Supporting Information for the manuscript entitled "Enhanced morphological transformation of human lung epithelial cells by continuous exposure to cellulose nanocrystals"

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Table S1. The average dimensions of CNC powder/gel and tremolite fibers

CNC	Diameter, nm	Length, nm	Width, nm
CNC Powder	149±2.6	158±97	54±17
CNC Gel	137.5±1.2	209±136	37±15
Tremolite Fibers		6560±210	310±10

The average dimensions of CNC materials were determined using DLS/AFM measurements. The CNC results are presented as mean ± SD. The reported values (CNC) correspond to the mean of ten separate runs. The average size of tremolite fibers were determined using TEM (n=817) with data presented as mean ± SEM.

Table S2. The degree of oxidative stress induced by CNC in BEAS-2B cells

	Control	CNC Powder	CNC Gel
GSH nmol/mg protein	10.72±0.18	7.52±0.71*	8.45±0.75*
SH nmol/mg protein	166.86±6.33	154.15±8.66	146.39±10.72
HNE-His μg/mg protein	15.85±0.26	17.77±0.49*	15.47±1.52
Carbonyls nmol/mg protein	0.22±0.01	0.25±0.01	0.26±0.02

The results are expressed as the mean ± SEM (n=3), * indicate significant differences from control cells ($p \leq 0.05$).

Table S3. Differential response in various inflammatory cytokines (CNC, 30 μg/cm^2, once a week/4 weeks)

pg/total cells, x10^{-6}	Control	CNC powder	CNC gel
IL-1β	0.05±0.01	0.11±0.01*$^{\alpha}$	0.05±0.01
IL-1rα	2.63±0.55	6.30±1.13*$^{\alpha}$	1.70±0.06
IL-2	0.28±0.07	0.67±0.10*$^{\alpha}$	0.30±0.02
IL-4	0.11±0.02	0.22±0.02*$^{\alpha}$	0.06±0.01
IL-6	10.40±2.12	51.70±6.19*$^{\alpha}$	8.39±1.32
IL-5	ND	ND	ND
IL-7	1.10±0.22	1.01±0.07	0.89±0.14
IL-8	22.92±4.60	147.38±14.78*$^{\alpha}$	28.60±3.66
IL-9	0.49±0.08	1.04±0.13*	0.55±0.07
IL-10	1.07±0.21	1.11±0.14	1.27±0.20
IL-12p70	19.26±4.04	19.76±3.18	16.54±2.98
IL-13	0.52±0.08	0.63±0.06	0.65±0.10
IL-15	0.34±0.08	0.74±0.06*	0.55±0.07
IL-17	3.38±0.67	5.85±1.00	4.21±0.58
Eotaxin	0.86±0.18	2.53±0.36*$^{\alpha}$	0.86±0.08
FGF-basic	1.48±0.29	2.13±0.31	1.52±0.26
G-CSF	3.53±0.69	14.68±2.52*$^{\alpha}$	2.92±0.42
GM-CSF	1.89±0.41	3.44±0.59	3.06±0.41
IFN-γ	10.22±2.09	13.94±2.45$^{\alpha}$	3.54±0.44
IP-10	7.80±1.86	30.19±0.92*$^{\alpha}$	3.70±0.28
MCP-1	599.77±125.23	1103.17±171.91	625.18±121.27
MIP-1α	0.05±0.01	0.07±0.01	0.05±0.0.01
PDGF-bb	17.55±4.22	3.71±0.93*	2.29±0.42*
MIP-1β	0.23±0.47	0.26±0.04	0.31±0.04
RANTES	1.14±0.21	1.52±0.09	0.51±0.05*$^{\beta}$
TNF-α	1.08±0.18	2.60±0.35*$^{\alpha}$	0.71±0.08
VEGF	ND	ND	ND

These measurements were performed using Bio-Rad 27 human assay kits, a combination of inflammatory cytokines with a subset of chemokine's/growth factors. The results are expressed as the mean ± SEM (n=3), *indicate significant differences from control cells, $^{\alpha}$ indicate significant difference from CNC gel-

exposed cells, $^{\beta}$ indicate significant difference from CNC powder-exposed cells ($p \leq 0.05$). VEGF and IL-5 were not detected (ND).

Table S4. Differential response in various inflammatory cytokines (TF, 2.5 μg/cm^2, once a week/4 weeks)

pg/total cells, x10^{-6}	Control	Tremolite
IL-1β	0.11±0.02	0.24±0.01*
IL-1rα	0.63±0.15	2.03±0.04*
IL-2	0.52±0.11	0.95±0.08*
IL-4	0.13±0.02	0.22±0.01*
IL-5	0.34±0.1	0.64±0.03*
IL-6	0.22±0.04	0.39±0.02*
IL-7	0.24±0.01	0.92±0.17
IL-8	0.49±0.02	0.76±0.04*
IL-9	ND	ND
IL-10	ND	ND
IL-12p70	0.29±0.01	0.38±0.01*
IL-13	0.01±0.00	0.05±0.00*
IL-15	0.51±0.07	0.94±0.11*
IL-17	4.91±0.90	8.39±0.63*
Eotaxin	1.47±0.23	2.73±0.15*
FGF-basic	229.85±56.55	561.03±65.63*
G-CSF	3.51±0.92	5.42±0.66
GM-CSF	0.28±0.02	0.45±0.03*
IFN-γ	5.34±1.04	9.28±0.54*
IP-10	0.91±0.17	1.61±0.09*
MCP-1	0.85±0.08	1.54±0.05*
MIP-1α	0.06±0.00	0.08±0.01
PDGF-bb	2.42±0.23	3.85±0.35*
MIP-1β	0.07±0.01	0.09±0.01
RANTES	0.46±0.06	0.73±0.05*
TNF-α	1.47±0.23	2.52±0.24
VEGF	8.18±0.48	13.48±0.91*

These measurements were performed using Bio-Rad 27 human assay kits, a combination of inflammatory cytokines with a subset of chemokine's/growth factors. The results are expressed as the mean ± SEM (n=2-3), *indicate significant differences from control cells ($p \leq 0.05$). IL-9 and IL-10 were not detected (ND).

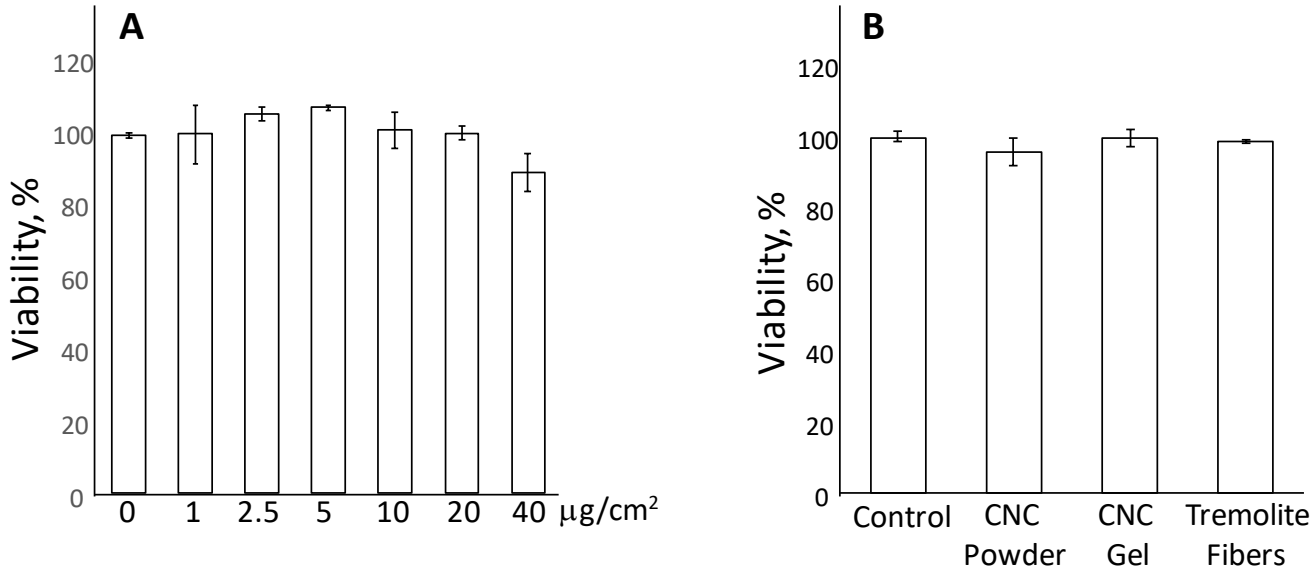

Figure S1. Cell cytotoxicity following exposure to CNC or TF: (A) viability of BEAS-2B cells exposed to different concentrations of TF for 24h; (B) viability of BEAS-2B cells exposed to CNC (powder or gel, 30 μg/cm^2) and TF (2.5 μg/cm^2) for 72h. The results are expressed as the mean ± SEM (n=2-3).

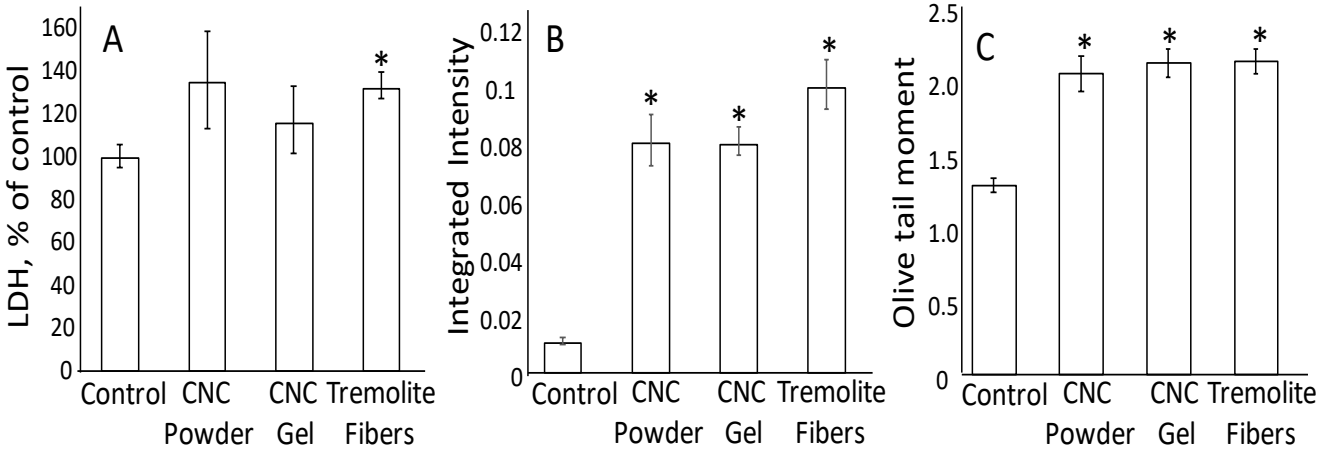

Figure S2. Cell cytotoxicity (A), intracellular ROS (B) and DNA damage represented by olive tail moment (C), after 72h of BEAS-2B cells exposure with CNC (powder or gel, 30 μg/cm^2) and TF (2.5 μg/cm^2). The results are expressed as the mean ± SEM (n=3), *indicate significant differences from control cells ($p \leq 0.05$).

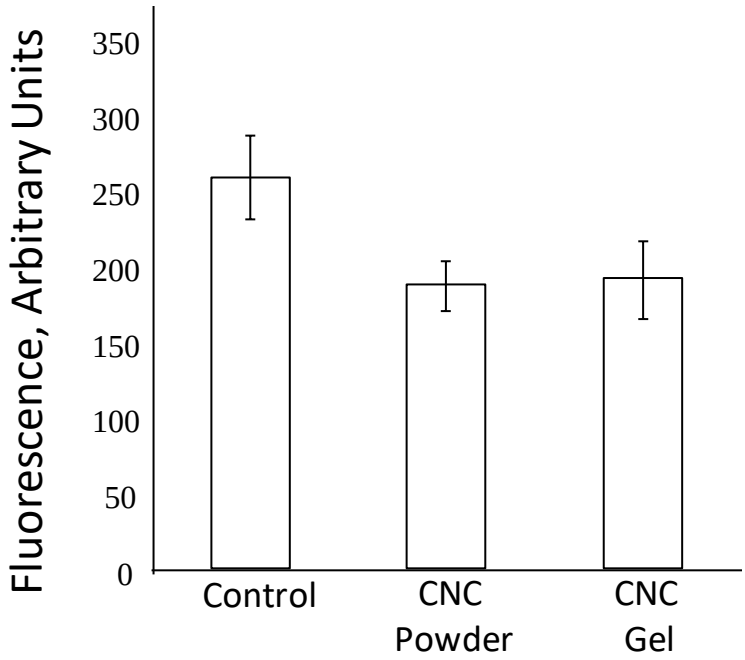

Figure S3. Apoptosis analysis in BEAS-2B cells exposed to CNC powder or gel (30 μg/cm^2/week) using Cell Meter TUNEL assay kit. Fluorescence intensity was monitored at Ex/Em 550/590nm. The results are expressed as the mean ± SEM of the three independent experiments.