

New Optical Sensing Reagent of Bis(2-oxidobenzylidene)benzenesulfonohydrazide Nickel(II) for the Detection of Formaldehyde in Fish

Wan Rusmawati Wan Mahamod¹, Yusnita Juahir¹, Norhayati Hashim¹,
Nurusaidah Abdul Rahim¹, Maizatul Najwa Jajuli¹, Norlaili Abu Bakar^{1*},
Nurul Hana Baharom² and Norlin Suhaiza Musali³

¹Department of Chemistry, Faculty of Science and Mathematics, Universiti Pendidikan Sultan Idris,
35900 Tanjung Malim, Perak, Malaysia

²Sekolah Menengah Sains, Jalan Sultan Suleiman Jugra, 42700 Banting, Selangor Darul Ehsan, Malaysia.

³Sekolah Menengah Kebangsaan Seri Jengka, Pekan Tajau S26500 Maran, Pahang

*Corresponding author (e-mail: norlaili@fsmt.upsi.edu.my)

An optical chemical sensor for the determination of formaldehyde in fish was developed based on bis(2-oxidobenzylidene)benzenesulfonohydrazide nickel(II) (Ni(SALBSH)₂). The Ni(SALBSH)₂ complex was immobilised onto a PVA/Chitosan membrane and designated as Ni(SALBSH)₂-PVA/Chi Probe. The absorbance intensity of the Ni(SALBSH)₂-PVA/Chi Probe decreased at a wavelength of 326 nm in the presence of formaldehyde. The reaction of the Ni(SALBSH)₂-PVA/Chi Probe and formaldehyde was optimal at pH 5 and a complex concentration of 300 mgL⁻¹. The dynamic range of the Ni(SALBSH)₂-PVA/Chi Probe for formaldehyde was 20 – 500 mgL⁻¹, and the limit of detection was 2.52 mgL⁻¹. This probe is suitable for detecting individual analytes in clean aqueous solutions but is not directly applicable to complex matrices, such as fish homogenates.

Keywords: Benzenesulfonylhydrazone, optical, formaldehyde, seafood safety, aquatic product

Received: February 2026; Accepted: August 2026

Fish offers numerous nutritional benefits that make it a valuable component of a healthy diet [1]. According to [2], the recognised nutrients contained in fish, such as a high concentration of omega-3 fatty acids, have essential functions in the body and are associated with a lower risk of all-cause mortality. However, despite the nutritional values present in fish, the toxicity of formaldehyde in fish is a well-known concern [3]. Formaldehyde is an intensely reactive solution or gas that is colourless, flammable, highly irritating, and asphyxiating [4]. It belongs to the aldehyde group, which has a pungent scent and is widely recognised as a poisonous substance that can affect human health [5].

Formaldehyde is commonly used by illicit traders in the pharmaceutical, wood, and textile industries and has also been used as a food preservative, especially in aquatic goods [6], [7]. Specific properties of formaldehyde, such as preservation, disinfection, and bleaching, have led to its use in various industries, including paint, cosmetics, foodstuffs, and adhesives. In comparison, formaldehyde, a known carcinogen, has been used as an industrial preservative in several foods, such as beef, fruits, and seafood [5]. In addition, formaldehyde has been ranked by the World Health Organization (WHO) as the third highest indoor chemical pollutant. More specifically, formaldehyde can cross-link proteins

and DNA molecules through a stable methylene bridge owing to its powerful capability of being a highly reactive carbonyl species [8, 9]. Formaldehyde contaminants in food and the atmosphere may also pose a significant threat to human life and health [4]. Formaldehyde solution is intentionally sprayed or dipped in fish to extend shelf life, stiffen, keep them looking new, and avoid spoilage [10]. The European Food Safety Authority (EFSA) has summarised the formaldehyde level in food, namely meat and poultry is 5.7–20 mg kg⁻¹, fish is 6.4–293 mg kg⁻¹, milk and milk-based products is 0.01–0.80 mg kg⁻¹, sugar and sweeteners is 0.75 mg kg⁻¹, fruit and vegetables are 6–35 mg kg⁻¹, coffee is 3.4–16 mg kg⁻¹ and alcoholic beverages is 0.27–3.0 mg kg⁻¹ [11, 12].

Studies have shown that abnormal accumulation of formaldehyde in humans can lead to a number of diseases, such as asthma, chronic liver disease, heart disease, Alzheimer's disease, lung cancer, nasopharynx, nasal oropharynx, and stomach [3]. Therefore, it is necessary to detect formaldehyde in fruit, water, and indoor environments easily and accurately [4]. In recent years, numerous methods have been developed for monitoring formaldehyde levels, including spectrophotometry, electrochemistry, and chromatography, which are reliable and consistent but entail complex, high-cost, and time-consuming procedures. There is also an urgent need to discover

a new method to measure the concentration of formaldehyde in food products [13, 14].

Over the last few decades, a range of analytical and testing approaches have been researched and developed to detect formaldehyde, such as colorimetric assays, gas chromatography, gas chromatography-mass spectrometry, and ultraviolet spectroscopy [15 – 17]. However, they typically suffer from certain drawbacks, including poor sensitivity, complicated procedures, and the need for intrusive destruction of biological tissues [4]. According to Kucukoglu et al. [18], the development of new hydrazone compounds has demonstrated that they exhibit a broad range of biological functions, such as antimicrobial, anticonvulsant, antidepressant, antiplatelet, antimalarial, anticancer, antifungal, antitubercular, antiviral, anti-inflammatory, analgesic, and cardioprotective activities. Hydrazones / azomethine imines represent a significant class of compounds for the production of novel drugs. A number of researchers have prepared and analysed the biological behaviours of hydrazones [19 – 20].

In the study conducted by Lijun et al. [21], a new Rhodamine B hydrazide hydrazone was synthesised and investigated as a colorimetric and fluorescent “off-on” sensor for the recognition of Cu^{2+} in $\text{CH}_3\text{CN}/\text{H}_2\text{O}$ (1:1 v/v). The study showed that the new rhodamine B-based sensor exhibited highly selective and rapid Cu^{2+} recognition behaviour *via* colorimetric and fluorescent detection modes. This study also shows that hydrazone derivative such as benzenesulfonylhydrazone has a property for sensing reagent. This study focused on highly stable metal complexes as sensing reagents. The complex used was a metal complex of salicylaldehyde benzenesulfonylhydrazone. The stability criteria of this complex provide an opportunity to overcome the leaching problem in the immobilisation process.

EXPERIMENTAL

Chemicals and Materials

All chemicals, namely 37% formaldehyde/formalin, chitosan, polyvinyl alcohol, acetic acid, potassium chloride, ammonium acetate, and sodium carbonate, used in this study were purchased from Sigma Aldrich and System. All solutions were prepared using double-deionised water. All chemicals were of analytical grade and used without further purification.

Preparation and Characterisation of $\text{Ni}(\text{SALBSH})_2$ – PVA/Chi Probe.

Chitosan solution (2%) was prepared with mixture of chitosan and acetic acid (1 %) undergoes 24 hours continuous stirring for homogeneous. Polyvinyl alcohol was prepared by dissolved powder of PVA into 100 mL hot distilled water and stirred for 30 minutes until homogeneous achieved. The metal

complex of bis(2-oxidobenzylidene) benzenesulfonylhydrazide) nickel(II), known as $\text{Ni}(\text{SALBSH})_2$, was synthesised using the method described by Juahir et al. (2009) [22]. The immobilisation of $\text{Ni}(\text{SALBSH})_2$ (light green colour) was prepared by mixing 7 mL of chitosan (1%), 7 mL of PVA (5%), 0.025 mL of glutaraldehyde, and 6 mL of 1000 mg/L $\text{Ni}(\text{SALBSH})_2$. The mixture was then stirred for 30 minutes. Then, 20 mL of the mixture was poured into a Petri dish and left overnight to dry. The membrane was peeled off and cut into 1.5 cm × 3.0 cm pieces for further studies. Three PVA: Chitosan ratios by volume (v/v) were prepared: 1:1, 1:2, and 2:1. The obtained membranes M1 (1:1), M2 (1:2), and M3 (2:1) with $\text{Ni}(\text{SALBSH})_2$ were known as $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi Probes and were kept in sample bottles for further experiments. In this study, the probes of $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi were characterised using FTIR Thermo Nicolet 6700 spectrophotometer in the range of 400 – 4000 cm^{-1} at room temperature using ATR method. The surface morphology of the probe was studied using an optical polarising microscope (OPM).

Sensing Characterisation of $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi Probe

The sensing characterisation was analysed by studying the response of the $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi Probe against formaldehyde which includes the optimum PVA/Chi ratio, pH, optimum $\text{Ni}(\text{SALBSH})_2$ concentration, leaching study, response time, effect of formaldehyde concentration, interference study, and dynamic range and performance of the probe against formaldehyde in real samples. The response of the $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi probe to the presence of formaldehyde in aqueous solution was tested using an Agilent UV-Visible Spectrophotometer. The absorbance spectrum of the membrane was recorded before and after the reaction with formaldehyde. This was done by immersing the $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi probe in 500 mgL^{-1} formaldehyde for 5 minutes. The probe was then dried and the absorbance spectrum was recorded using UV-Vis spectrophotometer. The difference in absorbance intensity at 326 nm before and after the reaction with formaldehyde was calculated using Equation 1 and is known as the relative absorbance.

$$\text{Relative Absorbance} = I_o - I_f \quad (1)$$

I_f = Absorbance intensity after reacting with analyte

I_o = Absorbance intensity before reacting with analyte

A sensing response study of the $\text{Ni}(\text{SALBSH})_2$ –PVA/Chi probe with different PVC/Chi ratios (M1, M2, and M3) against formaldehyde was carried out by immersing the probe in 500 mgL^{-1} formaldehyde for 5 minutes. The pH of the formaldehyde solution was adjusted using several buffer solutions with pH values of 2 (HCl and KCl), 3 – 5 (acetate buffer), 6 – 8 (phosphate buffer), and 9 – 10 (Trizma base, HCl,

and NaOH). A probe was immersed in a 500 mgL^{-1} formaldehyde solution at different pH values. The absorbance spectrum was recorded after 5 minutes. A study of the optimum concentration of the reagent was carried out in the range of 200 to 500 mg/L . The reagent $\text{Ni}(\text{SALBSH})_2$ at different concentrations was immobilised into PVA/Chitosan separately. Each membrane (fixed size) was immersed in 500 mgL^{-1} formaldehyde and a buffer solution (pH 5) separately. For the response time study, the $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe was immersed in 500 mgL^{-1} formaldehyde at optimum pH for 30, 60, 90, 120, 180, 240, 300, and 360 seconds. After a certain time, the probe was dried and the absorption spectrum was recorded using UV-Vis spectrophotometer to determine the absorbance intensity after the reaction. A dynamic range study was performed to investigate the effect of varying the concentration of formaldehyde on the response of the $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe. The concentration of formaldehyde was varied from 50 mgL^{-1} to 1000 mgL^{-1} . The $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe was immersed in $50 - 1000 \text{ mgL}^{-1}$ formaldehyde for 180 s separately. The $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe was then dried before the absorption spectrum was recorded using UV-Vis spectrophotometer. The $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe was tested for reagent leaching. The probe was immersed in 3 mL of acetate buffer solution (pH 5.0) for 0, 0.5, 1, 2, 3, 5 and 24 hours prior to the evaluation of membrane

performance with 500 mg/L formaldehyde. The absorbance intensity of the acetate buffer solution was recorded after immersion of the membrane. All experiments for the sensing characterisation were performed using an Agilent UV-Visible spectrophotometer. Relative absorbance was applied for all sensing characterisations of the $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi probe against formaldehyde in an aqueous medium.

RESULTS AND DISCUSSION

Characterization of $\text{Ni}(\text{SALBSH})_2$ Complex and $\text{Ni}(\text{SALBSH})_2$ -PVA/Chi Probe

Figure 1 shows the FTIR spectra of the free hydrazone ligand (SALBSH) and $\text{Ni}(\text{SALBSH})_2$ complex. The most significant structural difference was detected in the wavenumber range above 3000 cm^{-1} . The free ligand exhibited a broad absorption band at $3150 - 3200 \text{ cm}^{-1}$, attributed to the phenolic $\nu(\text{O-H})$ and/or hydrazide $\nu(\text{N-H})$ stretching vibrations. However, this band was not visible in the spectrum of the $\text{Ni}(\text{SALBSH})_2$ complex. According to Lira et al. (2025) [23] and Kalla et al. (2024) [24], the disappearance this phenolic $\nu(\text{O-H})$ band confirms the phenolic deprotonation process and the formation of a covalent Ni-O bond through the coordination of the phenolate oxygen atom.

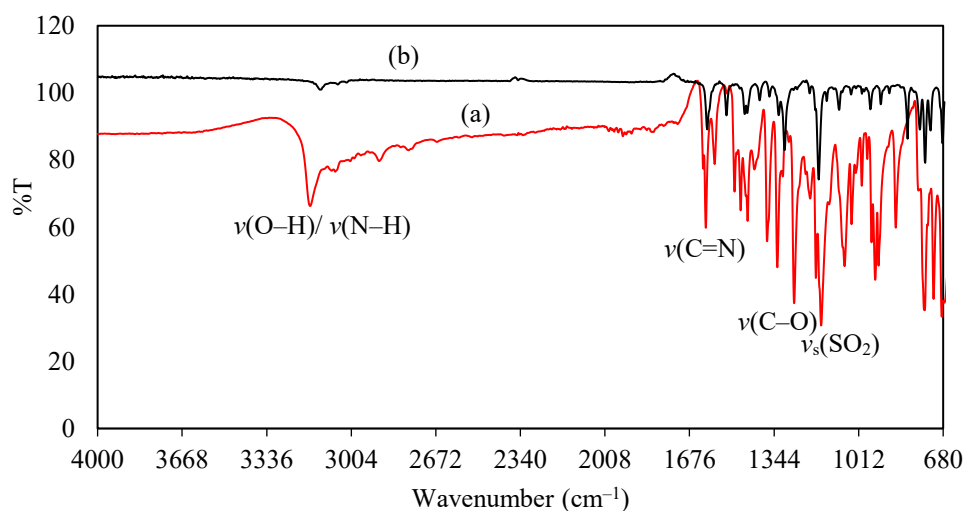


Figure 1. The FTIR Spectra of ligand (SALBSH) (a) and $\text{Ni}(\text{SALBSH})_2$ complex.

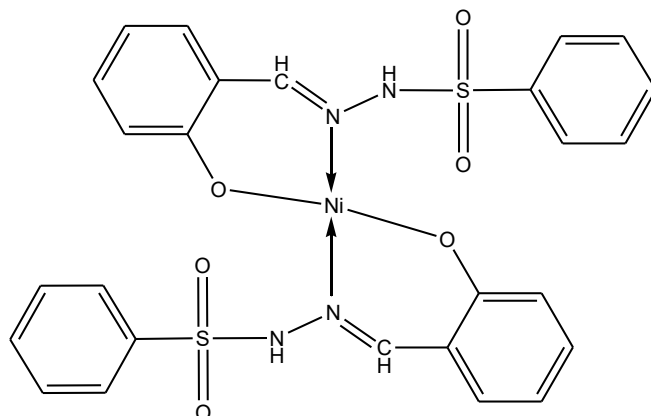


Figure 2. The molecular structure of metal complex bis(2-oxidobenzylidene)benzenesulfonohydrazide) nickel(II).

Furthermore, a shift in the azomethine stretching vibration occurs, where the sharp C=N bond band in the free ligand (1618 cm^{-1}) undergoes a bathochromic shift to 1600 cm^{-1} after complexation. This downward shift ($\Delta\nu \approx 18\text{ cm}^{-1}$) confirms the donation of the azomethine nitrogen's lone pair of electrons into the empty d orbital of the Ni(II) ion ($\text{N} \rightarrow \text{Ni}$). This condition reduces the electron density and weakens the intrinsic force constant of the C=N bond, as reported by Juahir et al. (2008) [25], Firmansyah et al. (2023) [26], and Vennila et al. (2024) [27]. Meanwhile, the shift of the phenolic stretching vibration, $\nu(\text{C}-\text{O})$, from 1250 cm^{-1} to 1290 cm^{-1} further supports the involvement of the phenolic oxygen atom in the coordination bond formation. Figure 2 shows the full structural configuration of the Ni(SALBSH)₂ complex.

The symmetric sulfonyl stretching vibration, $\nu_s(\text{SO}_2)$, located at $\sim 1150\text{ cm}^{-1}$, remained unshifted in the FTIR spectra of both the free SALBSH ligand and the Ni(SALBSH)₂ complex. This confirms that the sulfonyl group functions as a non-coordinating spectator substituent. In contrast, the emergence of weak new absorption bands in the lower wavenumber region ($< 600\text{ cm}^{-1}$) provides direct evidence for metal–ligand bond formation, assigned to $\nu(\text{Ni}-\text{O})$ and $\nu(\text{Ni}-\text{N})$ stretching modes. Overall, the absence of the phenolic $\nu(\text{O}-\text{H})$ band, the observed shifts in the $\nu(\text{C}=\text{N})$ and $\nu(\text{C}-\text{O})$ vibrations, alongside the appearance of new metal–ligand bands, collectively verify the successful synthesis of the Ni(SALBSH)₂ complex.

The PVA/Chitosan membrane displayed a strong, broad peak at approximately 3300 cm^{-1}

($3200\text{--}3500\text{ cm}^{-1}$), which was attributed to overlapping $\nu(\text{O}-\text{H})$ and primary amine $\nu(\text{N}-\text{H})$ stretching vibrations. According to Kadir et al. (2011) [28], this broad profile indicates comprehensive intra- and intermolecular hydrogen bonding within the host blend. The retention of this broad band in the Ni(SALBSH)₂-PVA/Chi probe confirms that the underlying polymer network remains structurally intact after immobilisation.

Successful immobilisation was confirmed in the fingerprint region ($1000\text{--}1700\text{ cm}^{-1}$) region. While the blank PVA/Chitosan matrix displayed a broad, simple absorption profile, the Ni(SALBSH)₂-PVA/Chi probe exhibited a composite spectrum featuring newly emerged sharp peaks at 1600 cm^{-1} , 1350 cm^{-1} , and 1150 cm^{-1} . These peaks serve as spectroscopic footprints for the coordinated metal-sulfonylhydrazone complex (aromatic C=C, azomethine C=N, and symmetric SO_2 modes) [25, 27]. Additionally, subtle frequency shifts and intensity fluctuations observed in the C–O–C/saccharide ring stretching region ($1000\text{--}1100\text{ cm}^{-1}$, attributed to PVA C–O and chitosan pyranose) signify non-covalent physical encapsulation, electrostatic interactions, and hydrogen bonding between the host functional groups ($-\text{OH}/-\text{NH}_2$) and guest molecules [23, 24]. The clear and sharp peaks observed in the wavenumber range of $< 900\text{ cm}^{-1}$ verify that the guest molecules are uniformly dispersed throughout the matrix. The preservation of the host polymer structure and the emergence of new coordination bands in the FTIR spectrum of the Ni(SALBSH)₂-PVA/Chi probe demonstrate the successful immobilisation of the Ni(SALBSH)₂ complex in the PVA/Chi membrane.

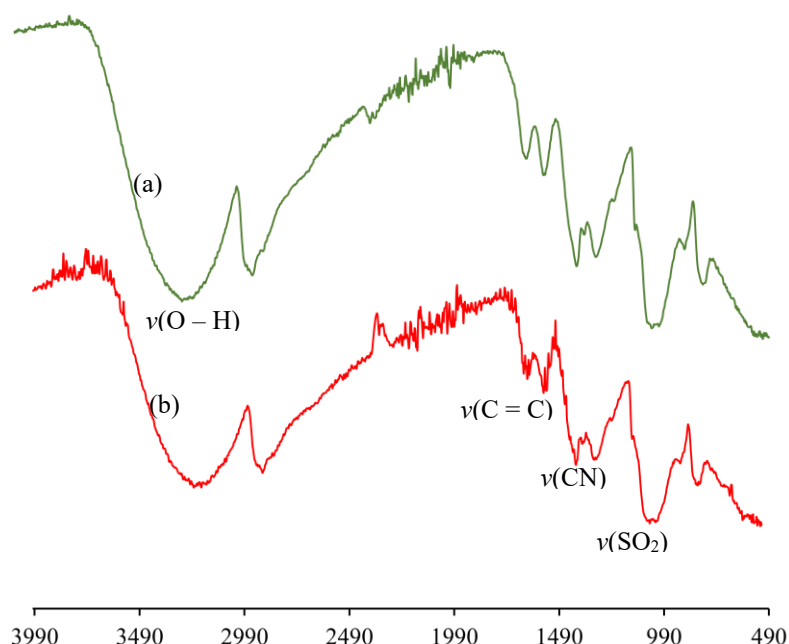


Figure 3. The FTIR spectrum of PVA/Chitosan membrane (M1) (a) and Ni(SALBSH)₂-PVA/Chi Probe (M1) (b).

Figure 4 shows the images of PVA/Chitosan membranes with different ratios (M1, M2, and M3). The PVA/Chitosan membrane (M1) exhibited a highly uniform, smooth, and homogeneous phase distribution. This structural homogeneity maximises the interfacial surface area while minimising steric hindrance, thereby facilitating unhindered analyte diffusion into the polymer matrix [29]. Consequently, this well-blended configuration promotes an optimal and uniform dispersion of the active Ni(SALBSH)₂ complex

throughout the matrix. The image also shows that membrane M1 exhibited the minimum cracking effect, and the surface was the smoothest compared to M2 and M3. Higher PVA loading increases the crystallisation tendency of the membrane, which inhibits polymer chain mobility and accelerates microcracking across the matrix [30]. Therefore, the blend ratio utilised in membrane M1 provides the necessary balance of polymer compatibility and structural morphology to achieve effective formaldehyde sensing.

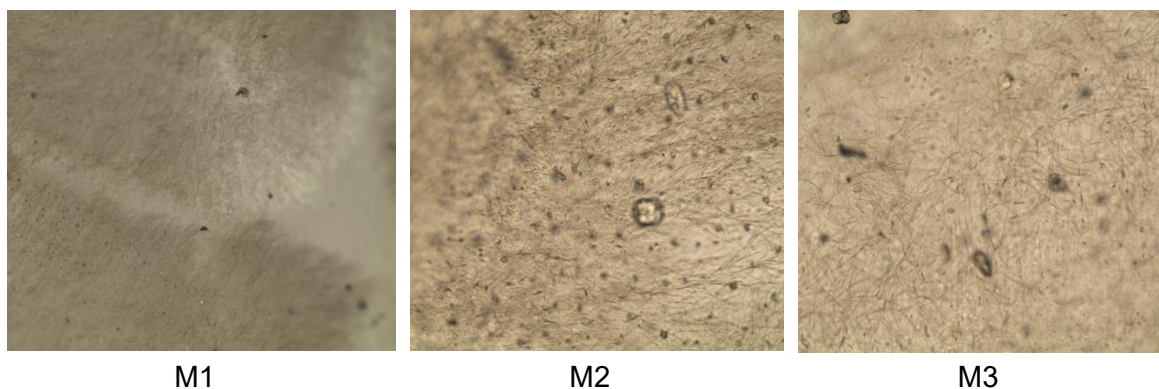


Figure 4. Optical microscopic image of Ni(SALBSH)₂-PVA/Chi Probe with different ratio of PVA: Chitosan (M1, M2, M3).

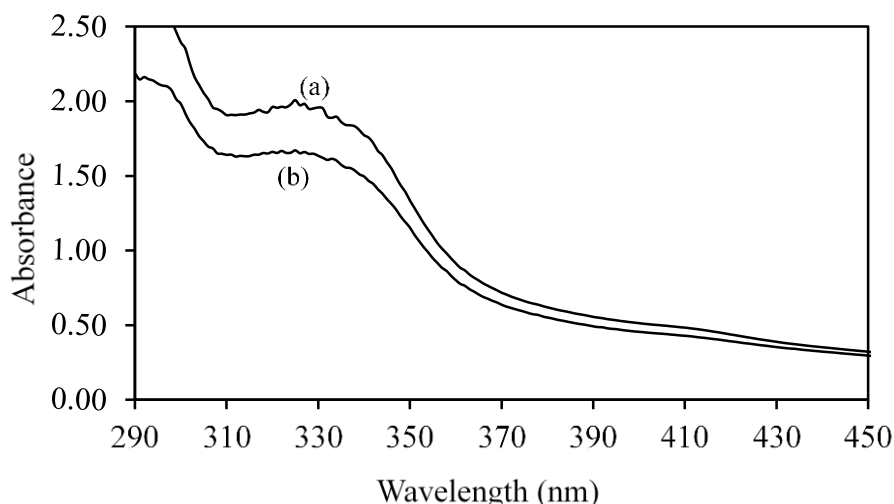


Figure 5. The absorption spectra of Ni(SALBSH)₂-PVA/Chi Probe M1 before (a) and after (b) reaction with formaldehyde. $\lambda_{\text{max}} = 326 \text{ nm}$, formaldehyde concentration = 500 mgL^{-1} .

The Absorbance Spectrum of Ni(SALBSH)₂ – PVA/Chi Probe

The ultraviolet–visible spectrum of the Ni(SALBSH)₂-PVA/Chi Probe against formaldehyde is shown in Figure 5. The absorbance intensity of the Ni(SALBSH)₂-PVA/Chi Probe at 326 nm decreased after the reaction with the formaldehyde solution. The decrease in absorbance upon reaction with formaldehyde in the nickel complex arises from the disruption of metal–ligand charge transfer and coordination, whereas the reaction with the free ligand does not significantly alter its chromophoric system [31].

Optimization of PVA/Chitosan Ratio

The PVA/Chitosan membrane M1 exhibited the highest relative absorbance, indicating an outstanding sensing interaction with formaldehyde compared to M2 and M3 (Figure 6). The decreased in optical performance can be directly elucidated by the surface morphologies detailed in the optical microscopic images (Figure 4). The surface morphologies of M2 and M3 induced extensive light scattering during spectrophotometric measurements, which directly contributed to their much lower relative absorbance intensity [32]. Thus, membrane M1 was used for further experiments in the sensing characterisation.

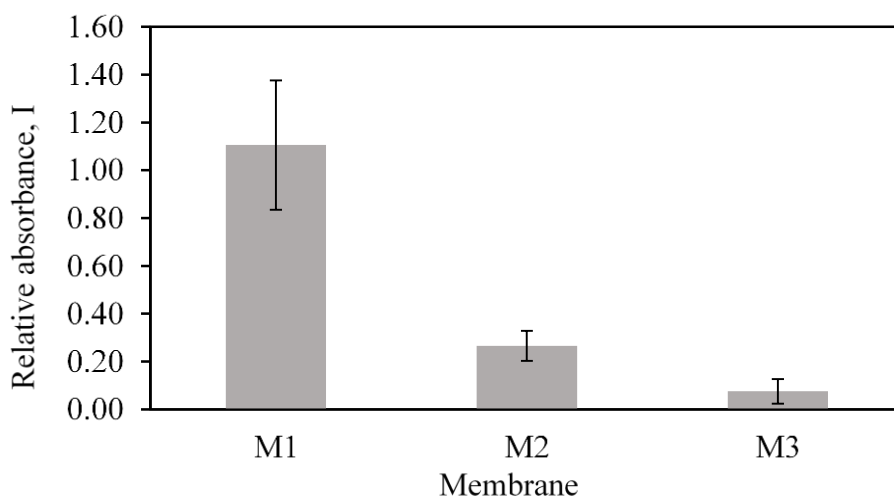


Figure 6. Relative Absorbance of Ni(SALBSH)₂-PVA/Chi Probe with different PVA/Chitosan composition (%v/v). The concentration of formaldehyde = 500 mgL^{-1} , $\lambda_{\text{max}} = 326 \text{ nm}$.

pH Study and Optimum Concentration of Reagent

The effect of pH on the response of the Ni(SALBSH)₂–PVA/Chi probe was investigated over the pH range of 2.0–10.0. The probe exhibited an optimum response at pH 5.0 in acetate buffer, as depicted in Figure 7. The larger error bar observed across different pH values is attributed to the varying ionic strengths and anion interactions from the distinct buffer systems (acetate, phosphate, and Tris) used across pH 2–10, which influence the swelling and light-scattering properties of the PVA/chitosan matrix in the low-absorbance regime (< 0.5 AU) [33]. In the study by Shahvar et al. [34], ammonium acetate was used to identify formaldehyde, and the optimum response was obtained

at pH 5.5. In the study by Chaiendoo et al. [35], a silver-amine complex ion [Ag(NH₃)₂], known as the Tollens reagent, was used for the identification of formaldehyde and showed strong pH absorption at 4.5. Absorption levels were diminished at other pH values.

Figure 8 shows that the absorbance intensity of the Ni(SALBSH)₂–PVA/Chi Probe increased with increasing Ni(SALBSH)₂ complex concentration. This increase was not reliable because the probe became less transparent at a concentration of 300 mgL^{−1} (Figure 9). This membrane condition causes the maximum peak of the absorption spectrum to be uneven (unsmooth), and the absorbance intensity at 326 nm is inaccurate.

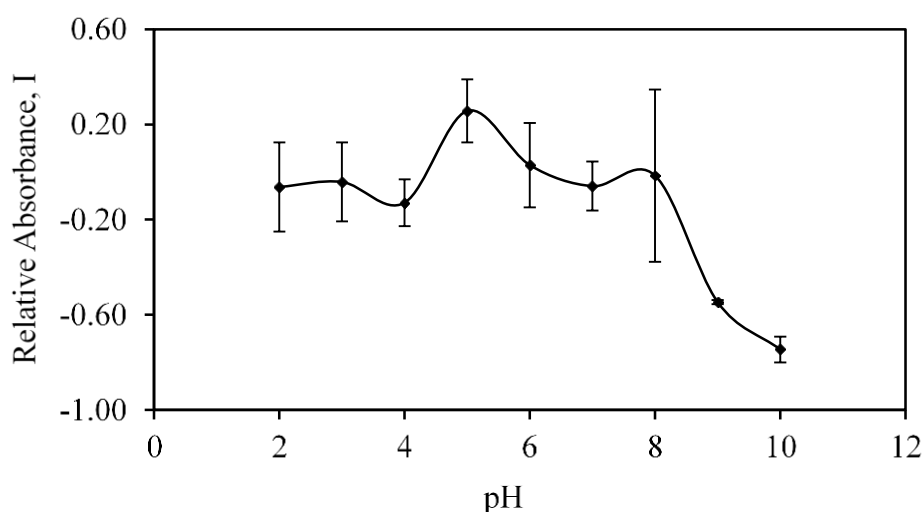


Figure 7. Effect of pH on the response of Ni(SALBSH)₂–PVA/Chi Probe M1 against formaldehyde. Formaldehyde concentration = 500 mgL^{−1}, λ_{max} = 326 nm.

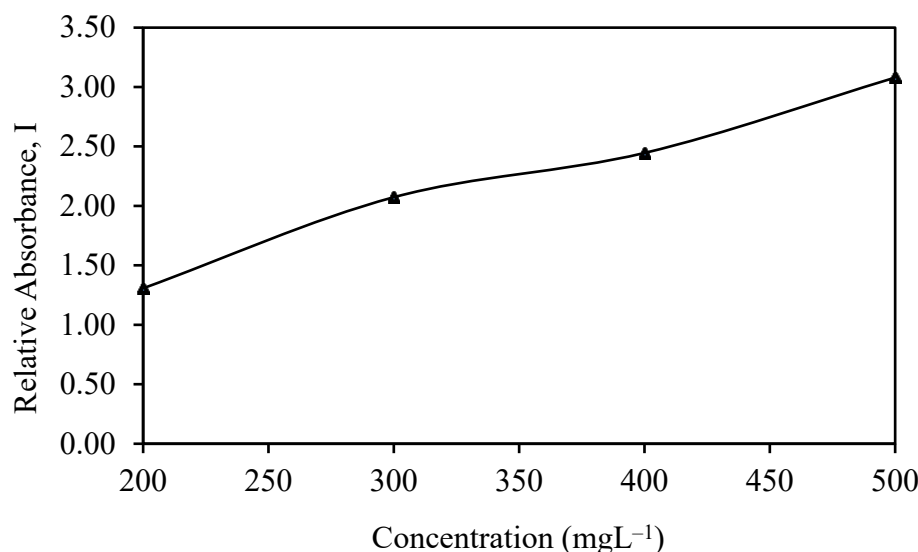


Figure 8. Effect of Ni(SALBSH)₂ complex concentration in PVA/Chitosan M1 against formaldehyde in aqueous solution. Concentration of formaldehyde = 500 mgL^{−1}, λ_{max} = 326 nm.

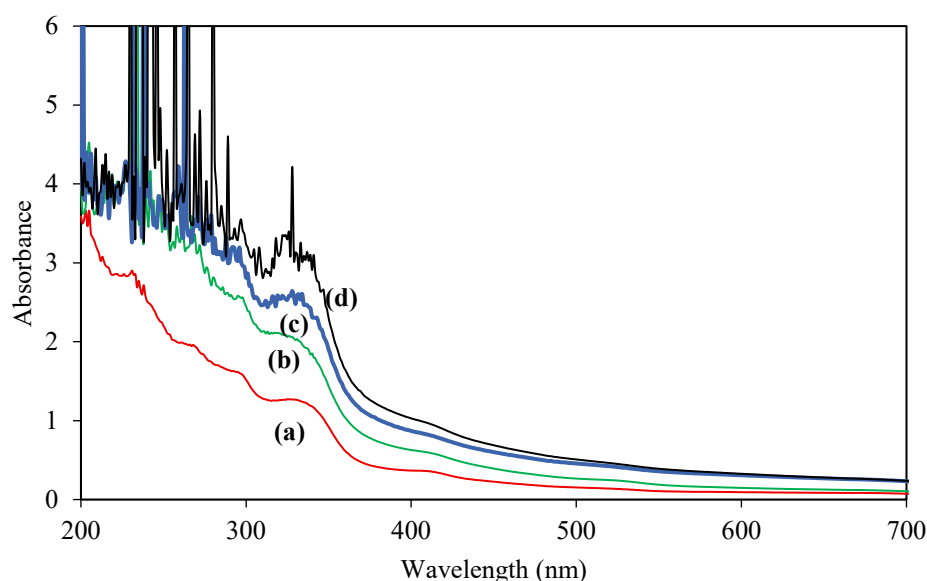


Figure 9. The absorption spectrum of Ni(SALBSH)₂-PVA/Chi Probe M1 with 200 mgL⁻¹ (a), 300 mgL⁻¹ (b), 400 mgL⁻¹ (c), and 500 mgL⁻¹ (d) complex concentration against formaldehyde in aqueous medium.

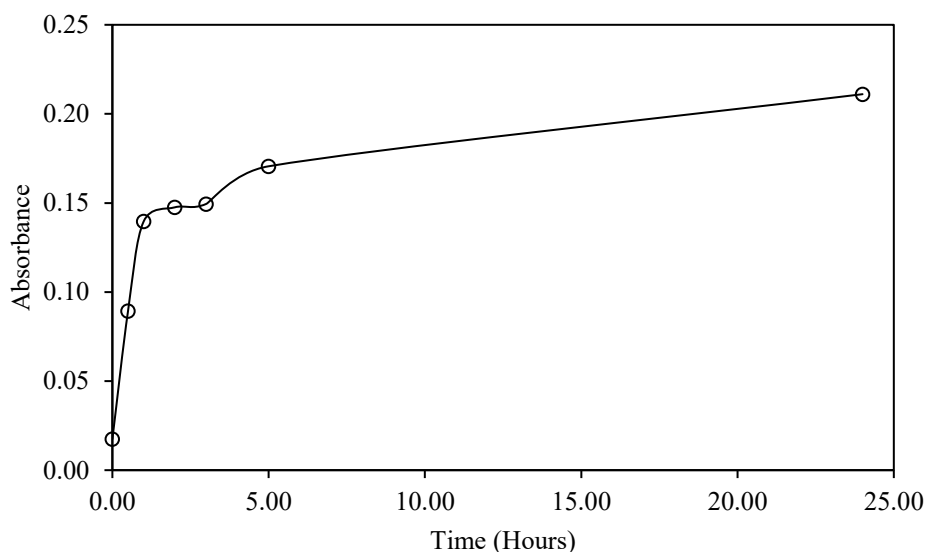


Figure 10. Leaching stability profile of Ni(SALBSH)₂-PVA/Chi Probe M1 in acetate buffer (pH 5.0) for 24 hours.

A reagent concentration of 300 mgL⁻¹ was chosen as the optimum because the membrane was more uniform and translucent and produced a smoother spectrum. According to the Beer–Lambert law, the measured absorbance is directly proportional to the molar absorptivity, optical path length, and concentration of the absorbing species at a specific wavelength [36, 37]. However, deviations from the Beer–Lambert law may occur when the physical

characteristics of the sensing membrane change, particularly at higher membrane concentrations. Factors such as increased membrane thickness, light scattering, sample heterogeneity, stray light, and spectral noise can reduce the linear relationship between absorbance and concentration by affecting the transmitted light intensity and signal-to-noise ratio [37, 38]. Therefore, the 300 mg L⁻¹ membrane concentration was considered optimal and selected for further study.

Leaching Study and Steady State Response Time

Figure 10 shows the leaching study of Ni(SALBSH)₂-PVA/Chi Probe. Within a 180-second (0.05 h) response time, complex leaching from the host matrix remained minimal ($\leq 2.5\%$), ensuring that $>97.5\%$ of the active species stays operational during signal acquisition. Such low leaching easily satisfies the allowable limit ($\leq 3\text{--}5\%$) required for stable optical sensing applications [39, 40].

Figure 11 shows the steady-state response times of the Ni(SALBSH)₂-PVA/Chi Probe for 500 mgL⁻¹ formaldehyde solution. The relative absorbance intensity dropped sharply and recovered

after 90 s, attaining a steady-state equilibrium at 180 s. Fast interfacial interactions and boundary layer disruption at the sensor-liquid interface cause a steep initial decrease at $t \leq 90$ s [41, 42]. This rapid surface binding temporarily alters the local dielectric microenvironment of the chromophore before full analyte penetration occurs. As analyte molecules diffuse into the bulk PVA/Chitosan matrix for coordination, thermodynamic equilibrium is restored, returning the signal to a stable baseline at 180 s [43]. Thus, 180 s was chosen as the steady-state response time. At this point, all the immobilised Ni(SALBSH)₂ complex was almost used up in the reaction, and further exposure to formaldehyde did not increase the absorbance intensity.

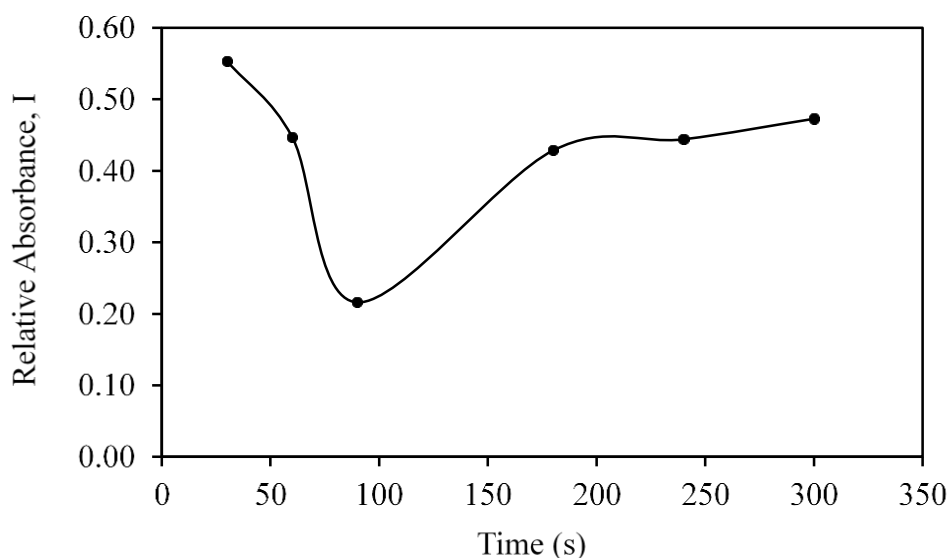


Figure 11. Steady state response time of Ni(SALBSH)₂-PVA/Chi Probe against formaldehyde. Concentration of formaldehyde = 500 mgL⁻¹, λ_{max} = 326 nm.

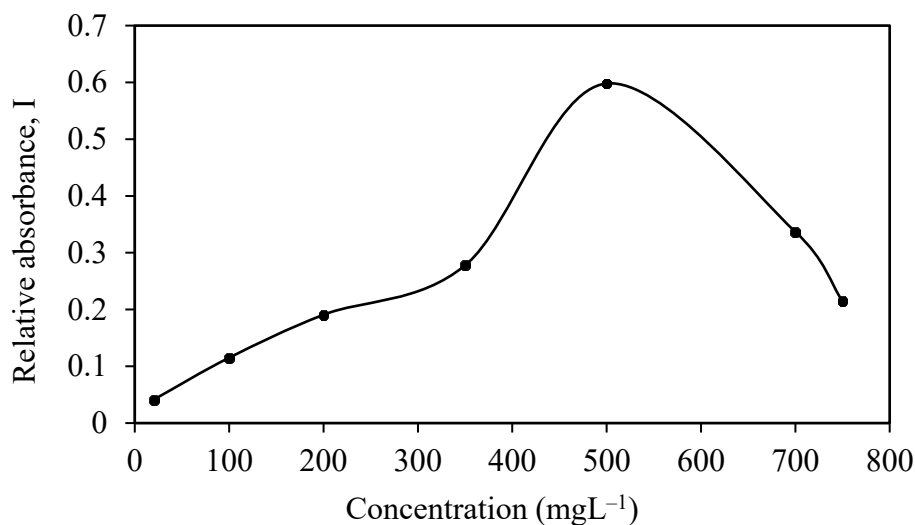


Figure 12. Effect of analyte concentration on Ni(SALBSH)₂-PVA/Chi Probe M1 response against formaldehyde in aqueous solution. λ_{max} = 326 nm, time = 180 s.

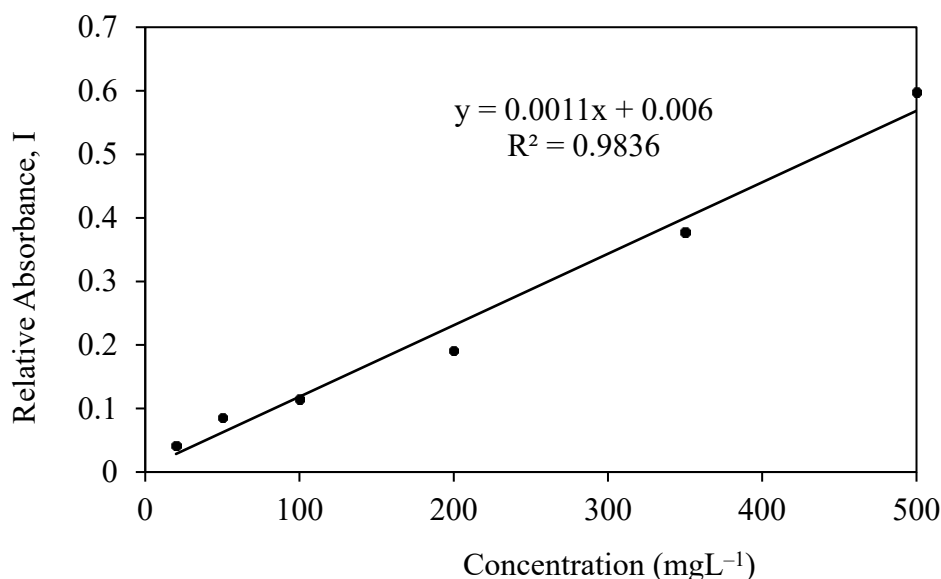


Figure 13. Linear range of Ni(SALBSH)₂-PVA/Chi Probe M1 response against formaldehyde in aqueous solution. $\lambda_{\text{max}} = 326 \text{ nm}$, time = 180 s.

Effect of Analyte Concentration Against Ni(SALBSH)₂-PVA/Chi Probe Response

The dynamic range was determined in the range of 20-700 mg/L of formaldehyde solution. Figures 12 and 13 show the effect of analyte concentration on the Ni(SALBSH)₂-PVA/Chi Probe. The relative absorbance of the membrane increased with increasing analyte concentration until 500 mgL⁻¹. The relative absorbance intensity decreased drastically after 500 mgL⁻¹. The same trend was reported by Tan et al. [44] in the determination of ammonium ions using a reagentless amperometric biosensor based on immobilised alanine dehydrogenase. The biosensor reaction was observed to increase the NH₄⁺ ion concentration from 10–500 mM and achieve saturation above 500 mM NH₄⁺ ions.

The absorbance strength decreased at concentrations above 500 mgL⁻¹. The linear concentration of formaldehyde was estimated in the range of 20 – 500 mgL⁻¹. This concentration range is broader than that reported by Hayun et al. [45], who found a range of 4 – 40 mgL⁻¹ for the assessment of formaldehyde content in wet noodles and that reported by Rungroadsri et al. [46], who found a range of 1 – 50 mgL⁻¹ for the identification of formaldehyde in food by digital image colorimetry.

Interference and Recovery Study

The response of the Ni (SALBSH) 2–PVA/Chi Probe was tested in the presence of formaldehyde and interfering ions. The results showed that the presence of foreign ions interfered with the detection of formaldehyde. Table 1 summarises the interference from various ions in the determination of formaldehyde. Carbonate and cations exhibited negative interference, and iron (III) and sodium ions were found to be the most prominent, with interference over 400%. This finding is conflicting, as Ehsan and Rehman [47] observed in their formaldehyde determination using a nanostructured nickel thin film. Two regular additions of interference species revealed only a few noisy fluctuations in the sensor signal, which can easily settle at the baseline, suggesting that the sensor is highly selective for the detection of formaldehyde.

The interference study was conducted in an aqueous medium; therefore, the investigation focused on selected inorganic ions that are commonly present as dissolved species in aqueous samples and may influence the analytical response of the Ni(SALBSH)₂-PVA/Chi probe. The selectivity toward structurally related organic compounds, including other aldehydes, was not evaluated in this study. Consequently, the reported selectivity is limited to the inorganic ions investigated, and the assessment of potential cross-reactivity with other aldehydes remains the subject of future work.

Table 1. Interference effect of various ions on Ni(SALBSH)₂-PVA/Chi Probe M1 in detection of formaldehyde.

Ions	% relative error			
	*Ratio	1:10	1:1	10:1
Fe ³⁺		-101.99	-115.29	-480.92
K ⁺		-318.78	-229.02	-215.45
Na ⁺		-492.85	-477.68	-236.25
NH ₄ ⁺		-242.30	-200.78	-261.74
CO ₃ ²⁻		-75.07	+30.34	+143.47
$*Ratio = \frac{\text{Concentration of formaldehyde (mgL}^{-1}\text{)}}{\text{Concentration of foreign ions (mgL}^{-1}\text{)}}$				

Table 2. Determination of formaldehyde in fish using Ni(SALBSH)₂-PVA/Chi Probe.

Fish samples	Concentration of formaldehyde added (mgL ⁻¹)	Concentration of formaldehyde found (mgL ⁻¹)	Recovery (%)
Red snapper	0	0.31	0.66
	100	0.98	
Toli shad	0	0.45	0.28
	100	0.72	

The sensor performance was evaluated in real sample analysis using the standard addition method. The results are presented in Table 2. The recovery percentages obtained were very low (< 1%) which is parallel to the highly interference effect. Therefore, the proposed sensor is best suited for the direct detection of formaldehyde in aqueous solutions. For real samples, an appropriate extraction procedure must be conducted prior to the application of the sensor.

CONCLUSION

This study demonstrates that the Ni(SALBSH)₂ complex was successfully immobilised onto a PVA/Chitosan membrane (Ni(SALBSH)₂-PVA/Chi probe) and exhibited a good response to formaldehyde in aqueous solution. The presence of formaldehyde decreased the absorbance intensity of the Ni(SALBSH)₂-PVA/Chi probe at 326 nm. The linearity of the probe response against formaldehyde was in the range of 20 – 500 mgL⁻¹ with a detection limit of 2.52 mgL⁻¹. Due to the high interference by common ions in real samples, the proposed sensor is highly recommended for the individual detection of formaldehyde in aqueous media and is not applicable to complex matrices.

ACKNOWLEDGEMENTS

The authors would like to extend their gratitude to the Ministry of Higher Education of Malaysia for their kind contribution of research grant FRGS-

EC/1/2024/STG04/UPSI/02/5 for financial support in this research.

REFERENCES

- de Boer, J., Schösler, H. and Aiking, H. (2020) Fish as an alternative protein – A consumer-oriented perspective on its role in a transition towards more healthy and sustainable diets. *Appetite*, **152**, 104721 (April).
- Zhao, L. G., Sun, J. W., Yang, Y., Ma, X., Wang, Y. Y. and Xiang, Y. B. (2016) Fish consumption and all-cause mortality: A meta-analysis of cohort studies. *European Journal of Clinical Nutrition*, **70**(2), 155–161.
- Kundu, A., Dey, P., Bera, R., Sarkar, R., Kim, B., Kacew, S., Lee, B. M., Karmakar, S. and Kim, H. S. (2020) Adverse health risk from prolonged consumption of formaldehyde-preserved carps in eastern region of Indian population. *Environmental Science and Pollution Research*, **27**(14), 16415–16425.
- Ding, H., Yuan, G., Peng, L., Zhou, L. and Lin, Q. (2020) TP-FRET-Based Fluorescent Sensor for Ratiometric Detection of Formaldehyde in Real Food Samples, Living Cells, Tissues, and Zebrafish. *Journal of Agricultural and Food Chemistry*, **68**(11), 3670–3677.

5. Jaman, N., Sazedul Hoque, M., Enamul Hoq, M., Md Sazedul Hoque, C., Chandra Chakraborty, S. and Pada Seal, H. (2015) Determination of formaldehyde content by spectrophotometric method in some fresh water and marine fishes of Bangladesh. *International Journal of Fisheries and Aquatic Studies*, **2**(6), 94–98.
6. Ma, Y., Zhao, C., Zhan, Y., Li, J., Zhang, Z. and Li, G. (2015) Separation and analysis of trace volatile formaldehyde in aquatic products by a MoO₃/polypyrrole intercalative sampling adsorbent with thermal desorption gas chromatography and mass spectrometry. *Journal of Separation Science*, **38**(8), 1388–1393.
7. Wang, X., Si, Y., Mao, X., Li, Y., Yu, J., Wang, H. and Ding, B. (2013) Colorimetric sensor strips for formaldehyde assay utilizing fluoral-p decorated polyacrylonitrile nanofibrous membranes. *Analyst*, **138**(17), 5129–5136.
8. Pontel, L. B., Rosado, I. V., Burgos-Barragan, G., Garaycochea, J. I., Yu, R., Arends, M. J., Chandrasekaran, G., Broecker, V., Wei, W., Liu, L., Swenberg, J. A., Crossan, G. P. and Patel, K. J. (2015) Endogenous Formaldehyde Is a Hematopoietic Stem Cell Genotoxin and Metabolic Carcinogen. *Molecular Cell*, **60**(1), 177–188.
9. Cole, P., Adami, H. O., Trichopoulos, D. and Mandel, J. S. (2010) Re: Mortality from lymphohematopoietic malignancies and brain cancer among embalmers exposed to formaldehyde. *Journal of the National Cancer Institute*, **102**(19), 1518–1519.
10. Rovina, K., Vonnice, J. M., Shaeera, S. N., Yi, S. X. and Halid, N. F. A. (2020) Development of biodegradable hybrid polymer film for detection of formaldehyde in seafood products. *Sensing and Bio-Sensing Research*, **27**, 100310.
11. European Food Safety Authority (EFSA) Panel on Additives and Products or Substances Used in Animal Feed (FEEDAP) (2014) *Scientific opinion on the safety and efficacy of formaldehyde for all animal species based on a dossier submitted by Adiveter S.L.* *EFSA Journal*, **12**(2), 3562.
12. Fappiano, L., Carrieria, F., Iannone, A., Notardonato, I. and Avino, P. (2022) A Review on Recent Sensing Methods for Determining Formaldehyde in Agri-Food Chain: A Comparison with the Conventional Analytical Approaches. *Foods*, **11**, 1351, 2 – 18.
13. Safavi, A., Ahmadi, R. and Ramezani, A. M. (2018) Vortex-assisted liquid-liquid micro-extraction based on hydrophobic deep eutectic solvent for determination of malondialdehyde and formaldehyde by HPLC-UV approach. *Microchemical Journal*, **143**, 166–174.
14. Gu, D. C., Liu, W., Yan, Y., Wei, W., Gan, J. Hong, Lu, Y., Jiang, Z. L., Wang, X. C. and Xu, C. H. (2019) A novel method for rapid quantitative evaluating formaldehyde in squid based on electronic nose. *Lwt-Food Science and Technology*, **101**, 382–388.
15. Wahed, P., Razzaq, M. A., Dharmapuri, S. and Corrales, M. (2016) Determination of formaldehyde in food and feed by an in-house validated HPLC method. *Food Chemistry*, **202**, 476–483.
16. Yeh, T. S., Lin, T. C., Chen, C. C. and Wen, H. M. (2013) Analysis of free and bound formaldehyde in squid and squid products by gas chromatography-mass spectrometry. *Journal of Food and Drug Analysis*, **21**(2), 190–197.
17. Zhang, Z., Zhao, C., Ma, Y. and Li, G. (2013) Rapid analysis of trace volatile formaldehyde in aquatic products by derivatization reaction-based surface enhanced Raman spectroscopy. *Analyst*, **139**(14), 3614–3621.
18. Kucukoglu, K., Gul, H. I., Taslimi, P., Gulcin, I. and Supuran, C. T. (2019) Investigation of inhibitory properties of some hydrazone compounds on hCA I, hCA II and AChE enzymes. *Bioorganic Chemistry*, **86**, 316–321.
19. Anbazhagan, R. and Sankaran, K. R. (2015) Design, synthesis, computational calculation and biological evaluation of some novel 2-thiazolyl hydrazones. *Spectrochimica Acta - Part A: Molecular and Biomolecular Spectroscopy*, **135**, 984–993.
20. Secci, D., Carradori, S., Petzer, A., Guglielmi, P., D'Ascenzio, M., Chimenti, P., Bagetta, D., Alcaro, S., Zengin, G., Petzer, J. P. and Ortuso, F. (2019) 4-(3-Nitrophenyl)thiazol-2-ylhydrazone derivatives as antioxidants and selective hMAO-B inhibitors: synthesis, biological activity and computational analysis. *Journal of Enzyme Inhibition and Medicinal Chemistry*, **34**(1), 597–612.
21. Tan, L., Guo, J. and Wang, N. (2013) A new rhodamine B hydrazide hydrazone derivative for colorimetric and fluorescent “off-on” recognition of copper(II) in aqueous media. *Bulletin of the Korean Chemical Society*, **34**(4), 1159–1162.
22. Juahir, Y., Subramaniam, P., Abdulla, M. A., Robinson, W. T. and Ali, H. M. (2009) Synthesis, Characterization and X-ray Structure of 2,6 diacetylpyridinebis(benzenesulfonylhydrazide)

- and its Zinc (II) Complex. *Journal of Chemical Crystallography*, **39**, 615 – 618.
23. Lira, R. S., Silva, M. A. and Santos, D. O. (2025) Synthesis and characterization of novel hydrazone complexes: Exploring DNA/BSA binding and antimicrobial potential. *ACS Omega*, **10**(6), 6123–6135.
 24. Kalla, R. M., Reddy, V. K. and Prakash, O. (2024) Synthesis, characterization, density functional theory studies and antioxidant activity of novel hydrazone ligands and their metal complexes. *Oriental Journal of Chemistry*, **39**(6), 1540–1549.
 25. Juahir, Y., Ali, H. M., Puvaneswary, S., Robinson, W. T. and Ng, S. W. (2008) Bis{2-bromo-4-chloro-6-[2-(phenylsulfonyl)hydrazonomethyl]phenolato- κ^2N, O^1 }copper(II). *Acta Crystallographica Section E: Structure Reports Online*, **E64**(8), 1039.
 26. Firmansyah, A., Arianingrum, R. and Handayani, S. (2023), Synthesis of Schiff base ligand based on 2-aminobenzothiazole and 4-hydroxybenzaldehyde and their nickel complexes. *Indonesian Journal of Chemistry*, **23**(5), 1668–1679.
 27. Vennila, P., Ahmed, S. B., Sumathi, P., Siva, V., Rajendran, P. and Kamal, C. (2024) Synthesis and spectroscopic characterization of Schiff base metal complexes, biological activity, and molecular docking studies. *ACS Omega*, **9**(12), 14210–14222.
 28. Kadir, M. F. Z., Aspanut, Z., Majid, S. R. and Arof, A. K. (2011) FTIR studies of plasticized poly(vinyl alcohol)-chitosan blend doped with NH_4NO_3 polymer electrolyte membrane. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **78**(3), 1068–1074.
 29. Amaregouda, Y. and Kamanna, K. (2023) Preparation and characterization of indicator films from chitosan/polyvinyl alcohol incorporated *Stachytarpheta jamaicensis* anthocyanins for monitoring chicken meat freshness. *Sustainable Food Technology*, **1**(5), 738-749.
 30. Wardhono, E. Y., Pinem, M. P., Susilo, S., Siom, B. J., Sudrajad, A., Pramono, A., Meliana, Y. and Guénin, E. (2022) Modification of Physio-Mechanical Properties of Chitosan-Based Films via Physical Treatment Approach. *Polymers*, **14**(23), 5216.
 31. Korusenko, P. M., Petrova, O. V., Vereshchagin, A. A., Nekipelov, S. V., Sivkov, D. V., Sivkov, V. N., Katin, K. P., Levin, O. V. and Vinogradov, A. S. I. (2023) A Comparative XPS, UV PES, NEXAFS, and DFT study of the Electronic Structure of the Salen Ligand in the H₂(Salen) Molecule and the [Ni(Salen)] Complex. *International Journal of Molecular Science*, **24**, 9868, 2 – 21.
 32. Cheedarala, R. K. and Song, J. I. (2021) Moderately transparent chitosan-PVA blended membrane for strong mechanical stiffness and as a robust bio-material energy harvester through contact-separation mode TENG. *Frontiers in Nanotechnology*, **3**, Article 667453.
 33. Steinegger, A., Wolfbeis, O. S. and Borisov, S. M. (2020) Optical sensing and imaging of pH values: Spectroscopies, materials, and applications. *Chemical Reviews*, **120**(24), 12357–12489.
 34. Shahvar, A., Saraji, M. and Shamsaei, D. (2018) Headspace single drop microextraction combined with mobile phone-based on-drop sensing for the determination of formaldehyde. *Sensors and Actuators, B: Chemical*, **273**, 1474–1478 (July).
 35. Chaiendoo, K., Sooksin, S., Kulchat, S., Promarak, V., Tuntulani, T. and Ngeontae, W. (2018) A new formaldehyde sensor from silver nanoclusters modified Tollens' reagent. *Food Chemistry*, **255**, 41–48.
 36. Skoog, D. A., Holler, F. J. and Crouch, S. R. (2018) Principles of Instrumental Analysis (7th ed.), Cengage Learning.
 37. Harris, D. C. and Lucy, C. A. (2020) Quantitative Chemical Analysis (10th ed.), W. H. Freeman and Company / Macmillan Learning.
 38. Rouessac, F. and Rouessac, A. (2013) Chemical Analysis: Modern Instrumentation Methods and Techniques (2nd ed.), John Wiley and Sons.
 39. Ghaedi, M., Naghiha, R., Karami, B., Asfaram, A. and Ghaedi, A. M. (2014) Preparation and evaluation of a new optical sensor for determination of trace amount of copper(II) ions in water and real samples. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **132**, 280–288.
 40. Mishra, Y. K., Kaushik, A. and Singh, A. (2024) Advanced materials for optical sensing and biomedical applications: Leaching kinetics and matrix stability in optical platforms. *Materials Advances*, **5**(15), 6346–6350
 41. Colon, B. A., Hassan, M. R., Saleheen, A., Baker, C. A. and Calhoun, T. R. (2020) Total Internal Reflection Transient Absorption Microscopy: An Online Detection Method for Microfluidics. *Journal of Physical Chemistry A*, **124**(20), 4160–4170.

146 Wan Rusmawati Wan Mahamod, Yusnita Juahir, Norhayati Hashim, Nurusaidah Abdul Rahim, Maizatul Najwa Jajuli, Norlaili Abu Bakar, Nurul Hana Baharom and Norlin Suhaiza Musali

New Optical Sensing Reagent of Bis(2-oxidobenzylidene)benzenesulfonohydrazide Nickel(II) for the Detection of Formaldehyde in Fish

42. Jokić, I., Djurić, Z., Radulović, K., Frantlović, M., Milovanović, G. V. and Krstajić, P. M. (2021). Stochastic Time Response and Ultimate Noise Performance of Adsorption-Based Microfluidic Biosensors. *Biosensors*, **11**(6), 194.
43. Soto, R. J., Hall, J. R., Brown, M. D., Taylor, J. B. and Schoenfisch, M. H. (2016) In Vivo Chemical Sensors: Role of Biocompatibility on Performance and Utility. *Analytical Chemistry*, **89**(1), 276–299.
44. Tan, L. L., Musa, A. and Lee, Y. H. (2011) Determination of ammonium ion using a reagentless amperometric biosensor based on immobilized alanine dehydrogenase. *Sensors*, **11**(10), 9344–9360.
45. Hayun, H., Harmita, K. and Pramudita, T. B. (2017) Determination of Formaldehyde Content

- in Wet Noodles by Thin Layer Chromatography-Densitometry After Derivatization With Nash Reagent. *Oriental Journal of Chemistry*, **33**(3), 1400-1405.
46. Rungroadsri, J., Limsakul, W., Wongniramaikul, W. and Choodum, A. (2017) Rapid Semi-Quantitative Analysis of Formaldehyde in Food by Digital Image Colorimetry. *International Journal of Chemical Engineering and Applications*, **8**(4), 294–298.
47. Ehsan, M. A. and Rehman, A. (2020) Facile and scalable fabrication of nanostructured nickel thin film electrodes for electrochemical detection of formaldehyde. *Analytical Methods*, **12**(32), 4028–4036.