## **Supplemental N Value Clarification**

The N values given for the various survival data groups represent the total numbers of animal subjects receiving 4 hour mechanical ventilation treatment respectively. However in order to generate our 1Hr data time points a number of additional subjects were ventilated and sacrificed after one hour. When possible these data were combined with the data collected at the 1Hr time point for our 4Hr subjects. The result is that certain N values for 1Hr data points exceed the N values of their 4Hr counterparts. Additionally, statistical outliers generated naturally through variations in FlexiVents measurements, incremental modifications to our data acquisition protocols, and random deaths of subjects cause a variation in the N values of some experimental groups across various statistical measures. The specific N values for data point are listed in the corresponding figure caption.

## **Supplemental Figure**

Title of Data: Lung tissue elastin stain

Description of data: Departafinized slides were stained using ACCUSTAIN elastic stain
(Sigma, HT25A-1KT) to characterize elastin and collagen content before and after
decellularization. Stained slides were dehydrated and mounted using Permount mounting
medium (Fisher). Stained slides were imaged using an Olympus IX71 Microscope (Olympus).
Representative Control and 4Hr histological elastin images of <u>A</u>. Young Non-Ventilated, <u>B</u>. Old
Non-Ventilated, <u>C</u>. Young LVT-HF, <u>D</u>. Old LVT-HF, <u>E</u>. Young HVT-HF, and <u>F</u>. Old HVT-HF
lung sections. In our older non-ventilated control subjects the elastin lining the alveolar walls is
noticeably sparser and less discretely located compared to their younger counterparts.
Additionally, elastin debris can be seen in alveoli of the older control subjects but not in that of

the young. Injurious mechanical ventilation was also seen to deteriorate elastin. It thinned and

fractured the elastin in the younger subjects and exacerbated these changes in the older subjects. Accordingly the elastin fiber distribution in our injuriously ventilated old subjects was markedly sparser than those of any other group.

## Supplemental 1

