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**Title:** Endogenous particles in exhaled air – Variability of particle number in healthy individuals

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**Body:** Endogenous particles in exhaled air (PEx) are formed during airway reopening following airway closure. Analysis of PEx may be used for monitoring biochemical alterations in respiratory tract lining fluid in the small airways. The number of exhaled particles is strongly dependent on breathing pattern but there is also a substantial difference between individuals. The aim of the study was to assess intra-individual variability of PEx number using a standardized breathing maneuver involving airway closure. 10 healthy subjects participated in the study. They were instructed to exhale slowly and completely, inhale to total lung capacity and exhale at 500 ml/s until functional residual capacity. Particles were counted in the final exhalation of the maneuver using an in-house developed instrument. The maneuver was repeated until 80 liters of exhaled air had been sampled. Sampling was performed 3 times (two mornings and one afternoon) for each subject. The average total number of PEx varied from 9700 to 93 000 particles/exhalation among individuals. The coefficient of variation of total PEx number/exhalation within individuals was 3.9-36%, median 21%. The coefficient of variation between individuals was 65%. There was no intra-individual difference in particle numbers between morning and afternoon samplings. The variability of PEx number was substantially higher between individuals than within individuals. The high variability between individuals may be due to differences in lung architecture and possibly to differences in the chemical composition of airway lining fluid. Our results support the fact that it is important to control for mass of PEx for any given concentration of non-volatile analyte.