

SUPPORTING INFORMATION

Caco-2 based co-culture model of the inflamed intestinal mucosa suitable for studies on particles

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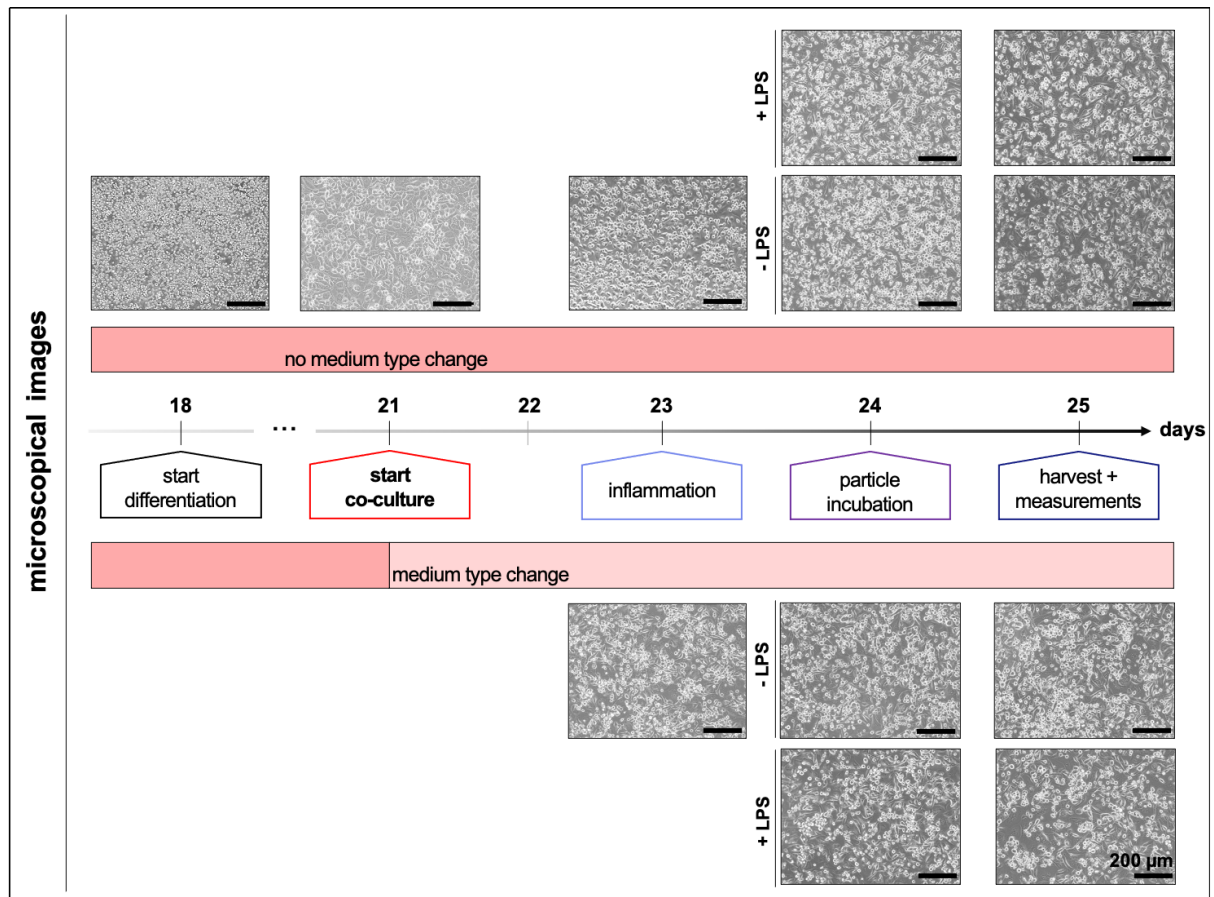


Figure S 1: Microscopical examination of the morphologic changes of PMA-derived THP-1 M0-macrophages during the co-culture simulation procedure. The upper images represent cells without medium type change (only RPMI+). The lower images represent cells cultivated with medium type change from RPMI+ to DMEM+ on day 21. Images were taken with a 100 x magnification. The scale bar displays a length of 200 μm.

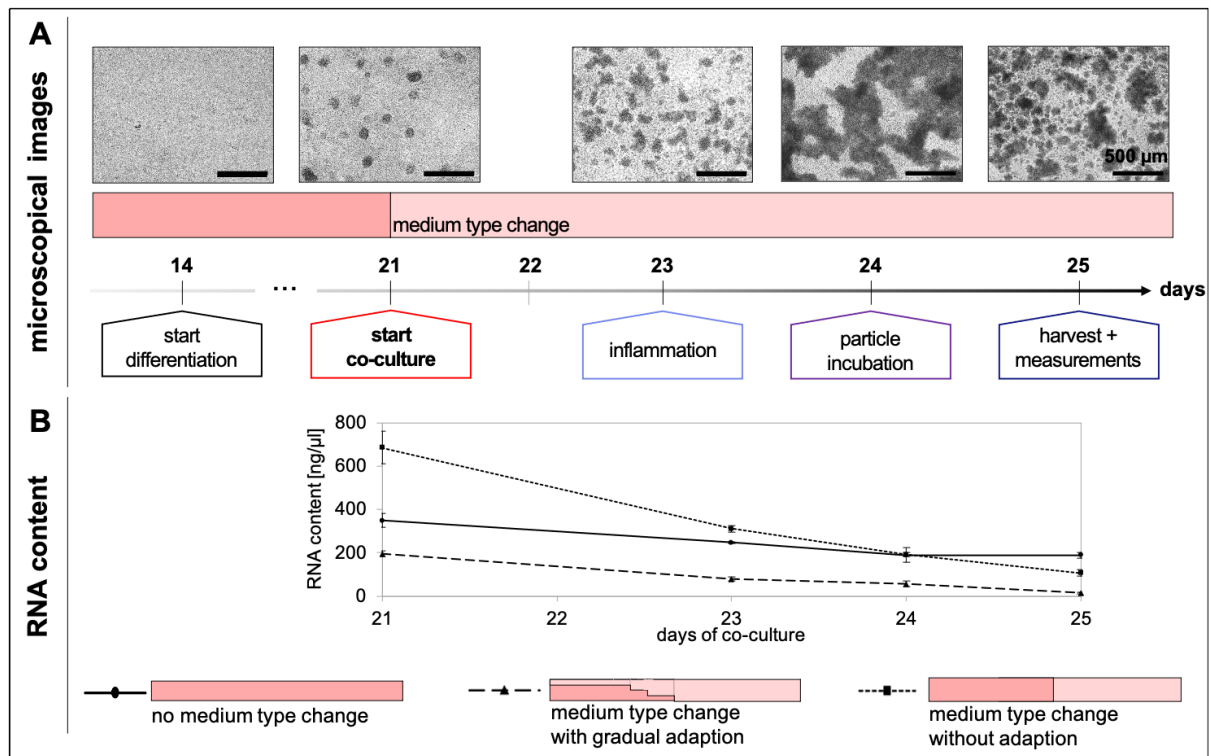


Figure S 2: Monitoring the survivability of differentiated MUTZ-3 dendritic cells after immediate medium type change from MUTZ-3 to DMEM+ during co-culture simulation procedure. **A:** Microscopical examination of MUTZ-3 cell morphology during differentiation in MEM+ and simulation of the co-culture in DMEM+. Images were taken with a 50 x magnification. The scale bar displays a length of 500 μm . **B:** Comparison of RNA content of MUTZ-3 cells harvested on days 21, 23, 24 and 25 after being differentiated in MEM+ and receiving 1) no medium type change 2) medium type change with gradual adaption or 3) medium type change without adaption during the co-culture simulation.

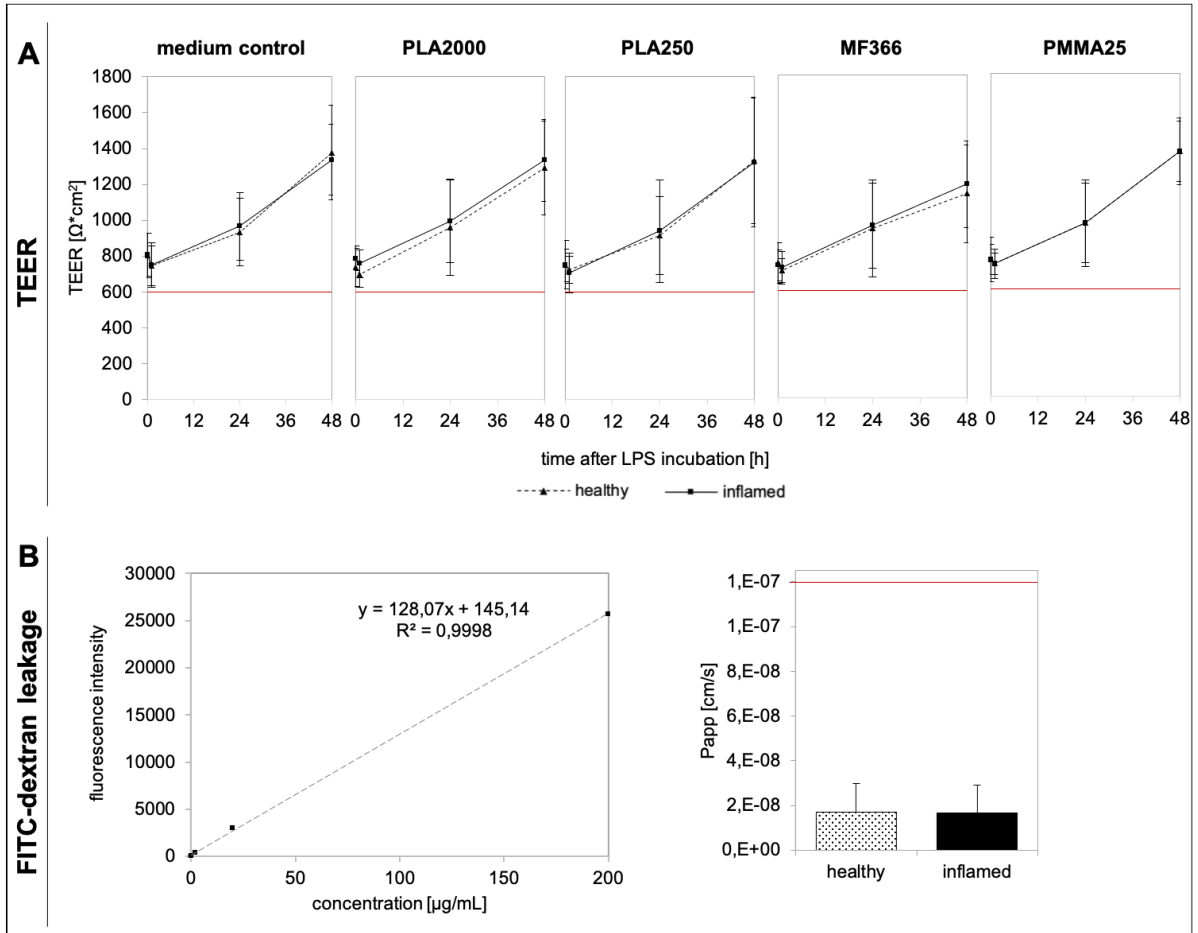


Figure S 3: Permeability measurements of the co-culture in healthy and inflamed state as well as after incubation with nano-, submicro- and microplastic particles. **A:** Changes in TEER values after 0 h, 1 h, 24 h and 48 h after LPS incubation. Red lines indicate lower limit for TEER values **B:** FITC-dextran leakage in the co-culture model. Left: representative standard curve of FITC-dextran, right: calculated P_{app} values from FITC-dextran leakage after 24 h incubation in the healthy and inflamed model. Red line indicates lower limit for leakage. All data are presented as means $\pm$ SD, $n = 3$.

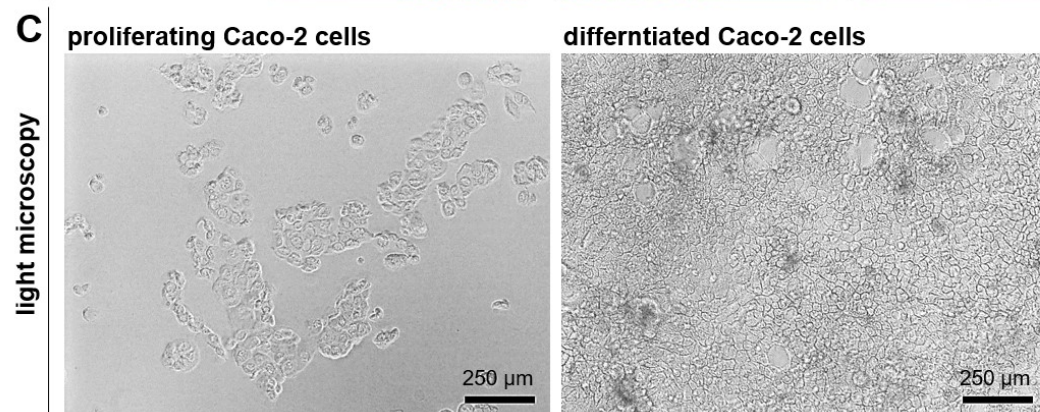
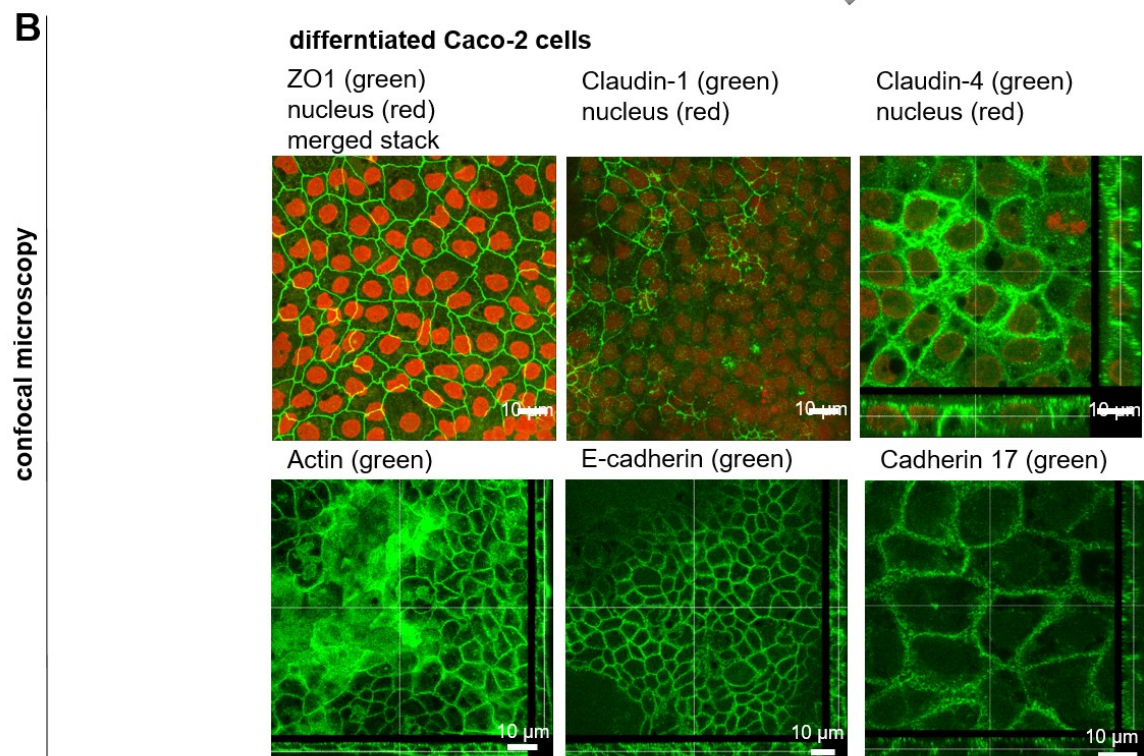
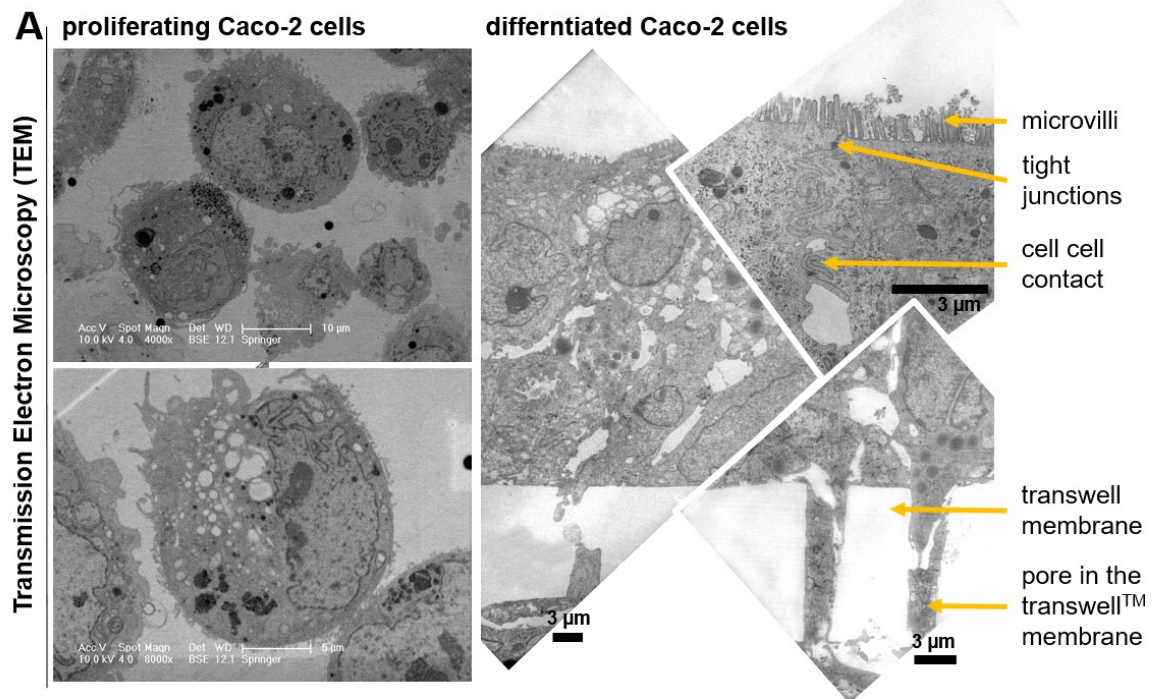


Figure S 4: Morphological characteristics of differentiated Caco-2 cells. A Transmissions Electron Microscopical images of proliferating and on TranswellTM membrane differentiated Caco-2 cells. Sample preparation: cells were fixed overnight with Karnovsky solution, pre-contrasted in 2% osmium tetroxide, dehydrated, embedded, ultra-thin sections were prepared on the Reichert Ultracuts microtome and then further contrasted with lead and uranyl acetate. Samples were viewed either on a Zeiss 10 CR or an FEI Tecnai G² 20 S-TWIN. *B* Confocal mikrosopical images of differentiated Caco-2 cells stained with antibodies against tight junction proteins and actin. Sample preparation: Caco-2 cells were differentiated on glass slides, fix, permeabilized and stained for analysis with SP5 from Leica Microsystems.

Table S1: overview expression markers.

cell marker	day	fold change in gene expression			
		MUTZ-3 without adaption	MUTZ-3 with adaption	THP-1 without adaption	THP-1 with adaption
CD14	14 or 18	0,9096	1,2510	1,0300	1,0300
	21	0,1078	0,0202	18,8280	18,8280
	23	0,9212	0,4855	223,9000	214,2840
	24 +	1,8272	1,6281	145,7660	143,2760
	25 +	2,5730	0,8373	161,7420	152,7060
	24 -	1,8872	0,8142	181,1067	143,5100
	25 -	2,4491	1,1078	28,7480	44,8980
CD1a	14 or 18	1,0183	1,0433	1,1133	1,1133
	21	144,9126	14,3687	206,6067	206,6067
	23	77,0747	25,6065	153,2433	179,0375
	24 +	126,6153	32,9841	46,1975	38,0650
	25 +	125,8015	15,1161	32,9775	44,9375
	24 -	119,3641	32,3858	9,2875	8,4800
	25 -	50,3344	37,3335	0,8725	49,5800
CD80	14 or 18	1,3582	0,4460	1,0067	1,0067
	21	47,6833	10,2889	0,5800	0,5800
	23	46,1654	9,6169	2,4850	5,3475
	24 +	27,8826	2,7872	44,7275	95,3875
	25 +	27,8574	0,8246	47,8075	29,7025
	24 -	45,8369	3,0708	21,5120	10,9750
	25 -	31,0697	1,3953	57,3950	160,7900
CD86	14 or 18	0,8742	0,8742	1,0250	1,0250
	21	4,4245	4,4245	22,3125	22,3125
	23	9,6747	9,6747	153,6700	94,6740
	24 +	12,0120	8,0763	94,2000	58,1675
	25 +	14,3753	9,5050	87,2233	79,5100
	24 -	6,9212	5,7715	141,1060	81,6175
	25 -	5,1677	3,5524	37,6460	42,2240
CD209	14 or 18	1,0231	0,6408	not measured	not measured
	21	130,7602	26,4726		
	23	17,3921	22,5741		
	24 +	13,1931	21,3094		
	25 +	14,3600	17,2245		
	24 -	16,3184	21,0896		
	25 -	18,5211	20,1939		
IL-10	14 or 18	0,9702	0,7014	1,0533	1,0533
	21	1,2351	0,1935	185,9500	185,9500
	23	1,2382	1,7346	423,7280	349,8975
	24 +	1,6398	2,7810	1007,4000	1450,5067
	25 +	2,7299	7,6979	4087,2525	3843,8417
	24 -	1,2824	1,5027	1384,8120	1374,9060
	25 -	1,8220	1,3902	1324,8175	2285,9475

IL-8	14 or 18	1,1350	1,3060	1,0002	1,0002
	21	4,0551	1,1251	5,1600	4,2067
	23	3,6889	1,7545	3,1550	3,7620
	24 +	2,3910	2,3703	13,5550	43,3625
	25 +	2,8888	2,9049	7,4017	20,0080
	24 -	1,6176	1,3302	4,6740	3,5550
	25 -	1,6747	1,6252	0,0740	0,1917
IL-6	14 or 18	0,7827	0,7827	1,0000	1,0000
	21	1,8403	1,8403	7,1700	7,1700
	23	1,4096	1,4096	2,0960	3,2180
	24 +	2,2503	1,8523	8,4040	7,3500
	25 +	2,4457	0,7056	6,4560	10,3900
	24 -	1,8445	1,9969	5,7300	2,2680
	25 -	1,6933	1,3645	5,1600	4,8317
TNF- α	14 or 18	0,9579	0,6231	1,0533	1,0533
	21	70,8333	11,3959	0,7380	0,7380
	23	37,4491	25,5557	9,1580	2,2160
	24 +	22,0109	17,8806	0,8680	0,9983
	25 +	33,3960	11,1376	1,9620	1,1540
	24 -	24,3259	12,9052	1,1900	1,1825
	25 -	55,7913	23,0738	0,9583	0,8760

Legend:

- 14 or 18 start of differentiation
- 21 start of co-cultivation
- 23 medium change or inflammation with LPS
- 24 + medium change, 24h after LPS inflammation
- 25 + medium change, 48h after LPS inflammation, potential incubation with substances
- 24 - medium change only
- 25 - medium change only, potential incubation with substances