Woolitmus-A Pyranine Embedded Wool Fabric as a Wearable pH Indicator and Sweat Analyzer

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This study introduces "*Woolitmus*" a textile-based wearable pH indicator and sweat analyzer developed using pyranine on wool substrate. Sweat pH analysis is crucial for monitoring health conditions associated with pH imbalances. This capitalizes on pyranine's unique acid dye properties, successfully dyeing on wool fabric to create a comprehensive pH sensor. The sensor exhibits pH responsiveness under both visible and UV light, offering potential as a real-time sweat patch for pH monitoring. The process of dyeing of wool with pyranine is detailed, elucidating the mechanism behind the pH sensitivity backed up by photophysical characterizations. Stability and reversibility tests also confirm the sensor's robustness and performance. The wool fabric-based pH sensor holds promising applications, including health monitoring, disease diagnosis, and lifestyle management. The reported sensor also can simultaneously collect and detect pH levels without support of any addition accessories.

1. Introduction

Sweat pH analysis is essential for detecting health issues related to temperature control and pH balance disruption. Normal sweat pH ranges from 4.5 to 7, but deviations indicate shifts toward acidity or alkalinity. Conditions like cystic fibrosis are evident through sweat's pH, which can approach to 9 due to bicarbonate imbalance. Monitoring sweat pH changes can provide early alerts for such concerns.^{1–3} Sweat pH also helps spot infections in wounds. Altered sweat pH (7 to 8.5) caused by bacteria and enzymes signals infected wounds, aiding in tracking the body's response. Moreover, sweat pH variations impact wound healing assessment. Changes during healing stages offer insights into treatment effectiveness, non-invasively gauging the body's reaction.^{4,5} Dermatological health benefits from sweat pH analysis too. Skin issues like irritant

contact dermatitis, atopic dermatitis, and acne correlate with sweat pH. Analyzing sweat composition aids diagnosis and monitoring, offering a non-invasive tool for dermatologists.⁶The connection between sweat pH and glucose levels offers further benefits.⁷ Sweat pH reveals physical exertion levels and dehydration signs. Evaluating sweat pH changes distinguishes exercise from dehydration.^{8–12} Hence, sweat pH analysis is versatile for diagnosing diverse health conditions. From metabolic disorders to infections and skin ailments, sweat pH provides insights into health and responses to internal and external factors.

Various pH sensor categories have been explored, including ionic liquid-based sensors^{13–16} metal complex-based sensors^{17,18} and carbon electrode, graphene-based sensors.^{19,20} However, each category presents certain limitations in the terms of recyclability, flexibility and compatibility. For example, liquid-based sensors face usage cycle constraints, while metal complex sensors excel in specificity and sensitivity but lack flexibility. Carbon and graphene electrode sensors offer specificity and responsiveness but are challenged by flexibility and compatibility issues. A common challenge across these sensors is the need of additional accessories to extract quantified data as the observed changes often requires conversion into readable data. Furthermore, the process of sweat collection and detection is not concurrent, necessitating separate devices for each step. Researchers have also investigated the integration of these sensors onto textile substrates.^{3,21–23} As textile substrates offer advantages like flexibility, biocompatibility and facilitating sweat flow from skin to the device. However, challenges like skin irritation, limited permeability, and user-hostile arrangements still remain there after the arrangement. Simply altering the substrate to textile and other system same may not fully address or mitigate these drawbacks

Considering these factors, there's a demand for sensors that harmonize with the human body, enabling simultaneous collection and detection. These sensors should offer sensitivity, realtime response, and non-destructive detection. A promising approach is the direct modification of textile substrates. In existing literature, curcumin-based sensors have been developed, directly printed onto textile substrates.²²These sensors exhibit strong responsiveness and biocompatibility but presents challenges like poor washing fastness and concern of durability. After careful consideration of all aspects, we introduce pyranine (8-Hydroxypyrene-1,3,6-trisulphonic acid) based sweat pH detector using wool fabric. This utilizes pyranine as an acid dye on wool fabric, creating a dyed wool fabric with the capability to detect sweat pH. This approach establishes pH sensor that combines colorimetric and fluorescence methods. This pyranine based sensor addresses several challenges observed in previously mentioned pH sensors. By utilizing pyranine, a well-established pH-responsive and biocompatible organic dye^{24–26} we could get real-time response, non-destructive detection, biocompatibility, sensitivity, and simultaneous sweat collection and pH detection. The application of pyranine as an acid dye on wool fabric not only capitalizes on its attributes but also ensures comfort and compatibility with the human body, similar to wearing ordinary clothing. This advancement offers improved durability and reversibility, enhancing its practicality and application potential.

2. Results and discussion

2.1. Acid dyeing of wool:

Most acid dyes are of sodium salts derived from sulfonic acids. Pyranine, being structured similarly, is introduced as an acid dye. In protein fibers like wool, silk, and nylon, there are amino and carboxyl groups. When the environment is slightly acidic, around pH 5, these groups become ionized, transforming into NH_3^+ and COO^{-1} .



Figure 1:Schematic representation illustrating dye molecules adhering to substrate fibres through different types of interactions.

The sulphonyl groups on pyranine interact with the NH_3^+ ions, resulting in ionic interactions. Besides these interactions, other factors such as hydrogen bonding and van der Waals forces also play a role in the dyeing process. The combined effect of all these interactions contributes to the overall dyeing effect. Ammonium groups and fiber's amorphous parts are crucial. Amorphous regions ease dye penetration; more ammonium groups intensify interaction with anions in dyes, improving dyeing uniformity and fastness. Comparing various protein fibers, wool excels as a substrate for pyranine acid dye due to its excess ammonium groups (20 times nylon, 5 times silk) and amorphous nature aiding dye absorption and diffusion.²⁷ Wool's ammonium groups and amorphous structure uniquely fosters pyranine dye acceptance with remarkable uniformity and steadfastness.

2.2 Mechanism of pH responsiveness:

The emission of pyranine is linked to its Pka values, with ground state Pka around 7.3 - 7.7 and excited-state Pka* around 0.4 - 1.3.^{24,28-30} This creates a dual pH-sensing mechanism. pH above the pKa, pyranine exists mainly as RO⁻, emitting RO^{-*} at around 510 nm upon excitation. Conversely, at pH below the pKa, pyranine is mostly in the ROH form, producing ROH* upon excitation. The excited-state proton transfer (ESPT) process then converts ROH* to RO^{-*} with emission at ~510 nm. This leads to different steady-state excitation spectra at pH above and below the pKa, indicating distinct pathways to reach RO^{-*}. Above the pKa, RO^{-*} emission primarily comes from RO⁻, while below the pKa, it mainly comes from ROH. When using pyranine as a fluorescent pH indicator, one can either monitor RO^{-*} emission with increasing pH during RO⁻ excitation, or measure the fluorescence excitation spectrum at the emission. The key sensing element in pyranine is its hydroxyl (OH) group. Even though ionic interactions with wool fibers occur during dyeing, they do not significantly impact pH sensing. Thus, the wool fabric maintains consistent pH-sensing behavior. Post-dyeing pH sensitivity remains constant, but it affects pyranine's interaction with wool. In acidic conditions, pyranine strongly adheres to wool, while in strong basic conditions, slight leaching occurs.

The fabric's pH-sensing capabilities were investigated in both its ground and excited states. As a ground state pH indicator, the fabric exhibits a reddish hue under acidic conditions, while it appears yellowish-green (fluorescent) in the basic range. This color change is directly proportional to its interaction with the substrate. In acidic environments, there is a strong ionic interaction between wool and pyranine, causing pyranine to tightly adhere to the substrate. Conversely, in basic mediums, the interaction between pyranine and wool weakens, leading to the release of pyranine from the interaction and supporting the slight leaching mentioned above. A chromaticity diagram in the CIE 1976 color space was generated for samples at pH 1(acidic), pH 7(neutral), and pH 11 (basic), utilizing reflectance values recorded with a CCM instrument under D-65 light. This diagram visually depicts the distribution of hue and saturation to an observer under D-65 light, thereby highlighting the potential of the dyed fabric

as a ground state pH indicator. Notably, the color coordinates of the dyed fabric closely align with those of pH 7, indicating that the dyed fabric can serve directly as a reference.

The distinct characteristics of pyranine remain unchanged in our prepared sample, leading to its unaltered behaviour in its excited state as well. So, Under UV light, a distinct tripartite pattern emerges resembling the traditional Acid, Neutral, and Base zones. This pattern is accompanied by significant shifts in fluorescence. The Acidic zone shifts from blue to cyan fluorescence, while the middle zone maintains the fabric's colour but with slightly enhanced fluorescence. The Basic zone exhibits intense green-yellow fluorescence with the highest luminosity. Validation is provided through emission which confirms the presence of three distinct zones. Emission in the acidic zone occurs around 426 nm, while the basic zone it emits around 525 nm. Interestingly, emission spectra from the middle zone show characteristics of both Acidic and basic profiles that can be attributed to formation of hybrid profile. This observations underscores wool's pH detection capability, shedding light on pyranine's behavior in an excited state. Emission in Acidic, Neutral, and Basic zones is ruled by interconversions of ROH and RO⁻ states across the pH variations. Comparing our findings with the Förster



Figure 3: a) CIE 1931 chromaticity diagram of pH 1,7 and 11 (b) Emission Spectra Illustrating Emission Changes at Varying pH Values of 1,7 and 12 along with reference dyed fabric sample.

cycle³⁰ framework reveals intriguing parallels. Emission peaks in the ROH state closely align with our recorded peak at 426 nm, a result of acidic protonation. Similarly, the RO⁻ state's emission peak coincides with our 525 nm peak, a consequence of basic deprotonation. This This congruence between the data herein and the Förster cycle, supports prepared sample's

alignment with established trends. However, a nuanced perspective emerges in the middle zone. In this intermediary state, pyranine's oxygen moiety engages in geminate ion pairing with buffer solution.^{31,32} The hydroxyl group partially detaches during the shift towards basicity and partially attaches during the shift towards acidity. This intermediate state exhibits characteristics of both peaks, affirming pyranine's intricate pH-Dependent Zones behavior across diverse environments and validating our sample's versatility. Amid multiple charge environments, pyranine's behavior becomes complex.

FTIR analysis (Figure S2) indicates the presence of tertiary alcohol ($1036-1040 \text{ cm}^{-1}$) and sulphonyl group ($1171-1182 \text{ cm}^{-1}$) in the dyed fabric, as well as in the acid and base-treated samples. The tertiary alcohol groups might play a role in facilitating the presence of pyranine on the fabric. Additionally, the broadening of OH⁻ bands ($3451-3454 \text{ cm}^{-1}$) in the base-treated



Figure 2:(a) Ground State Mechanism for pH Sensing (b) Excited state pH sensing Mechanism (c) Different pH treated dyed wool fabric under visible and UV light (d) Illustration of pH sensing mechanism on the basis of energy levels in ground and excited state.

same implies a potential interaction between the base and pyranine molecules, leading to the release of pyranine from the fabric. The SEM analysis (Figure S3), despite not showing any surface deposition of dye, suggests that the dye is effectively trapped within the wool fibers. This could be due to interactions and ionic bonding, which have led to the incorporation of the dye molecules within the fiber structure.

The stability analysis of the samples was conducted to assess their resistance against rubbing and washing, with readings documented as per the standard procedures out-lined in the experimental section. The readings obtained (**Table:1**) shows both the rubbing and washing resistance tests offers excellent durability and robustness of the samples, and have ability to withstand the challenges posed by rubbing and repeated washing.

Shade %	Dry rub fastness	Wet rub fastness	Wash Fastness	
5%	5	5	5	
Rating: 1 – Poor ,2- Fair,3- Good,4 – Very good 5- Excellent				

Table 1: Washing and rubbing (Dry and Wet) fastness rating

This comprehensive analysis contributes to performance and longevity of the samples. In terms of the reversibility of the prepared sample, it exhibits the capability to transit its color states reversibly between acidic \leftrightarrow basic, acidic \leftrightarrow neutral and basic \leftrightarrow neutral conditions in UV and Visible both as shown in **Figure 4(b)**. Analysis of the prepared sample is analyzed in the artificial sweat environment (**Table:2**) considering its application as a sweat analyzer. CIE 1976 and emission spectra were recorded to check the utility of the sample as a sweat patch with real time monitoring. The results obtained shows similar trend as witnessed during the testing in buffer solution, which underlines the potential of 'Woolitmus' in the proposed application.

2.3 Application:

In the provided illustration, the depicted concept involves a pH sensitive patch with a focus on real-time applications such as health monitoring and general fitness tracking. The construction and applications of Woolitmus is outlined (Figure: 5) along with its potential impact. One notable feature is its reversibility, allowing it to be reused after immersion in a neutral solution. This feature reduces waste and enhances the economic viability of continuous monitoring. By comparing the color change of the patch against a reference or a provided color scale, individuals can assess the pH level of sweat in real time. Detecting unusual change in pH indicates underlying health conditions and triggering the need for medical attention or early intervention. With further advancements in the same, this concept holds promise for improving healthcare and wellness on a broader scale.



Figure 5: Illustration of representing proposed sweat patch along with the construction, procedure of applications and reusability; Ideal for infants with cystic fibrosis or sweat pH issues. Continuous, comfortable monitoring.

3. Experimental section:

3.1 Acid Dyeing of wool using Pyranine:

For the fabrication of this textile-based sensor simply acid dyeing of wool in open bath was carried out. Various stages of dyeing procedure along with the temperature to be maintained is mentioned in (Figure S4). Referencing the same at the start by calculating material to liquor ratio (1:20); liquor and the fabric was inserted in the pot. After the time interval of 5 minutes first addition of 2 % formic acid and 5% galubers salt (A) was carried out. Following another 5 minutes pyranine was added according to shade percentage (B) addition was performed in the same pot. Afterwards again in the interval of 5 minutes increased the temperature to 100°C. After processing the fabric for 30-45 minutes in the same bath insertion of 2% Formic acid in combination with 2% sulphuric acid was added (C) to provide acidic environment for the electrostatic interaction and fixation of the pyranine dye on wool. Further wool fabric was processed for 30 more minutes in the same acidic environment before the washing in the nonionic soap solution and air drying was carried out.

3.2 Characterization

Scanning electron microscopy is used to analyze the sur-face morphology of the fabric to confirm the dyeing and not the surface deposition. Similarly, fourier transform infra-red (FT-IR) spectra of fabric (Dyed wool reference, fabric treated with pH 1 and pH 12) was performed on Shimadzu FTIR 8400S to detect the change in functional groups due to ionic environment. Prepared fabric-based sensor shows the change in UV and visible light both. To interpret the behavior in UV region, emission spectra of the sample recorded using JASCO 6000 fluorescence spectroscopy. To prove the changes with pH in visible light, CIE chromaticity diagram was plotted using colorfast color matching machine.

3.3 pH colorimetric Sensing Test

To prove the creditability of the prepared dyed wool fabric as pH sensor, the dyed fabric was checked against the viable pH range. To check the change, the pH solution was poured into the small beaker and the strip of the fabric was half dipped into the solution for one minute. As mentioned, the change occurs is detected and interpreted in UV and visible light using emission spectra and CIE color space values respectively. Investigate sample reversibility by testing against pH 1, 7, and 12. Immerse samples in corresponding buffers for one minute based on reversibility sequence (e.g., acid \leftrightarrow base, acid \leftrightarrow neutral, neutral \leftrightarrow base). Analyze various reversibility combinations. To analyze the pH sensitivity of the sample in the artificial sweat environment the preparation of acidic (pH 4.4) and basic (pH 8) sweat was prepared by

Perspiration	Reagent required for Alkaline perspiration	Reagent required for Acidic Perspiration
Histidine mono	0.5 gram/liter	0.5 gram/liter
hydrochloride monohydrate		
Sodium Chloride	0.5 gram/liter	0.5 gram/liter
Disodium Hydrogen	2.5gram/liter	
Orthophosphate		
Sodium dihydrogen		2.5gram/liter
orthophosphate		
0.1 N Sodium Hydroxide	To adjust pH 8	
0.1N Acetic Acid		To adjust pH 4.4

 Table 2: Recipe of acidic and basic sweat preparation

following recipe³³ (Table: 2). To analyze its efficiency the sample was dipped inside the prepared sweat solution for one minute and changes were studied accordingly.

3.4 Stability of sensor to real conditions

To look into the durability of the fabric sensor against the washing and rubbing considering its application. Washing fastness was performed using non-ionic soap solution of 2 gpl at 60°C for 30 minutes in the Laundrometer. Dry and wet rub fastness of dyed wool fabric samples were tested using a crock meter as per standard IS 766:1988 (Reaffirmed 2004) based on ISO 105-X12:2001 by mounting the yarn/fabric on panel and giving ten strokes for both dry and wet rub fastness tests.

4. Conclusion:

In this study, a pH sensor using pyranine dye was developed on wool as a substrate, showing pH responsiveness in visible and UV light. The sensor has potential to serve as a sweat patch for real-time pH monitoring. The pH sensitive patch offers real-time pH monitoring with applications in health and lifestyle management without any supporting accessory, contributing to the growing field of textile-based sensors for healthcare and allied areas.

Supporting Information

Supporting Information is available.

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