Unsupervised Clustering of multiparametric fluorescent images allows for the detection of new microfunctional units in living cells.

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Spectrally resolved fluorescence lifetime imaging can provide pixel-resolved multiparametric information. For example, solvatochromic probes reveal contextual information about the biophysical state of the membranes, like membrane hydration, microviscosity and the partition coefficient of the probe. Label free autofluorescence microscopy retrieves data on biophysical and biochemical state of the metabolites. However, the different parameters are analysed independently, and their correlations at pixel level are integrated out, thus providing only limited insights in unravelling the membrane phases of natural cell membranes and their spatial organization, or the activation of metabolic networks at the microscale. To overcome these issues, here we introduce an artificial intelligence-based analysis that, leveraging the multiparametric content of spectrally resolved lifetime images, allows to detect and classify, through an unsupervised learning approach, microfunctional units in living cells. This method, applied to the analysis of membrane phases through solvatochromic probes, allows to extend the spectrum of detectable membrane phases, and to study real-time phase transitions in cultured cells and tissues[1]. The same method, applied to label free autofluorescence analysis, allows for the detection of metabolic clusters, which are regions having almost uniform metabolic properties, giving information on the cellular mitochondrial turnover and on the metabolic activation state of intracellular compartments at the pixel level [2].

References

[1] Unsupervised clustering of multiparametric fluorescent images extends the spectrum of detectable cell membrane phases with sub-micrometric resolution
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[2] “Label-free metabolic clustering through unsupervised pixel classification of multiparametric fluorescent images”
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