



APPLIED SCIENTIFIC
INSTRUMENTATION

双向光片照明显微镜系统

这套设备可以安装在现有的反向显微镜，
或配置为一套完整的RAMM系统。

这套系统比传统的显微镜系统有很多优势：

使用常规安装/玻璃盖片

产生各向同性分辨率三维体积（330nm范围内的所有方向）

比共聚焦或旋转磁盘系统 轴向分辨率高2倍

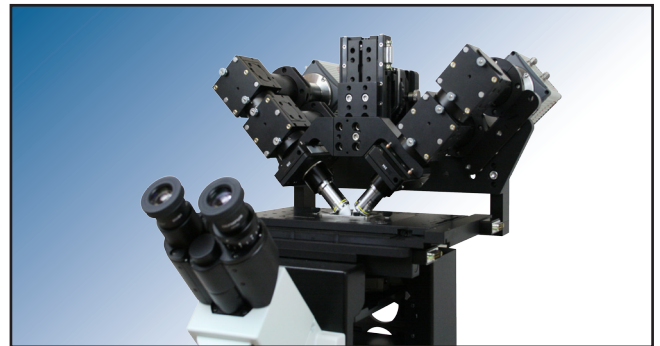
可实现7-10倍减少光漂白（约10倍更快于旋转盘显微镜）

采集速率达200帧每秒或2-5卷每秒

相比贝塞尔光束平面照明方法，

这套系统提供更完美的轴向分辨率（~330nm），

容积成像速率快10-100倍（0.5-1代替10-100s），
并且能够成像的超过10倍以上的时间点（1000而不是100），
由于较低的照射剂量和外部照明在焦平面的缺乏。
显微镜上有彩色的能力，并已在盖玻片培养细胞试验成功，
嵌入在胶原凝胶的细胞，和线虫，斑马鱼胚胎。



其他特点

1) 固定片系统（单面）

光片与片的位置固定，只有机械调整。

这两个目标是手动调整正确的集中表。

该样品扫描通过X和Z的阶段产生体积图像片。

该系统的优点是：

T是最昂贵的不需要振镜扫描仪或压目标定位器。

缺点是：

正确重叠的目标焦平面固定定位器和相对较慢的阶段，
比较难控制比较偏的角度

2) 单面系统标准。

从一个侧光片，排放在其他目的。

光片可使用电流计席卷样品体积扫描。

有一个发射目的压所以观察目的可以
定位跟踪光片是通过样品的扫描。

优点：快速扫描，直接设置。

缺点：XY Z分辨率更好的分辨率比。

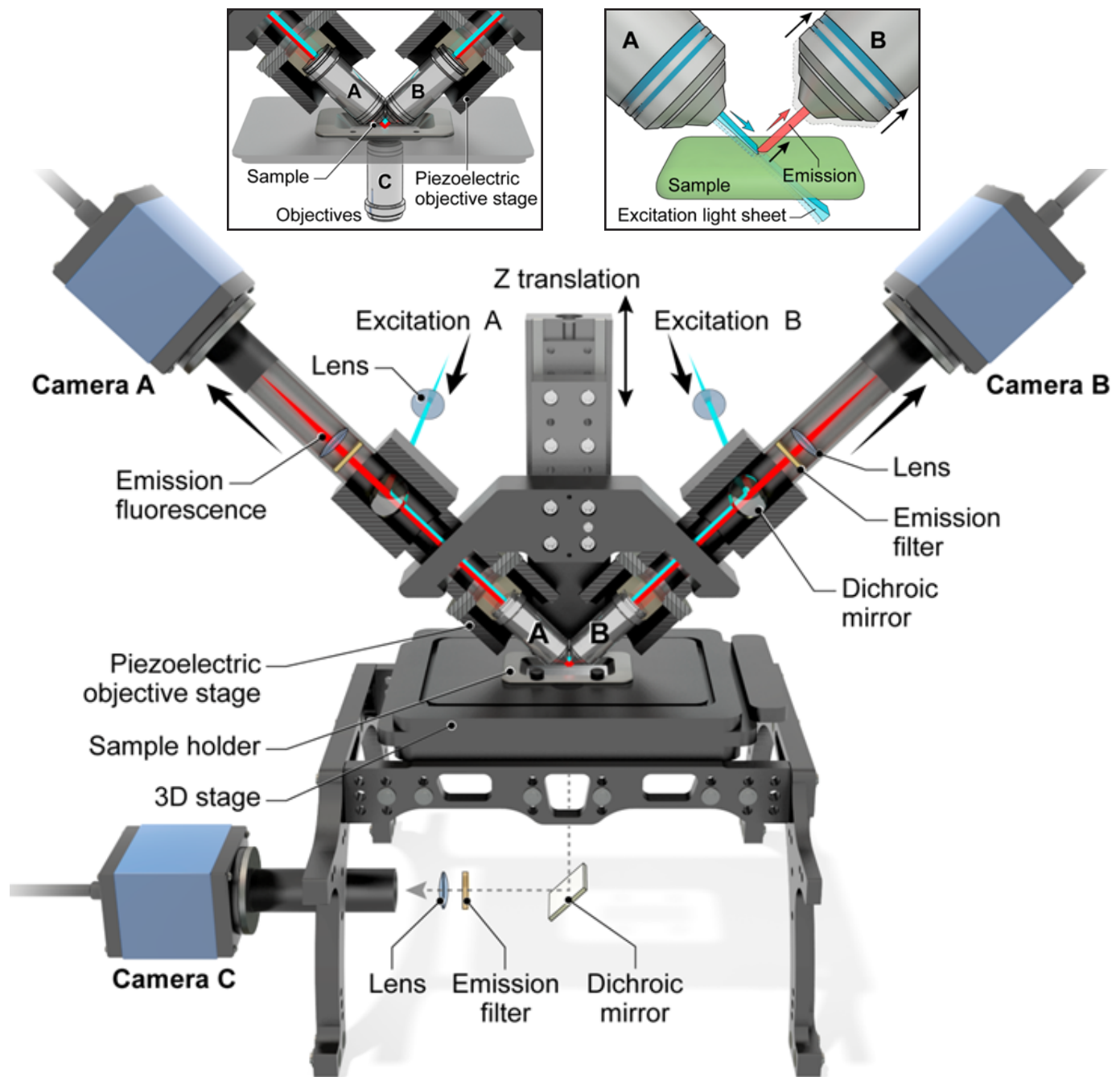


Fig. 1. Dual-View iSPIM Setup

0.8 NA water-immersion objectives (A/B) are mounted orthogonally onto a Z translation stage that is bolted directly onto the illumination pillar of an inverted microscope. In conjunction with other optics (Supplementary Fig. 1), both objectives produce a light sheet at the sample. Excitation A(B) occurs via objective A(B), and the resulting fluorescence is collected through perpendicular objective B(A), and imaged onto Camera B(A) via dichroic mirrors, emission filters, and lenses. Excitation (blue) and detection (red) are shown occurring simultaneously along both light paths in the lower schematic, but in reality volumetric imaging occurs sequentially as shown in the upper right inset. During acquisition, sample and objective A(B) are held stationary, the light sheet is scanned through the sample using galvanometric mirrors (not shown), and a piezoelectric objective stage moves objective B(A) in sync with the light sheet, ensuring that excitation/detection planes are coincident. The sample is mounted onto a rectangular coverslip that is placed onto a 3D translation stage, ensuring correct placement relative to objectives. The sample may also be viewed through objective C (see upper left inset), dichroic mirror, emission filter, lens and Camera C placed in the conventional light path of the inverted microscope. This objective is particularly useful in finding or screening samples.