



Supplementary Figure 2

Calibration using supported lipid bilayers.

(a) Periodicity of SAIM function at differing oxide heights. Theoretical intensity is plotted vs. angle for 488 nm (green lines) or 561 nm (red lines) laser illumination at 25 (light lines) nm or 50 nm (dark lines) from the surface of the oxide. Four oxide heights are represented: 500 nm, 1000 nm, 1500 nm and 1900 nm. **(b)** Measurement of oxide layer thickness. Three separate wafers were measured by ellipsometry at 8 positions spaced at increments of ~ 1 cm apart. Error bars represent standard deviation of these eight measurements. “All” depicts the mean and standard deviation of 24 measurements across three wafers (1930.5 ± 2.0 nm). “SAIM” depicts the mean and standard deviation for the deduced oxide height at three wavelengths (488 nm, 561 nm, 647 nm), on two different microscopes and on five separate days (1928.5 ± 5.6 nm) for wafer #3. **(c)** Using SAIM to deduce oxide thickness. A triple labeled bilayer (DiO, DiI, DiD) was measured by SAIM at three wavelengths (488 nm, 561 nm, 647 nm). These data were then fit using oxide thicknesses ranging from 1915 nm to 1935 nm, which is plotted here versus the resultant sample height. Vertical dashed line represents the oxide thickness measured by ellipsometry (1930.5 ± 2.0 nm). Horizontal dashed line represents the expected height of the supported lipid bilayer (6.4 nm) [Supplementary reference 10]. Deduced oxide height is the intersection of the horizontal line for true bilayer height and the data. **(d)** Daily wavelength-dependent variation in oxide height measurements. Data for “SAIM” in **b** are shown as individual data points to highlight day-to-day variability.