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Table and Figure Legends

Table 1. Serum and pulmonary levels of PC-SOD. Mice treated with or without bleomycin (5 mg/kg) once-only at day 0 were administered indicated doses of PC-SOD (kU/kg or kU/chamber) intravenously, intratracheally or by inhalation once daily for 3 days. Blood and pulmonary tissue were taken 6 h after the final administration of PC-SOD. Levels of PC-SOD in samples were determined by ELISA. Values are mean ± S.E.M. (n.d., not detected).

Table 2. Effect of PC-SOD on pulmonary level of hydrogen peroxide. Mice were administered indicated doses of PC-SOD (kU/kg or kU/chamber) intravenously or by inhalation once daily for 3 days. Lungs were removed and the amount of hydrogen peroxide was determined. Values are mean ± S.E.M. ** $P<0.01$.

Table 3. Effect of inhalation of U-SOD on bleomycin-induced inflammatory response. Mice were treated with bleomycin and the inflammatory response was

assessed as described in the legends of Fig. 1. Indicated doses of U-SOD (kU/chamber) were inhaled once per day for 3 days. Values are mean ± S.E.M.

Figure 1. Effect of intravenous administration of PC-SOD on bleomycin-induced inflammatory response. Mice treated with or without (vehicle) bleomycin (BLM) (5 mg/kg) once-only at day 0 were intravenously administered indicated doses of PC-SOD (kU/kg) once per day for 3 days (A-C). Total cell number, and numbers of alveolar macrophages, lymphocytes and neutrophils were determined after 3 days as described in Materials and Methods (A). Sections of pulmonary tissue were prepared after 3 days and subjected to TUNEL assay and DAPI staining. Similar results were obtained for at least three sections (B). The level of TGF-β1 in pulmonary tissue after 3 days was determined by ELISA (C). Values are mean ± S.E.M. * or $^{\#}P<0.05$; ** or $^{\#\#}P<0.01$ (A, C).

Figure 2. Effect of intravenous administration of PC-SOD on bleomycin-induced pulmonary fibrosis. Mice treated once-only with or without (Control) bleomycin

(BLM) (5 mg/kg) at day 0 were intravenously administered indicated doses of PC-SOD (kU/kg) once per day for 14 days (A-C). Mice treated once-only with bleomycin (BLM) (5 mg/kg) at day 0 were intravenously administered indicated doses of PC-SOD (kU/kg) once per day from day 7 to day 13 (D-F). Sections of pulmonary tissue were prepared after 14 days and subjected to histopathological examination (H & E staining (A, D) or Masson's trichrome staining (B, E)) as described in Materials and Methods. Similar results were obtained for at least three sections (A, B, D, E). The pulmonary hydroxyproline level was determined after 14 days as described in Materials and Methods. Values are mean ± S.E.M. * or $^{\#}P<0.05$; ** or $^{\#\#}P<0.01$ (C, F).

Figure 3. Effect of PC-SOD on cell death, expression of collagen and EMT *in vitro*. A549 (A, C) or HFL-I (B) cells were preincubated with the indicated concentration of PC-SOD for 1 h and further incubated with the indicated concentrations of menadione (A) or TGF-β1 (B, C) for 24 h in the presence of the same concentrations of PC-SOD as in the preincubation step. Cell viability was determined by MTT assay (A). Total RNA was extracted and subjected to real-time RT-PCR using a specific primer set for each

gene. Values were normalized to the actin gene, expressed relative to the control sample (B, C). Values shown are mean ± S.E.M. (n=3). $^{**}P<0.01$ (A-C).

Figure 4. Effect of concurrent administration of catalase on the ameliorative effect of PC-SOD on the bleomycin-induced inflammatory response and fibrosis. Mice were treated with bleomycin (BLM) and PC-SOD, and the inflammatory response (A) and pulmonary fibrosis (B-D) were assessed as described in the legends of Figs.1 and 2. Indicated dose of catalase (Cat) (kU/kg) was intravenously administered once per day for 3 days (A) or 14 days (B-D). Similar results were obtained for at least three sections (B, C). Values are mean ± S.E.M. * or $^{\#}P<0.05$; ** or $^{\#\#}P<0.01$.

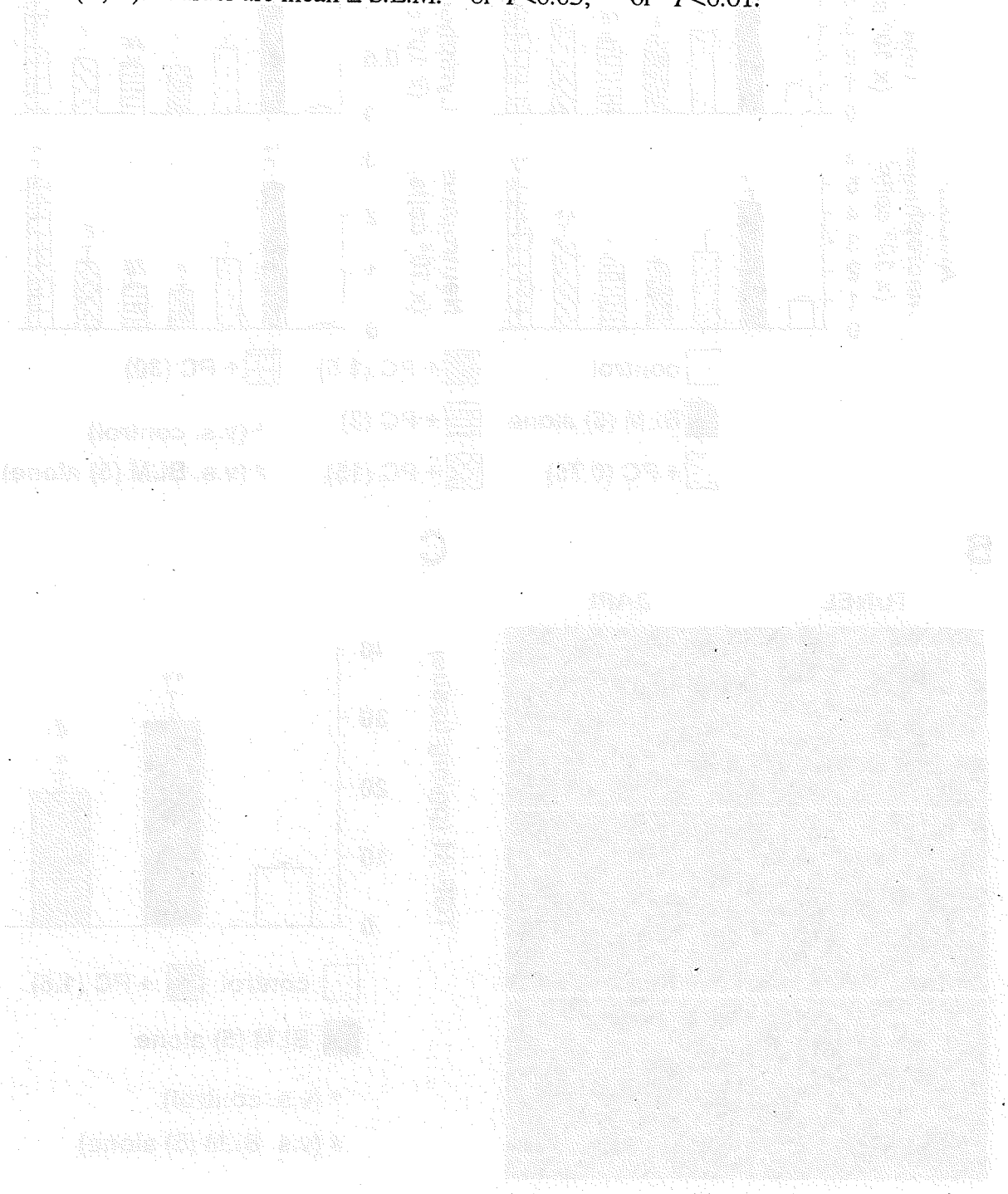
Figure 5. Effect of intratracheal administration of PC-SOD on bleomycin-induced inflammatory response and pulmonary fibrosis. Mice were treated with bleomycin (BLM) and the inflammatory response (A) and pulmonary fibrosis (B-D) were assessed as described in the legends of Figs. 1 and 2. Indicated doses of PC-SOD (kU/kg) were administered intratracheally once per day for 3 days (A) or 14 days (B-D). Similar

results were obtained for at least three sections (B, C). Values are mean ± S.E.M. * or $^{\#}P<0.05$; ** or $^{\#\#}P<0.01$.

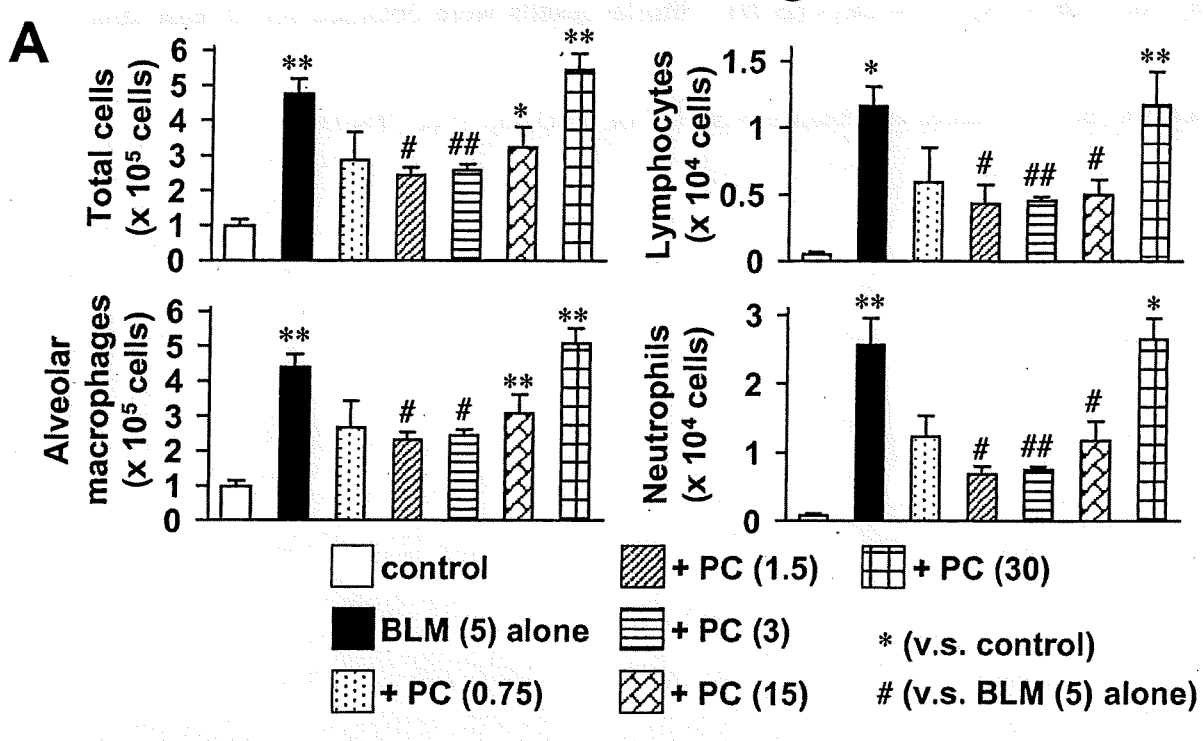
Figure 6. Distribution of PC-SOD in the lung. Mice were treated with bleomycin (BLM) and indicated doses of PC-SOD (kU/kg or kU/chamber) were administered intravenously, intratracheally, or by inhalation once per day for 3 days. Sections of pulmonary tissue (from the two regions shown in the figure) were prepared 6 h after the final administration of PC-SOD (after 3 days) and subjected to immunohistochemical analysis with an antibody against human Cu/Zn-SOD. Similar results were obtained for at least three sections.

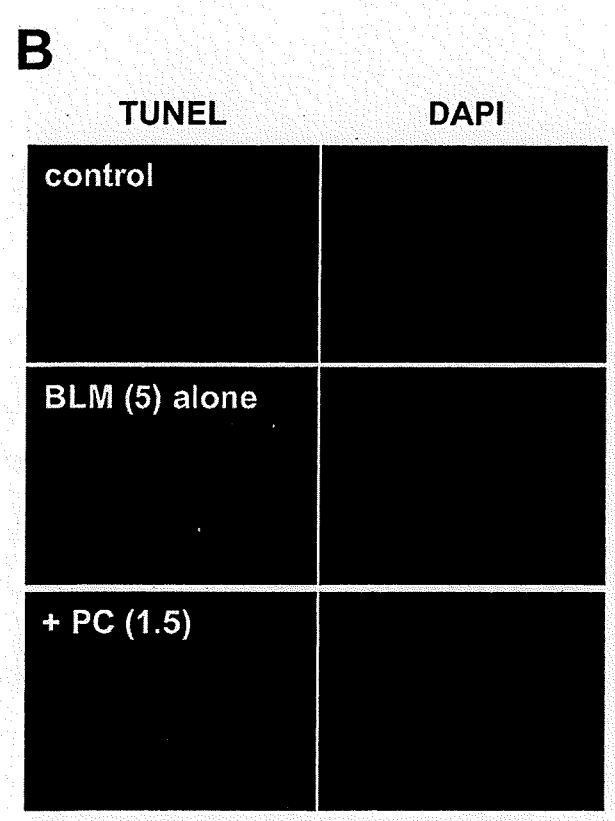
Figure 7. Effect of inhalation of PC-SOD on bleomycin-induced inflammatory response and pulmonary fibrosis. Mice were treated with bleomycin (BLM) and the inflammatory response (A) and pulmonary fibrosis (B-D) were assessed as described in the legends of Figs. 1 and 2. Indicated doses of PC-SOD (kU/kg) were inhaled once per

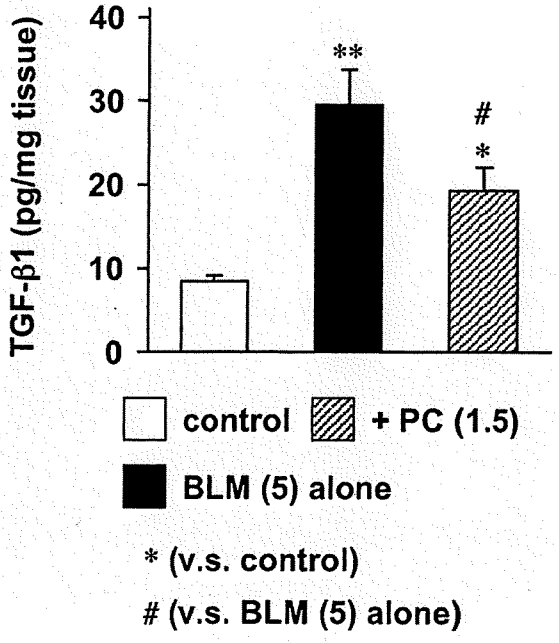
day for 3 days (A) or 14 days (B-D). Similar results were obtained for at least three sections (B, C). Values are mean ± S.E.M. * or $^{\#}P<0.05$; ** or $^{\#\#}P<0.01$.



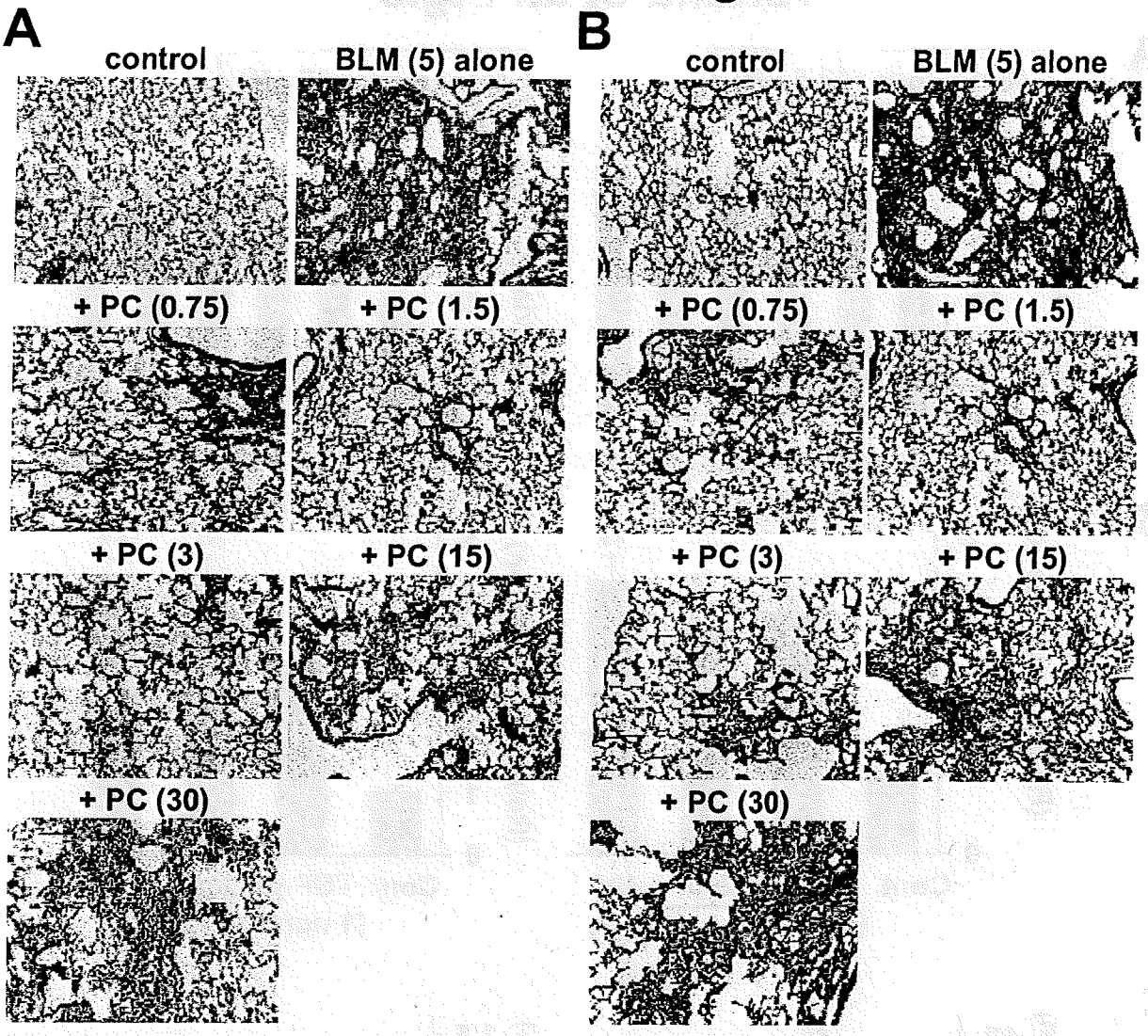
Tanaka *et al.* Fig.1

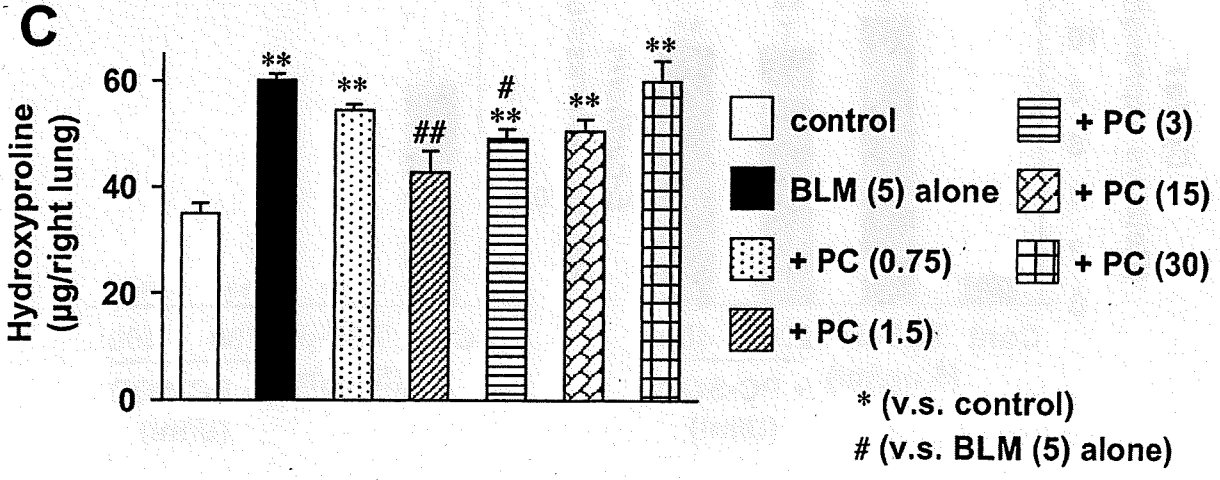






Tanaka *et al.* Fig.2





Tanaka *et al.* Fig.3