ADAPTIVE PHASE COMPENSATION IN MULTIPHOTON MICROSCOPY

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Shorter laser pulses should be able to yield increased signal intensity and improved resolution when applied to multiphoton microscopy. Yet, the dispersion introduced by high numerical aperture objectives and other dispersive elements in the laser beam path has prevented their effective use. Fig. 1 demonstrated the pulse duration expansion due to the group delay dispersion.

The addition of a pulse shaper controlled by an algorithm referred to as mutiphoton intrapulse interference phase scan (MIIPS) can measure and compensate the pulse dispersion [1]. The pulse width calculated by the spectrum of the laser was confirmed independently by interferometric autocorrelation measurements at the objective focus [2]. We report the comparison of two-photon imaging with MIIPS-compensated pulses and laser pulses corrected only for group delay dispersion with a standard prism-pair compressor. The resulting ≥5-fold improvement in signal magnitude is demonstrated on fixed and living cells, as well as in sections from mouse liver and brain.

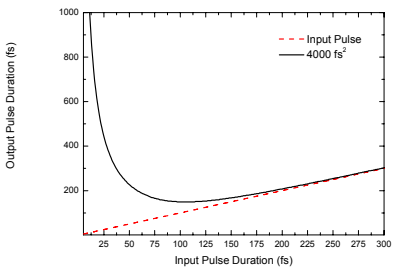

Fig. 1 The pulse expansion for dispersion of GDD at -4000fs^2. The amount of GDD is similar to that of a high NA objective.

References

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