

**Tiotropium attenuates virus-induced pulmonary inflammation in
cigarette smoke-exposed mice**

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Data Supplement

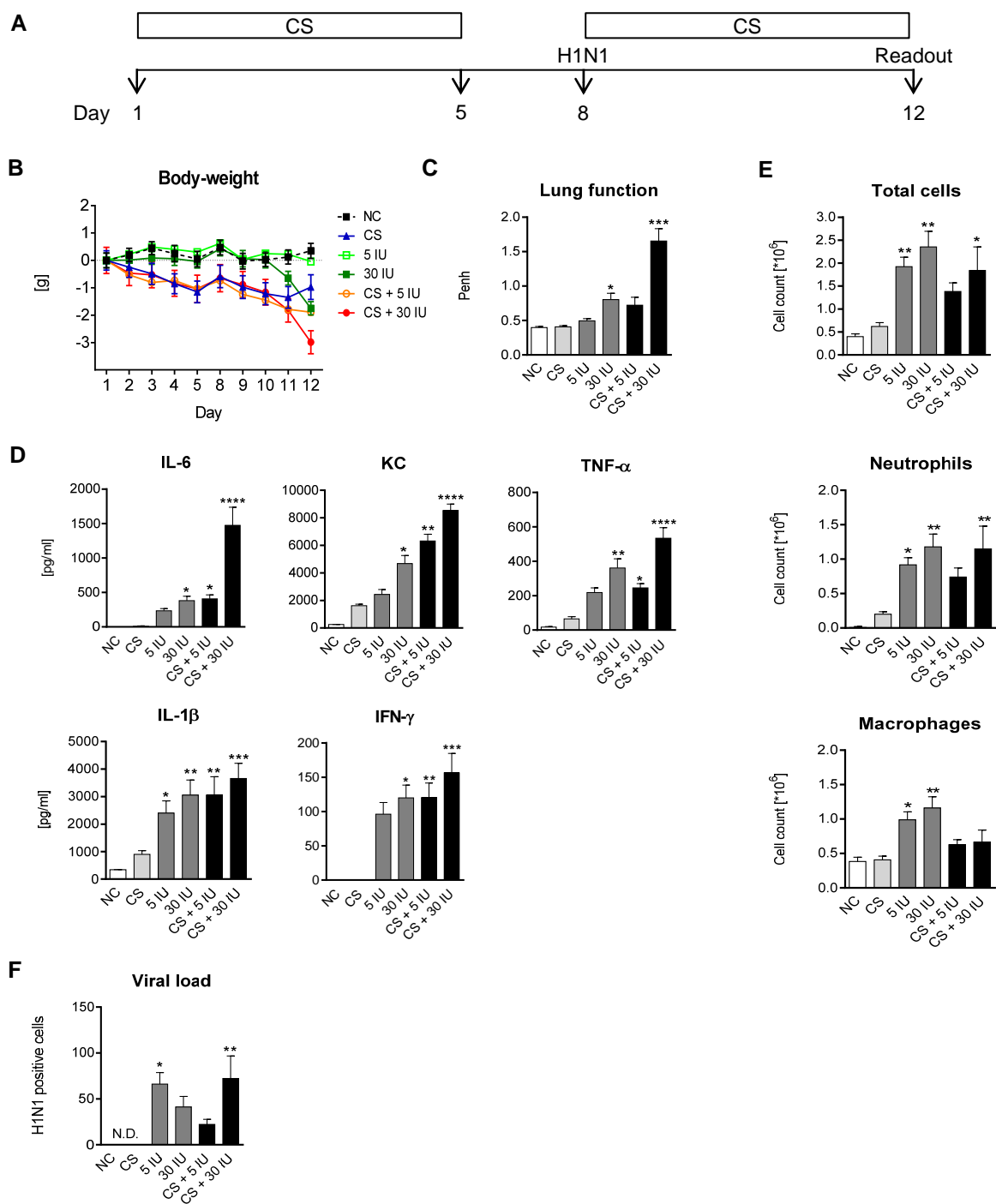


Figure E1. Infection of CS-exposed mice with different dosages of H1N1. (A) Experimental scheme: Mice were exposed to cigarette smoke (CS) (light gray bars), 5 IU H1N1 or 30 IU H1N1 (dark gray bars), CS-exposed and infected with 5 IU H1N1 or 30 IU H1N1 (black bars). White bars illustrate the results from negative control (NC) animals. (B) The body-weight was recorded on each day of the experiment. Negative control animals (black dotted line), CS-exposed (blue line), infected with 5 IU H1N1 (light green line), or 30 IU H1N1 (the dark green line), CS-exposed and infected with 5 IU H1N1 (orange line) or 30 IU H1N1 (red line) are shown. (C) Lung function, (D) cytokine levels in lung homogenate, (E) total cell, neutrophil and macrophage numbers in BAL fluid and (F) viral load in lung homogenate are shown. Mean \pm SEM of $n = 7-8$ animals per treatment group, and $n = 4$ for negative control are shown. **** $p < 0.0001$, *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$ represent significant differences compared to the negative control.

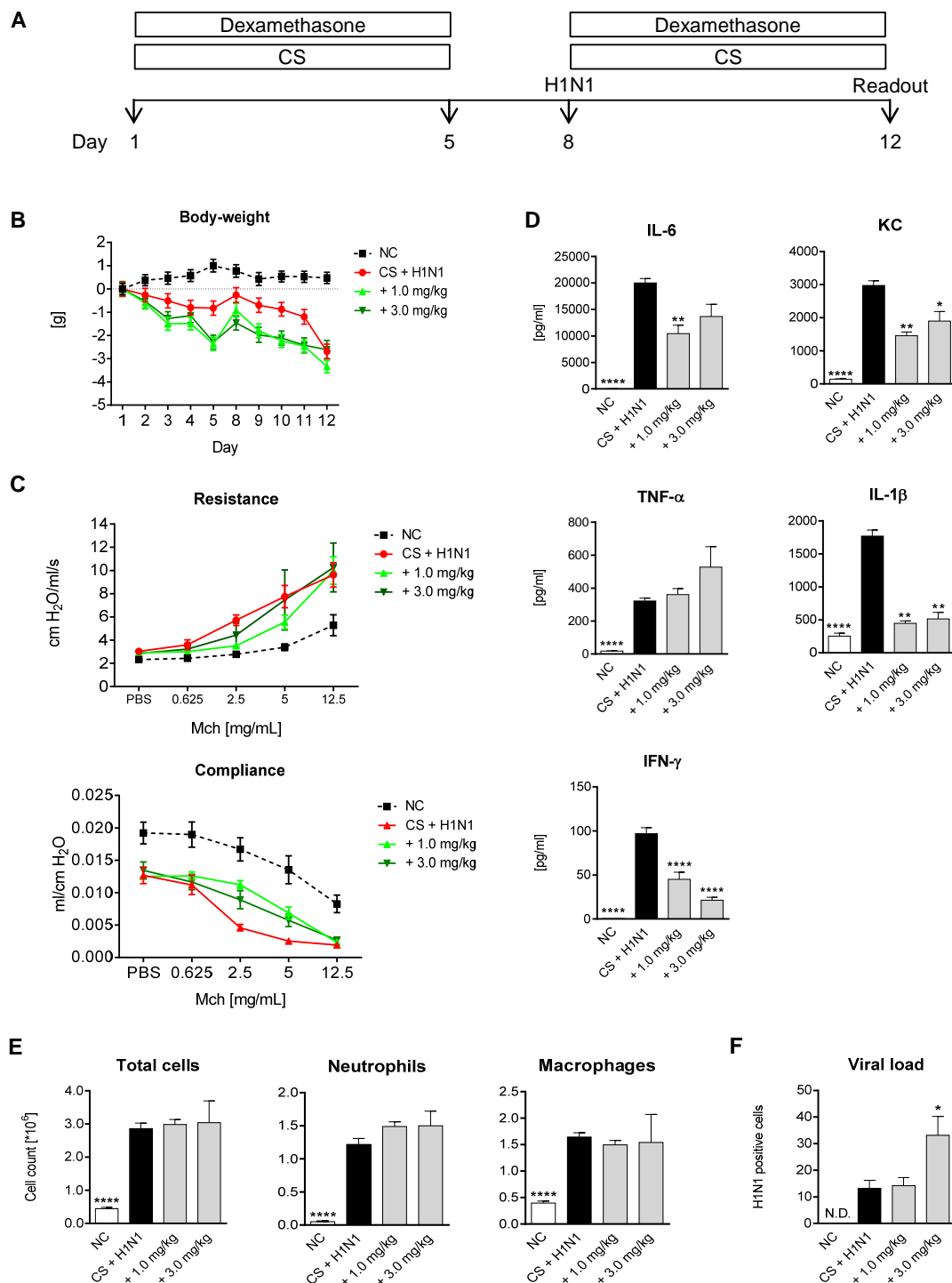


Figure E2. Dexamethasone treatment of CS-exposed and H1N1 infected mice. (A) CS-exposed and H1N1 infected mice (black bars) were treated for a total of ten days with 1.0 mg/kg or 3.0 mg/kg dexamethasone (gray bars). (B) Body-weight loss in negative control (NC) mice (black dotted line), untreated CS-exposed and H1N1 infected mice (red line), CS-exposed and H1N1 infected mice treated with 1.0 mg/kg (light green line) or 3.0 mg/kg (dark green line) dexamethasone is shown. (C) Resistance and compliance, (D) cytokine levels in Wlung homogenate, (E) total cell, neutrophil and macrophage numbers in BAL fluid and (F) viral load in lung homogenate are shown. Mean \pm SEM of $n = 7-8$ animals per group are shown. **** $p < 0.0001$, *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$ represent significant differences compared to the CS-exposed and H1N1 infected group.