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Technical Note

Low-cost production method for fluorescent particles for liquid flow velocimetry applications

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Abstract

In this Technical Note, we present a simple and low-cost production method for fluorescent particles perfectly suitable for velocimetry applications. We leverage the unique ability of Nile Red (NR) fluorophores to adsorb on the polymer surface and/or embed itself in between polymeric chains. The laboratory procedure to dye polyamide particles with NR and monetary advantages over commercially available fluorescent particles is outlined. Subsequently, the fluorescence behavior of the dyed particles is tested under a laser illumination source in polar and non-polar liquids. The distinct advantage of the emission spectrum of NR-dyed particles is demonstrated with sample test results.

Keywords: Nile Red, PIV/PTV, LIF, seeding particles

1. Introduction

The implementation of velocimetry techniques, ranging from correlation-based methodologies (2D/Stereo/Tomo-PIV) to particle tracking velocimetry (PTV) methods, requires the use of micro-sized seeding particles as tracers to acquire instantaneous velocity flow fields. With the advancement in high-speed cameras and the availability of superior particle tracking algorithms, high particle image density measurements have become feasible [1]. Therefore, the number of particles required grows, which increases exponentially with the larger capacity of the flow system, for example, in hydrological research facilities; refer [2] for a detailed review.

While non-fluorescent particles are the most cost-effective option for very large-scale flow facilities, it comes at the cost of particle field images suffering from surface reflections, skewed reflected light intensity (Mie scattering in hollow glass particles), unwanted background light intensity, air bubbles, undesirable contaminants, etc. The use of fluorescent particles combined with appropriate optics eliminates these issues, resulting in high-intensity fluorescent particle images. On the other hand, in simultaneous PIV and laser-induced fluorescence (LIF) measurements, it is crucial to select fluorescent particles for velocimetry and fluorescent dyes for LIF with emission peak wavelengths that are significantly separated. This ensures that each tracer remains distinct and avoids cross-visibility in the captured images. The present work stems from the desire to fulfill the experimental constraint of avoiding

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unwanted sources of noise in the images for simultaneous PIV and LIF measurements in a large-scale water tunnel facility.

Given the steep costs of higher-grade monodisperse fluorescent particles, see table 1, it becomes imperative to seek a low-cost alternative specifically for large-scale flow experiments. In our recent work [3], a cost-effective alternative to the expensive commercially available fluorescent particles, was necessary to perform 3D-PTV measurements. To this end, we followed an inexpensive particle fabrication method outlined by [4] where a recipe of epoxy resin and Rhodamine-WT is prepared, subsequently cured, pulverized, and sieved to obtain particles within the desired diameter range. Although the method is cost-effective, it produces non-spherical particles. Furthermore, the absence of a costly sieve shaker machine made the sieving process tedious and susceptible to contamination from foreign substances due to the lack of a clean-room environment. This also resulted in a comparatively lower degree of particle monodispersity. In particular, the monodispersity and sphericity of particles is crucial for the successful implementation of 3D-PTV particle reconstruction algorithms [1]. The lack of such particle quality, to some extent, contributed to a relatively low number of reconstructed particle trajectories in our work [3], despite a higher particle image density. Furthermore, the particles produced have a relatively higher density ($\sim 1.2\,\mathrm{g\,cm^{-3}}$) making them unsuitable for slower flow regions, such as near-wall flows as observed in our recent work [3]. In these areas, the local Stokes number would exceed acceptable limits, thus providing inaccurate local velocity information.

Nile Red (NR) has been successfully used in the detection of various types of microplastics found as contaminants in environmental water bodies, refer to [5] for detailed review of research on the use of NR in staining microplastics. Therein, the NR's solvatochromic behavior, i.e. varying emission characteristics with the binding polymer that facilitates successful classification of microplastics are described.

The current research leverages NR's adsorption properties to dye cost-effective non-fluorescent spherical polymer particles. This approach drastically reduces costs to approximately $1 per gram, producing fluorescent seeding particles ideal for velocimetry applications. In addition, they also facilitate simultaneous LIF and PIV/PTV measurements with the use of appropriate passive scalar dye and optical filters. Additionally, we demonstrate the appeal of using NR as a dyeing agent for polymers leading to a large Stokes shift ($\sim$80 nm, refer to figure 1), thus facilitating simultaneous velocity (using PIV) and concentration (using LIF) measurements.

The following sections detail the selection criteria for polymer particles and the dyeing methodology. Additionally, we provide sample images capturing the successful application of the dyed seeding particles in both polar (water) and non-polar (PolyDiMethylSiloxane or, PDMS) media. Through this work, our goal is to encourage researchers, specifically in the field of fluid mechanics, to adopt this simple and cost-effective method to produce fluorescent particles for furthering their experimental research.

Table 1. Cost of fluorescent particles suitable for Green laser illumination for liquid flow velocimetry applications. PMMA: Polymethylmethacrylate, PS: Polystyrene, PE: Polyethylene, RhB: Rhodamine-B, NR: Nile Red.

Polymer (Size, Fluorophore)	Cost per gram[a]	Manufacturer
PMMA (1–20 μm, RhB)	$50	Dantec Dynamics
PMMA (1–20 μm, RhB)	$150	LaVision Inc.
PS (1–20 μm, RhB)	$450	LaVision Inc.
PE (10–20 μm, NS[b])	$4800	Cospheric LLC
PS (10–14 μm, NR)	$11 000	Spherotech Inc.

[a] Prices for year 2024.

[b] Not Specified. Varied fluorescent products available and their peak emission wavelength vary from Green to Red region.

2. Polymer selection and dyeing procedure

In pursuit of microplastics detection and classification, Sturm *et al* [7] stained various types of polymers (synthetic and natural) and assessed their emission characteristics in different solvents. Among the tested micro-plastics, copolyamide and copolyester particles dyed with NR showed the most intense emission characteristics in the far red region of the spectrum. The authors also observed that the hydrophobic polymers, such as polyethylene and polypropylene dyed with NR showed significantly lower, $\lesssim 75\%$ emission intensity compared to hydrophilic polymers, such as copolyamide and copolyester. Unexpectedly, with further increase in polarity, for instance for polyvinylchloride, the intensity drops down significantly. As stated in [7], the adsorption of NR on the polymer surface and its embedding into the polymer network restrict molecular movement, leading to a significant variation in the emission spectrum. It seems that an ideal balance of surface adsorption and embedding of the NR molecule in the polymeric chain is achieved for copolyamide and copolyester, resulting in intense emission characteristics aiding the objectives of the present work.

Additionally, the hydrophilic polymer particles showed increasingly higher peak emission intensity with increasing polarity of the solvent. On the contrary, NR dye by itself shows decreasing absolute emission peak intensity with polarity of the solvent, refer to figure 1 in [9]. Regardless of whether NR dyed particles are suspended in a solvent or NR itself mixed in a solvent, it shows increasingly larger red-shift in emission spectrum with increasing polarity of the solvents. This observation is favorable to the objectives of the present work, and further discussion is provided in the next section. In addition, the emission spectrum for NR-dyed copolyamide suspended in neutral water is shown in figure 1, (refer to the red curve, with a peak intensity occurring around 630 nm [7]).

Upon careful examination of the above observations and cost analysis of candidate non-fluorescent particles, highly spherical polyamide (PA) particles of narrow size distribution with mean diameter of 20 μm (<10 μm, max 5% and >30 μm, max 5%) and density $1.03\,\mathrm{g\,cm^{-3}}$ supplied by LaVision Inc.

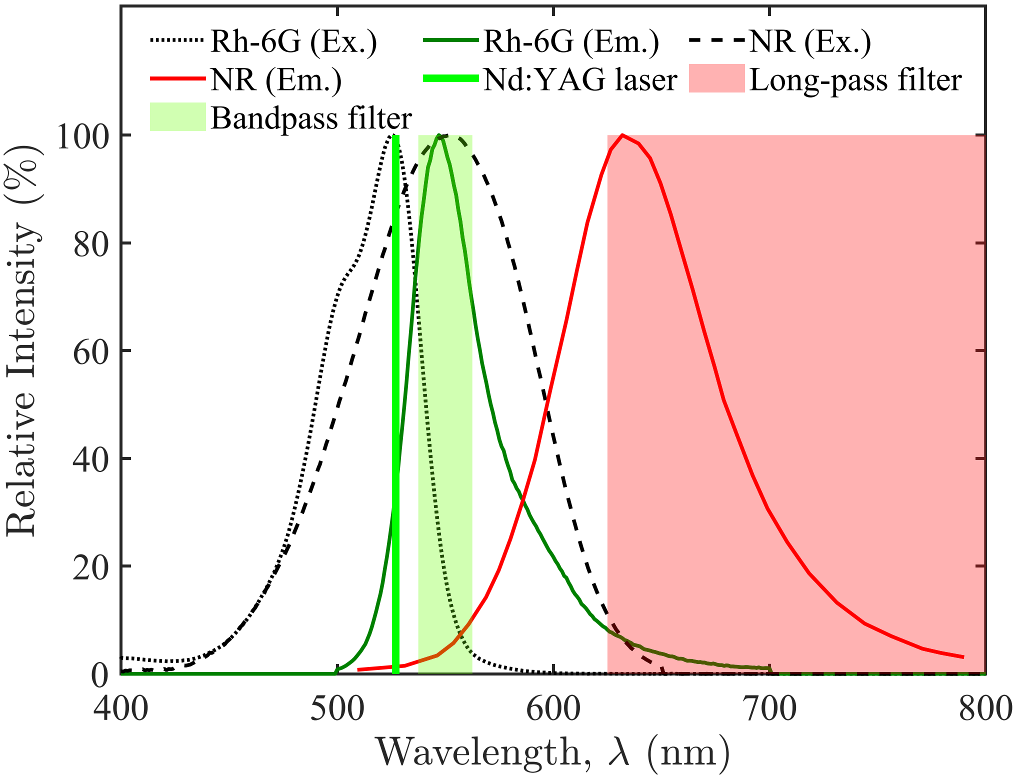

Figure 1. Excitation spectrum for NR (dashed line) is adapted from [6] and emission spectrum (solid red line) for NR adsorbed on copolyamide pieces in water is adapted from [7]. Excitation (dotted line) and emission (solid green line) spectra for Rhodamine-6G in water is adapted from figure 3 of [8]. Thick vertical green line corresponds to Nd: YLF laser source of wavelength ($\lambda \sim 527$ nm). Narrow green band: Bandpass filter range, 550 ± 12.5 nm. Wide red band: Long-pass filter, >625 nm.

(Article No.: 1108892, Cost: $\sim$44 ¢ g^{-1}) is procured. The particles are in white powder form and stable in liquids.

The laboratory dyeing method is similar to that adopted by [7] to dye various polymers to facilitate the detection of microplastics. A stock solution of concentration, 1 mg ml^{-1}, of Nile Red (CAS: 7385-67-3, Purity >98%, TCI Co., Ltd.) in acetone is prepared. Acetone is selected as a solvent because of its less detrimental effect on polymers. In addition, 1 g of PA particles is mechanically mixed in 150 ml of deionized (DI) water using a magnetic stirrer. Subsequently, 0.75 ml of the NR-acetone stock solution is added, resulting in a final NR concentration of $\sim$5 mg l^{-1}. The suspension is continued to be mechanically mixed for approximately 24 h at room temperature for adequate dyeing of the particles.

It is expected that most of the NR is adsorbed or embedded in the polymer, and negligible amount of NR remains in the solution because of its marginal solubility in water. Therefore, for a large-scale water tunnel flow system, the suspension can be mixed directly in the facility for velocimetry applications. However, the acquired flow images could be slightly affected by the marginal solubility of NR in water, thus producing a background fluorescent signal. To completely eliminate this possibility, the suspension is filtered through a quantitative filter paper (Whatman®, Grade 3, pore size: 6 µm), dried and desiccated with silica gel for 24 h, and suspensions of low particle concentration are prepared with fresh DI water and a non-polar solvent, PDMS oil (Gelest, Inc.) of viscosity 5 cSt and density 0.918 g cm^{-3} for final tests.

3. Test results

The test results shown in figure 2 are captured at an image repetition rate of 100 Hz using a high-speed Phantom camera (1280×800 pixels, pixel size: 28 µm, model v2512, Ametek Inc.) equipped with a 200 mm focal length lens set to an aperture of $f/8$. The flow region is illuminated with a $\sim$1 mm thin light sheet generated by manipulation of the Nd: YLF laser beam (wavelength $\sim$527 nm, Photonics Industries) with a combination of cylindrical lenses. To highlight potential sources of image contamination, background noise and undesirable pollutants are simulated by suspending non-fluorescent PA particles in water until the suspension exhibits slight turbidity under natural light. In addition, an acrylic piece was placed in the path of the light sheet to mimic the presence of a reflective surface. Figure 2(a) captures the mix of fluorescent and non-fluorescent particles in water (a polar medium) without using an optical filter at an estimated average laser power of 0.76 W. The bright vertical streak corresponds to the laser light reflecting off the acrylic piece. A few irregularly shaped contaminants can be observed in the captured image. There is also a notable background intensity

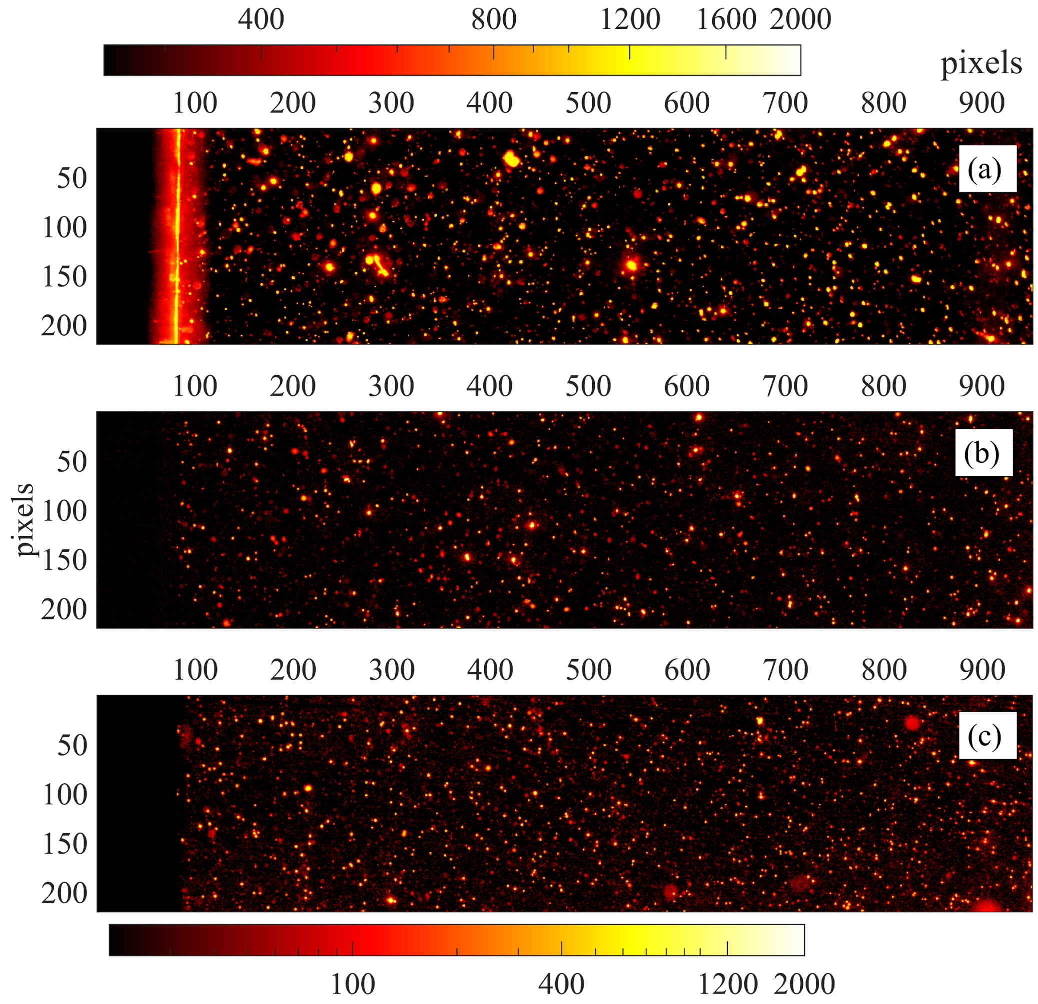

Figure 2. Raw image of NR-dyed and non-fluorescent PA particles suspended in water captured (a) without an optical filter, and (b) with the long-pass filter (wavelength cutoff: 625 nm). (c) NR-dyed PA particles suspended within PDMS oil. Note the bright vertical streak in (a) is surface reflection that is completely eliminated in (b) and (c). Top colorbar is for (a). Bottom colorbar is for both (b) and (c).

and halo effect around the bright particles. Nevertheless, the use of a long-pass optical filter with cutoff wavelength 625 nm (Edmund Optics Inc.) eliminates these sources of image contamination, as seen in figure 2(b). Note that an estimated average laser power of 2.8 W is used to obtain figures 2(b) and (c), approximately 3.7 times the power used for figure 2(a), corresponding to a 40% increase in laser diode current. This adjustment compensates for the light intensity attenuated by the optical filters, ensuring brighter particle images.

Furthermore, NR-dyed PA particles are tested in PolyDiMethylSiloxane (PDMS) oils (excluding the aforementioned image noise sources except for the reflective surface) to evaluate their suitability in non-polar medium (figure 2(c)). The particle intensity captured in these two media, illuminated at the same laser-diode current, showed insignificant variation demonstrating the effectiveness of NR-dyed PA particles for velocimetry applications in both polar and non-polar solvents. Additionally, since NR is soluble in PDMS, it indicates that the bonding of the NR molecules with the polymer surface is strong enough to preserve the emission characteristics and to prevent significant immediate diffusion into the PDMS oil. However, a mild background intensity, accounting for less than 5% of the peak intensity, is observed for this case. To address this, the background intensity value is subtracted from the raw image to produce figure 2(c). The background intensity may be attributed to slight diffusion of NR into the PDMS bulk. Further investigation into the diffusion of NR from the polymer surface into the PDMS over prolonged time would be necessary to further evaluate the performance of particles in PDMS; however, this is beyond the scope of the current work.

As discussed previously, the motivation to develop NR-dyed particles stemmed from the experimental necessity to simultaneously acquire velocity and concentration fields. To this end, a two-camera imaging system with optical filters (as specified in figure 1) meant for simultaneous PIV and LIF measurements is utilized to test the NR-dyed PA particles at the same laser power used to produce figures 2(b) and (c). Their peak emission in the far-red region complements the use of water-soluble Rhodamine-6G as a tracer to aid

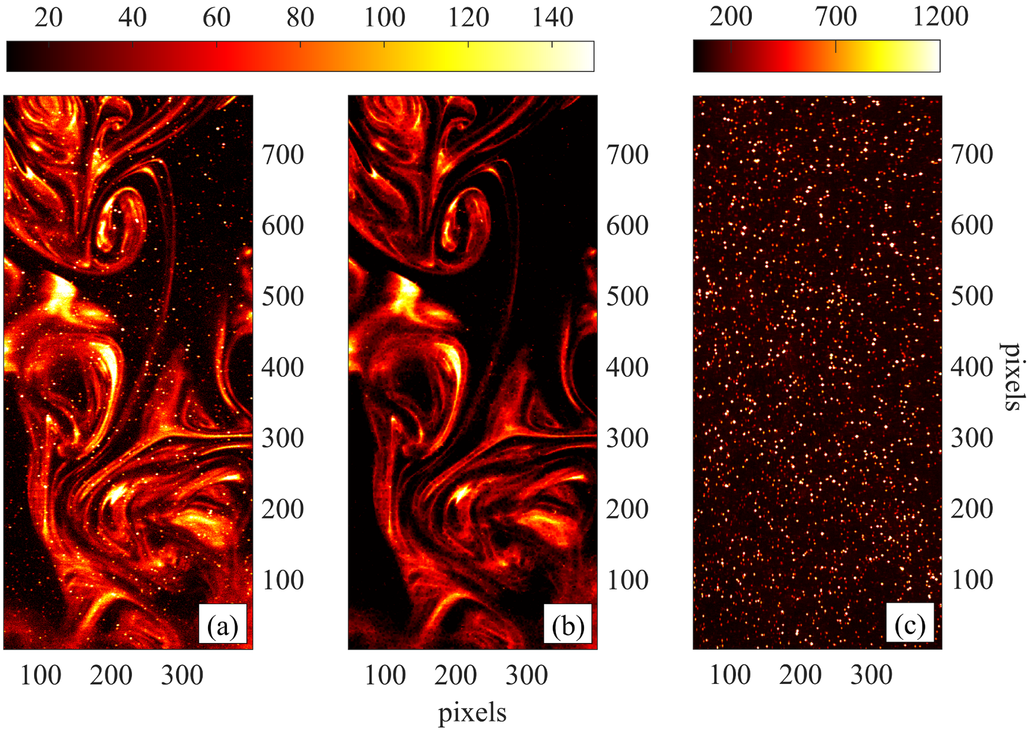

Figure 3. (a) Raw image captured using 550 nm band-pass filter. Particle images are visible due to leakage from the NR emission spectrum. (b) Post-processed image of (a) filtering out NR-leaked particles with erosion filter. (c) NR-dyed PA particles in neutral pH water captured using 625 nm long-pass filter in same flow region as (a). This optical filter attenuates the Rh-6G fluorescence enough to make it effectively undetectable and unnecessary to post-process.

simultaneous velocity and concentration field measurements (refer to figure 3). Figure 3(a) shows that the spatial intensity variation is directly proportional to the local concentration of Rhodamine-6G, thus providing a concentration variation field. In addition, it also shows weak-intensity NR particle images ($<10\%$ of peak image intensity), given a slight overlap of NR emission spectra and bandpass filter range (550 ± 12.5 nm, Edmund Optics Inc.) (refer to figure 1). The visibility may also have been enhanced due to the overlap between the Rh-6G emission and NR absorption spectra, inducing secondary fluorescence in the NR particles, thus enhancing its intensity.

Nonetheless, the spatial characteristic frequency of particle images is significantly higher than that of the LIF signal, which makes it possible to digitally filter out the particles without affecting the LIF noticeably. As shown in figure 3(b), particle images were eliminated during post-processing with an erosion filter (implemented through DaVis 10.2, LaVision Inc.). This is a non-linear digital filter similar to a median filter, but assigns intensity corresponding to the element before the median in an intensity-sorted array out of a 3 pixel × 3 pixel window. Figure 3(c) displays a raw image of particles captured through the long-pass filter (cutoff wavelength 625 nm). There is negligible cross-contamination of Rh-6G fluorescence. Hence, no post-processing is required on this image to eliminate the LIF signal. Particle images like in figure 3(c) are appropriate to educe the velocity field.

4. Summary

We present a simple procedure to produce high-quality fluorescent seeding particles using NR and PA particles at a significantly low cost, making them suitable for liquid flow velocimetry applications. The production cost is under $1 per gram, ideal for large-scale facilities prohibited by high costs associated with seeding particles. The resulting particles exhibit consistent particle image intensities in both water and PolyDiMethylSiloxane oils, demonstrating their suitability in both polar and non-polar liquids. Additionally, the large Stokes shift of the NR-dyed PA particles allows for their use with any fluorescent tracer with peak emission in the green region, enabling simultaneous capture of velocity and concentration fields in a PIV/PTV and LIF experimental setup. Testing in such a PIV-LIF setup revealed minimal cross-visibility between the passive scalar dye (Rhodamine-6G) and the NR-dyed PA particles, highlighting their excellent compatibility for simultaneous velocity and concentration field measurements.

Data availability statement

The data cannot be made publicly available upon publication because they are not available in a format that is sufficiently accessible or reusable by other researchers. The data that support the findings of this study are available upon reasonable request from the authors.

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Conflict of interest

The authors do not have any financial or personal interests regarding the results of this work.

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