A laser scanning microscope produces molecular excitation in a target material by simultaneous absorption of two photons to thereby provide intrinsic three-dimensional resolution. Fluorophores having single photon absorption in the short (ultraviolet or visible) wavelength range are excited by a stream of strongly focused subpicosecond pulses of laser light of relatively long (red or infrared) wavelength range. The fluorophores absorb at about one half the laser wavelength to produce fluorescent images of living cells and other microscopic objects. The fluorescent emission from the fluorophores increases quadratically with the excitation intensity so that by strongly focusing the laser light, fluorescence as well as photobleaching are confined to the vicinity of the focal plane. This feature provides depth of field resolution comparable to that produced by confocal laser scanning microscopes, and in addition reduces photobleaching. Scanning of the laser beam, by a laser scanning microscope, allows construction of images by collecting two-photon excited fluorescence from each point in the scanned object while still satisfying the requirement for very high excitation intensity obtained by focusing the laser beam and by pulse time compressing the beam. The focused pulses also provide three-dimensional spatially resolved photochemistry which is particularly useful in photolytic release of caged effector molecules.