

VISIBILITY ENHANCEMENT OF FLUORESCENT SUBSTANCE UNDER AMBIENT ILLUMINATION USING FLASH PHOTOGRAPHY

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ABSTRACT

Many natural and manmade objects contain fluorescent substance. To visualize the distribution of fluorescence emitting substance is of great importance for food freshness examination, molecular dynamics analysis and so on. Unfortunately, the presence of fluorescent substance is usually imperceptible under strong ambient illumination, since fluorescent emission is relatively weak compared with surface reflectance. Even assuming that surface reflectance could be somehow blocked out, shading effect on fluorescent emission that relates to surface geometry would still interfere with visibility of fluorescent substance in the scene. In this paper, we propose a visibility enhancement method to better visualize the distribution of fluorescent substance under unknown and uncontrolled ambient illumination. By using an image pair captured with UV and visible flash illumination, we obtain a shading-free luminance image that visualizes the distribution of fluorescent emission. We further replace the luminance of the RGB image under ambient illumination by using this fluorescent emission luminance, so as to obtain a full colored image. The effectiveness of our method has been verified when used to visualize weak fluorescence from bacteria on rotting cheese and meat.

Index Terms— Fluorescence, Visibility, Flash

1. INTRODUCTION

A lot of natural and manmade objects in daily life contain some fluorescent substance. The interaction between incident light and fluorescent substance is complicated, since, in addition to the usual reflectance, it will first absorb shorter wavelength light in the absorption spectral range, and emit longer wavelength light in the emission spectral range.

To visualize fluorescent emission (and thus the existence and distribution of fluorescent substance) is of great importance for many tasks, such as food freshness examination, molecular dynamics analysis labelled by fluorescent dyes and note anti-counterfeiting. For instance, bacteria on rotting cheese will emit fluorescence, and the abundance distribution of such fluorescence will be an intuitive index of cheese freshness, see Fig.1. Unfortunately, the mixture of reflectance and

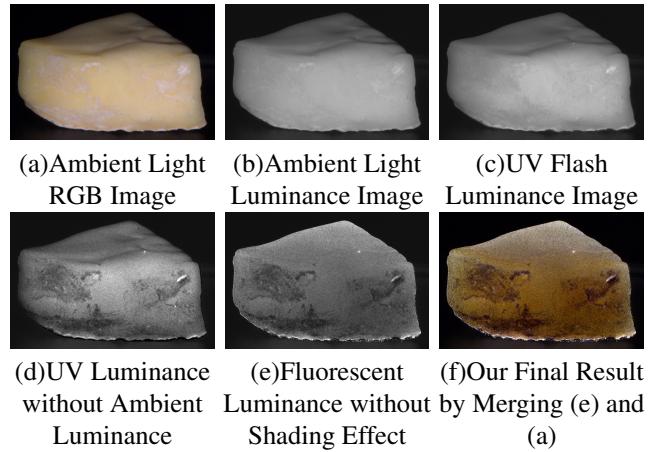


Fig. 1. Visibility enhancement of weak fluorescent substance on cheese. Note that the emitted fluorescence from bacteria is not noticeable in the RGB image under ambient light (a).

emission makes it difficult to visualize fluorescent emission only, which becomes especially true when the fluorescent emission is very weak compared with reflectance.

Under controlled environment where the reflectance can be avoided by blocking incident visible light, to visualize fluorescent emission becomes easier. This is the case to visualize fluorescent watermarks on flat currency note under UV light only in a dark space. However, to block out visible ambient light is not always feasible (or convenient), especially for food freshness examination on a portable device. In addition, the shading effect on fluorescent emission might cause incorrect abundance distribution estimate of fluorescent substances, since the incident irradiance is dependent on the surface geometry.

In this paper, we focus on visualization of fluorescence emitting substances on solid food surfaces, which will be excited mainly under UV light. We propose a novel visibility enhancement method by using flash lamps under unknown and uncontrolled ambient illumination, whose workflow is shown in Fig.2. We equip an eye-safe UV flash lamp and a visible flash lamp with a standard RGB camera, which are very close to each other such that their light rays are almost

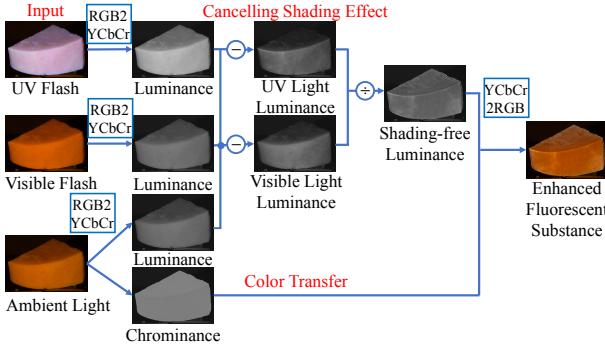


Fig. 2. The flowchart of our visibility enhancement method.

codirectional. An image pair under these two flash lamps are captured separately, from which a shading-free luminance image is obtained that visualizes the distribution of fluorescent emission. To retain the texture information and get a colored image, we further replace the luminance of the RGB image under ambient illumination by using this fluorescent emission luminance. The effectiveness of our method has been verified by using it to visualize weak fluorescence from bacteria on rotting cheese and meat.

2. RELATED WORKS

Fluorescent emission visualization has proven to be important in many research and application fields. For instance, the emitted fluorescence by chlorophyll in leafs can be used to examine the health condition of plants [1, 2], and molecular interaction can be directly seen by visualizing fluorescent markers added into molecules [3]. Usually, one has to use UV light to excite fluorescence and observe emission at a certain wavelength range through bandpass filters. This operation is usually conducted in dark room, so as to avoid the influence of reflectance of visible ambient light. Also, the shading effect on fluorescent emission relating to solid surface geometry has not been addressed.

In color computer vision, various computational imaging methods [4, 5, 6] have been proposed to separate fluorescent emission from reflectance. Although they can be applied to objects under ambient illumination in principle, their capability to visualize weak fluorescence is under question, and the shading effect remains in the separated results.

Flash photography has been widely used for image denoising by fusing a flash/non-flash pair. For example, Petschnigg et al. [7] used the bilateral filters for the flash image with details, and combined it with the image under the ambient light. Also, Krishnan et al. [8] suggested dazzle-free photography by using UV and IR flash in the non-visible spectral range. In contrast to these works, our major focus is on visibility enhancement of weak fluorescence by using flash illumination.

Fredembach et al. [9, 10] synthesized color images by

changing the color space and swap the brightness channel with IR intensity. Inspired by this method, we also generate full colored images by substituting the fluorescent emission luminance image into the luminance of the RGB image taken under ambient illumination.

3. COLOR IMAGING OF FLUORESCENCE

In this section, we introduce the color imaging model of a fluorescent surface, under the diffuse reflectance and fluorescent emission model [5, 4]. The appearance of a reflective and fluorescent surface point can be expressed as

$$P(\lambda_o, \lambda_i) = P_R(\lambda_o, \lambda_i) + P_F(\lambda_o, \lambda_i), \quad (1)$$

where $P_R(\lambda_o, \lambda_i)$ and $P_F(\lambda_o, \lambda_i)$ denote the reflective and fluorescent components. Specifically, the reflectance component resulting from surface and illumination interaction reads

$$P_R(\lambda_o, \lambda_i) = R(\lambda_o)L(\lambda_i)\delta(\lambda_o, \lambda_i), \quad (2)$$

where $R(\lambda_o)$, $L(\lambda_i)$ and $\delta(\cdot)$ are the reflectance at wavelength λ_o , the illuminance at wavelength λ_i and the unit impulse function where $\delta(0) = 1$ and $\delta(x) = 0, x \neq 0$.

The imaging model of fluorescence is more complex because of the wavelength shift effect, which can be described by the fluorescent excitation and emission process

$$P_F(\lambda_o, \lambda_i) = E_m(\lambda_o)E_x(\lambda_i)L(\lambda_i), \quad (3)$$

where $E_m(\lambda_o)$, $E_x(\lambda_i)$ and $L(\lambda_i)$ are the emission at wavelength λ_o , excitation at wavelength λ_i and illuminance at wavelength λ_i .

So far, we described the reflectance and fluorescence under narrow band illumination. When we consider under wide band illumination, the appearance at wavelength λ_o can be expressed as

$$\begin{aligned} P(\lambda_o) &= \int P(\lambda_o, \lambda_i)d\lambda_i \\ &= R(\lambda_o)L(\lambda_o) + E_m(\lambda_o) \int E_x(\lambda_i)L(\lambda_i)d\lambda_i, \end{aligned} \quad (4)$$

where $\int E_x(\lambda_i)L(\lambda_i)d\lambda_i$ is a constant under a given illumination. Note that the fluorescent emission spectrum is only scaled by the amount of the energy of the light, and the observed spectrum would have the same distribution over all wavelengths regardless of the spectrum of the illuminant, which is known as the color invariance of fluorescence.

A lot of objects in daily life, especially green plants and food, contain fluorescent substance, such as chlorophyll, fluorescent protein and bacteria [11, 12, 13]. The intensity and distribution of fluorescent emission can reflect the abundance of fluorescence substance, which can be used as an indicator of plant health condition and food freshness [14]. This motivates us to develop a visibility enhancement method for fluorescent substance by using flash photography, as will be detailed in the following section.

4. VISIBILITY ENHANCEMENT METHOD

We propose to use one UV flash lamp and one visible flash lamp to better visualize weak fluorescence on solid surfaces, by computationally cancelling out the unknown ambient illumination and the shading effect on fluorescent emission. We assume that these two lamps are placed close to each other, and their light rays are directional. The fluorescent substance in plants and food tends to absorb UV light, and emit visible light. For example, pseudomonas fluorescent breeding on the surface of food has a peak excitation at 384nm~398nm [15], while the absorption range of chlorophyll is between 360nm to 500nm. In this paper, we use a 365nm UV flash lamp, so as to make sure that the reflected UV light does not enter the RGB camera, whose spectral response range is usually between 400nm and 700nm. Meanwhile, to suppress fluorescent emission under the visible flash, we choose a 595nm flash lamp, whose spectral range goes beyond the absorption range of the fluorescent substance concerned in this paper.

According to Eqn.4, the intensity value $I_a(p)$ caused by the ambient light in a certain color channel of the p -th pixel can be expressed as

$$I_a(p) = \int L_a(\lambda)R(\lambda, p)C(\lambda)d\lambda + \int E_x(\lambda_i)L_a(\lambda_i)d\lambda_i \int E_m(\lambda_o)C(\lambda_o)d\lambda_o, \quad (5)$$

where $L_a(\lambda)$ and $C(\lambda)$ are the ambient light and the camera response function.

When the UV flash lamp is on, the intensity value $I_{UV}(p)$ caused by the UV flash and the ambient light of the p -th pixel can be expressed as

$$I_{UV}(p) = I_a(p) + \int E_x(\lambda_i, p)L_{UV}(\lambda_i)\cos\phi(p)d\lambda_i + \int E_m(\lambda_o, p)C(\lambda_o)d\lambda_o = I_a(p) + \cos\phi(p)F_{UV}(p), \quad (6)$$

where $L_{UV}(\lambda)$ is the illumination spectrum of the UV flash. $\cos\phi$ describes the cosine law irradiance change because of the angular difference between the surface normal and the UV light ray, which actually gives rise to the shading effect on fluorescent emission $F_{UV}(p)$.

To eliminate the shading effect $\cos\phi(p)$, a second image under the visible flash and the ambient light is taken, whose intensity value $I_{vis}(p)$ at the p -th pixel is expressed as

$$I_{vis}(p) = I_a(p) + \int L_{vis}(\lambda_v)R(\lambda_v, p)\cos\phi(p)C(\lambda_v)d\lambda_v = I_a(p) + \cos\phi(p)R_{vis}(p), \quad (7)$$

where $L_{vis}(\lambda)$ is the visible flash light spectrum. For targets concerned in this paper, like plants and food, the reflectance

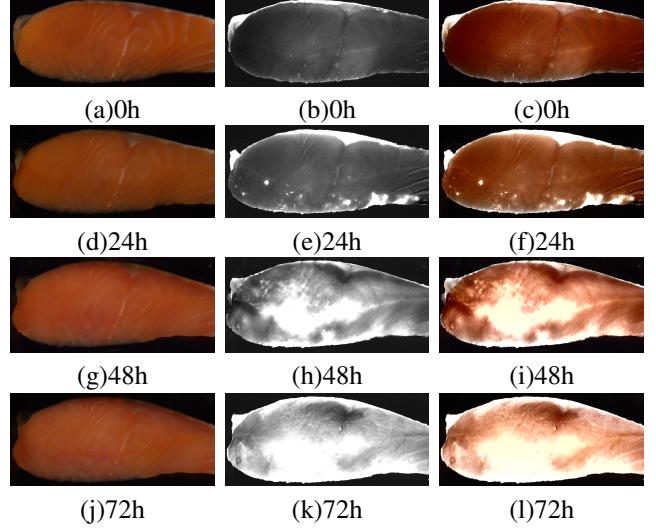


Fig. 3. Visibility enhancement of fluorescence emitting bacteria on rotting salmon across time up to 72 hours. For each row, the original RGB images under ambient light (left), the fluorescent emission luminance (middle) and the final colored image (right) are shown. Note that the fluorescence emission is almost invisible in the RGB images under ambient illumination.

material on the target surface is usually uniform. Therefore, Eqn.7 can be approximated into

$$I_{vis}(p) = I_a(p) + \cos\phi(p)R_{vis}. \quad (8)$$

From Eqns.5, 6, 8, the visualization of fluorescent emission can be achieved by

$$\frac{I_{UV}(p) - I_a(p)}{I_{vis}(p) - I_a(p)} = kF_{UV}(p), \quad (9)$$

where k is a constant value. Note that the shading effect on fluorescent emission has been eliminated.

In our implementation, we transform the original RGB images into the YCbCr color space, and calculate the fluorescent emission distribution image on the basis of the luminance channel. As mentioned in [9], this YCbCr color space is better than other color spaces like HSV, because the YCbCr color space separates luminance and chrominance, and it does not necessarily preserve saturation for our purpose.

Without enough color and texture information, it is difficult to figure out where the fluorescent substance lies. Inspired by [9, 10], we replace the Y channel of the color image under the ambient light by the calculated fluorescent emission distribution, so as to obtain a full colored image. The flowchart of our proposed method is shown in Fig.2.

5. EXPERIMENT RESULTS

In this section, we show how the distribution of fluorescent substance can be easily seen by using our visibility enhance-

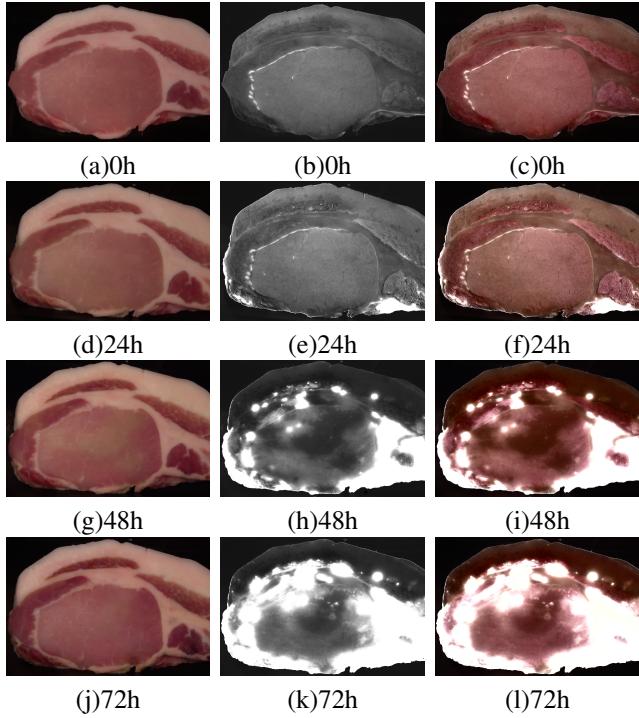


Fig. 4. Visibility enhancement of fluorescence emitting bacteria on rotting pork across time up to 72 hours. The subimages are aligned in the same way as in Fig.3.

ment method. We use a Nikon D4s camera to capture RGB images. We take all images in RAW format and then convert them into 16-bit TIFF images. By default, the Nikon conversion software performs white balancing and gamma correction as nonlinear operations. We thus turn off white balancing and gamma correction to avoid nonlinear transformation on the image intensity. All images are captured under bright ambient light from an incandescent lamp.

We first examine the distribution of fluorescent substance on a slice of rotten cheese, as shown in Fig.1. According to the RGB image (a) and the luminance image (b) under ambient light, the fluorescence emitted from bacteria at rotten spots is not noticeable at all. By cancelling out the ambient light, the fluorescence becomes visible (d), yet the shading effect remains, especially on the surface away from the camera viewing direction. The fluorescence emission luminance shown in (e) clearly reveals the abundance of fluorescence, without the shading effect arising from surface geometry. By merging the fluorescence luminance and the chrominance from the RGB image under ambient light, the ultimate visibility enhanced result with color in (f) looks visually satisfactory. To show food rotting process across time, we also use our visibility enhancement method to visualize the fluorescence emitting bacteria on salmon (Fig.3), pork (Fig.4) and tuna (Fig.5). We expose the meat samples at room temperature, and capture images every 24 hours. We can see that the abundance of flu-

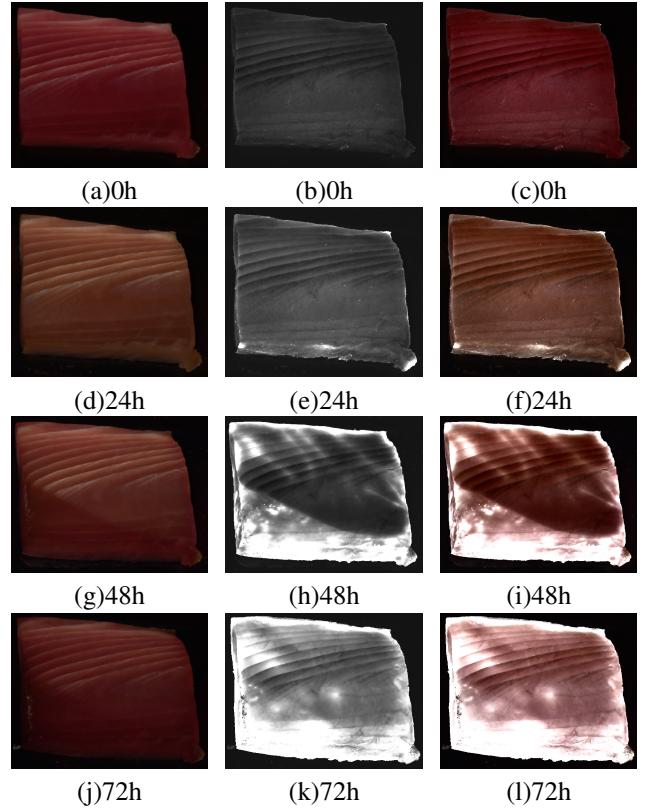


Fig. 5. Visibility enhancement of fluorescence emitting bacteria on rotting tuna across time up to 72 hours. The subimages are aligned in the same way as in Fig.3.

rescent emission grow gradually as time elapses. This reveals that the amount of fluorescence emitting bacteria increases as the meat goes bad. It is worthy to note that this rotting process is not so obvious by observing the RGB images under ambient illumination.

6. CONCLUSIONS

We have presented a visibility enhancement method to better visualize weak fluorescence emitting substance on a solid surface with unknown geometry and uncontrolled ambient light. By introducing one UV flash and one visible flash placed close to each other, our method is able to eliminate the effect of ambient illumination and shading effect on fluorescent emission. We have verified the effectiveness of our proposed method by using it to visualize weak fluorescence from bacteria on rotting cheese and meat.

7. ACKNOWLEDGEMENT

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