for this indication
186 due to potential adverse side effects, tissue toxicity and the increased development of
187 antimicrobial resistances. The only standardized RCT comparing IOWI with saline irrigation vs.
188 no irrigation after open appendectomies was published in 2000 and found a reduction of SSI
189 from 25% to 8.7% in the saline group [15]. Recently, PHX-based antiseptic solutions are
190 successfully and widely used in orthopedic and trauma surgery. Wound irrigation with PHX
191 showed a reduction of the SSI rate of almost 75% compared to Ringers solution in traumatic
192 dirty contaminated soft tissue wounds [16].

193

194 **4.3 The need for a trial**

195 SSIs contribute significantly to postoperative morbidity and mortality. In Germany approximately
196 128,000 SSIs are reported annually [17]. Studies have shown an increase of 6-24 days in the
197 mean length of hospital stay if SSI occurs [18]. In addition to the risk and discomfort for the
198 patient, SSIs dramatically increase treatment costs and indirect costs such as loss of workforce
199 or insurance payments. In Germany, postoperative SSIs account for approximately 1 million
200 extra days of hospitalization and additional costs of around € 3 billion per year [19, 20]. Clinical
201 guidelines and clinical practice vary largely in terms of the use of IOWI to reduce the incidence
202 of SSI [5]. The aim of this prospective, multicenter, randomized clinical trial is to show the
203 reduction of SSI rates by IOWI with PHX compared to saline or no irrigation. Individual patients
204 participating in this trial have the opportunity of directly benefitting of the anticipated positive
205 effect of PHX and/or saline irrigation, whilst no negative effects are to be expected. The results
206 of the trial will provide evidence for definite clinical recommendations that would change current
207 clinical guidelines and practice. A commercial interest is not expected as PHX solutions are
208 widely available and several companies offer this product in their portfolio. The trial further does
209 not request a certain product in order to avoid compliance conflicts, but encourages
210 collaborators to use the available product in their respective study sites.

211

212 **4.4 Summary and aims of the study**

213 SSI is one of the most common complications following abdominal visceral surgery (14-25%) [2-
214 4, 21] and dramatically increases length of hospital stay and costs. Hypothetically, IOWI before
215 skin closure with saline or antiseptics might be a potential pragmatic option to reduce SSI rates.
216 Currently, there are no official recommendations on its use and clinical practice varies largely.

217 Solutions containing the antiseptic agent PHX are approved for IOWI, and were shown to
218 promote wound healing [8, 9], but have not been evaluated in RCTs in abdominal visceral
219 surgery. Therefore, we designed a multicenter, randomized, observer-blinded clinical trial
220 evaluating the efficacy of IOWI with PHX solution or saline before skin closure after laparotomy.
221 Based on a meta-analysis on IOWI with various solutions, a sample-size of 540 patients was
222 calculated for a 3-armed study design (PHX- vs. saline irrigation vs. no irrigation). The trial shall
223 be conducted in 10 centers within the German surgical trial network *CHIR-Net*. All patients
224 undergoing visceral surgery by laparotomy within the recruitment period of 27 months will be
225 screened for the trial. The primary endpoint is the incidence of SSI 30 days postoperatively,
226 according to the CDC definition (attachment 1). The results of the trial will provide evidence for
227 definite clinical recommendations regarding the use of IOWI and influence current guidelines
228 and provide all participating patients the opportunity of an improved treatment.
229

230 **5. OUTCOME MEASURES**

231 **5.1 Rationale of outcome measures**

232 The primary efficacy endpoint of this trial is SSI within 30 days postoperatively, according to the
233 internationally accepted and recommended SSI definition by the CDC [1]. This endpoint has
234 been used in previous trials and assures comparability of the results [2-4, 21]. This endpoint is
235 further considered to be of clinical relevance as SSI increases morbidity and mortality of
236 individual patients, direct and indirect costs and prolongs hospital stay as outlined before. The
237 secondary endpoint of non-infectious wound complications was chosen to evaluate, if PHX
238 irrigation has an additional positive effect on wound healing. Furthermore, secondary endpoints
239 are morbidity and mortality within 30 days postoperatively. For safety analyses and the duration
240 of hospital stay to evaluate the potential economical benefit.

241

242 **5.2 Determination of primary and secondary measures**

243 The primary efficacy endpoint measure of the trial is the incidence of SSI within 30 days after
244 surgery diagnosed. Furthermore, in case of SSI, the depth of infection will be classified into one
245 of three categories according to CDC definition (superficial, deep, organ-space, see attachment
246 1). In addition, the following outcome measures have been defined as secondary endpoint
247 measures and will be determined by the unit given in parentheses: a) Duration of hospital stay
248 (in days); b) 30-days rate of reoperation in both groups (%); c) 30-days rate of non-infectious
249 wound complications in both groups (in %); d) 30-days rate of postoperative AE/SAE in both
250 groups (%); e) 30-days mortality in both groups (%); (f) 30-days morbidity in both groups (%). All
251 AE/SAEs that are surgical complications will be additionally classified according to the Clavien
252 Dindo classification of surgical complications (attachment 2) [22].

253

254 **6. FINANCING**

255 The clinical trial is financed by a grant from the German Research Society (Deutsche
256 Forschungsgemeinschaft; DFG), grant number: MU 3928/1-1. No co-financing by industry or
257 other third parties applies. There is no conflict of interest for the management of the study. All
258 participating trial sites have officially declared no conflict of interest within the eligibility
259 evaluation of the MSZ. A commercial interest does not apply as PHX solutions are widely
260 available and several companies offer this product in their portfolio. The trial further does not
261 request a certain product in order to avoid compliance conflicts, but encourages collaborators to
262 use the available product in their respective study sites.

263

264 **7. RISK / BENEFIT ANALYSIS**

265 No additional risks for study patients are anticipated, since IOWI represents a clinically
266 established standard method. PHX 0.04% irrigation solution is approved for surgical wound
267 irrigation of soft tissue wounds. The study will be planned, conducted and analysed according to
268 all relevant national and international rules and regulations according to AMG [23], ICH-GCP E6
269 [24], and the Declaration of Helsinki, 2008 (see 27.). No specific risks are expected because
270 IOWI is locally applied and neither application of PHX or saline will have systemic effects on the
271 participants. Safety of PHX solutions has been demonstrated before in the marketing studies.
272 Adverse effects may only be expected in the improbable event of accidental contamination of
273 the respective irrigation solutions or in case of unknown hypersensitivity to PHX. The potential
274 benefits of reduced SSIs outweigh the mentioned negligible adverse effects of PHX and saline.
275 The subjects' safety is ensured by regular study visits, enforcing GCP-guidelines. A subject-
276 insurance for all trial participants is mandatory according to AMG. The informed consent
277 process adheres to GCP-guidelines, which maximize patients' safety and guarantee
278 confidentiality.

279

280 **8. TRIAL IMPLEMENTATION**

281 **8.1 General study design**

282 This study is a prospective, randomized, controlled, observer and patient-blinded, multicenter,
283 surgical trial with three parallel comparison groups. Pre-screening of potential patients
284 (evaluation of inclusion and exclusion criteria) is possible up to 14 days prior to the planned
285 procedure. Patients can be included in the trial if inclusion and exclusion criteria apply and

286 written informed consent has been provided. In case of emergency procedures inclusion is
287 possible on the same day as the procedure, if the patient is able to understand and provide
288 written informed consent and has had a reasonable amount of time to think about the decision
289 (see 12.3). Included patients are randomized to no epifascial wound irrigation, epifascial wound
290 irrigation with saline 0.9% or epifascial wound irrigation with PHX 0.04% solution. Screened but
291 excluded patients will be documented in a screening log.

292

293 **8.2 Trial duration**

294 The estimated overall length of the study is 42 months, which assembles as follows:

295 I. Trial preparation: ~ 6 months

296 II. Execution of study: First patient in to last patient out: ~ 28 months

297 1. Begin of study: 3rd quarter, 2017

298 2. End of study: 4th quarter, 2019 (Completion of the last visit for the last patient
299 represents the end of study)

300 3. Recruitment period: ~ 27 months

301 4. Duration of treatment per patient:

302 a) Group with intervention 1: Surgery according to institutional standard, followed by
303 one-time wound irrigation with PHX 0.04% solution.

304 b) Group with intervention 2: Surgery according to institutional standard, followed by
305 one-time wound irrigation with saline 0.9% solution.

306 c) Control group: Surgery according to institutional standard, followed by no wound
307 irrigation.

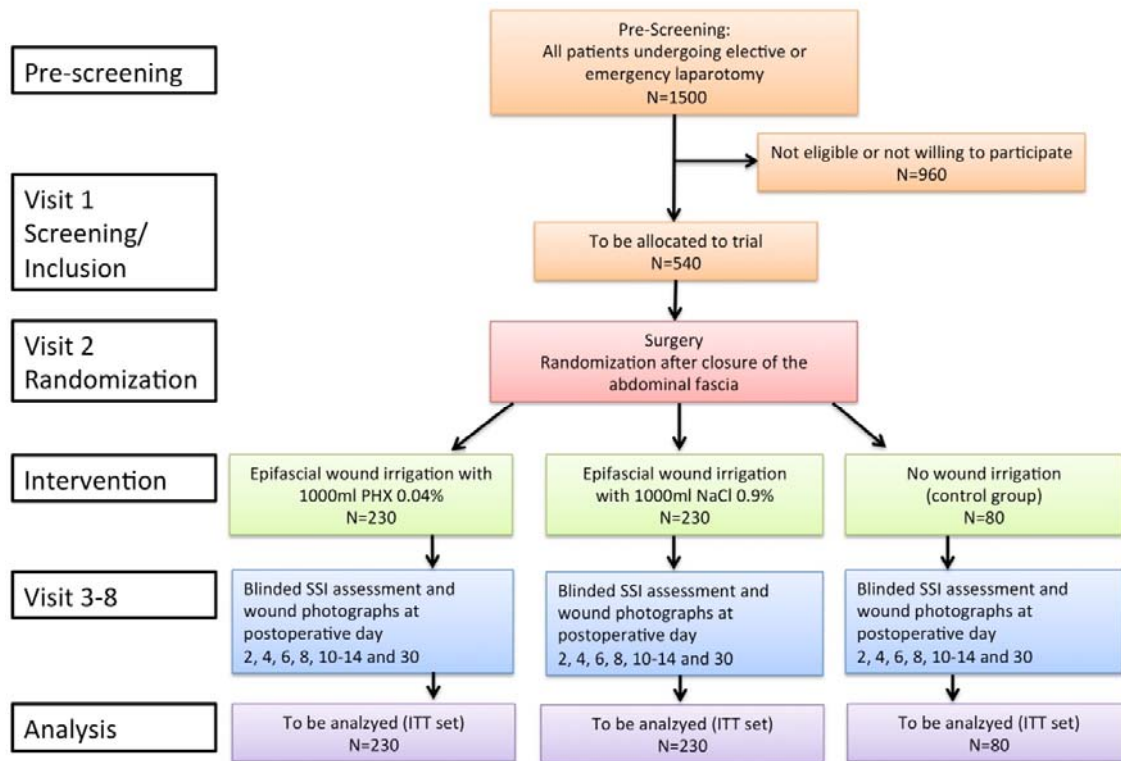
308 5. Duration of follow-up per patient: 30 days (+5 days at the latest)

309 For all three groups, documentation of the primary and secondary endpoints up to
310 postoperative day 30 is warranted.

311 III. Analysis, publication ~ 8 months

312

313 **Graph 1: IOWISI intervention scheme / trial flow**



314

315

316

Graph 2: IOWISI study visits (according to SPIRIT statement 2013 [25])

	STUDY PERIOD							
	INCLU.	RAND.	POST-ALLOCATION					CLOSE-OUT
STUDY VISIT	1	2	3	4	5	6	7	8
TIMEPOINT	- 1-3 days*	Surgery (day 0)	day 2	day 4	day 6	day 8	day 10-14	day 30 ^b
INCLUSION								
Informed consent	X							
Inclusion and exclusion criteria	X							
RANDOMIZATION								
		X						
INTERVENTIONS								
Intervention 1(IOWI with 1000ml PHX 0.04%)		X						
Intervention 2(IOWI with 1000ml NaCl 0.9%)		X						
Control group (no IOWI)		X						
ASSESSMENTS								
Demographical data	X							
Medical history	X							
Concurrent medication	X							
Physical examination	X							
NNSI Risk score	X							
Pregnancy test**	X**							
Blood sample***	X			X****				
Type of operation		X						
Duration of operation		X						
Level of contamination		X						
Type and length of incision		X						
Wound closure technique and suture material		X						
Creation of an enterostomy		X						
Administration and timing of antibiotic prophylaxis		X						
Intraoperative use of wound edge protectors		X						
Changing of gloves during operation		X						
Postoperative medication with effect on wound healing			X	X	X	X	X	X
Documentation of SSI			X	X	X	X	X	X
Documentation of other wound complications			X	X	X	X	X	X
Wound swab for microbiology [†]			X [†]	X [†]	X [†]	X [†]	X [†]	X [†]
Photograph of the wound			X	X	X	X	X	X
Documentation of re-operation			X	X	X	X	X	X
Documentation of AE/SAE		X	X	X	X	X	X	X
Duration of hospital stay								X

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* In case of emergency surgery enrolment is possible on the same day as the procedure

**For women of child-bearing potential only (serum or urine)

***Includes hemoglobin, hematocrit, platelets and white blood cell count, Na, K, Cr, Glu (non-fasting), AST/ASAT (SGOT), ALT/ALAT (SGPT), Bilirubin, Uric acid, Prothrombin time (PT), activated partial thromboplastin time (aPTT), international normalized ratio (INR) according to local in-house standards

****Between post-OP day 4-8 (visit 4-6)

[†] In case of SSI a swab will be taken from the wound or wound secretion for microbiological differentiation and testing of resistance to antibiotics according to local in-house standards

326
327
328

§ Visit window +6 days. If the patient is unable to attend visit 8 due to postoperative treatment in a rehabilitation facility or other medical reasons, a standardized protocol for evaluation and documentation of the wound will be sent to and filled out by the treating physician.

329

9. JUSTIFICATION OF DESIGN ASPECTS

330

9.1 Study design

331

This trial is a prospective, randomized, controlled, observer and patient-blinded, multicenter, surgical trial according to German drug law (AMG) phase IIIb with three parallel comparison groups. Reduction of SSI (according to CDC criteria) by IOWI after abdominal surgery is postulated. The IOWISI trial will be conducted in approximately 10 surgical departments (university and community hospitals), all of which are members of the trial network (CHIR-Net) of the German Surgical Society (*Deutsche Gesellschaft für Chirurgie*) and have experience in previous multicenter RCTs. Feasibility evaluation of all participating centers was done according to the SOPs of MSZ. All of the study personnel involved in the trial require GCP training and will be specifically instructed in all trial-specific procedures before initiation of the trial. According to AMG, the investigator requires 2 years' experience in drug trials. The leading surgeon of the operating team will perform the interventions since they represent standard techniques. All participating surgeons will be instructed and authorized by the investigator, prior to the first trial procedure.

344

345

9.2 Control and comparators

346

The WHO published the latest clinical guideline addressing the topic of IOWI in surgery in 2016. The consensus is that there is not sufficient evidence to support the use of IOWI with saline, diluted PVP-solutions should be considered and antibiotic solutions avoided. However, the underlying RCTs included all types of surgery (*i.e.* neuro-, orthopedic surgery.) and are of low level of evidence [6]. The guideline of the British National Institute of Clinical Excellence (NICE) from 2008 [7] states that, due to the lack of evidence any IOWI should be avoided. However, in clinical practice this advice is mostly not being followed. Most hospitals do not have standard protocols but leave the decision to irrigate or not to irrigate the wound up to the surgeon. Given these circumstances it is acceptable to recruit a control group receiving no intervention. So far, no gold standard was determined within RCTs in abdominal surgery. Therefore, the trial proposes an irrigation procedure on the best available evidence, which is either irrigation with PHX-solution or saline or no irrigation. PHX and saline solutions are widely used in clinical practice, but efficacy trials are not available momentarily. As PHX solution is a market-approved drug, safety is ensured and the trial subjects are not exposed to specific risks.

360

361 **9.3 Additional treatments**

362 No additional treatments will be performed within the trial. Antibiotic treatment 5 days prior to
363 surgery is an exclusion criterion. Pre-operative antibiotic treatment due to septic peritonitis (dirty
364 / contaminated wounds) after admission to the hospital is allowed, but has to be recorded in the
365 CRF. Application of routine intraoperative single shot antibiotic prophylaxis will be recorded in
366 the CRF (type and dose of antibiotics). The application of abdominal wall protectors is
367 recommended for contaminated procedures and has to be recorded in the CRF. A change of
368 gloves ahead of wound closure is recommended for contaminated procedures and has to be
369 recorded in the CRF. If indicated for medical reasons, all kind of medication is permitted during
370 the trial. Postoperative medication with adverse effects on wound healing (e.g. corticoids and
371 other immunosuppressive agents) will be recorded in the CRF. Any operative and / or
372 interventional revision of the wound will be documented as AE/ SAE and classified after Clavien
373 Dindo.

374 **9.4 Blinding**

375 The blinding procedure is restricted to participating patients, outcome assessors and the trial
376 statistician. Blinding of the surgical team that performs the intervention is impossible because
377 the control arm does not receive any wound irrigation. A member of the local study team, who
378 will not take part in postoperative patient visits, performs randomization after confirmed closure
379 of the abdominal fascia. A central online randomization tool of the MSZ (RANDOBASE) will
380 effectuate randomization. After informing the surgical team of the result, the investigator has to
381 print out, date and sign the randomization sheets. Subsequently, the randomization sheets have
382 to be stored away from the patient records, trial documents and ISF to ensure blinding of the
383 rest of the local study team.

384
385 Postoperatively, a GCP-trained investigator of the local study group, who is unaware of the
386 patient's intraoperative treatment, will assess wounds on 6 study visits and take a standardized
387 photograph of the wound at each visit which will be uploaded to a central database. However, in
388 case of SSI or any other AE/SAE that has to be reported the local investigator needs to be
389 unblinded.

390 In addition, independent, blinded outcome-assessors of spatially separated centers participating
391 in the trial will assess the pseudonymized wound photographs of every study visit in a
392 centralized database online. These online outcome-assessors receive training in rating of the
393 primary endpoints according to the CDC classification, which will be documented in a separate
394 training log. These independent outcome-assessors will only access the photo-database for
395 evaluation of the primary endpoint (SSI up to postoperative day 30) and will not be aware of the
396 randomization results or any other patient data. All treatment-specific data are documented in a

397 separate, undisclosed file. Wound photographs from all trial sites will be assessed by outcome
398 assessors of the coordinating study site in Munich. Photographs from the Munich study site will
399 be assessed in the study site Heidelberg.

400

401 **9.5 Exclusion of participants after initial inclusion**

402 Participants of the study can withdraw their consent to take part at any time without declaration
403 of reasons. All hitherto collected data are subject to analysis. The coordinating investigator or
404 the investigator may exclude patients from the study, if patients' safety is at risk or if there is
405 insufficient compliance of the patient. In order to generate a meaningful database, excluded
406 patients can be replaced by recruitment of new patients. If a patient does not receive PHX or
407 saline irrigation of the wound, this does not automatically lead to exclusion of the study.

408

409 **10. INCLUSION- AND EXCLUSION CRITERIA**

410 **10.1 Inclusion criteria**

- 411 • Clean-contaminated, contaminated or dirty surgery according to CDC classification
412 (attachment 3);
- 413 • Abdominal surgery by midline or transverse laparotomy; elective and emergency
414 procedures;
- 415 • Age \geq 18 years;
- 416 • American Society of Anesthesiologists (ASA) score \leq 3; (attachment 4)
- 417 • Ability to understand the nature and extent of the trial and to give written informed
418 consent

419

420 **10.2 Exclusion criteria**

- 421 • Pregnancy or breast feeding;
- 422 • Known hypersensitivity/allergy to PHX;
- 423 • Inability to give/understand informed consent;
- 424 • Critical medical condition of emergency patients, precluding informed consent or
425 sufficient time to reflect on the decision to participate in the trial;
- 426 • ASA $>$ 3;
- 427 • Inability to attend follow-up visits;
- 428 • Clean procedures according to the CDC classification or surgery without opening of the
429 abdominal cavity;
- 430 • Laparoscopic surgery;
- 431 • Revision-surgery (previous abdominal surgery within the last 30 days);
- 432 • Planned re-laparotomy within 30 days;

- 433 • Severe immunosuppression;
- 434 • Concurrent abdominal wall infections;
- 435 • Pre-operative systemic antibiotic therapy within 5 days prior to surgery (except
- 436 emergency pre-operative antibiotic treatment due to septic peritonitis after admission to
- 437 the hospital);
- 438 • Participation in another clinical trial that interferes with the primary or secondary
- 439 outcomes of this trial.

440

441 **10.3 Explanation of inclusion and exclusion criteria**

442 To enhance generalizability and representativeness, all patients undergoing elective and
443 emergency laparotomy (transverse or midline) for visceral surgery will be screened for this trial.
444 However, only clean-contaminated, contaminated or dirty (class II-IV), open abdominal surgery,
445 according to the CDC classification [1] will be eligible, since in clean (class I) procedures the
446 risk of SSI is low. Laparoscopic surgery as well as surgery without opening of the abdominal
447 cavity or revision surgery (previous abdominal surgery within the last 30 days or planned re-
448 laparotomy within the next 30 days of surgery) will be excluded, since these types of procedures
449 are not comparable in terms of SSI risk.

450 Pre-operative antibiotic therapy within 5 days prior to surgery was chosen to be an exclusion
451 criterion to avoid bias of the results, since this might lead to a lower individual risk of infection.
452 However, this does not apply to patients that receive pre-operative antibiotics after admission to
453 the hospital in an emergency situation of septic peritonitis. Furthermore, this does not include
454 standard intraoperative single shot antibiotic prophylaxis.

455 Patients have to be ≥ 18 years of age and able to understand and give written informed
456 consent. Any patient in a very bad general medical condition (ASA > 3) will be excluded to avoid
457 too many patient-related confounders. Emergency patients in a critical medical condition that
458 does not allow them to fully understand and provide informed consent or does not leave them
459 sufficient time to reflect on the decision to participate in the trial will not be included.
460 Furthermore, patients have to be able to attend follow-up visits.

461 Patients with severe immunosuppression (e.g. after: organ or bone marrow transplantation,
462 concurrent steroid treatment with >10 mg prednisone daily or an equivalent dose of any other
463 steroid), concurrent infliximab treatment or treatment with an equivalent immunosuppressive
464 substance, chemotherapy within the last 2 weeks prior to trial intervention) or patients with
465 severe pre-operative neutropenia ($\leq 0.5 \times 10^9/L$) or liver cirrhosis Child-Pugh B/C will not be
466 included. Pregnant or breast feeding women, as well as patients with a known
467 hypersensitivity/allergy to PHX will not be included in the trial either.

468 Patients that participate in other clinical trials that could interfere with the primary (SSI) or
469 secondary outcomes of the IOWISI trial will be excluded.

470

471 **11. FREQUENCY AND SCOPE OF TRIAL VISITS**

472 Graph 1 and 2 reflect the intervention scheme, trial flow, and visits for the IOWISI trial. Visits are
473 the same for all participants of the study, regardless the treatment group.

474

475 **11.1 Recruitment and screening**

476 Only surgical departments with adequate patient numbers, providing a written commitment on
477 their recruitment capacity were included in the trial to reach the target sample size. The
478 recruitment period is set to 27 months (first patient in to last patient out 28 months). In case of
479 elective procedures, pre-screening (this is just a pre-selection of eligible patients within the
480 study team) of patients can be performed up to 14 days prior to the scheduled surgical
481 procedure. Screening and inclusion of patients will be performed not earlier than 3 days and not
482 later than on the day before the planned surgical procedure, to ensure the patient has enough
483 time to consider the decision to participate. In case of emergencies, screening and inclusion can
484 take place on the day of admission to the hospital, which is usually the same day as surgery. All
485 screened patients are documented in a screening log. If patients do not wish to participate in the
486 study, reasons are documented accordingly. If patients fit inclusion/exclusion criteria and agree
487 to participate, they will need to give written informed consent to the local GCP-trained
488 investigator, after adequate time for consideration in order to participate in the study
489 (representing visit 1). Therefore, at the screening visit, a detailed description of the study and
490 further instructions are discussed with the patient, including methods of wound irrigation, risk-
491 benefit-ratio, and follow up schedule.

492

493 **11.2 Visit 1 (Inclusion)**

494 After the local investigator has reviewed the inclusion and exclusion criteria again and having
495 received written consent by a patient, demographical data / medical history (date of birth
496 [mm/yyyy], gender, body height, body weight, BMI, ASA, medical history, concurrent medication,
497 history of SSI, history of radio/chemotherapy, diabetes, smoking, alcohol consumption,
498 medication, duration of pre-operative hospital stay), diagnosis and the NNIS Risk score for
499 determining the intrinsic risk of SSI (attachment 5) will be documented according to the eCRF.
500 The investigator will perform a physical exam (blood pressure, heart frequency, condition of the
501 planned abdominal surgical incision area, clinical relevant findings [normal or abnormal (please
502 specify), respiratory system, cardiovascular system, liver, kidney, neurological or other free text

503 and date dd/mm/yyyy]) and take a blood sample (EDTA, Serum, and Citrate). Measurements of
504 the blood sample are:

- 505 • Hemoglobin
- 506 • Hematocrit
- 507 • Platelets
- 508 • White blood cell count
- 509 • Sodium
- 510 • Potassium
- 511 • Creatinine
- 512 • Non-fasting glucose
- 513 • AST/ASAT
- 514 • ALT/ALAT
- 515 • Bilirubin
- 516 • Uric acid
- 517 • Prothrombin time (PT)
- 518 • Activated partial thromboplastin time (aPTT)
- 519 • International normalized ratio (INR)

520 In case of women of child-bearing potential, a pregnancy test will be performed additionally
521 (serum or urine [negative/positive/not performed with specification of reason as free text]).

522

523 **11.3 Visit 2 (Surgery/Randomization)**

524 Documented parameters of the surgical procedure include the urgency (emergency/elective),
525 type of surgical procedure (colorectal and/or small bowel and/or,hepato-biliary and/or pancreatic
526 and/or splenectomy and/or gastric and/or esophageal and/or nephrectomy and/or urogenital
527 tract and/or others (freetext)) the duration of surgery (incision until complete skin closure,
528 minutes), the level of contamination according to CDC classification (class II-IV; see attachment
529 3), the intraoperative use of wound edge protectors (yes/no), and prophylactic changing of
530 gloves during of the operation (yes/no), type (transverse/midline) and length (cm) of the incision,
531 creation of an enterostomy (yes/no), the wound closure technique (subcutaneous sutures
532 (yes/no), stapler/suture, if suture: continuous/single) and used suture material, the
533 administration (yes/no) and timing (>1h/≤1h prior to incision) of antibiotic prophylaxis. If the
534 operating surgeon decides that incomplete closure of the wound and/or any other wound related
535 procedure after the study intervention (e.g. negative pressure treatment) is necessary for the
536 benefit of the patient, the patient will have to be excluded from the trial.

537

538 Randomization (see section 24.) will take place at the end of surgery, after closure of the
539 abdominal fascia, when the level of contamination is definitely determined by the surgeon. A

540 designated member of the local study team (who will not perform postoperative study visits) will
541 perform randomization instantly by using the online tool of the MSZ (RANDOBASE) and inform
542 the surgeon of the result and according treatment. Date of randomization (mm:hh, dd/mm/yyyy),
543 successful randomization (yes/no), and the result of the randomization process are
544 documented (Printout). Subsequently, the randomization sheets have to be stored away from
545 the patients file to ensure blinding.

546 Study treatment according to randomization:

- 547 • Wound irrigation with PHX 0,04% 1000ml
- 548 • Wound irrigation with NaCl 0,9% 1000ml
- 549 • No wound irrigation

550 Furthermore, any AE or SAE is documented during this visit.

551

552 **11.4 Visit 3 to 8 (Post-op days 2, 4, 6, 8, 10-14, and 30-36)**

553 Postoperatively, there will be 6 trial visits where an independent, blinded outcome assessor
554 trained in the diagnosis and classification of SSI according to CDC definitions will examine
555 wounds (SSI superficial or deep or organ/space, see attachment 1). In addition,
556 pseudonymized, electronic pictures of the wound will be uploaded to a centralized database for
557 independent and blinded evaluation (see 11.4). The assessors will not be aware of the study
558 procedure or other details of the examined wound photograph. Postoperative medication with
559 adverse effects on wound healing (e.g. corticoids and other immunosuppressive agents) will be
560 documented in the eCRF:

561 In case of SSI, microbiological swabs will be taken from the wound secretion for microbiological
562 differentiation and testing of resistance to antibiotics according to in-house standards by each
563 local institution. Other wound complications like seroma, hematoma, delayed healing or
564 necrosis will be documented as secondary endpoint. In case of any surgical complication,
565 including SSI, will be reported as AE/SAE and the Clavien Dindo classification (attachment 2)
566 will be applied to specify the severity and consequent treatment. Furthermore, the rate of re-
567 operations, mortality and occurrence of any AE or SAE will be documented (see 16).
568 Additionally, the duration of the hospital stay (from admission to discharge or day of the visit, in
569 days) will be documented on visit 8 (post-op day 30-36). To promote complete follow-up, a visit
570 window of 6 additional days was implemented. In addition, patients will be recompensed for any
571 travel expenses needed to attend study visit 8. If however, the patient is unable to attend visit 8
572 due to postoperative treatment in a rehabilitation facility or other medical reasons, a

573 standardized protocol for evaluation and documentation of the wound (incl. wound photograph)
574 will be sent to and filled out by the treating physician.

575 Between post-op day 4 and 8 (visit 4, 5 or 6) one study-specific, post-operative blood sample
576 will be taken, and the same measurements as upon visit 1 will be analyzed according to local
577 clinical routine:

- 578 • Hemoglobin
- 579 • Hematocrit
- 580 • Platelets
- 581 • White blood cell count
- 582 • Sodium
- 583 • Potassium
- 584 • Creatinine
- 585 • Non-fasting glucose
- 586 • AST/ASAT
- 587 • ALT/ALAT
- 588 • Bilirubin
- 589 • Uric acid
- 590 • Prothrombin time (PT)
- 591 • Activated partial thromboplastin time (aPTT)
- 592 • International normalized ratio (INR)

593

594 **12. DOSE, MODE AND SCHEME OF INTERVENTION**

595 After closure of the abdominal fascia, patients will be randomized stratified by level of
596 contamination of the operation. In the experimental group 1, the subcutaneous soft tissue will be
597 irrigated with 1000 ml of a 0.04% PHX solution, which is the recommended concentration for
598 surgical wound irrigation according to the SMPC. PHX solutions (0.04%) are approved for this
599 indication in Germany. The wound shall be carefully rinsed throughout with the irrigation solution
600 and the excess removed with suction. Debris and blood clots should be removed from the
601 wound using irrigation/suction. The wound shall not be rubbed dry with abdominal cloths, but left
602 moistened with the irrigation solution to ensure sufficient contact time for PHX to have the
603 desired antiseptic effect. After irrigation with PHX the wound shall not be irrigated with saline or
604 any other solution again. Since PHX is a cation-active substance, it is not compatible with
605 anionic organic substances (e.g. lactate). Furthermore, the combination of PHX with PVP-I
606 products should be avoided.

607 In the experimental group 2, the same intervention will be performed using 1000ml of isotonic
608 saline solution (NaCl 0.9%).

609 The irrigation volume of 1000ml was chosen to be sure that even large laparotomy wounds
610 would be sufficiently irrigated. This was determined by senior surgeons' clinical experience,
611 since so far no recommendations for the optimal volume of surgical irrigation exist. After
612 irrigation of the wound, the skin closure will be performed according to local standards, without
613 any further wound-related procedure.

614 In the control group, wounds will not be surgically irrigated, as is currently recommended in the
615 NICE guideline. PHX solutions or saline are to be purchased, stored, and distributed according
616 to the respective trial centers standard operating procedures. Trade name, dosage, batch and
617 dispensed amount will be documented on a separate form.
618

619 **13. PATIENT, STUDY AND SITE DISCONTINUATION**

620 **13.1 Patient discontinuation**

621 Patients have the right to voluntarily withdraw from the study at any time for reason. In addition,
622 the investigator has the right to withdraw a patient from the study at any time. Reasons for
623 withdrawal from the study may include but are not limited to the following:

- 624 ▪ Patient withdrawal of consent at any time;
- 625 ▪ Any medical condition that the investigator or sponsor determines may jeopardize the
626 patient's safety if he or she continues in the study;
- 627 ▪ If it is discovered that a study subject is pregnant or may have been pregnant at the time
628 of intervention (see point 16.9);
- 629 ▪ Investigator or sponsor determines it is in the best interest of the patient to discontinue
630 the study.

631 Every effort should be made to obtain information on patients who withdraw from the study. The
632 primary reason for withdrawal from the study should be documented on the appropriate eCRF.
633 However, patients will not be followed for any reason after consent has been withdrawn.
634 Patients who withdraw from the study will not be replaced.
635

636 **13.2 Study and site discontinuation**

637 The **sponsor** has the right to terminate this study at any time. Reasons for terminating the study
638 may include but are not limited to the following:

- 639 ▪ The incidence or severity of AEs in this or other studies indicates a potential health
640 hazard to patients;

- 641 ▪ Unsatisfactory patient enrolment;
642 ▪ The continuation of study is unethical or it has been proven that the therapy has a
643 clearly negative influence;
644 ▪ Unforeseen complications arise that no longer justify a continuation of the study;

645

646 The **sponsor** will notify the investigator of a decision to discontinue the study. The sponsor has
647 the right to **close a site** at any time.

648 Reasons for closing a site may include, but are not limited to, the following:

- 649 ▪ Excessively slow recruitment;
650 ▪ Poor protocol adherence;
651 ▪ Inaccurate or incomplete data recording;
652 ▪ Non-compliance with the ICH-GCP guideline;
653 ▪ No study activity (*i.e.* all patients have completed and all obligations have been fulfilled);

654 The **investigator** can discontinue the clinical study at his site at any time if he no longer
655 considers the continuation of the study, for example if there are ethical and/or medical concerns.

656

657 **14. ADVERSE EVENTS (AES)**

658 **14.1 Definition adverse event (AE)**

659 An AE is any untoward medical occurrence in a patient or in a clinical investigation subject
660 administered a pharmaceutical product, which does not necessarily have a causal relationship
661 with this treatment. An AE can therefore be any unfavorable and unintended sign (including an
662 abnormal laboratory finding), symptom or disease temporally associated with the use of a
663 medicinal product, whether or not related to the treatment. Any AE has to be documented in the
664 eCRF on the respective “Adverse Event Report Form”.

665

666 **14.2 Specific definitions of AEs in the IOWISI trial**

667 The obligation to document any AE in the study, starts with the randomization and ends with
668 completion of the last study visit. AE/SAEs are documented according to the standard grading
669 on the AE/SAE reporting forms. Surgical site infections (primary endpoint) and all other local
670 wound complications (secondary endpoint) will be documented as AE/SAE. In addition, their
671 severity and the consequent treatment will be documented according to the Clavien Dindo
672 classification (attachment 2). All laboratory values or events that will be assessed as “clinically
673 significant” in the eCRF have to be documented as an AE. The responsible medical investigator
674 will judge the clinical significance in the context of the postoperative course after laparotomy
675 and the correspondent laboratory values before intervention.

676

677 **14.3 Serious adverse events (SAE) and other definitions**

678 **Serious adverse events (SAEs)**

679 A SAE is defined as any clinical event that at any time during the study participation:

- 680 ▪ Results in death;
- 681 ▪ Is life-threatening (the term life-threatening refers to an event in which the subject was at
682 risk of death at the time of the event and not to an event which hypothetically might have
683 caused death if it was more severe);
- 684 ▪ Requires subject hospitalization or prolongation of existing hospitalization;
- 685 ▪ Results in persistent or significant disability/ incapacity.
- 686 ▪ Results in a congenital anomaly/birth defect or
- 687 ▪ Is rated as another significant event or condition by the investigator

688 Any SAE has to be reported to the MSZ immediately (*i.e.* within 24 hours after becoming aware
689 of the event, see chapter 16.7).

690 **Suspected Unexpected Serious Adverse Reaction (SUSAR)**

691 Serious AEs that are both suspected, *i.e.* possibly related to the investigational medicinal
692 product (IMP) and 'unexpected', *i.e.* the nature and/ or severity of which is not consistent with
693 the applicable product information, are to be classified as Suspected Unexpected Serious
694 Adverse Reactions (SUSARs). If the second assessor classifies the SAR as 'suspected' (the
695 relationship to the IMP is "related", "probable" or "possible") and unexpected, it will be
696 categorized as a SUSAR. All SUSARs are subject to an expedited reporting to the responsible
697 ethics committee(s), the competent federal authority (BfArM) and to all participating
698 investigators (see 16.7). Furthermore, a report on all observed SAEs / SARs / SUSARs will be
699 submitted once a year in the DSUR (Development Safety Update Report) format.

700 **Period of observation and documentation**

701 In this trial, all AEs that occur between the randomization (during surgery) and the last study
702 visit or premature study termination will be documented on the pages provided in the eCRF.
703 AEs must also be documented in the subject's medical records. All subjects who have AEs,
704 whether considered associated with the use of the trial medication or not, must be monitored to
705 determine the outcome. The clinical course of the AE will be followed up until resolution or
706 normalization of changed laboratory parameters or until it has changed to a stable condition.

707

708 **14.4 Evaluation of the severity**

709 The grading of AEs in this trial will be carried out on the basis of the 5-grade scale defined in the
710 CTCAE V4.0:

711 **Grade 1: Mild AE**

- 712 **Grade 2:** Moderate AE
713 **Grade 3:** Severe AE
714 **Grade 4:** Life-threatening AE or AE causing disablement
715 **Grade 5:** Death related to AE

716 The grading of all AEs listed in the CTCAE v4.0 will be based on the information contained
717 therein. The grading of all other AEs, i.e. those which are not listed in the CTCAE v4.0 will be
718 performed by a responsible investigator, based on definitions given above. In addition, surgical
719 complications will be evaluated according to the Clavien Dindo classification.

720

721 **14.5 Evaluation of the causal relationship**

722 Investigators will estimate the causal relationship between the AE/SAE and the treatment. When
723 estimating the causality the investigator may draw on known biophysical parameters,
724 incorporate previous knowledge on the AE profile of the investigational product and possible
725 simultaneously factor in the efficacy against other substances and the concomitant diagnoses of
726 the patient. The investigator will categorize each AE that occurred after administration of the
727 IMP regarding the coherency with the administration of the IMP as:

728 - **Related:** There is a reasonable possibility that the event may have been caused by the
729 IMP. A certain event has a strong temporal relationship and an alternative cause is
730 unlikely.

731 - **Probable:** An AE that has a reasonable possibility that the event is likely to have been
732 caused by the IMP. The AE has a timely relationship and follows a known pattern of
733 response, but a potential alternative cause may be present.

734 - **Possibility:** An AE that has a reasonable possibility that the event may have been
735 caused by the IMP. The AE has a timely relationship to the IMP; however, the pattern of
736 response is untypical, and an alternative cause seems more likely, or there is significant
737 uncertainty about the cause of the event.

738 - **Unlikely:** Only a remote connection exists between the IMP and the reported AE. Other
739 conditions including concurrent illness, progression or expression of the disease state or
740 reaction of the concomitant medication appear to explain the reported AE.

741 - **Not related:** An AE that does not follow a reasonable temporal sequence related to the
742 IMP and is likely to have been produced by the subject's clinical state, other modes of
743 therapy or other known aetiology.

744

745 **14.6 Outcome of AEs**

746 The outcome of an AE at the time of the last observation will be classified as:

- 747 - **Recovered/ Resolved:** All signs and symptoms of an AE disappeared without any
748 sequels at the time of the last interrogation.
- 749 - **Recovering/ Resolving:** The intensity of signs and symptoms has been diminishing
750 and/ or their clinical pattern has been changing up to the time of the last interrogation in a
751 way typical for its resolution. Further follow-up is possibly needed.
- 752 - **Not recovered/ Not resolved:** Signs and symptoms of an AE are mostly unchanged at
753 the time of the last interrogation. Further follow-up is possibly needed.
- 754 - **Recovered/ Resolved with sequels:** The patient recovered with sequels from the AE /
755 the AE resolved with sequels, *i.e.* the patient suffers from late complications or damage
756 resulting from the AE.
- 757 - **Fatal:** An AE resulting in death. If there are more than one AE only the AE leading to
758 death (possibly, related) will be characterized as 'fatal'.
- 759 - **Unknown:** The outcome is unknown or implausible and the information cannot be
760 supplemented or verified.

761 **14.7 Reporting of serious adverse events (SAEs)**

762 **Primary reporting of SAEs**

763 All SAEs must be reported immediately, by fax (number 089/4140-6480) by the investigator to
764 the responsible officer at the MSZ using the designated form.

765 München Studienzentrum

766 SAE-Reporting

767 Ismaninger Straße 22

768 81675 München

769 Tel.: +49/89/4140-6477

770 Fax: +49/89/4140-6480

771 Reporting should under no circumstances take place after more than 24 hours from the moment
772 the investigator becomes aware of the event.

773 The initial report must be as complete as possible including details of the current illness and
774 SAE and an assessment of the causal relationship between the event and the trial medication.

775 **Second assessment of SAEs**

776 All SAEs will be subject to a second assessment by a designated person. This person is elected
777 by the sponsor and will be independent from the sponsor and the reporting investigator. The
778 second assessor will fill out a 'Second Assessment Form' for each SAE. The 'Second
779 Assessment Form' will contain the following information:

- 780
- 781 I) Assessment of seriousness of the event (investigator and second assessor)

- 782 II) Assessment of relationship between SAE and IMP (investigator and second
783 assessor)
784 III) Assessment of expectedness of SAE, derived from IMP (second assessor)
785 IV) A statement if the benefit/ risk assessment for the trial did change as a result of
786 SAE (second assessor)

787 The responsible safety officer of the MSZ will carry out the expedited reporting. Only SUSARs/
788 SAEs occurring after administration of IMPs will undergo expedited reporting.

789

790 **14.8 Expedited reporting**

791 Pursuant to the German Drug Law (AMG) and the GCP Regulation, the ethics committee and
792 the competent federal authority (BfArM) will be informed of all suspected SUSARs and all SAEs
793 resulting in death or being life-threatening occurring during the trial. Both institutions and all
794 participating investigators will be informed in case the risk/ benefit assessment did change or
795 any others new and significant hazards for subjects' safety or welfare occur. The sponsor has to
796 ensure that all relevant information about a SUSAR, which occurs during the course of a clinical
797 trial and is fatal or life threatening is reported as soon as possible and not later than seven days
798 after the sponsor was first aware of the reaction. Any additional relevant information should be
799 sent within eight days of the report. A SUSAR, which is not fatal, or life threatening has to be
800 reported as soon as possible and in any event not longer than 15 days after the sponsor was
801 first aware of the reaction.

802

803 **14.9 Pregnancy**

804 If, following initiation of the investigational product, it is subsequently discovered that a study
805 subject is pregnant or may have been pregnant at the time of investigational product exposure,
806 the investigator must immediately notify the sponsor of this event via the "Report on the drug
807 exposure during pregnancy" within 24 hours and in accordance with SAE reporting procedures.
808 The patient will be withdrawn from the study. Follow-up information regarding the course of the
809 pregnancy, including perinatal and neonatal outcome and, where applicable, offspring
810 information must be reported on a "Report on the pregnancy outcome during drug exposure".
811 Any pregnancy occurring in a female partner of a male study participant the investigator
812 becomes aware of should be reported to the sponsor. Information on this pregnancy may also
813 be collected on the pregnancy reporting forms.

814

815 **15. SAFETY MONITORING BOARD (SMB)**

816 An independent Safety Monitoring Board (SMB according to the Guidance E3, ICH note for
817 Guidance E6, ICH note for Guidance E9, Directive 2001/20EC “relating to the implementation of
818 good clinical practice in the conduct of clinical trials on medicinal products for human use) is a
819 group of experts external to the study that addresses the patient’s safety and performs risk /
820 benefit assessments. According to its operating procedures the SMB reviews accumulating
821 safety data from ongoing trials to fulfill the safety monitoring. The rules of the SMB are
822 deposited in the SMB Charta, (SOP_MSZ_AE04-H-A01_V02). The aim of this Charta is to
823 define the composition, responsibilities, purpose and timing of meetings, details of the
824 operation, including documentation and reporting and specifying the procedures to ensure
825 confidentiality and appropriate communication of the SMB.
826

827 **16. ENSURING DATA QUALITY**

828 **16.1 Documentation**

829 All raw data such as patient records are declared as source documents. It must be ensured that
830 they are available during routine monitoring visits. Apart from that the investigator of each site
831 must maintain a separate patient identification list. The patient identification list will be
832 maintained at the site separate from the documentation. The eCRF covers all the important
833 forms, sorted according to visits. If a patient withdraws from the study, the reason must be
834 recorded on the eCRF.

835 **Data collection**

836 The documentation of the study data in adherence to the GCP-guidelines and the clinical trial
837 protocol is the responsibility of the investigator. Original data (source documents) remain in
838 hospital medical record and information on the eCRF must be traceable and consistent with the
839 original data. Source documents are e.g. laboratory results, photography, skin biopsy histology
840 description and quality of life questionnaire, EASI, Pruritus VAS, TSQM. Original written
841 informed consent signed by the patient is kept by the investigator and a signed copy will be
842 given to the patient. No information in source documents about the identity of the patients will
843 be disclosed. All data collected in this study must be entered in an eCRF which has to be
844 completed by the investigator or authorized trial personnel and signed by the investigator. This
845 also applies for those patients who do not complete the study. If a patient withdraws from the
846 study, the reason must be recorded on the eCRF. The investigator is responsible for ensuring
847 the accuracy, completeness, and timeliness of all data reported to the sponsor in the eCRFs
848 and in all required reports.

849 **Database management**

850 Data are administered and processed by data management of the MSZ with the support of a
851 study database (eCRF) according to the SOPs of the MSZ. A description of the study specific
852 processes is given in the Data Management Plan that details the key planning and control
853 elements for the data management component of the study.

854 The evaluation of the data takes place by programmed validity- and consistency checks. In
855 addition a manual/visual evaluation of plausibility is performed in accordance to the
856 requirements of GCP. Queries may occur, which will be visualized on the study database. The
857 investigator has to resolve all data discrepancies in the study database. After entry of all
858 collected data and clarification of all queries, the database will be closed at the completion of
859 the study. The database closure has to be documented. Data and results electronically
860 recorded will be archived according to legal guidelines at least 10 years after study termination.

861

862 **16.2 Audits and inspections**

863 As part of quality assurance according to GCP, the sponsor and the competent health
864 authorities have the right to audit/inspect the study sites and any other institutions involved in
865 the trial. The aim of an audit/inspection is to verify the validity, accuracy and completeness of
866 data, to establish the credibility of the clinical trial, and to check whether the trial subject's rights
867 and trial subject safety are being maintained. The sponsor may assign these activities to
868 persons otherwise not involved in the trial (auditors). These persons as well as inspectors are
869 allowed to access all trial documentation (especially the trial protocol, eCRFs, trial subjects'
870 medical records, drug accountability documentation, and trial-related correspondence).

871 The sponsor and all investigators of the participating study sites undertake to support auditors
872 and inspections by the competent authorities at all times and to allow the persons charged with
873 these duties access to the necessary original documentation. All persons conducting audits
874 undertake to keep all trial subject data and other trial data confidential.

875 After each external audit the investigator receives an audit confirmation from the responsible
876 auditor. This confirmation has to be stored in the ISF in order to provide access to it in case of
877 an inspection by the competent authorities. The audit report is provided to the sponsor for
878 control.

879 **16.3 Monitoring**

880 Monitoring activities are performed to ensure that the trial is conducted in accordance with the
881 trial protocol, the principles of GCP and local legislation. A monitoring manual describing the
882 scope of the monitoring activities in detail will be prepared.

883 The responsible monitor will contact the investigator and will be allowed, on request, to inspect
884 the various records of the trial (eCRF and other pertinent data) provided that patient
885 confidentiality is maintained in accord with local requirements. The monitor should have access

886 to patient records, any information needed to verify the entries in the eCRF and all necessary
887 information and essential study documents. The investigator agrees to cooperate with the
888 monitor to ensure that any problems detected in the course of these monitoring visits are
889 resolved. A monitoring visit report is prepared for each visit describing the progress of the
890 clinical trial and all identified problems.

891

892 **16.4 Archiving**

893 At the end of the clinical study all study-relevant data must be archived as required by law and
894 when indicated in addition according to the Clinical Trial Agreement. All documentation forms,
895 ICFs and other essential study documents must be retained as required by law. Patient ID lists
896 and patient files are retained in the respective study sites separately. The ICFs are kept in with
897 the study documents.

898

899 **17. ETHICAL AND REGULATORY ASPECTS**

900 **17.1 Sponsor's and investigator's responsibilities**

901 This study is conducted in compliance with all applicable laws and regulations and also the
902 Declaration of Helsinki. **The sponsor has the overall responsibility for the ethical and
903 scientific conduct of the study. All participating investigators agree to adhere to the
904 instructions and procedures described in the study protocol and thereby to adhere to the
905 principles of GCP that it conforms to.**

906 The responsible ethics committee of TUM and health authority (BfArM) will review the final study
907 documents. The ethics committee's and BfArM's decision concerning the conduct of the study
908 will be communicated in written form to the sponsor. The sponsor will assure submission of
909 required progress reports, annual safety reports and substantial amendments for approval to
910 the ethics committee and BfArM. Before initiating the study, the sponsor must submit any
911 required amendments to BfArM for review and acceptance to begin the trial according to § 42
912 AMG. Furthermore, the sponsor has to inform the ethics committee and BfArM within 90 days
913 about completion of the trial and provide a brief report of its outcome 1 year after completion of
914 the trial. Results of the study will be reported following ICH-GCP-E6 and published according to
915 the CONSORT statement.

916

917 **17.2 Independent ethics committees and health authorities**

918 Prior to the start of this study, the protocol and other required documents would have to be
919 reviewed and approved by the locally responsible ethics committees of each study site. Their
920 reports as well as a signed and dated approval by the BfArM must be obtained and assessed by
921 the leading ethics committee of the TUM before study initiation. Any amendments to the

922 protocol, other than administrative ones (of which the leading ethics committee and BfArM will
923 merely be informed), must be reviewed and approved by both authorities.

924 Before inclusion of the first patient the federal state authorities (*zuständige Regierungsbehörden*
925 *der Länder*) will be informed about the study. A copy of this report needs to be forwarded to
926 BfArM and needs to be filed in the ISF and TMF.

927

928 **17.3 Ethical performance of the study**

929 The study is conducted according to the ethical principles as defined in the Declaration of
930 Helsinki, version of 2008 (see 28.). The present clinical study is conducted in accordance with
931 principles published in the ICH-GCP Guideline and the applicable legal regulations (AMG, GCP-
932 V, see 19.1) These principles concern ethics committee procedures, patient information and
933 informed consent procedures, adherence to the protocol, administrative documents,
934 documentation of the study medication, data collection, patient records (source documents),
935 recording and reporting of AEs/SAEs, preparation of inspections and audits as well as storage
936 and safekeeping of the documents. All the investigators and personnel involved in the study
937 have been informed that international monitoring authorities, the competent federal authorities
938 and the sponsor are authorised to review the study documents and patient files.

939

940 **17.4 Public register**

941 Before the clinical study will be initiated, it will be filed at the German Clinical Trials Register
942 (DRKS), which is part of the International Clinical Trials Registry Platform (ICTRP) of the WHO.
943 After ethical approval the trial will be registered under the ID-number: DRKS00012251.

944

945 **17.5 Informed consent of the study participants**

946 A patient can only be included in the study, if he provides written consent after being informed
947 by a GCP-trained investigator (orally and in writing) about the nature, significance and scope of
948 the clinical study in an appropriate and understandable way. The investigator must fully explain
949 the purpose of the study to the patient or his/her guardian prior to entering the patient into the
950 study. The investigator is responsible for obtaining written informed consent from each patient
951 The person signing the consent form will receive a copy of the signed form. By providing such
952 consent the patient is declaring that he understands and accepts the recording of data that is
953 part of the study and its verification by authorised monitors or federal authorities. The patient will
954 be educated about the potential benefits and complications of the IMP used in the study. It must
955 be clear for him that he can withdraw his consent at any point of time without any disadvantages
956 to his further treatment. The original copy of the written ICF will be kept in the study folder of the
957 study site. The patient will be given the copies of the written patient information and ICF.
958 Additionally, copies of both documents will be filed in the patient's medical file. Patient

959 information and ICF are attached at the end of this protocol. The patient information and ICF will
960 be submitted to the responsible ethics committee for assessment before the study will be
961 initiated.
962

963 **18. INSURANCE FOR TRIAL PARTICIPANTS**

964 In the clinical trial of an IMP, all the participants are insured in accordance with the AMG. The
965 scope of the insurance coverage is derived from the insurance documents that are included in
966 the ISF. Before inclusion the insurance conditions shall be submitted to the patient for review
967 without request to do so. The insurance conditions should be furnished to the patient to take
968 with him before being included in the study on request and after inclusion in any case.
969 Insurance coverage is being provided by:

970 HDI-Gerling Industrie Versicherung AG
971 Niederlassung München
972 Vertragsservice/M-B
973 Ganghofer Strasse 37-39
974 80339 Munich
975 Tel.: +49 (89) 9243-420
976 Fax.: +49 (89) 9243-356
977 Insurance number: 65-963496-03037/390 (Studie: 2/17)

978 If an insured event is suspected to have occurred, the sponsor is to be notified immediately. He
979 then has to notify the insurance provider about the damages immediately. The patient will
980 receive a copy of the notification to the insurance provider. The patient may also inform the
981 insurance provider by bypassing the study personnel, reporting any claims. In this case, he
982 should be notified that the sponsor of the clinical trial should still be informed about the event.
983 Patients have to be informed about both options.

984 **19. DATA PRIVACY PROTECTION / CONFIDENTIALITY** 985 **PROTECTION**

986 The applicable local regulations of data privacy protection will be followed. The patients will be
987 informed that any patient-related data and materials will be appropriately made pseudonymous
988 (pursuant to § 12 and § 13 of the GCP Regulations) and that these data may be used for
989 analysis and publication purposes. Furthermore, the patients will be informed that their data
990 may be inspected by representatives of BfArM or of the sponsor for the purpose of validation of
991 a proper study conduct. Patients who do not provide consent for transmission of their data,
992 according to the data protection agreement included in the ICF, will not be included in the
993 clinical study.

995 **20. PROTOCOL AMENDMENTS OR CHANGES IN TRIAL** 996 **CONDUCT**

997 In order to insure comparable conditions in all study sites and in the interest of standardized
998 evaluations of the trial, changes in this protocol are not foreseen. However, changes in trial
999 conduct are possible. Any change (besides administrative changes) of this protocol requires a
1000 written protocol amendment that must be reviewed by the sponsor before implementation.
1001 Furthermore, consent needs to be obtained by the investigator of each participating center.
1002 Amendments that significantly affect the safety of subjects, the scope of the investigation or the
1003 scientific quality of the study, additionally require approval of the leading ethics committee and
1004 BfArM. A copy of the written approval of these amendments must be provided to the sponsor
1005 and the investigator at each study site. Examples of amendments requiring such approval are:

- 1006 - Significant changes in the study design;
- 1007 - Increases in the number of invasive procedures.

1008 However, these requirements for approval should in no way prevent the investigator or sponsor
1009 to take any immediate action in the interests of preserving patient safety. If the investigator feels
1010 an immediate change to the protocol is necessary and is implemented for safety reasons, the
1011 sponsor, ethics committee and BfArM must be informed immediately. Amendments affecting
1012 only administrative aspects of the study do not require formal protocol amendments or ethics
1013 committee and BfArM approval. However, the ethics committee and BfArM must still be notified
1014 about the changes.

1015

1016 **21. STATISTICAL CONSIDERATIONS**

1017 **21.1 Proposed sample size / Power calculations**

1018 The sample size was calculated (nQuery Advisor software version 7.0, Statistical Solutions Ltd,
1019 Cork, Ireland) based on the primary endpoints of the study, assuming SSI rates of 2.2% in the
1020 PHX group (assuming a 75% risk reduction according to the trial by Roth *et al.* [1]), 8.7% in the
1021 saline group (according to the results of the trial by Cervantes-Sanchez *et al.*[2]), and 16.2% in
1022 the control group (according to the meta-analysis by Mueller *et al.* [3]). The global significance
1023 level was set to 5%. Since the PHX arm will be used twice for a comparison, the Bonferroni-
1024 Holm procedure was used to set the local alpha level for test 1 (PHX vs. saline irrigation) to
1025 2.5% and for test 2 (PHX vs. no intervention) to 5%. If 230 patients are recruited in the PHX
1026 arm, 230 patients in the saline arm and 80 patients in the control arm (a total of 540 patients),
1027 the two-sided Fisher exact test for test 1 will have a power of 94% and for test 2 – a power of
1028 85% to detect differences between the treatment groups. The comparison saline irrigation vs.

1029 control is not included in the sample size calculation, as it will not be analyzed in a confirmatory
1030 manner. The low medical interest cannot justify the large increase in patient numbers.

1031 An interim analysis is not necessary since patients in this trial do not undergo any specific
1032 additional risks, as all products used are on-label in Germany. Baseline adjustments will be
1033 performed according to the pre-specified subgroup analyses. No dropout rates are calculated,
1034 as the analysis will be based on the ITT set.

1035

1036 **21.2 Statistical analysis**

1037 The primary and secondary endpoints will be analyzed on the Intention-To-Treat (ITT) set,
1038 consisting of all patients included in the study in the treatment arm they were randomized to.
1039 The safety analysis will be performed on the safety set, consisting of all patients randomized
1040 into the study and assigned to the treatment group of their actual treatment.

1041

1042 **21.3 Primary endpoint**

1043 Wound irrigation with PHX solution will be tested for superiority over no irrigation (Test 1) and
1044 irrigation with saline (Test 2) with respect to the incidence of SSI within 30 days of surgery using
1045 two Fisher exact tests with the following hypotheses:

1046 Test 1: $H_{1_0}: \pi_P = \pi_N$ vs. $H_{1_A}: \pi_P \neq \pi_N$

1047 Test 2: $H_{2_0}: \pi_P = \pi_S$ vs. $H_{2_A}: \pi_P \neq \pi_S$

1048 Where π_P, π_N , and π_S denote the incidence of SSI within 30 days of surgery in the PHX, no
1049 irrigation, and saline groups respectively. The tests will be performed two-sided and with a
1050 global significance level of 5%. Using the Bonferroni-Holm adjustment, the local significance
1051 level for Test 1 will be 2.5% and for Test 2 it will be 5%.

1052

1053 **21.4 Supportive analysis of the primary endpoint**

1054 Since randomization will be stratified by study center and level of contamination, supportive
1055 analysis of the primary endpoint will also be performed using a binary logistic regression model
1056 with dependent variable SSI and covariates treatment group, study center, and level of
1057 contamination. In case there are differences between the treatment groups in terms of baseline
1058 characteristics, those will also be included as covariates in the model. Operation related risk
1059 factors (e.g. type and duration of surgery, administration and timing of antibiotic prophylaxis, use
1060 of wound-edge protectors, intraoperative changing of gloves, presence of an ostomy) and
1061 patient related risk factors (e.g. NNIS risk score, ASA, BMI, age, diabetes, smoking, alcohol
1062 consumption, duration of preoperative hospital stay, history of SSI, history of

1063 radio/chemotherapy) might influence the outcome, which is why they will also be included as
1064 model covariates.

1065

1066 **21.5 Secondary endpoints**

1067 Secondary endpoints will be analyzed by treatment group on the ITT set, using appropriate
1068 descriptive statistics. Any explorative statistical testing will be performed using a significance
1069 level of 5%. Subgroup analyses or treatment group comparisons will be performed for rate of
1070 superficial/deep/organ space SSI (according to CDC [1], attachment 1) stratified by the NNIS
1071 risk score and by level of contamination (class II,III or IV) during surgery (according to CDC [1]
1072 attachment 3). All AEs including SSI and local wound complications will be analyzed with
1073 incidence rates by treatment group and according to severity. AEs rated as related to the study
1074 treatment will be listed separately. In addition, the duration of hospital stay in days will be
1075 compared between the three study groups.

1076

1077 **21.6 Missing data**

1078 First analysis will be based on all patients with complete follow-up. For sensitivity, multiple
1079 imputations will be used for missing primary endpoint data. A dropout rate of 8-10% is expected
1080 in this study.

1081

1082 **22. RANDOMIZATION AND METHODS AGAINST BIAS**

1083 Participating, GCP-certified investigators will perform the screening and recruitment of patients
1084 and will obtain the ICF prior to inclusion. Every patient fulfilling inclusion and exclusion criteria
1085 will be documented. Reasons for non-inclusion into the study will have to be documented as
1086 well in a screening-list. A GCP-trained member of the study group will perform randomization
1087 during surgery after closure of the abdominal fascia is completed using RANDOBASE, the
1088 online-randomization tool at MSZ. RANDOBASE uses pre-defined randomization lists, which
1089 will be created at IMSE and will be stratified by level of contamination of the surgical procedure
1090 (clean-contaminated, contaminated or dirty) and by study center. To assure balanced group
1091 sizes in the course of the accrual, a block-wise randomization is applied. Basic characteristics of
1092 the patient and day of randomization must be documented on the randomization sheets.
1093 Subsequently, randomization sheets must be printed out, dated, signed and stored away from
1094 the patient records, trial documents and ISF to ensure blinding. Details on the blinding
1095 procedure are presented under point 11.4.

1096

1097 **23. FINAL REPORTING**

1098 After completion of the trial, BfArM and the leading ethics committee (TUM) have to be informed
1099 within 90 days by a final study report. Within one year of the completion of the trial, BfArM and
1100 the ethics committee will be supplied with a summary of the final report on the clinical trial
1101 containing the principle results. The sponsor is responsible for the generation of these final
1102 reports.
1103

1104 **24. PUBLICATION OF STUDY RESULTS**

1105 After completion of the clinical study, a multi-center manuscript of the study results will be
1106 prepared for publication in a reputable scientific journal according to the CONSORT statement.
1107 For this manuscript, final analyses will be generated from the study database and it will be
1108 subject to review by the sponsor. The publication of the principal results from any single center
1109 experience within the trial is not allowed until the preparation and publication of the multi-center
1110 results. Exceptions to this rule require prior approval of the sponsor. For purposes of abstract
1111 presentation and publication, any secondary publications will be delegated to the appropriate
1112 principal authors. However, final analyses and manuscript review for all multi-center data will
1113 require the approval of the sponsor. The use of professional writers is not intended. Details on
1114 publication rules and author order will be provided in the Clinical Trial Agreement.
1115

1116 **25. DECLARATION OF HELSINKI**

1117 The Declaration of Helsinki, 2008 (Seoul), is attached to the protocol.
1118

1119

26. ATTACHMENTS

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Attachment 1: Definition and classification of SSI according to CDC

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<p>Superficial Incisional SSI</p>	<p>Infection occurs within 30 days after the operation and infection involves only skin or subcutaneous tissue of the incision and at least one of the following:</p> <ol style="list-style-type: none"> 1 Purulent drainage, with or without laboratory confirmation, from the superficial incision. 2 Organisms isolated from an aseptically obtained culture of fluid or tissue from the superficial incision. 3 At least one of the following signs or symptoms of infection: pain or tenderness, localized swelling, redness, or heat <i>and</i> superficial incision is deliberately opened by surgeon, <i>unless</i> incision is culture-negative. 4 Diagnosis of superficial incisional SSI by the surgeon or attending physician. <p><i>Notes:</i> Do <i>not</i> report the following conditions as SSI:</p> <ol style="list-style-type: none"> 1 Stitch abscess (minimal inflammation and discharge confined to the points of suture penetration).
<p>Deep Incisional SSI</p>	<p>Infection occurs within 30 days after the operation and the infection appears to be related to the operation and infection involves deep soft tissues (e.g. fascial and muscle layers) of the incision and at least one of the following:</p> <ol style="list-style-type: none"> 1 Purulent drainage from the deep incision but not from the organ/space component of the surgical site. 2 A deep incision spontaneously dehisces or is deliberately opened by a surgeon when the patient has at least one of the following signs or 3 Symptoms: fever (>38°C), localized pain, or tenderness, unless site is culture-negative. 4 An abscess or other evidence of infection involving the deep incision is found on direct examination, during reoperation, or by histopathologic or radiologic examination. 5 Diagnosis of a deep incisional SSI by a surgeon or attending physician. <p><i>Notes:</i></p> <ol style="list-style-type: none"> 1 Report infection that involves both superficial and deep incision sites as deep incisional SSI. 2 Report an organ/space SSI that drains through the incision as a deep incisional SSI.
<p>Organ/Space SSI</p>	<p>Infection occurs within 30 days after the operation and the infection appears to be related to the operation and infection involves any part of the anatomy (e.g., organs or spaces), other than the incision, which was opened or manipulated during an operation and at least one of the following:</p> <ol style="list-style-type: none"> 1 Purulent drainage from a drain that is placed through a stab wound into the organ/space. 2 Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ/space. 3 An abscess or other evidence of infection involving the organ/space that is found on direct examination, during reoperation, or by histopathologic or radiologic examination. 4 Diagnosis of an organ/space SSI by a surgeon or attending physician.

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Attachment 2: Clavien Dindo classification of surgical complications

Grade	Definition
Grade I	Any deviation from the normal postoperative course without the need for pharmacological treatment or surgical, endoscopic, and radiological interventions
Grade II	Allowed therapeutic regimens are: drugs as antiemetics, antipyretics, analgetics, diuretics, electrolytes, and physiotherapy. This grade also includes wound infections opened at the bedside
Grade III	Requiring pharmacological treatment with drugs other than such allowed for grade I complications Blood transfusions and total parenteral nutrition are also included Requiring surgical, endoscopic or radiological intervention
Grade IIIa	Intervention not under general anesthesia
Grade IIIb	Intervention under general anesthesia
Grade IV	Life-threatening complication (including CNS complications)* requiring IC/ICU management
Grade IVa	Single organ dysfunction (including dialysis)
Grade IVb	Multiorgan dysfunction
Grade V	Death of a patient
Suffix "d"	If the patient suffers from a complication at the time of discharge, the suffix "d" (for "disability") is added to the respective grade of complication. This label indicates the need for a follow-up to fully evaluate the complication.
	*Brain hemorrhage, ischemic stroke, subarachnoidal bleeding, but excluding transient ischemic attacks. CNS, central nervous system; IC, intermediate care; ICU, intensive care unit

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Attachment 3. Classification of wound contamination levels according to CDC

Class I/ Clean	These are uninfected operative wounds in which no inflammation is encountered and the respiratory, alimentary, genital, or uninfected urinary tracts are not entered. In addition, clean wounds are primarily closed, and if necessary, drained with closed drainage. Operative incisional wounds that follow non-penetrating (blunt) trauma should be included in this category if they meet the criteria. Laparoscopic surgeries, surgeries involving the skin (such as biopsies), eye or vascular surgeries are good examples.
Class II/ Clean-Contaminated	An operative wound in which the respiratory, alimentary, genital, or urinary tracts are entered under controlled conditions and without unusual contamination. Specifically, operations involving the biliary tract, appendix, vagina, and oropharynx are included in this category, provided no evidence of infection or major break in technique is encountered.
Class III/ Contaminated	Open, fresh, accidental wounds. In addition, operations with major breaks in sterile technique (e.g., open cardiac massage) or gross spillage from the gastrointestinal tract, and incisions in which acute, non-purulent inflammation is encountered are included in this category. Contaminated wounds are also created when an outside object comes in contact with the wound (e.g. a bullet, knife blade or other pointy object).
Class IV/ Dirty-Infected	Old traumatic wounds with retained devitalized tissue and those that involve existing clinical infection or perforated viscera or a foreign object lodged in the wound or any wound that has been exposed to pus or fecal matter. This definition suggests that the organisms causing postoperative infection were present in the operative field before the operation.

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This classification scheme has been shown in numerous studies to predict the relative probability that a wound will become infected. Clean wounds have a 1-5% risk of infection; clean-contaminated 3-11%; contaminated, 10-17%; and dirty over 27% (CDC).

1137
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Attachment 4: ASA classification

ASA Score	Patient's Preoperative Physical Status
1	Normally healthy patient
2	Patient with mild systemic disease
3	Patient with severe systemic disease that is not incapacitating
4	Patient with an incapacitating systemic disease that is a constant threat to life
5	Moribund patient who is not expected to survive for 24 hours with or without operation

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Attachment 5: NNIS risk index

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The NNIS risk index is operation-specific and applied to prospectively collected surveillance data. The index values range from 0 to 3 points and are defined by three independent and equally weighted variables. 0 indicating the lowest and 3 the highest risk of SSI.

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One point is scored for each of the following when present:

1145
1146

(1) American Society of Anesthesiologists (ASA) Physical Status Classification of >2

1147
1148

(2) Either contaminated or dirty/infected wound classification (class III and IV)

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(3) Length of operation >T hours, where T is the approximate 75th percentile of the duration of the specific operation being performed.

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1152

The T Point for Common Surgical Procedures (NNIS report 2004)

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Operation	T Point (hrs)
Bile duct, liver, or pancreatic surgery	4
Colonic surgery	3
Herniorrhaphy	2
Appendectomy	1
Other digestive	2
Laparotomy	2
Small bowel	3
Splenectomy	3
Cholecystectomy	2
Gastric	3
Nephrectomy	4
Organ transplant	6

1156
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1159

27. REFERENCES

- 1161 1. Mangram, A.J., et al., *Guideline for Prevention of Surgical Site Infection, 1999. Centers for*
1162 *Disease Control and Prevention (CDC) Hospital Infection Control Practices Advisory Committee.*
1163 *Am J Infect Control, 1999. 27(2): p. 97-132; quiz 133-4; discussion 96.*
- 1164 2. Mihaljevic, A.L., et al., *Multicenter Double-Blinded Randomized Controlled Trial of Standard*
1165 *Abdominal Wound Edge Protection With Surgical Dressings Versus Coverage With a Sterile*
1166 *Circular Polyethylene Drape for Prevention of Surgical Site Infections: A CHIR-Net Trial (BaFO;*
1167 *NCT01181206). Ann Surg, 2014. 260(5): p. 730-9.*
- 1168 3. Diener, M.K., et al., *Effectiveness of triclosan-coated PDS Plus versus uncoated PDS II sutures*
1169 *for prevention of surgical site infection after abdominal wall closure: the randomised controlled*
1170 *PROUD trial. Lancet, 2014. 384(9938): p. 142-52.*
- 1171 4. Pinkney, T.D., et al., *Impact of wound edge protection devices on surgical site infection after*
1172 *laparotomy: multicentre randomised controlled trial (ROSSINI Trial). Bmj, 2013. 347: p. f4305.*
- 1173 5. Barnes, S., et al., *Surgical wound irrigation: A call for evidence-based standardization of practice.*
1174 *American Journal of Infection Control, 2014. 42(5): p. 525-529.*
- 1175 6. *Global Guidelines for the Prevention of Surgical Site Infection. 2016, World Health Organization:*
1176 *Geneva.*
- 1177 7. *National Institute for Health and Clinical Excellence (NICE). Surgical Site Infection - Prevention*
1178 *and Treatment of Surgical Site Infection. NICE Clinical Guideline 74. 2008, RCGO Press:*
1179 *London, England.*
- 1180 8. Rohner, E., et al., *Preferred use of polyhexanide in orthopedic surgery. Orthopedics, 2011.*
1181 *34(10): p. e664-8.*
- 1182 9. Roth, B., et al., *[Recommendations for the use of polyhexanide-containing products for the*
1183 *treatment of wounds]. Praxis (Bern 1994), 2011. 100(9): p. 531-7.*
- 1184 10. Fournel, I., et al., *Meta-analysis of intraoperative povidone-iodine application to prevent surgical-*
1185 *site infection. Br J Surg, 2010. 97(11): p. 1603-13.*
- 1186 11. Chundamala, J. and J.G. Wright, *The efficacy and risks of using povidone-iodine irrigation to*
1187 *prevent surgical site infection: an evidence-based review. Can J Surg, 2007. 50(6): p. 473-81.*
- 1188 12. Charalambous, C.P., et al., *When should old therapies be abandoned? A modern look at old*
1189 *studies on topical ampicillin. J Infect, 2003. 47(3): p. 203-9.*
- 1190 13. McHugh, S.M., et al., *The role of topical antibiotics used as prophylaxis in surgical site infection*
1191 *prevention. J Antimicrob Chemother, 2011. 66(4): p. 693-701.*
- 1192 14. Mueller, T.C., et al., *Intra-operative wound irrigation to reduce surgical site infections after*
1193 *abdominal surgery: a systematic review and meta-analysis. Langenbecks Arch Surg, 2015.*
- 1194 15. Cervantes-Sanchez, C.R., et al., *Syringe pressure irrigation of subdermic tissue after*
1195 *appendectomy to decrease the incidence of postoperative wound infection. World J Surg, 2000.*
1196 *24(1): p. 38-41; discussion 41-2.*
- 1197 16. Roth B, A.O., Wurmitzer F, Kramer A, *[Surgical site infections after primary antiseptic cleansing*
1198 *of dirty-contaminated wounds by polyhexanide, PVP iodine resp. hydrogen peroxide]. GMS*
1199 *Krankenhaushyg Interdiszip 2007. 2(2): p. Doc58.*
- 1200 17. Wundinfektionen, K.S.S.p. 2013 [cited 2014; Available from: [http://www.nrz-](http://www.nrz-hygiene.de/surveillance/kiss/)
1201 [hygiene.de/surveillance/kiss/](http://www.nrz-hygiene.de/surveillance/kiss/)].
- 1202 18. de Lissovoy, G., et al., *Surgical site infection: incidence and impact on hospital utilization and*
1203 *treatment costs. Am J Infect Control, 2009. 37(5): p. 387-97.*
- 1204 19. Gastmeier, P., et al., *Postoperative Wundinfektionen nach stationären und ambulanten*
1205 *Operationen. Bundesgesundheitsblatt - Gesundheitsforschung - Gesundheitsschutz, 2004. 47(4):*
1206 *p. 339-344.*
- 1207 20. Geffers, C., M.D., *Postoperative Wundinfektionen*, B.M. e.V., Editor. 2011, Institut für Hygiene
1208 und Umweltmedizin der Charité, Berlin.
- 1209 21. Mihaljevic, A.L., et al., *Wound edge protectors in open abdominal surgery to reduce surgical site*
1210 *infections: a systematic review and meta-analysis. PLoS One, 2015. 10(3): p. e0121187.*
- 1211 22. Clavien, P.A., et al., *The Clavien-Dindo classification of surgical complications: five-year*
1212 *experience. Ann Surg, 2009. 250(2): p. 187-96.*
- 1213 23. *Gesetz über den Verkehr mit Arzneimitteln (Arzneimittelgesetz - AMG). 2005, Bundesministerium*
1214 *für Justiz und Verbraucherschutz.*
- 1215 24. *International Conference on Harmonisation of Technical Requirements for Registration of*
1216 *Pharmaceuticals for Human Use, ICH E6. 1996, International Conference on Harmonisation;*
1217 <http://www.ich.org>; Geneva.
- 1218 25. Chan, A.W., et al., *SPIRIT 2013 statement: defining standard protocol items for clinical trials. Ann*
1219 *Intern Med, 2013. 158(3): p. 200-7.*

TECHNISCHE UNIVERSITÄT MÜNCHEN KLINIKUM RECHTS DER ISAR

STUDY PROTOCOL

IOWISI

Intraoperative wound irrigation to prevent surgical site infection after laparotomy

Sponsor:

Technische Universität München (TUM)
Fakultät für Medizin
Represented by the Dean
Ismaninger Str. 22
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Sponsor Delegated Person (SDP):

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EudraCT Number: 2017-000152-26

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DRKS Number: DRKS00012251

Version: 3.0 AM2.0

Date 02.03.2021

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This document is confidential and should serve as a source of information for Investigators and other personnel involved in this clinical study, consultants and ethics committees and regulatory authorities. The contents of this document shall only be disclosed to others in agreement with the coordinating investigator and/or sponsor.

1222 **Responsibility**

1223

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1224

1225 **28. DECLARATION OF INVESTIGATOR**

1226

1227 I have read the trial protocol and I confirm that it contains all information to accordingly conduct
1228 the clinical trial. I pledge the clinical trial will be conducted at my trial center according to the
1229 protocol.

1230

1231 The first patient will be enrolled only after all ethical and regulatory requirements are fulfilled. I
1232 pledge that written informed consent for trial participation will be obtained from all patients.

1233

1234 I know the requirements for accurate notification of serious adverse events and I pledge to
1235 document and notify such events as described in the protocol.

1236

1237 I pledge to retain all trial-related documents and source data as described. All necessary
1238 documents will be provided before trial start. I agree that these documents will be submitted to
1239 the responsible regulatory authorities and ethics committees.

1240

29. SYNOPSIS

Sponsor	Technische Universität München, Fakultät für Medizin
Name of the trial	Intraoperative wound irrigation to prevent surgical site infection after laparotomy - IOWISI
Trial design	Prospective, randomized, controlled, observer and patient-blinded, multicenter, surgical trial according to German drug law (AMG) phase IIIb, with three parallel comparison groups
Objectives	To investigate whether the use of intraoperative, epifascial wound irrigation with polyhexanide (PHX) solution can reduce surgical site infections after laparotomy for visceral surgery compared to saline irrigation or no irrigation.
Interventions	<p><u>Experimental intervention/index test:</u></p> <ul style="list-style-type: none"> Intervention 1: Irrigation of the subcutaneous tissue after closure of the abdominal fascia with 1000ml PHX solution (0.04%) Intervention 2: Irrigation of the subcutaneous tissue after closure of the abdominal fascia with 1000ml saline solution (NaCl 0.9%) <p><u>Control intervention/reference test:</u> No epifascial wound irrigation</p> <p><u>Follow-up per patient:</u> Postoperative day 30 (+6 at the latest)</p> <p><u>Duration of intervention per patient:</u> One intraoperative application</p> <p><u>Experimental and/or control off-label or on-label in Germany:</u> All interventions are on-label in Germany</p>
Key inclusion and exclusion criteria	<p><u>Key inclusion criteria:</u></p> <ul style="list-style-type: none"> Clean-contaminated, contaminated or dirty surgery (class II-IV) according to Centre for Disease Control (CDC) classification; Abdominal surgery by midline or transverse laparotomy; elective and emergency procedures; Age \geq 18 years; American Society of Anesthesiologists (ASA) score \leq 3; Ability to understand the nature and extent of the trial and to give written informed consent; <p><u>Key exclusion criteria:</u></p> <ul style="list-style-type: none"> Pregnancy or breast feeding; Known hypersensitivity/allergy to PHX; Inability to give/understand informed consent; Critical medical condition of emergency patients, precluding informed consent or sufficient time to reflect on the decision to participate in the trial; ASA $>$3; Inability to attend follow-up visits; Clean procedures according to the CDC classification or surgery without opening of the abdominal cavity; Laparoscopic surgery; Revision-surgery (previous abdominal surgery within the last 30 days); Planned re-laparotomy within 30 days; Severe immunosuppression; Concurrent abdominal wall infections; Pre-operative systemic antibiotic therapy within 5 days prior to surgery (except emergency pre-operative antibiotic treatment due to septic peritonitis after admission to the hospital); Participation in another clinical trial that interferes with the primary or secondary outcomes of this trial.
Outcomes	<p><u>Primary efficacy endpoint:</u> SSI according to CDC criteria within 30 days postoperatively</p> <p><u>Key secondary endpoint(s):</u></p> <ul style="list-style-type: none"> Non-infectious wound complications (e.g. seroma, hematoma, delayed healing) within 30 days postoperatively

	<ul style="list-style-type: none"> • Duration of hospital stay • Mortality and morbidity within 30 days postoperatively • Incidence of reoperation within 30 days postoperatively • Incidence of AE/SAE within 30 days postoperatively <p>Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space), NNSI risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of wound-edge protectors and changing of gloves, presence of an enterostomy.</p> <p><u>Safety:</u> Adverse events (AE) and serious adverse events (SAE) are documented for all groups. Surgical complications will be additionally evaluated according to the Clavien-Dindo classification</p>
Study registry	German CTR (DRKS): DRKS00012251 / EudraCT: 2017-000152-26
Statistical analysis	<p><u>Efficacy:</u> The incidence of SSI within 30 days after surgery will be compared between three study groups in two ways: Test 1: PHX irrigation vs. no intervention Test 2: PHX irrigation vs. saline irrigation</p> <p><u>Description of the primary efficacy analysis and population:</u> The incidence of SSI within 30 days of surgery will be compared in test 1 and test 2 using two Fine and Gray subdistributional hazard models with SSI as main event and relaparotomy and death as competing risks. Since randomization is stratified by study centre and level of contamination, the models will include covariates treatment group, study centre, and level of contamination.. Both analyses will be performed on the ITT set, consisting of all patients included in the study in the treatment arm they were randomized to. The global significance level is set to 5%. Using the Bonferroni-Holm adjustment, the local significance level will be 2.5% and 5% in the order of increasing p-value.</p> <p><u>Missing data:</u> Missing primary endpoint data in the primary analysis will be dealt with using competing risks and censoring. Missing SSI evaluation due to death or relaparotomy will be considered a competing risk. Missing SSI for all other reasons will be censored. Data will not be imputed for other analyses such as secondary or subgroup analyses.</p> <p><u>Safety:</u> The assessment of safety will be based on the frequency of AE/SAE other than SSI within the safety population (according to CTCAE Version 4.03), consisting of all patients randomized into the study.</p> <p><u>Secondary endpoint(s):</u> Secondary endpoints will be analyzed on the ITT set using appropriate descriptive statistics. Subgroup analyses will be performed by use of logistic regression models involving main effects and interaction effects. Any explorative statistical testing will be performed two-sided using a significance level of 5%.</p>
Sample size	<p><u>To be assessed for eligibility (n):</u> approximately 5500 <u>To be assigned to the trial (n):</u> 680 <u>To be analyzed (n):</u> 680</p> <p>The sample size was adjusted based on the changed analysis of SSI. The global significance level was set to 5% (two-sided tests). Since the PHX arm will be used twice for a comparison, the Bonferroni-Holm procedure was used to set the local alpha level for test 1 (PHX vs. no intervention) to 2.5% and for test 2 (PHX vs. saline irrigation) to 5%. If 290 patients are recruited in the PHX arm, 290 patients in the saline arm and 100 patients in the control arm (a total of 680 patients), the two Fine and Gray sub-distributional hazard models will have a power of 80% each to detect differences between the treatments.</p>
Trial duration subject	<p><u>Intervention:</u> Single intraoperative intervention <u>Follow-up:</u> max. 36 days</p>
Trial duration project	<p><u>First patient in to last patient out (months):</u> 55 <u>Recruitment period (months):</u> 54 <u>Duration of the entire trial (months):</u> 61</p>
Participating centers	Planned: n about 11
Financing	Deutsche Forschungsgemeinschaft (DFG) grant number: MU 3928/1-1

1242 **30. ABBREVIATIONS**

1243	AE	Adverse Event
1244	ALT/ALAT	Alanine Aminotransferase
1245	AMG	Arzneimittelgesetz
1246	aPTT	Activated partial Thromboplastin time
1247	ASA	American Society of Anesthesiologists
1248	AST/ASAT	Aspartate Aminotransferase
1249	BfArM	Bundesinstitut für Arzneimittel und Medizinprodukte
1250	BMI	Body-Mass Index
1251	CDC	Centre for Disease Control and Prevention
1252	CI	Confidence Interval
1253	Cr	Creatinine
1254	CTCAE	Common Terminology Criteria for Adverse Events
1255	DFG	Deutsche Forschungsgemeinschaft
1256	DRKS	Deutsches Register Klinischer Studien
1257	DSUR	Development Safety Update Report
1258	eCRF	electronic Case Report Form
1259	EDTA	Ethylene-diamineteraacetic acid
1260	GCP	Good Clinical Practice
1261	Glu	Glucose
1262	ICF	Informed consent form
1263	ICH	International Conference on Harmonization
1264	ICMJE	International Committee of Medical Journal Editors
1265	IMP	Investigational Medicinal Product
1266	IMSE	Institut für Medizinische Statistik und Epidemiologie
1267	INR	International normalized ratio
1268	IOWI	Intraoperative wound irrigation
1269	ISF	Investigator site file
1270	ITT	Intention-To-Treat
1271	K	Potassium
1272	MeSH	Medical Subject Heading
1273	MRI	Klinikum München rechts der Isar
1274	MSZ	Münchener Studienzentrum
1275	Na	Sodium
1276	NaCl	Sodium chloride
1277	NICE	National Institute for Health and Clinical Excellence
1278	NNIS	National Nosocomial Infections Surveillance
1279	PHX	Polyhexanide
1280	PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
1281	PT	Prothrombin time
1282	PVP	Polyvinylpyrrolidone, Povidone
1283	RCT	Randomized Controlled Trial
1284	RDE	Remote Data Entry
1285	SAE	Serious Adverse Event
1286	SAR	Serious Adverse Reaction
1287	SAS	Statistical analysis system
1288	SGOT	Serum glutamic oxaloacetic transaminase
1289	SGPT	Serum glutamic pyruvic transaminase
1290	SMB	Safety Monitoring Board
1291	SmPC	Summary of product characteristics
1292	SOP	Standard operating procedure
1293	SSI	Surgical site infection
1294	SUSAR	Suspected Unexpected Serious Adverse events
1295	TUM	Technical University of Munich
1296	WHO	World Health Organization
1297		

1298 **31. INTRODUCTION**

1299 **31.1 The medical problem**

1300 Postoperative surgical site infection (SSI) represents the third most common hospital infection.
1301 According to the CDC's classification [1], SSI can be subdivided into infections of the
1302 subcutaneous tissue (superficial SSI), deep soft tissues such as fascial and muscle layers
1303 (deep SSI) and infections of organs or spaces (organ/space SSI) that occur within 30 days after
1304 surgery (attachment 1). In abdominal surgery, SSI rates are especially high. Recent high-level
1305 randomized controlled trials (RCTs) with standardized SSI definitions found rates between
1306 14.5% (BaFO trial) [2], 15.4% (PROUD trial) [3] and 25.0% (ROSSINI trial) [4] following
1307 laparotomy. Therefore, measures to prevent SSI in this field are urgently needed. Prophylactic
1308 intraoperative wound irrigation (IOWI) of the subcutaneous and deep soft tissue before skin
1309 closure with saline or antiseptic solutions hypothetically represents an easy and economical
1310 option to reduce SSI rates and is already frequently used in clinical practice, even though there
1311 are currently no definite recommendations on this practice [5]. The latest official guideline for the
1312 prevention of SSI by the World Health Organization (WHO) published in 2016, states that IOWI
1313 with saline is not efficient, but IOWI with diluted Polyvinylpyrrolidone (PVP)-iodine solutions has
1314 a potential benefit in preventing SSI, however, due to the low level of underlying evidence these
1315 recommendations are conditional and not limited to abdominal surgery [6]. In contrast, the
1316 clinical guidelines of the British National Institute for Health and Clinical Excellence (NICE) from
1317 2008 state that IOWI's efficacy is unproven and its use should be avoided at all. However, this
1318 recommendation too, is based on a small number of unstandardized RCTs evaluating different
1319 types of surgery and irrigation solutions [7]. Antiseptic PHX-based solutions are approved for
1320 intraoperative soft-tissue wound irrigation in surgery, and have been shown to be tissue
1321 tolerable and even promote wound healing. To our knowledge prophylactic PHX wound
1322 irrigation has not yet been evaluated in RCTs in abdominal, visceral surgery [8, 9].

1323

1324 **31.2 Evidence**

1325 Even though the literature concerning prevention of SSI is substantial, high-level evidence to
1326 guide decisions on the use of IOWI with saline or antiseptics remains scarce. Clinical trials
1327 investigating the efficacy of IOWI have been conducted mainly in the 1980-90's and their results
1328 are inconclusive and heterogeneous patient inclusion and outcome criteria were used. A few
1329 authors conducted systematic reviews and meta-analyses investigating specific irrigation
1330 solutions such as PVP-iodine or antibiotic solutions [10-13]. However, none of these reviews
1331 resulted in a definite conclusion, although they all observed a positive trend in the reduction of
1332 SSI rates through IOWI. Furthermore, more recent clinical trials have been conducted in the
1333 meantime. Therefore, we performed a large-scale meta-analysis in accordance with the
1334 Cochrane guidelines of the existing evidence on IOWI with saline, PVP-iodine or antibiotic

1335 irrigation solutions. Pubmed/MEDLINE, EMBASE, and the Cochrane Central Register of
1336 Controlled Trials (CENTRAL) were searched in May 2013. The following search terms were
1337 used in various combinations: prevention of surgical site infection, abdominal surgery, surgical
1338 wound infection/prevention and control [MeSH Terms], wound irrigation, wound lavage,
1339 incisional surgical site infection, intra operative irrigation, intra operative lavage, antibiotic
1340 irrigation, antibiotic irrigation solutions, iodine irrigation, povidone iodine irrigation, saline
1341 irrigation, and topical anti-infective agents [MeSH Terms]. The abstract and title search was
1342 limited to clinical trials published in English or German between January 1, 1970 and May 1,
1343 2013. In addition, all articles within the reference list of retrieved studies and reviews were
1344 hand-searched. The search was performed by two independent reviewers and followed the
1345 published protocol corresponding to the PRISMA statement and the Cochrane Handbook of
1346 systematic reviews of interventions. Prospective RCTs investigating the primary outcome of
1347 postoperative SSI after IOWI of the surgical incision after closure of the fascia or peritoneum
1348 and before skin closure were eligible for inclusion. Eligible irrigation solutions were saline, PVP-
1349 iodine, or topical antibiotics in different forms and concentrations (dry powder sprays or wound
1350 powder were also acceptable), irrespective of the closure and irrigation technique. Acceptable
1351 comparators were 'no irrigation' or irrigation with saline. All types of open abdominal surgeries
1352 were eligible, including visceral, gynecological, urological, or vascular procedures irrespective of
1353 the urgency of operation (elective or emergency). All trials reporting clinical SSI were included
1354 irrespective of the SSI definition used. Trials in which only one of the compared treatment arms
1355 received systemic prophylactic antibiotics were excluded, as this would have caused substantial
1356 bias. Methodological quality of individual clinical trials was assessed by examination of the
1357 allocation sequence, allocation concealment and double blinding using the Cochrane tool for
1358 assessing the risk of bias [21]. The risk of bias was graded as low, unclear, or high. In addition,
1359 the risk of publication bias was investigated by means of a funnel plot. Due to the naturally
1360 expected heterogeneity in performance of surgical procedures between different types of
1361 surgery, grade of contamination, and hence trials, random effect models with Mantel-Haenszel
1362 weights were used to estimate the average treatment effect and a corresponding 95 % CI.
1363 Forest plots were shown to illustrate treatment effects estimated for each trial and the estimated
1364 average treatment effect for all investigated subgroups. A two-sided level of significance of less
1365 than 5.0 % was considered for all tests. The results of this analysis show a risk reduction of 46
1366 % in the treatment group (IOWI with any irrigation solution). Incidence of SSI was 9% in the
1367 irrigation group compared to 16% in the untreated group [14]. However, the majority of included
1368 trials have been published from 1970 to 1990, and the quality assessment revealed that most of
1369 them were at a high risk of bias, mainly because of insufficient data reporting and
1370 methodological flaws. Methods of sequence generation, allocation concealment, and blinding
1371 were often inadequate or not reported. In addition, interventions, follow-up times, and definitions

1372 of SSI varied widely between studies, which might explain the large variance in overall SSI rates
1373 between 3.0 and 58.2%. Most studies used a non-standardized definition of SSI. The current
1374 internationally accepted CDC definition was not published until 1999. The funnel plot showed an
1375 asymmetry, which indicates a possible publication bias, as all included trials with a high
1376 standard error for the log odds ratio show a large benefit for the experimental group.
1377 Furthermore, PVP-I and antibiotic solutions are currently not recommended for this indication
1378 due to potential adverse side effects, tissue toxicity and the increased development of
1379 antimicrobial resistances. The only standardized RCT comparing IOWI with saline irrigation vs.
1380 no irrigation after open appendectomies was published in 2000 and found a reduction of SSI
1381 from 25% to 8.7% in the saline group [15]. Recently, PHX-based antiseptic solutions are
1382 successfully and widely used in orthopedic and trauma surgery. Wound irrigation with PHX
1383 showed a reduction of the SSI rate of almost 75% compared to Ringers solution in traumatic
1384 dirty contaminated soft tissue wounds [16].

1385

1386 **31.3 The need for a trial**

1387 SSIs contribute significantly to postoperative morbidity and mortality. In Germany approximately
1388 128,000 SSIs are reported annually [17]. Studies have shown an increase of 6-24 days in the
1389 mean length of hospital stay if SSI occurs [18]. In addition to the risk and discomfort for the
1390 patient, SSIs dramatically increase treatment costs and indirect costs such as loss of workforce
1391 or insurance payments. In Germany, postoperative SSIs account for approximately 1 million
1392 extra days of hospitalization and additional costs of around € 3 billion per year [19, 20]. Clinical
1393 guidelines and clinical practice vary largely in terms of the use of IOWI to reduce the incidence
1394 of SSI [5]. The aim of this prospective, multicenter, randomized clinical trial is to show the
1395 reduction of SSI rates by IOWI with PHX compared to saline or no irrigation. Individual patients
1396 participating in this trial have the opportunity of directly benefitting of the anticipated positive
1397 effect of PHX and/or saline irrigation, whilst no negative effects are to be expected. The results
1398 of the trial will provide evidence for definite clinical recommendations that would change current
1399 clinical guidelines and practice. A commercial interest is not expected as PHX solutions are
1400 widely available and several companies offer this product in their portfolio. The trial further does
1401 not request a certain product in order to avoid compliance conflicts, but encourages
1402 collaborators to use the available product in their respective study sites.

1403

1404 **31.4 Summary and aims of the study**

1405 SSI is one of the most common complications following abdominal visceral surgery (14-25%) [2-
1406 4, 21] and dramatically increases length of hospital stay and costs. Hypothetically, IOWI before
1407 skin closure with saline or antiseptics might be a potential pragmatic option to reduce SSI rates.
1408 Currently, there are no official recommendations on its use and clinical practice varies largely.

1409 Solutions containing the antiseptic agent PHX are approved for IOWI, and were shown to
1410 promote wound healing [8, 9], but have not been evaluated in RCTs in abdominal visceral
1411 surgery. Therefore, we designed a multicenter, randomized, observer-blinded clinical trial
1412 evaluating the efficacy of IOWI with PHX solution or saline before skin closure after laparotomy.
1413 Based on a meta-analysis on IOWI with various solutions, a sample-size of 540 patients was
1414 calculated for a 3-armed study design (PHX- vs. saline irrigation vs. no irrigation). The trial shall
1415 be conducted in 10 centers within the German surgical trial network *CHIR-Net*. All patients
1416 undergoing visceral surgery by laparotomy within the recruitment period of 27 months will be
1417 screened for the trial. The primary endpoint is the incidence of SSI 30 days postoperatively,
1418 according to the CDC definition (attachment 1). The results of the trial will provide evidence for
1419 definite clinical recommendations regarding the use of IOWI and influence current guidelines
1420 and provide all participating patients the opportunity of an improved treatment.
1421

1422 **32. OUTCOME MEASURES**

1423 **32.1 Rationale of outcome measures**

1424 The primary efficacy endpoint of this trial is SSI within 30 days postoperatively, according to the
1425 internationally accepted and recommended SSI definition by the CDC [1]. This endpoint has
1426 been used in previous trials and assures comparability of the results [2-4, 21]. This endpoint is
1427 further considered to be of clinical relevance as SSI increases morbidity and mortality of
1428 individual patients, direct and indirect costs and prolongs hospital stay as outlined before. The
1429 secondary endpoint of non-infectious wound complications was chosen to evaluate, if PHX
1430 irrigation has an additional positive effect on wound healing. Furthermore, secondary endpoints
1431 are morbidity and mortality within 30 days postoperatively. For safety analyses and the duration
1432 of hospital stay to evaluate the potential economical benefit.

1433

1434 **32.2 Determination of primary and secondary measures**

1435 The primary efficacy endpoint measure of the trial is the incidence of SSI within 30 days after
1436 surgery diagnosed. Furthermore, in case of SSI, the depth of infection will be classified into one
1437 of three categories according to CDC definition (superficial, deep, organ-space, see attachment
1438 1). In addition, the following outcome measures have been defined as secondary endpoint
1439 measures and will be determined by the unit given in parentheses: a) Duration of hospital stay
1440 (in days); b) 30-days rate of reoperation in both groups (%); c) 30-days rate of non-infectious
1441 wound complications in both groups (in %); d) 30-days rate of postoperative AE/SAE in both
1442 groups (%); e) 30-days mortality in both groups (%); (f) 30-days morbidity in both groups (%). All
1443 AE/SAEs that are surgical complications will be additionally classified according to the Clavien
1444 Dindo classification of surgical complications (attachment 2) [22].

1445

1446 **33. FINANCING**

1447 The clinical trial is financed by a grant from the German Research Society (Deutsche
1448 Forschungsgemeinschaft; DFG), grant number: MU 3928/1-1. No co-financing by industry or
1449 other third parties applies. There is no conflict of interest for the management of the study. All
1450 participating trial sites have officially declared no conflict of interest within the eligibility
1451 evaluation of the MSZ. A commercial interest does not apply as PHX solutions are widely
1452 available and several companies offer this product in their portfolio. The trial further does not
1453 request a certain product in order to avoid compliance conflicts, but encourages collaborators to
1454 use the available product in their respective study sites.

1455

1456 **34. RISK / BENEFIT ANALYSIS**

1457 No additional risks for study patients are anticipated, since IOWI represents a clinically
1458 established standard method. PHX 0.04% irrigation solution is approved for surgical wound
1459 irrigation of soft tissue wounds. The study will be planned, conducted and analysed according to
1460 all relevant national and international rules and regulations according to AMG [23], ICH-GCP E6
1461 [24], and the Declaration of Helsinki, 2008 (see 27.). No specific risks are expected because
1462 IOWI is locally applied and neither application of PHX or saline will have systemic effects on the
1463 participants. Safety of PHX solutions has been demonstrated before in the marketing studies.
1464 Adverse effects may only be expected in the improbable event of accidental contamination of
1465 the respective irrigation solutions or in case of unknown hypersensitivity to PHX. The potential
1466 benefits of reduced SSIs outweigh the mentioned negligible adverse effects of PHX and saline.
1467 The subjects' safety is ensured by regular study visits, enforcing GCP-guidelines. A subject-
1468 insurance for all trial participants is mandatory according to AMG. The informed consent
1469 process adheres to GCP-guidelines, which maximize patients' safety and guarantee
1470 confidentiality.

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1472 **35. TRIAL IMPLEMENTATION**

1473 **35.1 General study design**

1474 This study is a prospective, randomized, controlled, observer and patient-blinded, multicenter,
1475 surgical trial with three parallel comparison groups. Pre-screening of potential patients
1476 (evaluation of inclusion and exclusion criteria) is possible up to 14 days prior to the planned
1477 procedure. Patients can be included in the trial if inclusion and exclusion criteria apply and

1478 written informed consent has been provided. In case of emergency procedures inclusion is
1479 possible on the same day as the procedure, if the patient is able to understand and provide
1480 written informed consent and has had a reasonable amount of time to think about the decision
1481 (see 12.3). Included patients are randomized to no epifascial wound irrigation, epifascial wound
1482 irrigation with saline 0.9% or epifascial wound irrigation with PHX 0.04% solution. Screened but
1483 excluded patients will be documented in a screening log.

1484

1485 **35.2 Trial duration**

1486 The estimated overall length of the study is 42 months, which assembles as follows:

1487 IV. Trial preparation: ~ 6 months

1488 V. Execution of study: First patient in to last patient out: ~ 55 months

1489 1. Begin of study: 3rd quarter, 2017

1490 2. End of study: 2nd quarter, 2022 (Completion of the last visit for the last patient
1491 represents the end of study)

1492 3. Recruitment period: ~ 54 months

1493 4. Duration of treatment per patient:

1494 a) Group with intervention 1: Surgery according to institutional standard,
1495 followed by one-time wound irrigation with PHX 0.04% solution.

1496 b) Group with intervention 2: Surgery according to institutional standard,
1497 followed by one-time wound irrigation with saline 0.9% solution.

1498 c) Control group: Surgery according to institutional standard, followed by no
1499 wound irrigation.

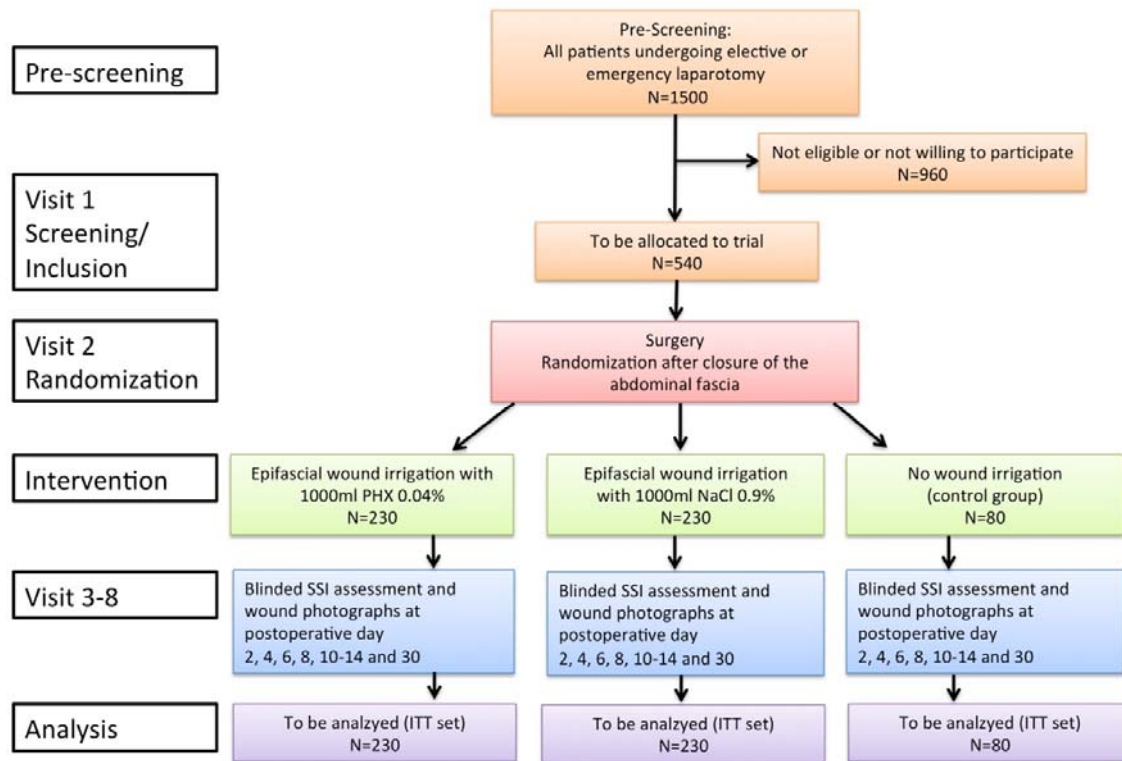
1500 5. Duration of follow-up per patient: 30 days (+6 days at the latest)

1501 For all three groups, documentation of the primary and secondary endpoints up to
1502 postoperative day 30 is warranted.

1503 VI. Analysis, publication ~ 7 months

1504

1505 **Graph 1: IOWISI intervention scheme / trial flow**



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Graph 2: IOWISI study visits (according to SPIRIT statement 2013 [25])

	STUDY PERIOD							
	INCLU.	RAND.	POST-ALLOCATION					CLOSE-OUT
STUDY VISIT	1	2	3	4	5	6	7	8
TIMEPOINT	- 1-3 days*	Surgery (day 0)	day 2	day 4	day 6	day 8	day 10-14	day 30 ^b
INCLUSION								
Informed consent	X							
Inclusion and exclusion criteria	X							
RANDOMIZATION								
		X						
INTERVENTIONS								
Intervention 1(IOWI with 1000ml PHX 0.04%)		X						
Intervention 2(IOWI with 1000ml NaCl 0.9%)		X						
Control group (no IOWI)		X						
ASSESSMENTS								
Demographical data	X							
Medical history	X							
Concurrent medication	X							
Physical examination	X							
NNSI Risk score	X							
Pregnancy test**	X**							
Blood sample***	X			X****				
Type of operation		X						
Duration of operation		X						
Level of contamination		X						
Type and length of incision		X						
Wound closure technique and suture material		X						
Creation of an enterostomy		X						
Administration and timing of antibiotic prophylaxis		X						
Intraoperative use of wound edge protectors		X						
Changing of gloves during operation		X						
Postoperative medication with effect on wound healing			X	X	X	X	X	X
Documentation of SSI			X	X	X	X	X	X
Documentation of other wound complications			X	X	X	X	X	X
Wound swab for microbiology [†]			X [†]	X [†]	X [†]	X [†]	X [†]	X [†]
Photograph of the wound			X	X	X	X	X	X
Documentation of re-operation			X	X	X	X	X	X
Documentation of AE/SAE		X	X	X	X	X	X	X
Duration of hospital stay								X

* In case of emergency surgery enrolment is possible on the same day as the procedure

**For women of child-bearing potential only (serum or urine)

***Includes hemoglobin, hematocrit, platelets and white blood cell count, Na, K, Cr, Glu (non-fasting), AST/ASAT (SGOT), ALT/ALAT (SGPT), Bilirubin, Uric acid, Prothrombin time (PT), activated partial thromboplastin time (aPTT), international normalized ratio (INR) according to local in-house standards

****Between post-OP day 4-8 (visit 4-6)

[†] In case of SSI a swab will be taken from the wound or wound secretion for microbiological differentiation and testing of resistance to antibiotics according to local in-house standards

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§ Visit window +6 days. If the patient is unable to attend visit 8 due to postoperative treatment in a rehabilitation facility or other medical reasons, a standardized protocol for evaluation and documentation of the wound will be sent to and filled out by the treating physician.

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36. JUSTIFICATION OF DESIGN ASPECTS

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36.1 Study design

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This trial is a prospective, randomized, controlled, observer and patient-blinded, multicenter, surgical trial according to German drug law (AMG) phase IIIb with three parallel comparison groups. Reduction of SSI (according to CDC criteria) by IOWI after abdominal surgery is postulated. The IOWISI trial will be conducted in approximately 10 surgical departments (university and community hospitals), all of which are members of the trial network (CHIR-Net) of the German Surgical Society (*Deutsche Gesellschaft für Chirurgie*) and have experience in previous multicenter RCTs. Feasibility evaluation of all participating centers was done according to the SOPs of MSZ. All of the study personnel involved in the trial require GCP training and will be specifically instructed in all trial-specific procedures before initiation of the trial. According to AMG, the investigator requires 2 years' experience in drug trials. The leading surgeon of the operating team will perform the interventions since they represent standard techniques. All participating surgeons will be instructed and authorized by the investigator, prior to the first trial procedure.

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36.2 Control and comparators

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The WHO published the latest clinical guideline addressing the topic of IOWI in surgery in 2016. The consensus is that there is not sufficient evidence to support the use of IOWI with saline, diluted PVP-solutions should be considered and antibiotic solutions avoided. However, the underlying RCTs included all types of surgery (*i.e.* neuro-, orthopedic surgery.) and are of low level of evidence [6]. The guideline of the British National Institute of Clinical Excellence (NICE) from 2008 [7] states that, due to the lack of evidence any IOWI should be avoided. However, in clinical practice this advice is mostly not being followed. Most hospitals do not have standard protocols but leave the decision to irrigate or not to irrigate the wound up to the surgeon. Given these circumstances it is acceptable to recruit a control group receiving no intervention. So far, no gold standard was determined within RCTs in abdominal surgery. Therefore, the trial proposes an irrigation procedure on the best available evidence, which is either irrigation with PHX-solution or saline or no irrigation. PHX and saline solutions are widely used in clinical practice, but efficacy trials are not available momentarily. As PHX solution is a market-approved drug, safety is ensured and the trial subjects are not exposed to specific risks.

1553 **36.3 Additional treatments**

1554 No additional treatments will be performed within the trial. Antibiotic treatment 5 days prior to
1555 surgery is an exclusion criterion. Pre-operative antibiotic treatment due to septic peritonitis (dirty
1556 / contaminated wounds) after admission to the hospital is allowed, but has to be recorded in the
1557 CRF. Application of routine intraoperative single shot antibiotic prophylaxis will be recorded in
1558 the CRF (type and dose of antibiotics). The application of abdominal wall protectors is
1559 recommended for contaminated procedures and has to be recorded in the CRF. A change of
1560 gloves ahead of wound closure is recommended for contaminated procedures and has to be
1561 recorded in the CRF. If indicated for medical reasons, all kind of medication is permitted during
1562 the trial. Postoperative medication with adverse effects on wound healing (e.g. corticoids and
1563 other immunosuppressive agents) will be recorded in the CRF. Any operative and / or
1564 interventional revision of the wound will be documented as AE/ SAE and classified after Clavien
1565 Dindo.

1567 **36.4 Blinding**

1568 The blinding procedure is restricted to participating patients, outcome assessors and the trial
1569 statistician. Blinding of the surgical team that performs the intervention is impossible because
1570 the control arm does not receive any wound irrigation. A member of the local study team, who
1571 will not take part in postoperative patient visits, performs randomization after confirmed closure
1572 of the abdominal fascia. A central online randomization tool of the MSZ (RANDOBASE) will
1573 effectuate randomization. After informing the surgical team of the result, the member of the
1574 study team has to print out, date and sign the randomization sheets. Subsequently, the
1575 randomization sheets have to be stored away from the patient records, trial documents and ISF
1576 to ensure blinding of the rest of the local study team.

1577 Postoperatively, a GCP-trained investigator of the local study group, who is unaware of the
1578 patient's intraoperative treatment, will clinically assess the primary endpoint (SSI) on 6 study
1579 visits.

1580 In addition, standardized photographs of the wound will be taken at each visit and uploaded to a
1581 central database. Independent, blinded outcome-assessors of spatially separated centers
1582 participating in the trial will assess those pseudonymized wound photographs in the database
1583 online. These online outcome-assessors receive training in rating of wounds according to the
1584 CDC classification of SSIs, which will be documented in a separate training log. These
1585 independent outcome-assessors will only access the photo-database for evaluation of SSI and
1586 will not be aware of the randomization results or any other patient data. All treatment-specific
1587 data are documented in a separate, undisclosed file. Wound photographs from all trial sites will

1588 be assessed by outcome assessors of the coordinating study site TUM in Munich. Photographs
1589 from the Munich TUM study site will be assessed in the study site Munich LMU.

1590 **36.5 Exclusion of participants after initial inclusion**

1591 Participants of the study can withdraw their consent to take part at any time without declaration
1592 of reasons. All hitherto collected data are subject to analysis. The coordinating investigator or
1593 the investigator may exclude patients from the study, if patients' safety is at risk or if there is
1594 insufficient compliance of the patient. In order to generate a meaningful database, excluded
1595 patients can be replaced by recruitment of new patients. If a patient does not receive PHX or
1596 saline irrigation of the wound, this does not automatically lead to exclusion of the study.

1597

1598 **37. INCLUSION- AND EXCLUSION CRITERIA**

1599 **37.1 Inclusion criteria**

- 1600 • Clean-contaminated, contaminated or dirty surgery according to CDC classification
1601 (attachment 3);
- 1602 • Abdominal surgery by midline or transverse laparotomy; elective and emergency
1603 procedures;
- 1604 • Age \geq 18 years;
- 1605 • American Society of Anesthesiologists (ASA) score \leq 3 (attachment 4);
- 1606 • Ability to understand the nature and extent of the trial and to give written informed
1607 consent

1608

1609 **37.2 Exclusion criteria**

- 1610 • Pregnancy or breast feeding;
- 1611 • Known hypersensitivity/allergy to PHX;
- 1612 • Inability to give/understand informed consent;
- 1613 • Critical medical condition of emergency patients, precluding informed consent or
1614 sufficient time to reflect on the decision to participate in the trial;
- 1615 • ASA $>$ 3;
- 1616 • Inability to attend follow-up visits;
- 1617 • Clean procedures according to the CDC classification or surgery without opening of the
1618 abdominal cavity;
- 1619 • Laparoscopic surgery;
- 1620 • Revision-surgery (previous abdominal surgery within the last 30 days);
- 1621 • Planned re-laparotomy within 30 days;
- 1622 • Severe immunosuppression;

- 1623 • Concurrent abdominal wall infections;
- 1624 • Pre-operative systemic antibiotic therapy within 5 days prior to surgery (except
- 1625 emergency pre-operative antibiotic treatment due to septic peritonitis after admission to
- 1626 the hospital);
- 1627 • Participation in another clinical trial that interferes with the primary or secondary
- 1628 outcomes of this trial.

1629

1630 **37.3 Explanation of inclusion and exclusion criteria**

1631 To enhance generalizability and representativeness, all patients undergoing elective and
1632 emergency laparotomy (transverse or midline) for visceral surgery will be screened for this trial.
1633 However, only clean-contaminated, contaminated or dirty (class II-IV), open abdominal surgery,
1634 according to the CDC classification [1] will be eligible, since in clean (class I) procedures the
1635 risk of SSI is low. Laparoscopic surgery as well as surgery without opening of the abdominal
1636 cavity or revision surgery (previous abdominal surgery within the last 30 days or planned re-
1637 laparotomy within the next 30 days of surgery) will be excluded, since these types of procedures
1638 are not comparable in terms of SSI risk.

1639 Pre-operative antibiotic therapy within 5 days prior to surgery was chosen to be an exclusion
1640 criterion to avoid bias of the results, since this might lead to a lower individual risk of infection.
1641 However, this does not apply to patients that receive pre-operative antibiotics after admission to
1642 the hospital in an emergency situation of septic peritonitis. Furthermore, this does not include
1643 standard intraoperative single shot antibiotic prophylaxis.

1644 Patients have to be ≥ 18 years of age and able to understand and give written informed
1645 consent. Any patient in a very bad general medical condition (ASA > 3) will be excluded to avoid
1646 too many patient-related confounders. Emergency patients in a critical medical condition that
1647 does not allow them to fully understand and provide informed consent or does not leave them
1648 sufficient time to reflect on the decision to participate in the trial will not be included.
1649 Furthermore, patients have to be able to attend follow-up visits.

1650 Patients with severe immunosuppression (e.g. after: organ or bone marrow transplantation,
1651 concurrent steroid treatment with >10 mg prednisone daily or an equivalent dose of any other
1652 steroid), concurrent infliximab treatment or treatment with an equivalent immunosuppressive
1653 substance, chemotherapy within the last 2 weeks prior to trial intervention) or patients with
1654 severe pre-operative neutropenia ($\leq 0.5 \times 10^9/L$) or liver cirrhosis Child-Pugh B/C will not be
1655 included. Pregnant or breast feeding women, as well as patients with a known
1656 hypersensitivity/allergy to PHX will not be included in the trial either.

1657 Patients that participate in other clinical trials that could interfere with the primary (SSI) or
1658 secondary outcomes of the IOWISI trial will be excluded.

1659

1660 **38. FREQUENCY AND SCOPE OF TRIAL VISITS**

1661 Graph 1 and 2 reflect the intervention scheme, trial flow, and visits for the IOWISI trial. Visits are
1662 the same for all participants of the study, regardless the treatment group.

1663

1664 **38.1 Recruitment and screening**

1665 Only surgical departments with adequate patient numbers, providing a written commitment on
1666 their recruitment capacity were included in the trial to reach the target sample size. The
1667 recruitment period is set to 54 months (first patient in to last patient out 55 months). In case of
1668 elective procedures, pre-screening (this is just a pre-selection of eligible patients within the
1669 study team) of patients can be performed up to 14 days prior to the scheduled surgical
1670 procedure. Screening and inclusion of patients will be performed not earlier than 3 days and not
1671 later than on the day before the planned surgical procedure, to ensure the patient has enough
1672 time to consider the decision to participate. In case of emergencies, screening and inclusion can
1673 take place on the day of admission to the hospital, which is usually the same day as surgery. All
1674 screened patients are documented in a screening log. If patients do not wish to participate in the
1675 study, reasons are documented accordingly. If patients fit inclusion/exclusion criteria and agree
1676 to participate, they will need to give written informed consent to the local GCP-trained
1677 investigator, after adequate time for consideration in order to participate in the study
1678 (representing visit 1). Therefore, at the screening visit, a detailed description of the study and
1679 further instructions are discussed with the patient, including methods of wound irrigation, risk-
1680 benefit-ratio, and follow up schedule.

1681

1682 **38.2 Visit 1 (Inclusion)**

1683 After the local investigator has reviewed the inclusion and exclusion criteria again and having
1684 received written consent by a patient, demographical data / medical history (date of birth
1685 [mm/yyyy], gender, body height, body weight, BMI, ASA, medical history, concurrent medication,
1686 history of SSI, history of radio/chemotherapy, diabetes, smoking, alcohol consumption,
1687 medication, duration of pre-operative hospital stay), diagnosis and the NNIS Risk score for
1688 determining the intrinsic risk of SSI (attachment 5) will be documented according to the eCRF.
1689 The investigator will perform a physical exam (blood pressure, heart frequency, condition of the
1690 planned abdominal surgical incision area, clinical relevant findings [normal or abnormal (please
1691 specify), respiratory system, cardiovascular system, liver, kidney, neurological or other free text]
1692 and take a blood sample (EDTA, Serum, and Citrate). Measurements of the blood sample are:

- 1693 • Hemoglobin
- 1694 • Hematocrit

- 1695 • Platelets
- 1696 • White blood cell count
- 1697 • Sodium
- 1698 • Potassium
- 1699 • Creatinine
- 1700 • Non-fasting glucose
- 1701 • AST/ASAT
- 1702 • ALT/ALAT
- 1703 • Bilirubin
- 1704 • Uric acid
- 1705 • Prothrombin time (PT)
- 1706 • Activated partial thromboplastin time (aPTT)
- 1707 • International normalized ratio (INR)

1708 In case of women of child-bearing potential, a pregnancy test will be performed additionally
 1709 (serum or urine [negative/positive/not performed with specification of reason as free text]).
 1710

1711 **38.3 Visit 2 (Surgery/Randomization)**

1712 Documented parameters of the surgical procedure include the urgency (emergency/elective),
 1713 type of surgical procedure (colorectal and/or small bowel and/or hepato-biliary and/or pancreatic
 1714 and/or splenectomy and/or gastric and/or esophageal and/or nephrectomy and/or urogenital
 1715 tract and/or others (freetext)) the duration of surgery (incision until complete skin closure,
 1716 minutes), the level of contamination according to CDC classification (class II-IV; see attachment
 1717 3), the intraoperative use of wound edge protectors (yes/no), and prophylactic changing of
 1718 gloves during of the operation (yes/no), type (transverse/midline) and length (cm) of the incision,
 1719 creation of an enterostomy (yes/no), the wound closure technique (subcutaneous sutures
 1720 (yes/no), stapler/suture, if suture: continuous/single) and used suture material, the
 1721 administration (yes/no) and timing (>1h/≤1h prior to incision) of antibiotic prophylaxis. If the
 1722 operating surgeon decides that incomplete closure of the wound and/or any other wound related
 1723 procedure after the study intervention (e.g. negative pressure treatment) is necessary for the
 1724 benefit of the patient, the patient will have to be excluded from the trial.

1725
 1726 Randomization (see section 24.) will take place at the end of surgery, after closure of the
 1727 abdominal fascia, when the level of contamination is definitely determined by the surgeon. A
 1728 designated member of the local study team (who will not perform postoperative study visits) will
 1729 perform randomization instantly by using the online tool of the MSZ (RANDOBASE) and inform
 1730 the surgeon of the result and according treatment. Date of randomization (mm:hh, dd/mm/yyyy),
 1731 successful randomization (yes/no), and the result of the randomization process are

1732 documented (printout). Subsequently, the randomization sheets have to be stored away from
1733 the patients file to ensure blinding.

1734 Study treatment according to randomization:

- 1735 • Wound irrigation with PHX 0,04% 1000ml
- 1736 • Wound irrigation with NaCl 0,9% 1000ml
- 1737 • No wound irrigation

1738 Furthermore, any AE or SAE is documented during this visit.

1739

1740 **38.4 Visit 3 to 8 (Post-op days 2, 4, 6, 8, 10-14, and 30-36)**

1741 Postoperatively, there will be 6 trial visits where an independent, blinded outcome assessor
1742 trained in the diagnosis and classification of SSI according to CDC definitions will examine
1743 wounds (SSI superficial or deep or organ/space, see attachment 1). In addition,
1744 pseudonymized, electronic pictures of the wound will be uploaded to a centralized database for
1745 independent and blinded evaluation (see 11.4). The assessors will not be aware of the study
1746 procedure or other details of the examined wound photograph. Postoperative medication with
1747 adverse effects on wound healing (e.g. corticoids and other immunosuppressive agents) will be
1748 documented in the eCRF.

1749 In case of SSI, microbiological swabs will be taken from the wound secretion for microbiological
1750 differentiation and testing of resistance to antibiotics according to in-house standards by each
1751 local institution. Other wound complications like seroma, hematoma, delayed healing or
1752 necrosis will be documented as secondary endpoint. Any surgical complication, including SSI,
1753 will be reported as AE/SAE and the Clavien Dindo classification (attachment 2) will be applied to
1754 specify the severity and consequent treatment. Furthermore, the rate of re-operations, mortality
1755 and occurrence of any AE or SAE will be documented (see 16). Additionally, the duration of the
1756 hospital stay (from admission to discharge or day of the visit, in days) will be documented on
1757 visit 8 (post-op day 30-36). To promote complete follow-up, a visit window of 6 additional days
1758 was implemented. In addition, patients can be recompensed for any travel expenses needed to
1759 attend study visit 8. If however, the patient is unable to attend visit 8 due to postoperative
1760 treatment in a rehabilitation facility or other medical reasons, a standardized protocol for
1761 evaluation and documentation of the wound (incl. wound photograph) will be sent to and filled
1762 out by the treating physician.

1763 Between post-op day 4 and 8 (visit 4, 5 or 6) one study-specific, post-operative blood sample
1764 will be taken, and the same measurements as upon visit 1 will be analyzed according to local
1765 clinical routine:

- 1766 • Hemoglobin
- 1767 • Hematocrit
- 1768 • Platelets
- 1769 • White blood cell count
- 1770 • Sodium
- 1771 • Potassium
- 1772 • Creatinine
- 1773 • Non-fasting glucose
- 1774 • AST/ASAT
- 1775 • ALT/ALAT
- 1776 • Bilirubin
- 1777 • Uric acid
- 1778 • Prothrombin time (PT)
- 1779 • Activated partial thromboplastin time (aPTT)
- 1780 • International normalized ratio (INR)

1781

1782 **39. DOSE, MODE AND SCHEME OF INTERVENTION**

1783 After closure of the abdominal fascia, patients will be randomized stratified by level of
1784 contamination of the operation. In the experimental group 1, the subcutaneous soft tissue will be
1785 irrigated with 1000 ml of a 0.04% PHX solution, which is the recommended concentration for
1786 surgical wound irrigation according to the SMPC. PHX solutions (0.04%) are approved for this
1787 indication in Germany. The wound shall be carefully rinsed throughout with the irrigation solution
1788 and the excess removed with suction. Debris and blood clots should be removed from the
1789 wound using irrigation/suction. The wound shall not be rubbed dry with abdominal cloths, but left
1790 moistened with the irrigation solution to ensure sufficient contact time for PHX to have the
1791 desired antiseptic effect. After irrigation with PHX the wound shall not be irrigated with saline or
1792 any other solution again. Since PHX is a cation-active substance, it is not compatible with
1793 anionic organic substances (e.g. lactate). Furthermore, the combination of PHX with PVP-I
1794 products should be avoided.

1795 In the experimental group 2, the same intervention will be performed using 1000ml of isotonic
1796 saline solution (NaCl 0.9%).

1797 The irrigation volume of 1000ml was chosen to be sure that even large laparotomy wounds
1798 would be sufficiently irrigated. This was determined by senior surgeons' clinical experience,
1799 since so far no recommendations for the optimal volume of surgical irrigation exist. After

1800 irrigation of the wound, the skin closure will be performed according to local standards, without
1801 any further wound-related procedure.

1802 In the control group, wounds will not be surgically irrigated, as is currently recommended in the
1803 NICE guideline. PHX solutions or saline are to be purchased, stored, and distributed according
1804 to the respective trial centers standard operating procedures. Trade name, dosage, batch and
1805 dispensed amount will be documented on a separate form.
1806

1807 **40. PATIENT, STUDY AND SITE DISCONTINUATION**

1808 **40.1 Patient discontinuation**

1809 Patients have the right to voluntarily withdraw from the study at any time for reason. In addition,
1810 the investigator has the right to withdraw a patient from the study at any time. Reasons for
1811 withdrawal from the study may include but are not limited to the following:

- 1812 ▪ Patient withdrawal of consent at any time;
- 1813 ▪ Any medical condition that the investigator or sponsor determines may jeopardize the
1814 patient's safety if he or she continues in the study;
- 1815 ▪ If it is discovered that a study subject is pregnant or may have been pregnant at the time
1816 of intervention (see point 16.9);
- 1817 ▪ Investigator or sponsor determines it is in the best interest of the patient to discontinue
1818 the study.

1819 Every effort should be made to obtain information on patients who withdraw from the study. The
1820 primary reason for withdrawal from the study should be documented on the appropriate eCRF.
1821 However, patients will not be followed for any reason after consent has been withdrawn.
1822 Patients who withdraw from the study will not be replaced.

1823

1824 **40.2 Study and site discontinuation**

1825 The **sponsor** has the right to terminate this study at any time. Reasons for terminating the study
1826 may include but are not limited to the following:

- 1827 ▪ The incidence or severity of AEs in this or other studies indicates a potential health
1828 hazard to patients;
- 1829 ▪ Unsatisfactory patient enrolment;
- 1830 ▪ The continuation of study is unethical or it has been proven that the therapy has a
1831 clearly negative influence;
- 1832 ▪ Unforeseen complications arise that no longer justify a continuation of the study;

1833

1834 The **sponsor** will notify the investigator of a decision to discontinue the study. The sponsor has
1835 the right to **close a site** at any time.

1836 Reasons for closing a site may include, but are not limited to, the following:

- 1837 ▪ Excessively slow recruitment;
- 1838 ▪ Poor protocol adherence;
- 1839 ▪ Inaccurate or incomplete data recording;
- 1840 ▪ Non-compliance with the ICH-GCP guideline;
- 1841 ▪ No study activity (*i.e.* all patients have completed and all obligations have been fulfilled);

1842 The **investigator** can discontinue the clinical study at his site at any time if he no longer
1843 considers the continuation of the study, for example if there are ethical and/or medical concerns.
1844

1845 **41. ADVERSE EVENTS (AES)**

1846 **41.1 Definition adverse event (AE)**

1847 An AE is any untoward medical occurrence in a patient or in a clinical investigation subject
1848 administered a pharmaceutical product, which does not necessarily have a causal relationship
1849 with this treatment. An AE can therefore be any unfavourable and unintended sign (including an
1850 abnormal laboratory finding), symptom or disease temporally associated with the use of a
1851 medicinal product, whether or not related to the treatment. Any AE has to be documented in the
1852 eCRF on the respective “Adverse Event Report Form”.
1853

1854 **41.2 Specific definitions of AEs in the IOWISI trial**

1855 The obligation to document any AE in the study, starts with the randomization and ends with
1856 completion of the last study visit. AE/SAEs are documented according to the standard grading
1857 on the AE/SAE reporting forms. Surgical site infections (primary endpoint) and all other local
1858 wound complications (secondary endpoint) will be documented as AE/SAE. In addition, their
1859 severity and the consequent treatment will be documented according to the Clavien Dindo
1860 classification (attachment 2). All laboratory values or events that will be assessed as “clinically
1861 significant” in the eCRF have to be documented as an AE. The responsible medical investigator
1862 will judge the clinical significance in the context of the postoperative course after laparotomy
1863 and the correspondent laboratory values before intervention.
1864

1865 **41.3 Serious adverse events (SAE) and other definitions**

1866 **Serious adverse events (SAEs)**

1867 A SAE is defined as any clinical event that at any time during the study participation:

- 1868 ▪ Results in death;

- 1869 ▪ Is life-threatening (the term life-threatening refers to an event in which the subject was at
1870 risk of death at the time of the event and not to an event which hypothetically might have
1871 caused death if it was more severe);
- 1872 ▪ Requires subject hospitalization or prolongation of existing hospitalization;
- 1873 ▪ Results in persistent or significant disability/ incapacity.
- 1874 ▪ Results in a congenital anomaly/birth defect or
- 1875 ▪ Is rated as another significant event or condition by the investigator
- 1876 Any SAE has to be reported to the MSZ immediately after becoming aware of the event (see
1877 chapter 16.7).

1878 **Suspected Unexpected Serious Adverse Reaction (SUSAR)**

1879 Serious AEs that are both suspected, *i.e.* possibly related to the investigational medicinal
1880 product (IMP) and ‘unexpected’, *i.e.* the nature and/ or severity of which is not consistent with
1881 the applicable product information, are to be classified as Suspected Unexpected Serious
1882 Adverse Reactions (SUSARs). If the second assessor classifies the SAR as ‘suspected’ (the
1883 relationship to the IMP is “related”, “probable” or “possible”) and unexpected, it will be
1884 categorized as a SUSAR. All SUSARs are subject to an expedited reporting to the responsible
1885 ethics committee(s), the competent federal authority (BfArM) and to all participating
1886 investigators (see 16.7). Furthermore, a report on all observed SAEs / SARs / SUSARs will be
1887 submitted once a year in the DSUR (Development Safety Update Report) format.

1888 **Period of observation and documentation**

1889 In this trial, all AEs that occur between the randomization (during surgery) and the last study
1890 visit or premature study termination will be documented on the pages provided in the eCRF.
1891 AEs must also be documented in the subject’s medical records. All subjects who have AEs,
1892 whether considered associated with the use of the trial medication or not, must be monitored to
1893 determine the outcome. The clinical course of the AE will be followed up until resolution or
1894 normalization of changed laboratory parameters or until it has changed to a stable condition.

1895

1896 **41.4 Evaluation of the severity**

1897 The grading of AEs in this trial will be carried out on the basis of the 5-grade scale defined in the
1898 CTCAE V4.03:

1899 **Grade 1:** Mild AE

1900 **Grade 2:** Moderate AE

1901 **Grade 3:** Severe AE

1902 **Grade 4:** Life-threatening AE or AE causing disablement

1903 **Grade 5:** Death related to AE

1904 The grading of all AEs listed in the CTCAE v4.03 will be based on the information contained
1905 therein. The grading of all other AEs, i.e. those which are not listed in the CTCAE v4.03 will be
1906 performed by a responsible investigator, based on definitions given above. In addition, surgical
1907 complications will be evaluated according to the Clavien Dindo classification.

1908 **41.5 Evaluation of the causal relationship**

1909 Investigators will estimate the causal relationship between the AE/SAE and the treatment. When
1910 estimating the causality the investigator may draw on known biophysical parameters,
1911 incorporate previous knowledge on the AE profile of the investigational product and possible
1912 simultaneously factor in the efficacy against other substances and the concomitant diagnoses of
1913 the patient. The investigator will categorize each AE that occurred after administration of the
1914 IMP regarding the coherency with the administration of the IMP as:

1915 - **Related:** There is a reasonable possibility that the event may have been caused by the
1916 IMP. A certain event has a strong temporal relationship and an alternative cause is
1917 unlikely.

1918 - **Probable:** An AE that has a reasonable possibility that the event is likely to have been
1919 caused by the IMP. The AE has a timely relationship and follows a known pattern of
1920 response, but a potential alternative cause may be present.

1921 - **Possibility:** An AE that has a reasonable possibility that the event may have been
1922 caused by the IMP. The AE has a timely relationship to the IMP; however, the pattern of
1923 response is untypical, and an alternative cause seems more likely, or there is significant
1924 uncertainty about the cause of the event.

1925 - **Unlikely:** Only a remote connection exists between the IMP and the reported AE. Other
1926 conditions including concurrent illness, progression or expression of the disease state or
1927 reaction of the concomitant medication appear to explain the reported AE.

1928 - **Not related:** An AE that does not follow a reasonable temporal sequence related to the
1929 IMP and is likely to have been produced by the subject's clinical state, other modes of
1930 therapy or other known aetiology.

1931

1932 **41.6 Outcome of AEs**

1933 The outcome of an AE at the time of the last observation will be classified as:

1934 - **Recovered/ Resolved:** All signs and symptoms of an AE disappeared without any
1935 sequels at the time of the last interrogation.

1936 - **Recovering/ Resolving:** The intensity of signs and symptoms has been diminishing
1937 and/ or their clinical pattern has been changing up to the time of the last interrogation in a
1938 way typical for its resolution. Further follow-up is possibly needed.

1939 - **Not recovered/ Not resolved:** Signs and symptoms of an AE are mostly unchanged at
1940 the time of the last interrogation. Further follow-up is possibly needed.

- 1941 - **Recovered/ Resolved with sequels:** The patient recovered with sequels from the AE /
1942 the AE resolved with sequels, *i.e.* the patient suffers from late complications or damage
1943 resulting from the AE.
1944 - **Fatal:** An AE resulting in death. If there are more than one AE only the AE leading to
1945 death (possibly, related) will be characterized as 'fatal'.
1946 - **Unknown:** The outcome is unknown or implausible and the information cannot be
1947 supplemented or verified.

1948

1949 **41.7 Reporting of serious adverse events (SAEs)**

1950 **Primary reporting of SAEs**

1951 All SAEs must be reported immediately, by fax (number 089/4140-6480) by the investigator to
1952 the responsible officer at the MSZ using the designated form.

1953 München Studienzentrum

1954 SAE-Reporting

1955 Ismaninger Straße 22

1956 81675 München

1957 Tel.: +49/89/4140-6477

1958 Fax: +49/89/4140-6480 or Email: sae-msz@mri.tum.de

1959 Reporting should be immediately after the investigator becomes aware of the event.

1960 The initial report must be as complete as possible including details of the current illness and
1961 SAE and an assessment of the causal relationship between the event and the trial medication.

1962 **Second assessment of SAEs**

1963 All SAEs will be subject to a second assessment by a designated person. This person is elected
1964 by the sponsor and will be independent from the sponsor and the reporting investigator. The
1965 second assessor will fill out a 'Second Assessment Form' for each SAE. The 'Second
1966 Assessment Form' will contain the following information:

1967 II) Assessment of seriousness of the event (investigator and second assessor)

1968 II) Assessment of relationship between SAE and IMP (investigator and second
1969 assessor)

1970 III) Assessment of expectedness of SAE, derived from IMP (second assessor)

1971 IV) A statement if the benefit/ risk assessment for the trial did change as a result of
1972 SAE (second assessor)

1973 The responsible safety officer of the MSZ will carry out the expedited reporting. Only SUSARs/
1974 SAEs occurring after administration of IMPs will undergo expedited reporting.

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41.8 Expedited reporting

Pursuant to the German and applicable EU laws and regulations, the ethics committee and health authorities will be informed of all suspected SUSARs and all SAEs resulting in death or being life-threatening occurring during the trial. Both institutions and all participating investigators will be informed in case the risk/ benefit assessment did change or any others new and significant hazards for subjects' safety or welfare occur. The sponsor has to ensure that all relevant information about a SUSAR, which occurs during the course of a clinical trial and is fatal or life threatening is reported as soon as possible and not later than seven days after the sponsor was first aware of the reaction. Any additional relevant information should be sent within eight days of the report. A SUSAR, which is not fatal, or life threatening has to be reported as soon as possible and in any event not longer than 15 days after the sponsor was first aware of the reaction.

41.9 Pregnancy

If, following initiation of the investigational product, it is subsequently discovered that a study subject is pregnant or may have been pregnant at the time of investigational product exposure, the investigator must immediately notify the sponsor of this event via the "Report on the drug exposure during pregnancy" and in accordance with SAE reporting procedures. The patient will be withdrawn from the study. Follow-up information regarding the course of the pregnancy, including perinatal and neonatal outcome and, where applicable, offspring information must be reported on a "Report on the pregnancy outcome during drug exposure". Any pregnancy occurring in a female partner of a male study participant the investigator becomes aware of should be reported to the sponsor. Information on this pregnancy may also be collected on the pregnancy reporting forms.

42. SAFETY MONITORING BOARD (SMB)

An independent Safety Monitoring Board (SMB according to the Guidance E3, ICH note for Guidance E6, ICH note for Guidance E9, Directive 2001/20EC "relating to the implementation of good clinical practice in the conduct of clinical trials on medicinal products for human use) is a group of experts external to the study that addresses the patient's safety and performs risk / benefit assessments. According to its operating procedures the SMB reviews accumulating safety data from ongoing trials to fulfill the safety monitoring. The rules of the SMB are deposited in the SMB Charta, (SOP_MSZ_AE04-H-A01_V02). The aim of this Charta is to define the composition, responsibilities, purpose and timing of meetings, details of the operation, including documentation and reporting and specifying the procedures to ensure confidentiality and appropriate communication of the SMB.

2012

2013 **43. ENSURING DATA QUALITY**

2014 **43.1 Documentation**

2015 All raw data such as patient records are declared as source documents. It must be ensured that
2016 they are available during routine monitoring visits. Apart from that the investigator of each site
2017 must maintain a separate patient identification list. The patient identification list will be
2018 maintained at the site separate from the documentation. The eCRF covers all the important
2019 forms, sorted according to visits. If a patient withdraws from the study, the reason must be
2020 recorded on the eCRF.

2021 **Data collection**

2022 The documentation of the study data in adherence to the GCP-guidelines and the clinical trial
2023 protocol is the responsibility of the investigator. Original data (source documents) remain in
2024 hospital medical record and information on the eCRF must be traceable and consistent with the
2025 original data. Source documents are e.g. laboratory results, photography, skin biopsy histology
2026 description and quality of life questionnaire, EASI, Pruritus VAS, TSQM. Original written
2027 informed consent signed by the patient is kept by the investigator and a signed copy will be
2028 given to the patient. No information in source documents about the identity of the patients will
2029 be disclosed. All data collected in this study must be entered in an eCRF which has to be
2030 completed by the investigator or authorized trial personnel and signed by the investigator. This
2031 also applies for those patients who do not complete the study. If a patient withdraws from the
2032 study, the reason must be recorded on the eCRF. The investigator is responsible for ensuring
2033 the accuracy, completeness, and timeliness of all data reported to the sponsor in the eCRFs
2034 and in all required reports.

2035 **Database management**

2036 Data are administered and processed by data management of the MSZ with the support of a
2037 study database (eCRF) according to the SOPs of the MSZ. A description of the study specific
2038 processes is given in the Data Management Plan that details the key planning and control
2039 elements for the data management component of the study.

2040 The evaluation of the data takes place by programmed validity- and consistency checks. In
2041 addition a manual/visual evaluation of plausibility is performed in accordance to the
2042 requirements of GCP. Queries may occur, which will be visualized on the study database. The
2043 investigator has to resolve all data discrepancies in the study database. After entry of all
2044 collected data and clarification of all queries, the database will be closed at the completion of
2045 the study. The database closure has to be documented. Data and results electronically
2046 recorded will be archived according to legal guidelines at least 10 years after study termination.

2047

2048 **43.2 Audits and inspections**

2049 As part of quality assurance according to GCP, the sponsor and the competent health
2050 authorities have the right to audit/inspect the study sites and any other institutions involved in
2051 the trial. The aim of an audit/inspection is to verify the validity, accuracy and completeness of
2052 data, to establish the credibility of the clinical trial, and to check whether the trial subject's rights
2053 and trial subject safety are being maintained. The sponsor may assign these activities to
2054 persons otherwise not involved in the trial (auditors). These persons as well as inspectors are
2055 allowed to access all trial documentation (especially the trial protocol, eCRFs, trial subjects'
2056 medical records, drug accountability documentation, and trial-related correspondence).

2057 The sponsor and all investigators of the participating study sites undertake to support auditors
2058 and inspections by the competent authorities at all times and to allow the persons charged with
2059 these duties access to the necessary original documentation. All persons conducting audits
2060 undertake to keep all trial subject data and other trial data confidential.

2061 After each external audit the investigator receives an audit confirmation from the responsible
2062 auditor. This confirmation has to be stored in the ISF in order to provide access to it in case of
2063 an inspection by the competent authorities. The audit report is provided to the sponsor for
2064 control.

2065 **43.3 Monitoring**

2066 Monitoring activities are performed to ensure that the trial is conducted in accordance with the
2067 trial protocol, the principles of GCP and local legislation. A monitoring manual describing the
2068 scope of the monitoring activities in detail will be prepared.

2069 The responsible monitor will contact the investigator and will be allowed, on request, to inspect
2070 the various records of the trial (eCRF and other pertinent data) provided that patient
2071 confidentiality is maintained in accord with local requirements. The monitor should have access
2072 to patient records, any information needed to verify the entries in the eCRF and all necessary
2073 information and essential study documents. The investigator agrees to cooperate with the
2074 monitor to ensure that any problems detected in the course of these monitoring visits are
2075 resolved. A monitoring visit report is prepared for each visit describing the progress of the
2076 clinical trial and all identified problems.

2077

2078 **43.4 Archiving**

2079 At the end of the clinical study all study-relevant data must be archived as required by law and
2080 when indicated in addition according to the Clinical Trial Agreement. All documentation forms,
2081 ICFs and other essential study documents must be retained as required by law. Patient ID lists

2082 and patient files are retained in the respective study sites separately. The ICFs are kept in with
2083 the study documents.

2084

2085 **44. ETHICAL AND REGULATORY ASPECTS**

2086 **44.1 Sponsor's and investigator's responsibilities**

2087 This study is conducted in compliance with all applicable laws and regulations and also the
2088 Declaration of Helsinki. **The sponsor has the overall responsibility for the ethical and**
2089 **scientific conduct of the study. All participating investigators agree to adhere to the**
2090 **instructions and procedures described in the study protocol and thereby to adhere to the**
2091 **principles of GCP that it conforms to.**

2092 The responsible ethics committee of TUM and health authority (BfArM) will review the final study
2093 documents. The ethics committee's and BfArM's decision concerning the conduct of the study
2094 will be communicated in written form to the sponsor. The sponsor will assure submission of
2095 required progress reports, annual safety reports and substantial amendments for approval to
2096 the ethics committee and BfArM. Before initiating the study, the sponsor must submit any
2097 required amendments to BfArM for review and acceptance to begin the trial according to § 42
2098 AMG. Furthermore, the sponsor has to inform the ethics committee and BfArM within 90 days
2099 about completion of the trial and provide a brief report of its outcome 1 year after completion of
2100 the trial. Results of the study will be reported following ICH-GCP-E6 and published according to
2101 the CONSORT statement.

2102

2103 **44.2 Independent ethics committees and health authorities**

2104 Prior to the start of this study, the protocol and other required documents would have to be
2105 reviewed and approved by the locally responsible ethics committees of each study site. Their
2106 reports as well as a signed and dated approval by the BfArM must be obtained and assessed by
2107 the leading ethics committee of the TUM before study initiation. Any amendments to the
2108 protocol, other than administrative ones (of which the leading ethics committee and BfArM will
2109 merely be informed), must be reviewed and approved by both authorities.

2110 Before inclusion of the first patient the federal state authorities (*zuständige Regierungsbehörden*
2111 *der Länder*) will be informed about the study. A copy of this report needs to be filed in the ISF
2112 and TMF.

2113

2114 **44.3 Ethical performance of the study**

2115 The study is conducted according to the ethical principles as defined in the Declaration of
2116 Helsinki, version of 2008 (see 28.). The present clinical study is conducted in accordance with
2117 principles published in the ICH-GCP Guideline and the applicable legal regulations (AMG, GCP-

2118 V, see 19.1) These principles concern ethics committee procedures, patient information and
2119 informed consent procedures, adherence to the protocol, administrative documents,
2120 documentation of the study medication, data collection, patient records (source documents),
2121 recording and reporting of AEs/SAEs, preparation of inspections and audits as well as storage
2122 and safekeeping of the documents. All the investigators and personnel involved in the study
2123 have been informed that international monitoring authorities, the competent federal authorities
2124 and the sponsor are authorised to review the study documents and patient files.

2125

2126 **44.4 Public register**

2127 Before the clinical study will be initiated, it will be filed at the German Clinical Trials Register
2128 (DRKS), which is part of the International Clinical Trials Registry Platform (ICTRP) of the WHO.
2129 After ethical approval the trial will be registered under the ID-number: DRKS00012251.

2130

2131 **44.5 Informed consent of the study participants**

2132 A patient can only be included in the study, if he provides written consent after being informed
2133 by a GCP-trained investigator (orally and in writing) about the nature, significance and scope of
2134 the clinical study in an appropriate and understandable way. The investigator must fully explain
2135 the purpose of the study to the patient or his/her guardian prior to entering the patient into the
2136 study. The investigator is responsible for obtaining written informed consent from each patient
2137 The person signing the consent form will receive a copy of the signed form. By providing such
2138 consent the patient is declaring that he understands and accepts the recording of data that is
2139 part of the study and its verification by authorised monitors or federal authorities. The patient will
2140 be educated about the potential benefits and complications of the IMP used in the study. It must
2141 be clear for him that he can withdraw his consent at any point of time without any disadvantages
2142 to his further treatment. The original copy of the written ICF will be kept in the study folder of the
2143 study site. The patient will be given the copies of the written patient information and ICF.
2144 Additionally, copies of both documents will be filed in the patient's medical file. Patient
2145 information and ICF are attached at the end of this protocol. The patient information and ICF will
2146 be submitted to the responsible ethics committee for assessment before the study will be
2147 initiated.

2148

2149 **45. INSURANCE FOR TRIAL PARTICIPANTS**

2150 In the clinical trial of an IMP, all the participants are insured in accordance with the AMG. The
2151 scope of the insurance coverage is derived from the insurance documents that are included in
2152 the ISF. Before inclusion the insurance conditions shall be submitted to the patient for review
2153 without request to do so. The insurance conditions should be furnished to the patient to take

2154 with him before being included in the study on request and after inclusion in any case.
2155 Insurance coverage is being provided by:

2156 HDI-Gerling Industrie Versicherung AG
2157 Niederlassung München
2158 Vertragsservice/M-B
2159 Ganghofer Strasse 37-39
2160 80339 Munich
2161 Tel.: +49 (89) 9243-420
2162 Fax.: +49 (89) 9243-356
2163 Insurance number: 65-963496-03037/390 (Studie: 2/17)

2164 If an insured event is suspected to have occurred, the sponsor is to be notified immediately. He
2165 then has to notify the insurance provider about the damages immediately. The patient will
2166 receive a copy of the notification to the insurance provider. The patient may also inform the
2167 insurance provider by bypassing the study personnel, reporting any claims. In this case, he
2168 should be notified that the sponsor of the clinical trial should still be informed about the event.
2169 Patients have to be informed about both options.

2170 **46. DATA PRIVACY PROTECTION / CONFIDENTIALITY** 2171 **PROTECTION**

2172 The applicable local regulations of data privacy protection will be followed. The patients will be
2173 informed that any patient-related data and materials will be appropriately made pseudonymous
2174 (pursuant to § 12 and § 13 of the GCP Regulations) and that these data may be used for
2175 analysis and publication purposes. Furthermore, the patients will be informed that their data
2176 may be inspected by representatives of BfArM or of the sponsor for the purpose of validation of
2177 a proper study conduct. Patients who do not provide consent for transmission of their data,
2178 according to the data protection agreement included in the ICF, will not be included in the
2179 clinical study.

2180 **47. PROTOCOL AMENDMENTS OR CHANGES IN TRIAL** 2181 **CONDUCT**

2182 In order to insure comparable conditions in all study sites and in the interest of standardized
2183 evaluations of the trial, changes in this protocol are not foreseen. However, changes in trial
2184 conduct are possible. Any change (besides administrative changes) of this protocol requires a
2185 written protocol amendment that must be reviewed by the sponsor before implementation.
2186 Furthermore, consent needs to be obtained by the investigator of each participating center.
2187 Amendments that significantly affect the safety of subjects, the scope of the investigation or the
2188 scientific quality of the study, additionally require approval of the leading ethics committee and

2189 BfArM. A copy of the written approval of these amendments must be provided to the sponsor
2190 and the investigator at each study site. Examples of amendments requiring such approval are:
2191 - Significant changes in the study design;
2192 - Increases in the number of invasive procedures.

2193 However, these requirements for approval should in no way prevent the investigator or sponsor
2194 to take any immediate action in the interests of preserving patient safety. If the investigator feels
2195 an immediate change to the protocol is necessary and is implemented for safety reasons, the
2196 sponsor, ethics committee and BfArM must be informed immediately. Amendments affecting
2197 only administrative aspects of the study do not require formal protocol amendments or ethics
2198 committee and BfArM approval. However, the ethics committee and BfArM must still be notified
2199 about the changes.
2200

2201 **48. STATISTICAL CONSIDERATIONS**

2202 **48.1 Proposed sample size / Power calculations**

2203 Due to the unexpected high number of dropouts, the sample size was adjusted based on the
2204 changed analysis of SSI (see section 23.3). The sample size was calculated (Sample Size
2205 Software, Sample Size Tables, D. Machin et al., 2009) based on the primary endpoints of the
2206 study, assuming SSI rates (event of interest) of 2.2% in the PHX group (assuming a 75% risk
2207 reduction according to the trial by Roth *et al.* [1]), 8.7% in the saline group (according to the
2208 results of the trial by Cervantes-Sanchez *et al.*[2]), and 16.2% in the control group (according to
2209 the meta-analysis by Mueller *et al.* [3]). The incidence rate of SSI over all study arms is then
2210 expected to be 7%, given the approximate 3:3:1 group assignment. The actual SSI rate up to
2211 now is 7.2%, which is very close to our assumption and we consider it valid. The incidence rate
2212 for the competing risks of death or re-laparotomy is estimated to be a total of 13.4% in all arms.
2213 This estimation is done based on the data collected up to now.

2214 The global significance level was set to 5% (two-sided tests). Since the PHX arm will be used
2215 twice for a comparison, the Bonferroni-Holm procedure was used to set the local alpha level for
2216 test 1 (PHX vs. no intervention) to 2.5% and for test 2 (PHX vs. saline irrigation) to 5%. If 290
2217 patients are recruited in the PHX arm, 290 patients in the saline arm and 100 patients in the
2218 control arm (a total of 680 patients, an increase of 140 in the sample size), the two Fine and
2219 Gray sub-distributional hazard models will have a power of 80% each to detect differences
2220 between the treatment groups. The comparison saline irrigation vs. control is not included in the
2221 sample size calculation, as it will not be analyzed in a confirmatory manner. The low medical
2222 interest cannot justify the large increase in patient numbers.

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48.2 Statistical analysis

The primary and secondary endpoints will be analyzed on the Intention-To-Treat (ITT) set, consisting of all patients included in the study in the treatment arm they were randomized to. The safety analysis will be performed on the safety set, consisting of all patients randomized into the study and assigned to the treatment group of their actual treatment.

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48.3 Primary endpoint

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Wound irrigation with PHX solution will be tested for superiority over no irrigation (Test 1) and irrigation with saline (Test 2) with respect to the incidence of SSI within 30 days of surgery using two Fine and Gray sub-distributional hazard models with SSI as main event and relaparotomy and death as competing risks. Since randomization is stratified by study centre and level of contamination, the models will include covariates treatment group, study centre, and level of contamination. The global significance level is set to 5%. Using the Bonferroni-Holm adjustment, the local significance level will be 2.5% and 5% in the order of increasing p-value.

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48.4 Supportive analysis of the primary endpoint

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Since randomization will be stratified by study center and level of contamination, supportive analysis of the primary endpoint will also be performed using a binary logistic regression model with dependent variable SSI and covariates treatment group, study center, and level of contamination. In case there are differences between the treatment groups in terms of baseline characteristics, those will also be included as covariates in the model. Operation related risk factors (e.g. type and duration of surgery, administration and timing of antibiotic prophylaxis, use of wound-edge protectors, intraoperative changing of gloves, presence of an ostomy) and patient related risk factors (e.g. NNIS risk score, ASA, BMI, age, diabetes, smoking, alcohol consumption, duration of preoperative hospital stay, history of SSI, history of radio/chemotherapy) might influence the outcome, which is why they will also be included as model covariates.

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Additionally, the incidence rates within 30 days for SSI, re-laparotomy, death, and lost to follow-up for other reasons will be displayed per treatment group and compared using Fisher's exact test in order to better understand the distribution of missing values.

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48.5 Secondary endpoints

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Secondary endpoints will be analyzed by treatment group on the ITT set, using appropriate descriptive statistics. Any explorative statistical testing will be performed using a significance level of 5%. Subgroup analyses or treatment group comparisons will be performed for rate of superficial/deep/organ space SSI (according to CDC [1], attachment 1) stratified by the NNIS

2259 risk score, level of contamination (class II,III or IV) during surgery (according to CDC [1]
2260 attachment 3) ASA score, BMI, age, diabetes, smoking, alcohol consumption, history of SSI,
2261 history of radio-/chemotherapy, preoperative hospital stay >2d, administration and timing of
2262 antibiotic prophylaxis, type and duration of surgery, intraoperative use of wound-edge protectors
2263 and changing of gloves, presence of an enterostomy by use of a binary logistic regression
2264 model with a main effect for treatment, the subgroup defining variable and a respective
2265 interaction effect. Description of treatment costs will be summarized as additional costs with
2266 respect to the no intervention group. All AEs including SSI and local wound complications will be
2267 analyzed with incidence rates by treatment group and according to severity. AEs rated as
2268 related to the study treatment will be listed separately. In addition, the duration of hospital stay in
2269 days will be compared between the three study groups.

2270

2271 **48.6 Missing data**

2272 Missing primary endpoint data in the primary analysis will be dealt with using competing risks
2273 and censoring. Missing SSI evaluation due to death or relaparotomy will be considered a
2274 competing risk. Missing SSI for all other reasons will be censored. Data will not be imputed for
2275 other analyses such as secondary or subgroup analyses.

2276

2277 **49. RANDOMIZATION AND METHODS AGAINST BIAS**

2278 Participating, GCP-certified investigators will perform the screening and recruitment of patients
2279 and will obtain the ICF prior to inclusion. Every patient fulfilling inclusion and exclusion criteria
2280 will be documented. Reasons for non-inclusion into the study will have to be documented as
2281 well in a screening-list. A GCP-trained member of the study group will perform randomization
2282 during surgery after closure of the abdominal fascia is completed using RANDOBASE, the
2283 online-randomization tool at MSZ. RANDOBASE uses pre-defined randomization lists, which
2284 will be created at IMSE and will be stratified by level of contamination of the surgical procedure
2285 (clean-contaminated, contaminated or dirty) and by study center. To assure balanced group
2286 sizes in the course of the accrual, a block-wise randomization is applied. Basic characteristics of
2287 the patient and day of randomization must be documented on the randomization sheets.
2288 Subsequently, randomization sheets must be printed out, dated, signed and stored away from
2289 the patient records, trial documents and ISF to ensure blinding. Details on the blinding
2290 procedure are presented under point 11.4.

2291

2292 **50. FINAL REPORTING**

2293 After completion of the trial, BfArM and the leading ethics committee (TUM) have to be informed
2294 within 90 days by a final study report. Within one year of the completion of the trial, BfArM and
2295 the ethics committee will be supplied with a summary of the final report on the clinical trial
2296 containing the principle results. The sponsor is responsible for the generation of these final
2297 reports.
2298

2299 **51. PUBLICATION OF STUDY RESULTS**

2300 After completion of the clinical study, a multi-center manuscript of the study results will be
2301 prepared for publication in a reputable scientific journal according to the CONSORT statement.
2302 For this manuscript, final analyses will be generated from the study database and it will be
2303 subject to review by the sponsor. The publication of the principal results from any single center
2304 experience within the trial is not allowed until the preparation and publication of the multi-center
2305 results. Exceptions to this rule require prior approval of the sponsor. For purposes of abstract
2306 presentation and publication, any secondary publications will be delegated to the appropriate
2307 principal authors. However, final analyses and manuscript review for all multi-center data will
2308 require the approval of the sponsor. The use of professional writers is not intended. Details on
2309 publication rules and author order will be provided in the Clinical Trial Agreement.
2310

2311 **52. DECLARATION OF HELSINKI**

2312 The Declaration of Helsinki, 2008 (Seoul), is attached to the protocol.
2313

2314

53. ATTACHMENTS

2315

Attachment 1: Definition and classification of SSI according to CDC

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<p>Superficial Incisional SSI</p>	<p>Infection occurs within 30 days after the operation and infection involves only skin or subcutaneous tissue of the incision and at least one of the following:</p> <ol style="list-style-type: none"> 5 Purulent drainage, with or without laboratory confirmation, from the superficial incision. 6 Organisms isolated from an aseptically obtained culture of fluid or tissue from the superficial incision. 7 At least one of the following signs or symptoms of infection: pain or tenderness, localized swelling, redness, or heat <i>and</i> superficial incision is deliberately opened by surgeon, <i>unless</i> incision is culture-negative. 8 Diagnosis of superficial incisional SSI by the surgeon or attending physician. <p><i>Notes:</i> Do <i>not</i> report the following conditions as SSI:</p> <ol style="list-style-type: none"> 2 Stitch abscess (minimal inflammation and discharge confined to the points of suture penetration).
<p>Deep Incisional SSI</p>	<p>Infection occurs within 30 days after the operation and the infection appears to be related to the operation and infection involves deep soft tissues (e.g. fascial and muscle layers) of the incision and at least one of the following:</p> <ol style="list-style-type: none"> 6 Purulent drainage from the deep incision but not from the organ/space component of the surgical site. 7 A deep incision spontaneously dehisces or is deliberately opened by a surgeon when the patient has at least one of the following signs or 8 Symptoms: fever (>38°C), localized pain, or tenderness, unless site is culture-negative. 9 An abscess or other evidence of infection involving the deep incision is found on direct examination, during reoperation, or by histopathologic or radiologic examination. 10 Diagnosis of a deep incisional SSI by a surgeon or attending physician. <p><i>Notes:</i></p> <ol style="list-style-type: none"> 3 Report infection that involves both superficial and deep incision sites as deep incisional SSI. 4 Report an organ/space SSI that drains through the incision as a deep incisional SSI.
<p>Organ/Space SSI</p>	<p>Infection occurs within 30 days after the operation and the infection appears to be related to the operation and infection involves any part of the anatomy (e.g., organs or spaces), other than the incision, which was opened or manipulated during an operation and at least one of the following:</p> <ol style="list-style-type: none"> 5 Purulent drainage from a drain that is placed through a stab wound into the organ/space. 6 Organisms isolated from an aseptically obtained culture of fluid or tissue in the organ/space. 7 An abscess or other evidence of infection involving the organ/space that is found on direct examination, during reoperation, or by histopathologic or radiologic examination. 8 Diagnosis of an organ/space SSI by a surgeon or attending physician.

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Attachment 2: Clavien Dindo classification of surgical complications

Grade	Definition
Grade I	Any deviation from the normal postoperative course without the need for pharmacological treatment or surgical, endoscopic, and radiological interventions
Grade II	Allowed therapeutic regimens are: drugs as antiemetics, antipyretics, analgetics, diuretics, electrolytes, and physiotherapy. This grade also includes wound infections opened at the bedside
Grade III	Requiring pharmacological treatment with drugs other than such allowed for grade I complications Blood transfusions and total parenteral nutrition are also included Requiring surgical, endoscopic or radiological intervention
Grade IIIa	Intervention not under general anesthesia
Grade IIIb	Intervention under general anesthesia
Grade IV	Life-threatening complication (including CNS complications)* requiring IC/ICU management
Grade IVa	Single organ dysfunction (including dialysis)
Grade IVb	Multiorgan dysfunction
Grade V	Death of a patient
Suffix "d"	If the patient suffers from a complication at the time of discharge, the suffix "d" (for "disability") is added to the respective grade of complication. This label indicates the need for a follow-up to fully evaluate the complication.
	*Brain hemorrhage, ischemic stroke, subarachnoidal bleeding, but excluding transient ischemic attacks. CNS, central nervous system; IC, intermediate care; ICU, intensive care unit

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Attachment 3. Classification of wound contamination levels according to CDC

Class I/ Clean	These are uninfected operative wounds in which no inflammation is encountered and the respiratory, alimentary, genital, or uninfected urinary tracts are not entered. In addition, clean wounds are primarily closed, and if necessary, drained with closed drainage. Operative incisional wounds that follow non-penetrating (blunt) trauma should be included in this category if they meet the criteria. Laparoscopic surgeries, surgeries involving the skin (such as biopsies), eye or vascular surgeries are good examples.
Class II/ Clean-Contaminated	An operative wound in which the respiratory, alimentary, genital, or urinary tracts are entered under controlled conditions and without unusual contamination. Specifically, operations involving the biliary tract, appendix, vagina, and oropharynx are included in this category, provided no evidence of infection or major break in technique is encountered.
Class III/ Contaminated	Open, fresh, accidental wounds. In addition, operations with major breaks in sterile technique (e.g., open cardiac massage) or gross spillage from the gastrointestinal tract, and incisions in which acute, non-purulent inflammation is encountered are included in this category. Contaminated wounds are also created when an outside object comes in contact with the wound (e.g. a bullet, knife blade or other pointy object).
Class IV/ Dirty-Infected	Old traumatic wounds with retained devitalized tissue and those that involve existing clinical infection or perforated viscera or a foreign object lodged in the wound or any wound that has been exposed to pus or fecal matter. This definition suggests that the organisms causing postoperative infection were present in the operative field before the operation.

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This classification scheme has been shown in numerous studies to predict the relative probability that a wound will become infected. Clean wounds have a 1-5% risk of infection; clean-contaminated 3-11%; contaminated, 10-17%; and dirty over 27% (CDC).

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Attachment 4: ASA classification

ASA Score	Patient's Preoperative Physical Status
1	Normally healthy patient
2	Patient with mild systemic disease
3	Patient with severe systemic disease that is not incapacitating
4	Patient with an incapacitating systemic disease that is a constant threat to life
5	Moribund patient who is not expected to survive for 24 hours with or without operation

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2335

Attachment 5: NNIS risk index

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The NNIS risk index is operation-specific and applied to prospectively collected surveillance data. The index values range from 0 to 3 points and are defined by three independent and equally weighted variables. 0 indicating the lowest and 3 the highest risk of SSI.

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One point is scored for each of the following when present:

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(1) American Society of Anesthesiologists (ASA) Physical Status Classification of >2

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(2) Either contaminated or dirty/infected wound classification (class III and IV)

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(3) Length of operation >T hours, where T is the approximate 75th percentile of the duration of the specific operation being performed.

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The T Point for Common Surgical Procedures (NNIS report 2004)

Operation	T Point (hrs)
Bile duct, liver, or pancreatic surgery	5
Colonic surgery	3
Herniorrhaphy	2
Appendectomy	1
Other digestive	2
Laparotomy	2
Small bowel	3
Splenectomy	3
Cholecystectomy	2
Gastric	3
Nephrectomy	4
Organ transplant	6

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54. REFERENCES

- 2356 1. Mangram, A.J., et al., *Guideline for Prevention of Surgical Site Infection, 1999. Centers for*
2357 *Disease Control and Prevention (CDC) Hospital Infection Control Practices Advisory Committee.*
2358 *Am J Infect Control, 1999. 27(2): p. 97-132; quiz 133-4; discussion 96.*
- 2359 2. Mihaljevic, A.L., et al., *Multicenter Double-Blinded Randomized Controlled Trial of Standard*
2360 *Abdominal Wound Edge Protection With Surgical Dressings Versus Coverage With a Sterile*
2361 *Circular Polyethylene Drape for Prevention of Surgical Site Infections: A CHIR-Net Trial (BaFO;*
2362 *NCT01181206). Ann Surg, 2014. 260(5): p. 730-9.*
- 2363 3. Diener, M.K., et al., *Effectiveness of triclosan-coated PDS Plus versus uncoated PDS II sutures*
2364 *for prevention of surgical site infection after abdominal wall closure: the randomised controlled*
2365 *PROUD trial. Lancet, 2014. 384(9938): p. 142-52.*
- 2366 4. Pinkney, T.D., et al., *Impact of wound edge protection devices on surgical site infection after*
2367 *laparotomy: multicentre randomised controlled trial (ROSSINI Trial). Bmj, 2013. 347: p. f4305.*
- 2368 5. Barnes, S., et al., *Surgical wound irrigation: A call for evidence-based standardization of practice.*
2369 *American Journal of Infection Control, 2014. 42(5): p. 525-529.*
- 2370 6. *Global Guidelines for the Prevention of Surgical Site Infection.* 2016, World Health Organization:
2371 Geneva.
- 2372 7. *National Institute for Health and Clinical Excellence (NICE). Surgical Site Infection - Prevention*
2373 *and Treatment of Surgical Site Infection. NICE Clinical Guideline 74.* 2008, RCGO Press:
2374 London, England.
- 2375 8. Rohner, E., et al., *Preferred use of polyhexanide in orthopedic surgery.* *Orthopedics, 2011.*
2376 *34(10): p. e664-8.*
- 2377 9. Roth, B., et al., *[Recommendations for the use of polyhexanide-containing products for the*
2378 *treatment of wounds]. Praxis (Bern 1994), 2011. 100(9): p. 531-7.*
- 2379 10. Fournel, I., et al., *Meta-analysis of intraoperative povidone-iodine application to prevent surgical-*
2380 *site infection. Br J Surg, 2010. 97(11): p. 1603-13.*
- 2381 11. Chundamala, J. and J.G. Wright, *The efficacy and risks of using povidone-iodine irrigation to*
2382 *prevent surgical site infection: an evidence-based review. Can J Surg, 2007. 50(6): p. 473-81.*
- 2383 12. Charalambous, C.P., et al., *When should old therapies be abandoned? A modern look at old*
2384 *studies on topical ampicillin. J Infect, 2003. 47(3): p. 203-9.*
- 2385 13. McHugh, S.M., et al., *The role of topical antibiotics used as prophylaxis in surgical site infection*
2386 *prevention. J Antimicrob Chemother, 2011. 66(4): p. 693-701.*
- 2387 14. Mueller, T.C., et al., *Intra-operative wound irrigation to reduce surgical site infections after*
2388 *abdominal surgery: a systematic review and meta-analysis. Langenbecks Arch Surg, 2015.*
- 2389 15. Cervantes-Sanchez, C.R., et al., *Syringe pressure irrigation of subdermic tissue after*
2390 *appendectomy to decrease the incidence of postoperative wound infection. World J Surg, 2000.*
2391 *24(1): p. 38-41; discussion 41-2.*
- 2392 16. Roth B, A.O., Wurmitzer F, Kramer A, *[Surgical site infections after primary antiseptic cleansing*
2393 *of dirty-contaminated wounds by polyhexanide, PVP iodine resp. hydrogen peroxide]. GMS*
2394 *Krankenhaushyg Interdiszip 2007. 2(2): p. Doc58.*
- 2395 17. Wundinfektionen, K.S.S.p. 2013 [cited 2014; Available from: [http://www.nrz-](http://www.nrz-hygiene.de/surveillance/kiss/)
2396 [hygiene.de/surveillance/kiss/](http://www.nrz-hygiene.de/surveillance/kiss/)].
- 2397 18. de Lissovoy, G., et al., *Surgical site infection: incidence and impact on hospital utilization and*
2398 *treatment costs. Am J Infect Control, 2009. 37(5): p. 387-97.*
- 2399 19. Gastmeier, P., et al., *Postoperative Wundinfektionen nach stationären und ambulanten*
2400 *Operationen. Bundesgesundheitsblatt - Gesundheitsforschung - Gesundheitsschutz, 2004. 47(4):*
2401 *p. 339-344.*
- 2402 20. Geffers, C., M.D., *Postoperative Wundinfektionen*, B.M. e.V., Editor. 2011, Institut für Hygiene
2403 und Umweltmedizin der Charité, Berlin.
- 2404 21. Mihaljevic, A.L., et al., *Wound edge protectors in open abdominal surgery to reduce surgical site*
2405 *infections: a systematic review and meta-analysis. PLoS One, 2015. 10(3): p. e0121187.*
- 2406 22. Clavien, P.A., et al., *The Clavien-Dindo classification of surgical complications: five-year*
2407 *experience. Ann Surg, 2009. 250(2): p. 187-96.*
- 2408 23. *Gesetz über den Verkehr mit Arzneimitteln (Arzneimittelgesetz - AMG).* 2005, Bundesministerium
2409 für Justiz und Verbraucherschutz.
- 2410 24. *International Conference on Harmonisation of Technical Requirements for Registration of*
2411 *Pharmaceuticals for Human Use, ICH E6.* 1996, International Conference on Harmonisation;
2412 <http://www.ich.org>; Geneva.
- 2413 25. Chan, A.W., et al., *SPIRIT 2013 statement: defining standard protocol items for clinical trials.* *Ann*
2414 *Intern Med, 2013. 158(3): p. 200-7.*

2415 **1.3 CSP summary of changes**

2416

2417 **Approvals and Amendments**

2418 **Initial Approval:** Bundesinstitut für Arzneimittel und Medizinprodukte (BfArM): 27.06.2017 (Nr.
2419 4042099); Ethics Committee (EC): 27.06.2017 (Nr. 173/17-Af) Clinical Study Protocol (CSP)
2420 Version (V) 2.0 06.06.2017

2421

2422 **Amendment 1:** Addition of Study sites #11 Würzburg and #12 Mannheim

2423 Approval of AM1: EC: 14.05.2019

2424

2425 **Amendment 2:** Major changes: Extension of study duration, Changes in statistical analysis plan
2426 because of an unexpected high number of drop-outs due to relaparotomy. To maintain statistical
2427 power, the sample size had to be increased to 680.

2428 Approval AM2: BfArM: 08.03.2021 EC: 24.03.2021, CSP V 3.0 02.03.2021

2429 During the course of the clinical trial BfArM/EC were informed of/approved further changes (e.g.
2430 due to addition of study sites and changes of PIs).

2431

2432

2433	<u>2. Statistical Analysis Plan (SAP)</u>		
2434	<u>2.1. SAP Original Version 1.0 (28.04.2021)</u>		
2435			
2436	Table of Contents		
2437			
2438	1	INTRODUCTION	86
2439	1.1	Background and rationale	86
2440	1.2	Study objectives.....	86
2441	1.3	Study endpoints	86
2442	2	STUDY METHODS	87
2443	2.1	Trial design	87
2444	2.2	Randomization	87
2445	2.3	Sample size	87
2446	2.4	Framework.....	88
2447	2.5	Statistical interim analyses and stopping guidance	88
2448	2.6	Timing of final analysis.....	88
2449	2.7	Timing of outcome assessments.....	88
2450	3	STATISTICAL PRINCIPLES	89
2451	3.1	Confidence intervals and P values	89
2452	3.2	General calculation rules.....	89
2453	3.3	Adherence and protocol deviations	89
2454	3.4	Analysis populations	89
2455	3.5	Event and censor times for the Kaplan-Meier analyses.....	89
2456	4	ANALYSIS.....	90
2457	4.1	Trial population.....	90
2458	4.1.1	Screening data.....	90
2459	4.1.2	Eligibility	90
2460	4.1.3	Recruitment.....	90
2461	4.1.4	Withdrawal/follow-up.....	90
2462	4.1.5	Baseline patient characteristics.....	91
2463	4.2	Outcome definitions.....	91
2464	4.3	Analysis methods for the primary and secondary endpoints.....	92
2465	4.3.1	Primary endpoint	92
2466	4.3.2	Supportive analysis of the primary endpoint.....	92
2467	4.3.3	Secondary endpoint analysis.....	92
2468	4.3.3.1	Non-infectious wound complications	93
2469	4.3.3.2	Hospital stay.....	93
2470	4.3.3.3	Thirty-day mortality	93
2471	4.3.3.4	Re-operation rate.....	93

2472	4.3.3.5	Subgroup analysis	93
2473	4.3.3.6	Treatment costs	94
2474	4.3.3.7	Adverse events	94
2475	4.4	Missing data.....	94
2476	4.5	Additional analyses.....	94
2477	4.6	Statistical software.....	94
2478	5	TABLE SHELLS	95
2479			
2480			

2481 **2 INTRODUCTION**

2482 **2.1 Background and rationale**

2483 Surgical site infection (SSI) is one of the most common complications following
2484 abdominal visceral surgery and dramatically increases length of hospital stay and costs.
2485 Hypothetically, intraoperative wound irrigation (IOWI) before skin closure with saline or
2486 antiseptics might be a potential pragmatic option to reduce SSI rates. Currently, there
2487 are no official recommendations on its use and clinical practice varies largely. Solutions
2488 containing the antiseptic agent polyhexanide (PHX) are approved for IOWI, and were
2489 shown to promote wound healing, but have not been evaluated in RCTs in abdominal
2490 visceral surgery. Therefore, we designed a multicenter, randomized, observer-blinded
2491 clinical trial evaluating the efficacy of IOWI with PHX solution or saline before skin
2492 closure after laparotomy. The primary endpoint is the incidence of SSI 30 days
2493 postoperatively, according to the CDC definition. The results of the trial will provide
2494 evidence for definite clinical recommendations regarding the use of IOWI and influence
2495 current guidelines and provide all participating patients the opportunity of an improved
2496 treatment.

2497 **2.2 Study objectives**

2498 To investigate whether the use of intraoperative, epifascial wound irrigation with PHX
2499 solution can reduce surgical site infections after laparotomy for visceral surgery
2500 compared to saline irrigation or no irrigation.

2501 **2.3 Study endpoints**

2502 Primary efficacy endpoint:

2503 SSI according to CDC criteria within 30 days postoperatively

2504 Secondary endpoints:

- 2505 • Non-infectious wound complications (e.g. seroma, hematoma, delayed
2506 healing) within 30 days postoperatively
- 2507 • Duration of hospital stay
- 2508 • Mortality and morbidity within 30 days postoperatively
- 2509 • Incidence of reoperation within 30 days postoperatively
- 2510 • Incidence of AE/SAE within 30 days postoperatively
 - 2511 ○ Surgical complications will be additionally evaluated according to the
2512 Clavien-Dindo classification.

2513 Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space),
2514 NNSI risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history
2515 of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration
2516 and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of
2517 wound-edge protectors and changing of gloves, presence of an enterostomy.

2518 **3 STUDY METHODS**

2519 **3.1 Trial design**

2520 This study is prospective, randomized, controlled, observer and patient-blinded,
2521 multicenter, surgical trial according to German drug law (AMG) phase III-b, with three
2522 parallel comparison groups.

2523 Patients are randomised to one of the following treatment arms:

2524 **Arm 1 (Intervention 1):**

2525 Irrigation of the subcutaneous tissue after closure of the abdominal fascia with
2526 1000ml PHX solution (0.04%)

2527 **Arm 2 (Intervention 2):**

2528 Irrigation of the subcutaneous tissue after closure of the abdominal fascia with
2529 1000ml saline solution (NaCl 0.9%)

2530 **Arm 3 (Control):**

2531 No epifascial wound irrigation

2532 A total of 680 patients (290 patients in arm 1, 290 patients in arm 2, and 100 patients in
2533 arm 3) in up to 15 centres will be enrolled in the study with duration of 34 days per
2534 patient (up to three days prior to surgery, day of surgery, and 30 days post-surgery).

2535 A total of 8 visits are scheduled during the study period.

2536 **3.2 Randomization**

2537 Patients are randomised blockwise ca. 3:3:1 to the treatment arms with stratification by
2538 centre and level of contamination of the surgical procedure (clean-contaminated,
2539 contaminated, or dirty) during surgery after closure of the abdominal fascia using
2540 RANDOBASE, the online-randomization tool at MSZ. RANDOBASE uses pre-defined
2541 randomization lists, which are created at IMedIS using Rancode Professional 2015. Two
2542 sets of sealed envelopes were produced: one for emergency unblinding at site, one for
2543 the same purpose at the MSZ-safety management department.

2544 **3.3 Sample size**

2545 Justification and calculation of the sample size can be found in the study protocol
2546 section 23.1.

2547 Sample size adjustment (CSP approved Version 3 Amendment 2 02.03.2021):

2548 Due to the unexpected high number of dropouts, the sample size was adjusted based
2549 on the changed analysis of SSI (see section 5.2.1). The sample size was calculated
2550 (Sample Size Software, Sample Size Tables, D. Machin et al., 2009) based on the
2551 primary endpoints of the study, assuming SSI rates (event of interest) of 2.2% in the
2552 PHX group (assuming a 75% risk reduction according to the trial by Roth et al. (14)),
2553 8.7% in the saline group (according to the results of the trial by Cervantes-Sanchez et
2554 al. (13)), and 16.2% in the control group according to results of the previously
2555 conducted meta-analysis (12). The incidence rate of SSI over all study arms is then
2556 expected to be 7%, given the approximate 3:3:1 group assignment. The actual SSI rate
2557 up to now is 7.2%, which is very close to our assumption and we consider it valid. The

2558 incidence rate for the competing risks of death or re-laparotomy is estimated to be a
2559 total of 13.4% in all arms. This estimation is done based on the data collected up to
2560 now.

2561 The global significance level was set to 5% (two-sided tests). Since the PHX arm will be
2562 used twice for a comparison, the Bonferroni-Holm procedure was used to set the local
2563 alpha level for test 1 (PHX vs. no intervention) to 2.5% and for test 2 (PHX vs. saline
2564 irrigation) to 5%. If 290 patients are recruited in the PHX arm, 290 patients in the saline
2565 arm and 100 patients in the control arm (a total of 680 patients), the two Fine and Gray
2566 sub-distributional hazard models will have a power of 80% each to detect differences
2567 between the treatments.

2568 **3.4 Framework**

2569 This study tests for superiority of (1) PHX over no intervention and (2) PHX over saline
2570 irrigation with respect to SSI rate within 30 days postoperatively.

2571 **3.5 Statistical interim analyses and stopping guidance**

2572 No interim analyses are planned for this study.

2573 **3.6 Timing of final analysis**

2574 The final analysis will be performed collectively at the end of the study.

2575 **3.7 Timing of outcome assessments**

2576 **Baseline** data will be collected at visit 1 and 2, which means up to day 0.

2577 **Day 0** is defined as the time of closure of the abdominal fascia.

2578 All other day definitions are relative to day 0.

2579 The primary outcome will be accessed up to visit 8, which means on day 30 and up to
2580 day 36 when the time window is considered.

Visit	Day	Time window
1	-3 to -1	Up to day 0
2	0 (surgery)	
3	2	
4	4	
5	6	
6	8	
7	10	Up to day 14
8	30	Up to day 36

2581

2582

2583 **4 STATISTICAL PRINCIPLES**

2584 **4.1 Confidence intervals and P values**

2585 All statistical tests will be performed two-sided at the global significance level of 5%.
2586 Adjustment for multiplicity will be done for the primary endpoint only, where the
2587 Bonferroni-Holm adjustment method will be used (see also section 5.2.1).

2588 Confidence intervals will be two-sided and 95%.

2589 **4.2 General calculation rules**

2590 1. Percentages will always be quoted using number of 'known' values in the
2591 denominator unless otherwise stated.

2592 2. P-values will be quoted to three decimal places only. Confidence intervals will
2593 also be quoted to three decimal places. However, if the statistical software SAS,
2594 which is used for analysis, prints four decimal places, values will not be rounded
2595 again, but printed to four decimal places.

2596 3. Chi-square tests in contingency tables will be replaced by Fisher's exact tests if
2597 any expected cell frequency is less than five.

2598 **4.3 Adherence and protocol deviations**

2599 The number and percent of patients per treatment group who received IOWI will be
2600 reported.

2601 Major protocol deviations will be listed per patient.

2602 **4.4 Analysis populations**

2603 The **Intention-to-Treat (ITT) population** will contain all randomised patients with
2604 results attributed to the treatment group they were randomised to.

2605 The **safety analysis (SA) population** will consist of all randomised subjects with
2606 results attributed to the treatment group of their actual treatment.

2607 Safety analysis will be performed on the SA population. All other analyses will be
2608 performed in the ITT population.

2609 **4.5 Event and censor times for the Kaplan-Meier analyses**

2610 The following definitions will apply to the time-to-event analysis of SSI:

Parameter:	SSI
Main Event Time	time of SSI
Competing Event Times	time of death time of re-laparotomy
Censor Time	time of last follow-up* / time of competing event#

2611 * for patients without SSI

2612 # for patients with re-laparotomy and for patients who died

2613

2614 **5 ANALYSIS**

2615 **5.1 Trial population**

2616 **5.1.1 Screening data**

2617 The overall number of screened patients will be presented in the report.

2618 The reasons for non-eligibility will also be summarized.

2619 **5.1.2 Eligibility**

2620 **Key inclusion criteria:**

- 2621 • Clean-contaminated, contaminated or dirty (according to CDC classification)
- 2622 • Abdominal surgery by midline or transverse laparotomy (elective or emergency)
- 2623 • Age ≥ 18 years
- 2624 • American Society of Anesthesiologists (ASA) score ≤ 3

2625 **Key exclusion criteria:**

- 2626 • Pregnancy or breast feeding
- 2627 • Known hypersensitivity/allergy to PHX
- 2628 • Inability to understand/give informed consent
- 2629 • Inability to attend follow-up visits
- 2630 • Revision-surgery (previous abdominal surgery within the last 30 days)
- 2631 • Planned re-laparotomy within 30 days
- 2632 • Severe immunosuppression
- 2633 • Concurrent abdominal wall infections
- 2634 • Pre-operative antibiotic therapy (within 5 days prior to surgery)

2635 The full set of inclusion and exclusion criteria can be found in the study protocol
2636 sections 12.1 and 12.2.

2637 **5.1.3 Recruitment**

2638 Tables will contain the following absolute and relative frequencies per treatment group
2639 and overall:

- 2640 • patients who entered the study.
- 2641 • patients within each analysis set including reasons for exclusion. Patients in the
2642 SA will be considered in the group of their actual treatment.
- 2643 • patients per centre on the ITT set.

2644 **5.1.4 Withdrawal/follow-up**

2645 Since study treatment is given only once at the beginning of the study, withdrawal from
2646 study treatment is not of interest.

2647 The number of patients per treatment group who did not complete the study will be
2648 given in a summary table by reason (multiple reasons possible) including absolute and
2649 relative frequencies.

2650 **5.1.5 Baseline patient characteristics**

2651 The following characteristics will be summarized per treatment group on the ITT set:

2652 Demographics: sex, age, BMI.

2653 Medical history: main diagnosis leading to operation (malign/benign), ASA classification,
2654 diabetes (including type and treatment), allergies, comorbidities (11 pre-defined and
2655 other; multiple comorbidities are possible), previous abdominal surgery (no, single,
2656 multiple), time since last abdominal surgery, history of SSI (no, single, multiple), time
2657 since last SSI, location of last SSI, history of radiotherapy (no, single, multiple), time
2658 since end of last radiotherapy, dose (Gy), history of chemotherapy (no, single, multiple),
2659 smoking (no, previously, currently), packyears, regular alcohol consumption (no,
2660 previously, currently), glasses/week.

2661 Surgery: duration of preoperative hospital stay (days), urgency and type of procedure,
2662 duration of surgery (min), antibiotic prophylaxis (no, yes >1h prior OP, yes ≤1h prior
2663 OP), type of skin disinfectant, type of incision, length of incision, intra-OP change of
2664 gloves (y/n), intra-OP use of wound edge protectors (y/n), enterostomy created (y/n),
2665 type of abdominal fascia closure, use of mesh (no, yes-sublay, yes-onlay), level of
2666 contamination (class I to IV), NNIS risk score, wound closure (complete/incomplete),
2667 subcutaneous sutures used (y/n), skin closure type (stapler, continuous suture, single
2668 suture).

2669 Absolute and relative frequencies will be presented for categorical variables. Number of
2670 valid cases, mean, standard deviation, median, minimum, and maximum will be
2671 displayed for continuous variables.

2672

2673 **5.2 Outcome definitions**

2674 The primary endpoint is the frequency of physician-assessed SSI up to 30 days post-
2675 surgery. According to the time-definitions, the SSI assessment may take place up to day
2676 36. Surgical site infections will additionally be classified in three groups: superficial
2677 incisional, deep incisional, and organ/space.

2678 The secondary endpoints of this study are:

2679 Frequency of non-infectious wound complications (e.g. seroma, hematoma, delayed
2680 healing) within 30 days postoperatively;

2681 Duration of hospital stay;

2682 Mortality within 30 days postoperatively;

2683 Rate of reoperation within 30 days post-surgery, which will be identified during medical
2684 review based on the AE records.

2685 Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space),
2686 NNSI risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history
2687 of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration
2688 and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of
2689 wound-edge protectors and changing of gloves, presence of an enterostomy.

2690 Assessment of safety:

2691 The assessment of safety will be based on the frequency of AE/SAE other than SSI
2692 within the safety population (according to CTCAE V. 4.3). Surgical complications will be
2693 additionally evaluated according to the Clavien-Dindo classification.

2694

2695 **5.3 Analysis methods for the primary and secondary endpoints**

2696 **5.3.1 Primary endpoint**

2697 Wound irrigation with PHX solution will be tested for superiority over no irrigation (Test
2698 1) and irrigation with saline (Test 2) with respect to the incidence of SSI within 30 days
2699 of surgery using two Fine and Gray sub-distributional hazard models with SSI as main
2700 event and re-laparotomy and death as competing risks. The model actually compares
2701 time to event, although the timing of SSI is of less interest than the occurrence of SSI
2702 within 30 days of surgery.

2703 Since randomization is stratified by study center and level of contamination, the models
2704 will include covariates treatment group, study center, and level of contamination. The
2705 global significance level is set to 5%. Using the Bonferroni-Holm adjustment, the local
2706 significance level will be 2.5% and 5% in the order of increasing p-value.

2707 In case there are small study centers which would not allow the model to converge,
2708 center will not be used as covariate.

2709 **5.3.2 Supportive analysis of the primary endpoint**

2710 The frequency of SSI will be presented graphically by treatment and study center using
2711 a clustered bar chart.

2712 In case there are differences between the treatment groups in terms of baseline
2713 characteristics, those will also be included as covariates in the models. Type and
2714 duration of operation, use of wound-edge protectors, intraoperative changing of gloves
2715 and patient related risk factors (NNSI risk score, BMI, age, diabetes) might influence the
2716 outcome, which is why they will also be included as model covariates.

2717 Descriptive statistics for the primary endpoint will be presented per treatment group:
2718 absolute and relative frequencies of

- 2719 • SSI (overall and by class: superficial incisional, deep incisional, and organ/space)
- 2720 • Re-laparotomy (will be identified during medical review based on the AE records)
- 2721 • Death
- 2722 • Lost to follow-up
- 2723 • Completed study without event

2724 Those incidences will be compared between treatment groups using Fisher's exact test
2725 in order to better understand the distribution of missing values.

2726 **5.3.3 Secondary endpoint analysis**

2727 Secondary endpoints will be analysed by study group on the ITT set using appropriate
2728 descriptive statistics. Any explorative statistical testing will be performed two-sided
2729 using a significance level of 5%.

2730 **5.3.3.1 Non-infectious wound complications**

2731 Non-infectious wound complications (seroma, hematoma, delayed healing, necrosis)
2732 within 30 days postoperatively will be summarized by treatment group using absolute
2733 and relative frequencies. The two types of irrigation will be compared to no irrigation
2734 using the χ^2 -test or the Fisher exact test, as appropriate.

2735 **5.3.3.2 Hospital stay**

2736 Duration of hospital stay – overall and post-surgery. The later will be calculated by
2737 subtracting the duration of preoperative hospital stay from the (overall) duration of
2738 hospital stay, recorded at study end (visit 8). All durations will be measured in days.
2739 Number of valid cases, mean, standard deviation, median, minimum, and maximum of
2740 post-surgical hospital stay will be displayed per treatment group. The two types of
2741 irrigation will be compared to no irrigation using the independent t-test or the Mann-
2742 Whitney-U test, as appropriate.

2743 **5.3.3.3 Thirty-day mortality**

2744 Mortality within 30 days postoperatively will be summarized by treatment group using
2745 absolute and relative frequencies. The two types of irrigation will be compared to no
2746 irrigation using the χ^2 -test or the Fisher exact test, as appropriate.

2747 **5.3.3.4 Re-operation rate**

2748 Rate of reoperation within 30 days post-surgery will be identified during medical review
2749 based on the AE records and will be summarized by treatment group using absolute
2750 and relative frequencies. The two types of irrigation will be compared to no irrigation
2751 using the χ^2 -test or the Fisher exact test, as appropriate.

2752 **5.3.3.5 Subgroup analysis**

2753 The primary and secondary endpoints will be additionally summarized within the
2754 following subgroups:

- 2755 • Type of SSI (superficial, deep, organ space)
- 2756 • NNSI - surgical infection risk index (0, 1, 2, 3)
- 2757 • ASA score (1, 2, 3)
- 2758 • BMI (<18.5, 18.5≤BMI<25, 25≤BMI<30, ≥30)
- 2759 • Age (18≤age<40, 40≤age<65, 65≤age<85, ≥85)
- 2760 • Diabetes (Type I, Type II - insulin, Type II - oral antidiabetics, Type II - dietary)
- 2761 • Smoker (current, former, no)
- 2762 • Alcohol consumption (current, former, no)
- 2763 • History of SSI (multiple, single, no)
- 2764 • History of radiotherapy (multiple, single, no)
- 2765 • History of chemotherapy (multiple, single, no)
- 2766 • Preoperative hospital stay >2d (y/n)
- 2767 • Antibiotic prophylaxis (yes >1h prior OP, yes ≤1h prior OP, no)

- 2768 • Type of surgery (intestinal, hepato-biliary, other):
 - 2769 1 = Colo-rectal= colon, rectum, appendix
 - 2770 2 = Hepato-biliary= pancreas, bile duct, hepatic
 - 2771 3 = Oesophago-gastric = esophageal and gastric
 - 2772 4 = Other
- 2773 • Duration of surgery taken from NNIS: Length of OP >T hours (y/n)
- 2774 • Intraoperative use of wound-edge protectors (y/n)
- 2775 • Intraoperative changing of gloves (y/n)
- 2776 • Presence of an enterostomy (y/n)
- 2777 • Level of contamination (Class II, III, IV)

2778 Absolute and relative frequencies will be presented for categorical variables. Number of
 2779 valid cases, mean, standard deviation, median, minimum, and maximum will be
 2780 displayed for continuous variables. Tests will only be performed in case clinically
 2781 meaningful differences are observed between treatment groups.

2782 **5.3.3.6 Treatment costs**

2783 Analysis of treatment costs will be done indirectly through the between-group
 2784 comparisons of hospital stay and surgical complications. This analysis is already
 2785 described in sections 4.3.3.1, 4.3.3.2, 4.3.3.4, 4.3.1 and 4.3.2.

2786 **5.3.3.7 Adverse events**

2787 All AEs other than SSI will be analysed on the safety set. The overall incidence of non-
 2788 SAE-adverse events, SAEs, and related SAEs will be tabulated using MedDRA System
 2789 Organ Class and Preferred Term by treatment group (see table shells). The overall
 2790 occurrences as well as the number of affected patients are of interest. SAEs and related
 2791 SAEs which resulted in death will also be tabulated.

2792 **5.4 Missing data**

2793 Missing primary endpoint data in the primary analysis will be dealt with using competing
 2794 risks and censoring. Missing SSI evaluation due to death or re-laparotomy will be
 2795 considered a competing risk. Missing SSI for all other reasons will be censored. Data
 2796 will not be imputed for other analyses such as secondary or subgroup analyses.

2797 **5.5 Additional analyses**

2798 The following parameters will be summarized by treatment group using absolute and
 2799 relative frequencies: use of concomitant medications of special interest (antibiotics,
 2800 immunosuppression, anticoagulants), SSI (no/yes-new/yes-ongoing), Type of SSI
 2801 (including their Clavien-Dindo classification), non-SSI wound complications (seroma,
 2802 hematoma, delayed healing, necrosis, other wound intervention (none, VAC, bedside
 2803 wound revision, re-operation, other), abnormal lab values.

2804 The vital parameters body temperature, systolic blood pressure, diastolic blood
 2805 pressure, and pulse will be summarized by treatment group and visit using mean and
 2806 SD.

2807 **5.6 Statistical software**

2808 Analysis will be performed with SAS version 9.4.

2809 **6 TABLE SHELLS**

2810

2811 SAE, related-SAE, SAE resulting in death, related-SAE resulting in death, and non-
2812 SAE-AEs (5 tables)

System Class	Organ Class	Exposed to Treatment 1 N=			Exposed to Treatment 2 N=			Exposed to Treatment 3 N=			
		Preferred Term	Events	Subjects affected		Events	Subjects affected		Events	Subjects affected	
				n	%*		n	%*		n	%*
		OVERALL	x	x	(x)	x	x	(x)	x	x	(x)
		SOC1	x	x	(x)	x	x	(x)	x	x	(x)
		PT1	x	x	(x)	x	x	(x)	x	x	(x)
		PT2	x	x	(x)	x	x	(x)	x	x	(x)
		PT3	x	x	(x)	x	x	(x)	x	x	(x)
		SOC2	x	x	(x)	x	x	(x)	x	x	(x)
		PT4	x	x	(x)	x	x	(x)	x	x	(x)
		PT5	x	x	(x)	x	x	(x)	x	x	(x)
		...									

2813 * with respect to the number of exposed subjects

2814

2815 Subjects enrolled per age group

Age group	Treatment 1		Treatment 2		Treatment 3		Total
	n	%	n	%	n	%	n
Total	x	(x)	x	(x)	x	(x)	x
In utero	x	(x)	x	(x)	x	(x)	x
Preterm newborn - gestational age < 37 wk	x	(x)	x	(x)	x	(x)	x
Newborns (0-27 days)	x	(x)	x	(x)	x	(x)	x
Infants and toddlers (28 days-23 months)	x	(x)	x	(x)	x	(x)	x
Children (2-11 years)	x	(x)	x	(x)	x	(x)	x
Adolescents (12-17 years)	x	(x)	x	(x)	x	(x)	x
Adults (18-64 years)	x	(x)	x	(x)	x	(x)	x
From 65 to 84 years	x	(x)	x	(x)	x	(x)	x
85 years and over	x	(x)	x	(x)	x	(x)	x

2816 Include only existing categories for the current study in the summary table.

2817

2818

2819 **2.2 SAP Final Version 1.1 (08.11.2022)**

2820			
2821		Table of Contents	
2822			
2823	1	INTRODUCTION	98
2824	1.1	Background and rationale	98
2825	1.2	Study objectives.....	98
2826	1.3	Study endpoints	98
2827	2	STUDY METHODS	99
2828	2.1	Trial design	99
2829	2.2	Randomization	99
2830	2.3	Sample size	99
2831	2.4	Framework.....	100
2832	2.5	Statistical interim analyses and stopping guidance	100
2833	2.6	Timing of final analysis.....	100
2834	2.7	Timing of outcome assessments.....	100
2835	3	STATISTICAL PRINCIPLES	101
2836	3.1	Confidence intervals and P values	101
2837	3.2	General calculation rules.....	101
2838	3.3	Adherence and protocol deviations	101
2839	3.4	Analysis populations	101
2840	3.5	Event and censor times for the Kaplan-Meier analyses.....	101
2841	4	ANALYSIS.....	102
2842	4.1	Trial population.....	102
2843	4.1.1	Screening data.....	102
2844	4.1.2	Eligibility	102
2845	4.1.3	Recruitment.....	102
2846	4.1.4	Withdrawal/follow-up.....	102
2847	4.1.5	Baseline patient characteristics.....	103
2848	4.2	Outcome definitions.....	103
2849	4.3	Analysis methods for the primary and secondary endpoints	104
2850	4.3.1	Primary endpoint	104
2851	4.3.2	Supportive analysis of the primary endpoint.....	104
2852	4.3.3	Secondary endpoint analysis.....	104
2853	4.3.3.1	Non-infectious wound complications	105
2854	4.3.3.2	Hospital stay	105
2855	4.3.3.3	Thirty-day mortality	105
2856	4.3.3.4	Re-operation rate.....	105
2857	4.3.3.5	Subgroup analysis	105
2858	4.3.3.6	Treatment costs	106
2859	4.3.3.7	Adverse events	106

2860	4.4	Missing data.....	106
2861	4.5	Additional analyses.....	106
2862	4.6	Statistical software.....	106
2863	5	TABLE SHELLS.....	107
2864			
2865			

2866 **7 INTRODUCTION**

2867 **7.1 Background and rationale**

2868 Surgical site infection (SSI) is one of the most common complications following
2869 abdominal visceral surgery and dramatically increases length of hospital stay and costs.
2870 Hypothetically, intraoperative wound irrigation (IOWI) before skin closure with saline or
2871 antiseptics might be a potential pragmatic option to reduce SSI rates. Currently, there
2872 are no official recommendations on its use and clinical practice varies largely. Solutions
2873 containing the antiseptic agent polyhexanide (PHX) are approved for IOWI, and were
2874 shown to promote wound healing, but have not been evaluated in randomized clinical
2875 trials (RCTs) in abdominal visceral surgery. Therefore, we designed a multicenter,
2876 randomized, observer-blinded clinical trial evaluating the efficacy of IOWI with PHX
2877 solution or saline before skin closure after laparotomy. The primary endpoint is the
2878 incidence of SSI 30 days postoperatively, according to the Center of Disease Control
2879 (CDC) definition. The results of the trial will provide evidence for definite clinical
2880 recommendations regarding the use of IOWI and influence current guidelines and
2881 provide all participating patients the opportunity of an improved treatment.

2882 **7.2 Study objectives**

2883 To investigate whether the use of intraoperative, epifascial wound irrigation with PHX
2884 solution can reduce surgical site infections after laparotomy for visceral surgery
2885 compared to saline irrigation or no irrigation.

2886 **7.3 Study endpoints**

2887 Primary efficacy endpoint:

2888 SSI according to CDC criteria within 30 days postoperatively

2889 Secondary endpoints:

- 2890 • Non-infectious wound complications (e.g. seroma, hematoma, delayed
2891 healing) within 30 days postoperatively
- 2892 • Duration of hospital stay
- 2893 • Mortality and morbidity within 30 days postoperatively
- 2894 • Incidence of reoperation within 30 days postoperatively
- 2895 • Incidence of AE/SAE within 30 days postoperatively
 - 2896 ○ Surgical complications will be additionally evaluated according to the
2897 Clavien-Dindo classification.

2898 Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space),
2899 NNIS risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history
2900 of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration
2901 and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of
2902 wound-edge protectors and changing of gloves, presence of an enterostomy.

2903 **8 STUDY METHODS**

2904 **8.1 Trial design**

2905 This clinical trial is a prospective, randomized, controlled, observer and patient-blinded,
2906 multicenter, surgical trial according to German drug law (AMG) phase III-b, with three
2907 parallel comparison groups.

2908 Patients are randomised to one of the following treatment arms:

2909 **Arm 1 (Intervention 1):**

2910 Irrigation of the subcutaneous tissue after closure of the abdominal fascia with
2911 1000ml PHX solution (0.04%)

2912 **Arm 2 (Intervention 2):**

2913 Irrigation of the subcutaneous tissue after closure of the abdominal fascia with
2914 1000ml saline solution (NaCl 0.9%)

2915 **Arm 3 (Control):**

2916 No epifascial wound irrigation

2917 A total of 680 patients (290 patients in arm 1, 290 patients in arm 2, and 100 patients in
2918 arm 3) in up to 15 centres will be enrolled in the study with duration of 34 days per
2919 patient (up to three days prior to surgery, day of surgery, and 30 days post-surgery).

2920 A total of 8 visits are scheduled during the study period.

2921 **8.2 Randomization**

2922 Patients are randomised blockwise ca. 3:3:1 to the treatment arms with stratification by
2923 centre and level of contamination of the surgical procedure (clean-contaminated,
2924 contaminated, or dirty) during surgery after closure of the abdominal fascia using
2925 RANDOBASE, the online-randomization tool at MSZ. RANDOBASE uses pre-defined
2926 randomization lists, which are created at IMedIS using Rancode Professional 2015. Two
2927 sets of sealed envelopes were produced: one for emergency unblinding at site, one for
2928 the same purpose at the MSZ-safety management department.

2929 **8.3 Sample size**

2930 Justification and calculation of the sample size can be found in the study protocol
2931 section 23.1.

2932 Sample size adjustment (CSP approved Version 3 Amendment 2 02.03.2021):

2933 Due to the unexpected high number of dropouts, the sample size was adjusted based
2934 on the changed analysis of SSI (see section 5.2.1). The sample size was calculated
2935 (Sample Size Software, Sample Size Tables, D. Machin et al., 2009) based on the
2936 primary endpoints of the study, assuming SSI rates (event of interest) of 2.2% in the
2937 PHX group (assuming a 75% risk reduction according to the trial by Roth et al. (14)),
2938 8.7% in the saline group (according to the results of the trial by Cervantes-Sanchez et
2939 al. (13)), and 16.2% in the control group according to results of the previously
2940 conducted meta-analysis (12). The incidence rate of SSI over all study arms is then
2941 expected to be 7%, given the approximate 3:3:1 group assignment. The actual SSI rate
2942 up to now is 7.2%, which is very close to our assumption and we consider it valid. The

2943 incidence rate for the competing risks of death or re-laparotomy is estimated to be a
2944 total of 13.4% in all arms. This estimation is done based on the data collected up to
2945 now.

2946 The global significance level was set to 5% (two-sided tests). Since the PHX arm will be
2947 used twice for a comparison, the Bonferroni-Holm procedure was used to set the local
2948 alpha level for test 1 (PHX vs. no intervention) to 2.5% and for test 2 (PHX vs. saline
2949 irrigation) to 5%. If 290 patients are recruited in the PHX arm, 290 patients in the saline
2950 arm and 100 patients in the control arm (a total of 680 patients), the two Fine and Gray
2951 sub-distributional hazard models will have a power of 80% each to detect differences
2952 between the treatments.

2953 **8.4 Framework**

2954 This study tests for superiority of (1) PHX over no intervention and (2) PHX over saline
2955 irrigation with respect to SSI rate within 30 days postoperatively.

2956 **8.5 Statistical interim analyses and stopping guidance**

2957 No interim analyses are planned for this study.

2958 **8.6 Timing of final analysis**

2959 The final analysis will be performed collectively at the end of the study.

2960 **8.7 Timing of outcome assessments**

2961 **Baseline** data will be collected at visit 1 and 2, which means up to day 0.

2962 **Day 0** is defined as the time of closure of the abdominal fascia.

2963 All other day definitions are relative to day 0.

2964 The primary outcome will be accessed up to visit 8, which means on day 30 and up to
2965 day 36 when the time window is considered.

Visit	Day	Time window
1	-3 to -1	Up to day 0
2	0 (surgery)	
3	2	
4	4	
5	6	
6	8	
7	10	Up to day 14
8	30	Up to day 36

2966

2967 **9 STATISTICAL PRINCIPLES**

2968 **9.1 Confidence intervals and P values**

2969 All statistical tests will be performed two-sided at the global significance level of 5%.
2970 Adjustment for multiplicity will be done for the primary endpoint only, where the
2971 Bonferroni-Holm adjustment method will be used (see also section 5.2.1).

2972 Confidence intervals will be two-sided and 95%.

2973 **9.2 General calculation rules**

2974 4. Percentages will always be quoted using number of 'known' values in the
2975 denominator unless otherwise stated.

2976 5. P-values will be quoted to three decimal places only. Confidence intervals will
2977 also be quoted to three decimal places. However, if the statistical software SAS,
2978 which is used for analysis, prints four decimal places, values will not be rounded
2979 again, but printed to four decimal places.

2980 6. Chi-square tests in contingency tables will be replaced by Fisher's exact tests if
2981 any expected cell frequency is less than five.

2982 **9.3 Adherence and protocol deviations**

2983 The number and percent of patients per treatment group who received IOWI will be
2984 reported.

2985 Major protocol deviations will be listed per patient.

2986 **9.4 Analysis populations**

2987 The **Intention-to-Treat (ITT) population** will contain all randomised patients with
2988 results attributed to the treatment group they were randomised to.

2989 The **safety analysis (SA) population** will consist of all randomised subjects with
2990 results attributed to the treatment group of their actual treatment.

2991 Safety analysis will be performed on the SA population. All other analyses will be
2992 performed in the ITT population.

2993 **9.5 Event and censor times for the Kaplan-Meier analyses**

2994 The following definitions will apply to the time-to-event analysis of SSI:

Parameter:	SSI
Main Event Time	time of SSI
Competing Event Times	time of death time of re-laparotomy
Censor Time	time of last follow-up*

2995 * for patients without SSI

2996

2997 **10 ANALYSIS**

2998 **10.1 Trial population**

2999 **10.1.1 Screening data**

3000 The overall number of screened patients will be presented in the report.

3001 The reasons for non-eligibility will also be summarized.

3002 **10.1.2 Eligibility**

3003 **Key inclusion criteria:**

- 3004 • Clean-contaminated, contaminated or dirty (according to CDC classification)
- 3005 • Abdominal surgery by midline or transverse laparotomy (elective or emergency)
- 3006 • Age ≥ 18 years
- 3007 • American Society of Anesthesiologists (ASA) score ≤ 3

3008 **Key exclusion criteria:**

- 3009 • Pregnancy or breast feeding
- 3010 • Known hypersensitivity/allergy to PHX
- 3011 • Inability to understand/give informed consent
- 3012 • Inability to attend follow-up visits
- 3013 • Revision-surgery (previous abdominal surgery within the last 30 days)
- 3014 • Planned re-laparotomy within 30 days
- 3015 • Severe immunosuppression
- 3016 • Concurrent abdominal wall infections
- 3017 • Pre-operative antibiotic therapy (within 5 days prior to surgery)

3018 The full set of inclusion and exclusion criteria can be found in the study protocol
3019 sections 12.1 and 12.2.

3020 **10.1.3 Recruitment**

3021 Tables will contain the following absolute and relative frequencies per treatment group
3022 and overall:

- 3023 • patients who entered the study.
- 3024 • patients within each analysis set including reasons for exclusion. Patients in the
3025 SA will be considered in the group of their actual treatment.
- 3026 • patients per centre on the ITT set.

3027 **10.1.4 Withdrawal/follow-up**

3028 Since study treatment is given only once at the beginning of the study, withdrawal from
3029 study treatment is not of interest.

3030 The number of patients per treatment group who did not complete the study will be
3031 given in a summary table by reason (multiple reasons possible) including absolute and
3032 relative frequencies.

3033 **10.1.5 Baseline patient characteristics**

3034 The following characteristics will be summarized per treatment group on the ITT set:

3035 Demographics: sex, age, BMI.

3036 Medical history: main diagnosis leading to operation (malign/benign), ASA classification,
3037 diabetes (including type and treatment), allergies, comorbidities (11 pre-defined and
3038 other; multiple comorbidities are possible), previous abdominal surgery (no, single,
3039 multiple), time since last abdominal surgery, history of SSI (no, single, multiple), time
3040 since last SSI, location of last SSI, history of radiotherapy (no, single, multiple), time
3041 since end of last radiotherapy, dose (Gy), history of chemotherapy (no, single, multiple),
3042 smoking (no, previously, currently), packyears, regular alcohol consumption (no,
3043 previously, currently), glasses/week.

3044 Surgery: duration of preoperative hospital stay (days), urgency and type of procedure,
3045 duration of surgery (min), antibiotic prophylaxis (no, yes >1h prior OP, yes ≤1h prior
3046 OP), type of skin disinfectant, type of incision, length of incision, intra-OP change of
3047 gloves (y/n), intra-OP use of wound edge protectors (y/n), enterostomy created (y/n),
3048 type of abdominal fascia closure, use of mesh (no, yes-sublay, yes-onlay), level of
3049 contamination (class I to IV), NNIS risk score, wound closure (complete/incomplete),
3050 subcutaneous sutures used (y/n), skin closure type (stapler, continuous suture, single
3051 suture).

3052 Absolute and relative frequencies will be presented for categorical variables. Number of
3053 valid cases, mean, standard deviation, median, minimum, and maximum will be
3054 displayed for continuous variables.

3055

3056 **10.2 Outcome definitions**

3057 The primary endpoint is the frequency of physician-assessed SSI up to 30 days post-
3058 surgery. According to the time-definitions, the SSI assessment may take place up to day
3059 36. Surgical site infections will additionally be classified in three groups: superficial
3060 incisional, deep incisional, and organ/space.

3061 The secondary endpoints of this study are:

3062 Frequency of non-infectious wound complications (e.g. seroma, hematoma, delayed
3063 healing) within 30 days postoperatively;

3064 Duration of hospital stay;

3065 Mortality within 30 days postoperatively;

3066 Rate of reoperation within 30 days post-surgery, which will be identified during medical
3067 review based on the AE records.

3068 Pre-specified subgroup analysis by category of SSI (superficial, deep, organ space),
3069 NNIS risk score, ASA score, BMI, age, diabetes, smoking, alcohol consumption, history
3070 of SSI, history of radio-/chemotherapy, pre-operative hospital stay >2d, administration
3071 and timing of antibiotic prophylaxis, type and duration of surgery, intraoperative use of
3072 wound-edge protectors and changing of gloves, presence of an enterostomy.

3073 Assessment of safety:

3074 The assessment of safety will be based on the frequency of AE/SAE other than SSI
3075 within the safety population (according to CTCAE V. 4.3). Surgical complications will be
3076 additionally evaluated according to the Clavien-Dindo classification.

3077

3078 **10.3 Analysis methods for the primary and secondary endpoints**

3079 **10.3.1 Primary endpoint**

3080 Wound irrigation with PHX solution will be tested for superiority over no irrigation (Test
3081 1) and irrigation with saline (Test 2) with respect to the incidence of SSI within 30 days
3082 of surgery using two Fine and Gray sub-distributional hazard models with SSI as main
3083 event and re-laparotomy and death as competing risks. The model actually compares
3084 time to event, although the timing of SSI is of less interest than the occurrence of SSI
3085 within 30 days of surgery.

3086 Since randomization is stratified by study center and level of contamination, the models
3087 were planned to include covariates study center and level of contamination in addition to
3088 treatment group. After reviewing the blinded data, it was decided to combine
3089 contamination classes I with II and III with IV. Classes I and IV contain two patients
3090 each. Since there are small study centers which would not allow the model to converge,
3091 center will not be used as covariate (Langen N=5, Düsseldorf N=6).

3092 The global significance level is set to 5%. Using the Bonferroni-Holm adjustment, the
3093 local significance level will be 2.5% and 5% in the order of increasing p-value.

3094 **10.3.2 Supportive analysis of the primary endpoint**

3095 In case there are differences between the treatment groups in terms of baseline
3096 characteristics, those will also be included as covariates in the models.

3097 Type and duration of operation, use of wound-edge protectors, intraoperative changing
3098 of gloves and patient related risk factors (NNIS risk score, BMI, age, diabetes) might
3099 influence the outcome, which is why they will also be included as model covariates.

3100 Descriptive statistics for the primary endpoint will be presented per treatment group:
3101 absolute and relative frequencies of

- 3102 • SSI (overall and by class: superficial incisional, deep incisional, and organ/space)
- 3103 • Re-laparotomy (will be identified during medical review based on the AE records)
- 3104 • Death
- 3105 • Lost to follow-up
- 3106 • Completed study without event

3107 Those incidences will be compared between treatment groups using Fisher's exact test
3108 in order to better understand the distribution of missing values.

3109 **10.3.3 Secondary endpoint analysis**

3110 Secondary endpoints will be analysed by study group on the ITT set using appropriate
3111 descriptive statistics. Any explorative statistical testing will be performed two-sided
3112 using a significance level of 5%.

3113 **10.3.3.1 Non-infectious wound complications**

3114 If wound complications are recorded on the same day as an SSI or an ongoing SSI,
3115 then they will not be included in the analysis, as they are not non-infectious (and are
3116 therefore already accounted for in the SSI analysis).

3117 Non-infectious wound complications (seroma, hematoma, delayed healing, necrosis)
3118 within 30 days postoperatively will be summarized by treatment group using absolute
3119 and relative frequencies. The two types of irrigation will be compared to no irrigation
3120 using the Fisher exact test.

3121 **10.3.3.2 Hospital stay**

3122 Duration of hospital stay – overall and post-surgery. The later will be calculated by
3123 subtracting the duration of preoperative hospital stay from the (overall) duration of
3124 hospital stay, recorded at study end (visit 8). All durations will be measured in days.
3125 Number of valid cases, mean, standard deviation, median, minimum, and maximum of
3126 post-surgical hospital stay will be displayed per treatment group. The two types of
3127 irrigation will be compared to no irrigation using the Mann-Whitney-U test.

3128 **10.3.3.3 Thirty-day mortality**

3129 Mortality within 30 days postoperatively will be summarized by treatment group using
3130 absolute and relative frequencies. The two types of irrigation will be compared to no
3131 irrigation using the Fisher exact test.

3132 **10.3.3.4 Re-operation rate**

3133 Rate of reoperation within 30 days post-surgery will be identified during medical review
3134 based on the AE records and will be summarized by treatment group using absolute
3135 and relative frequencies. The two types of irrigation will be compared to no irrigation
3136 using the χ^2 -test.

3137 **10.3.3.5 Subgroup analysis**

3138 The primary and secondary endpoints will be additionally summarized within the
3139 following subgroups:

- 3140 • Type of SSI (superficial, deep, organ space)
- 3141 • NNIS - surgical infection risk index (0, 1, 2, 3)
- 3142 • ASA score (1, 2, 3)
- 3143 • BMI (<18.5 , $18.5 \leq \text{BMI} < 25$, $25 \leq \text{BMI} < 30$, ≥ 30)
- 3144 • Age ($18 \leq \text{age} < 40$, $40 \leq \text{age} < 65$, $65 \leq \text{age} < 85$, ≥ 85)
- 3145 • Diabetes (Type I, Type II - insulin, Type II - oral antidiabetics, Type II - dietary)
- 3146 • Smoker (current, former, no)
- 3147 • Alcohol consumption (current, former, no)
- 3148 • History of SSI (multiple, single, no)
- 3149 • History of radiotherapy (multiple, single, no)
- 3150 • History of chemotherapy (multiple, single, no)

- 3151 • Preoperative hospital stay >2d (y/n)
- 3152 • Antibiotic prophylaxis (yes >1h prior OP, yes ≤1h prior OP, no)
- 3153 • Type of surgery (intestinal, hepato-biliary, other):
 - 3154 1 = Colo-rectal= colon, rectum, appendix
 - 3155 2 = Hepato-biliary= pancreas, bile duct, hepatic
 - 3156 3 = Oesophago-gastric = esophageal and gastric
 - 3157 4 = Other
- 3158 • Duration of surgery taken from NNIS: Length of OP >T hours (y/n)
- 3159 • Intraoperative use of wound-edge protectors (y/n)
- 3160 • Intraoperative changing of gloves (y/n)
- 3161 • Presence of an enterostomy (y/n)
- 3162 • Level of contamination (Class II, III, IV)

3163 Absolute and relative frequencies will be presented for categorical variables. Number of
 3164 valid cases, mean, standard deviation, median, minimum, and maximum will be
 3165 displayed for continuous variables. Tests will only be performed in case clinically
 3166 meaningful differences are observed between treatment groups.

3167 **10.3.3.6 Treatment costs**

3168 Analysis of treatment costs will be done indirectly through the between-group
 3169 comparisons of hospital stay and surgical complications. This analysis is already
 3170 described in sections 4.3.3.1, 4.3.3.2, 4.3.3.4, 4.3.1 and 4.3.2.

3171 **10.3.3.7 Adverse events**

3172 All AEs other than SSI will be analysed on the safety set. The overall incidence of non-
 3173 SAE-adverse events, SAEs, and related SAEs will be tabulated using MedDRA System
 3174 Organ Class and Preferred Term by treatment group (see table shells). The overall
 3175 occurrences as well as the number of affected patients are of interest. SAEs and related
 3176 SAEs which resulted in death will also be tabulated.

3177 **10.4 Missing data**

3178 Missing primary endpoint data in the primary analysis will be dealt with using competing
 3179 risks and censoring. Missing SSI evaluation due to death or re-laparotomy will be
 3180 considered a competing risk. Missing SSI for all other reasons will be censored. Data
 3181 will not be imputed for other analyses such as secondary or subgroup analyses.

3182 **10.5 Additional analyses**

3183 The following parameters will be summarized by treatment group using absolute and
 3184 relative frequencies: use of concomitant medications of special interest (antibiotics,
 3185 immunosuppression, anticoagulants), SSI (no/yes-new/yes-ongoing), Type of SSI
 3186 (including their Clavien-Dindo classification), non-SSI wound complications (seroma,
 3187 hematoma, delayed healing, necrosis, other wound intervention (none, VAC, bedside
 3188 wound revision, re-operation, other), abnormal lab values.

3189 **10.6 Statistical software**

3190 Analysis will be performed with SAS version 9.4.

3191 **11 TABLE SHELLS**

3192

3193 SAE, related-SAE, SAE resulting in death, related-SAE resulting in death, and non-
 3194 SAE-AEs (5 tables)

System Organ Class Preferred Term	Exposed to Treatment 1 N=			Exposed to Treatment 2 N=			Exposed to Treatment 3 N=		
	Events	Subjects affected		Events	Subjects affected		Events	Subjects affected	
		n	%*		n	%*		n	%*
OVERALL	x	x	(x)	x	x	(x)	x	x	(x)
SOC1	x	x	(x)	x	x	(x)	x	x	(x)
PT1	x	x	(x)	x	x	(x)	x	x	(x)
PT2	x	x	(x)	x	x	(x)	x	x	(x)
PT3	x	x	(x)	x	x	(x)	x	x	(x)
SOC2	x	x	(x)	x	x	(x)	x	x	(x)
PT4	x	x	(x)	x	x	(x)	x	x	(x)
PT5	x	x	(x)	x	x	(x)	x	x	(x)
...									

3195 * with respect to the number of exposed subjects

3196

3197 Subjects enrolled per age group

Age group	Treatment 1		Treatment 2		Treatment 3		Total
	n	%	n	%	n	%	n
Total	x	(x)	x	(x)	x	(x)	x
Adults (18-64 years)	x	(x)	x	(x)	x	(x)	x
From 65 to 84 years	x	(x)	x	(x)	x	(x)	x
85 years and over	x	(x)	x	(x)	x	(x)	x

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3200

3201 **2.3 SAP Summary of Changes**

3202 Due to the large number of missing data that the unexpected high number of drop-outs would have
3203 meant, we adapted the analysis strategy, so the primary endpoint was analysed using the Fine and
3204 Gray sub-distributional hazard models with SSI as main event and relaparotomy and death as
3205 competing risks. The rest of the missing SSI was censored at time of last follow-up. The sample size
3206 was adjusted based on this changed analysis of SSI. The sample size was calculated based on the
3207 primary endpoints of the study, assuming SSI rates (event of interest) of 2.2% in the PHX group
3208 (assuming a 75% risk reduction according to the trial by Roth et al.), 8.7% in the saline group
3209 (according to the results of the trial by Cervantes-Sanchez et al.), and 16.2% in the control group
3210 according to results of the previously conducted meta-analysis. The incidence rate of SSI over all
3211 study arms was then expected to be 7%, given the approximate 3:3:1 group assignment. The
3212 incidence rate for the competing risks of death or re-laparotomy was estimated to be a total of 13.4%
3213 in all arms. This estimation was done based on the data collected up to that timepoint. The global
3214 significance level was set to 5% (two-sided tests). Since the PHX arm was used twice for a
3215 comparison, the Bonferroni-Holm procedure was used to set the local alpha level for test 1 (PHX vs.
3216 no intervention) to 2.5% and for test 2 (PHX vs. saline irrigation) to 5%. If 290 patients were recruited
3217 in the PHX arm, 290 patients in the saline arm and 100 patients in the control arm (a total of 680
3218 patients), the two Fine and Gray sub-distributional hazard models had a power of 80% each to detect
3219 differences between the treatments. These changes were submitted to and approved by the ethics
3220 committee and health authority (BfARM) as a second amendment of the study protocol on the 8th and
3221 24th March 2021 respectively. No other changes were made in the trial conduct in respect to the
3222 original design.

3223