Dissertation

Prevention of Anastomotic Leaks in Colorectal Surgery

submitted by Dr. der Philosophie

GERHARD ERNST STEYER

for the Academic Degree (equivalent to PhD) of Doctor of Medical Science /

Doctor scientiae medicae (Dr.scient.med.)

at the Medical University of Graz, Europe / Medizinische Universität Graz, Europa

Division of General, Visceral and Transplant Surgery

under the Supervision (Doctoral Father) of Prof.Dr.med.univ. Johann PFEIFER

2024

Declaration

I hereby declare that this thesis is my own original work and that I have fully acknowledged by name all of those individuals and organisations that have contributed to the research for this thesis. Due acknowledgement has been made in the text to all other material used. Throughout this thesis and in all related publications I followed the "Guidelines of the Medical University of Graz on Good Scientific Practice".

Graz, 1.4.2024 Gerhard Ernst Steyer *e.h.*

In Liebe und Verehrung

meiner Frau

für den gemeinsamen Weg im letzten halben Jahrhundert

und die Führung unserer immer grösser werdenden Familie

I geh grad voraus - und wart dann a Weil
bis ihr nachkommts - hat aber ka Eil.
Nach einem Totenbrett im bayerischen Wald.

Disclosures

This research received no external funding. This trial was investigator-initiated and investigator-driven and designed for the third doctoral thesis of the author at a University in Graz, Europe, for this time at the Medical University of Graz (Doctoral School of Lifestyle-Related Diseases, Speaker: Sandra Holasek) by *G.E.S.* under the supervision of *Johann Pfeifer* and *Markus Puchinger* to enable patients to make use of the results of this investigation.

This study (Mugthesis 7950) was conducted according to the guidelines of the Declaration of Helsinki and approved by the Ethics Committee of the Medical University of Graz for the protocol and patient consent (decision number 30-183 ex 17/18).

The author declares no conflicts of interest. Without a sponsor and without any financial support the results of this academic study were interpreted independently and without any conflicts of interest.

Parts of this dissertation have been published according to the mandatary rules of the Medical University of Graz in (see also: Appendix A.1, page 75 to this thesis and Permission to reproduce published text and figures Appendix A.2, page 75):

Steyer GE, Puchinger M, Pfeifer J. Successful Clinical Avoidance of Colorectal Anastomotic Leakage through Local Decontamination. Antibiotics (Basel). 2024 Jan 15;13(1):79.

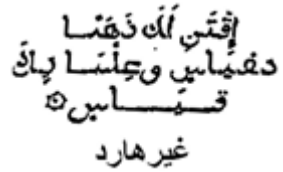
doi: 10.3390/antibiotics13010079. PMID: 38247638; PMCID: PMC10812415.

and the Poster (Abstract):

Steyer GE, Pfeifer J. Colorectal Anastomosis Leakless.

Poster for "Doctoral Day 2023" at the Medical University of Graz, 9.2.2023.

Preface[1]



ERWIRB DIR GOLD SOVIEL DU BRAUCHST
UND WEISHEIT SOVIEL DU KANNST

Fig.1: *Engravings in the mosque at the park of Castle Schwetzingen, Germany, Europe, in Arabic (showing original typos) and in German (correct, no typo), from a photo by the author.*
The quotation means in english language: Acquire gold as much you need / And wisdom as much you can

[1] ***„Opa schau, so geht das, kinderleicht!****“, thus spoke [1] my grandson last year when he was two years old and I answered: „**Herzbinkerl schau, ich bin glücklich, ich habe Enkel.**“ Because he helped me, while he pressed the button “print“ on my very old personal computer - and he really managed to print some literature for this dissertation, my # 3 (see also „Acknowlwdgements“).*

It is not worth, from my point of view, to write a preface for this English Edition, because it is optinal, according to the rules of the Medical University of Graz.- So you are reading a footnote to a non existing preface, but if one is interested in more details, please consult the - until now unpublished - German Deluxe Edition [2] of this dissertation.

When the author started at the University (of course “ex propriis“ and when he gave lectures in different medical topics) half a century ago, the beloved University incarnates: **Solitude and Liberty**. In my opinion, both can be found no more in our days at the University, this University changed - to become a school (creating rules and standard operating procedures, and mandatary rules instead of proposals).

Once upon a time I had - among other topics - a scientific discussion with Johann Pfeifer, because he was wondering how an anastomotic leak could happen. *"Even under the most expert care, a properly constructed intestinal anastomosis can fail to heal resulting in leakage of its contents, peritonitis and sepsis"* [22] and death. The method of surgery was always the same, but in a few cases a re-do surgery was needed.
But why and when, and if it is possible (but how) to avoid those re-do surgery? We also discussed a paper, that was published at that time by the German surgeon Hans Martin Schardey [47] where he described his experiences so far.

Making of: A study concept was born. So we decided to start a literature research. It came out, that surgeons discussed anastomotic leaks since decades, without a solution in sight [4]. Two publications turned the spotlight on microbiota: One paper described the role of *P. aeruginosa* and *E. faecalis* on activating MMP9 (Matrix metallo protease-9) [8], the other one from a German surgeon ventilated the possibility to reduce anastomotic leaks by an antibiotic mixture, supplemented by an antimycotic substance to avoid overgrowth [29].

That was pleasing to the ears of the author and he subsequently got in contact to this surgeon; in a personal discussion [not published and therefore unfortunately not citable] Hans Martin Schardey from Agatharied Academic Teaching Hospital of the Ludwig-Maximilians-University Munich, Germany, Europe, shared his expertise and wrote about his long experiences he made in his hospital to reduce anastomotic leaks by his method. That was great help for our next step: We decided to perform an own independent investigation. I carried out a study protocol and Johann Pfeifer became my doctor father for my third dissertation in Graz (Europe). During the last half century we had allready published together from time to time some articles on different topics, including microbiota, also when I became lecturer on Immunology and the Microbiome at the University of Krems (Austria, Europe) (see citations in [4]).

Tempora mutantur …. Over the time, some rules have been changed. In 1981 the full-scale University of Graz initially tried to prohibit, that the author becomes twice "Doctor of Philosophy" at the end of his **different** studies (scientific and humanistic, see Fig. 1), but then, of course, his dissertations have been approved, and he passed doctoral examinations;

Fig.1: Studienbuch GES, 1981: „**ZUSÄTZLICHE RIGOROSEN OHNE PROMOTION**"

nowadays it was mandatary for the author to show a degree (Doctor of Philosophy) **to start** an education for Philosophy Doctor (PhD-equivalent). *Conclusion:* He prefered to learn (study) through life and to perform dissertations himself as much he wants, instead of awarding a "Golden Doctoral Diploma" (50 years later) or honoris causa. **This is my legacy to coming generations** (valid as well for all universities)**:**

LEARN AS MUCH YOU CAN

Graz, 16.5.2024

Acknowledgements of the author

The author wants to thank *Hans Martin Schardey*, Agatharied Academic Teaching Hospital of the Ludwig-Maximilians-University Munich, Germany, Europe, for his pioneering works in anastomotic leak and for his input regarding his experience on the magistral preparation of the antibiotic mixture investigated in his studies.

The author is greatful to his Doctoral father *Johann Pfeifer* for his permanent extraordinary supervision; before this study was started, we published together from time to time different scientific topics and had an amusing time at the Medical University of Graz, Austria, Europe; together with my son *Gernot Ernst Steyer* (Senior Physician at the Clinical Department of Oral and Maxillofacial Surgery, Medical University of Graz, Austria, Europe) he was editor and author in a Reference Book [3] : “Microbiota”, published 2019, very helpful for the overview of this Dissertation; my son also was Coordinator of this study.- My grandson *Gerald Ernst Steyer* helped me to print some literature for this dissertation (see also preface on page 5 and his collected quotations in reference [1]).

The author thanks *James Elvis Waha,* surgeon at the Division of General, Visceral and Transplant Surgery, Medical University of Graz, Austria, Europe, for the guidance to use MEDOCS at our University and to find patients data.

The head of the Division of General, Visceral and Transplant Surgery, Medical University of Graz, Austria, Europe, *Robert Sucher* enabled the fees for the mandatary publication (Open access) in a Journal.

The statistical supervisor *Markus Puchinger* from the Division of General, Visceral and Transplant Surgery, Medical University of Graz, Austria, Europe, helped with very quick response to questions of the author.

For good compliance the author thanks our patients and the staff of the Surgical Clinic, maybe some patients need not undergo a re-do surgery because of this study.

For the study protocol *Alexander Avian*, Medical Informatics, Statistics and Documentation, at the Medical University of Graz, Austria, Europe, calculated alpha-value for the sample-size, and therefore the author is much obliged to him.

The dissertation was carried out at the Doctoral School of Lifestyle-Related Diseases (Medical University of Graz, Austria, Europe); Speaker *Sandra Holasek* supported this investigation whenever needed.

1. Table of Contents

2. Abbreviations

mGES ….. mural GutEpithelialShelter / muraler GedärmeEpithelSchutz (named after the method proposed by Pfeifer)

& …… et (Latin) / in english: and (ampersand; “and per se and“)

&c ……. et cetera

…… Hash mark, hashing is used across several domains, including medicine : “libra Pondo“

© …… Copyright

MMXXIV….. 2024 in roman numerals

3. Definitions

- The method **mGES** (mural GutEpithelialShelter / muraler GedärmeEpithelSchutz) is named after the method used by Johann Pfeifer, the first successful investigator to prevent Anastomotic Leak in emergency and elective patients by an antibiotic mixture (100 g Polymixin B, 80 g Gentamicin and 125 g Vancomycin) and without the addition of an antimycotic substance; it consists of a short course of intake of an antibiotic mixture (every six hour for five consecutive days after colorectal surgery).

- **Anastomotic Leak** is a clinically obvious defect of the intestinal wall integrity at the colorectal anastomosis (including suture) that leads to a communication between the intra- and extraluminal compartments, requiring re-do surgery within 7 postoperative days. (See also chapter 3 and [4]).

4. List of Figures

5. List of Tables

6. Zusammenfassung Vermeidung kolorektaler Anastomoseninsuffizienz (durch mGES / muraler GedärmeEpithelSchutz, benannt nach der Methode von Pfeifer, bei Notfall- und elektiven Patienten)

Forschungshintergrund: Eine Dickdarmoperation ist ein häufiger chirurgischer Eingriff, jedoch kann es nach der Operation zu einer Insuffizienz der Dickdarmanastomose kommen, einer unvorhersehbaren Komplikation, die möglicherweise eine erneute Operation erforderlich macht. Studien haben gezeigt, dass potenziell pathogene Bakterien (akzidentelle Pathogene) wie Pseudomonas, Enterococcus et cetera das Auftreten einer Anastomoseninsuffizienz im Dickdarm fördern können, indem sie Kollagen abbauen und die Gewebematrix-Metalloproteinase-9 aktivieren. Daher ist die mikrobielle Bakterienbesiedlung im Darm der Schlüssel zur Verhinderung einer Anastomoseninsuffizienz nach einer Dickdarmoperation. **Forschungsinhalte:** Ziel dieser Studie war es zu untersuchen, ob eine selektive Dekontamination mit einer neuartigen Mischung lokal wirkender Antibiotika, die speziell gegen Pseudomonas und Enterokokken wirkt, das Auftreten früher symptomatischer Leckagen reduzieren oder sogar stoppen kann. Bei den Studienteilnehmern handelte es sich um stationäre Patienten, die sich an unserer Universitätsklinik einer Dickdarmoperation unterzogen hatten; diese wurden in zwei Gruppen eingeteilt. Die Interventionsgruppe erhielt 5 Tage lang alle 6 Stunden 100 mg Polymyxin B, 80 mg Gentamicin und 125 mg Vancomycin, während die Kontrollgruppe keine solche Intervention erhielt. Die Ergebnisse zeigten, dass insgesamt 301 Patienten analysiert wurden, davon 152 in der Kontrollgruppe, 11 eine Dickdarmanastomoseninsuffizienz hatten und 149 in der Interventionsgruppe waren bei denen keine Dickdarmanastomoseninsuffizienz auftrat. Der Unterschied zwischen den beiden Gruppen war statistisch hoch signifikant. **Bedeutung:** Die in dieser Studie gegen potentiell pathogene Bakterien verwendete Antibiotikamischung (Polymyxin B, Gentamicin und Vancomycin) verhinderte das Auftreten einer Anastomoseninsuffizienz im Dickdarm vollständig. Basierend auf der Definition einer Dickdarmanastomoseninsuffizienz ist nach lokaler perioperativer Dekontamination kein weiterer chirurgischer Eingriff erforderlich. Die Innovation dieser Studie besteht darin, dass durch den Einsatz einer erstmals in einer Studie verwendeten Antibiotikamischung das Auftreten einer Anastomoseninsuffizienz im Dickdarm erfolgreich verhindert werden konnte, was eine neue Methode für die Prävention von Komplikationen nach einer Dickdarmoperation darstellt.

7. Summary (see also Appendix A.3)

Prevention of Anastomotic Leaks in Colorectal Surgery (through mGES / mural GutEpithelialShelter, named after the method of Pfeifer for emergency and elective patients)

Research background:

Colon surgery is a common surgical procedure, but anastomotic leakage of the colon can occur after surgery, an unpredictable complication that may require reoperation. Studies have shown that potential pathogenic bacteria such as Pseudomonas, Enterococcus &c can promote the occurrence of colonic anastomotic leakage by degrading collagen and activating tissue matrix metalloproteinase-9. Therefore, the microbial community is key to preventing anastomotic leakage after colon surgery.

Research content:

The aim of this study was to investigate whether selective decontamination using a novel mixture of locally acting antibiotics, specifically targeting Pseudomonas and Enterococci, could reduce or even stop the occurrence of early symptomatic leaks. The study subjects were inpatients who underwent colon surgery at our university hospital and were divided into two groups. The intervention group received 100 mg polymyxin B, 80 mg gentamicin, and 125 mg vancomycin every 6 hours for 5 days, while in the control group no such intervention was accepted. The results showed that from a total of 301 analyzed patients, 11 had colonic anastomotic leakage from 152 in the control group, but 149 from the intervention group, had no colonic anastomotic leakage at all. The difference between the two groups was highly significant.

Significance:

The antibiotic mixture (polymyxin B, gentamicin, and vancomycin) used in this study for local decontamination completely prevented the occurrence of colonic anastomotic leakage. Based on the definition of colonic anastomotic leakage, no further surgery is required after local perioperative decontamination. The innovation of this study is that by using a new antibiotic mixture, the occurrence of colonic anastomotic leakage was successfully prevented, providing a new idea for the prevention of complications after colon surgery.

8. Introduction

For successful treatment of colorectal diseases like cancer, inflammatory bowel disease, or diverticulitis a healty bowel connection (anastomosis) after surgery is required for successful treatment [5], but "*up to 20% of surgeries from anastomotic leak resulting in significant patient morbidity and mortality plus an increased length of hospital stay and economic burden*" [5].

Anastomotic leak is an unpredictable postoperative complication during recovery period from colorectal surgery [4]. It may require re-operation. To date, interdisciplinary teams have found that the *"microbiome is the key to preventing anastomotic leak in colorectal surgery, representing a critical missing piece in this puzzle – it modulates the innate immune respond to anastomotic wound healing"* [6, 7]. For decades, surgeons have tried to find out the reasons that cause anastomotic wound healing,

8.1 A Leaking Gut after Anastomosis?

"*For a variety of illnesses including cancer, diverticulitis, or intestinal obstruction ….. resection of a segment of the gastrointestinal tract is a common procedure* [8]" and "*intestinal continuity is reestablished by carefully suturing … the remaining ends together to create a viable and well-sealed connection referred to as an ´anastomosis´* [8]". Such an anastomosis "*therefore is a significant tissue injury given that the surgeon must first divide all of the blood vessels feeding the proposed resection site, all layers of the intestinal wall are divided meaning that their entire thickness from mucosa to serosa is disrupted, ….. then tissues are approximated with sutures and staples that represent foreign bodies embedded into area of healing tissue* [8]." But leaks can occur, despite excellent surgical technique, if there is a lack of healing of the anastomosis. If such a leak occurs, intraluminal gut contents can spillage into the peritoneal cavity, and when that happened, it can be associated with sepsis, a quick re-do surgery is needed, because the patient could die.

That makes an anastomotic leak an unpredictable postoperative complication. It happens during recovery from colorectal surgey, it requires a re-operation (see definition, page 12).

Surgeons (and of course patients) are afraid of associated high mortality and morbidity rates,“*ranging from intra-abdominal abscess to peritonitis, sepsis, and even death*“ [5]. Therefore they tried since decades worldwide to find out the underlying reason, many congresses dealed with this topic, much wisdom was published. The academic discussion was unsuccessful: some thought, the surgical technique is unsufficient (a tension-free anastomosis and adequate blood supply are required), others thougt, the patient bears the blame (and called it patient-related-factors) [9, 10]. “*Discussions of ischemia, tension, suture material, device choice, technique, dog ears* [22, 12]*, are voiced by experts coming to no consensus on the actual cause of the leak* [13]“. Such discussions are *„counterproductive to advancing our understanding of the root causes of anastomotic leak* [13]“ (see also next two chapters).

8.1.1 The Mechanism of Anastomotic Leak

Williamson [14] describes, that *“it is well known that the physiological stress of surgery can itself cause damage and ischemia remote from the anatomic site of surgery which can alter intestinal mucus production, blood flow, cytokine production, and tight junction permeability”,* and *“mucus detachment from the colonic wall … allowing bacteria to have direct contact with intestinal epithelium. Reperfusion injury can also allow goblet cells to release mucus that was able to clear previously bound bacteria”.*

Since studies showed [15] that antibiotic protection of colon anastomoses is possible and needed, the bacterial species involved in the mechanism of anastomotic leaks remained unknown for decades. Now we know, that potentially pathogenic bacteria like Pseudomonas (and Enterococcus), which occur in small amounts in the commensal gut flora, “*contributes to the pathogenesis of anastomotic leak*“ [8, 22, 16, 17], also *Bacillus subtilis* is involved and has been investigated [18].

“*Bacteria that colonize an anastomosis can detect subtle changes present in this site such as cytokines, chemokines, and end products of ischemia to which they often respond with enhanced virulence (i.e., expression of the collagenolytic phenotype)“* [14, 19].

Very important is, that ischemia is not causing anstomotic leak. Hoeppner, a German surgeon from the University of Freiburg, Europe, investigated “an experimental model for colonic healing after ischemia in colonic anastomoses [20]“. He constructed an end-to-end anastomosis at descending colon in twelve German domestic pigs, and compared three groups: “*in Group A, the anastomosis he applied with a leak of 18 mm, Group B addinitionally created an arteficial ischemia oft he proximal anastomotic segment, by ligation of supplying vessels over 5 cm, and in Group C he combined an intentional leakage and an anastomotic ischemia also by ligation of supplying vessels“.* He concluded, that *„large anastomotic dehiscence and local ischemia oft he bowel wall are not reliable factors for the development of intra-abdominal abscess, peritonitis, or sepsis“* [20] (see also chapter 11.8, page 52, and [21]).

8.1.2 The Dispute is over now, Accidential Pathogens are the Key!

The initiating cause behind the pathogenesis of anastomic leak are - luminal bacteria [22]; they are at the interface between pathogenicity and symbiosis, they are also „the driving force behind the pathogenesis [22]“. The incidence in the European literature is high [22, 23, 24]. “*When an anastomotic leak is discussed at a typical surgical morbidity and mortality”* (M&M) *“conference, it is often presented as a due to an error in surgical technique involving ischemia, tension, or device failure.* [13].”

In a multiple analysis in 28.271 patients, recieving an anastomosis after gut resection Kube found [24], that risk factors on anastomotic leak are *„lange Eingriffsdauer, hoher ASA-Score, männliches Geschlecht, Ileus, Tumor im Linkskolon, kardiovaskuläre, hepatogene Begleiterkrankungen, … einreihige Handnaht, Vorliegen einer intraoperativen Komplikation, BMI >30 kg/m^2 “* [direct quotation from [24]]. Those days are gone, never to return, because *“without direct visual analysis of the leak site and its tissue histology, an ex post facto claim*

that an anastomotic leak is due to an error in surgical technique remains speculative [13]", and because to date, interdisciplinary teams have found out that the "*microbiome is the key to preventing anastomotic leaks in colorectal surgery, it modulates the innate immune system response to anastomotic wound healing*" [6, 7]. Now we know that **microbiota from the patient`s own gut can shift perioperatively to a pathogenic microbiome** [22, 8, 13, 25]. A shift to a pathogenic microbiome can also occur due to medication or radiation [26, 27], or be caused by diet [5]. The identified potentially pathogenic bacteria have the capacity "*to degrade collagen and to activate tissue matrix metalloprotease-9 (MMP9) in host intestinal tissues*" perioperatively at the anastomosis [8].

In another research Shogan showed [22], that "*an anastomotic injury appears to significantly alter the tissue-associated microbiota*" and "*this alteration and virulence factor expression was not detected in the luminal contents* [14]", a fact, "*strongly suggesting that the micribiome within anastomotic tissues are likely to play more of a role in healing, rather than species detected in the luminal contents alone* [14]". By quorum sensing, meaning to sense changes in its environment, opportunistic pathogens like *P. aeruginosa* and *E. faecalis "use its virulence activation system to express tissue destroying enzymes" and "respond accordingly as a mechanism for nutrient acquisitation, ….. and survival* [14]*". "A phenotype shift is induced [bacteria that colonize an anastomosis can detect e.g. end products of ischemia and can respond with enhanced virulence like expression of the collagenolytic phenotype"* [19]*. "The human intestinal tract can become colonized by hospital associated pathogens … following severe catabolic injury or exposure to extreme physiologic stress … that replaces the normal commensal flora"* [19].

In some works in the clinical setting a reduction in anastomotic leak after local decontamination with oral, local acting antibiotics was found (see Table 1, page XX).

Shardey was the first to suggest that *P. aeruginosa* might play a causative role in anastomotic leak [28], he found in his milestone work [29] after total gastrectomy a remarkable reduction for anastomotic leak from 10.6% (control group) to 2.9% (in patients treated with a mixture of local acting antibiotics).

8.1.3 Anastomotic Leak and Emergency Patients

An analysis of intervention-related risk factors [24] found that the frequency of anastomotic leak was higher in emergency resections, but *„ob ein Notfall auch tatsächlich mit einem höheren Risiko einer Anastomoseninsuffizienz assoziiert war, hing von der Art des Notfalls ab. Nur beim Ileus bestand eine deutlich erhöhte Anastomoseninsuffizienz-Rate, nicht bei einer lokalen oder diffusen Peritonitis bzw. einem Abszess“* [direct quotation from [24]]. Intraoperative bleeding requiring transfusion (> 2 transfusion units of packed red cells) were also associated with an increased rate of anastomotic leak [24]. In emergency surgery - common at a University hospital - there is not enough time to start “preparations of the patient“ like cleaning the gut by mechanical bowel preparation starting the day before surgery.

8.1.4 Matrix Metallo Protease-9

Collagen is necessary in the recovery period after anastomotic colorectal surgery for wound healing. *P. aeruginosa* and *E. faecalis* produce collagenase when cued by local factors present at anastomotic tissues [8]. Intestinal matrix metallo protease-9 of the host is a proteolytic enzyme, activation from an inactive proform by high collagenase producing strains (like *E. faecalis or P. aeruginosa*) leads to collagen degradation (including type IV collagen), to inflammation of the gut and - to anastomotic leak [8]. In rats it was investigated, that *P. aeruginosa* caused anastomotic leak via production of collagenase [28]. It was shown by Lee [30] that the two highest collagenase producing organisms in human microbiota (*P. aeruginosa* and *E. faecalis)* are the two most isolated organism from anastomotic leak.

Remember (and check chapter 8.1.7 on page 15), that a so called “multispecies probiotic” contains *E. faecalis* and the investigators “found (?!)” a positive effect in prevention of anastomic leak [31].

“Adequate anastomotic healing requires collagen deposition and remodeling.- Enterococcus faecalis and Pseudomonas aeruginosa can be collagenolytic and may activate additional enzymatic pathways that lyse connective tissue. Furthermore, these microbes are enriched in post-operative anastomotic tissue as identified using 16S rRNA sequencing“ [32].

8.1.5 Creating Selective Decontamination of the Digestive Tract

Looking in the medical wisdom for a proper antibiotic to combat *P. aeruginosa* and *E. faecalis* in a study protocol leads to the method called *“selective decontamination of the digestive tract”* [33], used to decontaminate the digestive tract in critically ill patients in the intensive care medicine (to prevent infections acquired in the intensive care unit, particularly respiratory infections / pneumonia in long-term ventilated patients) [34]:

- **It was very inventive to use antibiotics for prevention (!), and not for therapy!**
- **Also proposal of local acting (= non absorbable) antibiotics is very original!**
- **“Selective“ means, the local acting antibiotic respects anaerobic microbiota!**

But:

“Selective decontamination of the digestive tract (SDD) is one of the most controversial topics in the treatment of critically ill patients [35]”. This method was proposed 40 years ago by Stoutenbeek1983 [33], and until to date (2024) Carlet [34] counts in an editorial *that* “*a huge amount of peer-reviewed papers (n=1.010), randomized controlled trials (n=283) performed in the intensive care unit, meta-analysis (n=72) and pro/con debates … have been published“*, but the topic remains controversial and *“unfortunately, these new studies do not close the book on SDD* [35]*“*: in October 2022 in JAMA (Journal of the American Medical Association) have been published (October 26) two original investigations dealing with selective decontamination of the digestive tract - one a randomized clinical trial [36] followed by the other one, a systematic review and meta-analysis [37] - but reporting controversial results. Here we have to consider, that the original concept from Stoutenbeek [33] has been

modified in many studies by the additional use of systemic antibiotics (!), and this use *"did not appear in the acronym ´SDD´* [34]".

Johanson described [38] in 1969 an increased prevalence of Gram-negative bacilli he found in patients with hospital exposure rising markedly in patients with illnesses of varying severity compared with physiologically normal patients among the oropharyngeal bacterial flora that was not correlated with inhalation therapy or antibiotic administration.

The increased incidence was not dependent on duration of hospital stay, but correlated best with the clinical severity of illness. "*… most bacterial pneumonias begin with the aspiration in the lung of bacteria present in the upper respiratory tract, that may represent an important initial step in the pathogenesis of pneumonia due to Gram-negative bacilli* [38]*".* "*Suggesting that pharyngeal clearance mechanisms are impaired in these patients"* [38] *„and an increased exposure to these organisms alone does not adequately explain these findings* [38]*"*.

Besides the importance of clinical severity the description of colonization resistance 1972 by van der Waij [39] and the study by Bodey with local acting antibiotics in high concentration [40] made the proposal of SDD by Stoutenbeek scientific [33]. Bodey initiated his study (Beta-lactam antibiotics alone or in combination with gentamicin) to determine, "*whether antibiotic combination are superior to single antibiotics for the treatment of Gram-negative bacillary infections in neutropenic patients"*. He found (limited by the small number of patients studied), that "*combination therapy may be superior to single antibiotic therapy"*.

In a textbook, published by Springer-Verlag in 2008 [41], edited by van der Voort and van Saene, in extenso the whole story of Selective digestive tract decontamination in intensive care medicine is described as a practical guide to controlling infection to reduce pneumonia. After a description of the history of selective decontamination of the digestive tract by H.K.F. van Saene, H.J. Rommes and D.F. Zandstra the concept is explained by H.J. Rommes. Different authors debate the effect on mortality and that resistance does not appear to be a clinical problem. In the preface both editors pointed out, that "*it has been shown that the results obtained by individual intensive care units vary in the degree of success in*

decontamination and the outcomes they reflect" [Preface of the textbook]. A proper understanding of the described principles is needed, because "*the effects of the decontamination … can be completely lost in a multicentre study if … basic conditions are not all equally in place".*

Author	Year	Milestone
Johanson	1969	reports change of digestive flora according to disease severity
van der Waij	1972	"colonization resistance"
Bodey	1976	non-absorbable antibiotics (in high concentration) eliminates GNB (Gram negative bacteria)
Stoutenbeek	1983	proposed SDD (colistin/tobramycin/amphotericinB)
Unertl	1987	modification of SDD in the first trial published (polymyxinB/gentamicin)
van Saene	2008	Textbook SDD
Silvestri	2012	reports 66 trials, 11 meta-analyses on SDD
Hammond	2022	meta-analysis (24.389 participants), NO resistance
SuDDICU	2022	Y/N Effect of SDD on Mortality (mechanical ventilation)
Rodríguez-Gascón	2024	NO antibiotic resistance with SDD
Carlet	2024	SDD: Make decision, on this 40-year old controversy

Table 1: Milestones of the historical developement of SDD. [Johanson 1969 [38]*,* van *der Waij 1972* [39]*, Bodey 1976* [40]*, Stoutenbeek 1983* [33]*, Unertl 1987* [42]*, van Saene 2008* [41]*, Silvestri 2012* [43]*, Hammond 2022* [37]*, SuDDICU 2022* [36]*, Rodriguez-Gascón 2024* [44]*, Carlet 2024* [34]*].*

Anyway, the first randomized trial with selective decontamination of the digestive tract was published by Unertl, a German surgeon from Ludwig-Maximilians-University of Munic, in 1987. "*Subsequently, 66 randomized controlled trials .. and 11 meta-analyses of RCTs [randomized controlled trials] of selective decontamination of the digestive tract have been published* [43]" until 2012. In an other metaanalysis Hammond found 2022 [37] no antibiotic resistance (24.389 participants).

The first clinical experience was published 1983 by Stoutenbeck and van Saene [33], both from Groningen, The Netherlands, a city, that became the birthplace of selective decontamination of the digestive tract. The term "selective" does not mean just "selective suppression of pathogens", but also that *„eine Selektion von Patientengruppen nach Art und Schwere der Grunderkrankung getroffen wird"* [direct quotation [45]].

Selective decontamination of the digestive tract is a prophylactic strategy (usually tobramycin, colistin {inactive prodrug, polymyxin E is the active antibiotic} and amphotericin B [45]) to minimize infections and to reduce mortality in the intensive care unit, caused by potentially pathogenic micro-organisms [43]].

Amphotericin B was chosen, because „*neben der Kolonisation der Trachea*" in ventilated patients at an intensive care unit „*ist auch die Kolonisation der inneren Oberfläche von Tubus und Beatmungsschläuchen für die Entstehung von Pneumonien bedeutsam, ….. im Kondenswasser können innerhalb weniger Stunden ….. Bakterien anwachsen, die ursprünglich aus der Mikroflora des Oropharynx und der Trachea stammen"* [direct quotations from [45]; [46]]".

The purpose of the treatment is to eradicate potential pathogenic microorganisms from the oropharynx and digestive tract of patients at risk for nosocomial infections (ventilated patients, neutropenic patients and neonates); the targeted potential pathogenic microorganisms include aerobic Gram-negative bacteria (GNB), methicillin-susceptible ***Staphylococcus aureus*** and yeasts, and once a patient has been successfully decolonized the unaffected anaerobic flora would offer prevention against new

colonization with potential pathogenic microorganisms [48], a principle called colonization resistance [39].

The composition of the antibiotics for selective decontamination of the digestive tract was modified by Unertl from Munic; he has chosen [45] a combination of polymyxin B and **gentamicin**, because "*Gram-negative rods, especially Pseudomonas aeruginosa were the predominating colonizing organism in ventilated patients and were found to be sensitive to at least one oft he two agents. … Both agents exert a high and prolonged local activity when given orally, show no systemic toxicity as they are not absorbed through intact mucosa and are well tolerated, … cross-resistance between polymyxin B and other antimicrobial agents is uncommon, and amikain could be expected to be a suitable substitute if resistance again gentamicin developed* [42]." To prevent yeast colonization amphotericin B was added; Unertl expected to have only minor effects upon other oral microbiota [42].

8.2 Leakless *Anastomosis*

For anastomotic surgery the situation differs from that, described for ventilated patients at an intensive care unit, but the target (eradicate *Pseudomonas aeruginosa*) is the same! In the one case, *Pseudomonas aeruginosa* (coming from the gut) causes pneumonia, in the other case the bacteria can lead to collagen degradation and disturb wound healing (via activating matrix metallo protease-9) when shifted to a pathogenic bacteria.
Shogan [8] showed, that parenteral cefoxitin (given in standard procedures of colorectal surgery) did not kill *E. faecalis* and did not prevent anasomotic leakage.

8.2.1 If a Method Works, why not Try it in Another Set? (Milestones)

Anyway, Schardey first introduced in the year 1997 a local decontamination when he performed a prospective, randomized, double-blind placebo-controlled multicenter trial for the prevention of anastomotic leakage after total gastrectomy through local decontamination [29]. His article in Coloproctology [47] triggered this dissertation (see preface). Following a literature research (see Table 1 in the chapter results), we decided: Never change a winning team. The remaining question was, what is the proper mixture for a local decontamination to perform a study leading to prevention of anastomotic leak for the maximalized benefit for patients (and surgeons too)? Similar to Schardey, who wrote [cited in direct quotations] in the overview „*sind Darmbakterien an der Entstehung der Anastomoseninsuffizienz beteiligt ?*“ [47], that reduction in anastomotic leak (in the investigations of his group by an impressive rate from 11,9 % to 5 %) was not sufficiant for him, and „*wir wollten diese Möglichkeit der Optimierung nutzen*“ [47], so we returned „*zurück … zu Vancomycin als Teil unseres Dekontaminationsschemas, die wir im Rahmen unserer Studien getestet hatten*“[47]; we decided also to include vancomycin to improve results, because „*addition of vancomycin can improve the effectiveness against Enterococci*“ [47], but we did not add amphotericin B (against yeast).

Bura mat dekho, bura mat suno, bura mat bolo.

Mahatma Ghandi [knowlwdege from schooldays]

8.2.2 `Antibiotic Resistance` and `Overgrowht Resistance`

Resistance is not always the same. Antibiotic resistance is well defined in medical textbooks, the term `colonisation resistance` was introduced 1972 by van der Waaij [39] and means, that the unaffected anaerobic flora in a successful decolonized patient safeguards itself against new colonization (and invasion) of potential pathogenic microorganisms [48]. Selective decontamination of the digestive tract is a method, that "*involves the application of topical, nonabsorbable antimicrobial agents (usually colistin, tobramycin, and nystatin) that selectively spare the anaerobic flora*" [44].

It was controversally discussed if this method can lead to an antibiotic resistance. Franz Daschner, an "antibiotic opinion leader" from Freiburg, made a major contribution to critizise strongly the antibiotic concept of selective decontamination of the digestive tract (SDD): he "*has written repeatedly since 1987 that SDD promotes resistance; he still continues in this vein today*" [41, p 16]; van Saene was referring to the article from the group of Daschner, dealing with "*bacterial resistance and overgrowth due to selective decontamination of the digestive tract*" [49], and reminded, that "*during the mid-1980s he [Daschner] swore ´SDD mit allen Mitteln nieder zu machen´ "* [41, p16]. But was the "opinion" of Daschner correct? The answer is cristal clear: See, hear but also speak no evil [Mahatma Ghandi, knowledge from schooldays].

In 2002 the first trial was published by Unertl [42] "*demonstrating a significant survival benefit in patients receiving SDD*" [41]. And in a recent [Epub ahead of print] "*long-term comparison of two intensive care units (with and without SDD) of the same tertiary hospital*" [44] the authors point out their point of view, and that "*in settings with relatively low prevalence of multidrug-resistant pathogens, such as ICUs [Intensive Care Units] in the Netherlands, Australia, and New Zealand, evidence that the implementation of SDD has a negative impact in resistance ecology is scarce*" [44, 35, 50].

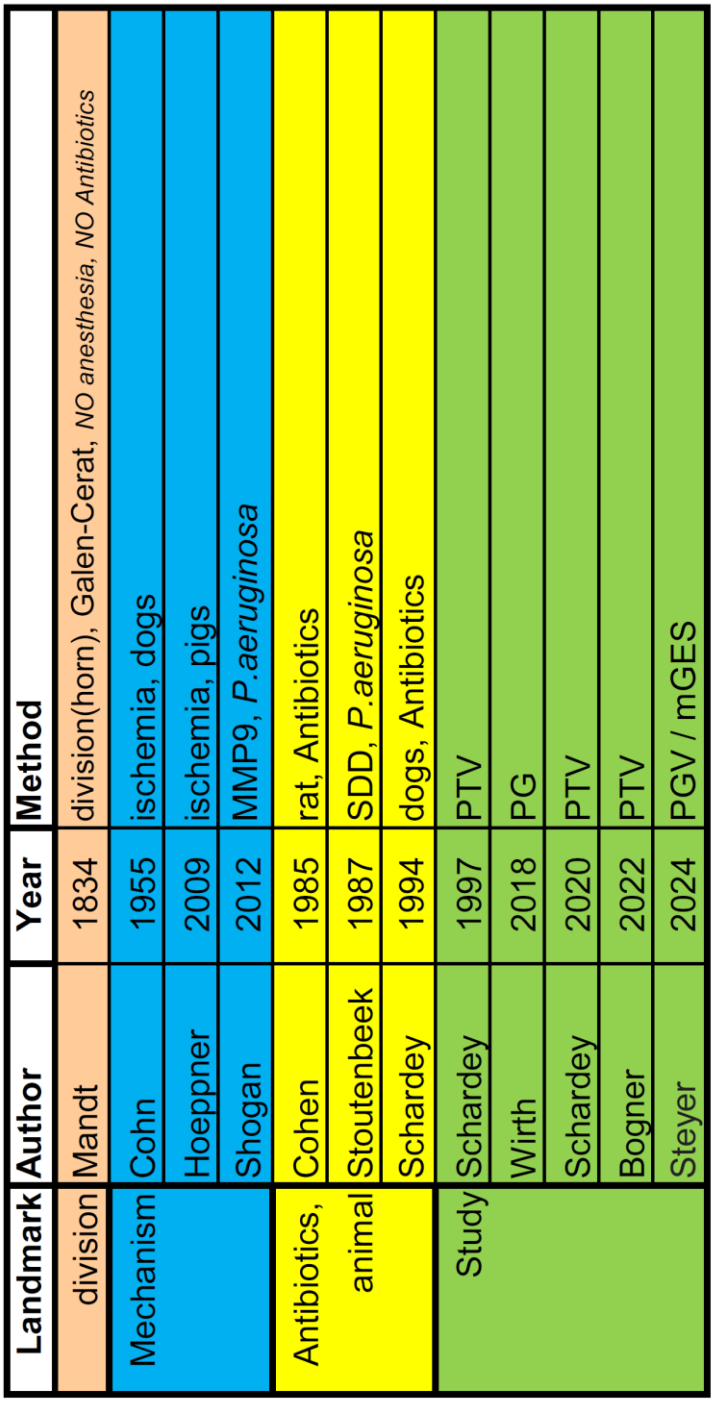
How can it happen, that "***SDD is one of the most widely evaluated interventions in critically ill patients**, yet its use is not widespread"* [51]? "*Selective digestive decontamination (SDD) is a prophylactic strategy aimed at preventing or eradicating bacterial overgrowth in the intestinal flora that precedes the development of most infections in the Intensive Care Unit.* And the administered non-absorbable antibiotics "*must meet the following criteria:*

- *SDD prevents serious infections,*
- *reduces mortality,*
- *is cost-effective,*
- *has no adverse effects,*
- *and its short- or long-term use is not associated with any significant increase in antimicrobial resistance"* [51].

The method was developed following some reports, like that from Johanson who described in 1969 *"that the digestive flora of patients **changes [!]** after a few days of hospital admission, with a predominance of Gram-negative bacteria. … The main factor underlying this change is **disease severity**"* [38]. The prevalence of gram-negative bacilli among the oropharyngeal bacterial flora ….. correlated best with the clinical severity of illness [38], "*many systemic antimicrobials can sterilize the lungs, blood and bladder, but are usually unable to eliminate such GNB from theoropharynx and/or intestine. He found that the enteraladministration of non-absorbable antibiotics can eliminatethe GNB* [Gram-negative bacteria] *from the gastrointestinal tract, as a result of the high drug concentrations reached in the intestinallumen. The combination of polymyxin E (colistin) and tobramycin was chosen given its efficacy against GNB, including Pseudomonas spp."* [40].

8.2.3 The Genesis of mGES (muraler GedärmeEpithelSchutz)

To create a **synergistic antibiotic combination (mixture)** to combat *P. aeruginosa* & partners in a new study a literature search was started (see chapter "Results", Table 3 and 7a + b).



Landmark	Author	Year	Method
division	Mandt	1834	division(horn), Galen-Cerat, *NO anesthesia, NO Antibiotics*
Mechanism	Cohn	1955	ischemia, dogs
	Hoeppner	2009	ischemia, pigs
	Shogan	2012	MMP9, *P.aeruginosa*
Antibiotics, animal	Cohen	1985	rat, Antibiotics
	Stoutenbeek	1987	SDD, *P.aeruginosa*
	Schardey	1994	dogs, Antibiotics
Study	Schardey	1997	PTV
	Wirth	2018	PG
	Schardey	2020	PTV
	Bogner	2022	PTV
	Steyer	2024	PGV / mGES

Table 2: Landmark studies in the developement of mGES. [von Mandt 1834 [52], Cohn 1955 [53], Hoeppner 2009 [20], Shogan 2012 []22, Cohen 1985 [54], Stoutenbeek 1987 [55], Schardey 1994 [56], Schardey 1997 [29], Wirth 2018 [57], Schardey 2020 [58], Bogner 2022 [59], Steyer 2024 [4]].

We found, that the combination of polymyxin B, gentamycin and vancomycin fits best and decided not to add an antimycotic substance (like amphotericin B).

8.2.4 Verification of mGES

In this trial, we wanted to verify that the study medication and the method mGES can reduce anastomotic leak.

Shogan [8] describes, that "the *E. faecalis* genes *gelE* and *sprE* were required for *E. faecalis*-mediated matrix metallo protease-9 activation". Very important for our protocol was, that an application of intravenous antibiotics, as required by clinical standard procedure given in patients undergoing anastomoses surgery, eliminated *E. faecalis* at anastomotic tissue, also no prevention of anastomotic leak was shown; but through topical acting antibiotics it was possible to prevent anastomotic leak [8].
In his investigation, Shogan [8] could also show, that in patients undergoing colon surgery, their anastomotic tissue still contained *"E. faecalis and other bacterial strains with collagen-degrading matrix metallo proease-9 when treated with the standard recommended intravenous antibiotics"*, suggesting that these bacteria *"break down collagen in the gut tissue contributing to anastomotic leak* [8]".

8.2.4.1 Justification of the Research Question

From literature we know, that anastomotic leak in colorectal surgery is an unsolved problem. We wanted to investigate, wether a new method can reduce anastomotic leak in emergency and elective patients in routine surgery at a University hospital.

8.2.4.2 Aim of the Dissertation

The objective of this investigation was to assess the effectiveness of local acting antibiotics in reducing anastomotic leak in colorectal surgery in elective and emergency patients, and to find a method to reduce anastomotic leak by combatting *P. aeruginosa,* and create an indirect proof of involvement of this bacteria in causing anastomotic leak.

8.2.4.3 Hypotheses

In the present study we presumed

- a significant reduction of anastomotic leak in the group of patients recieving the local acting antibiotic mixture (polymyxin B – 100 mg, gentamycin – 80 mg, vancomycin – 125 mg) compared with a control group (not recieving the antibiotics), and that
- a short course of 5 days intake (every 6 hours per day) of mGES is sufficiant for emergency and elective patients undergoing colorectal surgery recieving an anastomosis with respect to anastomotic leak.

8.2.4.4 Novelty Value

We used for the first time in a study mGES, the antibiotic mixture of three antibiotics (polymyxin B – gentamycin – vancomycin).

8.2.4.5 Limitations of the Research Topic

We could not investigate in labor analyses the presence of *P. aeruginosa* or *E. faecalis,* but we know from recent literature, that these bacteria cause leaks by acting colagenolytic via activating matrix metallo protease-9 [8].

We could not perform dose finding experiments, important perhaps for very obese patients (BMI > 35).

9. Material and Methods

9.1 Defining `Graded Anastomotic Leak`

In the medical literature there is a surprising lack of a standardized definition of anastomotic leak based on the heterogenity in the anastomotic leak definition in many studies [60]. Bruce identified [61] a total of 56 different definitions of anastomotic leak in 97 reviewed studies at three sites: *„lower gastrointestinal (29 definitions), hepatopancreaticobiliary (14 definitions) and upper gastrointestinal (13 definitions)“*. That is really very surprising, even though anastomotic leak represents a major dreaded complication after colorectal surgery, resulting in a widely reported [62] variation from 4 to 20 % [4].

Our first aim in the present study therefore was to define anastomotic leak itself fort he study protocol submitted to the ethics committee, ist proposed feasible cause, the entity (early vs, long) and, consequently, the endpoint and the duration of the observation time. We follow [4] the study results from Floodeen [63] in that early and late symptomatic leakage may be viewed as different entities. The severity of anastomotic leakage should be graded according to the impact on clinical management [60]. Grade A: anastomotic leakage results in no change in the patient's management. Grade B: leakage requires conservative management but no re-do surgery. Grade C: anastomotic leakage requires re-do surgery. In this study protocol, we defined Grade C as the end of the investigation.

Anastomotic leak can be defined primarily as a leak of luminal contents from a surgical join between two hollow viscera [64]. In our study protocol, we add on two more clinically important factors (see also chapter 11.8): point of time [65] of anastomotic leak and, secondly, the requirement of re-do surgical intervention (=severity of AL). Therefore, we determined anastomotic leak in daily, routine procedures at a university hospital which was modified from the literature [60, 65, 66] as follows: **Anastomotic leak is a clinically obvious defect of the intestinal wall integrity at the colorectal anastomosis site (including suture) that leads to a communication between the intra- and extraluminal compartments, requiring re-do surgery within 7 postoperative days.**

9.2 The Observation Time in this Study

The temporal occurrence of anastomotic leak during recovery from colorectal surgery also has to be defined for the protocol [4] because it is well known that symptomatic anastomotic leakage that requires re-do surgery following surgical resection can occur during the initial hospital stay (up to 7 postoperative days) or later, after hospital discharge. The point in time of anastomotic leak is very important [65] with respect to the severity and management of this complication. Late anastomotic leak is defined [66, 67, 68, 69] as leakage that occurs after 30 postoperative days, but this is a rare event [62] (occurring in less than 4% of colorectal cases in one study [70]. By contrast, early anastomotic leak is associated with severe peritonitis, emergency re-do surgery and increased mortality, with a median occurrence time of 5 - 6 postoperative days [65, 78].

9.3 Patients [4]

We included all consecutive patients (older than 18 years of age) from our clinic who required left-sided colorectal anastomosis; that means not “only elective resections” (being an academic inclusion criteria in many studies) but also urgent patients, representing the daily routine at a university hospital. We did not include pregnant females (because of the antibiotic intervention) or persons with known antibiotic allergy. No restrictions were applied regarding gender. We compared an intervention group with a retrospective control group.

During the inclusion period, we had a slight delay in our timetable due to COVID-19 (SARS-CoV-2); there was no change in the suture material (polydioxanone 4-0 or 5-0), stapling device or technical management of gut surgery. Clinical data were retrospectively obtained from our internal documentation system (MEDOCS).

9.4 Design

In our mono-center study of anastomotic leak after left-sided colorectal anastomosis we compared an intervention group and a retrospective control group (without local antibiotic decontamination). In the peri- and postoperative period, the clinical decisions were identical in both groups. If, in the emerging situation, a diverting stoma was required, topical administration of the study drug was given transanally. Day of inclusion (T0) was the day of surgery. The intervention period followed from T0 to T5. On day T7, the observation time ended (see definition of anastomotic leak) [4].

9.5 Antibiotic Intervention (mGES) to Combat Accidential Pathogens

In the intervention group, patients received (in addition to the standard antibiotic prophylaxis starting preoperatively, which was an i.v. of piperacillin/tazobactam - a broadspectrum ß-Lactam antibiotic that can act via penetration in Gram-negative bacteria, as well as against *P. aeruginosa*, but when used alone, it lacks strong activity against the Grampositive pathogens) an antibiotic mixture 4 times a day for 5 consecutive days after surgery, beginning the day before surgery in elective patients and as soon as possible in emergency patients. With respect to previous studies, we chose a mixture (PGV) that could combat Gram-positive and Gram-negative pathogens (like *P. aeruginosa* or *E. faecalis*, respectively): polymyxin B (100 mg), gentamicin (80 mg) and vancomycin (125 mg). The PGV medication was chosen by our study group according to the approved study protocol and was then prepared by our pharmacy at the Medical University of Graz and filled in capsules.

9.6 Statistics [4]

Patients' characteristics are presented as medians and expressed as a percentage. For the comparisons, we used the Mann–Whitney U test for continuous data and Fisher's exact test for binary data. A p value < 0.05 was considered statistically significant, and a p value $<$

0.001 highly significant. Statistical analyses were carried out using IBM SPSS Statistics v29 (IBM Corp., Armonk, NY, USA).

10. Results

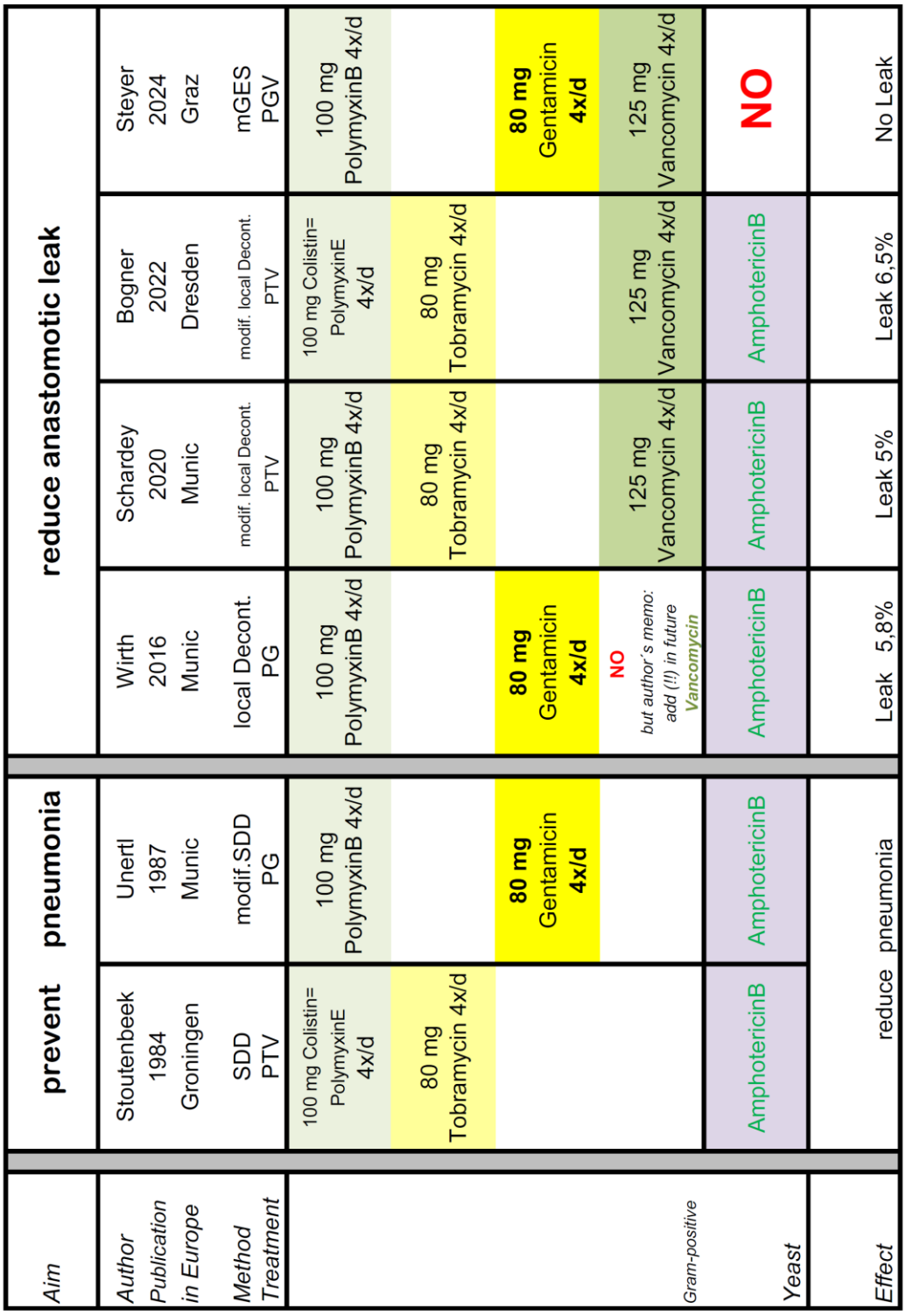
10.1 Results for the evolution of mGES

Different studies proofed the effect of selective decontamination in intensive care units for preventing or reduce pneumonia, the used antibiotic mixture was modified [33, 42]. In his description of the history of selective decontamination van Saene described [71, 33] reasons, why he added amphotericin B, an antimicrobial substance: in the presence of antibiotic prophylaxis he wanted to avaiod potential superinfections with fungi.

Schardey and his group showed, that an adapted concept of that method (local acting antibiotics combined with antimicrobial substances) can be helpful to combat *P. aeruginosa* also in colorectal surgery (see Table 3).

Other combinations have shown an reducing effect for anastomotic leak. Wirth 2018 [57] reported a reduction to 5.8 %, he did not include vancomycin B; Schardey found [58] 5 % in a follow-up study 2020, and in Dresden, Germany, Europe, Bogner published [59] a reduction from a high level in his clinic from 14.9 % to 6.5 %).

We decided in our study, not to include amphotericin B (500 mg), and to use gentamicin, tested in other studies, but not published combined with polymyxin B and vancomycin in a study to reduce or prevent anastomotic leak. Analyzing the results from our study, we found a high level of emergency patients in both groups.



Aim	prevent pneumonia			reduce anastomotic leak			
Author Publication in Europe	Stoutenbeek 1984 Groningen	Unertl 1987 Munic	Wirth 2016 Munic	Schardey 2020 Munic	Bogner 2022 Dresden	Steyer 2024 Graz
Method Treatment	SDD PTV	modif.SDD PG	local Decont. PG	modif. local Decont. PTV	modif. local Decont. PTV	mGES PGV
Gram-positive	100 mg Colistin= PolymyxinE 4x/d	100 mg PolymyxinB 4x/d	100 mg PolymyxinB 4x/d	100 mg PolymyxinB 4x/d	100 mg Colistin= PolymyxinE 4x/d	100 mg PolymyxinB 4x/d
	80 mg Tobramycin 4x/d			80 mg Tobramycin 4x/d	80 mg Tobramycin 4x/d	
		80 mg Gentamicin **4x/d**	**80 mg** Gentamicin **4x/d**			**80 mg** Gentamicin **4x/d**
			NO *but author´s memo: add (!!) in future* ***Vancomycin***	125 mg Vancomycin 4x/d	125 mg Vancomycin 4x/d	125 mg Vancomycin 4x/d
Yeast	AmphotericinB	AmphotericinB	AmphotericinB	AmphotericinB	AmphotericinB	**NO**
Effect	reduce pneumonia		Leak 5,8%	Leak 5%	Leak 6,5%	No Leak

Table 3: Genesis of mGES. [Stoutenbeek 1984 [33], Unertl 1987 [42], Wirth 2016 [57], Schardey 2020 [58], Bogner 2022 [59], Steyer 2024 [4]].

10.2 Results of Interventions for Preventing anastomotic leak in the Previous Literature

We found [4] in our literature search three studies using PT (=polymyxin B {or E} and tobramycin) for decontamination [72, 73, 74, 57], resulting in a minor (in two studies, not significant) reduction in anastomotic leak (from 9.7% to 6.6%).

Another study [57] had no control group and used PG (=polymycin B and gentamicin) and reported anastomotic leak in 5.8%. Three other studies [29, 58, 59] used PTV (=polymyxin B (or E), tobramycin and vancomycin but no gentamicin) and reported, respectively, a reduction in anastomotic leak from 14.9% to 6.5%, 20% to 5% and 10.6% to 2.9%. Also, these three studies investigated elective patients only.

In the previous literature, we saw a remarkable reduction in anastomotic leak through local decontamination, but our antibiotic mixture mGES that was used in this trial for the first time in a trial.

10.3 Patients' Demographic Data

The two cohorts did not differ in baseline parameters (see Table 4, compare [4]); Control Gropup and Intervention Group were comparable with respect to the patients' characteristics:

		IG n=149		CC n=152		
		No. (%)	Median	No. (%)	Median	p value
Gender						
	Male	80 (53.69%)		72 (47.36%)		0.163
	Female	69 (46.30%)		80 (52.63%)		
Age			63		63	0.405
BMI			25		25	0.574
ASA						
	I	14		12		0.398
	II	66		63		0.351
	III	52		59		0.279
	IV	17		18		0.525
AL						
Redo-surgery		0.0		11 (= 7.23 %)		<0.001

Table 4: Patients´ characteristics. IG = Intervention Group; CG = Control Group; No. = number of patients; BMI = Body Mass Index; ASA score = American Society of Anesthesiology Classification System; AL = Anastomotic Leakage. A p value <0.05 was considered statistically significant, and a p value <0.001 highly significant.

10.4 Results of the Intervention

In the Control Group (n = 152), we found 11 (= 7.23%) necessary re-do operations vs. none in the treated Intervention Group (n = 149). With the new local decontamination mGES; this

result is highly significant and in consensus with medical studies and with the literature [knowledge from Textbook].

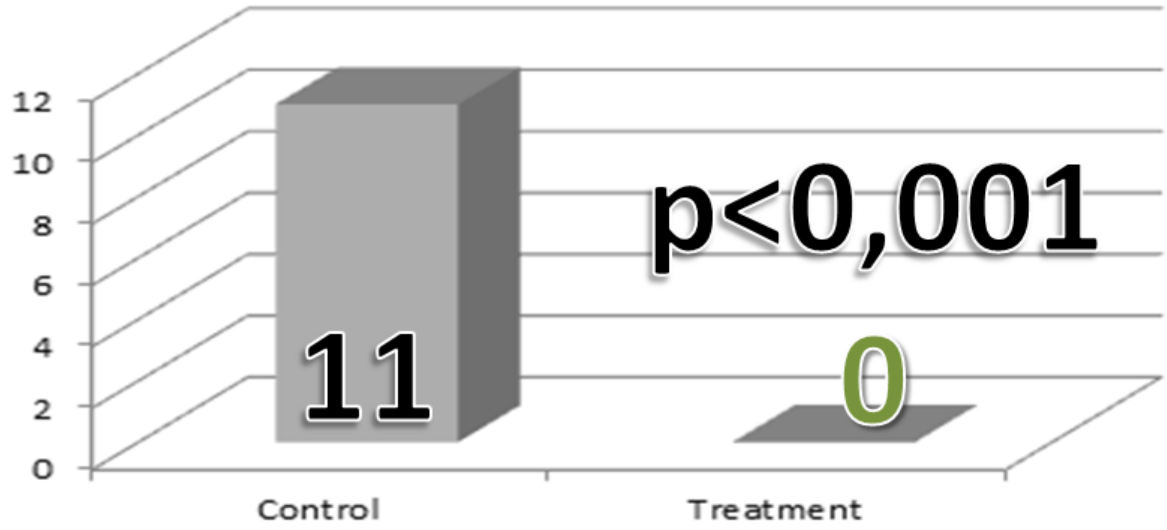


Fig. 2: Results of anastomotic leak in this study. According to our protocol clinically we did not find a single case of AL in the treatment group, but in the control group we found 11 cases with necessary redo-surgery (n=301); that result is highly significant (p<0,001 in the Fisher exact test).

11. Discussion and Conclusions

11.1 Requirements [4]

- Anastomosis must heal in the presence of fecal contamination [75].
- Microbiota, perioperatively, can shift to a pathogenic flora and contribute to the pathogenesis of AL through its capacity to activate MMP9 and degrade collagen in host intestinal tissue [8].
- A mixture of locally acting antibiotics - specially designed against Pseudomonas aeruginosa and Enterococcus faecalis - can reduce anastomotic leak [58].

11.2 The Proper Antibiotic Mixture for Prevention?

At the same time in 2018 we found two articles dealing with anastomotic leak: one review came from Schardey [47], describing his results with local decontamination, an other one came from Alverdy [76] and described the mechanism of surgical infection and the role of *P. aeruginosa*. That helped us to perform a study protocol.

11.3 Alternatives for Reducing Anastomotic Leak?

To combat a bacteria like the nosocomial hospital bug *P. aeruginosa,* a strong antibacterial substance is needed.

The use of oral polyphosphate [77] to suppress bacterial collagenase production is not sufficient investigated and available for routine use.

Also stool transplant is not applicable. And always remember the use of the method in emergency patients.

11.4 Multispecies Probiotics do NOT Work, have NO Mechanism of Action

Do detect, how studies dealing with “multispecies probiotics” very often are designed, it is helpful to discuss for example a “clinical investigation”, published 2012 by Zhang [31]. The study group describes, that preoperative given probiotics decrease postoperative infectious complications (anastomotic leakage or fistulae) of colorectal cancer. *"Perineal infection was characterized by an abscessed perineal wound that discharged spontaneously or required surgical draining. Anastomotic leakage designated a symptomatic leak that was verified by a water-soluble contrast enema .."* [31]. Two (extremely small) groups of 30 patients were compared in this double-blind placebo controlled “study”. 82 patients were included in this study and then 22 were exluded. One group received placebo, the other group received for (only) 3 preoperative days 3 times a day oral capsules, each of which contained 0.21 g (10^8 cfu/g) of *B. longum, L. acidophilus* and *E. faecalis*. **Remember:** *E. faecalis* is causing (!) anastomotic leaks by activating matrix metallo protease-9. They concluded, that a *“preoperative oral bifid triple viable probiotics minimize the postoperative occurence of infectious complications, with possible mechanisms attributed to the maintenance of the intestinal flora and restriction of bacterial translocation from the intestine”* [31].

Looking into details, patients also received additionally a lot of antibiotics (not described in the abstract): a conventional bowel preparation was performed on preoperative day 2 (days -2 and -1), **including** the administration of a full liquid diet, oral **gentamicin** (80,000 U, **three times a day**), **metronidazol** (0.4 g, **three times a day**) …, a postoperative prophylactic regimen of 3 g of cefuroxime sodium and 1 g of **metronidazole** … was intravenously infused **twice daily for 3 to 5 days**.

Is a comment really necessary? Is it really a miracle, what worked to decrease postoperative infectious complications (anastomotic leakage)?-

For elective patients only, another group named their study a priori “Miracle” ! [79]. They compared a series of 131 elective patients recieving oral antibiotics, mechanical bowel preparation and perioperative probiotics to a control group of 500 patients recieving a standard protocol in a single high-volume centre. The multispecies probiotic was administered oral twice a day starting 5 days before surgery and was continued until 4 days after surgery, and during the operation two doses were instilled into the lumen of the proximal anastomotic stump; in intracorporal anastomoses the solution (20 cc) was injected

by a cannula. The multispecies probiotic contained 8 different strains: *Streptococcus thermophilus, Bifdobacterium breve, Bifdobacterium longum, Bifdobacterium infantis, Lactobacillus acidophilus, Lactobacillus plantarum, Lactobacillus paracasei* and *Lactobacillus delbrueckii subsp. Bulgaricus.*

Looking also into details in this study, patients received additionally a lot of antibiotics (not described in the abstract): *"oral antimicrobial prophylaxis was obtained with amoxicillin and clavulanic acid 1 g every 12 h and metronidazole 250 mg every 8 h from the day before surgery until the frst postoperative day. On the day of the surgical procedure, oral antibiotics was administered two hours before surgery, intravenous antibiotics were completely abolished. In the case of penicillin allergy, oral ciprofloxacin 500 mg was administered twice a day* [79]". The aim of the study was *"to confirm the safety of the protocol and validate its efcacy compared to a standard protocol in reducing the anastomotic leak rate … after colorectal laparoscopic surgery* [79]".

Of course, the method is not applicable in emergency patients. Additionally the list of exclusion criteria is very long: excluded were *"ASA IV stage, diverticular and infammatory diseases, emergency procedures, transanal microsurgery or minimally invasive surgery, intraoperative major complications or anastomotic defects, temporary diverting stomas or no anastomosis construction (such as abdominoperineal resection or colonic resections with terminal colostomy)* [79]". The primary outcome was anastomotic leakage. The incidence of anastomotic leaks was reduced from 6.5 % in the control group (n=500) compared to 1.7 % in the multispecies/antibiotic group (n=131), reduction of surgical site infections did not reach statistical significance in this miracle study [Marcellinaro 2023]. Interestingly the authors are aware of the publication of Guyton 2017 [80] and cite, *"that intestinal bacteria respond to host environment changes with a phenotypic shift, and some of them become virulent pathogens, producing collagenases, which can contribute to the development of anastomic leaks"*, because *"bacterial collagenases have a double action: they breakdown collagen I and activate local tissue matrix metallo proteinase-9, which destroy collagen IV. resulting in rupture of the anastomotic tissue."*

But what do they conclude to improve their protocol?

Do they add a propper antibiotic?

To add "*amoxicillin, clavulanic acid and metronidazole"* does not combat *P. aeruginosa* and reduce anastomotic leak, but it is just causing a disturbed microbiota.

Do they add a proper probiotic, specially designed for the given aim? Do and can they explain the mechanism of action for the probiotic mixture of 8 accidential strains (in unknown amounts)? Intake of multispezies probiotics in studies was sometimes very problematic, because of bad taste patients´ comlience was extremely low, the price for a multispecies product is also to high for routine handling except in sponsored trials and lack of good studies is evident.

Instead of answering all this question in their paper. authors declare affiliation to a Emergency Department (Remember: emergency patients are an exclusion criteria in their investigation), but anyway: **La Sapienca** in Rome is their home; does it mean, the protocol is automatically **science** or still a miracle? Who will buy access to that content in a journal (and pay only EUR 39.99)? Bura mat dekho, bura mat suno, bura mat bolo … make science based recommendations (do not use terms from advertisments of selling companies like "good, bad und ugly bacteria" instead of defining properties of bacteria when selling a supplement)

- and please, keep away (those) multispecies probiotics from patients´ gut. Every surgeon shall be aware, if a patient trusts advertising of "available products".

11.5 Discussion of Former Findings in the Literature [4]

To reach our aim (=to reduce anastomotic leak in daily routine patients undergoing left-sided colorectal surgery in our hospital which has a high rate of emergency surgery), we had to find the proper intervention drug.

A literature search now listed in a chronologically way, in my opinion starts with a case report series from the German surgeon Martin Wilhelm von Mandt [52] published during his time at the University of Greifswald, Europe; this milestone article (see also Table 5 on page 48), published 1834 in Berlin in *„Magazin für die gesammte Heilkunde, mit besonderer Berücksichtigung auf das allgemeine Sanitäts-Wesen im Königlich Preussischen Staate"*

(edited by his teacher Johann Nepomuk Rust), explains the successful dividing of gut content and surgical anastomosis (this paper I found in the private library of my grandfather, and follow the citation given in that book, because in the year 1825 this magazine was renumbered: Neue Folge 1, resulting in number 18 instead of number 42 for the new series in the year 1835). Mandt describes a *„Resektion eines Theiles des Intestini recti“* in detail, in his words „in extenso“. By intuition he realized, that it was necessary for a successful rectal surgery (at this time without anaesthesia and without antibiotics), to create a special construction by a division horn made of ivory, and to introduce his new surgical method. He explained [direct quotation, Mandt 1834 [58, p.21ff]], *„da ich die unmittelbare Berührung der Fäcalmaterien mit der frischen Wundfläche vermeiden wollte, so hatte ich vor der Operation eine trichterförmige Röhre von 4 Zoll Länge … aus einer ganz dünnen Hornplatte* [Elfenbein] *anfertigen lassen“, … „aus dem Angeführten wurde es mir wahrscheinlich, dass die organischen Veränderungen innerhalb des Darmes als die Produkte von Entzündung zu betrachten sein dürften“.* By suture he fixed the division horn and hoped *„wenigstens die nächsten Tage hindurch meinen Zweck zu erreichen, und späterhin die Befestigung dadurch zu veranlassen, dass durch die Trichterröhre ein Wachsbougie in den Darm eingeschoben … und durch eine schickliche Bandage in dieser Lage erhalten wurde“*. *„Der Kranke war sehr erschöpft, die Operation hatte etwa eine halbe Stunde gedauert.“* For the healing of the wound he tried to reach „die höchst mögliche Reinlichkeit der Wunde“, and he *„liess ein 4 ½ Zoll langes, dickes und conisches Bourdonnet anfertigen, mit Cerat* [an oil, known as Galen-Cerat] *bestreichen, und führte dasselbe so hoch in den Darm ein als ich konnte“.* On day 7 after surgery he recognized, that *„die Wunde bereits zu vernarben anfing; der Verband … musste nach jeder Entleerung sorgfältig erneuert werden“*. … *„Nach 6 Wochen war die Heilung beendigt, so dass er aus dem Clinico entlassen werden konnte“*. Mandt received one year later a message, that the patient was again able to work. From this historical pioneer case we learn, how successful it is, to divide stool content from the healing process at the epithelial layer of the gut, we now call it mGES (muraler GedärmeEndothelSchutz / mural GutEndothelialShelter) – just the method has become more mature.

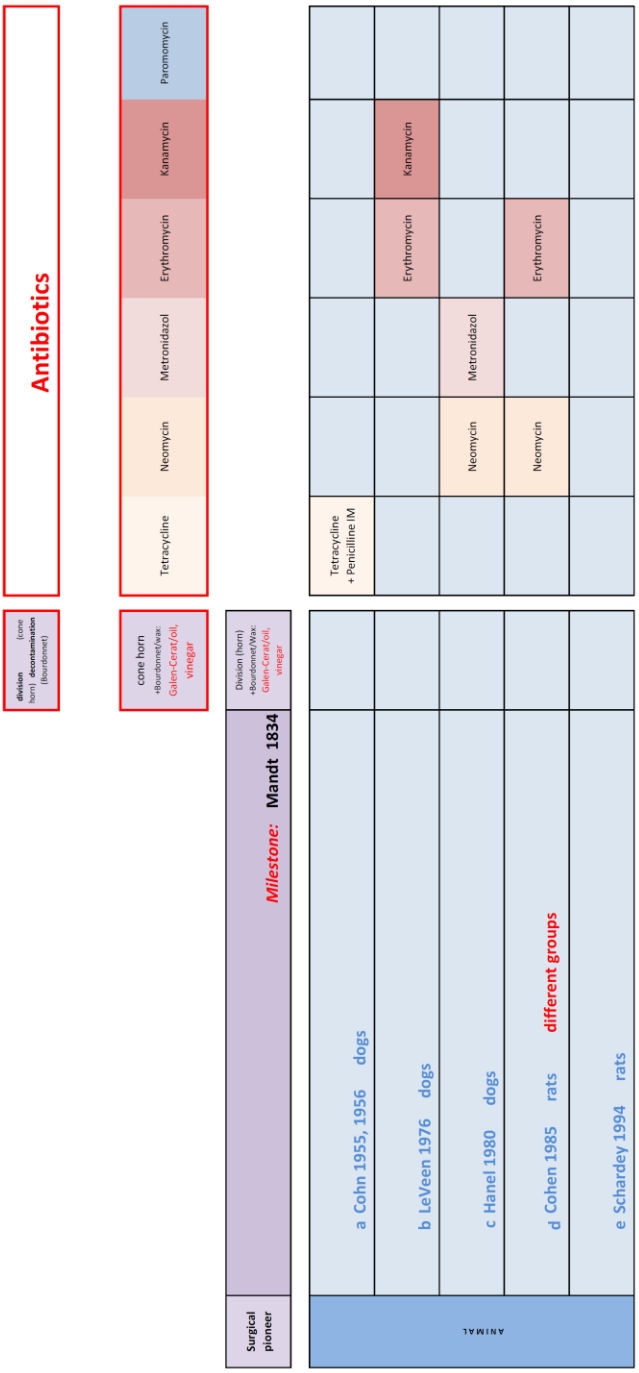


Table 5: Milestones and animal studies. [Cohn 1955 [53], Cohn 1956 [15], LeVeen 1976 [75], Hanel 1980 [80], Cohen 1985]54], Schardey 1994 [56]].

Also several animal studies [75, 53, 15, 80, 54, 56] investigated wheather use of antibiotics is helpful in reducing anastomotic leak.

Cohn [15] was one of the first, who investigated and published 1955 (together with Rives) his findings in antibiotic protection of colon anastomoses in dogs. In December 1954 he presented in Florida before the Southern Surgical Association three factors being responsible „for the danger of leakage at the suture line:

- the presence of solid feces and gas,
- the vulnerable circulation and
- the profuse bacterial flora.

He thought, „that antibacterial therapy should be continued after surgery and throughout the healing period", because *"simple dilution oft he feces, which also dilutes the antibiotic agent, permits active bacterial growth". … "He found a broad spectrum antibiotic even more effective. But this method attacks bacteria only after they have invaded viable tissues. Organisms in the lumen of the bowel … are almost, if not completely, unaffected"* [53]. In his study he used achromycin (a broad-spectrum antibiotic, belonging to the group of tetracyclines) and intramuscularly penicillin, begun at operation.

Over the time, patient studies [82, 83, 84, 85, 86] followed, investigating different antibiotics to reduce anastomotic leak in elective patients (one [84] without success in significant reduction in anastomotic leak; one [86] a meta-analysis of thirteen studies, with very different antibiotics) [4]. There were four published studies (three studies [72, 87, 57] and the underpowered SELECT-trial [73, 74] that were using PT/PG (PT: polymyxin/tobramycin; PG: polymyxin/gentamycin but no vancomycin) and amphotericin B (an antimycotic acting substance) for decontamination, all showing a slight reduction in anastomotic leak for elective patients [4].

Some unsuccessful studies with different antibiotics were performed (see Table 6, page 51)

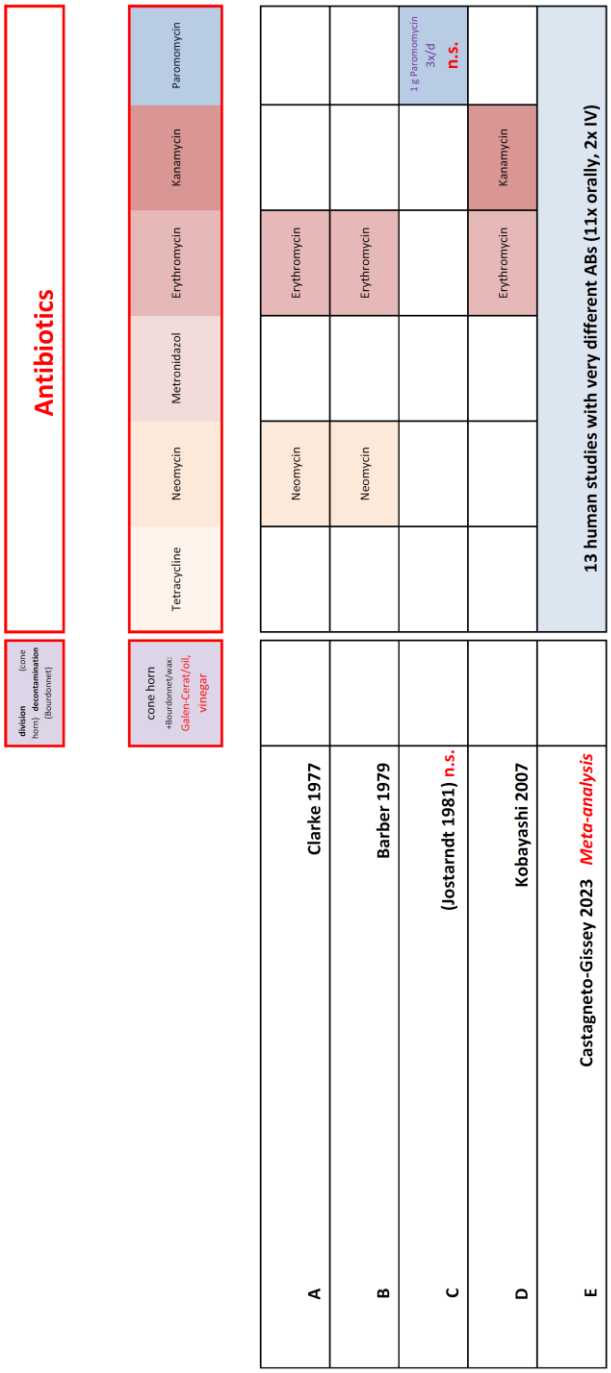


Table 6: Studies with different antibiotics. [Clarke 1977 [82], Barber 1979 [83], Kobayashi 2007 [85]], *a meta-analysis followed* [Castagneto-Gissey 2023 [86]].

In 1997, Hans Martin Schardey (Agatharied Academic Teaching Hospital of the Ludwig-Maximilians-University Munich, Germany) published his milestone trial [29], using PTV (PTV: polymyxin/tobramycin/vancomycin) and amphotericin B for locally effective acting decontamination in patients' gut with a remarkable reduction in anastomotic leak. Based on the excellent evidence, perioperative local decontamination was implemented at our surgical institution at the University of Graz; the occurrence of anastomotic leak with regard to this change was an aim of our trial. The next study by the group of Schardey [58] was cancelled after an interim analysis because of a death in the control group, not in the group using the investigation drug [4].

This settings and concepts were also investigated by a group of German researchers at the University of Dresden [59] for elective patients only, and it resulted in a reduction in anastomotic leak in the decontamination group. A human body exists in symbiosis with its microbiota, which can be defined as the ecosystem of the body. A microbial dysbiosis contributes [4] to "diseases" and consists of changes in the microbial metabolism; this influences the regulation of inflammation in the case of anastomotic leak, a change in the release of metabolites influences the gut barrier and wound healing after gut surgery.

It has been well known for a long time that microbiota can modulate the restitution during a postoperative period, not only by influencing the immune system but also by influencing metabolites [4] "Bacterial metabolites can be either degraded or absorbed depending on competitive microbes" [76] and "surgical injury . . . can shift the phenotype of a potentially pathogen from innocuous colonizer to invasive and virulent pathogen" [8].

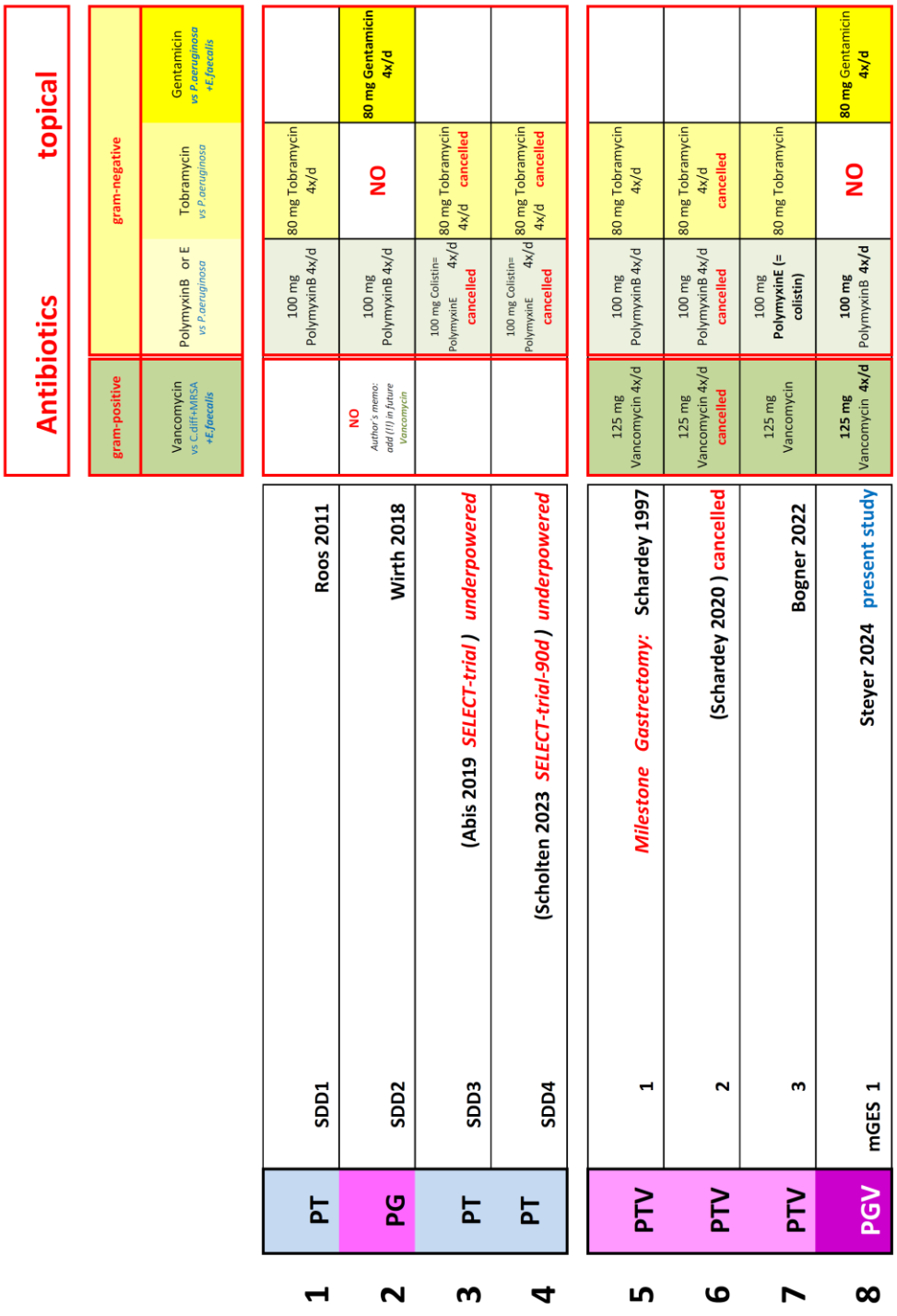


Table 7a: Studies with local acting antibiotics to reduce or prevent anastomotic leak.

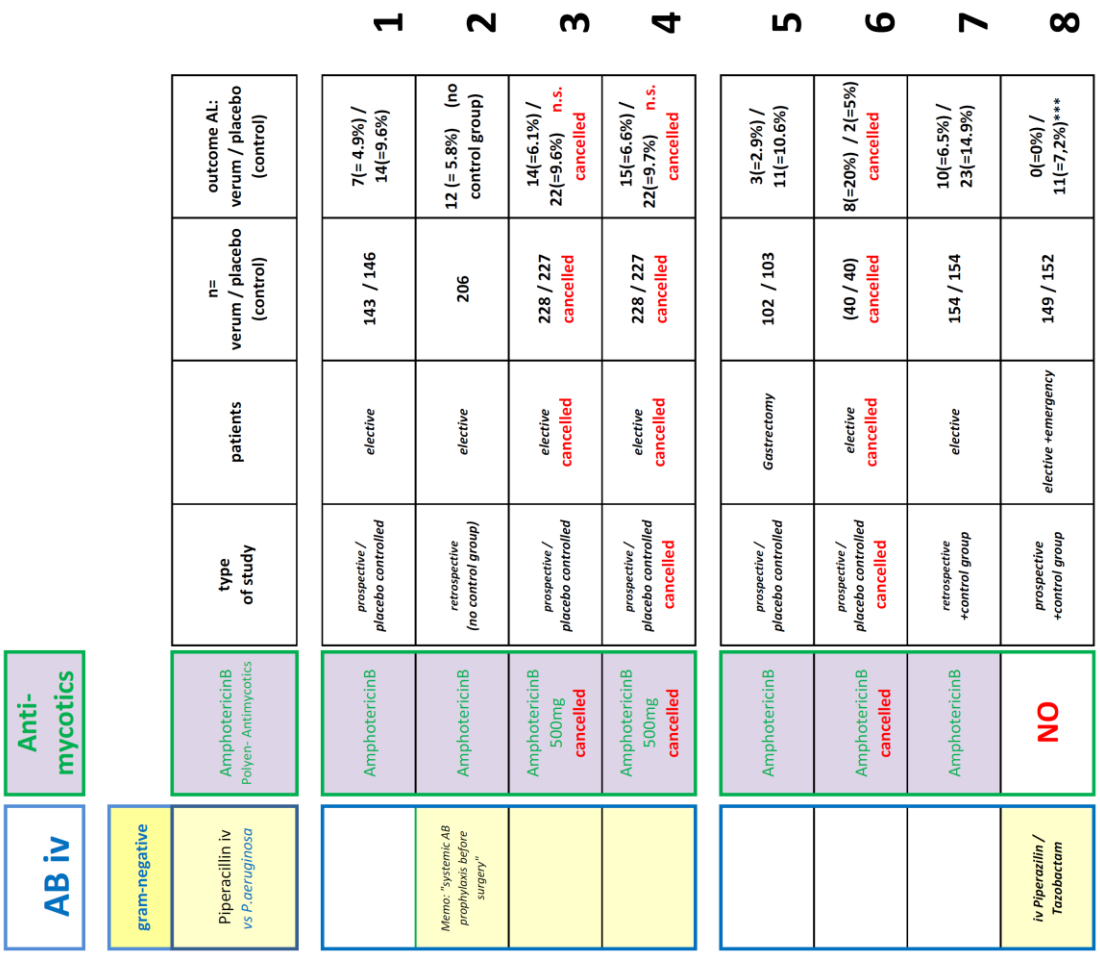


Table 7b: Results of studies with local acting antibiotics to reduce or prevent anastomotic leak. [Roos 2011 [72], Wirth 20018 [57], Abis 2019 [74], Scholten 2023 [87], Schardey 1997 [29], Schardey 2020 [58], Bogner 2022 [59], Steyer 2024 [4]].

The release of host stress factors during surgery can activate bacterial virulence genes. Until now, *P. aeruginosa* and *E. faecalis* have been identified and investigated [8, 28] to shift to a pathogen phenotype and act after gut surgery via the activation of matrix metallo protease-9,

followed by the degradation of collagen [4]. An “adequate anastomotic healing requires collagen deposition and remodeling through post-translational modification” [27].

To date, we have identified [4] four published studies [72, 73, 74, 87, 57] in the medical literature on anastomotic leak in patients who received an antibiotic mixture that is known under the name Selective Decontamination in Digestive tract (see chapter XX) and is helpful for avoiding respiratory infections in emergency care units, especially against *P. aeruginosa*. Selective Decontamination of the Digestive tract contains polymyxin B and tobramycin (an aminoglycoside antibiotic). In anastomotic patients, amphotericin B (a polyeneantimycotic agent) was always added to the mixture; this intervention results in a moderate reduction in anastomotic leak. Two trials had to be stopped after an interim analysis because those studies were statistically underpowered [4].

A modification of the agents for Selective Decontamination of the Digestive tract was used in three other trials [29, 58, 59]. Even here, one trial was stopped (because of an unexpected side effect in the control group); also, these trials resulted in a remarkable reduction in anastomotic leak [4]. Based on these, we designed our study in elective and emergency patients undergoing colon surgery (representing the routine cases in a university hospital). Therefore, we used P + G (P: polymyxin; G: gentamicin) for the decontamination of the Gram-negative bacteria (like *P. aeruginosa* and E. faecalis). For the decontamination of Gram-positive bacteria (like *C. difficile* and its toxins we added in our mGES (murine GutEpithelialShelter / muriner GedärmeEpithelSchutz) the well-investigated antibiotic vancomycin (but no antimycotic like amphotericin B), resulting in zero anastomotic leak (vs. eleven cases in the control group). The only difference concerning treatment in our two groups was the use of PGV in the treatment group because both groups received systemic piperacillin/tazobactam following the standard protocol in our clinic [88; 4].

11.6 Discussion of the Hypothesis of this Study

In 1987, Unertl [42] published his study on the *"prevention of colonization and respiratory infections in long-term ventilated patients by local antimicrobial prophylaxis".* He investigated a mixture of polymyxin B and gentamicin because *"gram-negative rods, especially Pseudomonas aeruginosa . . .. . . were found to be sensitive to at least one of the two agents, ... gentamicin covered more than 90% of the isolates of Enterobacteriaceae . . .. . ., but only 80% of the isolates of P. aeruginosa. Both agents exert a high and prolonged local activity when given orally, show no systemic toxicity as they are not absorbed through intact mucosa and are well tolerated. Cross-resistance between polymyxin B and other antimicrobial agents is uncommon, and amikacin could be expected to be a suitable substitute if resistance against gentamicin developed".* We decided in the protocol of our study to investigate this mixture (polymyxin B and gentamicin) combined with vancomycin, a glycopeptide antibiotic, covering Gram-positive microbes like *C. difficile*, MRSA and *E. faecalis*. We wanted to combat mainly *E. faecalis* and *P. aeruginosa* (because of their property to activate matrix metallo protease-9 and consecutively collagenase resulting in degradation of collagen at the anastomosis), and therefore we did not add amphotericin B [4].

Our study hypothesis, that the occurrence of anastomotic leak can be reduced through a locally acting unabsorbable antibiotic mixture, has been completely validated and we showed, indirectly, that matrix metallo protease-9-activating microbes are involved in the disturbed healing process of an anastomotic leak [4].

11.7 Discussion of the Intervention in this Study

However, by using the described intervention with a mixture of polymyxin B, gentamicin and vancomycin, we stopped anastomotic leak in our study completely compared to the control group. This combination of antibiotics was used for the first time in this study to prevent AL - and the game was worth the candle. A strength of this study is that the intervention worked beside the high rate of emergency surgery in our clinic. Future studies should be done and randomized [4].

We have been able to successfully combat nosocomial hospital microbes with reserve antibiotics. Until now, three main bacteria that cause anastomotic leak have been investigated: *P. aeruginosa*, *E. faecalis* and *B. subtilis*. The antibiotic mixture therefore includes polymyxin B (a polypeptide antibiotic) to fight Gram-negative bacteria like *P. aeruginosa*, and gentamycin (instead of tobramycin, fighting *P. aeruginosa*), an aminoglycoside antibiotic, to fight *P. aeruginosa* and *E. faecalis*, and, furthermore, vancomycin (a glycopeptide antibiotic) to fight Gram-positive bacteria like *E. faecalis*; our study did not include the antimycotic acting amphotericin B (a polyene antimycotic acting substance). *Bacillus subtilis* was incriminated to cause anastomotic leak [18] and is also fought by vancomycin. Patients additionally received a piperacillin i.v., a systemic antibiotic prophylaxis. Piperacillin must be administered parenterally, as after oral administration, only minimal absorption occurs. It is well known [88, 89] that, if the broad-spectrum penicillin piperacillin is given *"in combination with aminoglycoside antibiotics"* (like gentamicin in our study), *"additive and synergistic increases in activity can often be achieved against Enterobacteriaceae and especially against Pseudomonas aeruginosa"* [4].

The decontamination medication (mGES) was prepared by our institutional pharmacy.

"The enteral antimicrobial combination of colistin (polymyxinE), tobramycin and amphotericin B (or nystatin) complied with these criteria when it was first described, but the appearance of outbreaks and endemics due to MRSA and MRGNB in some cases makes it necessary to modify the original combination [51]". Gentamicin could now again be replaced by tobramicin in both settings, selective decontimination of the digestive tract [51] and mGES (=local selective decontamination of the digestive tract) [see chapter 11.11, "future studies"].

"most leaks can be managed without reexploration" [13]

11.8 Discussion of Results of this Study

It is very important, to recognize, that, *"today most leaks can be managed without reexploration and therefore leaking tissues cannot be examined directly* [13]". But we had to define an endpoint of the study.

There are two factors we had to consider:

1. the duration of the mean hospital stay after surgery (normally 5 to 7 days), and
2. the main occurence of anastomotic leak after surgery (normally 5 to 7 days),

and also the time needed for healing and to start cicatrice.

So we decided, to follow Rahbari [60], that re-do surgery required within 7 postoperative days to be the end of the observation time in this study.

Discussing the mechanism of action for anastomotic leak (see chapter 8.1.1, page 18), that ischemia would be a cause of leak, we look again at the study of Shakhsheer [21], who investigated in a rat model the influence of ischemia on anastomotic leak. Ligating the feeding blood vessels of a colorectal anastomosis he did not find evidence of ischemia two weaks later. But by this concept of the study only "evidence of enhanced neovascularity and angiogenesis was observed [13]", and "even if it were possible to examine intestinal tissues postoperatively" it is nearly impossible "to account for the time-dependent adaption of the … ativation of vascular endothelial growth factor … and blood vessel growth that occurs following anastomotic construction [90]".

In our study, ASA score did not significantly influence the leak rate; that is in consensus with the medical literature [91], and risk factors were not an aim of our investigation. The PGV intervention in our study showed that AL can be stopped successfully (or at least reduced remarkably), but this means that selectively killing bacteria like *P. aeruginosa* and E. faecalis with a topic-acting antibiotic mixture is also indirect proof that these killer microbes are strongly involved in the development of anastomotic leak [4].

A re-operation is always associated with different risks. A lifelong stoma can be a consequence, or increased mortality and morbidity [92].

11.9 Implication on Future Practice and Research

By using a new antibiotic mixture (mGES) for perioperative decontamination, it was possible to stop anastomotic leak in elective and emergency patients. Future (randomized) studies should investigate whether mGES has to be administered routinely to all patients or just in selected patients. In a systematic review and meta-analysis, a Canadian Study Group [93] *"detected no relation between the use of selective decontamination of the digestive tract . . . and the development of antimicrobial resistance in pathogens in patients in the intensive care units".*

Long-term effects like antimicrobial resistance should be investigated carefully. Also, the question remains if antimycotics (like Amphotericin) have to be added to the antimicrobial decontamination to avoid mycotic overgrowth [4].

11.10 Reducing Costs

It is not primarily a medical issue to reduce costs, but for anastomotic leak, it has been shown in some publications concerning anastomotic leak that decontamination can remarkably reduce the rate of re-operations and, in this way, healthcare costs [29, 59]. Some bookkeepers - from a patient's point of view - also take increasing costs into consideration and perform an *"economic evaluation from a societal perspective as a costeffectiveness and cost-utility analysis"* [73, 74]. Ultimately, and not surprisingly, number crunchers recently presented *"the first cost-analysis study of anastomotic leak after colorectal surgery"* [94] in colorful pictures, and "concluded" that *"the appearance of anastomotic leak generates a considerable increase in the consumption of health resources, mainly due to an increase in hospital stays"* and *"the more complex the anastomotic leak, the higher the cost associated with its treatment"* - cost analyses on this topic have already been completed and are therefore obviously not an aim of our investigation [4]. They found out, that analyses show, that 2 surgical operations (anastomosis and re-do surgery after an anastomotic leak) are not cheaper than 1 operation; do they take into consideration patient´s needs (and the feeling of

the surgeon in charge) or just money (gold), and is that worth to be called a “scientific” statistical evaluation)?

These bookkeepers are only interested in reducing costs, but they do not take into consideration which medical implications exist: *“each and every anastomotic leak … disrupts the implementation of adjuvant chemotherapy,“* (increases cancer recurrance), *“prolongs hospitalization, requires multiple intervention,”* and *“extends exposure to” (other) “antibiotics* [13]“ that can disturb the microbiota in totum, because they are not acting local and selective.

11.11 Coming Next (or “Future Studies”)?

Looking for Lifestyle-Related Diseases there is a lot to be done for next generations (or artificial intelligence) and it is a main task for the Medical University of Graz, Austria, Europe. We have to consider, to investigate conditions at *“anastomotic sites (i.e., nutrient depletion, loss of mucus, absence of competiting microbiota, presence of opioids, etc.) that trigger collagenolytic pathogens to express the appropriate phenotype necessary to cause clinically relevant anastomotic leak* [13]”. But *“the mere presence of collagenolytic bacteria at anastomotic sites alone is not sufficient to cause a leak; rather environmental cues .. are needed, that signal bacteria to express collagenase* [13]”, and *“collagenolytic bacteria are necessary” – “but alone not sufficient”* is the proper scientific conclusion from studies [13].

For future decisions helpful would be dose finding studies (with respect to BMI), in a double blind manner, conceptualized as a multicenter investigation. Maybe it is also better to change gentamycin for tobramycin, or leave the coast and find new oceans: stool transplant (instead of gut transplant), improve technical surgery, or oral polyphosphate (to suppress bacterial collagenase production in *P. aeruginosa* [77] or in *E. faecalis* [95] to prevent anastomotic leak).

It is necessary to evaluate, if an antimicrobial decontamination is needed to avoid mycotic overgrowth. Stool transfer can be an option (for elective patients), or …

Ultimately helpful is of course, just to gender the language, create cost analyses, or to create new rules, or even to pray.

But the author of this dissertation has a dream (in German of course, because he never ever dreamed in English):

If, in near future, we have to give students a medico-historical lession to tell them what an “anastomotic leak” per se is, because they do not learn and recognize it as a feared surgical complication and it does not longer “exist” in daily routine, this trial has had a positive effect on patient´s **Lifestyle-Related Diseases**,

and then the author wakes up in red tape. Glückwunsch.

12. References

1. Steyer GE. Also sprach Gerald. Aussprüche meines Enkels im Alter von 2 Jahren. Graz, 2024, *in print*.
2. Steyer GE. Darmanastomoseninsuffizienzprophylaxe. Meine Dissertation Nummer 3, Graz, MedUni, 2024, *in print*.
3. Pfeifer J, Steyer GE (eds). Mikrobiom. Reference book 2019. Overview of the current state of research. 1st edn, Medical Tribune, Vienna, pp 24-27 ISBN 978-3-200-06207-8.
4. Steyer GE, Puchinger M, Pfeifer J. Successful Clinical Avoidance of Colorectal Anastomotic Leakage through Local Decontamination. Antibiotics (Basel). 2024 Jan 15;13(1):79. doi: 10.3390/antibiotics13010079. PMID: 38247638; PMCID: PMC10812415.
5. Boatman S, Kohn J, Jahansouz C. The Influence of the Microbiome on Anastomotic Leak. Clin Colon Rectal Surg. 2023 Jan 17;36(2):127-132. doi: 10.1055/s-0043-1760718. PMID: 36844711; PMCID: PMC9946719.
6. Kinross JM. Microbiome is the key to preventing anastomotic leak in colorectal surgery. Gut. 2024 Jan 5;73(2):212-214. doi: 10.1136/gutjnl-2022-329281. PMID: 37197907.
7. Hajjar R, Gonzalez E, Fragoso G, Oliero M, Alaoui AA, Calvé A, Vennin Rendos H, Djediai S, Cuisiniere T, Laplante P, Gerkins C, Ajayi AS, Diop K, Taleb N, Thérien S, Schampaert F, Alratrout H, Dagbert F, Loungnarath R, Sebajang H, Schwenter F, Wassef R, Ratelle R, Debroux E, Cailhier JF, Routy B, Annabi B, Brereton NJB, Richard C, Santos MM. Gut microbiota influence anastomotic healing in colorectal cancer surgery through modulation of mucosal proinflammatory cytokines. Gut. 2023 Jun;72(6):1143-1154. doi: 10.1136/gutjnl-2022-328389. Epub 2022 Dec 30. PMID: 36585238.
8. Shogan BD, Smith DP, Christley S, Gilbert JA, Zaborina O, Alverdy JC. Intestinal anastomotic injury alters spatially defined microbiome composition and function. Microbiome. 2014 Sep 15;2:35. doi: 10.1186/2049-2618-2-35. PMID: 25250176; PMCID: PMC4171717.

9. Slieker JC, Daams F, Mulder IM, Jeekel J, Lange JF. Systematic review of the technique of colorectal anastomosis. JAMA Surg. 2013 Feb;148(2):190-201. doi: 10.1001/2013.jamasurg.33. PMID: 23426599.
10. Kitaguchi D, Ito M. Optimal anastomotic technique in rectal surgery to prevent anastomotic leakage. Ann Coloproctol. 2023 Apr;39(2):97-105. doi: 10.3393/ac.2022.00787.0112. Epub 2023 Jan 3. PMID: 36593572; PMCID: PMC10169556.
11. Valmasoni M, Capovilla G, Pierobon ES, Moletta L, Provenzano L, Costantini M, Salvador R, Merigliano S. A Technical Modification to the Circular Stapling Anastomosis Technique During Minimally Invasive Ivor Lewis Procedure. J Laparoendosc Adv Surg Tech A. 2019 Dec;29(12):1585-1591. doi: 10.1089/lap.2019.0461. Epub 2019 Oct 3. PMID: 31580751.
12. Zhuo C, Liang L, Ying M, Li Q, Li D, Li Y, Peng J, Huang L, Cai S, Li X. Laparoscopic Low Anterior Resection and Eversion Technique Combined With a Nondog Ear Anastomosis for Mid- and Distal Rectal Neoplasms: A Preliminary and Feasibility Study. Medicine (Baltimore). 2015 Dec;94(50):e2285. doi: 10.1097/MD.0000000000002285. PMID: 26683958; PMCID: PMC5058930.
13. Alverdy JC, Schardey HM. Anastomotic Leak: Toward an Understanding of Its Root Causes. J Gastrointest Surg. 2021 Nov;25(11):2966-2975. doi: 10.1007/s11605-021-05048-4. Epub 2021 Jun 7. PMID: 34100248; PMCID: PMC8815793.
14. Williamson AJ, Alverdy JC. Influence of the Microbiome on Anastomotic Leak. Clin Colon Rectal Surg. 2021 Nov 23;34(6):439-446. doi: 10.1055/s-0041-1735276. PMID: 34853567; PMCID: PMC8610638.
15. Cohn I Jr, Rives JD. Antibiotic protection of colon anastomoses. Ann Surg. 1955 May;141(5):707-17. doi: 10.1097/00000658-195505000-00016. PMID: 14362409; PMCID: PMC1609892.
16. Christley S, Shogan B, Levine Z, Koo H, Guyton K, Owens S, Gilbert J, Zaborina O, Alverdy JC. Comparative genetics of Enterococcus faecalis intestinal tissue isolates before and after surgery in a rat model of colon anastomosis. PLoS One. 2020 Apr 28;15(4):e0232165. doi: 10.1371/journal.pone.0232165. PMID: 32343730; PMCID: PMC7188289.

17. Anderson DI, Keskey R, Ackerman MT, Zaborina O, Hyman N, Alverdy JC, Shogan BD. *Enterococcus faecalis* Is Associated with Anastomotic Leak in Patients Undergoing Colorectal Surgery. Surg Infect (Larchmt). 2021 Dec;22(10):1047-1051. doi: 10.1089/sur.2021.147. Epub 2021 Jul 13. PMID: 34255574; PMCID: PMC8851212.
18. van Praagh JB, Luo JN, Zaborina O, Alverdy JC. Involvement of the Commensal Organism *Bacillus subtilis* in the Pathogenesis of Anastomotic Leak. Surg Infect (Larchmt). 2020 Dec;21(10):865-870. doi: 10.1089/sur.2019.345. Epub 2020 Apr 20. PMID: 32310731; PMCID: PMC7703376.
19. Fink D, Romanowski K, Valuckaite V, Babrowski T, Kim M, Matthews JB, Liu D, Zaborina O, Alverdy JC. Pseudomonas aeruginosa potentiates the lethal effect of intestinal ischemia-reperfusion injury: the role of in vivo virulence activation. J Trauma. 2011 Dec;71(6):1575-82. doi: 10.1097/TA.0b013e31821cb7e5. PMID: 22002612; PMCID: PMC3245766.
20. Hoeppner J, Crnogorac V, Hopt UT, Weiser HF. The pig as an experimental model for colonic healing. Study of leakage and ischemia in colonic anastomosis. J Invest Surg. 2009 Jul-Aug;22(4):281-5. doi: 10.1080/08941930903040098. PMID: 19842904.
21. Shakhsheer BA, Lec B, Zaborin A, Guyton K, Defnet AM, Bagrodia N, Kandel JJ, Zaborina O, Hernandez SL, Alverdy J. Lack of evidence for tissue hypoxia as a contributing factor in anastomotic leak following colon anastomosis and segmental devascularization in rats. Int J Colorectal Dis. 2017 Apr;32(4):539-547. doi: 10.1007/s00384-016-2737-9. Epub 2016 Dec 20. PMID: 27999936.
22. Shogan BD, An GC, Schardey HM, Matthews JB, Umanskiy K, Fleshman JW Jr, Hoeppner J, Fry DE, Garcia-Granereo E, Jeekel H, van Goor H, Dellinger EP, Konda V, Gilbert JA, Auner GW, Alverdy JC. Proceedings of the first international summit on intestinal anastomotic leak, Chicago, Illinois, October 4-5, 2012. Surg Infect (Larchmt). 2014 Oct;15(5):479-89. doi: 10.1089/sur.2013.114. Epub 2014 Sep 12. PMID: 25215465; PMCID: PMC4382716.
23. Wright CD, Kucharczuk JC, O'Brien SM, Grab JD, Allen MS; Society of Thoracic Surgeons General Thoracic Surgery Database. Predictors of major morbidity and mortality after esophagectomy for esophageal cancer: a Society of Thoracic

Surgeons General Thoracic Surgery Database risk adjustment model. J Thorac Cardiovasc Surg. 2009 Mar;137(3):587-95; discussion 596. doi: 10.1016/j.jtcvs.2008.11.042. Erratum in: J Thorac Cardiovasc Surg. 2009 Jun;137(6):1581. PMID: 19258071.

24. Kube R, Mroczkowski P, Steinert R, Sahm M, Schmidt U, Gastinger I, Lippert H. Anastomoseninsuffizienzen nach Kolonkarzinomresektionen : Multiple Analyse der Risikofaktoren. Chirurg. 2009 Dec;80(12):1153-9. doi: 10.1007/s00104-009-1725-9. PMID: 19533064.
25. Schardey HM. Pathogenese und Prävention der Nahtinsuffizienz in der onkologischen Kolonchirurgie. In: Benz S, Grützmann R, Stinner B. (eds) Chirurgie des Kolonkarzinoms, Springer, Berlin, 2021, pp 365-381. ISBN 978-3-662-60452-6.
26. Lederer AK, Pisarski P, Kousoulas L, Fichtner-Feigl S, Hess C, Huber R. Postoperative changes of the microbiome: are surgical complications related to the gut flora? A systematic review. BMC Surg. 2017 Dec 4;17(1):125. doi: 10.1186/s12893-017-0325-8. PMID: 29202875; PMCID: PMC5715992.
27. Chang J, Guyton K. A Pathologic Microbiome Impacts Post-Operative Anastomotic Healing. Surg Infect (Larchmt). 2023 Apr;24(3):238-244. doi: 10.1089/sur.2023.003. PMID: 37010973.
28. Olivas AD, Shogan BD, Valuckaite V, Zaborin A, Belogortseva N, Musch M, Meyer F, Trimble WL, An G, Gilbert J, Zaborina O, Alverdy JC. Intestinal tissues induce an SNP mutation in Pseudomonas aeruginosa that enhances its virulence: possible role in anastomotic leak. PLoS One. 2012;7(8):e44326. doi: 10.1371/journal.pone.0044326. Epub 2012 Aug 31. PMID: 22952955; PMCID: PMC3432121.
29. Schardey HM, Joosten U, Finke U, Schauer R, Staubach KH, Exner H, Schildberg FW. Kostensenkung durch Dekontamination zur Prävention der Nahtinsuffizienz nach Gastrektomie. Chirurg. 1997 Apr;68(4):416-24. doi: 10.1007/s001040050207. PMID: 9206638.
30. Lee DS, Ryu JA, Chung CR, Yang J, Jeon K, Suh GY, Lee WY, Park CM. Risk factors for acquisition of multidrug-resistant bacteria in patients with anastomotic leakage after colorectal cancer surgery. Int J Colorectal Dis. 2015 Apr;30(4):497-504. doi: 10.1007/s00384-015-2161-6. Epub 2015 Mar 4. PMID: 25735927.

31. Zhang JW, Du P, Gao J, Yang BR, Fang WJ, Ying CM. Preoperative probiotics decrease postoperative infectious complications of colorectal cancer. Am J Med Sci. 2012 Mar;343(3):199-205. doi: 10.1097/MAJ.0b013e31823aace6. PMID: 22197980.
32. Chang J, Guyton K. A Pathologic Microbiome Impacts Post-Operative Anastomotic Healing. Surg Infect (Larchmt). 2023 Apr;24(3):238-244. doi: 10.1089/sur.2023.003. PMID: 37010973.
33. Stoutenbeek CP, Van Saene HK, Miranda DR, Zandstra DF. A new technique of infection prevention in the intensive care unit by selective decontamination of the digestive tract. Acta Anaesthesiol Belg. 1983 Sep;34(3):209-21. PMID: 6316704.
34. Carlet J. Selective digestive decontamination: We must make an international decision on this 40-year old controversy. Intensive Care Med. 2024 Feb;50(2):272-274. doi: 10.1007/s00134-023-07289-0. Epub 2023 Dec 20. PMID: 38117320.
35. Bonten M. Selective Decontamination of the Digestive Tract: An Answer at Last? JAMA. 2022 Dec 20;328(23):2310-2311. doi: 10.1001/jama.2022.18623. PMID: 36378275.
36. SuDDICU Investigators for the Australian and New Zealand Intensive Care Society Clinical Trials Group; Myburgh JA, Seppelt IM, Goodman F, Billot L, Correa M, Davis JS, Gordon AC, Hammond NE, Iredell J, Li Q, Micallef S, Miller J, Mysore J, Taylor C, Young PJ, Cuthbertson BH, Finfer SR. Effect of Selective Decontamination of the Digestive Tract on Hospital Mortality in Critically Ill Patients Receiving Mechanical Ventilation: A Randomized Clinical Trial. JAMA. 2022 Nov 15;328(19):1911-1921. doi: 10.1001/jama.2022.17927. PMID: 36286097; PMCID: PMC9607966.
37. Hammond NE, Myburgh J, Seppelt I, Garside T, Vlok R, Mahendran S, Adigbli D, Finfer S, Gao Y, Goodman F, Guyatt G, Santos JA, Venkatesh B, Yao L, Di Tanna GL, Delaney A. Association Between Selective Decontamination of the Digestive Tract and In-Hospital Mortality in Intensive Care Unit Patients Receiving Mechanical Ventilation: A Systematic Review and Meta-analysis. JAMA. 2022 Nov 15;328(19):1922-1934. doi: 10.1001/jama.2022.19709. PMID: 36286098; PMCID: PMC9607997.
38. Johanson WG, Pierce AK, Sanford JP. Changing pharyngeal bacterial flora of hospitalized patients. Emergence of gram-negative bacilli. N Engl J Med. 1969 Nov 20;281(21):1137-40. doi: 10.1056/NEJM196911202812101. PMID: 4899868.

39. van der Waaij D, Berghuis-de Vries JM, Lekkerkerk-van der Wees. Colonization resistance of the digestive tract and the spread of bacteria to the lymphatic organs in mice. J Hyg (Lond). 1972 Jun;70(2):335-42. doi: 10.1017/s0022172400022385. PMID: 4626276; PMCID: PMC2130037.
40. Bodey GP, Feld R, Burgess MA. Beta-lactam antibiotics alone or in combination with gentamicin for therapy of gram-negative bacillary infections in neutropenic patients. Am J Med Sci. 1976 Mar-Apr;271(2):179-86. doi: 10.1097/00000441-197603000-00006. PMID: 773180.
41. van der Voort PHJ, van Saene HKF (eds). Selective Digestive Tract Decontamination in Intensive Care Medicine: a Practical Guide to Controlling Infection. Springer-Verlag Milan 2008, 200 pp, ISBN 978-88-470-0652-2.
42. Unertl K, Ruckdeschel G, Selbmann HK, Jensen U, Forst H, Lenhart FP, Peter K. Prevention of colonization and respiratory infections in long-term ventilated patients by local antimicrobial prophylaxis. Intensive Care Med. 1987;13(2):106-13. doi: 10.1007/BF00254795. PMID: 3033040.
43. Silvestri L, de la Cal MA, van Saene HK. Selective decontamination of the digestive tract: the mechanism of action is control of gut overgrowth. Intensive Care Med. 2012 Nov;38(11):1738-50. doi: 10.1007/s00134-012-2690-1. Epub 2012 Sep 22. PMID: 23001446.
44. Rodríguez-Gascón A, Lloréns-Villar Y, Solinís MÁ, Barrasa H, Canut-Blasco A. Does selective digestive decontamination (SDD) increase antibiotic resistance? Long-term comparison of two intensive care units (with and without SDD) of the same tertiary hospital. Eur J Clin Microbiol Infect Dis. 2024 Mar 9. doi: 10.1007/s10096-024-04792-0. Epub ahead of print. PMID: 38460030.
45. Krueger WA, Heininger A, Unertl KE. Selektive Darmdekontamination in der Intensivmedizin. Grundlagen und aktuelle Bewertung. Anaesthesist. 2003 Feb;52(2):142-52. German. doi: 10.1007/s00101-002-0443-1. PMID: 12624700.
46. Craven DE, Connolly MG Jr, Lichtenberg DA, Primeau PJ, McCabe WR. Contamination of mechanical ventilators with tubing changes every 24 or 48 hours. N Engl J Med. 1982 Jun 24;306(25):1505-9. doi: 10.1056/NEJM198206243062501. PMID: 7043269.

47. Schardey HM, Rogers SK, Schopf SK, von Ahnen T, Wirth U. Sind Darmbakterien an der Entstehung der Anastomoseninsuffizienz beteiligt? / Are gut bacteria associated with the development of anastomotic leaks? Coloproctology. 2017; 40(S1), 1–8. doi:10.1007/s00053-017-0145-4 (Online) and (Print) 39(2):94-100. doi: 10.1007/s00053-016-0136-x.
48. Plantinga NL, Bonten MJ. Selective decontamination and antibiotic resistance in ICUs. Crit Care. 2015 Jun 24;19(1):259. doi: 10.1186/s13054-015-0967-9. PMID: 26104045; PMCID: PMC4479224.
49. Ebner W, Kropec-Hübner A, Daschner FD. Bacterial resistance and overgrowth due to selective decontamination of the digestive tract. Eur J Clin Microbiol Infect Dis. 2000 Apr;19(4):243-7. doi: 10.1007/s100960050470. PMID: 10834811.
50. Oostdijk EA, de Smet AM, Blok HE, Thieme Groen ES, van Asselt GJ, Benus RF, Bernards SA, Frénay IH, Jansz AR, de Jongh BM, Kaan JA, Leverstein-van Hall MA, Mascini EM, Pauw W, Sturm PD, Thijsen SF, Kluytmans JA, Bonten MJ. Ecological effects of selective decontamination on resistant gram-negative bacterial colonization. Am J Respir Crit Care Med. 2010 Mar 1;181(5):452-7. doi: 10.1164/rccm.200908-1210OC. Epub 2009 Dec 3. PMID: 19965807.
51. Tejerina-Álvarez EE, de la Cal López MÁ. Selective decontamination of the digestive tract: concept and application. Med Intensiva (Engl Ed). 2023 Oct;47(10):603-615. doi: 10.1016/j.medine.2023.05.018. PMID: 37858367.
52. von Mandt MW. Resection eines Theiles des Intestini recti. In: Rust JN (Hg.): Magazin für die gesammte Heilkunde, mit besonderer Rücksicht auf das allgemeine Sanitäts-Wesen im Königl. Preussischen Staate. Berlin NF 18, 1834, p. 11-35.
53. Cohn I Jr, Rives JD. Antibiotic protection of colon anastomoses. Ann Surg. 1955 May;141(5):707-17. doi: 10.1097/00000658-195505000-00016. PMID: 14362409; PMCID: PMC1609892.
54. Cohen SR, Cornell CN, Collins MH, Sell JE, Blanc WA, Altman RP. Healing of ischemic colonic anastomoses in the rat: role of antibiotic preparation. Surgery. 1985 Apr;97(4):443-6. PMID: 3983820.
55. Stoutenbeek CP, van Saene HK, Miranda DR, Zandstra DF, Langrehr D. The effect of oropharyngeal decontamination using topical nonabsorbable antibiotics on the incidence of nosocomial respiratory tract infections in multiple trauma patients. J

Trauma. 1987 Apr;27(4):357-64. doi: 10.1097/00005373-198704000-00003. PMID: 3033259.

56. Schardey HM, Kamps T, Rau HG, Gatermann S, Baretton G, Schildberg FW. Bacteria: a major pathogenic factor for anastomotic insufficiency. Antimicrob Agents Chemother. 1994 Nov;38(11):2564-7. doi: 10.1128/AAC.38.11.2564. PMID: 7872748; PMCID: PMC188242.
57. Wirth U, Rogers S, Haubensak K, Schopf S, von Ahnen T, Schardey HM. Local antibiotic decontamination to prevent anastomotic leakage short-term outcome in rectal cancer surgery. Int J Colorectal Dis. 2018 Jan;33(1):53-60. doi: 10.1007/s00384-017-2933-2. Epub 2017 Nov 8. PMID: 29119289.
58. Schardey HM, Wirth U, Strauss T, Kasparek MS, Schneider D, Jauch KW. Prevention of anastomotic leak in rectal cancer surgery with local antibiotic decontamination: a prospective, randomized, double-blind, placebo-controlled single center trial. Int J Colorectal Dis. 2020 May;35(5):847-857. doi: 10.1007/s00384-020-03544-8. Epub 2020 Feb 27. PMID: 32103326.
59. Bogner A, Stracke M, Bork U, Wolk S, Pecqueux M, Kaden S, Distler M, Kahlert C, Weitz J, Welsch T, Fritzmann J. Selective decontamination of the digestive tract in colorectal surgery reduces anastomotic leakage and costs: a propensity score analysis. Langenbecks Arch Surg. 2022 Sep;407(6):2441-2452. doi: 10.1007/s00423-022-02540-6. Epub 2022 May 13. PMID: 35551468; PMCID: PMC9468075.
60. Rahbari NN, Weitz J, Hohenberger W, Heald RJ, Moran B, Ulrich A, Holm T, Wong WD, Tiret E, Moriya Y, Laurberg S, den Dulk M, van de Velde C, Büchler MW. Definition and grading of anastomotic leakage following anterior resection of the rectum: a proposal by the International Study Group of Rectal Cancer. Surgery. 2010 Mar;147(3):339-51. doi: 10.1016/j.surg.2009.10.012. Epub 2009 Dec 11. PMID: 20004450.
61. Bruce J, Krukowski ZH, Al-Khairy G, Russell EM, Park KG. Systematic review of the definition and measurement of anastomotic leak after gastrointestinal surgery. Br J Surg. 2001 Sep;88(9):1157-68. doi: 10.1046/j.0007-1323.2001.01829.x. PMID: 11531861.
62. Maeda H, Okamoto K, Namikawa T, Akimori T, Kamioka N, Shiga M, Dabanaka K, Hanazaki K, Kobayashi M. Rarity of late anastomotic leakage after low anterior

resection of the rectum. Int J Colorectal Dis. 2015 Jun;30(6):831-4. doi: 10.1007/s00384-015-2207-9. Epub 2015 Apr 11. PMID: 25862318.

63. Floodeen H, Hallböök O, Rutegård J, Sjödahl R, Matthiessen P. Early and late symptomatic anastomotic leakage following low anterior resection of the rectum for cancer: are they different entities? Colorectal Dis. 2013 Mar;15(3):334-40. doi: 10.1111/j.1463-1318.2012.03195.x. PMID: 22889325.

64. Chiarello MM, Fransvea P, Cariati M, Adams NJ, Bianchi V, Brisinda G. Anastomotic leakage in colorectal cancer surgery. Surg Oncol. 2022 Mar;40:101708. doi: 10.1016/j.suronc.2022.101708. Epub 2022 Jan 24. PMID: 35092916.

65. Li YW, Lian P, Huang B, Zheng HT, Wang MH, Gu WL, Li XX, Xu Y, Cai SJ. Very Early Colorectal Anastomotic Leakage within 5 Post-operative Days: a More Severe Subtype Needs Relaparatomy. Sci Rep. 2017 Jan 13;7:39936. doi: 10.1038/srep39936. PMID: 28084305; PMCID: PMC523396.

66. Lim SB, Yu CS, Kim CW, Yoon YS, Park IJ, Kim JC. Late anastomotic leakage after low anterior resection in rectal cancer patients: clinical characteristics and predisposing factors. Colorectal Dis. 2016 Apr;18(4):O135-40. doi: 10.1111/codi.13300. PMID: 26888300.

67. Leahy J, Schoetz D, Marcello P, Read T, Hall J, Roberts P, Ricciardi R. What is the risk of clinical anastomotic leak in the diverted colorectal anastomosis? J Gastrointest Surg. 2014 Oct;18(10):1812-6. doi: 10.1007/s11605-014-2588-z. Epub 2014 Jul 15. PMID: 25022256.

68. Morks AN, Ploeg RJ, Sijbrand Hofker H, Wiggers T, Havenga K. Late anastomotic leakage in colorectal surgery: a significant problem. Colorectal Dis. 2013 May;15(5):e271-5. doi: 10.1111/codi.12167. PMID: 23398601.

69. Hyman N, Manchester TL, Osler T, Burns B, Cataldo PA. Anastomotic leaks after intestinal anastomosis: it's later than you think. Ann Surg. 2007 Feb;245(2):254-8. doi: 10.1097/01.sla.0000225083.27182.85. PMID: 17245179; PMCID: PMC1876987.

70. Park JS, Huh JW, Park YA, Cho YB, Yun SH, Kim HC, Lee WY. Risk Factors of Anastomotic Leakage and Long-Term Survival After Colorectal Surgery. Medicine (Baltimore). 2016 Feb;95(8):e2890. doi: 10.1097/MD.0000000000002890. PMID: 26937928; PMCID: PMC4779025.

71. van Saene, H., Silvestri, L., Bams, J., van der Voort, P., Zandstra, D.

Selective Decontamination of the Digestive Tract: Use in Liver Transplantation is Evidence Based. Crit Care Med. 2003;31(5):1600-1601. doi:10.1097/01.CCM.0000065673.52359.A3.

72. Roos D, Dijksman LM, Oudemans-van Straaten HM, de Wit LT, Gouma DJ, Gerhards MF. Randomized clinical trial of perioperative selective decontamination of the digestive tract versus placebo in elective gastrointestinal surgery. Br J Surg. 2011 Oct;98(10):1365-72. doi: 10.1002/bjs.7631. Epub 2011 Jul 12. PMID: 21751181.
73. Abis GS, Oosterling SJ, Stockmann HB, van der Bij GJ, van Egmond M, Vandenbroucke-Grauls CM, Bonjer HJ. Perioperative selective decontamination of the digestive tract and standard antibiotic prophylaxis versus standard antibiotic prophylaxis alone in elective colorectal cancer patients. Dan Med J. 2014 Apr;61(4):A4695. PMID: 24814583.
74. Abis GSA, Stockmann HBAC, Bonjer HJ, van Veenendaal N, van Doorn-Schepens MLM, Budding AE, Wilschut JA, van Egmond M, Oosterling SJ; SELECT trial study group. Randomized clinical trial of selective decontamination of the digestive tract in elective colorectal cancer surgery (SELECT trial). Br J Surg. 2019 Mar;106(4):355-363. doi: 10.1002/bjs.11117. Epub 2019 Feb 25. PMID: 30802304.
75. Le Veen HH, Wapnick S, Falk G, Olivas O, Bhat D, Gaurdre M, Patel M. Effects of prophylactic antibiotics on colonic healing. Am J Surg. 1976 Jan;131(1):47-53. doi: 10.1016/0002-9610(76)90419-0. PMID: 1247153.
76. Alverdy JC, Hyoju SK, Weigerinck M, Gilbert JA. The gut microbiome and the mechanism of surgical infection. Br J Surg. 2017 Jan;104(2):e14-e23. doi: 10.1002/bjs.10405. PMID: 28121030; PMCID: PMC7938814.
77. Hyoju SK, Klabbers RE, Aaron M, Krezalek MA, Zaborin A, Wiegerinck M, Hyman NH, Zaborina O, Van Goor H, Alverdy JC. Oral Polyphosphate Suppresses Bacterial Collagenase Production and Prevents Anastomotic Leak Due to Serratia marcescens and Pseudomonas aeruginosa. Ann Surg. 2018 Jun;267(6):1112-1118. doi: 10.1097/SLA.0000000000002167. PMID: 28166091; PMCID: PMC5540820.
78. Škrabec CG, Carné AV, Pérez MC, Corral J, Pujol AF, Cuadrado M, Troya J, Ibáñez JJ, Parés D. Early and late anastomotic leak after colorectal surgery: A systematic review of the literature. Cir Esp (Engl Ed). 2023 Jan;101(1):3-11. doi: 10.1016/j.cireng.2022.07.011. Epub 2022 Jul 23. PMID: 35882311.

79. Marcellinaro R, Grieco M, Spoletini D, Troiano R, Avella P, Brachini G, Mingoli A, Carlini M. How to reduce the colorectal anastomotic leakage? The MIRACLe protocol experience in a cohort in a single high-volume centre. Updates Surg. 2023 Sep;75(6):1559-1567. doi: 10.1007/s13304-023-01588-3. Epub 2023 Jul 15. PMID: 37452926.
80. Guyton K, Alverdy JC (2017) The gut microbiota and gastrointestinal surgery. Nat Rev Gastroenterol Hepatol 14(1):43–54.
81. Hanel KC, King DW, McAllister ET, Reiss-Levy E. Single-dose parenteral antibiotics as prophylaxis against wound infections in colonic operations. Dis Colon Rectum. 1980 Mar;23(2):98-101. doi: 10.1007/BF02587603. PMID: 7379664.
82. Clarke JS, Condon RE, Bartlett JG, Gorbach SL, Nichols RL, Ochi S. Preoperative oral antibiotics reduce septic complications of colon operations: results of prospective, randomized, double-blind clinical study. Ann Surg. 1977 Sep;186(3):251-9. doi: 10.1097/00000658-197709000-00003. PMID: 889372; PMCID: PMC1396345.
83. Barber MS, Hirschberg BC, Rice CL, Atkins CC. Parenteral antibiotics in elective colon surgery? A prospective, controlled clinical study. Surgery. 1979 Jul;86(1):23-9. PMID: 377539.
84. Jostarndt L, Thiede A, Sonntag HG, Hamelmann H. Die systemische Antibioticumprophylaxe in der elektiven Colonchirurgie. Ergebnisse einer kontrollierten Studie. Chirurg. 1981 Jun;52(6):398-402. German. PMID: 6265162.
85. Kobayashi M, Mohri Y, Tonouchi H, Miki C, Nakai K, Kusunoki M; Mie Surgical Infection Research Group. Randomized clinical trial comparing intravenous antimicrobial prophylaxis alone with oral and intravenous antimicrobial prophylaxis for the prevention of a surgical site infection in colorectal cancer surgery. Surg Today. 2007;37(5):383-8. doi: 10.1007/s00595-006-3410-7. Epub 2007 Apr 30. PMID: 17468819.
86. Castagneto-Gissey L, Russo MF, Casella-Mariolo J, Serao A, Marcellinaro R, D'Andrea V, Carlini M, Casella G. The Role of Antibiotic Prophylaxis in Anastomotic Leak Prevention during Elective Colorectal Surgery: Systematic Review and Meta-Analysis of Randomized Controlled Trials. Antibiotics (Basel). 2023 Feb 16;12(2):397. doi: 10.3390/antibiotics12020397. PMID: 36830306; PMCID: PMC9951989.

87. Scholten J, Reuvers JRD, Stockmann HBAC, van Stralen KJ, van Egmond M, Bonjer HJ, Kazemier G, Abis GSA, Oosterling SJ; SELECT study group. Selective Decontamination with Oral Antibiotics in Colorectal Surgery: 90-day Reintervention Rates and Long-term Oncological Follow-up. J Gastrointest Surg. 2023 Jul 5. doi: 10.1007/s11605-023-05746-1. Epub ahead of print. PMID: 37407901.
88. Holmes B, Richards DM, Brogden RN, Heel RC. Piperacillin. A review of its antibacterial activity, pharmacokinetic properties and therapeutic use. Drugs. 1984 Nov;28(5):375-425. doi: 10.2165/00003495-198428050-00002. PMID: 6391888.
89. Piperacillin – ein neues Breitbandspekrtum-Penicillin. Z Chemotherapie (from 2015 Z Infektionstherapie) 5:1980.
90. Morgan A, Zheng A, Linden KM, Zhang P, Brown SA, Carpenter JP, Spitz FR, Kwiatt ME. Locally Transplanted Adipose Stem Cells Reduce Anastomotic Leaks in Ischemic Colorectal Anastomoses: A Rat Model. Dis Colon Rectum. 2020 Jul;63(7):955-964. doi: 10.1097/DCR.0000000000001667. PMID: 32168095.
91. Biondo S, Parés D, Kreisler E, Ragué JM, Fraccalvieri D, Ruiz AG, Jaurrieta E. Anastomotic dehiscence after resection and primary anastomosis in left-sided colonic emergencies. Dis Colon Rectum. 2005 Dec;48(12):2272-80. doi: 10.1007/s10350-005-0159-9. PMID: 16228841.
92. Standl T. Niemandem schaden. Anaesthesist. 2015 May;64(5):345-6. German. doi: 10.1007/s00101-015-0041-7. PMID: 25986409. Morgan A, Zheng A, Linden KM, Zhang P, Brown SA, Carpenter JP, Spitz FR, Kwiatt ME. Locally Transplanted Adipose Stem Cells Reduce Anastomotic Leaks in Ischemic Colorectal Anastomoses: A Rat Model. Dis Colon Rectum. 2020 Jul;63(7):955-964. doi: 10.1097/DCR.0000000000001667. PMID: 32168095.
93. Daneman N, Sarwar S, Fowler RA, Cuthbertson BH; SuDDICU Canadian Study Group. Effect of selective decontamination on antimicrobial resistance in intensive care units: a systematic review and meta-analysis. Lancet Infect Dis. 2013 Apr;13(4):328-41. doi: 10.1016/S1473-3099(12)70322-5. Epub 2013 Jan 25. PMID: 23352693.
94. Flor-Lorente B, Noguera-Aguilar JF, Delgado-Rivilla S, García-González JM, Rodriguez-Martín M, Salinas-Ortega L, Casado MÁ, Álvarez M. The economic impact of anastomotic leak after colorectal cancer surgery. Health Econ Rev. 2023 Feb

16;13(1):12. doi: 10.1186/s13561-023-00425-y. PMID: 36795234; PMCID: PMC9933261.

95. Wiegerinck M, Hyoju SK, Mao J, Zaborin A, Adriaansens C, Salzman E, Hyman NH, Zaborina O, van Goor H, Alverdy JC. Novel de novo synthesized phosphate carrier compound ABA-PEG20k-Pi20 suppresses collagenase production in Enterococcus faecalis and prevents colonic anastomotic leak in an experimental model. Br J Surg. 2018 Sep;105(10):1368-1376. doi: 10.1002/bjs.10859. Epub 2018 Apr 16. PMID: 29658991; PMCID: PMC6827486.
96. ÖNORM A26262. Wissenschaftliche Abschlussarbeiten: Angaben für den bibliographischen Nachweis, valid since 1.11.2023, (replaced ÖNORM 2662:1993), 8p.
97. Aschbacher, C. Entwurf eines Führungsstils für innovative Unternehmen. Dissertation, Slowakische Technische Universität Bratislava, 2020, 134p.
98. Steyer GE. Anatomie und Anatomen an der Universität Graz. Medizinhistorische Dissertation, Karl-Franzens-Universität Graz. In: Steyer GE. (ed): Grazer medizinhistorische Schriften, vol 1, ^{3}1988, 391p.
99. Steyer A. Physiologie und Physiologen an der Universität Graz. Medizinhistorische Dissertation, Karl-Franzens-Universität Graz. In: Steyer GE. (ed): Grazer medizinhistorische Schriften, vol 3, ^{2}1988, 470p.
100. Pschyrembel W. Klinisches Wörterbuch. ^{269}2023.

Appendix:

A.1 Publications Resulting from this Dissertation (mandatory chapter)

See mandatary chapter “Disclosures“, page 4.

A.2 Declaration of Copyright for this Dissertation

All co-authors who contributed to the published research

Steyer GE, Puchinger M, Pfeifer J. Successful Clinical Avoidance of Colorectal Anastomotic Leakage through Local Decontamination. Antibiotics (Basel). 2024 Jan 15;13(1):79.

doi: 10.3390/antibiotics13010079. PMID: 38247638; PMCID: PMC10812415.

agreed to the use of their contributes in this dissertation, the publication in this Journal was enabled open access.

A.3 *It´s all Greek to me:* Abstracts in Mandarin, Hindi and Spain. *„Mi sembra tedesco“*

Taking account of the ***number of native speakers***, it seems to the author, to be very important to add and include - at least in the appendix of this dissertation - abstracts in Mandarin, Hindi and Spanish.

According to the rules of the Medical University of Graz, Austria, Europe, a dissertation has to be submitted in a PDF/a version, …

> … but unfortunately this mandatary version can only handle Arial and not instance letters in Chinese, Greek or Cyrillic script, even not, if they are used correctly. So, an automatic translation from the „Zusammenfassung“ was done, using an extension of a language checking programme from “Google Translate v.2.0.13“, a pre-programme of artificial intelligence. It is easy, to perform a re-translation.

通过局部去污成功避免结直肠吻合口瘘

研究背景

结肠手术是一种常见的外科手术，但手术后可能会发生结肠吻合口漏，这是一种不可预测的并发症，可能需要再次手术。研究表明，潜在的致病菌如假单胞菌和肠球菌会通过降解胶原蛋白和激活组织基质金属蛋白酶-9来促进结肠吻合口漏的发生。因此，微生物群落是预防结肠手术后吻合口漏的关键。

研究内容

本研究旨在探讨使用一种新型的局部作用抗生素混合物，特别针对假单胞菌和肠球菌的选择性脱菌术，是否能够减少或甚至停止早期症状性漏出的发生。研究对象为在我们大学医院接受结肠手术的住院患者，分为两组，干预组接受多粘菌素B、庆大霉素和万古霉素，每6小时一次，连续5天，而对照组则没有接受这种干预。结果显示，共分析了301名患者，其中152名为对照组，11名发生结肠吻合口漏，而149名为干预组，没有发生结肠吻合口漏，两组之间的差异非常显著。

研究意义

本研究使用的抗生素混合物（多粘菌素B、庆大霉素和万古霉素）用于局部脱菌，完全阻止了结肠吻合口漏的发生。根据结肠吻合口漏的定义，在局部围手术期脱菌后不需要进行进一步的手术。这项研究的创新点在于，通过使用新型的抗生素混合物，成功预防了结肠吻合口漏的发生，为结肠手术后并发症的预防提供了新思路。

स्थानीय परिशोधन के माध्यम से कोलोरेक्टल एनास्टोमोटिक रिसाव का सफल नैदानिक बचाव

अनुसंधान बैकग्राउंड

कोलन सर्जरी एक सामान्य सर्जिकल प्रक्रिया है, लेकिन सर्जरी के बाद कोलन का एनास्टोमोटिक रिसाव हो सकता है, एक अप्रत्याशित जटिलता जिसके लिए दोबारा ऑपरेशन की आवश्यकता हो सकती है। अध्ययनों से पता चला है कि स्यूडोमोनास और एंटरोकोकस जैसे संभावित रोगजनक बैक्टीरिया कोलेजन को क्षीण करके और ऊतक मैट्रिक्स मेटालोप्रोटीनेज-9 को सक्रिय करके कोलोनिक एनास्टोमोटिक रिसाव की घटना को बढ़ावा दे सकते हैं। इसलिए, बृहदान्त्र सर्जरी के बाद एनास्टोमोटिक रिसाव को रोकने के लिए माइक्रोबियल समुदाय महत्वपूर्ण है।

अनुसंधान सामग्री

इस अध्ययन का उद्देश्य यह जांच करना था कि क्या स्थानीय रूप से काम करने वाले एंटीबायोटिक दवाओं के एक नए मिश्रण का उपयोग करके चयनात्मक नसबंदी, विशेष रूप से स्यूडोमोनस और एंटरोकोकी को लक्षित करके, प्रारंभिक रोगसूचक रिसाव की घटना को कम या यहां तक कि रोक सकता है। अध्ययन के विषय वे रोगी थे जिनकी हमारे विश्वविद्यालय अस्पताल में बृहदान्त्र सर्जरी हुई थी और उन्हें दो समूहों में विभाजित किया गया था। हस्तक्षेप समूह को 5 दिनों के लिए हर 6 घंटे में पॉलीमीक्सिन बी, जेंटामाइसिन और वैनकोमाइसिन प्राप्त हुआ, जबकि नियंत्रण समूह को ऐसा कोई हस्तक्षेप स्वीकार नहीं किया गया। परिणामों से पता चला कि कुल 301 रोगियों का विश्लेषण किया गया, जिनमें से 152 नियंत्रण समूह में थे, 11 में कोलोनिक एनास्टोमोटिक रिसाव था, और 149 हस्तक्षेप समूह में थे, और कोई कोलोनिक एनास्टोमोटिक रिसाव नहीं हुआ। दोनों समूहों के बीच अंतर बहुत था महत्वपूर्ण।

महत्व

स्थानीय डिबैक्टीरिया के लिए इस अध्ययन में उपयोग किए गए एंटीबायोटिक मिश्रण (पॉलीमीक्सिन बी, जेंटामाइसिन और वैनकोमाइसिन) ने कोलोनिक एनास्टोमोटिक रिसाव की घटना को पूरी तरह से रोक दिया। कोलोनिक एनास्टोमोटिक लीकेज की परिभाषा के आधार पर, स्थानीय पेरिऑपरेटिव डिस्टरलाइजेशन के बाद किसी और सर्जरी की आवश्यकता नहीं होती है। इस अध्ययन की नवीनता यह है कि एक नए एंटीबायोटिक मिश्रण का उपयोग करके, कोलोनिक एनास्टोमोटिक रिसाव की घटना को सफलतापूर्वक रोका गया, जिससे कोलन सर्जरी के बाद जटिलताओं की रोकथाम के लिए एक नया विचार मिला।

Evitar con éxito la fístula anastomótica colorrectal mediante descontaminación local

Antecedentes de la investigación

La cirugía de colon es un procedimiento quirúrgico común, pero puede ocurrir fuga anastomótica del colon después de la cirugía, una complicación impredecible que puede requerir una nueva operación. Los estudios han demostrado que bacterias patógenas potenciales como Pseudomonas y Enterococcus pueden promover la aparición de fugas anastomóticas del colon al degradar el colágeno y activar la metaloproteinasa-9 de la matriz tisular. Por tanto, la comunidad microbiana es clave para prevenir la fuga anastomótica después de la cirugía de colon.

Contenido de la investigación

El objetivo de este estudio fue investigar si la esterilización selectiva utilizando una nueva mezcla de antibióticos de acción local, dirigida específicamente a Pseudomonas y Enterococci, podría reducir o incluso detener la aparición de fugas sintomáticas tempranas. Los sujetos del estudio fueron pacientes hospitalizados sometidos a cirugía de colon en nuestro hospital universitario y se dividieron en dos grupos: el grupo de intervención recibió polimixina B, gentamicina y vancomicina cada 6 horas durante 5 días, mientras que el grupo de control recibió No se aceptó dicha intervención. Los resultados mostraron que se analizaron un total de 301 pacientes, de los cuales 152 estaban en el grupo de control, 11 tenían fuga anastomótica colónica y 149 estaban en el grupo de intervención, y no se produjo fuga anastomótica colónica. La diferencia entre los dos grupos fue muy significativo.

Significado

La mezcla de antibióticos (polimixina B, gentamicina y vancomicina) utilizada en este estudio para las desbacterias locales evitó por completo la aparición de fuga anastomótica colónica. Según la definición de fuga anastomótica colónica, no se requiere cirugía adicional después de la desesterilización perioperatoria local. La innovación de este estudio es que mediante el uso de una nueva mezcla de antibióticos, se evitó con éxito la aparición de fugas anastomóticas del colon, lo que proporciona una nueva idea para la prevención de complicaciones después de la cirugía de colon.

The content „kommt mir spanisch vor,“

the author remembers a witticism from his schooldays, attributed to Holy Roman Emperor and Archduke of Austria Carlos I / Karl V.(I.), that he communicatet in different languages - depending on the recipient: In different languages he used to speak to his GOD, to men and to wifes, to horses and donkeys - he spoke Latin, French, German or Spanish, but nobody really knows, if **he** understood the response of his donkey (and had a speach “with“ and not only “to“ his vis-á-vis). But his way to gender (speak Italian to women, and French to men) could be a prototype for the Medico-Philologico University of Graz, Austria, Europe.

Please remember page 5 (Preface):

Acquire gold as much you need
And wisdom as much you can

To communicate in different languages is not a loss of identity (that is true especially for using English instead of German for scientia medicae at our Statistico-Medical-Philologico University), **it is not important in toto, it´s just another way of communication** expressing the mindset and knowledge of the **user** (appreciating, that additionally pluralism in a language itself is a given condition). And languages are complementary sciences (like mathematics for physics, or statistics for empirical sciences) for science, but always remember: **surgery (and medicine) is not a science,** it is an **art** (of healing)!

A.4 Afterword (optional)

Red tapes are lovely,
the author will give an example: to create the title page of a dissertation, there exists an important rule (different by different Universities, because a University is independent, and science?) following ÖNORM, the number is A26262 [96] („Wissenschaftliche Abschlussarbeiten: Angaben für den bibliographischen Nachweis, valid since 1.11.2023, replaced ÖNORM 2662:1993“. Sic!). But the ÖNORM is behind a paywall: a student can buy this rule [96] from the editor, it is cheap: for 8 pages (title page and table of content included) the costs are at the moment EUR 71.45 for a paper print (for a download price is reduced to 47.63). That is great, the prices are very creatively arranged, and helpful. The University Library of the Medical University of Graz does not have the A26262, the author would have paid Euro 7.- for interlibrary loan, but it was impossible, even the “Austrian National Library” has no copy. The same is true with some literature when you try to work scientific. Additional informations on the title page are possible, but they are censored and have to be removed. During the inclusion period, we had a slight delay in our timetable due to COVID-19 (SARS-CoV-2), but the study has not been slowed down by barnacles like other dissertations [97] written in clumsy German and submitted at the Technical University of Bratislava, Slovacia.

Remember every day: The Medical University of Graz is not new founded, but in good tradition, in reality, the logo and a hymn were changed with youthful enthusiasm. Surgical teaching in Graz started in the „Totenkammerl” at “St. George Cementary” [98] (my first Dissertation) in the year 1777, later „Medizinisch-Chirurgische Lehranstalt Graz” was founded 1786 and 1863 renamed „Medizinische Fakultät der Karl-Franzens-Universität Graz“ [99].

According to the rules of the Medical University of Graz, Austria, Europe, the author preferred to use direct quotations in the original language (for example, he preferred in citation [47] to cite the published German version).

If you check out soon after reading some more pages coming, don´t forget to check my Easter-Eggs placed (as usual in any of my dissertations) in the text, as „Steinlaus” (first described in 1983 [100]), or sarcophagus, English (or who cares if it is Greek, or perhaps

Latin?), pertaining to the distinctive vocabulary of the educated, meaning (look, it´s an **aorist**), a “stone feeds meat”, because when the sarcophagus is re-opened after some time, the meat has disappeared unfortunately - or some thought, “sarcos” (that is Greek in Latin letters) was eaten by the stone - and still bones are left [“scientific” medical knowledge from schooldays, therefore no citation is needed according to the rules]).

Please, don´t judge me too hard. *Danke.*