

The background of the slide is a close-up photograph of several white, oval-shaped tablets. The tablets are arranged in a grid-like pattern, typical of a blister pack. The lighting is soft, highlighting the texture and slight shadows of the tablets. A dark grey rectangular box is overlaid on the left side of the image, containing the title and other text.

# Survival of *Lactobacillus Rhamnosus* in a Tableting Process

April 26<sup>th</sup> 2023

Marek Lachmann



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- Short introduction into PROBIOTICS
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# Short introduction into PROBIOTICS

# Short introduction into PROBIOTICS

- Probiotics are live, apathogenic microorganisms (e.g. bacteria or yeasts) that are considered / proven to bring about distinct health benefits in humans or animals:

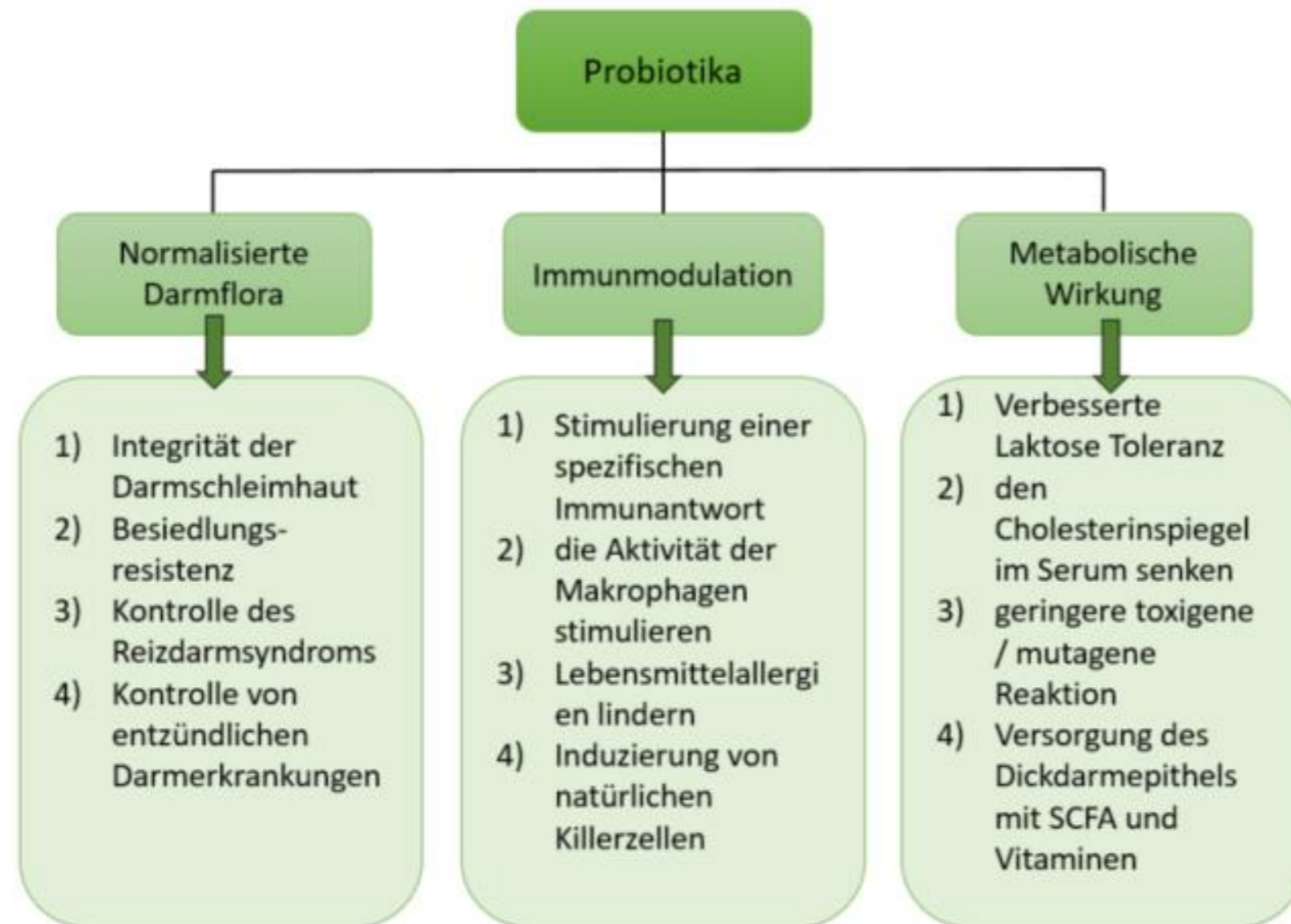


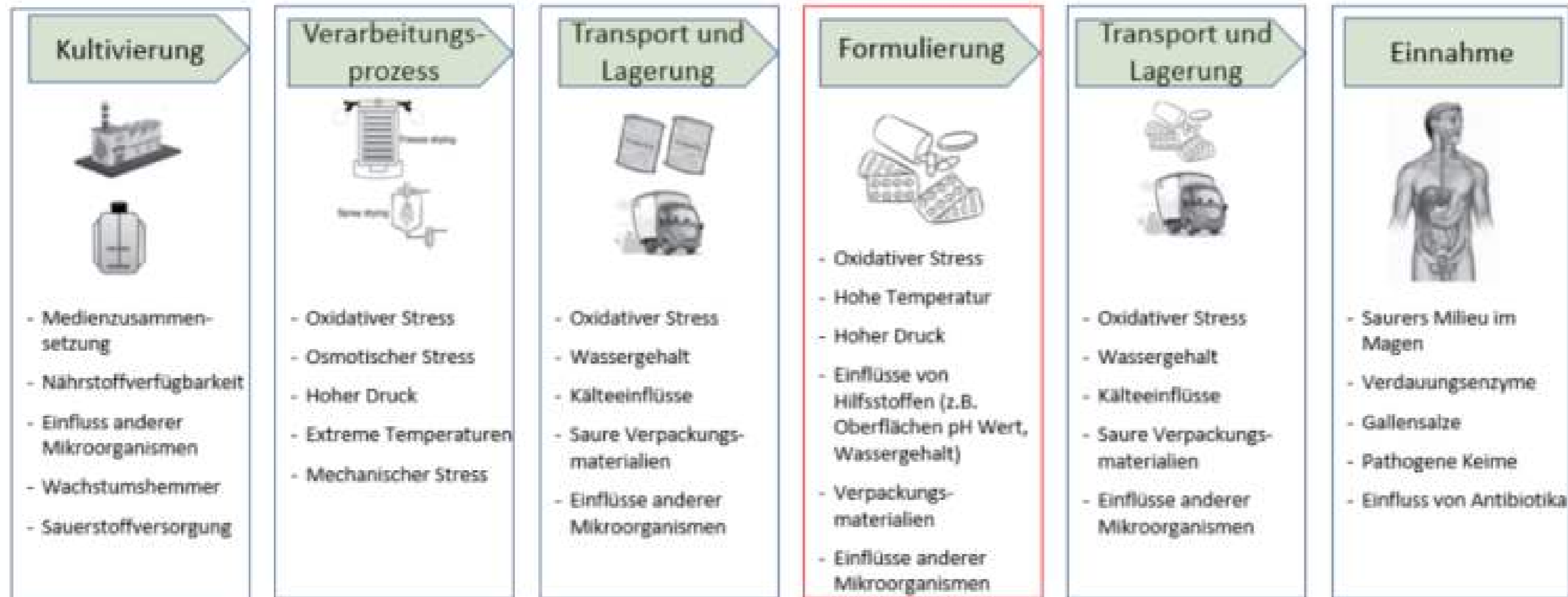
Abbildung 2: Schematische Darstellung über die verschiedenen Funktionen von Probiotika (in Anlehnung an <sup>20)</sup>)



# Influences on survival of probiotic bacteria

# Influences on survival of probiotic bacteria

- From their cultivation to consumption probiotic microorganisms are exposed to various type of stress that can reduce viability of the strains:



**Abbildung 1: negative Einflüsse auf die Vitalität von probiotischen Kulturen von der Kultivierung bis zur Einnahme Probiotika (in Anlehnung an <sup>1, 6)</sup>)**



Influences of tableting  
temperature and compression  
force on the survival of  
*L. rhamnosus*

# Influences of tableting temperature and compression force on the survival of *L. rhamnosus*

Aims: Evaluation of machine and formulation influences on the survival of *L. rhamnosus*

Tablet Compositions: 200 mg Tablets, RoTab T rotary lab press; convex punches;  $\varnothing = 7$  mm; feeding speed 20 rpm

Materialien	M_LGG1	M_LGG2	M_LGG3	M_LGG4	M_LGG5	6M_LGG6	M_LGG7
<b>DCPA</b>	57,6	15,4	57,6	61,6	14,4	14,4	59,6
<b>MCC</b>	14,4	61,6	14,4	15,4	57,6	57,6	14,9
<b>DIS</b>	2,0	2,0	2,0	2,0	2	2	2,0
<b>LUB</b>	1,0	1,0	1,0	1,0	1	1	1,0
<b>LGG</b>	10,0	10,0	10,0	10,0	10	10	10,0
<b>MAB</b>	10,0	10,0	10,0	10,0	10	10	10,0
<b>L-HPC</b>	5,0	-	-	-	-	5	-
<b>HPC</b>	-	-	5,0	-	5	-	2,5



# Influences of tableting temperature and compression force on the survival of *L. rhamnosus*

## Tablet Compositions:

**Tabelle 3: Rezeptur von CompactCel® F 900.17 DIS [**

Flohsamenschalen
Hafer Fasern
Bio-Kartoffelstärke
Agave-Inulin-Pulver

**Tabelle 4: Rezeptur von CompactCel® F 200.28 LUB**

Mikrokristalline Cellulose (Typ 105)
Hafer Fasern
Sonnenblumenöl

**Tabelle 5: Rezeptur von CompactCel® F 200.25 MAB**

Mikrokristalline Cellulose (Typ 200)
Isomalt
Calcium Carbonat
Vorgelatinierte Stärke

# Influences of tableting temperature and compression force on the survival of *L. rhamnosus*

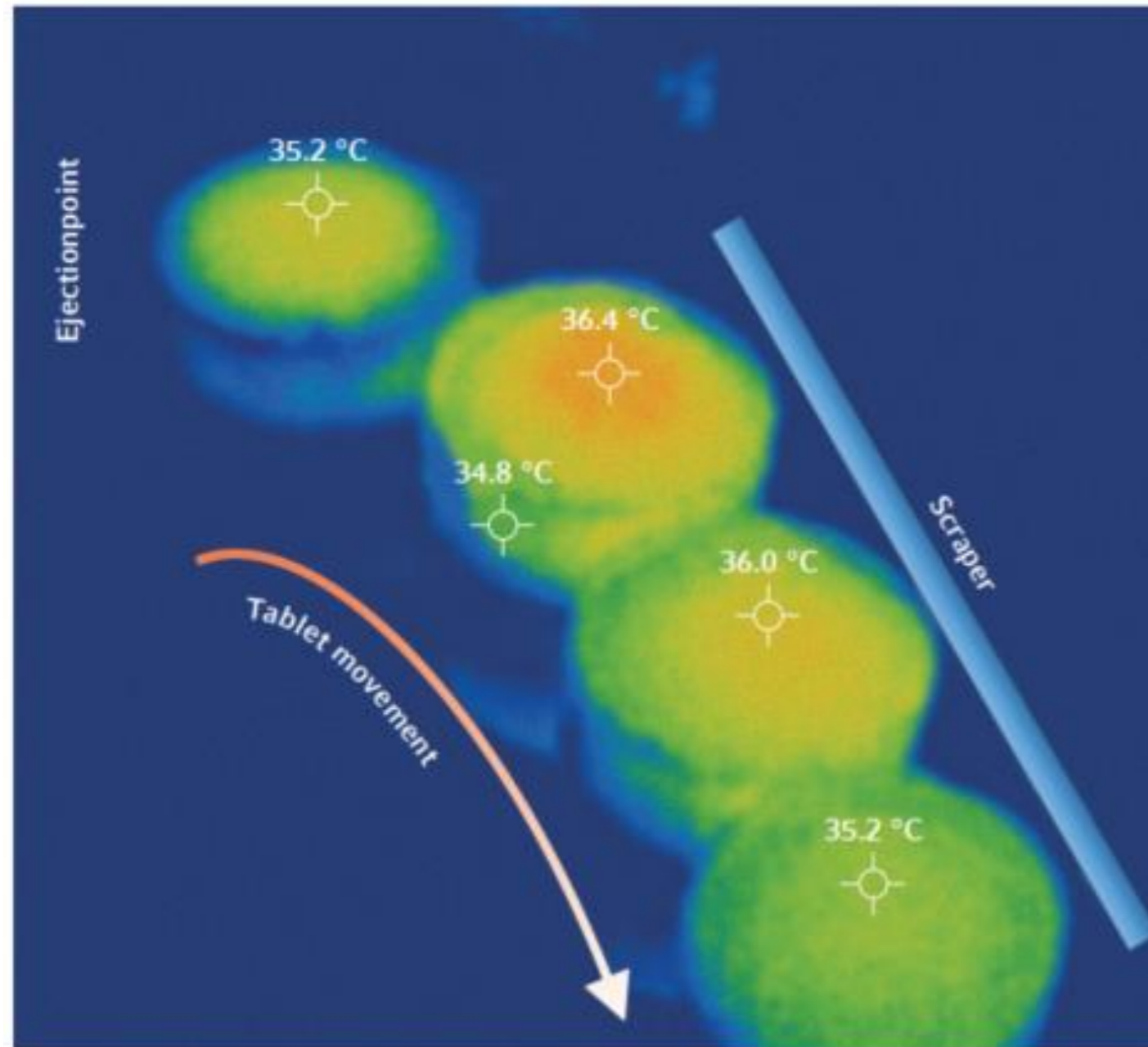
## Tableting Parameters:

DOE settings for the tablet press with variations of MCF and rotational speed

Name	Rezeptur	Hauptpresskraft [kN]	Umdrehungszahl [rpm]
<b>N-PLA1 v N-LGG1</b>	M_PLA1 v M_LGG1	4	10
<b>N-PLA2 v N-LGG2</b>	M_PLA2 v M_LGG2	4	10
<b>N-PLA3 v N-LGG3</b>	M_PLA3 v M_LGG3	12	10
<b>N-PLA4 v N-LGG4</b>	M_PLA2 v M_LGG2	12	40
<b>N-PLA5 v N-LGG5</b>	M_PLA4 v M_LGG4	4	40
<b>N-PLA6v N-LGG6</b>	M_PLA5v M_LGG5	4	40
<b>N-PLA7 v N-LGG7</b>	M_PLA4 v M_LGG4	12	40
<b>N-PLA8 v N-LGG8</b>	M_PLA6v M_LGG6	12	40
<b>N-PLA9 v N-LGG9</b>	M_PLA7 v M_LGG7	8	25
<b>N-PLA10 v N-LGG10</b>	M_PLA7 v M_LGG7	8	25
<b>N-PLA11 v N-LGG11</b>	M_PLA7 v M_LGG7	8	25

# Influences of tableting temperature and compression force on the survival of *L. rhamnosus*

Tablet Analysis : Breaking Force, Friability, Ejection Force, Tablet Temperature, CFU count



<https://www.optris.global/optris-xi-400>

# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- Results of the 11 rund within the DOE framework

Batch	Bruchkraft	SD	RSD	Zugfestigkeit	SD	RSD	Zerfallszeit	SD	RSD	Ausstoßkraft	SD	RSD	Temperatur	SD	RSD
	[N]	[N]	[%]	[N/mm <sup>2</sup> ]	[N/mm <sup>2</sup> ]	[%]	[s]	[s]	[%]	[N]	[N]	[%]	[°C]	[°C]	[%]
N-LGG1	51,6	2,7	5,2	1,1	0,1	4,6	40	11	26,7	279	28	10,0	29,9	0,3	1,0
N-LGG2	114,3	8,4	7,3	2,2	0,2	7,1	120	35	29,6	235	47	20,0	31,6	0,1	0,4
N-LGG3	175,6	14,5	8,3	4,6	0,4	8,5	3804	530	13,9	457	55	12,0	36,3	0,2	0,4
N-LGG4	256,2	15,9	6,2	6,0	0,4	6,2	3480	439	12,6	466	90	19,3	36,1	0,2	0,6
N-LGG5	35,0	3,7	10,6	0,7	0,1	9,8	29	4	13,7	271	21	7,7	31,2	0,2	0,5
N-LGG6	103,9	16,8	16,2	2,0	0,3	16,2	564	275	48,7	213	21	9,9	32,7	0,4	1,3
N-LGG7	129,7	8,5	6,6	3,3	0,2	6,6	678	157	23,1	438	45	10,2	39,7	0,1	0,4
N-LGG8	250,6	13,7	5,5	5,6	0,3	6,0	2021	170	8,4	341	67	19,7	40,6	0,2	0,6
N-LGG9	96,4	14,1	14,7	2,3	0,3	14,9	799	211	26,5	381	39	10,0	34,5	0,1	0,3
N-LGG10	97,3	6,0	6,2	2,4	0,2	7,6	976	223	22,9	391	51	13,0	35,2	0,2	0,7
N-LGG11	101,5	10,9	10,7	2,5	0,3	10,7	1024	230	22,5	375	41	10,9	35,2	0,2	0,5

Materialien	M_LGG1	M_LGG2	M_LGG3	M_LGG4	M_LGG5	6M_LGG6	M_LGG7
DCPA	57,6	15,4	57,6	61,6	14,4	14,4	59,6
MCC	14,4	61,6	14,4	15,4	57,6	57,6	14,9
DIS	2,0	2,0	2,0	2,0	2	2	2,0
LUB	1,0	1,0	1,0	1,0	1	1	1,0
LGG	10,0	10,0	10,0	10,0	10	10	10,0
MAB	10,0	10,0	10,0	10,0	10	10	10,0
L-HPC	5,0	-	-	-	-	5	-
HPC	-	-	5,0	-	5	-	2,5

Name	Rezeptur	Hauptpresskraft [kN]	Umdrehungszahl [rpm]
N-PLA1 v N-LGG1	M_PLA1 v M_LGG1	4	10
N-PLA2 v N-LGG2	M_PLA2 v M_LGG2	4	10
N-PLA3 v N-LGG3	M_PLA3 v M_LGG3	12	10
N-PLA4 v N-LGG4	M_PLA2 v M_LGG2	12	40
N-PLA5 v N-LGG5	M_PLA4 v M_LGG4	4	40
N-PLA6v N-LGG6	M_PLA5v M_LGG5	4	40
N-PLA7 v N-LGG7	M_PLA4 v M_LGG4	12	40
N-PLA8 v N-LGG8	M_PLA6v M_LGG6	12	40
N-PLA9 v N-LGG9	M_PLA7 v M_LGG7	8	25
N-PLA10 v N-LGG10	M_PLA7 v M_LGG7	8	25
N-PLA11 v N-LGG11	M_PLA7 v M_LGG7	8	25

# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- Survival (viability) of *L. rhamnosus* within the DOE framework

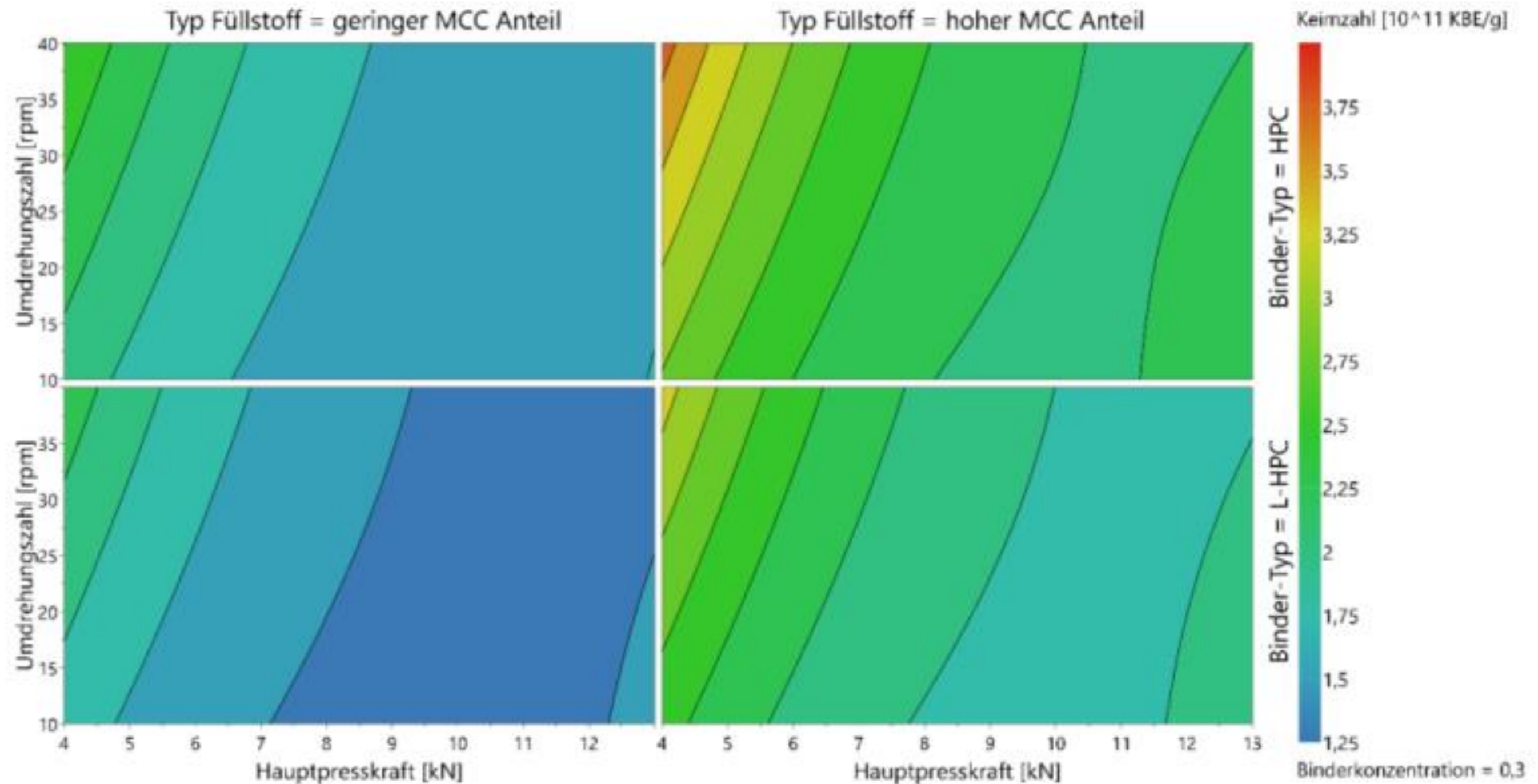
Batch	Keimzahl	SD	RSD
	[10 <sup>11</sup> KBE/g]	[KBE/g]	[%]
<b>N-LGG1</b>	1,15	6,92*10 <sup>10</sup>	60,4
<b>N-LGG2</b>	2,77	5,49*10 <sup>10</sup>	19,8
<b>N-LGG3</b>	1,13	3,39*10 <sup>10</sup>	30,0
<b>N-LGG4</b>	1,84	8,92*10 <sup>10</sup>	48,6
<b>N-LGG5</b>	2,12	8,25*10 <sup>10</sup>	38,9
<b>N-LGG6</b>	2,58	4,12*10 <sup>10</sup>	16,0
<b>N-LGG7</b>	1,38	2,55*10 <sup>10</sup>	18,5
<b>N-LGG8</b>	1,29	1,26*10 <sup>10</sup>	9,8
<b>N-LGG9</b>	1,51	4,93*10 <sup>10</sup>	32,6
<b>N-LGG10</b>	1,24	8,28*10 <sup>9</sup>	6,7
<b>N-LGG11</b>	1,15	2,60*10 <sup>10</sup>	22,5

Materialien	M_LGG1	M_LGG2	M_LGG3	M_LGG4	M_LGG5	6M_LGG6	M_LGG7
<b>DCPA</b>	57,6	15,4	57,6	61,6	14,4	14,4	59,6
<b>MCC</b>	14,4	61,6	14,4	15,4	57,6	57,6	14,9
<b>DIS</b>	2,0	2,0	2,0	2,0	2	2	2,0
<b>LUB</b>	1,0	1,0	1,0	1,0	1	1	1,0
<b>LGG</b>	10,0	10,0	10,0	10,0	10	10	10,0
<b>MAB</b>	10,0	10,0	10,0	10,0	10	10	10,0
<b>L-HPC</b>	5,0	-	-	-	-	5	-
<b>HPC</b>	-	-	5,0	-	5	-	2,5

Name	Rezeptur	Hauptpresskraft [kN]	Umdrehungszahl [rpm]
<b>N-PLA1 v N-LGG1</b>	M_PLA1 v M_LGG1	4	10
<b>N-PLA2 v N-LGG2</b>	M_PLA2 v M_LGG2	4	10
<b>N-PLA3 v N-LGG3</b>	M_PLA3 v M_LGG3	12	10
<b>N-PLA4 v N-LGG4</b>	M_PLA2 v M_LGG2	12	40
<b>N-PLA5 v N-LGG5</b>	M_PLA4 v M_LGG4	4	40
<b>N-PLA6 v N-LGG6</b>	M_PLA5 v M_LGG5	4	40
<b>N-PLA7 v N-LGG7</b>	M_PLA4 v M_LGG4	12	40
<b>N-PLA8 v N-LGG8</b>	M_PLA6 v M_LGG6	12	40
<b>N-PLA9 v N-LGG9</b>	M_PLA7 v M_LGG7	8	25
<b>N-PLA10 v N-LGG10</b>	M_PLA7 v M_LGG7	8	25
<b>N-PLA11 v N-LGG11</b>	M_PLA7 v M_LGG7	8	25

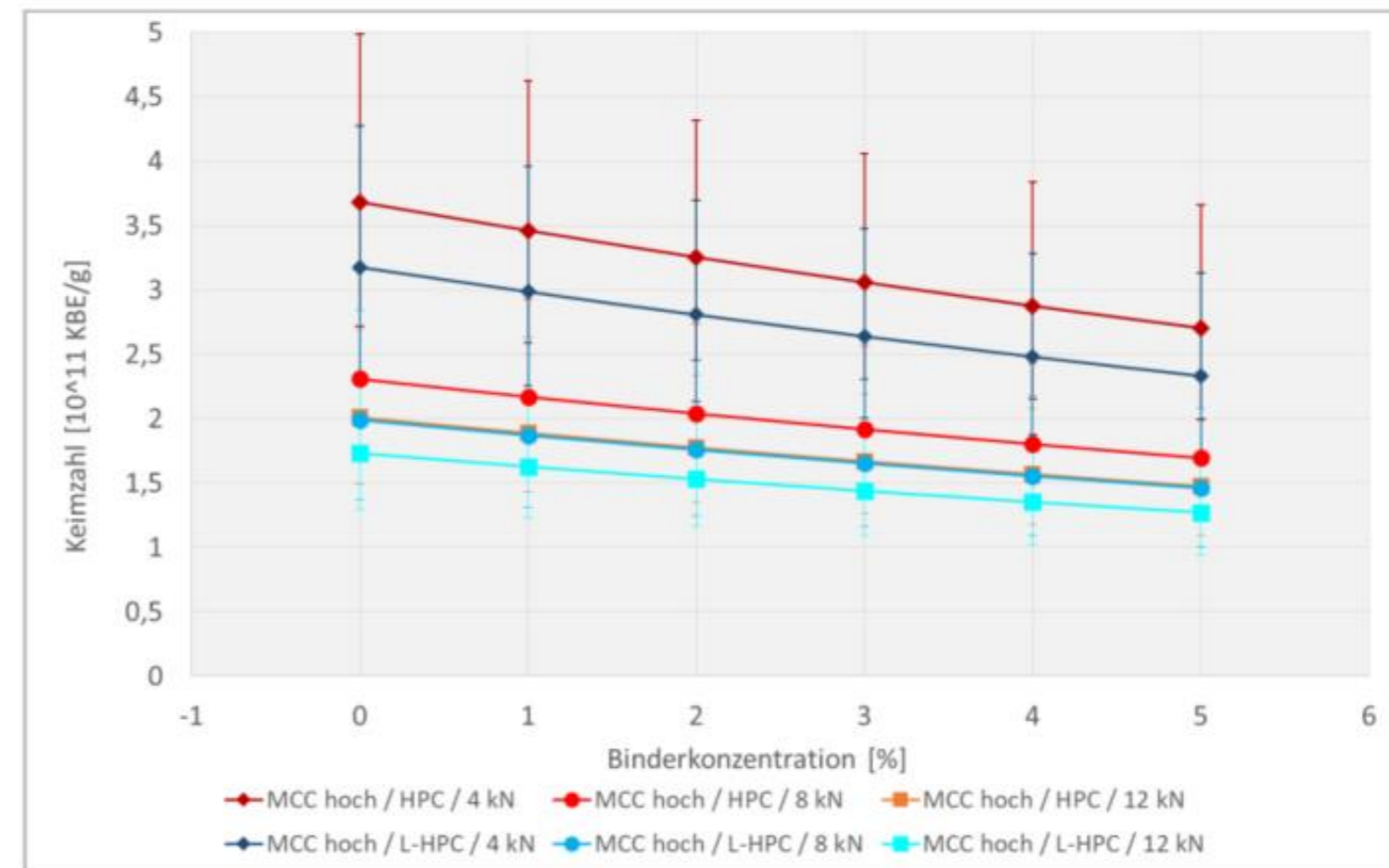
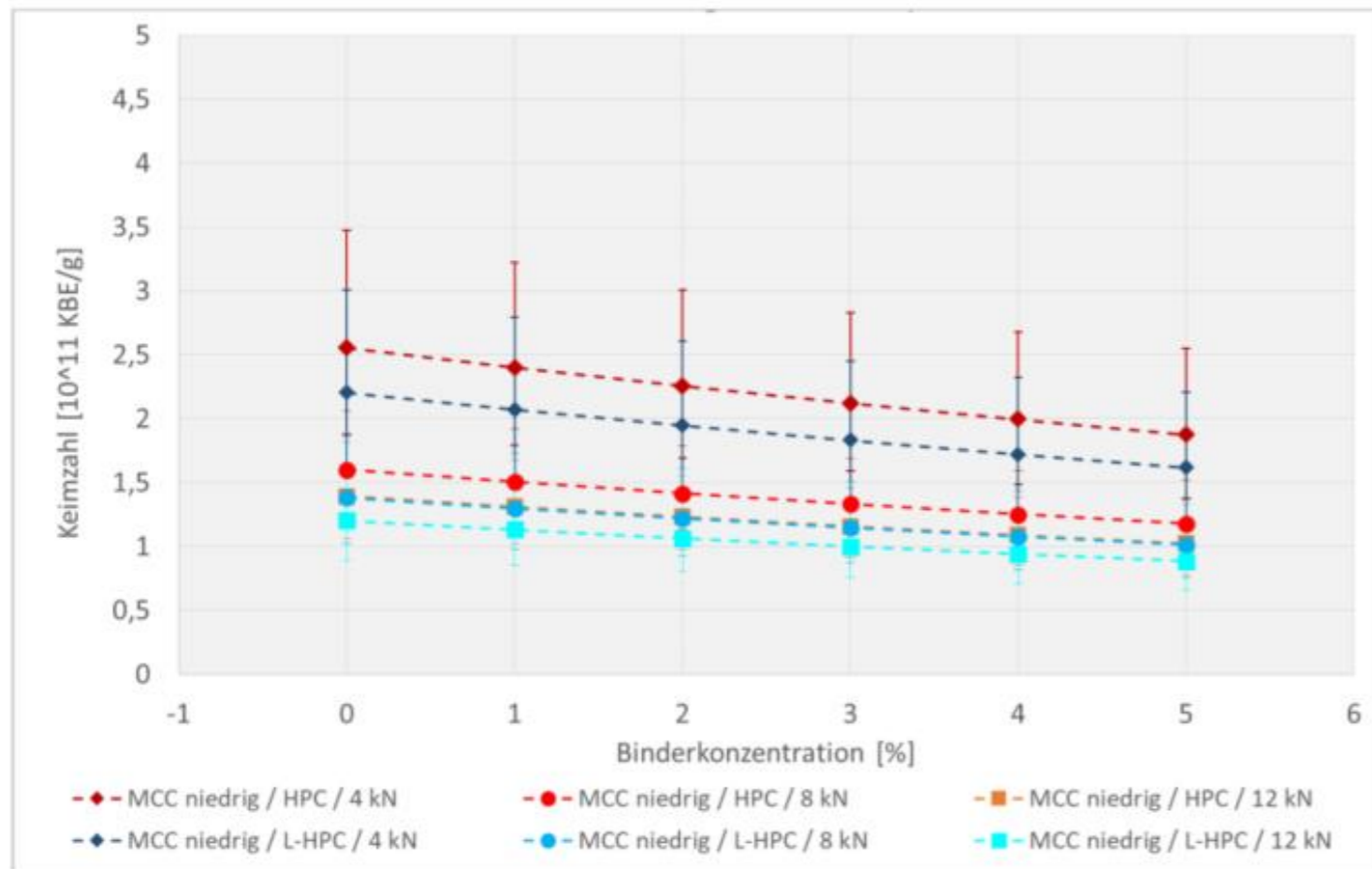
# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- Modelling of the data showed that the formulations with low concentrations of MCC resulted in decreased number of viable bacteria
- Additionally the compaction force plays a pivotal role
- L-HPC was found to be the less favourable binder of the two candidates



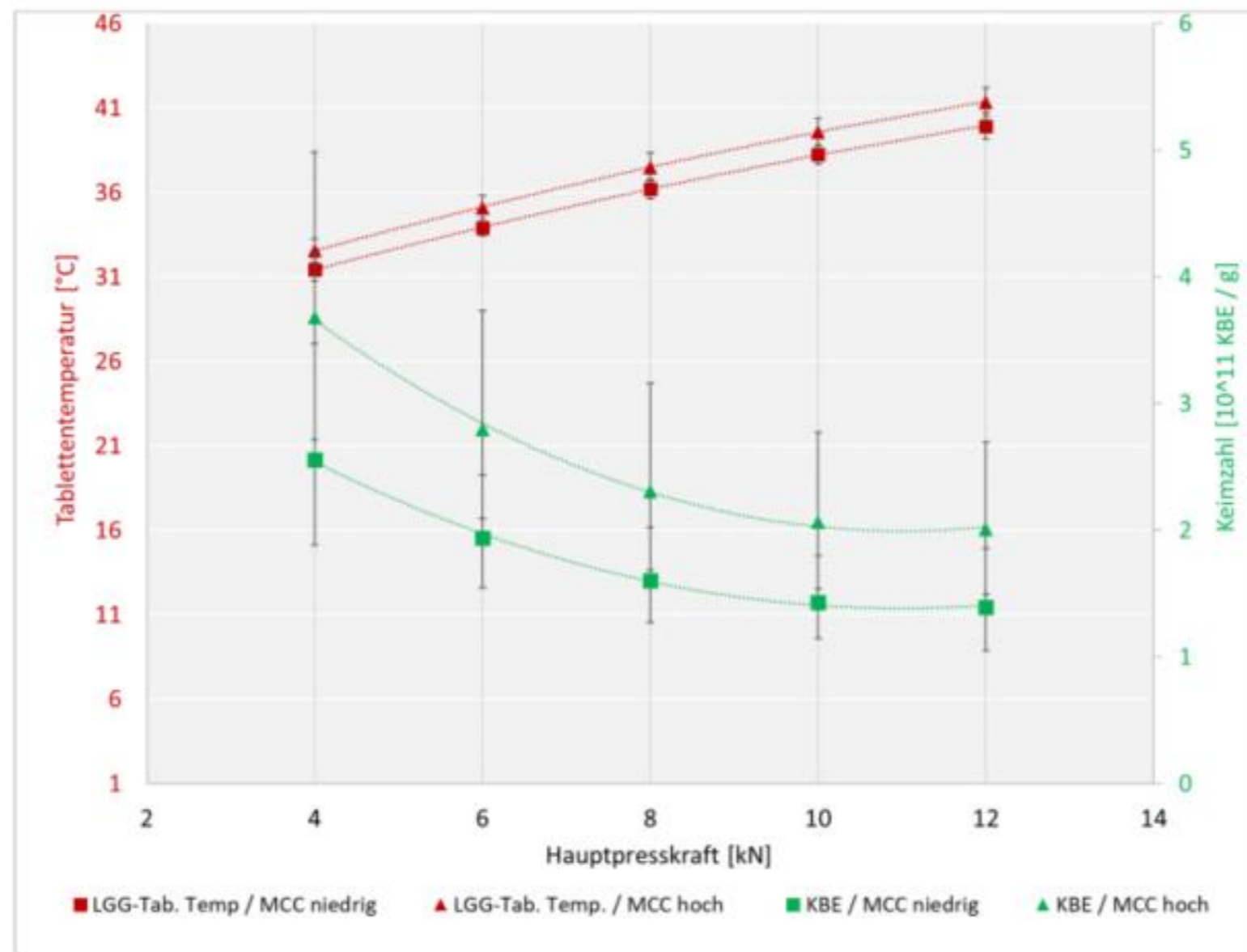
# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- The influences of the binders were not shown in the previous graph and are summarised below (data generated from data modelling)



# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

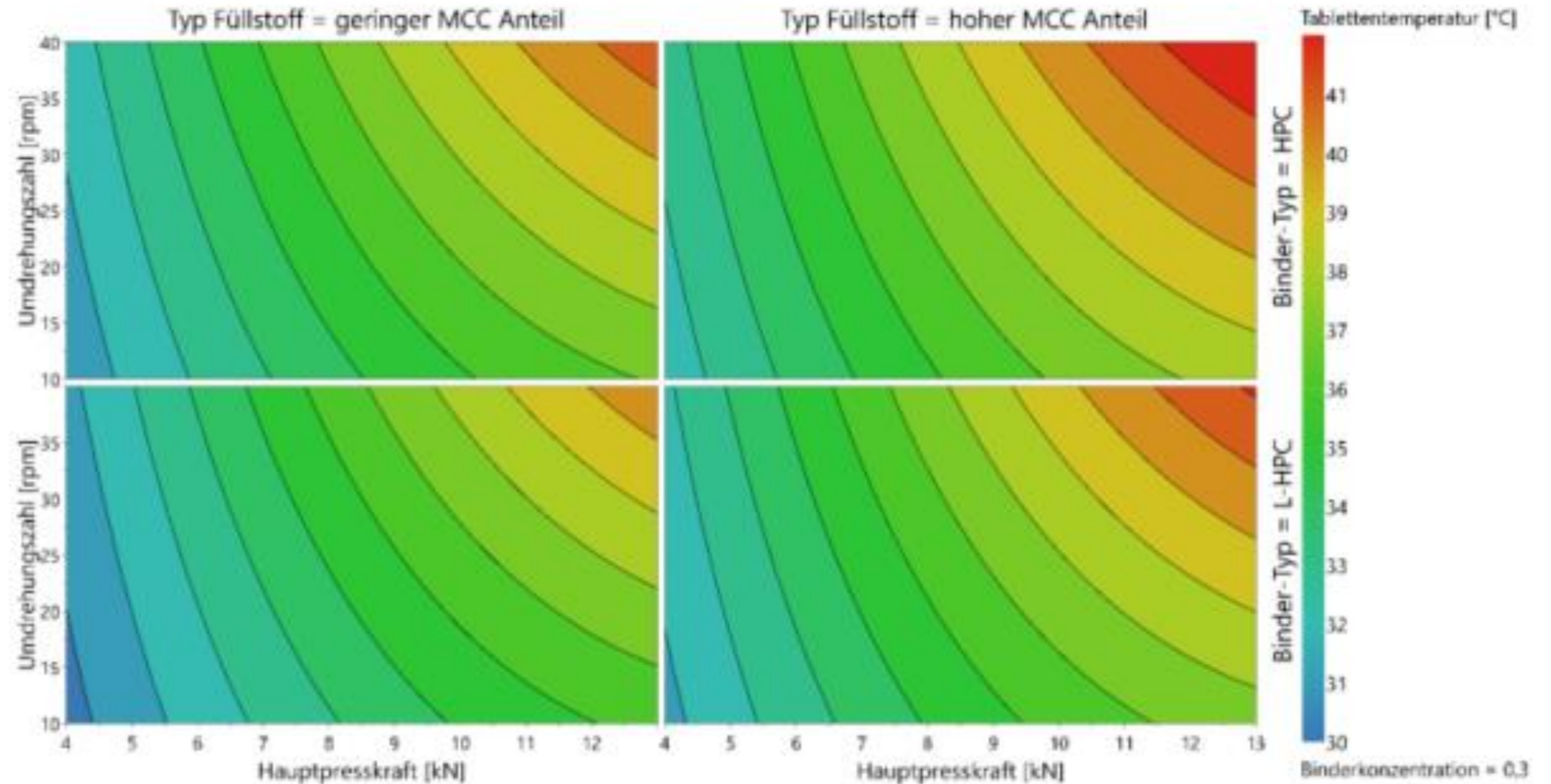
- Looking closer at the influence of tablet temperature we see that there is a correlation between viability and compaction force but little correlation between tablet temperature and viability.





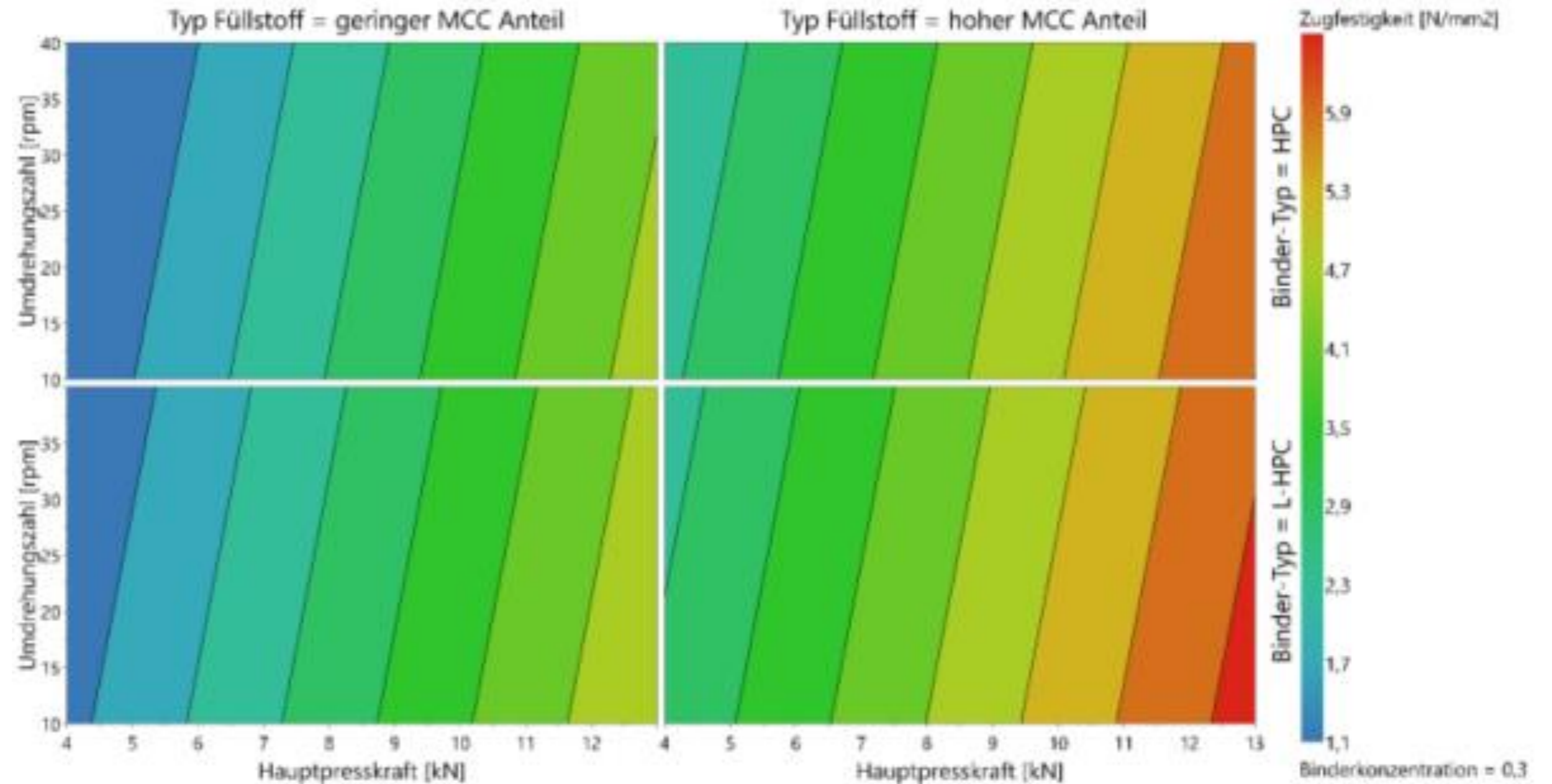
# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- Tablet temperatures were higher with the plastically deforming (high MCC) mixtures
- This is congruent with the literature and is correlated to the deformation behaviour and heat capacity of the used materials
- As shown on the previous slide the higher temperature of the tablets with the high MCC content did not have a negative effect on bacteria survival



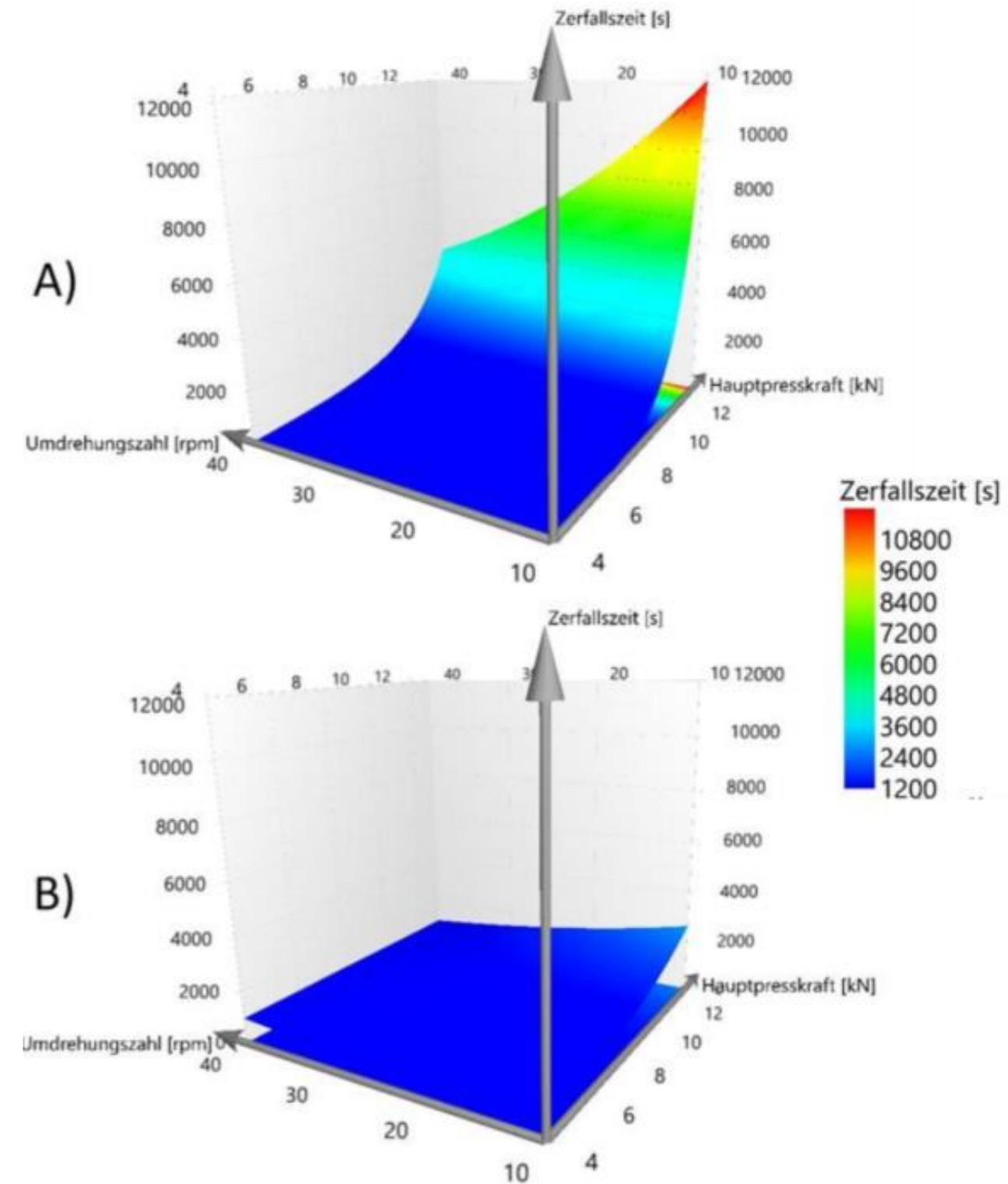
# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- As expected the plastically deforming mixtures yielded a higher tablet hardness than those made with brittle components
- The binder had little effect at the shown concentration of 0,3 %
- Increasing the tableting speed resulted in decreased tablet hardness



# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*: Results

- Surprisingly the use of L-HPC (A) resulted in a strong delay of disintegration times at high compaction forces in comparison to the use of HPC (B)
- At lower compaction forces the use of the binders had little effect on the disintegration times



# Influences of tableting temperature and compression force on the survival of *L. Rhamnosus*

- It was found that the main compression force plays the most important role regarding the viability of the bacteria
- However, formulation factors were of great significance too
- Brittle (hard) materials seem to be very unfavorable for use with live microorganisms
- It can be assumed that the microorganisms get ripped apart during the compaction with such materials due to sharp particle edges
- Higher compaction speeds had a positive impact by reducing the load impact time on the bacteria
- Increasing the binder concentrations had an adverse effect on the viability

<b>LGG-Tabletten</b>	<b>Keimzahl</b>
<b>Typ Füllstoff</b>	+
<b>Hauptpresskraft</b>	+++
<b>Umdrehungszahl</b>	+
<b>Bindertyp</b>	+
<b>Binderkonzentration</b>	++
<b>HPK<sup>2</sup></b>	+++
<b>Interaktion HPK+ UDZ</b>	++



# Formulation Optimization

# Formulation Optimization

Aims: Improve compaction behaviour of mixture at low compaction force  
Control: Tablet Tensile Strength, Friability, Disintegration Time, Powder Flow

Tablet Compositions:

Compaction Force	MCC	Magnesium Carbonate	Isomalt	preg Starch	Psyllium Husk	Oat Fibre	Inulin Powder	LGG	Mg-Stearate	HPC
kN	%	%	%	%	%	%	%	%	%	%
4	50 - 90	0-5	0 - 25	0- 25	0-5	0 - 5	0 - 5	10	1	0,5 - 2,5

Tableting Parameters:

200 mg Tablets, RoTab T rotary lab press; convex punches;  $\varnothing = 7$  mm; feeding speed 20 rpm, turret speed 25 rpm

# Formulation Optimization

- Powder flow was sufficient for all mixtures with only minor differences between the mixtures
- Friability was also low for most tablets
- The Ejection force was negligible for all formulations

Exp Name	AOR [°]	Funnel Flow [g/min]
N1	30,1	4,95
N2	30,5	4,75
N3	29,2	4,70
N4	30,5	4,75
N5	31,0	4,05
N6	30,5	5,75
N7	30,5	4,75
N8	30,5	5,45
N9	30,5	4,50
N10	30,1	4,55
N11	30,1	4,40
N12	29,7	3,35
N13	30,5	4,15
N14	30,5	4,05
N15	30,1	3,95

Exp Name	Friability [%]	Ejection Force [N]
N1	0,003051	0,01
N2	0,411795	0,01
N3	0,045326	0,01
N4	0,001	4
N5	0,156867	0,27
N6	0,001	0,01
N7	0,001	0,01
N8	0,001	0,01
N9	0,319969	0,01
N10	0,236438	0,01
N11	0,001	0,01
N12	0,097356	0,01
N13	0,001	0,01
N14	0,001	0,01
N15	0,001	0,01

# Formulation Optimization

- While little variation was observed for EJF, powder flow and friability large differences were observed for the tablet hardness and disintegration times
- Therefore, these were subjected to further data modelling and optimization

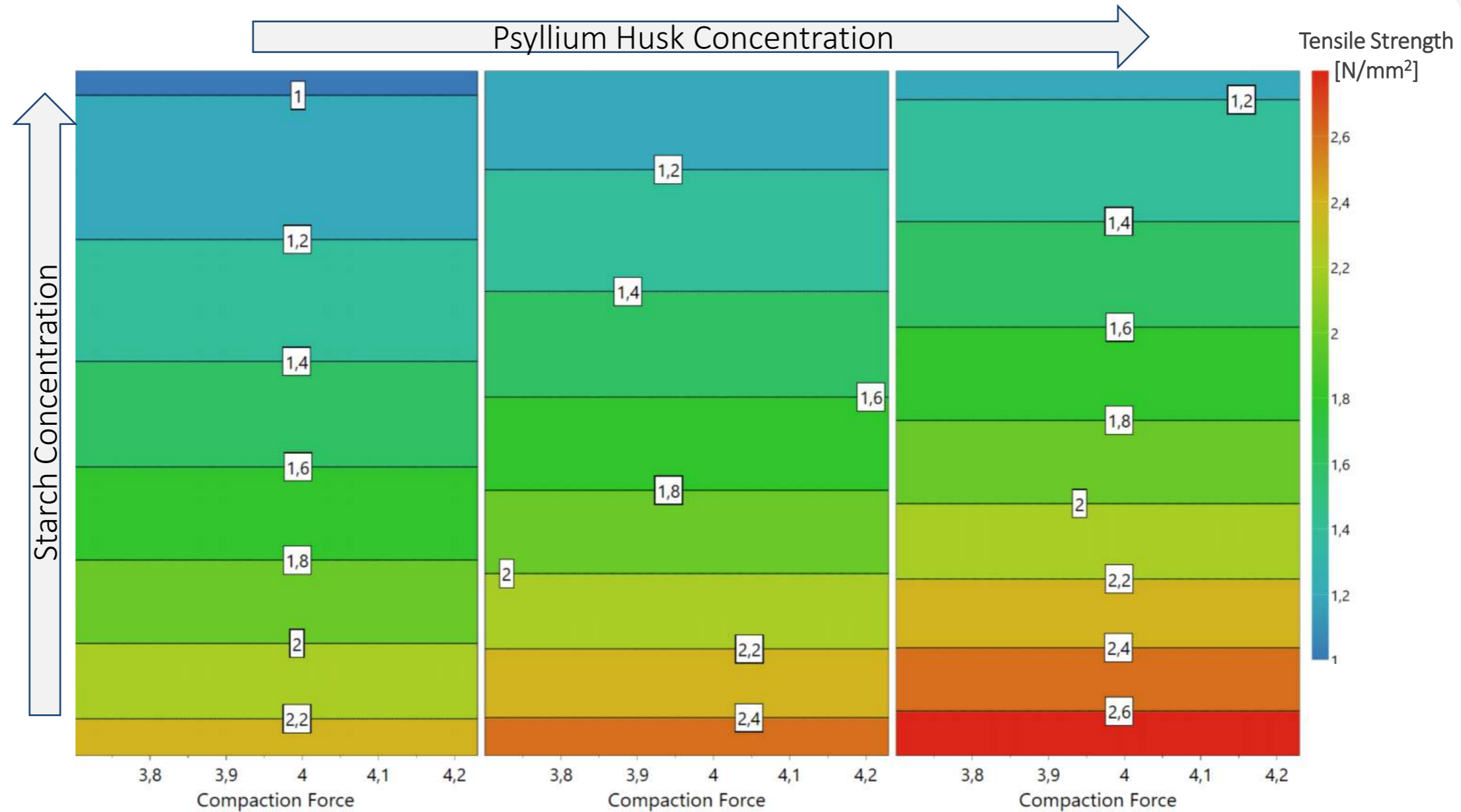
Exp Name	Tensile Strength [N/mm <sup>2</sup> ]	Disintegration time [min]
N1	2,031	2,110
N2	0,732	11,790
N3	2,476	11,170
N4	2,536	14,900
N5	1,214	4,483
N6	2,441	15,833
N7	2,915	8,372
N8	2,548	19,906
N9	0,825	6,944
N10	0,882	13,144
N11	2,555	6,006
N12	0,936	18,256
N13	1,831	8,833
N14	1,572	11,294
N15	1,522	10,944





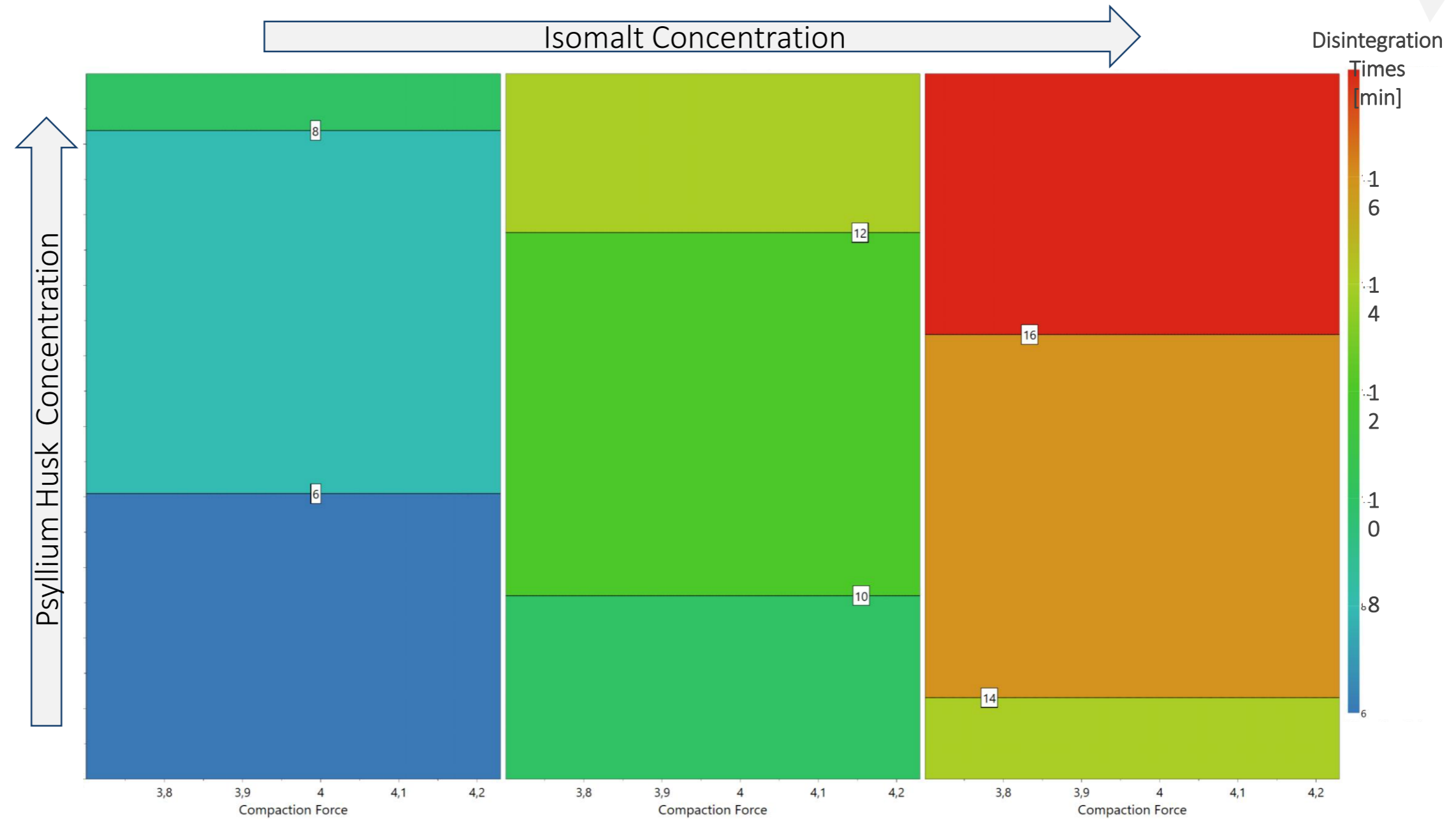
# Formulation Optimization

- It was found that the tensile strength of the tablets depends mostly on the isomalt, starch and psyllium husk concentrations



# Formulation Optimization

- It was found that the disintegration time of the tablets depends mostly on the isomalt and psyllium husk concentrations





# Compaction Simulation

# Compaction Simulation



**Aims:** Checking if the last two optimization candidates work well in high speed industrial tableting processes

Evaluate if high speed tableting processes have unexpected / un-modelled effects on bacteria viability

**Tablet Compositions:** Two formulations that contain MCC, Magnesium Carbonate, Isomalt, pregelatinized starch, psyllium husk, oat fibre, inulin powder, L. rhamnosus, Mg-St and HPC super fine powder

**Tableting Parameters:** StylOne Evolution compaction simulator ; convex punches;  $\varnothing = 7,0$  mm; Main Compression Force 1, 2, 3, 4, 5 kN, Pre Compression Force 20% of MCF ; 200 mg tablets

# Compaction Simulation

- A StylOne Evolution compaction simulator was used to simulate the compaction behaviour on a Romaco – Kilian KTP 720X 85 high speed press

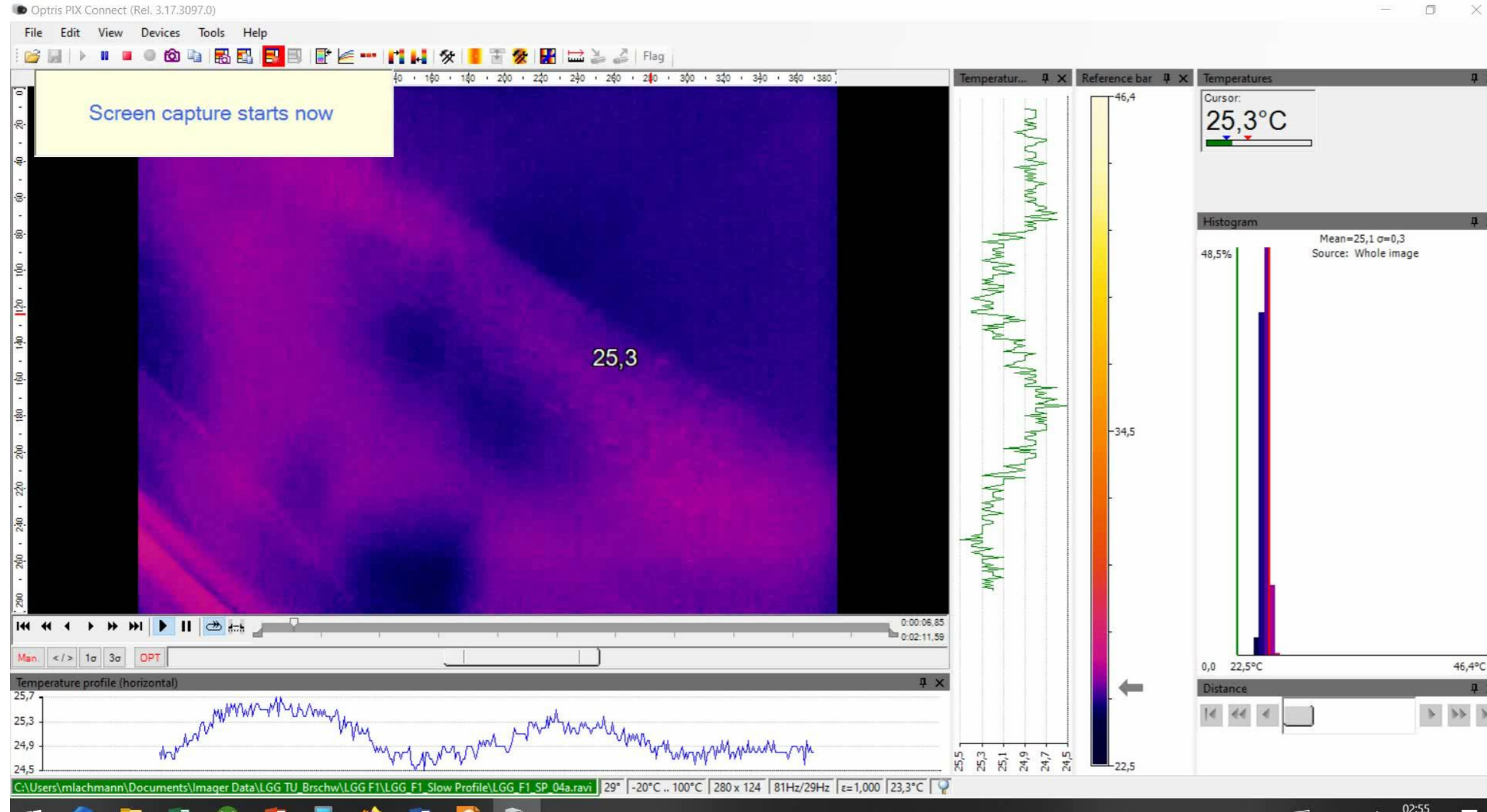
Technical Data	KTP 720X			
Machine configurations	Mono-layer; Bi-layer; Core			
Number of press stations	85	77	63	51
Tool type* (EU and TSM)	B	B	B	D
Die type	BBS	BB	B	D
Maximum tablet diameter (mm)	11	13	16	25
Maximum output** (tablets/hour)	1,020,000	924,000	753,000	550,000
Maximum die filling (mm)	18		20	
Maximum pre-compression force (kN)	100***			
Maximum main compression force (kN)	100***			
Power (kW)	38			
Standard voltage (V), frequency (Hz)	400, 50/60			
Compressed air (bar)	6			
Weight (kg)	5,600			



file:///C:/Users/mlachmann/Downloads/Romaco\_DB\_Kilian\_KTP\_720X-EN.pdf

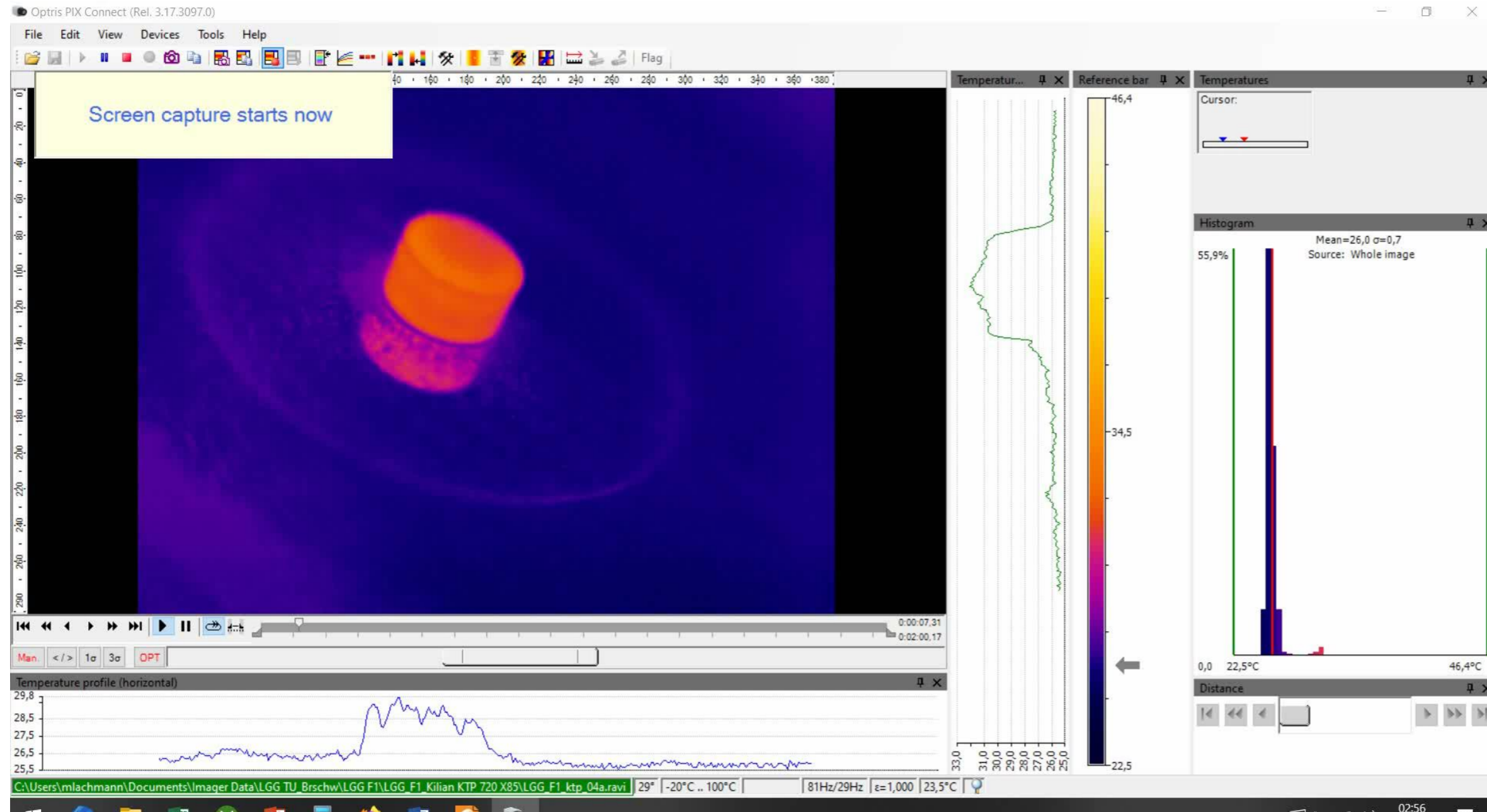
# Compaction Simulation

- Video Capture of slow profile tableting on a Medelphamr StylOne compaction simulator



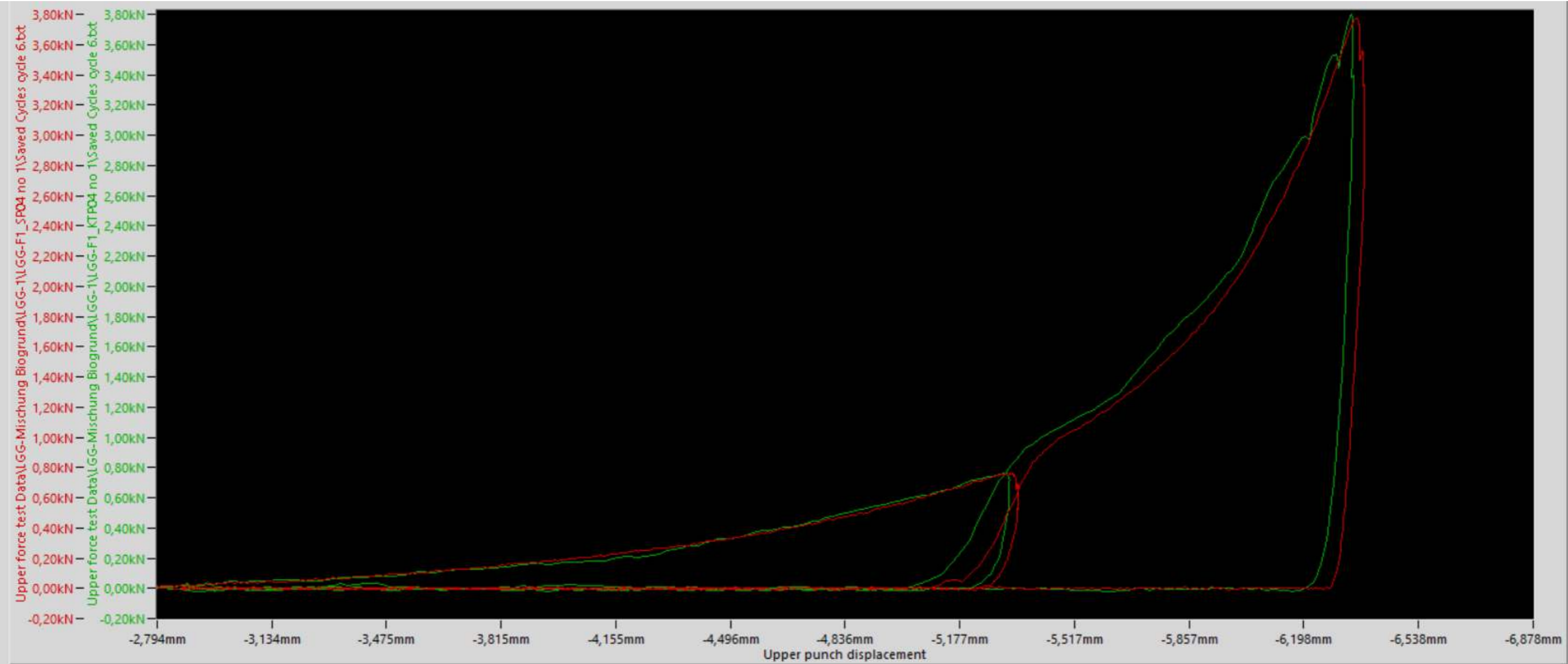
# Compaction Simulation

- Video Capture of the simulation of a KTP 720X high speed press



# Compaction Simulation

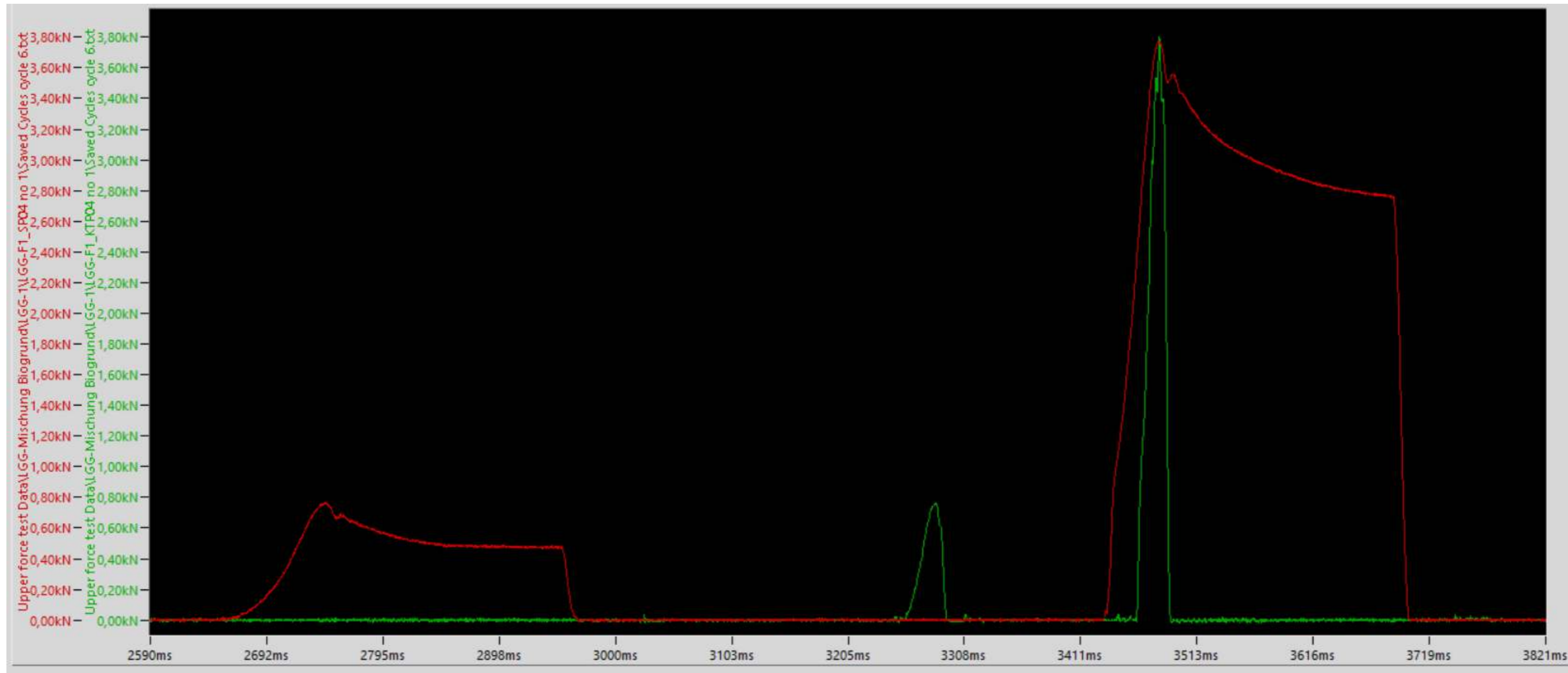
- Force – Distance profiles for a slow and fast compaction process at 4 kN MCF





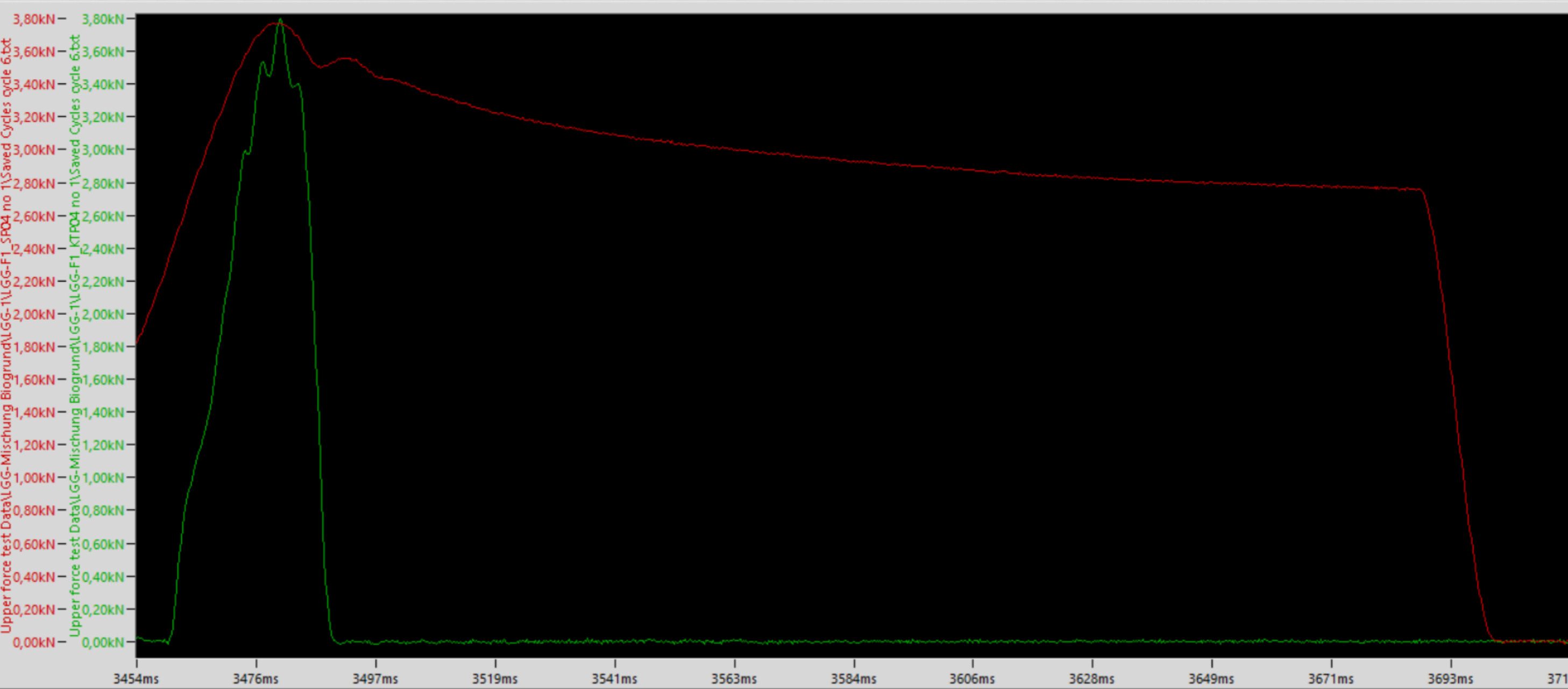
# Compaction Simulation

- Force – Time curve for a slow and fast compaction process at 4 kN MCF

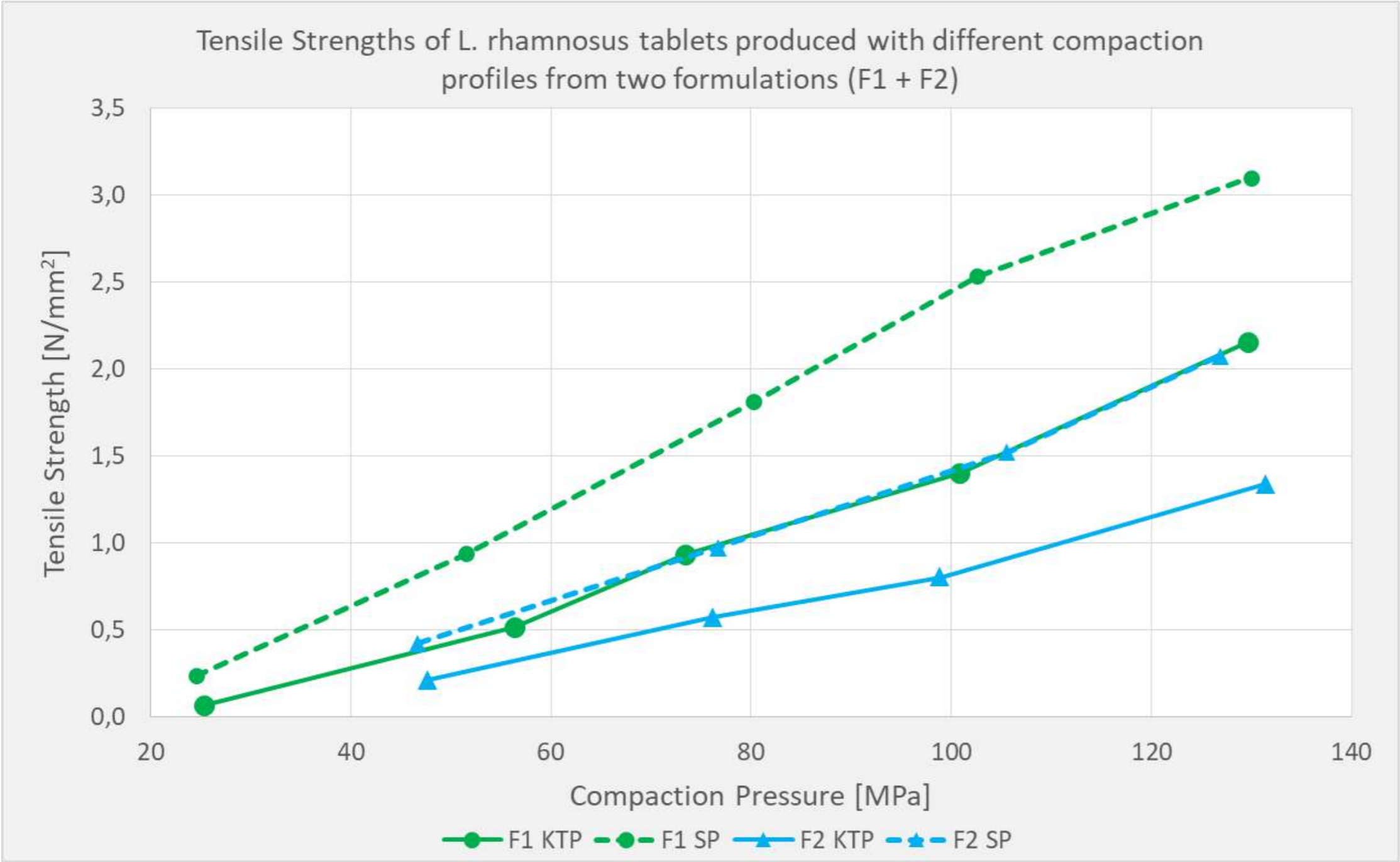


# Compaction Simulation

- Dwell time from 10 ms to 200 ms



# Compaction Simulation

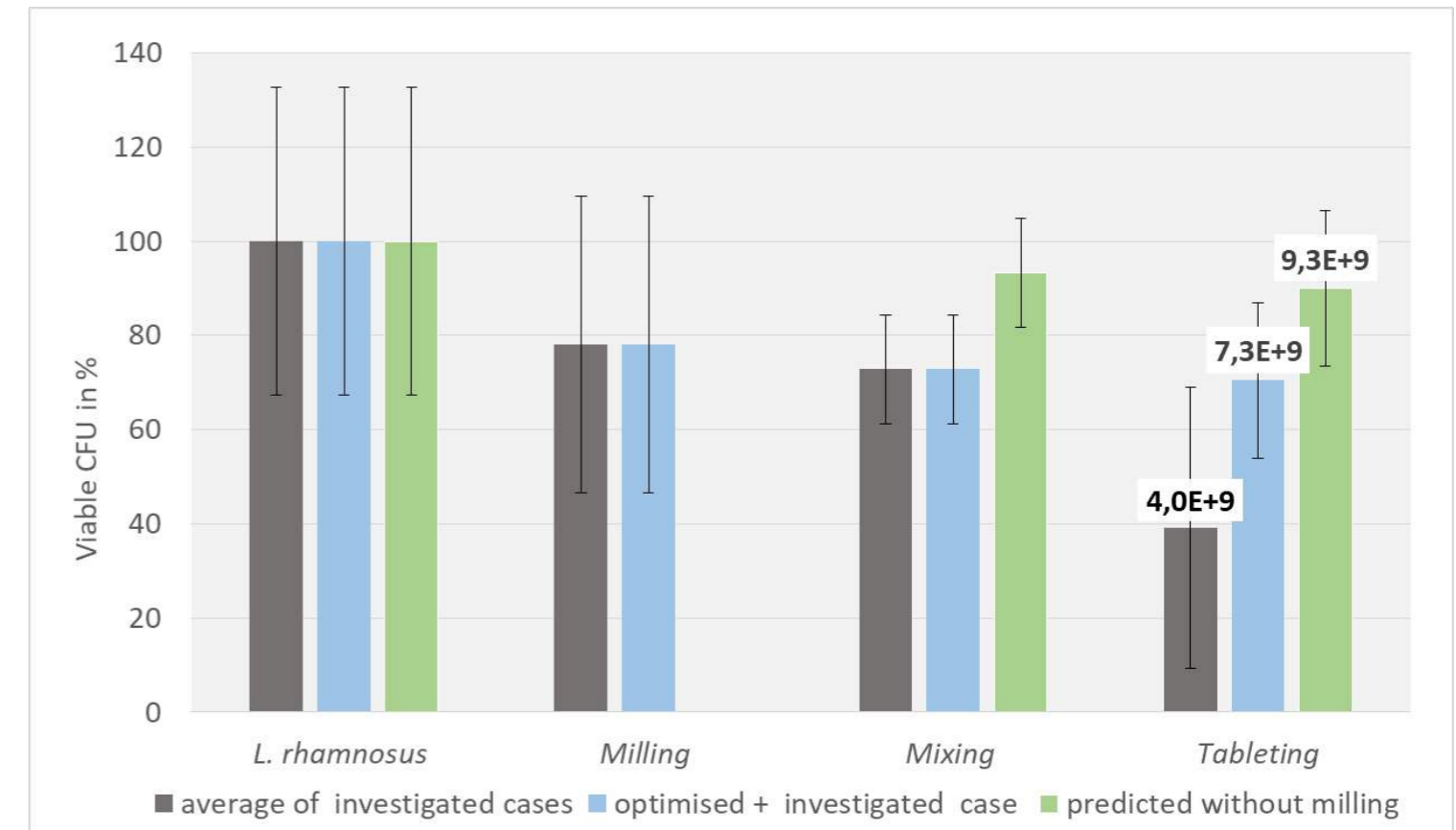




## New BonuTab<sup>®</sup> for Probiotics

# NEW BIOGRUND BonuTab<sup>®</sup> XXX.xx

- A new BonuTab<sup>®</sup> type for tableting of probiotic bacteria
- The new BonuTab<sup>®</sup> offers compaction at high speed and low compaction forces
- The mixture was optimized to enhance survival of bacteria
- So far the mixture was only tested with *L. rhamnosus* but we are confident that it should work with other probiotic strains too
- The mixture also contains the prebiotic inulin
- According to our concurrent investigation and data modelling loss of bacteria can be reduced to roughly 20 % even during high speed tableting



<b>MCC</b>	<b>Oat Fibre</b>
<b>Magnesium Carbonate</b>	<b>Inulin powder</b>
<b>Isomalt</b>	<b>LGG</b>
<b>preg Starch Lycatab</b>	<b>Mg-Stearate</b>
<b>Psyllium Husk</b>	<b>HPC</b>



# Publication

# Publication

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## Survival of *Lactobacillus Rhamnosus* in a Tableting Process

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### Introduction:

Probiotics are living microorganisms that have been proven to confer beneficial effects on overall health and immunity<sup>1,2</sup>. *Lactobacillus Rhamnosus* (GC 822), a probiotic strain with many reported health benefits<sup>3,4</sup> was chosen for the present study.

From selection to the packaging and transport of the final dosage forms, probiotic bacteria are subjected to extensive chemical, thermal and mechanical stress that can lead to significant reduction of the CFU stability.

Most of the probiotic formulations available on the market today are hard capsules, sometimes reducing the mechanical stress during the dosage form production steps. However, tablets are more cost-effective solutions to manufacture than capsules<sup>5,6</sup>. The aim of the presented work was to investigate the influence of tableting process parameters and recipe composition on GC 822 survival.

### Materials:

All materials were used as delivered by the manufacturers. *Lactobacillus Rhamnosus* GC822 (Strain 822) (Probiotec AG, Bielefeld, DE), CAPTOP® 4120 (DCCG Pharma GmbH, Germany), Compocel® 200 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 200 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 300 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 400 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 500 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 600 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 700 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 800 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 900 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 1000 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 1200 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 1400 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 1600 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 1800 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 2000 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 2200 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 2400 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 2600 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 2800 (MCC (Christoph Chemiefabrik GmbH, Germany), Compocel® 3000 (MCC (Christoph Chemiefabrik GmbH, Germany).

### Methods:

#### Formulation (Basic)

100 mg GC 822 lyophilate was added to 10 ml of purifier water, containing 0.05 % NaCl and 0.1 % of preservative. The mixture was prepared before the bacterial count was determined (using an optical density measurement). The suspension was prepared using the 'back-to-front' method with 1 ml of the sample and 9 ml of water. The CFU count for suspensions and tablets was determined (dilution by using the equivalent mass of 1 g GC) comprising 10 % of powder content. The procedures were conducted at 20°C for 24h.

#### Tableting (Basic)

Portable size of GC 822 lyophilate was enhanced to improve powder handling and filling accuracy (5 mm manual milling in a mortar and passage through a 200 µm sieve). The milled material was prepared with a water-soluble binder (10% w/w) to improve powder flow and to avoid lumping. Finally, the GC 822 material was combined with the remaining components of the tablet formulation as shown in table 1.

Table 1: Formulation Compositions in %

Ingredient	F1	F2	F3	F4	F5	F6	F7
GC822	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10
Excipient	10	10	10	10	10	10	10

#### Tablet Production and Analysis

Tablets with a target mass of 100 mg (100 µm) were produced on a Fette 25 rotary tableting press (Göteborg, Germany) (Fette 25, rotary press). Compression and ejection forces were monitored at all times. Tablet temperatures were measured using a Delta 10 (Delta 10, Germany). Tablet length and diameter were measured using a digital indicator CH 200 (Mitutoyo, Germany). Tablet breaking force was determined with a FTB 1110 (FTB, Germany). For application, GC 822 material was added to the corresponding granules, stored and using the 30-minute procedure. Finally, GC 822 material was used to create a functional bacterial formulation design. Main Compression Pressure was varied from 1 to 12 kN depending on 100 to 500 MPa. For Compression Pressure was kept constant at 1 kN (100 MPa). Tableting speed was varied between 10 and 30 rpm.

#### Results

All formulations yielded tablets with a sufficient tensile strength (1.5 N/mm) and a low friability (0.1 %). The main driver for the survival of GC 822 in the course of tableting procedure was more likely mechanical stress during the tableting process (as shown in Figure 1) in relation to the starting material, having a CFU count of 10<sup>10</sup> CFU/g. The type of binder also having an effect while the mechanical stress of the tablet material in the process is. Particularly, there is a small difference in survival, with higher survival during more GC 822 content. Most likely the effect of the pressure on the bacteria is reduced at shorter dwell times.

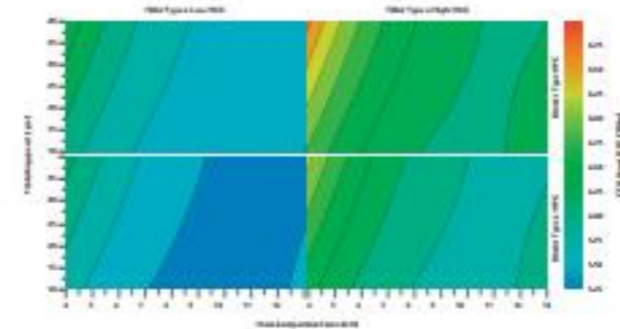


Figure 2: CFU counts of tablets

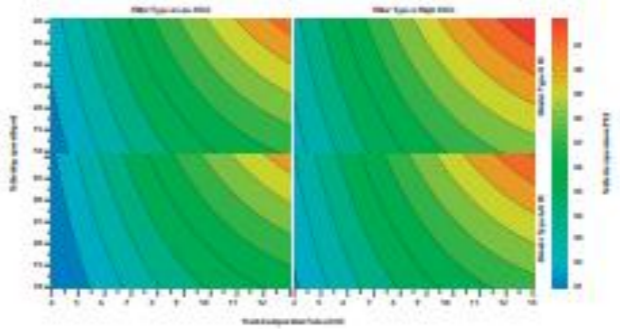


Figure 3: Tablet temperatures

For all formulations, the tablet temperature increased with higher compression pressure and increased tableting speed (Figure 3 and 4). However, within the temperature range reached in this study, the influence of the GC 822 content on survival was not significant. In general, the bacterial counts with higher MCC content yielded higher tablet temperatures. This being evaluated in different tableting conditions and energy for compression of particles due to differences in particle shape. It was found that binder types have only a minimal influence on tablet temperatures.

Figure 3 shows an example of the thermal imaging for formulation F1 at 1 kN (100 MPa), 30 rpm. The temperature of the tablet is higher on the top of the tablet and lower in the tableting region. Therefore, it can be concluded for the investigated, well-hydrated formulation, that the distribution is not the main reason for heat generation. From the point of distribution, the tablet undergoes a change in temperature profile with increasing compression force, being reached when the tablet enters during the compression. This can be seen as an additional indicator that the main source of heat generation can be prepared in the tableting and deformation of the particles inside the tablet.

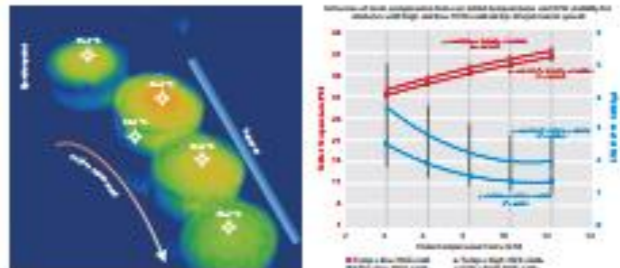


Figure 4: Thermal imaging of Tablets

#### With each processing step a loss of viable bacteria can be observed.

Especially for the milling process, and the tableting process results in a high mortality of GC 822. Figure 5 shows the loss of viable bacteria during the process. The survival of GC 822 in the course of tableting procedure was more likely mechanical stress during the tableting process (as shown in Figure 1) in relation to the starting material, having a CFU count of 10<sup>10</sup> CFU/g. The type of binder also having an effect while the mechanical stress of the tablet material in the process is. Particularly, there is a small difference in survival, with higher survival during more GC 822 content. Most likely the effect of the pressure on the bacteria is reduced at shorter dwell times.

Figure 5: Loss of Viability of *L. Rhamnosus* from raw material in tablet dosage forms (GC content per tablet as data taken) up to 30 min of tablet tableting.

#### Conclusion

The survival of the bacteria depends on the formulation ingredients whereas "water" materials are being preferable. Compression pressure is having a major impact on the survival of the bacterial and therefore it is desirable to use moisture materials to increase the production of tablets of sufficient tensile strength at low compression pressure. However, dwell times seem to have a positive effect on bacterial survival. This is especially important since higher tableting speed leads to more cost-effective production of probiotic formulations.

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#### References

- (1) Liu, G. et al. Efficacy of probiotics in human gut microbiome-associated diseases. *Life*, United Kingdom, 2018, **8**, 1330-1348 (2018).
- (2) Capone, L. et al. *Lactobacillus Rhamnosus* GC 822. *Clinical Gastroenterology*, **23**, 63-63 (2019).
- (3) Radler, G. et al. Advancements in the Microencapsulation of Probiotic Dosage Forms and Formulation Technology. *Int J Pharmaceut*, **451**, 105-124 (2018).
- (4) Gaudin, L. et al. Temperature changes during tableting measured using infrared thermography. *Int. J. Pharm.*, **351**, 132-144 (2008).



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