

*Dedicated to My Beloved*

*Parents*

*Yumnam Shanta Devi & Yumnam Yaima Singh*

*and*

*My Supervisor*

*Prof. Poonam Mishra*

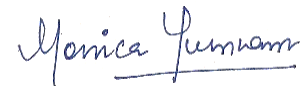
## Declaration by the Candidate

---

The thesis entitled “**Design and Development of a Smartphone Sensing System for Determination of Freshness of Fish fillets During Storage**” is being submitted to *School of Engineering, Tezpur University* in partial fulfillment for the award of the degree of *Doctor of Philosophy* in the *Department of Food Engineering and Technology* is a record of bonafide research work accomplished by me under the supervision of **Prof. Poonam Mishra**.

All help from various sources have been duly acknowledged.

No part of the thesis has been submitted elsewhere for the award of any other degree.



**Monica Yumnam**

**FEP18107**

Department of Food Engineering and Technology

School of Engineering, Tezpur University

Tezpur, Assam- 784028 (India)

Date: 17.10.2024

Place: **Tezpur**



**Department of Food Engineering and Technology**  
**Tezpur University**  
**(A Central University)**  
**Napaam, Tezpur - 784028, Assam**

Dr. Poonam Mishra  
Professor

Email: [poonam@tezu.ernet.in](mailto:poonam@tezu.ernet.in)  
Phone: 91(03712)-27-5707

---

**Certificate of the Supervisor**

This is to certify that the thesis entitled “**Design and Development of a Smartphone Sensing System for Determination of Freshness of Fish fillets During Storage**” submitted to the **School of Engineering**, Tezpur University in partial fulfillment for the award of the degree of Doctor of Philosophy in **Food Engineering and Technology** is a record of research work carried out by **Ms. Monica Yumnam** under my supervision and guidance.

All help received by her from various sources have been duly acknowledged.

No part of this thesis has been submitted elsewhere for award of any other degree.

**(Poonam Mishra)**

Date: 17.10.2024

Place: **Tezpur**

## Acknowledgments

---

As I reach the end of my Ph.D. journey, I am overwhelmed with gratitude for all the support, guidance, and encouragement that I have received along the way. This thesis is the result of years of hard work and dedication, as well as the combined efforts of numerous remarkable individuals.

First and foremost, I would like to express my heartfelt gratitude to my supervisor, Prof. Poonam Mishra, Department of Food Engineering and Technology, Tezpur University, Tezpur, Assam, whose unwavering support, insightful feedback, and continuous encouragement helped shape this thesis. I am deeply grateful for her love, care, and valuable recommendations throughout the entire process, from beginning to end. Her passion for research and dedication to excellence have inspired me to strive for the highest standards in my work.

I express my deep sense of gratitude to Hon. Vice-Chancellor Prof. Shambhu Nath Singh, Tezpur University, Tezpur, Assam for providing the opportunity to carry out the thesis work. I am profoundly grateful to Prof. Partha Pratim Sahu, Dean of the School of Engineering, Prof. Sankar Chandra Deka, Controller of Examinations, and Prof. Laxmikant S. Badwaik, Head, Department of Food Engineering and Technology, Tezpur University, for their unwavering support throughout the thesis.

With a sense of gratitude and great pleasure, my thanks go to the esteemed members of my Doctoral Committee, Prof. Manuj Kumar Hazarika, Department of Food Engineering and Technology, Prof. Pabitra Nath, Prof. Dambarudhar Mohanta, Department of Physics, Tezpur University, Tezpur, Assam, for their valuable suggestions and encouragement at various stages of investigation and thesis writing.

Heartfelt thanks are due to the members of the Departmental Research Committee for extending all sorts of help and guidance throughout my research work. I acknowledge Prof. Charu Lata Mahanta, Prof. Brijesh Srivastava, Prof. Nandan Sit, Dr. Nishant R. Swami Hulle, Dr. Tabli Ghosh, Dr. Nickhil C, Dr. Soumya Ranjan Purohit, Dr. Amit Baran Das, the faculty members of the Department of Food Engineering and Technology and External Committee

---

members, Prof. Tarun K. Maji, Department of Chemical Sciences and Prof. Dilip Dutta, Department of Mechanical Engineering, Tezpur University, Assam for their constructive reviews and encouragement throughout my research.

Thanks are due to the technical staff, Dr. Arup Jyoti Das, Dr. Dipankar Kalita, Mr. Bhaskar Jyoti Kalita, Mr. Labadeep Kalita, and office staff, Mr. Krishna Borah and Mr. Anjan Keot of the Department for providing the necessary help and assistance in carrying out my research work in the Department of Food Engineering and Technology, Tezpur University, Tezpur, Assam.

I wish to express my sincere thanks to Mr. Wungshim Zimik, Department of Food Engineering and Technology, Dr. Diganta Hatiboruah, Department of Physics, Tezpur University, and Mr. Rahul Mishra, Department of Electronic and Communication, University of Allahabad, for their assistance and support in completing my research.

I would like to acknowledge the funding sources that made this research possible. The support from LSRB-DRDO (Life Science Research Board- Defence Research and Development Organisation) (Sanction No.: 314) and Tezpur University in terms of Research and Innovation grant (File no. DoRD/RIG/10-73/1544-A) has been essential in enabling to pursue my research.

I would like to express my heartfelt gratitude to my loving lab mates (Ms. Manisha Medhi, Dr. Arun Kumar Gupta, Ms. Parismita Koch, Mr. Subhamoy Dhua, Ms. Krishna Gopalakrishnan, Ms. A. Sangeeta, Ms. Mridusmita Burman, and Mr. Keshram Dulait), and my juniors for their continuous support throughout my research.

To my colleagues and friends, Ms. Sonam Kumari, Ms. Ruchi Rani, Ms. Lourebam Monika Devi, Ms. Arjuara Begum, Ms. Tridisha Bordoloi, Ms. Fogila Begum, Mr. Amardeep Kumar, Ms. Nongmaithem Sofia, Ms. Somya Singhal, Ms. Indrani Chetia, who supported me and making this journey enjoyable, the countless discussion and moments of laughter that we shared. Special thanks to Mr. Satyabanta Singh Laishram for his constant love and support and for being there throughout this journey.

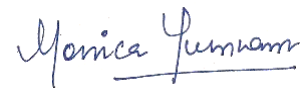
I want to thank everyone who has supported me in some way, whether directly or indirectly, and whose names have not been stated explicitly.

A heartfelt thank you to my family for their unconditional love and encouragement. To my parents, Yumnam Yaima Singh and Yumnam Shanta Devi, your belief in me has been a constant source of strength. To my, elder sister (Yumnam Smeeta John), brother-in-law (Arvind John), elder brother (Yumnam Ranju Singh), sister-in-law (Yumnam (O) Vama Devi), elder brother (Yumnam Mahesh Singh), younger brother (Yumnam Mukesh Singh), and sister-in-law (Yumnam (O) Lalleima Chanu) for their love, understanding, and unwavering support have been my anchor through the highs and lows of this journey.

I would like to acknowledge the unwavering love and support of my close friends and family, who have kept me going throughout my Ph.D. program.

To everyone who has been a part of this journey, thank you for your contributions and for believing in me.

Above all, I am thankful to “The God Almighty” who has showered his blessings on me to complete my PhD thesis.



Monica Yumnam

17<sup>th</sup> Oct 2024

## List of Tables

<b>Table No.</b>	<b>Title</b>	<b>Page No.</b>
2.1.	Typical composition of fish (%) (Edible portion)	7
2.2.	Criteria of EU scheme	11
2.3.	Consumer QIM for assessment of raw whole non-specific fish species (adapted from Hyldig et al. 2004)	12
2.4	Summary of the recent smartphone-based sensor used for the detection of fish freshness	40-43
5.1.	The linearity and the uncertainty of the calibration curve	112
5.2.	Percent (%) Recovery for ammonia solution using the developed smartphone sensor	113
6.1.	Estimated ammonia concentration (ppm) of the Rohu, Mullet, and Common carp fish stored at ambient temperature using the smartphone sensor and spectrophotometer	127
6.2.	Estimated ammonia concentration (ppm) of the Rohu and Common carp fish stored at refrigeration temperature using the smartphone sensor and spectrophotometer	128
7.1.	Chemical compounds of pomelo peel essential oil	152
7.2.	Zone of inhibition	156
7.3.	Log CFU/g for 15 days of storage of essential oil-treated Rohu, Bahu, and Silver carp fish fillets	162

## List of Figures

Figure No.	Title	Page No.
2.1.	Schematic diagram of the internal anatomy of fish. (Source: Wikipedia [ <a href="https://en.wikipedia.org/wiki/Fish_anatomy">https://en.wikipedia.org/wiki/Fish_anatomy</a> ])	7
2.2.	Mechanism of fish spoilage	9
2.3.	The diagnostic adapter for on-site marine toxins created by Fang et al. (2016)	24
2.4.	(i) A portable electrochemical platform deployed for point-of-use analyses; (ii) Smartphonescreenshots showing the Tongue Metrix Android App- Initial screen, (iii) typical cyclic voltammogram for the Fe (CN) <sub>6</sub> <sup>4-/3-</sup> redox probe, (iv) optional data transmission by email, and (v) interface screen for PCA treatment (adapted from Giordano et al., 2016)	29
2.5.	Smartphone-based Labelled and Label-free mode dual-sensing system or onsite determination of Aflatoxin B1 (AFB1) contamination in foodstuffs (adapted from Ma et al., 2023)	31
2.6.	(i) Schematic of TVB-N sensing for fish freshness monitoring by the smartphone-integrated hydrogel sensing platform, (ii) Real-time monitoring of fish freshness stored at 37, 25, and 4 °C, respectively by the sensing hydrogel with color evolution photographs as a function of time, and corresponding ratio value changes of R/(R + G + B), and (iii) The changes of TVB-N detected by the hydrogel sensing method and the semimicro-Kjeldahl method of fish (grass carp, crucian carp, and grouper) stored at 37 °C and correlation of the results between the semimicro-Kjeldahl method and the hydrogel sensing method in monitoring the	35



---

	freshness of three kinds of fish. Reproduced with permission (adapted from Zhang et al., 2021)	
<b>2.7.</b>	(i) A schematic illustration of the color measurement technique, (ii) The calibration curve with 90% and 95% confidence intervals for the NH <sub>3</sub> analyte with the corresponding color changes and changes in the value of DE at different g with increasing time, and (iii) Color changes of the black carrot extract (BCE) in solutions with increasing pH (adapted from Mastnak et al., 2023)	36
<b>2.8.</b>	(i) A schematic illustration of the color measurement technique, (ii) The calibration curve with 90% and 95% confidence intervals for the NH <sub>3</sub> analyte with the corresponding color changes and changes in the value of DE at different g with increasing time, and (iii) Color changes of the black carrot extract (BCE) in solutions with increasing pH (adapted from Mastnak et al., 2023)	36
<b>2.9.</b>	Schematic illustration of the working mechanism and application of smartphone-adaptable fluorescent sensing tag for monitoring the freshness of fish stored under different conditions (adapted from Jiang et al. 2022)	39
<b>2.10.</b>	Application of the ratiometric fluorescent sensing label based on aggregation-induced emissive (AIE) polymers in salmon samples, and a conceptual design of smartphone-based reader and smartphone applications. Reproduced with permission (adapted from Liu et al. 2022)	39
<b>2.11.</b>	Keyword network analysis (KNA) of smartphone-based sensors application in fish spoilage	44
<b>3.1.</b>	Synthesis of PANI and PANI label	67
<b>3.2.</b>	PANI label exposed to ammonia vapor	68

---

---

<b>3.3.</b>	PANI labels exposed to different concentrations of ammonia vapor	68
<b>3.4.</b>	Tilapia ( <i>Oreochromis nilotica</i> ) fish	69
<b>3.5.</b>	PANI label exposed to fish fillets	69
<b>3.6.</b>	Mechanism of PANI synthesis and its forms	73
<b>3.7.</b>	Fourier Transform Infrared (FTIR) spectra of aniline and PANI	73
<b>3.8.</b>	XRD of PANI	74
<b>3.9.</b>	FESEM image of PANI label deposited with 1 mL (A) 10,000X, (B) 20,000X, (C) 30,000X, and (D) 40,000X	74
<b>3.10.</b>	Response of fresh and exposed PANI label to 400 ppm ammonia vapor (a) 0.2 mL PANI deposited, (b) 0.5 mL PANI deposited, and (c) 1 mL PANI deposited	75
<b>3.11.</b>	Response time of PANI label with 1 mL (0.05 g) of PANI deposition	75
<b>3.12.</b>	Calibration curve of PANI label response using UV-visible spectrophotometer	76
<b>3.13.</b>	Spectrophotometer response of Tilapia fish, and (b) Percent (%) intensity reduction at ambient room temperature	77
<b>3.14.</b>	Percent (%) intensity reduction for spectrophotometer response of Tilapia fish at refrigeration temperature	77
<b>3.15.</b>	TVB-N value of the fish sample at ambient room temperature	79
<b>3.16.</b>	pH and TVC (total viable count) of fish sample at ambient room temperature	80

---

---

<b>3.17.</b>	TVB-N value of fish sample at refrigeration temperature	80
<b>3.18.</b>	pH and TVC (total viable count) at refrigeration temperature	81
<b>3.19.</b>	Pseudomonas count of fish sample at refrigeration temperature	81
<b>4.1.</b>	BPD label exposed to different concentrations of ammonia vapor	89
<b>4.2.</b>	(a) Rohu (Labeorohita), and (b) BDP label exposed to fish fillets	90
<b>4.3.</b>	Response label toward ammonia vapor (0-400 ppm) using UV-vis spectrophotometer (a) 0.2 mL dye deposition, (b) 0.5 mL dye deposition, and (c) 1 mL dye deposition	92
<b>4.4.</b>	(a) Response time of dye label, and (b) Calibration curve of dye label with ammonia vapor using UV-vis spectrophotometer.	92
<b>4.5.</b>	Percent (%) intensity reduction for spectrophotometer response of Rohu fish at ambient room temperature	94
<b>4.6.</b>	TVB-N value of Rohu fish sample at ambient temperature	96
<b>4.7.</b>	TVC (total viable count) and pH value of Rohu fish sample at ambient temperature	96
<b>4.8.</b>	Leaching test of the developed labels	97
<b>5.1.</b>	Schematic diagram of smartphone sensing system	105
<b>5.2.</b>	3D design drawing of the cradle set up using Google SketchUp 2020	105

---

---

<b>5.3.</b>	(a) Process diagram of the optical-based smartphone sensing system, and (b) Cradle setup with optical component integrated into the camera of the smartphone	106
<b>5.4.</b>	Spectral image of red and blue laser light obtained using the developed sensor	107
<b>5.5.</b>	Spectral image of PANI label exposed to ammonia vapor obtained using the developed sensor and the spectral data extracted using ImageJ software	108
<b>5.6.</b>	Pixels were converted to wavelength	111
<b>5.7.</b>	Linear relationship between pixel and calibrated wavelength	111
<b>5.8.</b>	Calibration curve of PANI label response using smartphone-based sensor	112
<b>6.1.</b>	(a) Rohu ( <i>Labeo rohita</i> ), (b) Mullet ( <i>Mugil cephalus</i> ), and (c) Common Carp ( <i>Cyprinus carpio</i> )	119
<b>6.2.</b>	Graphical representation of developed smartphone-based sensor equipped with PANI sensor for determination of fish spoilage	120
<b>6.3.</b>	The PANI label regeneration process	122
<b>6.4.</b>	PANI label response using smartphone sensor	123
<b>6.5.</b>	(a) Percent (%) intensity reduction of smartphone sensor response, and (b) Percent (%) intensity reduction of spectrophotometer response at ambient room temperature	126
<b>6.6.</b>	(a) Percent (%) intensity reduction of smartphone sensor response, and (b) Percent (%) intensity reduction of spectrophotometer response	126

---

---

	(a) TVB-N value, (b) pH value, and (c) Total viable count (TVC) of three different varieties of fish fillet stored at ambient temperature	131
<b>6.7.</b>		
	(a) TVB-N value, (b) pH value, and (c) Total viable count (TVC) of three different varieties of fish fillet stored at ambient temperature	132
<b>6.8.</b>		
	Texture profile analysis of (A) Hardness, (B) Springiness, (C) Cohesiveness, and (D) Gumminess of Rohu, Mullet, and Common carp fish fillets during storage at ambient temperature	134
<b>6.9.</b>		
	Texture profile analysis of (A) Hardness, (B) Springiness, (C) Cohesiveness, and (D) Gumminess of Rohu and Common carp fish fillets during storage at refrigeration temperature	135
<b>6.10.</b>		
	Response of PANI label stored at three different relative humidity (40- 60 %, 70 %, and 85 % and at ambient room temperature (ART) and refrigeration temperature (RT)	136
<b>6.11.</b>		
	Regeneration study of PANI label.	136
<b>6.12.</b>		
	Process of web application utilization	137
<b>6.13.</b>		
	(a) Rohu ( <i>Labeo rohita</i> ), (b) Bahu ( <i>Labeo calbahu</i> ), and (c) Silver carp ( <i>Hypophthalmichthys molitrix</i> )	148
<b>7.1.</b>		
	Pre-treatment of fish with pomelo peel essential oil	148
<b>7.2.</b>		
	Antimicrobial properties of Essential oil (From 1 to 8: <i>Candida albicans</i> , <i>Listeria monocytogenes</i> , <i>Pseudomonas aeruginosa</i> , <i>Mycobacterium smegmatis</i> , <i>Staphylococcus aureus</i> , <i>Bacillus cereus</i> , <i>Escherichia coli</i> , and <i>Yersinia pestis</i> )	155
<b>7.3.</b>		

---

---

<b>7.4.</b>	Percent (%) intensity reduction of smartphone sensor response, (a) Rohu fish fillet, and (b) Common carp fish fillet	157
<b>7.5.</b>	Percent (%) intensity reduction of spectrophotometer response, (a) Rohu fish fillet, and (b) Common carp fish fillet	158
<b>7.6.</b>	(a) TVB-N value of essential oil treated and untreated Rohu fish fillet, (b) TVB-N value of essential oil treated and untreated Common carp fish fillet, (c) pH value of essential oil treated and untreated Rohu fish file, and (d) pH value of essential oil treated and untreated Common carp fish fillet	160
<b>7.7.</b>	Total viable count (TVC) of essential oil treated and untreated (a) Rohu fish fillet, (b) Common carp fish fillet, (c) Pseudomonas count of treated and untreated Rohu fish fillet, and (d) Pseudomonas count of treated and untreated Common carp fish fillet	163
<b>7.8.</b>	Texture profile analysis of (a) Hardness, (b) Springiness, (c) Cohesiveness, and (d) Gumminess of essential oil treated and untreated Rohu fish fillets during storage at refrigeration temperature	165
<b>7.9.</b>	Texture profile analysis of (a) Hardness, (b) Springiness, (c) Cohesiveness, and (d) Gumminess of essential oil treated and untreated Common carp fish fillets during storage at refrigeration temperature	166

---

## List of Abbreviations

<b>Abbreviations</b>	<b>Full Form</b>
<b>AF</b>	Aflatoxin
<b>ATP</b>	Adenosine Triphosphate
<b>Au-SPE</b>	Gold Screen Printed Electrode
<b>BPB</b>	Bromophenol Blue
<b>CCD</b>	Charge Coupled Device
<b>CMOS</b>	Complementary Metal Oxide Semiconductor
<b>CSA</b>	Colorimetric Sensor Array
<b>DFT</b>	Density functional theory
<b>DHA</b>	Docosahexaenoic Acid
<b>DMA</b>	Dimethylamine
<b>EC</b>	Electrochromic
<b>EEM</b>	Excitation Emission Matrics
<b>EIS</b>	Electrochemical Impedance Spectroscopy
<b>EPA</b>	Eicosapentaenoic Acid
<b>EU</b>	Union Scheme
<b>FAO</b>	Food and Agriculture Organization
<b>FBN</b>	Formalin-bound Nitrogen
<b>FESEM</b>	Field Emission Scanning Electron Microscopic
<b>FFA</b>	Free Fatty Acid
<b>FT-IR</b>	Fourier Transform Infrared
<b>FTO</b>	Fluorine-doped tin oxide
<b>GC-MS</b>	Gas chromatography Mass Spectrometry
<b>GOI</b>	Government of India
<b>GRAS</b>	Generally Recognized as Safe
<b>HIS</b>	Histamine
<b>IOPG</b>	Inverse Opal Photonic Gel
<b>IoT</b>	Internet of Things
<b>LED</b>	Light Emoting Diode
<b>MIP</b>	Molecularly Imprinted Polymer
<b>MSME</b>	Ministry of Micro, Small, Medium Enterprises
<b>NiNPs</b>	Nickel Nanoparticles

---

<b>OA</b>	Okadaic Acid
<b>PANI</b>	Polyaniline
<b>PET</b>	Polyethylene Terephthalate
<b>PMMSY</b>	Pradhan Mantri Matsya Sampada Yojana
<b>POC</b>	Point of Care
<b>PUFA</b>	Polyunsaturated Fatty Acid
<b>PV</b>	Peroxide value
<b>QIM</b>	Quality Index Method
<b>RGB</b>	Red Green Blue
<b>RGO</b>	Reduced Graphene Oxide
<b>SBCA</b>	Smartphone Based Colorimetric Analyzer
<b>SDAE-NN</b>	Double Sacked Deionizing Autoencoder
<b>SPBS</b>	Smartphone Based Sensor
<b>STX</b>	Saxitoxin
<b>TBA</b>	Thiobarbituric Acid
<b>TCA</b>	Trichloroacetic acid
<b>TMA</b>	Trimethylamine
<b>TMAO</b>	Trimethylamine Oxide
<b>TPC</b>	Total Plate Count
<b>TVB-N</b>	Total Volatile Basic Nitrogen
<b>VOCs</b>	Volatile Organic Compounds
<b>XO</b>	Xanthine Oxidase
<b>XPD</b>	X-ray diffraction
<b>Zn-TPP</b>	Zinc Tetraphenylporphyrin
<b>LSPR</b>	Localized Surface Plasma Resonance
<b>HPLC</b>	High Performance Liquid Chromatography
<b>ML</b>	Machine learning
<b>XOD</b>	Xanthine Oxidase
<b>AIE</b>	Aggregation Induced Emissive
<b>PMAA</b>	Poly Methacrylic Acid
<b>TPE</b>	Tetraphenylethylene
<b>EMI</b>	Electromagnetic Interference
<b>LOD</b>	Limit of Detection

---



---

<b>LOQ</b>	Limit of Quantification
<b>PLA</b>	Polylactic Acid
<b>ISE</b>	Ion Selective Electrode
<b>RH</b>	Relative Humidity
<b>LB</b>	Luria Broth
<b>PDB</b>	Potato Dextrose Broth
<b>PDA</b>	Potato Dextrose Agar
<b>LBA</b>	Luria Broth Agar
<b>TVC</b>	Total Viable Count
<b>TPC</b>	Total Plate Count
<b>BPD</b>	Bromocresol Purple Dye
<b>SD</b>	Standard Deviation
<b>RSD</b>	Relative Standard Deviation
<b>ZOI</b>	Zone of Inhibition
<b>WHO</b>	World Health Organization

---