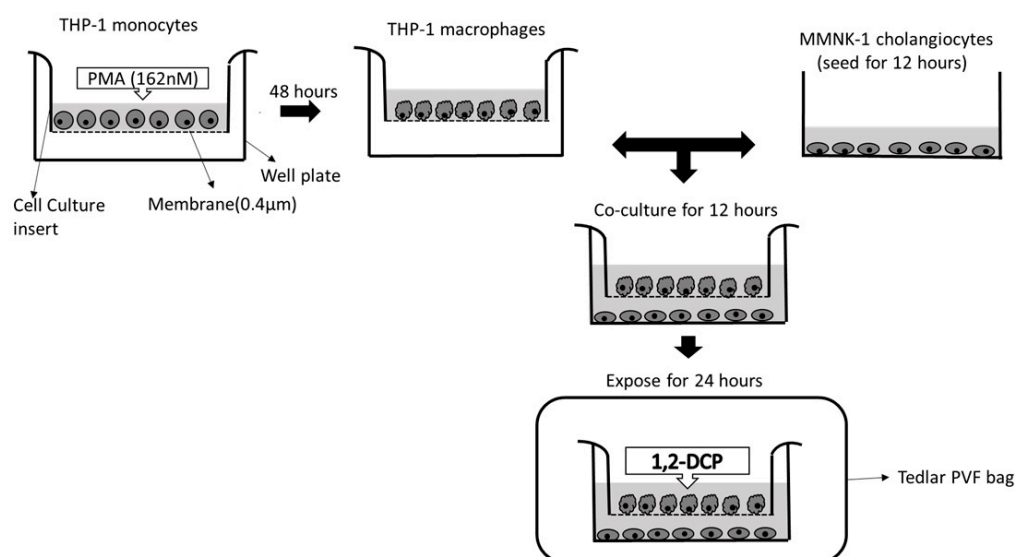


# Supplementary Materials: Role of Macrophages in Cytotoxicity, Reactive Oxygen Species Production and DNA Damage in 1,2-Dichloropropane-Exposed Human Cholangiocytes In Vitro

Abigail Ekuban, Cai Zong, Frederick Adams Ekuban, Yusuke Kimura, Ryoya Takizawa, Kota Morikawa, Kazuo Kinoshita, Sahoko Ichihara, Seiichiroh Ohsako and Gaku Ichihara



**Figure S1.** Illustration of the co-culture model. THP-1 monocytes were differentiated into macrophages by PMA at 162 nM concentration by seeding into cell culture inserts (pore size membrane: 0.4 μm) for 48 hours. MMNK-1 cholangiocytes were seeded in well plate for 12 hours and then combined with THP-1 macrophages. MMNK-1 cholangiocytes and THP-1 macrophages were co-cultured for 12 hours and then exposed to different concentrations of 1,2-DCP for 24 hours. The MMNK-1 cholangiocytes and THP-1 macrophages were placed into Tedlar polyvinyl fluoride (PVF) gas sampling bags to prevent evaporation of 1,2-DCP and incubated at 37°C.

**Table S1.** Protein concentration values for proinflammatory cytokines expression analysis by ELISA.

1,2-DCP Concentration(mM)	Protein Concentration (pg/ml)	
	TNF- $\alpha$	IL-1 $\beta$
0	41 $\pm$ 10	0.9 $\pm$ 0.7
0.2	301 $\pm$ 91	8 $\pm$ 2
0.4	232 $\pm$ 14	6 $\pm$ 3
0.8	568 $\pm$ 457	187 $\pm$ 43*

Protein concentrations are expressed as mean  $\pm$ SD, \*p<0.05, compared with corresponding control by one-way ANOVA followed by Dunnett's test multiple comparison test.