

2 **Study of a pH-sensitive Hologram for Biosensing Applications**3 **KOMAL SHARMA**^{1,2}, **HEENA**¹, **GIRISH C. MOHANTA**^{1,2}, **RAJ KUMAR**^{1,2,*}

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8
9 **Abstract:** Photopolymers are widely utilized as holographic recording media due to their ease of
10 preparation and no wet chemistry post-processing. Holographic sensors constructed from pH-
11 sensitive photopolymer film have several applications in biosensors and medical diagnostic field.
12 However, the stability of photopolymer films in an aqueous medium is one of the important
13 challenges in their application for biosensing. Furthermore, pH of the solution is another important
14 parameter for biochemical reactions. In this work, we have compared the pH sensitivity and stability
15 of our holographic grating against two widely utilized classes of buffers; Phosphate Buffered Saline
16 (PBS) and Tris-Acetate-EDTA (TAE) at two different pH values of 7.36 and pH 8.3, respectively. It
17 was observed that physiological pH (pH 7.4) had a negligible effect on the diffraction efficiency of
18 the holographic sensor while it significantly deteriorated at a higher value of ~ pH 8.3. This high
19 sensitivity towards the minute pH difference of our holographic sensor could potentially be
20 exploited for pH-based biosensing applications such as urea detection.

21
22 **Keywords:** Holographic Sensors; Photopolymer Film; Biosensors, pH-Sensing

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25 **1. Introduction**

26 The quantitative and qualitative measurement of different physical and chemical
27 processes is very important in the field of medical, agriculture, industry and
28 environmental applications. Holography is one of the optical techniques in which one
29 can observe the physical and chemical change in the material. It is a method for storing
30 and retrieving object information using light diffraction and interference. This technique
31 uses an object beam and reference beam that are both captured on the photopolymer film
32 to produce an interference pattern. This technique has advantage of no use of additional
33 re-chargeable power unit, versatility, robustness and ease of preparation. These benefits
34 led to its use in the field of bio sensing applications like glucose, lactose, pH detection
35 and drug detection, among other things [1]. In order to achieve the necessary results with
36 accuracy, selectivity, and sensing capacity, the photopolymer films must be prepared
37 with all point of care mainly in biosensing applications.

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Recently, it is reported that the polyvinyl alcohol (PVA) based photopolymer due to its non-toxicity, swelling properties and good adhesive properties are used widely[2]. For biosensing applications like drug detection, pH sensors, this binder is not desirable due to its hygroscopic nature. Also, cellulose acetate based photopolymer are biocompatible, sustainable, cheap, flexible, lightweight and biodegradable [3] but have limitation in film formation due to volatile nature of the solvent used.

In this paper, we propose to analyse the effect of pH change on the diffraction efficiency of transmission gratings recorded in commercially available photopolymer as holographic recording material by dipping it in the buffer solutions for different time intervals. The diffraction efficiency of the recorded grating can be measured as;

$$D.E. (\%) = \frac{I_{D+1}}{I_I} \times 100 \text{ -----(1)}$$

Where, I_{D+1} and I_I are the intensity of first order diffracted beam and incident beam respectively.

2. Materials and methods

For biosensing applications, sensitivity of the material towards pH plays a crucial role. In order to study its effect change in diffraction efficiency of recorded holograms is calculated at different pH exposure.

2.1. Recording Process

Transmission gratings are recorded in a two beam holographic optical setup (Figure 1) using a continuous wave DPSS laser (532 nm). The beam is allowed to pass through a spatial filter (S.F.) followed by lens (L) to obtain collimated beam which is further divided into two by beam splitter (B.S.). These two beams are directed by the mirrors M1 and M2 on the recording plate (photopolymer film, LLPF465, Light Logics). This film requires the dosage of 12 mJ/cm² at 532nm wavelength. The angle between the interfering beams is 25°. The intensity of these beams are adjusted to equal values by using a neutral density filter. A shutter is used to control the exposure from the laser and the total recording intensity and exposure time are 12.8 mJ/cm² and 740 ms, respectively. After the recording process, gratings are characterized by diffraction efficiency measurement (Figure 2) of the first diffracted order using an optical power monitor.

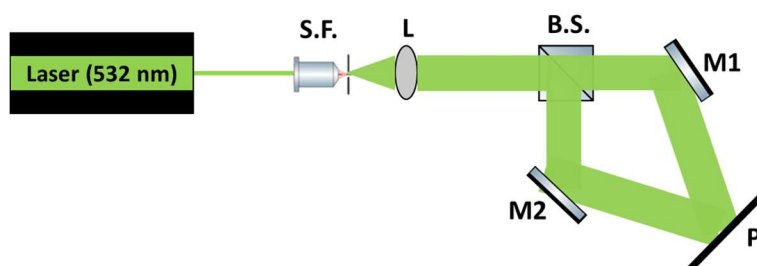


Figure 1. Schematic of experimental setup for recording of transmission gratings

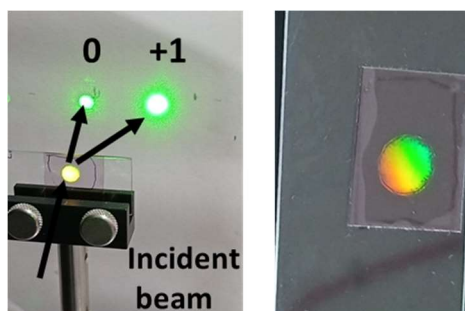


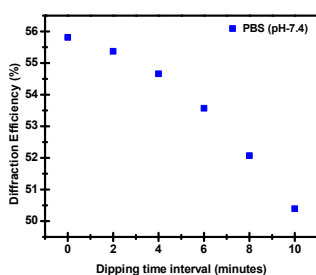
Figure 2. Photograph of diffracted orders and recorded grating

2.2. pH sensitivity measurement

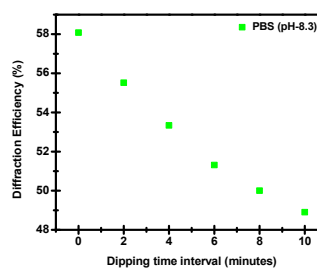
In order to study the effect of pH sensitivity, three widely used buffer solutions are prepared having different pH values namely Tris-Acetate-EDTA (pH-8.3) and Phosphate Buffered Saline (pH- 7.4 and 8.3). The motive behind using PBS at two different pH levels is to examine the variation in diffraction efficiency of hologram with varying pH of same solution without relying on the pKa value. pKa value is used to measure the acidity of a particular molecule in the solvent. Experiments are performed to see the effect on the photopolymer film by varying pH when these gratings are dipped into different solutions for different time intervals and subsequent change in diffraction efficiency is measured. The total volume of solution is 30 mL for complete immersion of glass slides.

3. Results and Discussion

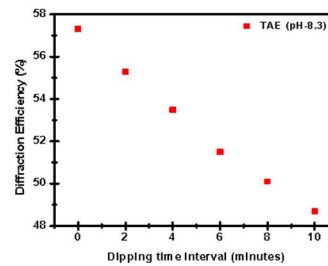
The recorded gratings exhibit diffraction efficiency > 55% before any exposure to pH solutions. Once these are allowed to completely dip into different pH solutions, the variation in the diffraction efficiency values is observed. The observed change is different for different pH values. For neutral pH i.e., 7.4, the change in diffraction efficiency is slight within 5 % of original values (Figure 3 (a)). On the other hand, when the pH is increased to 8.3, the deterioration is much more for both PBS (Figure 3 (b)) and TAE (Figure 3 (c)) solutions, the percentage change is up to 10 % for the given dipping time intervals. With this we can infer that the recorded holograms show sensitivity towards alkaline pH and can be used for biosensing applications.



(a)



(b)



(c)

Figure 3. The diffraction efficiency response with dipping time intervals at different pH : **a)** PBS pH -7.4; **(b)** PBS pH -8.3; **c)** TAE pH -8.3.

4. Conclusion

In this work, we have recorded holographic transmission gratings in photopolymer recording material and compared the pH sensitivity and stability of our holographic grating against two widely utilized classes of buffers; Phosphate Buffered Saline (PBS) and Tris-Acetate-EDTA (TAE) at two different pH values of 7.36 and pH 8.3, respectively. The sensitivity of the recorded hologram changed with pH, and by increasing its value from 7.4 to 8.3; the variance in diffraction efficiency decreased substantially. This study will be helpful in further development of holographic sensors for bio-sensing industry, including glucose sensors, urea sensors, lactose sensors, and drug detection sensors.

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