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Saliency-guided compressive fluorescence microscopy

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***Abstract*— A novel saliency-guided approach is proposed for improving the acquisition speed of compressive fluorescence microscopy systems. By adaptively optimizing the sampling probability density based on regions of interest instead of the traditional unguided random sampling approach, the proposed saliency-guided compressive fluorescence microscopy approach can achieve high-quality microscopy images using less than half of the number of fluorescence microscopy data measurements required by existing compressive fluorescence microscopy systems to achieve the same level of quality.**

I. Introduction

Fluorescence microscopy has broad applications in molecular studies of individual proteins and living cells [7]. A key benefit of fluorescence microscopy over those based on optical density changes and chemiluminescent emission is its greater sensitivity and range [7]. A popular fluorescence microscopy approach is scanning confocal microscopy, where samples are scanned by a laser to reconstruct an image [6]. However, such an approach has been traditionally limiting in terms of acquisition speed due to the image being acquired pixel by pixel.

To address this limitation, the concept of compressive fluorescence microscopy (CFM) was proposed to greatly increase acquisition speed while preserving high reconstruction quality [10], [11]. CFM is based around compressive sensing (CS) [4], [5], which allows for greatly reduced fluorescence microscopy acquisition times by accurately recovering the image from sparse, sub-Nyquist measurements. Wu et al. [11] illustrated the applicability of CFM for fast optical-sectioning imaging, while Studer et al. [10] illustrated its applicability for fast hyperspectral imaging.

Much of the research in CFM systems focuses on hardware design and reconstruction design, thus leaving the design of the sampling procedure largely unexplored. However, given that many applications of fluorescence microscopy involve regions of interest with structured characteristics, the design of the sampling procedure can have a tremendous effect on acquisition speed and reconstruction quality.

Motivated by the importance of the sampling procedure on CFM performance, the main contribution of this paper is the introduction of a saliency-guided compressive fluorescent microscopy system, which incorporates a saliency-guided sparse measurement model [9] that was developed to significantly improves reconstruction quality and acquisition speed for situations where regions of interest have structured characteristics.

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II. Saliency-guided CFM framework

Existing CFM systems employs a traditional sampling scheme that sample the entire scene in the same manner regardless of the underlying data. However, such an approach is limited for many practical applications of fluorescence microscopy, which involve distinct regions of interest with highly salient structural characteristics. Given that such regions are of greater interest for analysis purposes, one is motivated to obtain higher quality reconstructions for these regions than the background regions. To tackle this issue with traditional sampling approaches, we propose the optimization and integration of the state-of-the-art saliency-guided sparse measurements model [9] into the CFM imaging framework.

Let the scene being imaged via fluorescence microscopy contain $M \times N$ sampling locations organized in a finite, rectangular lattice $\Omega_{M\times N}$:

$$\Omega_{M\times N} = \{(m,n) \mid m = 0,\ldots,M-1, n = 0,\ldots,N-1\} \quad (1)$$

Given such a lattice, let us partition $\Omega_{M\times N}$ into three complementary sets Ω_D, Ω_S and Ω_{DS}^c (Ω_D represents highly salient locations, Ω_S represents sparse sampling, and Ω_{DS}^c represent non-sampled locations) such that

$$\Omega_{M\times N} = \Omega_D \cup \Omega_S \cup \Omega_{DS}^c, \text{ with } \Omega_D \cap \Omega_S \cap \Omega_{DS}^c = \emptyset, \quad (2)$$

whose cardinalities are equal to $\#(\Omega_D + \Omega_S) = T$ and $\#\Omega_{DS}^c = MN - T$, respectively.

Since Ω_D represents highly salient locations, Ω_D is defined based on a function $\Gamma(m,n)$ that quantifies saliency at a given location (m,n):

$$\Gamma(m,n) = \begin{cases} 1, & \text{if } (m,n) \in \Omega_D \\ 0, & \text{if } (m,n) \notin \Omega_D \end{cases} \quad (3)$$

Since regions of interest in many CFM applications are characterized by large spatial intensity variations (which relates to structural characteristics), one can define a saliency function $\Gamma(m,n)$ (3) as:

$$\Gamma(m,n) \to \Omega_D, \text{ if } S(m,n) > t_a, \ \forall(m,n) \quad (4)$$

where

$$S(m,n) = \|I_\mu - I(m,n)\| \quad (5)$$

where I_μ is the mean, $I(m,n)$ is the corresponding vector of the Laplacian of the Gaussian filtered data, and t_a is the threshold value (set at two times the mean saliency $S(m,n)$ of a given data [1]).

Now, with the fluorescence microscopy measurements representing discrete intensity quantities $f : \Omega_{M\times N} \to \mathbb{R}$,

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given a collection of $K \leq NM$ *sampling functions* $\{\varphi_k\}_{k=1}^{K}$, the linear measurements of f can be expressed as:

$$y_k = \langle f, \varphi_k \rangle + e_k = \sum_{m=0}^{M-1} \sum_{n=0}^{N-1} f(m,n)\, \varphi_k(m,n) + e_k, \quad (6)$$

where $k = 1, 2, \ldots, K$ and e_k represents the combined effect of measurement and quantization noises.

In the proposed saliency-guided CFM framework, since there are no measurements in Ω_{DS}^c and the sampling distributions of Ω_D and Ω_S are different, the sampling function at each location can be defined by:

$$\varphi_k(m,n) = \begin{cases} \varphi_k^D(m,n), & \text{if } (m,n) \in \Omega_D \\ \varphi_k^S(m,n), & \text{if } (m,n) \in \Omega_S \\ 0, & \text{if } (m,n) \in \Omega_{DS}^c \end{cases} \quad (7)$$

where $\varphi_k^D(m,n)$ are realizations of a Gaussian distributed random variable x:

$$p_D(x \mid \mu, \sigma) = \eta(x|\mu, \sigma^2), \quad (8)$$

and $\varphi_k^S(m,n)$ are realizations of a Gauss-Bernoulli distributed random variable x:

$$p_S(x \mid \pi, \sigma) = \pi\, \delta(x) + (1-\pi)\, \mathcal{N}(x|0, \sigma^2), \quad (9)$$

where $p_S = 0$ with probability π and p_S is Gaussian distributed with probability $(1-\pi)$. In this implementation, $\mu = 0$, $\sigma = 1$, and π was selected to be 0.9 (represents 90% compression rate or 10% sampling) as the maximum compression rate where CFM can still produce reasonably reconstructed images.

III. Implementation

Based on the above theory, the saliency-guided CFM framework can be implemented in two phases. In the first phase, a traditional sampling procedure is employed where f is sampled sparsely via $\varphi_k^S(m,n)$ with the pdf p_S (9) using only 10% of the sampling locations. The saliency function $\Gamma(m,n)$ (4,5) is then employed to determine subset Ω_D based on the fluorescence microscopy image reconstructed from these samples.

In the second phase, f is sampled by $\varphi_k^D(m,n)$ with pdf p_D(8) to sample highly salient regions of interest with greater density. The samples from the two subsets Ω_D and Ω_S are then combined and used to reconstruct the fluorescence microscopy image at a higher accuracy than that achieved in the first phase.

To reconstruct the fluorescence microscopy image from the acquired samples, a $\ell 1$-based total variation minimization approach was employed:

$$\arg\min_{f} \left\{ \lambda \|f\|_{TV_{l_1}} + \frac{1}{2} \|\Phi \bar{f} - \bar{y}\|_2^2 \right\} \quad (10)$$

where $\|\cdot\|_{TV_{l_1}}$ denotes the $\ell 1$-based anisotropic total variation norm defined by [2], and can be solved using Fast Iterative Shrinkage-Thresholding Algorithm (FISTA) approach [3], [2].

IV. Experimental results and discussion

Two sets of experiments were performed to investigate and evaluate the capabilities of the proposed saliency-guided compressive fluorescence microscopy (SGCFM) approach for improving reconstruction quality over existing unguided random sampling approaches employed in existing CFM systems. In the first set of experiments, the reconstruction performance of SGCFM is evaluated at different compression rates to study the quality gains in a systematic and quantitative manner. It is important to note that an increase in compression rate results in a decrease in the number of samples used for reconstruction, and hence an decrease in acquisition time. In the second set of experiments, the reconstruction performance of SGCFM is evaluated qualitatively using real noisy fluorescence microscopy data. For comparison purposes, the traditional unguided sampling strategy employed in existing CFM systems [10], [11] is also evaluated as a baseline reference.

A. Experimental Setup

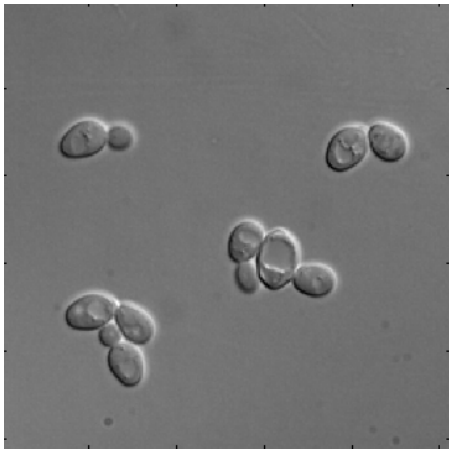
Both experiments were performed with existing fluorescence microscopy data sets [8]. The data set used for the first set of experiments is YRC PIR ID: 64, which is largely noiseless and has the following imaging settings: exposure time: 0.05s, pixel size: 0.12758 μm $\times$ 0.12758 μm, objective: 100$\times$ and image size 512$\times$512. An example of fully sampled data (where each sampling location is measured), and the corresponding synthetic noise contaminated version used in the first set of experiments, are shown in Fig. 1.

In the second set of experiments, the data set used is YRC PIR ID: 8565, which is heavily contaminated by noise and has the following imaging settings: exposure time: 0.40 s, pixel size: 0.12758 μm $\times$ 0.12758 μm, objective: 100$\times$ and image size 512$\times$512. An example of noisy fully sampled data used in the second set of experiments is shown in Fig. 2.

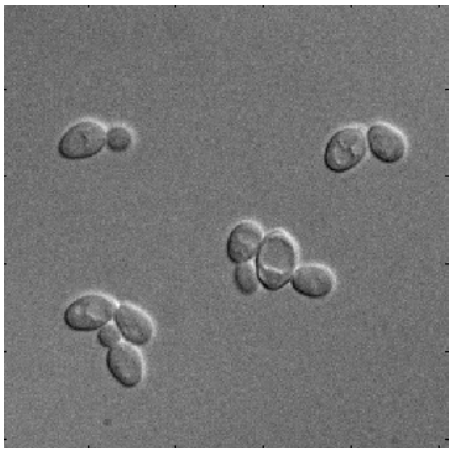
B. Experiment 1 - compression rate sensitivity tests

In the first set of experiments, the reconstruction performance of SGCFM was evaluated using a comprehensive parametric analysis approach, where the signal-to-noise ratio (SNR) of the reconstructed fluorescence microscopy image was computed for a wide range of compression rates, where the compression rate ρ is defined as one minus the ratio between the number of sampling locations measured and the total number of sampling locations. As such, the higher the compression rate, the fewer measurements are made and the faster the acquisition speed. For illustrative purposes, the SNR was measured for the reconstructed results of fluorescence microscopy images (contaminated by Gaussian noise with noise with a standard deviation that is 3% of the dynamic range) across the compression rate range of 0% - 80% (as illustrated in Fig. 3).

Fig. 3 shows that the proposed SGCFM approach can achieve similar SNR performance as traditional CFM approaches using significantly fewer samples, which translates to significant increases in acquisition speed. For example, the SGCFM approach achieves an SNR of 25 dB using



(a)



(b)

Fig. 1. Example of (a) fully sampled fluorescence microscopy image, and (b) the corresponding noise contaminated version (standard deviation = 3%) used in the first set of experiments.

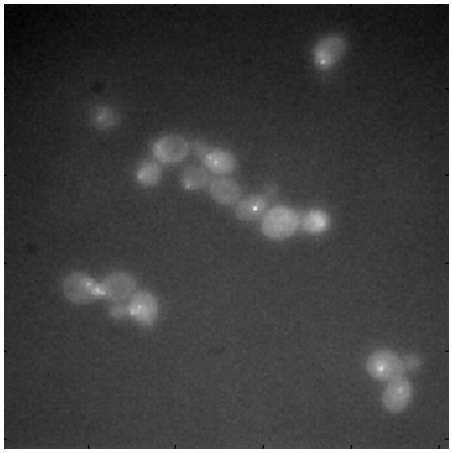


Fig. 2. An example of noisy fully sampled data used in the second set of experiments.

just 39% of the samples (61% compression rate), while the traditional sampling strategy employed in existing CFM systems requires 87% of the samples (13% compression rate) to achieve similar performance. Another way of looking at

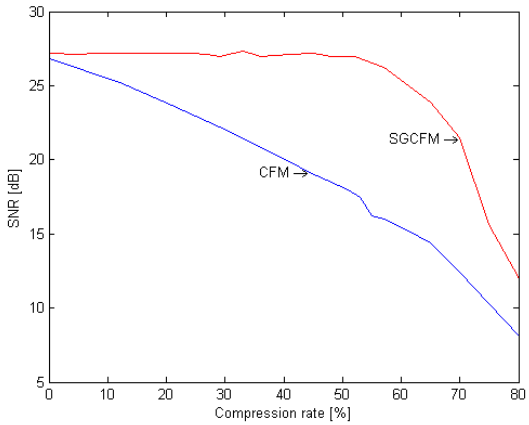


Fig. 3. SNR vs. compression rate for reconstruction results of fluorescence microscopy images contaminated by Gaussian noise with noise with a standard deviation that is 3% of the dynamic range

it is that the proposed SGCFM approach requires less than half (44%) of the samples needed by traditional sampling approaches to achieve the same level of performance.

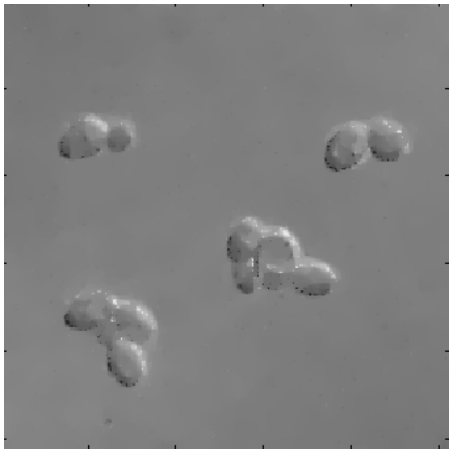
For visual inspection purposes, examples of fluorescence microscopy images reconstructed from Gaussian noise-contaminated measurements are shown in Fig. 4. It can be seen that the fluorescence microscopy images reconstructed using the proposed SGCFM approach captures significantly more important detail than that reconstructed using the traditional unguided sampling strategy. This is particularly noticeable within the individual cells, where weaker yet important structural details are noticeably better captured in the fluorescence microscopy images produced using the proposed SGCFM approach.

C. Experiment 2 - Noisy fluorescence microscopy reconstruction tests

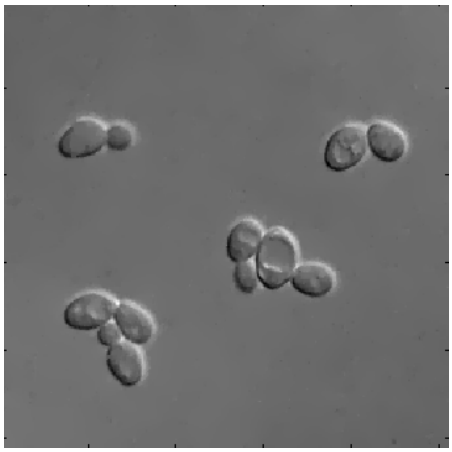
In the second set of experiments, a qualitative investigation is made into reconstruction performance for real-world situations where the fluorescence microscopy measurements that are heavily contaminated by real-world noise. As can be seen from Fig. 5, the proposed SGCFM approach noticeably outperforms the traditional sampling approach employed in existing CFM systems in terms of image quality, with significant improvements in structural definition in important regions of interests.

V. Conclusions and Future Work

In this paper, we propose the concept of saliency-guided compressive fluorescence microscopy for the purpose of improving acquisition speed and reconstruction quality in CFM systems. The saliency-guided CFM framework adaptively optimizes the sampling probability to the regions of interest rather than the common unguided random sampling used in existing CFM systems. The performance of the proposed SGCFM approach was demonstrated to provide improved fluorescence microscopy data acquisition speed and reconstruction quality in experiments with synthetic noise-contaminated measurements, as well as experiments

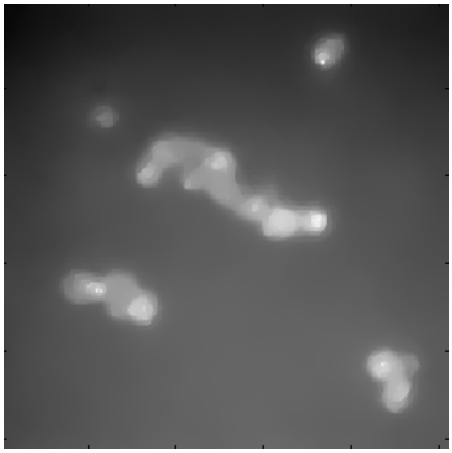


(a) Traditional unguided sampling reconstruction

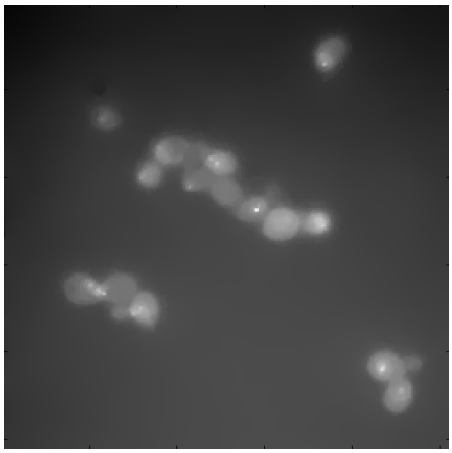


(b) SGCFM reconstruction

Fig. 4. Example reconstruction results for first set of experiments at 75% compression rate and 3% noise level



(a) Traditional unguided sampling reconstruction



(b) SGCFM reconstruction

Fig. 5. Example reconstruction results for second set of experiments at 80% compression rate from real noise-contaminated measurements.

with real noise-contaminated measurements. As the proposed SGCFM framework can act as the foundation for future research in CFM, it would be interesting to investigate the integration of different alternatives for optimizing the sampling probability density functions in the interest of further improving acquisition speed and image quality.

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