Automated cell treatment for competence and transformation of *Escherichia coli* in high throughput

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Source Code S1: Hamilton Script Overview

	Method
1	Assignment 'main_runID' = '325'
2	HSL code. main_current_date_str = TimGetFormattedDate("%Y-%m-%d") + "_" + TimGetFormattedTime("%H_%M_%S");
3	Assignment 'main_cultivation_dilution' = '10'
4	Assignment 'main_cultivation_cycletime_min' = '60'
5	Assignment 'main_incubation_onIce' = '1800'
6	Assignment 'main_Operator_Number' = "491775724287"'
7	Grouping Konstanten
50	Grouping Initialize
61	Grouping Cultivation
138	Grouping Filtrieren
150	Grouping Resuspendiren
159	Grouping Incubation on ice
164	Grouping Transformation
174	Grouping Incubation on Ice & Prepair fresh media plate
181	Grouping Heatshock
189	Grouping Cultivation2
198	Grouping Ausplatieren
231	

Source Code S2: Hamilton Script detail

	Method
1	X=0 Assignment 'main_runID' = '325'
2	HSL code. main_current_date_str = TimGetFormattedDate("%Y-%m-%d") + "_" + TimGetFormattedTime("%H_%M_%S");
3	X=0 Assignment 'main_cultivation_dilution' = '10'
4	X=0 Assignment 'main_cultivation_cycletime_min' = '60'
5	X=0 Assignment 'main_incubation_onIce' = '1800'
6	X=0 Assignment 'main_Operator_Number' = '"4917757 24287"'
7	Grouping Konstanten
8	X=0 Assignment 'main_Gen5_PlateCounter' = '2'
9	X=0 Assignment 'main_PreCulture_KryoVol' = '5'
10	Comment <main_PreCulture_IncubationTime in Hours!>
11	Array: Declare / Set Size Set array 'main_PreCulture_IncubationTime' to empty size.
12	X=0 Assignment 'main_PreCulture_dilution' = '10'
13	X=0 Assignment 'main_cultivationVol' = '170'

14	HSL code.
	// Berechne Preculture Media Volume
	main_PreCulture_MediaVol = main_cultivationVol - main_PreCulture_KryoVol;
	
	// Teile PreCulte Cultivation Time in 3 Intervalle ein (1 /3,5 / 3,5 = 8)
	// Umrechnen der Kultivierungszeit von Stunden in Sekunden
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime.AddAsLast(3600);
	main_PreCulture_IncubationTime_temp = 0;
	
	main_PreCulture_LoopNum = main_PreCulture_IncubationTime.GetSize();
	
	// Rechne Verdünnungen für die ODMessung aus
	main_PreCulture_VolSample = main_cultivationVol / main_PreCulture_dilution;
	main_PreCulture_VolNaCl = main_cultivationVol - main_PreCulture_VolSample;

15	X=0 Assignment 'main_reader_RowName' = '"OD600"'
16	Array: Declare / Set Size Set array 'main_reader_PreCultureValues_1' to empty size.
17	Array: Declare / Set Size Set array 'main_reader_PreCultureValues_2' to empty size.
18	Array: Declare / Set Size Set array 'main_reader_PreCultureValues_3' to empty size.
19	X=0 Assignment 'main_OD600_threshold' = '0,041'
20	X=0 Assignment 'main_OD600_correctionFactor' = '2'

	Method
21	X=0 Assignment 'main_cultivation_VolPreCulture' = '10'
22	HSL code. main_cultivation_VolMedium = main_cultivationVol - main_cultivation_VolPreCulture;
23	X=0 Assignment 'main_cultivation_VolSample' = '20'
24	HSL code. main_cultivation_VolNaCl = 200 - main_cultivation_VolSample;
25	X=0 Assignment 'main_cultivation_slot' = '4'
26	X=0 Assignment 'main_cultivation_temp' = '37'
27	X=0 Assignment 'main_cultivation_speed' = '1000'
28	X=0 Assignment 'main_reader_path2resultsFolder' = '"C:\\Dokumente und Einstellungen\\Peter Neubauer\\Eigene Dateien\\Sebastian\\Robot"'
29	X=0 Assignment 'main_reader_path2experimentFolder' = '"C:\\Programme\\BioTek\\Gen5 1.09\\Experiments\\Sebastian\\Robot"'
30	HSL code. Shell("C:\WINDOWS\system32\\cmd.exe /C mkdir \"" + main_reader_path2resultsFolder + "\\" + main_current_date_str + "\"", 2, 1); Shell("C:\WINDOWS\system32\\cmd.exe /C mkdir \"" + main_reader_path2experimentFolder + "\\" + main_current_date_str+ "\"", 2, 1); Shell("C:\WINDOWS\system32\\cmd.exe /C copy \"" + main_reader_path2experimentFolder + "\\" + "_default_OD600.xpt\"" + " \"" + main_reader_path2experimentFolder + "\\" + main_current_date_str + "\\" +
31	X=0 Assignment 'main_cultivation_abort' = '0'
32	X=0 Assignment 'main_filter_VolFromCultur' = '180'

33	HSL code.
	// Rechne Cycletime in Sec
	main_cultivation_cycletime_sec = main_cultivation_cycletime_min * 60;
	
	// Ermittle wieviele Stämme vorhanden sind
	main_numStrains = ML_STAR._Res_Kryos.GetTotal();

34	X=0 Assignment 'main_pump_ID' = '1'
35	X=0 Assignment 'main_pump_COMPort' = '3'
36	X=0 Assignment 'main_pump_dPressure' = '200'
37	X=0 Assignment 'main_pump_tresholdPressure' = '10'
38	X=0 Assignment 'main_filter_duration' = '20'
39	X=0 Assignment 'main_wash_Volume' = '200'
40	X=0 Assignment 'main_wash_cycles' = '3'
41	X=0 Assignment 'main_resuspend_Vol' = '150'
42	X=0 Assignment 'main_incubation_time1' = '7200'
43	X=0 Assignment 'main_dna_Vol' = '2'

	Method
44	Assignment 'main_incubation_heatShock' = '120'
45	Assignment 'main_cultivation2_Vol' = '150'
46	HSL code. main_cultivaiton2_VolKultur = 200 - main_cultivation2_Vol;
47	Assignment 'main_cultivation2_time' = '3600'
48	Assignment 'main_plating_vol' = '200'
49	Grouping
50	Grouping Initialize
51	If, Else (main_runID is NOT equal to 0)
52	iLAB_connect of BVTlib_ilab_connector iLAB_connect()
53	getBioreactorIDs of BVTlib_ilab_connector main_bioID_array = getBioreactorIDs(main_runID)
54	Assignment 'main_bioID' = 'main_bioID_array[1]'
55	End If

56	HSL code.
	main_StatusLTU = MessageBox("Is it Plate of the LTU on the site of the Hamilton?","Status LTU", 4);
	if (main_StatusLTU == 6) {
	Position_Tecan();
	}

57	Initialize (Single Step) on ML_STAR Always initialize: Off 3 return value(s)
58	initialize_Fame of unifiedlibrary UNIFIEDLIBRARY::initialize_Fame(ML_STAR, main_cultivation_temp, 2, main_cultivation_speed, ML_STAR._Plate_Cultivation)
59	BVSInitialize of HSLVacuuBrandPump HSLStarBVSLib::BVSInitialize(main_pump_ID, main_pump_COMPort)
60	Grouping
61	Grouping Cultivation
62	Comment <Prepair PreCulture Plate>
63	Sub_01_AddFromContainer_sterilTips of KompetenteZellen_v0.0.1 Sub_01_AddFromContainer_sterilTips(ML_STAR._Res_Medium, ML_STAR._Plate_Preculture, main_PreCulture_MediaVol)
64	Sub_02_AddFromSequence of KompetenteZellen_v0.0.1 Sub_02_AddFromSequence(ML_STAR._Res_Kryos, ML_STAR._Plate_Preculture, main_PreCulture_KryoVol)
65	Comment <Start Preculture>
66	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(2, ML_STAR._Plate_Preculture, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
67	Timer: Start Start timer 'main_timer_preculture', set to relative time: '0' [s]

	Method
68	Cleaning_0050_Tips of SM_SH_Cleanning SM_SH_CLEANNING::Cleaning_0050_Tips(ML_STAR, ML_STAR._Tips_0050_1, ML_STAR._Res_EtOH, ML_STAR._Res_EtOH_Lid, ML_STAR._Res_EtOH_Lid_Storage,
69	sendWhatsApp of SM_Gluocse SM_GLUOCSE::sendWhatsApp(main_Operator_Number, "First Timer is started ...")
70	Loop 'main_PreCulture_LoopNum' times 'main_PreCulture_Loop' used as loop counter variable
71	Grouping Timer
72	Timer: Wait for Wait for timer 'main_timer_preculture', showtimer display, is stoppable
73	Timer: Start Start timer 'main_timer_preculture', set to relative time: 'main_PreCulture_IncubationTime[main_PreCulture_Loop]' [s]
74	sendWhatsApp of SM_Gluocse SM_GLUOCSE::sendWhatsApp(main_Operator_Number, "Next Timer is started ...")
75	Grouping
76	Comment <Take new Plate>
77	1000µl Channel CO-RE Grip Get Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_Storage, Sequence counting: (1) Automatic, Channel to be used: 8 3 return value(s)
78	1000µl Channel CO-RE Grip Place Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_working, Sequence counting: (0) Manually, Eject tool: (1) Yes 3 return value(s)
79	Sub_01_AddFromContainer of KompetenteZellen_v0.0.1 Sub_01_AddFromContainer(ML_STAR._Res_NaCl, ML_STAR._Plate_working, main_PreCulture_VolNaCl)
80	Comment <Hole Platte aus dem Inkubator>
81	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(1, ML_STAR._Plate_FromIncubator, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
82	Comment <Messe OD>
83	Sub_02_AddFromSequence_96 of KompetenteZellen_v1.1.0 Sub_02_AddFromSequence_96(ML_STAR._Plate_FromIncubator, ML_STAR._Plate_working, main_PreCulture_VolSample)
84	Comment <Lege Platte zurück in den Inkubator>
85	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(2, ML_STAR._Plate_FromIncubator, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
86	Grouping ReaderMessung und Daten erreichnen

	Method

87	HSL code.
	// Erstelle Path zum Experiment
	main_reader_path2experiment = main_reader_path2experimentFolder + "\\" + main_current_date_str + "\\" + main_current_date_str + "_OD600.xpt";
	// Erstelle Phat zur Exceldatei mit den Ergebnissen
	main_reader_path2results = main_reader_path2resultsFolder + "\\" + main_current_date_str;
	
	main_reader_filenameResults = main_current_date_str + "_OD600_Preculture_" + IStr(main_PreCulture_Loop);
	main_reader_TabName = "Plate " + IStr(main_Gen5_PlateCounter) + " - Data";
	main_Gen5_PlateCounter++;
	main_reader_filenameReturn = "C:\\Dokumente und Einstellungen\\Peter Neubauer\\Eigene Dateien\\Sebastian\\Robot\\" + main_current_date_str + "\\" + main_reader_filenameResults + ".xls";

88	Readermessung of unifiedlibrary UNIFIEDLIBRARY::Readermessung(ML_STAR, HxGen5, ML_STAR._Plate_working, main_reader_path2experiment, main_reader_path2results, main_reader_filenameResults, main_cultivation_dilution, main_cultivation_start, main_reader_filenameReturn)
89	Comment <Werte OD aus>
90	readRow of BVTlib_ExcelImport main_reader_resultes = readRow(main_reader_filenameReturn, main_reader_TabName, main_reader_RowName)
91	subArray of BVTlib_ExcelImport main_reader_resultesSub = subArray(main_reader_resultes, 1, main_numStrains)
92	subtractArray of BVTlib_ExcelImport main_reader_resultesSub = subtractArray(main_reader_resultesSub, main_OD600_threshold)
93	multiplyArray of BVTlib_ExcelImport main_reader_resultesSub = multiplyArray(main_reader_resultesSub, main_PreCulture_dilution)
94	multiplyArray of BVTlib_ExcelImport main_reader_PreCultureValues = multiplyArray(main_reader_resultesSub, main_OD600_correctionFactor)
95	HSL code. main_TimeStamp_ilab = TimGetFormattedDate("%Y-%m-%d") + " " + TimGetFormattedTime("%H:%M:%S");
96	If, Else (main_runID is NOT equal to 0)
97	sendData_long of BVTlib_ilab_connector sendData_long(main_runID, main_bioID, "OD600", main_reader_PreCultureValues, 1, main_TimeStamp_ilab, main_PreCulture_dilution)
98	End If
99	arrayMinValue of BVTlib_ExcelImport main_ODmin = arrayMinValue(main_reader_PreCultureValues)
100	If, Else (main_ODmin is greater than OR equal to 0,75)
101	Loop: Break
102	End If
103	Grouping
104	Grouping performing dilution

	Method
105	If, Else (main_runID is NOT equal to 0)
128	Grouping
129	Comment <Plate2waste>
130	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._Plate_working' to 'ML_STAR.platewaste' 1 return value(s) :
131	Cleaning_0050_Tips of SM_SH_Cleanning SM_SH_CLEANNING::Cleaning_0050_Tips(ML_STAR, ML_STAR._Tips_0050_1, ML_STAR._Res_EtOH, ML_STAR._Res_EtOH_Lid, ML_STAR._Res_EtOH_Lid_Storage, 1, 0)
132	Cleaning_0300_Tips of SM_SH_Cleanning SM_SH_CLEANNING::Cleaning_0300_Tips(ML_STAR, ML_STAR._Tips_0300_1, ML_STAR._Res_EtOH, ML_STAR._Res_EtOH_Lid, ML_STAR._Res_EtOH_Lid_Storage, 0, 1)
133	End Loop
134	Comment <Hole Plate zurück aus dem Inkubator>
135	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(1, ML_STAR._Plate_FromIncubator, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
136	sendWhatsApp of SM_Gluocse SM_GLUOCSE::sendWhatsApp(main_Operator_Number, "Cultivation ended")
137	Grouping
138	Grouping Filtrieren
139	Comment <Start filtration>
140	Sub_02_AddFromSequence_300 of KompetenteZellen_v0.0.1 Sub_02_AddFromSequence_300(ML_STAR._Plate_FromIncubator, ML_STAR._Plate_Filter, main_filter_VolFromCultur)
141	ReqActualPressure of HSLVacuuBrandPump main_temp = HSLVacuuBrandPump::ReqActualPressure(main_pump_ID)
142	BVSVacuum of HSLVacuuBrandPump HSLStarBVSLib::BVSVacuum(main_pump_ID, main_pump_dPressure, main_filter_duration, 1, main_pump_tresholdPressure, main_pump_returnPressure)
143	Loop 'main_wash_cycles' times 'main_Loop_Washing' used as loop counter variable
144	HSL code. Trace("CaCl - Loop"); ML_STAR._Plate_Filter.SetCurrentPosition(0);
145	Sub_01_AddFromContainer of KompetenteZellen_v0.0.1 Sub_01_AddFromContainer(ML_STAR._Res_CaCl2, ML_STAR._Plate_Filter, main_wash_Volume)
146	BVSVacuum of HSLVacuuBrandPump HSLStarBVSLib::BVSVacuum(main_pump_ID, main_pump_dPressure, main_filter_duration, 1, main_pump_tresholdPressure, main_pump_returnPressure)
147	End Loop
148	BVSTerminate of HSLVacuuBrandPump HSLStarBVSLib::BVSTerminate(main_pump_ID)
149	Grouping
150	Grouping Resuspendiren

	Method
151	HSL code. Trace("Resuspend - Loop"); ML_STAR._Plate_Filter.SetCurrentPosition(1); main_resuspend_LoopCounter = main_numStrains/8;
152	Loop 'main_resuspend_LoopCounter' times 'main_Loop_resuspend' used as loop counter variable
153	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Res_CaCl2, Volume [µl]: main_resuspend_Vol 0 return value(s) .
154	1000µl Channel Dispense on ML_STAR Sequence: ML_STAR._Plate_Filter, Volume [µl]: main_resuspend_Vol 0 return value(s) .
155	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Plate_Filter, Volume [µl]: main_resuspend_Vol 0 return value(s) .
156	1000µl Channel Dispense on ML_STAR Sequence: ML_STAR._Plate_PCR, Volume [µl]: Remaining volume inclusive blowout air 0 return value(s) .
157	End Loop
158	Grouping
159	Grouping Incubation on ice
160	Timer: Start Start timer 'main_timer_incutation', set to relative time: 'main_incubation_time1' [s]
161	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._Plate_FromIncubator' to 'ML_STAR.platewaste' 1 return value(s) .
162	Timer: Wait for Wait for timer 'main_timer_incutation', show timer display, is stoppable timer.
163	Grouping
164	Grouping Transformation
165	HSL code. Trace("Starting Loop - Transformation"); ML_STAR._Plate_PCR.SetCurrentPosition(1);
166	_Sub05_TransportLid of KompetenteZellen_v0.0.1 _Sub05_TransportLid(ML_STAR._Res_Plasmid_Lid, ML_STAR._LidStorage)
167	Loop 'main_resuspend_LoopCounter' times 'main_Loop_DNA' used as loop counter variable
168	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Res_Plasmid, Volume [µl]: main_dna_Vol 0 return value(s) .
169	1000µl Channel Dispense on ML_STAR Sequence: ML_STAR._Plate_PCR, Volume [µl]: Remaining volume inclusive blowout air 0 return value(s) .
170	End Loop
171	_Sub05_TransportLid of KompetenteZellen_v0.0.1 _Sub05_TransportLid(ML_STAR._LidStorage, ML_STAR._Res_Plasmid_Lid)
172	HSL code. Trace("Transformation done");
173	Grouping

	Method
174	Grouping Incubation on Ice & Prepair fresh media plate
175	HSL code. Trace("Incubation on ice");
176	Timer: Start Start timer 'main_timer_incutationOnIce', set to relative time: 'main_incubation_onIce' [s]
177	Sub_01_AddFromContainer_sterilTips of KompetenteZellen_v0.0.1 Sub_01_AddFromContainer_sterilTips(ML_STAR._Res_Medium, ML_STAR._Plate_Cultivaiton_2, main_cultivation2_Vol)
178	Timer: Wait for Wait for timer 'main_timer_incutationOnIce', show timer display, is stoppable timer.
179	HSL code. Trace("Incubation on ice done");
180	Grouping
181	Grouping Heatshock
182	HSL code. Trace("Starting Heatshock");
183	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._Plate_PCR' to 'ML_STAR._Plate_PCR_heatshock' 1 return value(s) .
184	Timer: Start Start timer 'main_timer_incutationHeatshock', set to relative time: 'main_incubation_heatShock' [s]
185	Timer: Wait for Wait for timer 'main_timer_incutationHeatshock', show timer display, is stoppable timer.
186	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._Plate_PCR_heatshock' to 'ML_STAR._Plate_PCR' 1 return value(s) .
187	HSL code. Trace("Heatshock Done");
188	Grouping
189	Grouping Cultivation2
190	HSL code. Trace("Starting Cultivation 2");
191	Sub_02_AddFromSequence of KompetenteZellen_v0.0.1 Sub_02_AddFromSequence(ML_STAR._Plate_PCR, ML_STAR._Plate_Cultivaiton_2, main_cultivaiton2_VolKultur)
192	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(2, ML_STAR._Plate_Cultivaiton_2, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
193	Timer: Start Start timer 'main_timer_cultivation2', set to relative time: 'main_cultivation2_time' [s]
194	Timer: Wait for Wait for timer 'main_timer_cultivation2', show timer display, is stoppable timer.
195	fame_transport of unifiedlibrary UNIFIEDLIBRARY::fame_transport(1, ML_STAR._Plate_FromIncubator, main_cultivation_slot, main_cultivation_temp, main_cultivation_speed, ML_STAR)
196	HSL code. Trace("Cultivation 2 Done");
197	Grouping
198	Grouping Ausplatieren

	Method
199	Sequence: Set Current Position current position of sequence 'ML_STAR._Plate_Cultivaiton_2' = '1'

200	HSL code.
	Trace("Start Plating");
	main_temp = 0;
	
	// Errechne wie viele Platen ausplattiert werden muessen
	main_platting_numPlates = main_numStrains%6;

201	Assignment 'main_CytomatPos' = '148'
202	Loop 'main_platting_numPlates' times 'main_loop_platting' used as loop counter variable
203	Grouping Get Plate from cytomat
204	HSL code. sShellCmd = "C:\Dokumente und Einstellungen\Peter Neubauer\Eigene Dateien\Sebastian\Programms\Remote_Trick\ClientConsole.exe C:\Users\BVT-Administrator\Anaconda3\python.exe C:\Tecan_Scripts\Remoting\CytomatAgarplate.py -m 1 -s " + IStr(main_CytomatPos); Trace(sShellCmd); Shell(sShellCmd, 2,1);
205	Position_Hamilton of BVTlib_LTU Position_Hamilton()
206	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._move_Tecan' to 'ML_STAR._Plate_AgarTransport' 1 return value(s) .
207	Grouping
208	Sequence: Set Current Position current position of sequence 'ML_STAR._LidAgar' = '1'
209	_Sub05_TransportLid_Agar of KompetenteZellen_v0.0.1 _Sub05_TransportLid_Agar(ML_STAR._Plate_Agar_StorePlated_Lids, ML_STAR._LidAgar)
210	1000µl Channel CO-RE Grip Get Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_Agar_StorePlated, Sequence counting: (0) Manually, Channel to be used: 8 3 return value(s) .
211	1000µl Channel CO-RE Grip Place Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_Agar, Sequence counting: (0) Manually, Eject tool: (0) No 3 return value(s) .
212	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Res_Medium, Volume [µl]: 190 0 return value(s) .
213	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Res_Medium, Volume [µl]: 10 0 return value(s) .
214	1000µl Channel Aspirate on ML_STAR Sequence: ML_STAR._Plate_FromIncubator, Volume [µl]: 50 0 return value(s) .
215	1000µl Channel Dispense on ML_STAR Sequence: ML_STAR._Plate_Agar, Volume [µl]: Remaining volume inclusive blowout air 0 return value(s) .
216	Sub_05_Shaking of KompetenteZellen_v0.0.1 Sub_05_Shaking()
217	Sequence: Set Current Position current position of sequence 'ML_STAR._Plate_Agar' = '1'

	Method
218	1000µl Channel CO-RE Grip Get Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_Agar, Sequence counting: (0) Manually, Channel to be used: 8 3 return value(s) .
219	1000µl Channel CO-RE Grip Place Plate (Single Step) on ML_STAR Transport mode: (0) Plate only, Sequence: ML_STAR._Plate_Agar_StorePlated, Sequence counting: (0) Manually, Eject tool: (0) No 3 return value(s) .
220	Sequence: Set Current Position current position of sequence 'ML_STAR._LidAgar' = '1'
221	_Sub05_TransportLid_Agar of KompetenteZellen_v1.1.0 _Sub05_TransportLid_Agar(ML_STAR._LidAgar, ML_STAR._Plate_Agar_StorePlated_Lids)
222	Grouping Transfer Agarplate to Cytomat
223	iSWAP Transport on ML_STAR Transport labware from 'ML_STAR._Plate_AgarTransport' to 'ML_STAR._move_Tecan' 1 return value(s) .
224	Position_Tecan of BVTlib_LTU Position_Tecan()
225	HSL code. sShellCmd = "C:\Dokumente und Einstellungen\Peter Neubauer\Eigene Dateien\Sebastian\Programms\Remote_Trick\ClientConsole.exe C:\Users\BVT-Administrator\Anaconda3\python.exe C:\Tecan_Scripts\Remoting\CytomatAgarplate.py -m 0 -s " + IStr(main_CytomatPos); Trace(sShellCmd); Shell(sShellCmd, 2,1);
226	Assignment with Calculation 'main_CytomatPos' = 'main_CytomatPos' + '1'
227	Grouping
228	End Loop
229	HSL code. Trace("Plating done");
230	Grouping
231	

Figure S3: Tecan LHS

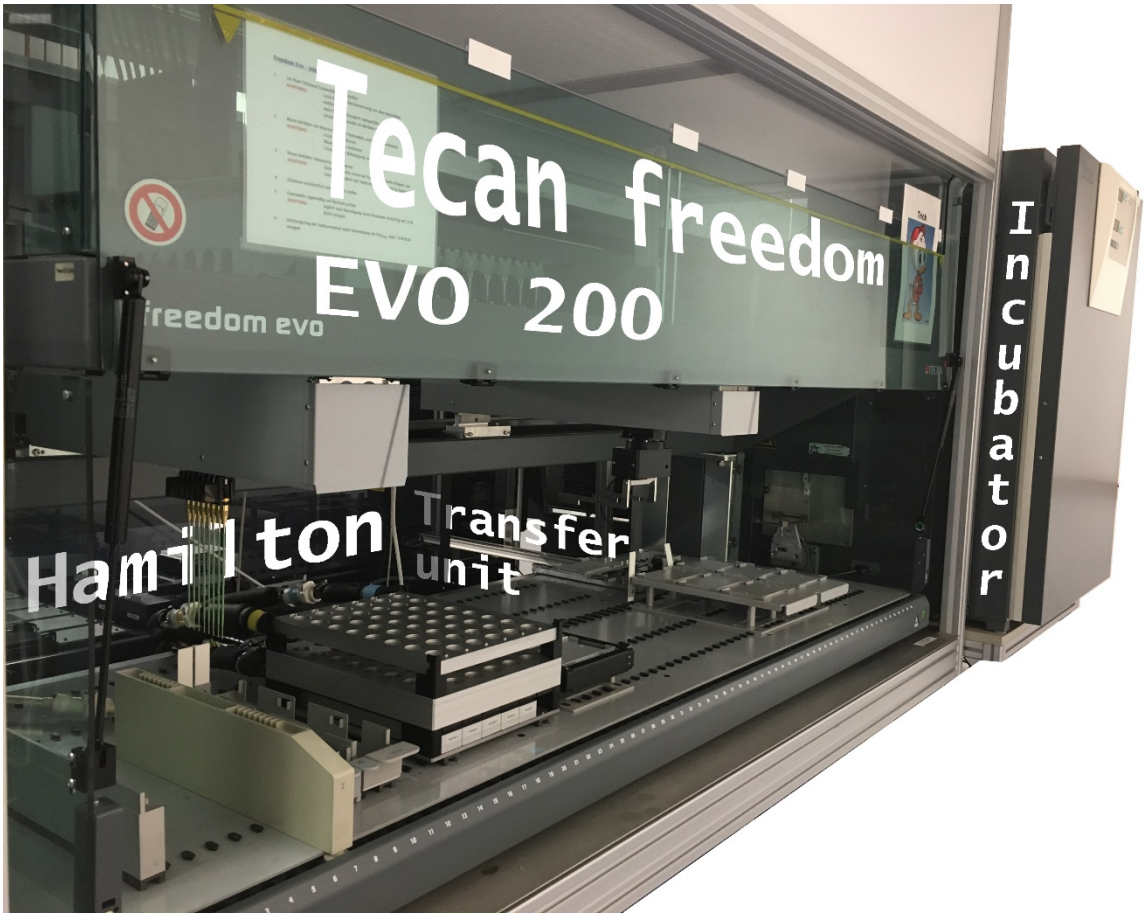

Figure 1: Second used Liquid handling station (LHS). Plates are stored at the incubator on the right site of the LHS. On a command of the Hamilton LHS a plate is moved from the incubator to the Transfer unit. A second command was used to return the plate from the transfer unit into the incubator.

Source Code S4 MATLAB Source Code

```
function DiluteForChemostat(iRunID)
sim = 0;
%% Ziel OD:
    fTragetOD = 0.8;
    iCultivationVol = 170; % [µL]

%% Connect to ilab
    if ispc % check if running on Windows
        ilab = actxserver('BVT_iLabDriver.ilab_net_class');
% %         ilab = actxserver('ILAB_COM_BVT.ILAB_COM_BVT');
    else
        ilab = iLab_driver_universal;
    end
    ilab.SQL_Close;
    ilab.SQL_Connect;

%     ilab.run_id = iRunID;
%     ilab = ilab.get_bioID(iRunID);
%     iExpID = ilab.getExpIDByBioID();
%     iProfID = ilab.getProfIDByBioID();
    iBioID = ilab.getBioreactorIDs(iRunID);
    iBioID = iBioID(1);
    iExpID = ilab.getExperimentIDs(iBioID);
    iProfID = ilab.getProfilIDs(iBioID);
    iNumOfExperiments = length(iExpID);



    %% Calling Measurments
    fOD600 = ilab.getExperimentMeasurements(iExpID,'OD600');
    fTime = fOD600(:,1)/3600;
    fOD600 = fOD600(:,2:end);

    iSizeOfData = size(fOD600);
    iNumOfMeasurments = iSizeOfData(1);

    %% get last Setpoints to compute the last dilution factor
    fDilutionOld =   ones(1,length(iExpID));
    if iNumOfMeasurments > 1
        for ci = 1:iNumOfExperiments
            fDilutionOld(ci) = (iCultivationVol-ilab.SetpointgetCurrent(iProfID(ci),
'Puls_Medium'))/iCultivationVol;
        end
        % Berechne vorletzte OD neu:
        fOD600(end-1,:) = fOD600(end-1,:).*fDilutionOld;
    end

    %% create timestamp:
    iTime = ilab.getCultivationTime(iRunID);

    %% Compute µ / OD set
    % Erstelle Array's
    fODNext =       zeros(1,length(iExpID));
    fDilution =  ones(1,length(iExpID));
    fVolRemain =  zeros(1,length(iExpID));
    fVolAdd =       zeros(1,length(iExpID));
    fVolRemove =    zeros(1,length(iExpID));

    fMu = zeros(iNumOfMeasurments-1, length(iExpID));
    if ~(iNumOfMeasurments > 1) % if no µ values can be calculated
        for ci = 1:length(iExpID)
            fVolAdd(ci) = 20;
            fVolRemove(ci) = 0;
            if ~sim
                ilab.SetpointSet(int32(iProfID(ci)), 'Puls_Medium', iTime, fVolAdd(ci));
                ilab.SetpointSet(int32(iProfID(ci)), 'Probe_Volume', iTime,
fVolRemove(ci));
            end
        end
    else % normal process ...
        for ci = 2:iNumOfMeasurments
            for cj = 1:length(iExpID)
```

```
                fMu(ci, cj) = (log(fOD600(ci,cj))-log(fOD600(ci-1,cj))) / (fTime(ci)-
fTime(ci-1));
            end
        end
        for ci = 1:length(iExpID)
            fODNext(ci) = fTragetOD / exp(fMu(end,ci) * (fTime(end)-fTime(end-1)));
            if fODNext(ci)~=0
                fDilution(ci) = fODNext(ci) / fOD600(end, ci);
            end
            fVolRemain(ci) = fDilution(ci) * iCultivationVol;
            if fVolRemain(ci) > (iCultivationVol - 20)
                fVolRemain(ci) = (iCultivationVol - 20);
            elseif fVolRemain(ci) < 45
                 fVolRemain(ci) = 45;
            end
            % Compute Volumes
            fVolRemove(ci) = (iCultivationVol - 20) - fVolRemain(ci);
            fVolAdd(ci) = iCultivationVol - fVolRemain(ci);
            %% Write data to database
            if ~sim
                ilab.SetpointSet(int32(iProfID(ci)), 'Puls_Medium', iTime, fVolAdd(ci));
                ilab.SetpointSet(int32(iProfID(ci)), 'Probe_Volume', iTime,
fVolRemove(ci));
            end
        end
    end
end
```