



**Fig. S1.** Effects of *trans*-10-hydroxy-2-decenoic acid (10H2DA) on: a) HCT-116 and b) SW-480 cell viability assessed with MTT assay

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**Table S1.** Forward and reverse primer sequences of target genes

Gene	Forward sequence	Reverse sequence
<i>β-actin</i>	5'-AAGCAGGAGTATGACGAGTCCG-3'	5'-GCCTTCATACATCTCAAGTTGG-3'
<i>E-cadherin</i>	5'-GAACAGCACGTACACAGCCCT-3'	5'-GCAGAACTGTCCCTGTCCAG-3'
<i>β-catenin</i>	5'-AAAATGGCAGTGCCTTTAG-3'	5'-TTTGAAGGCAGTCTGTCTGA-3'
<i>N-cadherin</i>	5'-GACGGTTCGCCATCCAGAC-3'	5'-TCGATTGGTTTGACCACGG-3'
<i>vimentin</i>	5'-GGCTCAGATTCAGGAACAGC-3'	5'-AGCCTCAGAGAGGTCAGCAA-3'
<i>Snail</i>	5'-TCAGACGAGGACAGTGGGAAAG-3'	5'-GCTTGTGGAGCAGGGACATTC-3'

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**Table S2.** The effect of royal jelly and *trans*-10-hydroxy-2-decenoic acid (10H2DA) on the migration of HCT-116 and SW-480 cells. The analysis of wound space is shown as a relative level of changes of wound space compared to control cells (100 %)

HCT-116	Relative wound space/%		
	t/h		
$\gamma$ (royal jelly)/( $\mu$ g/mL)	0	12	24
0	100.00 $\pm$ 0.01	(52.4 $\pm$ 0.9) <sup>a</sup>	(38.4 $\pm$ 0.8) <sup>a</sup>
10	100.00 $\pm$ 0.01	(77.2 $\pm$ 1.1) <sup>ab</sup>	(60.1 $\pm$ 1.2) <sup>ab</sup>
100	100.00 $\pm$ 0.01	(88.1 $\pm$ 0.7) <sup>ab</sup>	(77.6 $\pm$ 0.4) <sup>ab</sup>
c(10H2DA)/ $\mu$ M			
0	100.00 $\pm$ 0.01	(52.4 $\pm$ 0.9) <sup>a</sup>	(38.4 $\pm$ 0.8) <sup>a</sup>
10	100.00 $\pm$ 0.01	(80.6 $\pm$ 1.2) <sup>ab</sup>	(75.0 $\pm$ 0.8) <sup>ab</sup>
100	100.00 $\pm$ 0.01	(92.5 $\pm$ 1.0) <sup>ab</sup>	(80.8 $\pm$ 0.8) <sup>ab</sup>
SW-480			
$\gamma$ (royal jelly)/( $\mu$ g/mL)			
0	100.00 $\pm$ 0.01	(64.9 $\pm$ 1.0) <sup>a</sup>	(51.4 $\pm$ 0.5) <sup>a</sup>
10	100.00 $\pm$ 0.01	(87.8 $\pm$ 1.2) <sup>ab</sup>	(65.1 $\pm$ 0.5) <sup>ab</sup>
100	100.00 $\pm$ 0.01	(83.4 $\pm$ 0.6) <sup>ab</sup>	(76.1 $\pm$ 1.1) <sup>ab</sup>
c(10H2DA)/ $\mu$ M			
0	100.00 $\pm$ 0.01	(64.9 $\pm$ 1.0) <sup>a</sup>	(51.4 $\pm$ 0.5) <sup>ab</sup>
10	100.00 $\pm$ 0.01	(87.9 $\pm$ 2.4) <sup>ab</sup>	(75.7 $\pm$ 3.8) <sup>ab</sup>
100	100.00 $\pm$ 0.01	(98.5 $\pm$ 1.6) <sup>b</sup>	(85.2 $\pm$ 2.4) <sup>ab</sup>

Results are presented as mean value $\pm$ S.E. of two independent experiments performed in four repeats. <sup>a</sup>p<0.05 is considered as statistically significant difference between treatments compared to control at the same time, and <sup>b</sup>p<0.05 is considered as statistically significant difference between concentrations in a treated group

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**Table S3.** Migratory and invasive potential of HCT-116 and SW-480 cells after 24 h of treatment with different concentrations of royal jelly and *trans*-10-hydroxy-2-decenoic acid (10H2DA)

HCT-116	Index of migration or invasion/ $A_{595\text{ nm}}$	
	Migration	Invasion
Control	0.449 $\pm$ 0.04	0.303 $\pm$ 0.03
$\gamma$ (royal jelly)/( $\mu$ g/mL)		
10	(0.34 $\pm$ 0.03) <sup>a</sup>	0.27 $\pm$ 0.0
100	(0.37 $\pm$ 0.01) <sup>a</sup>	0.28 $\pm$ 0.01
c(10H2DA)/ $\mu$ M		
10	(0.35 $\pm$ 0.01) <sup>a</sup>	0.25 $\pm$ 0.01
100	(0.34 $\pm$ 0.02) <sup>a</sup>	(0.22 $\pm$ 0.01) <sup>ab</sup>
SW-480		
Migration		
Control	0.32 $\pm$ 0.01	0.27 $\pm$ 0.02
$\gamma$ (royal jelly)/( $\mu$ g/mL)		
10	0.31 $\pm$ 0.01	0.23 $\pm$ 0.01
100	(0.25 $\pm$ 0.01) <sup>ab</sup>	0.22 $\pm$ 0.02
c(10H2DA)/ $\mu$ M		
10	0.32 $\pm$ 0.01	(0.20 $\pm$ 0.01) <sup>a</sup>
100	0.32 $\pm$ 0.01	(0.19 $\pm$ 0.00) <sup>a</sup>

The results are shown as the index of migration or invasion/absorbance ( $A_{595\text{ nm}}$ ) of a mean value $\pm$ S.E. from two representative experiments with four repeats. <sup>a</sup>p<0.05 is considered as statistically significant difference between treatments compared to control (0 h), and <sup>b</sup>p<0.05 is considered as statistically significant difference between concentrations in a treated group

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**Table S4.** Protein expression of E-cadherin, cytoplasmic  $\beta$ -catenin, nuclear  $\beta$ -catenin, N-cadherin, vimentin and Snail in HCT-116 and SW-480 cells after 24 h of treatments

HCT-116	Relative fluorescence per cell				
	Control	$\gamma$ (royal jelly)/( $\mu$ g/mL)			$c$ (10H2DA)/ $\mu$ M
	0	10	100	10	100
E-cadherin	229063 $\pm$ 14286	(315815 $\pm$ 42224) <sup>ab</sup>	225086 $\pm$ 10569	233211 $\pm$ 12262	256808 $\pm$ 17822
Cyt. $\beta$ -catenin	233982 $\pm$ 21882	285954 $\pm$ 41082	300825 $\pm$ 40804	(441501 $\pm$ 8548) <sup>a</sup>	(420314 $\pm$ 45492) <sup>a</sup>
Nucl. $\beta$ -catenin	236446 $\pm$ 14331	(86391 $\pm$ 4873) <sup>a</sup>	(98592 $\pm$ 14193) <sup>a</sup>	(111902 $\pm$ 15083) <sup>a</sup>	(116397 $\pm$ 11744) <sup>a</sup>
N-cadherin	1175936 $\pm$ 78681	(235817 $\pm$ 48593) <sup>a</sup>	(73712 $\pm$ 15143) <sup>ab</sup>	(236668 $\pm$ 23124) <sup>a</sup>	(71845 $\pm$ 13543) <sup>ab</sup>
vimentin	265795 $\pm$ 29716	(104279 $\pm$ 8012) <sup>a</sup>	(112400 $\pm$ 3427) <sup>a</sup>	(86382 $\pm$ 5056) <sup>a</sup>	(27611 $\pm$ 7289) <sup>ab</sup>
Snail	1059201 $\pm$ 69763	(313915 $\pm$ 12316) <sup>a</sup>	(64239 $\pm$ 16773) <sup>ab</sup>	(126957 $\pm$ 25256) <sup>a</sup>	(61528 $\pm$ 2826) <sup>a</sup>
<b>SW-480</b>					
E-cadherin	135741 $\pm$ 5721	142876 $\pm$ 6156	(276734 $\pm$ 15416) <sup>ab</sup>	(187225 $\pm$ 8306) <sup>a</sup>	(167475 $\pm$ 9402) <sup>a</sup>
Cyt. $\beta$ -catenin	586952 $\pm$ 38904	(143203 $\pm$ 16065) <sup>a</sup>	(192632 $\pm$ 23267) <sup>a</sup>	(86647 $\pm$ 9801) <sup>a</sup>	(147070 $\pm$ 16421) <sup>a</sup>
Nucl. $\beta$ -catenin	121893 $\pm$ 19868	107706 $\pm$ 5552	(171573 $\pm$ 27781) <sup>b</sup>	120369 $\pm$ 24178	143990 $\pm$ 22778
N-cadherin	170722 $\pm$ 26364	(41748 $\pm$ 5924) <sup>a</sup>	(12941 $\pm$ 1507) <sup>a</sup>	(17400 $\pm$ 751) <sup>a</sup>	(14387 $\pm$ 1460) <sup>a</sup>
Vimentin	283305 $\pm$ 13283	(43283 $\pm$ 3202) <sup>a</sup>	(43138 $\pm$ 3482) <sup>a</sup>	(56968 $\pm$ 3534) <sup>a</sup>	(82762.0 $\pm$ 3469) <sup>ab</sup>
Snail	267145 $\pm$ 20043	(11190 $\pm$ 10631) <sup>a</sup>	(61938 $\pm$ 8098) <sup>ab</sup>	(66069 $\pm$ 8487) <sup>a</sup>	(53633 $\pm$ 3912) <sup>a</sup>

Values are presented as a mean value $\pm$ S.E. of the triplicates from two representative experiments. <sup>a</sup> $p$ <0.05 is considered as statistically significant difference between treatments compared to control (0 h), and <sup>b</sup> $p$ <0.05 is considered as statistically significant difference between concentrations in a treated group. Cyt=cytoplasmic, Nucl=nuclear

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**Table S5.** Results of gene expression are presented as the fold change in mRNA expression in a target sample, normalized to a reference gene and relative to the control sample

HCT-116	<i>E-cadherin</i>	$\beta$ -catenin	<i>N-cadherin</i>	<i>vimentin</i>	<i>Snail</i>
Control	1	1	1	1	1
$\gamma$ (royal jelly)/( $\mu$ g/mL)					
10	(3.2 $\pm$ 0.2) <sup>ab</sup>	(1.2 $\pm$ 0.3) <sup>a</sup>	(0.7 $\pm$ 0.2) <sup>a</sup>	(0.75 $\pm$ 0.09) <sup>a</sup>	(0.8 $\pm$ 0.3) <sup>a</sup>
100	1.3 $\pm$ 0.2	(1.4 $\pm$ 0.2) <sup>ab</sup>	(0.20 $\pm$ 0.02) <sup>ab</sup>	(0.48 $\pm$ 0.04) <sup>ab</sup>	(0.72 $\pm$ 0.08) <sup>ab</sup>
$c$ (10H2DA)/ $\mu$ M					
10	(1.3 $\pm$ 0.1) <sup>ab</sup>	(2.71 $\pm$ 0.08) <sup>ab</sup>	(0.57 $\pm$ 0.08) <sup>a</sup>	(0.4 $\pm$ 0.1) <sup>a</sup>	(0.81 $\pm$ 0.02) <sup>a</sup>
100	1.0 $\pm$ 0.4	(2.37 $\pm$ 0.02) <sup>a</sup>	(0.42 $\pm$ 0.07) <sup>ab</sup>	(0.16 $\pm$ 0.04) <sup>ab</sup>	(0.4 $\pm$ 0.1) <sup>ab</sup>
<b>SW-480</b>					
Control	1	1	1	1	1
$\gamma$ (royal jelly)/( $\mu$ g/mL)					
10	(1.35 $\pm$ 0.08) <sup>a</sup>	(0.18 $\pm$ 0.03) <sup>a</sup>	(0.13 $\pm$ 0.03) <sup>ab</sup>	(0.69 $\pm$ 0.09) <sup>a</sup>	(0.48 $\pm$ 0.08) <sup>a</sup>
100	(2.89 $\pm$ 0.04) <sup>ab</sup>	(0.61 $\pm$ 0.03) <sup>ab</sup>	(0.15 $\pm$ 0.03) <sup>a</sup>	(0.63 $\pm$ 0.09) <sup>ab</sup>	(0.18 $\pm$ 0.08) <sup>ab</sup>
$c$ (10H2DA)/ $\mu$ M					
10	(1.41 $\pm$ 0.03) <sup>a</sup>	(0.53 $\pm$ 0.001) <sup>a</sup>	(0.11 $\pm$ 0.001) <sup>a</sup>	(0.10 $\pm$ 0.01) <sup>a</sup>	(0.35 $\pm$ 0.02) <sup>a</sup>
100	(2.1 $\pm$ 0.1) <sup>ab</sup>	(0.58 $\pm$ 0.004) <sup>ab</sup>	(0.10 $\pm$ 0.003) <sup>ab</sup>	(0.10 $\pm$ 0.01) <sup>ab</sup>	(0.11 $\pm$ 0.02) <sup>ab</sup>

Values of relative gene expression were calculated according to  $2^{-\Delta\Delta Ct}$  method. <sup>a</sup> $p$ <0.05 indicates a significant difference compared to control, while <sup>b</sup> $p$ <0.05 indicates a significant difference between concentrations in treated group