Biomarkers for early sepsis detection

Bettina Leber, PhD,[1,2*] Angela Horvath, PhD,[1,2*] Irina Komarvoa,[3] MD, Bianca Schmerboeck, BSc,[1,2] Ingeborg Klymiuk, PhD,[4] Marija Durdevic, MSc,[5] Florian Rainer[3], MD, Andreas Blesl[3], MD, Philipp Stiegler, MD,[1] Vanessa Stadlbauer, MD,[3]

[1]Department of Transplantation Surgery, Medical University of Graz, Austria, [2]Center for Biomarker Research in Medicine (CBmed), Austria,[3]Department of Gastroenterology and Hepatology, Medical University of Graz, Austria, [4]Center for Medical Research, Core Facility Molecular Biology, Medical University of Graz, Austria. [5]Core Facility Computational Bioanalytics, Medical University of Graz, Austria

Introduction

- Sepsis
  - Systemic, deleterious host response to infection
  - affects millions of people around the world each year, lethality of 25%-50%
- therapeutic goal: “Hit early, hit fast, hit hard(right)”
- loss of gut wall integrity: pivotal role
- gut microbiome composition: important mechanism
- no data on gut barrier function and gut microbiome composition in early sepsis available

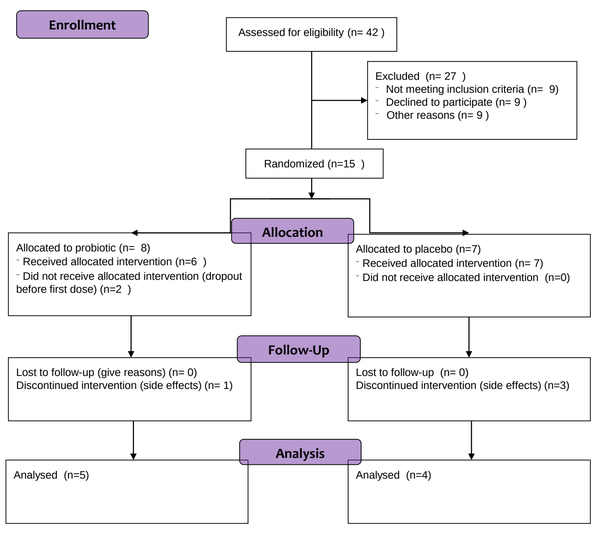
Aims

- identification of potential biomarkers for early sepsis
- proof of concept: effects of a multispecies probiotic on gut microbiota and gut wall integrity

Methods

- Pilot clinical trial on patients with early sepsis receiving a multispecies probiotic (Winclove 607) or placebo for 28 days
- Gut microbiome in stool samples (16s sequencing, bioinformatics)
- Markers of gut wall integrity (fecal zonulin and calprotectin, serum diamino oxidase)
- Markers of bacterial translocation (LPS, sCD14, LBP, peptidoglykane, bacterial DNA)
- Methods: ELISA, HEK-Blue reporter cells

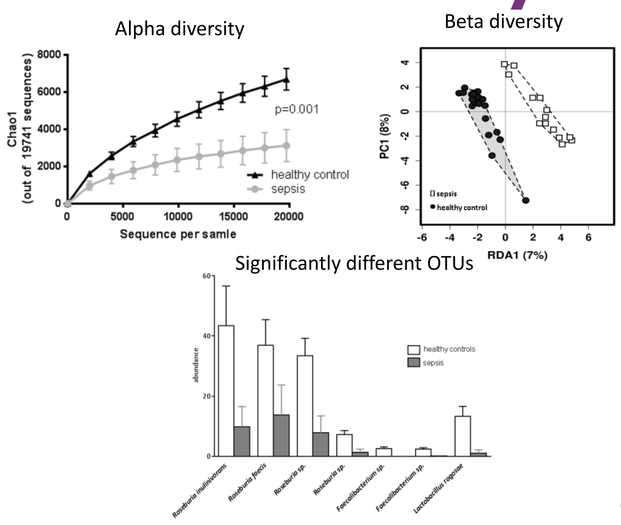
Enrolment and patient characteristics



	Early sepsis (n=15*)	Healthy controls (n=21)
Age (years)	54 +/- 17	58 +/- 7
Gender female/male	7/8	12/9
BMI kg/m²	26 +/- 5	25 +/- 3
White blood cells	12.7 +/- 9.1**	5.6 +/- 1.8
CRP	160.3 +/- 124.9***	2.1 +/- 2.2

The reasons for dropout were diarrhoea (n=1, placebo group), nausea (n=3, 2 placebo, 1 probiotic) and withdrawal of consent before the first dose of the study product was taken (n=2, both probiotic).
Dropouts were therefore not related to the intervention.

Microbiome composition and function in early sepsis



Pathway	Bonferroni p value	mean abundance sepsis patients	mean abundance healthy controls
Amino sugar and nucleotide sugar metabolism	0,00024374	2594555	707802
D-Alanine metabolism	0,00024374	172116	47000
Glutamatergic synapse	0,00024374	202790	46155
Glutathione metabolism	0,00024374	292896	76620
Glycosphingolipid biosynthesis - globo series	0,00024374	235468	52671
Glycosyltransferases	0,00024374	539846	137969
Lipid metabolism	0,00024374	232120	67962
Membrane and intracellular structural molecules	0,00024374	909137	185946
N-Glycan biosynthesis	0,00024374	39225	8185
Novobiocin biosynthesis	0,00024374	227841	56277
Other ion-coupled transporters	0,00024374	2138788	552342
Phosphatidylinositol signaling system	0,00024374	144648	37578
Primary bile acid biosynthesis	0,00024374	73455	17639
Prostate cancer	0,00024374	81028	17300
Proteasome	0,00024374	76249	16868
Protein digestion and absorption	0,00024374	47867	5842
Riboflavin metabolism	0,00024374	394239	89225
Ribosome Biogenesis	0,00024374	2364573	614470
Thiamine metabolism	0,00024374	866051	219568
Valine, leucine and isoleucine degradation	0,00024374	318558	76041

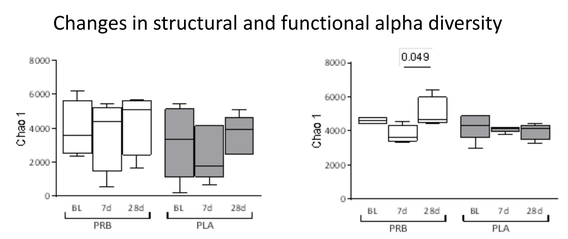
- Early sepsis is associated with
  - decreased alpha diversity and beta diversity of the gut microbiome
  - Loss of beneficial species
  - Decreased functional content

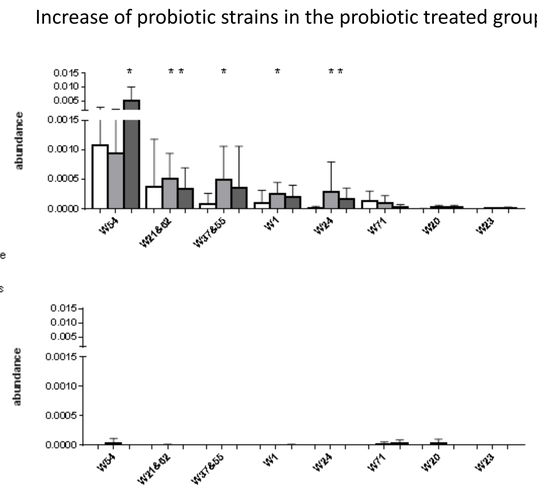
Gut permeability and bacterial translocation in early sepsis

	baseline	day 7	day 28	Healthy controls
Calprotectin in stool (ng/ml)	191 (45; 568)	44 (32;621)	62 (26;199)	56 (27;80)
Zonulin in stool (ng/ml)	49 (17; 96)	74 (30; 116)	57 (54; 193)	66 (57; 79)
DAO (U/ml)	11 (7; 23)	9 (7;14)	7 (5;10)	9 (8; 15)
Endotoxin (EU/ml)	4 (1;51) **	5 (1; 41) *	3 (1; 6) *	2 (1; 3)
sCD14 (µg/ml)	2.8 (2.1; 3.6)***	2.2 (1.6; 3.6)*	2.2 (1.6; 3.2)*	1.7 (1.5; 1.8)
LBP (mg/ml)	39 (30; 62)***	23 (16; 31)	13 (9; 16)	17 (15; 21)
Bacterial DNA (µM)	17 (11;19)	12 (10;30)	13 (10;19)	10 (7,18)
Peptidoglycane (ng/ml)	1.8 (1.4;2.7)	1.6 (1.0;2.4)	1.6 (1.3;2.3)	0.8 (0.5;1.1)

- Early sepsis is associated with
  - Increased endotoxin and endotoxin binding proteins
  - No changes in gut barrier function

Effect of Winclove 607 on the gut microbiome in early sepsis





- Probiotic modulation
  - Increases functional diversity
  - Increases abundance of probiotic strains

Conclusions

- Sepsis is associated with early structural and functional changes in microbiome composition
- Alpha Diversity may be a useful biomarker
- Gut permeability is not severely altered
- Probiotic modulation of the gut microbiome is feasible in early sepsis

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CONTACT
CBmed GmbH
CENTER FOR BIOMARKER RESEARCH IN MEDICINE
Stiftingtalstrasse 5, 8010 Graz
Phone: +43 316 385 28801
office@cbmed.at