

## The heterogeneity and complexity of *Cannabis* extracts as antitumor agents

### SUPPLEMENTARY MATERIALS

#### PHYTOCANNABINOID EXTRACTION AND SAMPLE PREPARATION

*Cannabis* plant samples were ground to a fine powder using an electrical grinder. Prior to phytocannabinoid extraction, several of the flowers underwent heat-decarboxylation in an oven at 120 °C for 1 h. Approximately 5 g of the natural or decarboxylated flowers were accurately weighed and extracted with 50 ml ethanol. Samples were sonicated in an ultrasonic bath for 30 min and then agitated in an orbital shaker at 25 °C for 15 min. Samples were then filtered under pressure through Whatman filter paper number 4 and the ethanol was evaporated under reduced pressure at 38 °C using a rotary evaporator (Laborata 4000; Heidolph Instruments GmbH & Co. KG; Germany).

#### PHYTOCANNABINOID IDENTIFICATION AND QUANTIFICATION

Phytocannabinoid analyses were performed using a Thermo Scientific ultra-high-performance liquid chromatography (UHPLC) system coupled with a Q Exactive™ Focus Hybrid Quadrupole-Orbitrap MS (Thermo Scientific, Bremen, Germany). The chromatographic separation was achieved using a Halo C18 Fused Core column (2.7 µm, 150 mm × 2.1 mm i.d.) with a guard column (2.7 µm, 5 mm × 2.1 mm i.d.) (Advanced Materials Technology, Delaware, USA) and a ternary A/B/C multistep gradient (solvent A: 0.1 % acetic acid in Milli Q water, solvent B: 0.1% acetic acid in acetonitrile, and solvent C: methanol, all solvents were

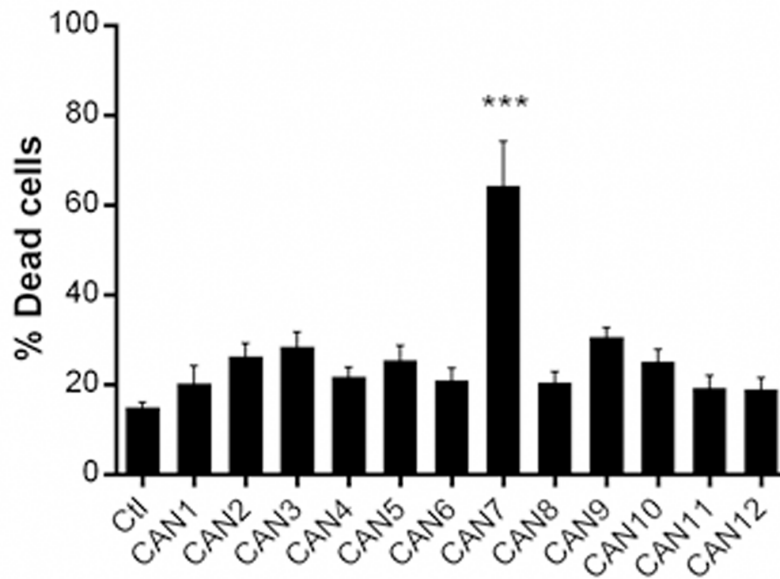
of LC/MS grade). The multistep gradient program was established as follows: initial conditions were 50% B raised to 67% B until 3 min, held at 67% B for 5 min, and then raised to 90% B until 12 min, held at 90 % B until 15 min, decreased to 50% B over the next min, and held at 50 % B until 20 min for re-equilibration of the system prior to the next injection. Solvent C was initially 5% and then lowered to 3% until 3 min, held at 3 % until 8 min, raised to 5 % until 12 min and then kept constant at 5 % throughout the run. A flow rate of 0.25 ml/min was used, the column temperature was 30 °C and the injection volume was 1 µL. MS acquisition was carried out with a heated electro spray ionization (HESI-II) ion source operated in negative mode. Source parameters were as follows: sheath gas flow rate, auxiliary gas flow rate and sweep gas flow rate: 50, 20 and 0 arbitrary units respectively; capillary temperature: 350 °C; heater temperature: 50 °C; spray voltage: 3.00 kV. The scan range was 150–550 m/z for all acquisition events. MS was operated in full MS<sup>1</sup> mode at 70,000 resolution, and the AGC target was set to 10<sup>6</sup> with a maximum IT of 100 ms.

#### POLYMERASE CHAIN REACTION (PCR) ANALYSIS

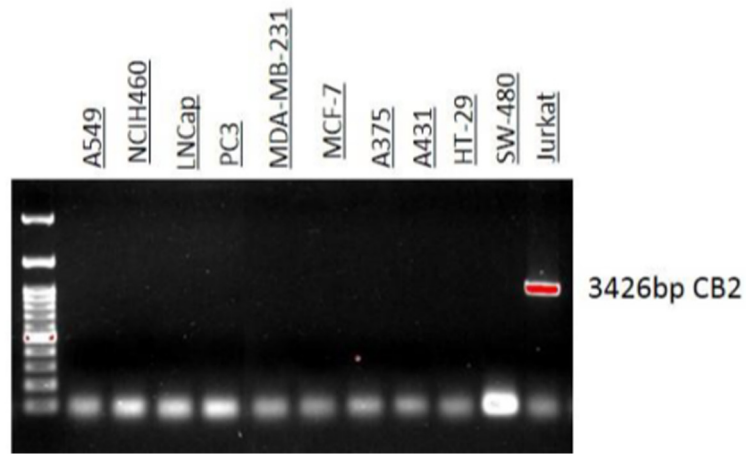
mRNA expression levels of the human CNR2 gene (CB2) were quantified in 50 ng cDNA using Red Load Taq Master PCR assay. PCR amplification was performed using the following primers: Forward: atggaggaatgctgggtgacagaga Reverse: gcaatcagagaggtct agatctctggaatct. Results were visualized using 1.2% gel electrophoresis.

	CAN6	CAN4	CAN11	CAN7	CAN5	CAN12	CAN2	CAN9	CAN3	CAN8	CAN1	CAN10			
A <sup>+</sup> -THCtype	THCA	55.0065	48.0668	29.9010	0.0000	0.3049	0.2170	0.0000	0.0000	0.0000	1.1003	16.6407	0.0000	C - %w/w	
	THC	6.1633	15.9636	1.7548	67.8032	56.5847	35.1295	25.9923	2.3489	1.4977	0.0000	3.1207	0.0000	C<0.02	
	THCA-C4	0.3514	0.2030	0.1169	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0665	0.0000	0.02<C<0.1%	
	THC-C4	0.0000	0.0000	0.0000	0.1969	0.1085	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.1%<C<1%
	THCVA	1.1172	0.3818	0.3895	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0360	0.1295	0.0000	0.0000	1%<C<10%
	THCV	0.0000	0.0000	0.0000	0.6137	0.2072	0.2246	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	10%<C<30%
	THCOA	0.2371	0.3631	0.0676	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.1178	0.0000	0.0000	C>30%
THCMA	0.0617	0.0896	0.0297	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0351	0.0000	0.0000		
CBDtype	CBDA	0.1922	0.2139	0.0787	0.0000	0.0000	0.0093	0.1294	6.2145	0.0249	42.4938	31.1696	0.0848		
	CBD	0.0000	0.0000	0.0000	0.2107	0.2125	0.0776	35.8411	55.5851	27.1263	4.4839	1.8886	0.8097		
	CBDA-C4	0.0000	0.0000	0.0000	0.0000	0.0015	0.0000	0.0023	0.0206	0.0000	0.1345	0.0466	0.0000		
	CBD-C4	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0564	0.1803	0.0521	0.0172	0.0000	0.0000		
	CBDVA	0.0014	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0701	0.0000	0.4627	0.0433	0.0000		
	CBDV	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0589	0.7227	0.1896	0.0573	0.0000	0.0210		
	CBDOA	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0085	0.0000	0.0307	0.0085	0.0000		
CBGtype	CBGA	12.7114	4.4696	0.3930	0.2240	0.0725	0.0739	0.0000	0.1960	0.0000	0.8667	1.1989	0.0196		
	CBG	0.8920	0.6684	0.1866	12.6179	4.4623	0.5928	1.8095	1.7364	1.9181	0.3457	0.1926	54.6694		
	CBGA-C4	0.0192	0.0222	0.0026	0.0362	0.0408	0.0114	0.0131	0.0000	0.0000	0.0034	0.0032	0.0000		
	CBG-C4	0.0000	0.0000	0.0000	0.0094	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.1466		
	CBGV	0.0000	0.0000	0.0000	0.0071	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.8819		
	CBGO	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0651		
	Sesqui-CBG	0.0130	0.0277	0.0110	0.0160	0.0310	0.0187	0.0273	0.0374	0.0350	0.0173	0.0161	0.0358		
CBE/CEN type	CBCA	0.3390	0.4440	0.1316	0.0061	0.0060	0.0252	0.0031	0.0763	0.0000	0.6815	0.6925	0.0000		
	CBC	0.0749	0.2113	0.0163	0.8269	1.0927	0.3350	1.9335	2.4134	1.5208	0.1785	0.1491	0.7137		
	CBCVA	0.0042	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0031	0.0000	0.0277	0.0037	0.0000		
	CBCV	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0880	0.0255	0.0000	0.0000	0.0417		
	CBNA	0.7241	1.3076	0.4984	0.0000	0.0121	0.0000	0.0000	0.0000	0.0000	0.0488	0.2313	0.0000		
	CBN	0.0417	0.3022	0.0171	1.0339	2.7877	0.3699	1.0495	0.0830	1.5111	0.0143	0.0252	0.0000		
	CBEA	0.0109	0.0105	0.0000	0.0000	0.0000	0.0000	0.0000	0.0362	0.0000	0.1430	0.0837	0.0000		
CBE	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.2917	0.2958	0.9250	0.0156	0.0000	0.0543			
CBTtype	CBNDA	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0043	0.0000	0.0651	0.0296	0.0000		
	CBL	0.0000	0.0000	0.0000	0.0000	0.0458	0.0000	0.0582	0.0386	0.0848	0.0000	0.0000	0.0444		
	CBTA-1	1.5635	1.2756	1.8236	0.0000	0.0166	0.0037	0.0000	0.0000	0.0000	0.0466	0.3494	0.0000		
	CBT-1	1.0314	2.6370	0.6330	3.3258	4.1060	1.8548	1.2396	0.2518	0.7204	0.1128	0.3086	0.0000		
	CBTV-1	0.0000	0.0000	0.0000	0.0295	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		
	CBTA-3	0.4617	0.2567	0.3411	0.0000	0.0087	0.0000	0.0000	0.0000	0.0000	0.0612	0.0935	0.0000		
	CBT-3	0.1403	0.4385	0.0731	1.6640	2.6390	0.5477	0.7158	0.0948	0.3649	0.0344	0.0572	0.0000		
	CBT-2	0.0000	0.0215	0.0000	0.0000	0.0000	0.0000	0.0607	0.0484	0.0000	0.0000	0.0000	0.0000		
	329-11a	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0554	0.2582	0.0523	0.0276	0.0000	0.0000		
	329-11b	0.0000	0.0139	0.0169	0.1524	0.1491	0.2630	0.1943	0.0550	0.1050	0.0000	0.0247	0.0277		
	329-11c	0.0000	0.0000	0.0000	0.0170	0.0000	0.0000	0.0737	0.1308	0.0000	0.0330	0.0000	0.0187		
	329-11d	0.1285	0.5413	0.1145	0.7637	1.1899	0.3622	0.2969	0.0424	0.0789	0.0190	0.0703	0.0000		
	329-11e	0.0000	0.0000	0.0000	0.0554	0.0843	0.0176	0.0368	0.0000	0.0000	0.0000	0.0000	0.0000		
	373-12a	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0100	0.0000	0.0455	0.0088	0.0000		
373-12b	0.0234	0.0276	0.0807	0.0000	0.0000	0.0000	0.0000	0.0035	0.0000	0.0413	0.0630	0.0000			
373-12c	0.0297	0.0452	0.0124	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000			
327-13a	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.1053	0.7707	0.0314	0.0000	0.0000	0.0000			
327-13b	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0157	0.1360	0.0317	0.0146	0.0000	0.0000			
327-13c	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.2999	0.2335	0.1199	0.2178	0.0772	0.0000			
371-14a	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0331	0.0000	0.0341	0.0030	0.0000			
371-14b	0.1078	0.1063	0.0815	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0046	0.0193	0.0000			
417-15a	0.0467	0.0585	0.0626	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0226	0.0000			
373-15b	0.2426	0.2337	0.0787	0.1663	0.1712	0.1308	0.1767	0.1204	0.1346	0.1241	0.1380	0.0000			
373-15c	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0413	0.1523	0.0000	0.1298	0.1851	0.0000			
357-16a	0.0143	0.0290	0.0068	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0352	0.0441	0.0000			
313-16b	0.0000	0.0000	0.0000	0.0103	0.0173	0.0000	0.1367	0.2006	0.1009	0.0917	0.0711	0.0000			
361-17a	0.0000	0.0962	0.0000	0.0236	0.0454	0.0265	0.0000	0.0000	0.0163	0.0000	0.0000	0.0000			
361-17b	0.0000	0.0000	0.0000	0.0256	0.0333	0.0000	0.0154	0.0000	0.0203	0.0000	0.0000	0.0000			
331-18a	0.0000	0.0000	0.0000	0.0396	0.0435	0.0000	1.2613	2.0983	1.0447	0.1981	0.1158	0.0686			
331-18b	1.1828	0.8041	0.3370	0.7599	0.4643	0.3771	0.5084	0.7992	0.2322	0.6126	0.6776	0.7082			
331-18d	0.1087	0.2432	0.1558	0.0392	0.0670	0.0908	0.0543	0.0713	0.0000	0.0810	0.2207	0.0000			
375-19a	0.0087	0.0094	0.0000	0.0000	0.0000	0.0000	0.0000	0.0191	0.0000	0.5218	0.3494	0.0000			
375-19b	0.0209	0.0123	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0030	0.0000			
375-19c	0.0234	0.0210	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0042	0.0057	0.0000			

**Supplementary Figure 1: Phytocannabinoid analysis of the *Cannabis* extracts.** ECI-LC/MS phytocannabinoid profiling of the *Cannabis* extracts which were prepared as described in Materials and Methods in the main text. *Cannabis* extracts were ordered into clusters according to cannabinoid content (% w/w) in each extract with values color-coded according to order of magnitude of concentration. Only cannabinoids identified at a concentration of at least 0.2 (%w/w) in at least one extract were included in this figure.



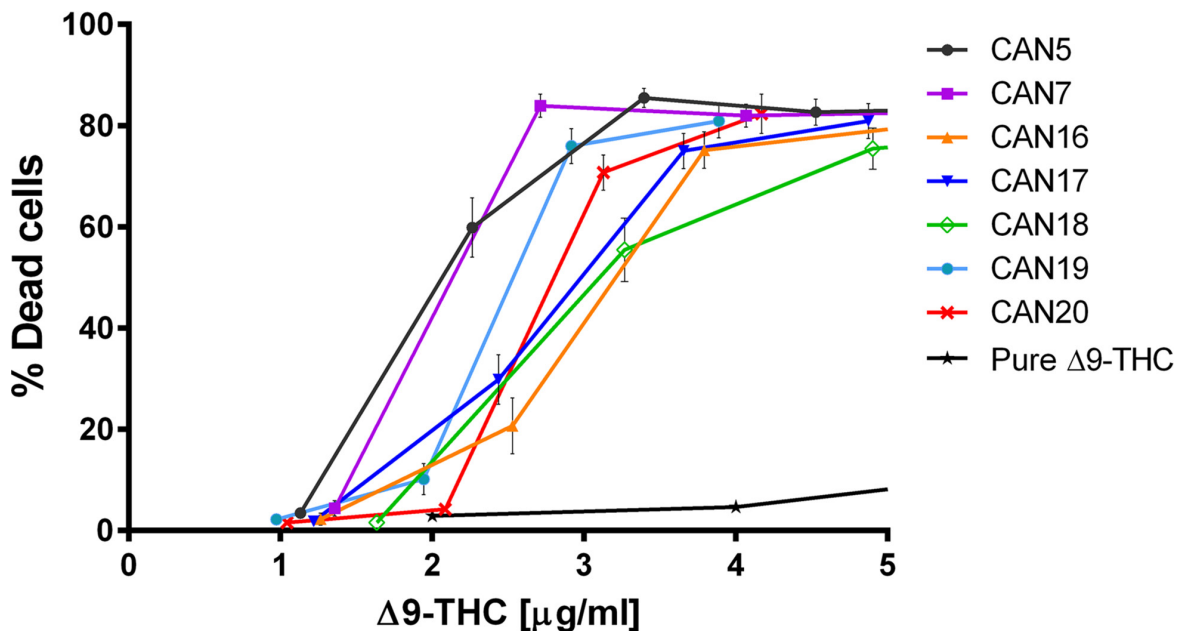
**Supplementary Figure 2: The effect of various *Cannabis* extracts on primary airway epithelial cells.** Normal primary epithelial cells were incubated for 24 hours incubation with or without (control) 4  $\mu\text{g/ml}$  of CAN1-CAN12. This was followed by the addition of Hoechst and PI and imaged by IXM Micro system, 20 $\times$  magnification. Live and dead cells were analyzed by the MetaXpress™ software, and data are reported as mean  $\pm$  SE of % dead cells out of total cells (N=3). Asterisks indicate statistical difference compared to control (\* $P$ <0.05, one-way ANOVA).



**Supplementary Figure 3: Evaluation of CNR2 mRNA expression in ten different cancer cell lines.** CNR2 mRNA expression levels were evaluated by PCR, using cDNA. As described in Supplementary Materials.

	CAN23	CAN19	CAN20	CAN21	CAN24	CAN25	CAN5	CAN13	CAN26	CAN17	CAN14	CAN16	CAN7	CAN18	
THCA	1.1502	1.5803	0.6912	3.5042	4.1435	0.1274	0.3049	5.0325	0.4736	0.0000	0.1255	0.3122	0.0000	0.6908	C - %w/w
THC	46.0497	48.6068	52.1128	52.2002	53.2992	56.1284	56.5847	56.5917	57.3547	60.9241	62.2031	63.1325	67.8032	81.7029	C<0.02
THCA-C4	0.0000	0.0000	0.0000	0.0128	0.0198	0.0000	0.0000	0.0213	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.02<C<0.1%
THC-C4	0.1160	0.1188	0.1696	0.1184	0.1498	0.0000	0.1085	0.1196	0.1278	0.1720	0.2003	0.2159	0.1969	0.1275	0.1%<C<1%
THCVA	0.0213	0.0000	0.0000	0.0452	0.0532	0.0000	0.0000	0.0552	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	1%<C<10%
THCV	0.3933	0.2408	0.4409	0.3724	0.3426	0.2343	0.2072	0.2938	0.3090	0.4601	0.4692	0.4479	0.6137	0.4426	10%<C<30%
THCOA	0.0000	0.0000	0.0000	0.0132	0.0134	0.0000	0.0000	0.0255	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	C>30%
CBDA	0.0917	0.0109	0.0270	0.0546	0.0379	0.0089	0.0000	0.0665	0.0246	0.0000	0.0175	0.0750	0.0000	0.0285	
CBD	0.7689	0.1115	0.1174	0.0846	0.0964	0.1240	0.2125	0.1095	0.1204	0.1325	0.1679	0.3280	0.2107	0.1598	
CBGA	0.1783	0.1988	0.2964	1.1113	0.4298	0.1016	0.0725	0.2913	0.2919	0.0579	0.0595	0.1588	0.2240	0.4754	
CBG	1.3528	1.7086	1.1200	1.7429	1.1701	0.9061	4.4623	0.8703	1.4233	1.6082	0.6369	1.0870	12.6179	2.2239	
CBGA-C4	0.0116	0.0077	0.0076	0.0151	0.0192	0.0094	0.0408	0.0272	0.0102	0.0000	0.0000	0.0132	0.0362	0.0148	
Sesqui-CBG	0.0246	0.0248	0.0114	0.0154	0.0404	0.0320	0.0340	0.0292	0.0594	0.0280	0.0336	0.0525	0.0160	0.0275	
CBCA	0.0376	0.0305	0.0466	0.1657	0.0991	0.0273	0.0060	0.1257	0.0378	0.0215	0.0274	0.0435	0.0061	0.0721	
CBC	1.1255	0.8699	0.6844	0.7319	0.7240	0.7091	1.0927	0.4670	0.5878	0.8653	0.6813	0.6439	0.8269	1.0350	
CBNA	0.0550	0.0334	0.1016	0.1903	0.0788	0.0000	0.0121	0.1660	0.0236	0.0000	0.0296	0.0229	0.0000	0.0617	
CBN	1.6068	0.5138	2.4148	0.4405	0.5195	0.4269	2.7877	0.6746	0.6110	0.5453	1.5265	0.6243	1.0339	1.5517	
CBNV	0.0142	0.0000	0.0207	0.0024	0.0023	0.0000	0.0083	0.0029	0.0000	0.0036	0.0117	0.0043	0.0096	0.0082	
CBL	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0458	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	
CBTA-1	0.0160	0.0194	0.0068	0.0237	0.0406	0.0000	0.0166	0.0340	0.0000	0.0000	0.0000	0.0000	0.0000	0.0050	
CBT-1	2.2479	1.6424	1.7820	2.3609	2.1782	1.1004	4.1060	2.5065	1.2334	0.9925	1.2721	1.3111	3.3258	1.8195	
CBTV-1	0.0175	0.0000	0.0139	0.0152	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0295	0.0000	
CBT-3	0.8889	0.7212	1.3833	0.5757	0.4832	0.6269	2.6390	0.6647	0.8046	0.3887	0.5495	0.5503	1.6640	1.9103	
329-11b	0.0875	0.5470	0.1020	0.0522	0.0709	0.0409	0.1491	0.0748	0.0987	0.0358	0.1108	0.3803	0.1524	0.1389	
329-11c	0.0000	0.0909	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0170	0.0000	
329-11d	0.5344	0.4950	0.3515	0.4551	0.4780	0.2922	1.1899	0.3604	0.2622	0.3361	0.4742	0.4629	0.7637	0.6024	
329-11e	0.0400	0.0233	0.0350	0.0235	0.0456	0.0172	0.0843	0.0354	0.0206	0.0069	0.0478	0.0373	0.0554	0.0473	
373-15b	0.2644	0.2457	0.0522	0.3520	0.3424	0.0885	0.1712	0.4375	0.1482	0.1348	0.2806	0.3367	0.1663	0.1918	
373-15c	0.5388	0.1661	0.4819	0.3271	0.1323	0.0000	0.0000	0.2561	0.2482	0.0000	0.1311	0.3467	0.0000	0.0000	
361-17a	0.2379	0.0497	0.2154	0.0819	0.0544	0.0297	0.0454	0.1339	0.0764	0.0000	0.1601	0.0484	0.0236	0.1958	
361-17b	0.0649	0.0211	0.0458	0.0283	0.0256	0.0265	0.0333	0.0511	0.0346	0.0177	0.0404	0.0266	0.0256	0.0764	
331-18a	0.0437	0.0158	0.0192	0.0148	0.0163	0.0215	0.0435	0.0201	0.0212	0.0184	0.0259	0.0267	0.0396	0.0331	
331-18b	0.3667	0.7235	0.2291	0.7731	0.7236	0.3291	0.4643	0.6252	0.6119	0.0000	0.0000	0.8514	0.7599	0.8409	
331-18d	0.0558	0.1570	0.1614	0.0924	0.0000	0.1497	0.0670	0.1388	0.1024	0.0000	0.0349	0.0000	0.0392	0.1833	

**Supplementary Figure 4: Phytocannabinoid analysis of the *Cannabis* extracts.** ECI-LC/MS phytocannabinoid profiling of 14  $\Delta^9$ -THC-rich *Cannabis* extracts which were prepared as described in Materials and Methods. *Cannabis* extracts are ordered by increasing content of  $\Delta^9$ -THC (% w/w) in each extract with values color-coded according to concentration order of magnitude of concentration. Only cannabinoids identified at a concentration of at least 0.2 (%w/w) in at least one extract were included in this figure.



**Supplementary Figure 5: The effect of different high  $\Delta^9$ -THC extracts on the survival of A549 cells.** A dose-response curve of A549 cells after 24 h incubation with 2-10  $\mu\text{g/ml}$  of seven high- $\Delta^9$ -THC *Cannabis* extracts or pure  $\Delta^9$ -THC, according to  $\Delta^9$ -THC concentration in each extract. Data are reported as mean  $\pm$  SE of % dead cells out of total cells (N=5).

**Supplementary Table 1: Effect of *Cannabis* extracts on the survival of cancer cell lines**

	A549	NCIH460	PC3	LNcAP	HT29	SW480	A431	A375	MDA231	MCF7	U87MG	T98G
CAN1	>10	>10	>10	7.91 ± 0.75	>10	>10	>10	>10	>10	>10	>10	>10
CAN2	3.89 ± 0.17	4.61 ± 0.57	6.07 ± 0.36	5.99 ± 0.19	>10	3.92 ± 0.36	6.26 ± 0.33	4.75 ± 0.16	3.57 ± 0.36	7.04 ± 0.31	3.32 ± 0.14	3.18 ± 0.25
CAN3	7.43 ± 0.15	7.88 ± 0.56	9.39 ± 0.22	>10	>10	6.33 ± 0.39	9.16 ± 0.44	7.82 ± 0.30	5.74 ± 0.47	>10	5.94 ± 0.20	5.12 ± 0.21
CAN4	5.84 ± 0.20	6.59 ± 0.39	7.86 ± 0.22	4.44 ± 0.35	>10	6.16 ± 0.50	8.92 ± 0.39	6.75 ± 0.20	7.35 ± 0.52	>10	7.55 ± 0.42	6.69 ± 0.28
CAN5	3.65 ± 0.19	4.68 ± 0.77	5.94 ± 0.22	6.22 ± 0.27	>10	3.70 ± 0.39	5.90 ± 0.45	4.97 ± 0.21	4.54 ± 0.17	6.21 ± 0.23	4.16 ± 0.13	3.53 ± 0.20
CAN6	8.30 ± 0.21	7.70 ± 0.63	9.14 ± 0.18	4.29 ± 0.35	>10	5.75 ± 0.43	>10	7.92 ± 0.34	8.35 ± 0.55	>10	>10	8.54 ± 0.41
CAN7	3.06 ± 0.18	4.11 ± 0.81	3.85 ± 0.41	5.21 ± 0.30	>10	3.71 ± 0.38	5.74 ± 0.42	4.51 ± 0.33	3.24 ± 0.19	5.11 ± 0.21	2.97 ± 0.13	3.07 ± 0.17
CAN8	>10	>10	>10	7.34 ± 0.60	>10	>10	>10	>10	>10	>10	>10	>10
CAN9	4.90 ± 0.14	5.17 ± 0.55	6.36 ± 0.28	7.77 ± 0.42	>10	4.42 ± 0.30	6.99 ± 0.30	4.92 ± 0.20	3.90 ± 0.31	8.11 ± 0.47	3.71 ± 0.15	3.25 ± 0.17
CAN10	4.33 ± 0.15	4.92 ± 0.42	5.69 ± 0.33	6.03 ± 0.20	>10	4.81 ± 0.53	9.96 ± 0.58	4.97 ± 0.36	3.77 ± 0.27	7.82 ± 0.59	3.64 ± 0.13	3.15 ± 0.22
CAN11	>10	>10	>10	7.39 ± 0.53	>10	>10	>10	>10	>10	>10	>10	>10
CAN12	6.85 ± 0.15	8.04 ± 0.48	8.92 ± 0.11	>10	>10	7.01 ± 0.48	9.23 ± 0.48	8.35 ± 0.31	8.64 ± 1.13	>10	6.39 ± 0.12	6.40 ± 0.16
Δ <sup>9</sup> -THC	7.37 ± 0.18	7.73 ± 1.02	>10	>10	>10	9.94 ± 0.82	9.41 ± 1.14	9.23 ± 0.96	7.24 ± 0.32	>10	6.08 ± 0.29	6.23 ± 0.35

Various cancer cell lines were treated with (2-10 µg/ml) concentrations of *Cannabis* extracts and 2-10 µg/ml of pure Δ<sup>9</sup>-THC for 24 h. Cells were imaged via ImageXpress Micro XLS (20× magnification) after adding Hoechst and PI dyes to differentiate between live and dead cells. The number of live and dead cells were analyzed by MetaXpress® software. Data are reported as mean ± S.E. of LC50 values (µg/ml) (n ≥ 5).

**Supplementary Table 2: Correlation between phytocannabinoid content and LC50 values of 14  $\Delta^9$ -THC-rich extracts on the A549 cell line**

<b>Cannabinoid</b>	<b>r</b>	<b>p-value</b>	<b>Adjusted p-value</b>
<b>CBG</b>	0.6664	0.0002	0.0031
<b>CBT-3</b>	0.4621	0.0045	0.0582
<b>THC</b>	0.4553	0.0049	0.0582
<b>329-11d</b>	0.4346	0.0062	0.0677
<b>373-15c</b>	0.3237	0.0198	0.1976
<b>CBC</b>	0.2995	0.0250	0.2246
<b>CBT-1</b>	0.2151	0.0540	0.4316
<b>THCA</b>	0.1805	0.0729	0.5103
<b>THCV</b>	0.1335	0.1087	0.6521
<b>CBN</b>	0.0712	0.1831	0.9154
<b>331-18b</b>	-0.0383	0.4847	1.0000
<b>CBD</b>	-0.0416	0.5011	1.0000
<b>CBGA</b>	-0.0689	0.6940	1.0000
<b>329-11b</b>	-0.0798	0.8470	1.0000

r- Pearson correlation coefficient, adjusted p-value is according to the Holm-Bonferroni correction method for multiple tests.