In order to confirm a possible association between UU and abnormal sperm function, a prospective study was conducted comparing semen between UU positive (n = 17), UU negative (n = 33), and specimens from healthy donors (n = 21). Measurements of reactive oxygen species (ROS), leukocytospermia, sperm motion characteristics, acrosome reaction (AR), and mannose binding assay (MBA) were conducted. The most striking results were the following: UU positive patients had significantly higher ROS (2,391.82 ± 1,141.54) than UU negative patients (1,038.22 ± 555.63) (P = 0.006), or donors (63.9 ± 30.9) (P = 0.02). Elevated ROS levels induce lipid peroxidation which decreases membrane fluidity and sperm fertilizing capacity. Surprisingly, leukocytospermia was detected in only 1 of the 17 (6%) UU positive specimens. Our findings have significant clinical implications as leukocytospermia detection is widely used as a screening tool for infection in the infertile male. As demonstrated by the abnormal ROS measurements, UU may potentially impair sperm function and would otherwise go undetected in this setting unless specific cultures were obtained. In conclusion, UU infection is associated with absence of leukocytospermia; therefore, cultures are recommended for evaluation of infertile men.