National Institute of Science,
Technology and Innovation

STI Focus

SCIENCE, TECHNOLOGY AND INNOVATION

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FOREWORD

Science, Technology, and Innovation (STI) continue to serve as fundamental drivers of national development under the Royal Government of Cambodia's Pentagonal Strategy Phase I. In alignment with these priorities, the Ministry of Industry, Science, Technology & Innovation (MISTI) and the National Institute of Science, Technology and Innovation (NISTI) remain steadfast in their commitment to fostering a knowledge-based economy through research, capacity building, and public dissemination of scientific information. STI Focus is an integral mechanism supporting this mission, providing a trusted platform for sharing scientific evidence and advancing STI awareness across all sectors.

This edition presents key research findings that reflect both the growing capabilities of Cambodian researchers and the expanding relevance of STI in national policymaking, industrial development, and societal progress. The contributions in this volume highlight the essential role of science and innovation in enhancing productivity, strengthening competitiveness, and ensuring the Kingdom's sustainable and inclusive growth.

I extend my sincere appreciation to the authors whose expertise and dedication have enriched this publication. Their work not only broadens public understanding but also inspires emerging researchers, students, and practitioners to contribute to the advancement of STI in Cambodia. Continued engagement from the scientific community, including both established scholars and emerging talents, is vital to realizing our national vision for a resilient, innovative, and prosperous Cambodia.

Phnom Penh, January 5, 2026

Minister

HEM Vanndy

EDITORIAL NOTE

With great excitement, we present STI Focus's latest issue for the year 2025. which brings together studies from both the Scientific Findings and Technology Trends. This issue reflects the increasing commitment of Cambodian researchers to addressing practical challenges through scientific inquiry, technological innovation, and evidence-based approaches. Although the topics span diverse disciplines, they collectively demonstrate how STI supports community needs and contributes to national development priorities.

The Scientific Findings section covers several important areas: food safety, quality and food development, biotech and engineering, as well as digital, environmental, and geospatial technologies. Readers will find studies on Molecular Genetic Identification of *Escherichia coli* O157:H7 Isolated from Fresh Vegetables in Phnom Penh, Knowledge and Consumption of Ultra Processed Food and Their Health Impact Among University Students, Leveraging Hyperspectral Imagery Data for Vegetation Analysis in Advancing Agriculture Practices, Organic Waste as a Source of Bioactive Compounds: Extraction and Evaluation of Their Capacity by Vitro Test, Evaluation of Cherry Tomato Shelf Life Stored in Different Types of Packaging, and Base Formula Development of Syrup from Tomatoes and Jam from Tomato.

The Technology Trends section introduces Converting Agricultural Waste into High-Quality Compost Using Lactic Acid Bacteria and CRISPR/Cas9 Technology in Biomedical Science. The explanations are presented in an accessible manner, making the content suitable for a broad readership while still offering meaningful insights. We extend our sincere appreciation to all contributing authors for their dedication, time, and expertise. Their continued participation ensures that STI Focus remains a valuable platform for promoting scientific knowledge, encouraging research collaboration, and strengthening Cambodia's STI ecosystem.

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SCIENTIFIC FINDINGS

1

Molecular Genetic Identification of *Escherichia coli* O157:H7 Isolated from Fresh Vegetables in Phnom Penh

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Highlight

- Fresh-eaten vegetables are one of the main sources of infection if contaminated with pathogenic microorganisms.
- This study aims to identify *Escherichia coli (E. coli)* O157:H7 in cucumber, saw leaf, and lettuce (curly-leafy vegetable) collected from different markets in Phnom Penh.
- Lettuce samples were more contaminated with pathogenic *E. coli* than saw leaf and cucumber ones.

Molecular Genetic Identification of *Escherichia coli* O157:H7 Isolated from Fresh Vegetables in Phnom Penh

Figure 1. Microbial contamination of fresh vegetables.

1. Introduction

The role of fresh vegetables in nutrition and healthy diets is well recognized and has undertaken various initiatives to encourage consumers to eat more of these products (FAO, 2020). Meanwhile, the Interagency Food Safety Analytics Collaboration released a report which outbreaks data to update previous analyses to estimate which foods are responsible for illness related to foodborne pathogens. Highlights of the findings noted in the report including *E. coli* O157:H7 illnesses were most often linked to fresh vegetables such as leafy greens (Gdoura et al., 2018). Bacteria as well as other parasites may infect vegetables during planting, growth, transportation, and up to post-harvest or unhygienic cooking (De Silva et al., 2014). Interestingly, there is rarely a report associated with microbial contamination in local foods, whereas chemical contaminations associated with the use of insecticides (Neufeld et al., 2010). Therefore, it is very important to investigate the contamination by pathogenic strain of *E. coli* in vegetables, if not in all food sources. Our study aims to isolate DNA and to identify *E. coli* O157:H7 from fresh vegetables including cucumber, saw-leaf herb, and lettuce from different markets in Phnom Penh using Polymerase Chain Reaction (PCR) technique.This research will raise the awareness of probable microbial contaminates thus reducing adverse health impacts.

2. Materials and Methods

2.1. Bacterial DNA Preparation

All vegetable samples were collected and performed bacterial isolation between July and September in 2017 (Phoeurk et al., 2019). Bacterial culture stocks were preserved as glycerol stocks and stored at -20 °C. Forty-two bacterial stocks were inoculated for DNA isolation where its protocol was adapted and modified from Puregene and Fanglian methods (He, 2011). The quality of obtained DNA samples was analyzed by agarose gel electrophoresis, 1% agarose gel was pre-casted with Gel RED stain and visualized under the UV light.

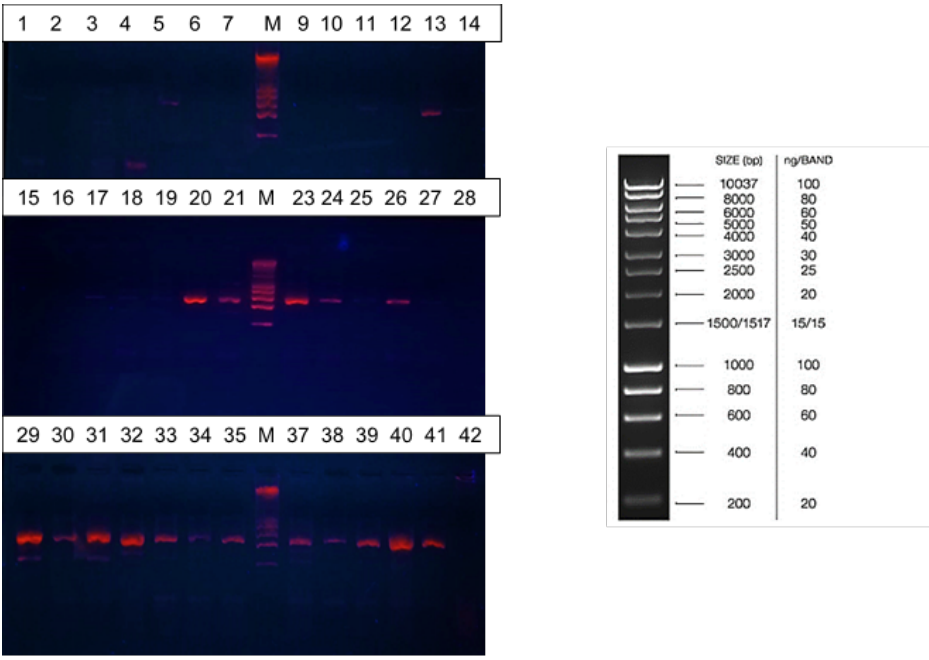

Figure 2. PCR product results of *E. coli* O157:H7 from tested fresh vegetable samples. The left-hand side picture illustrates PCR product of *E. coli* O157:H7 using stx gene with the expected size 513bp, while the right-hand side picture demonstrate the base pairs of the used DNA marker. There are 45 lanes include 42 samples and 3 markers in the middle (lane M). The lane 1-14 are the PCR product from cucumber samples in different markets, lane 15-28 are from saw leaf samples, and lane 29-42 are from lettuce samples.

2.2 Polymerase Chain Reaction

The protocol was followed to the BIOLINE manufacturer's instruction. A 0.1µl isolated DNA was used as the template for PCR. Primers for the specific and its PCR conditions are listed in the Table 1. The amplified PCR products were analyzed by gel electrophoresis where 2% agarose gel was used.

Table 1. The stx primers and its PCR conditions

Gene	Primer Sequences	Size	PCR Condition
Shiga toxin gene of *E. coli*	• *stx*-F: 5'-CAGTTAATGTGGTTGCGAAG-3' • *stx*-R: 5'-CTGTCACAGTAACAAACCGT-3'	513bp	• 1×94℃ for 5min; • 35× (94℃ for 1min, 64℃ for 30s, 72℃ for 1 min); • 1×72℃ for 7 min

3. Results and Discussion

The target stx gene amplicon, with the size of 513 base pairs using stx primer(s) to identify pathogenic *E. coli* O157:H7 that is virulence factor for human health, of the forty-two selected samples from cucumber, lettuce, saw leaf was shown as in Figure 1. The result showed that, none of 14 cucumber samples from different market sites (from lane 1 to 14) was positive, while 5 of 14 saw leaf samples (35%), and 12 of 14 lettuce samples (85%) were presented with the specific target virulence gene (positive) of *E. coli* O157:H7. Therefore, the leafy vegetables (lettuce and saw leaf herb) showed a higher bacterial contamination than fruit vegetable, cucumber. The results from our study are consistent to other previous studies that found the leafy vegetable is the most bacterial contamination due to it has larger surface than fruit vegetable, which is a good position for bacteria to stay there (Pang et al., 2017).

The vegetables in these selected markets are sold mostly in the open area with warm temperature and humid. These practices expose the vegetables to open environment thus may be easily contaminated by airborne pathogens. In addition, all the selected vegetables in this study are mostly consumed as fresh by Cambodian people (Mosimann et al., 2023). The high prevalence of bacterial contamination demonstrated in both of curly and flat surfaces; therefore, it is important to wash them with the clean water more carefully and repeatedly (Santos et al., 2010). Phoeurk et al., 2019 reported that washing vegetables (lettuce, saw-leaf herbs, and cucumber) with tap water can significantly reduce and eliminate the number of bacteria, particularly *E. coli.* Furthermore, the molecular technique (PCR) that use to complete main objectives in this study is an appropriate culture method in identifying virulence factor of bacteria with lower cost than other techniques (Gurtler et al., 2013).

4. Conclusion

In conclusion, our study found that the contamination of pathogenic *E. coli* O157:H7 occurred mostly in the leafy vegetables, lettuce samples and saw-leaf herbs. The wider surface area of leafy vegetables the higher possibility of bacterial contamination. Even the fruity vegetables are less contaminated; it is still in need to wash with clean water to remove both microbes and chemicals in prior to eating. Washing with tap water is highly recommended to remove biological contaminants as well as bacteria from the fresh vegetables effectively.

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2

Knowledge and Consumption of Ultra Processed Food and Their Health Impact Among University Students

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Highlight

- Ultra-processed foods (UPFs) are products made with multiple ingredients, including salt, sugar, oils, fats, and additives like flavorings, colorings, sweeteners, and emulsifiers, to mimic the taste and appearance of natural foods.
- UPFs are very popular because these foods are tasty, easy to eat and cheap.
- high UPF consumption significantly increased the risk of diabetes, cardiovascular disease, depressive symptoms, and cancer.

Knowledge and Consumption of Ultra Processed Food and Their Health Impact Among University Students

1. Introduction

Currently, approximately 2 billion adults worldwide are overweight or obese, a dramatic increase from just over 100 million in 1975. This rise has been largely attributed to a dietary shift from minimally processed foods (MPFs) toward more ultra-processed foods (UPFs) (Dicken et al., 2023). UPFs are highly popular due to their convenience, palatability, and affordability, leading to widespread consumption. At the same time, the intake of fresh fruits and vegetables has declined. Many students, in particular, favor UPFs because they are quick to eat, require little preparation, and are budget-friendly, especially when they are busy with school or other activities. In addition to personal preferences, environmental and social factors also influence the frequent consumption of UPFs among students (Juliana Sarmiento-Santos et al., 2022).

Ultra-processed foods (UPFs) are formulation made from multiple ingredients, often including salt, sugar, oils, fats, flavorings, colorings, sweeteners, emulsifiers, and other additive designed to mimic the sensory qualities of unprocessed or minimally processed foods. Common examples include soft drinks, instant noodles, packaged snacks, and fast foods. The growing global population has increased the demand for food, prompting the development of many food products with extended shelf lives (Machín et al., 2016). According to findings from the NutriNet-Santé study, a large prospective cohort study in France, indicates that high UPF consumption is significantly associated with increased risk of diabetes, cardiovascular disease, depressive symptoms, and cancer (Pagliai et al., 2020). Despite these risks, some students may be unaware of the potential negative health effects of UPFs or may underestimate their harm. Therefore, this study aims to investigate the knowledge and frequency of UPF consumption among university students as well as identifying the main factors that influence students' choices to consume ultra-processed foods.

2. Methodology

2.1 Study Design

The understanding of the term UPF was explored through Google form questionnaires by face-to-face interaction. Convenience sampling method was chosen in this study. The convenience sampling is a non-probability sampling technique where participants are selected because they are easy to access, readily available, or willing to participate (Etikan, I., 2016). A total of 100 participants from University of Puthisastra (UP) were randomly selected and surveyed by google form. In addition, no students were excluded from the data analysis.

2.2 Study Questionnaire

The study questionnaire consists of four parts. The first part collected general information, including participants' name, age, gender, major, year of study, and people who live with. The second part assessed the knowledge of ultra-processed food, including misconceptions, awareness about what ultra-processed food is, ingredients and examples of health impacts. The third part showed participants' consumption habits of ultra-processed food, including opinions, feelings, personal beliefs about UPFs, convenience, testing and danger. The final part focused on participants' health effects of consuming ultra-processed food, including frequency of UPFs consumption, eating habits, food choices in daily life, the situation where UPFs are eating and whether students try to avoid them or not. The questionnaires were developed based on a thorough literature review, and before conducting interviews, they were pre-tested to ensure clarity and effectiveness.

2.3 Study Limitation

This study has some limitations that should be acknowledged. First, the sample size was relatively small and limited to 100 participants from the University of Puthisastra, which may not fully represent the broader student population or other universities. Since convenience sampling was used, the findings may be subject to selection bias and lack generalizability. Therefore, the results should be interpreted with caution, and further research with larger, more diverse, and randomly selected samples is recommended to enhance the validity and applicability of the findings.

2.4 Ethical Considerations and Consent

All information was kept in private and will be removed after the three years of research are up. Every information was kept on a computer and wasn't allowed a third party access. All participants were provided a consent form and they are free to decline not to answer the question if they felt uncomfortable with it. Also, participants had an option to leave the study at any time.

2.5 Data Analysis

The collected data was analyzed using descriptive statistics to summarize participants' knowledge of ultra-processed food. Statistical tools like Microsoft Excel was used to calculate the frequency, create graphs, and present the date in percentages.

3. Results and Discussion

3.1 General Information of Participants

General information of participants shown in Table 1. showed a total of 100 participants took part in the study, of whom 69 (69%) were female and 31 (31%) were male. In terms of age, the largest group was 18–20 years old, accounting for 67% of the sample, with 51 females (51%) and 16 males (16%). Participants aged 21–23 years represented 30%, consisting of 16 females (16%) and 14 males (14%). Only a small number were aged 24–26 years (2%), with all being female, while 1% were aged 27–29 years, represented solely by one male participant. No participants were aged 30 years or older.

Regarding their field of study, the highest proportion were enrolled in Medicine (23%), including 12 females (12%) and 11 males (11%). Dentistry accounted for 20% of participants, equally divided between females (10%) and males (10%). Pharmacy comprised 18%, with a predominance of females (16%) compared to males (2%). Science Research represented 13%, including 10 females (10%) and 3 males (3%). Nursing accounted for 12% of participants, mostly female (10%) with only 2% male. Laboratory Science contributed 7%, with 5% female and 2% male. Smaller groups were observed in Midwifery (3% female only), IT (1% female and 2% male), and Business (1% female only).

In terms of living arrangements, most participants lived with family (68%), consisting of 50 females (50%) and 18 males (18%). A total of 22% reported living alone, including 12 females (12%) and 10 males (10%), while 10% lived with friends, of whom 3% were female and 7% were male.

Table 1. General Information of Participants

Characteristics	Frequency (N=100)	Female (N%)	Male (N%)
Gender	100	69 (69%)	31 (31%)
Age group			
18-20	67	51 (51%)	16 (16%)
21-23	30	16 (16%)	14 (14%)
24-26	2	2 (2%)	0 (0%)
27-29	1	0 (0%)	1 (1%)
30-other	0	0 (0%)	0 (0%)
Major of study			
Medicine	23	12 (12%)	11 (11%)
Pharmacy	18	16 (16%)	2 (2%)
Dentistry	20	10 (10%)	10 (10%)
Nursing	12	10 (10%)	2 (2%)
Science Research	13	10 (10%)	3 (3%)
Laboratory Science	7	5 (5%)	2 (2%)
Midwife	3	3 (3%)	0 (0%)
Business	1	1 (1%)	0 (0%)
IT	3	1 (1%)	2 (2%)
Students live with			
Family	68	50 (50%)	18 (18%)
Friends	10	3 (3%)	7 (7%)
Alone	22	12 (12%)	10 10%)

3.2 Student's Knowledge and Consumption Habit of UPFs

The findings of this study revealed important insights into students' awareness and consumption patterns of ultra-processed foods (UPFs) as showed in Table 2. A majority of the participants (70%) reported having heard of UPFs, with female students (49%) demonstrating higher awareness compared to their male counterparts (21%). However, 30% of students, equally distributed between genders, had not heard of UPFs, highlighting that a significant proportion of students still lack familiarity with the concept.

With regard to the sources of information, social media and the internet were identified as the dominant channels (45%), particularly among female students (30%). In contrast, formal sources such as universities and academic literature accounted for only 11%, while family and friends contributed minimally (6% and 8%, respectively). This indicates that students predominantly rely on informal and digital sources for knowledge about UPFs, rather than structured educational platforms.

In terms of consumption frequency, the majority of students (61%) consumed UPFs two to three times per week, with females (45%) consuming more frequently than males (16%). About 28% of participants consumed UPFs once per week, while 9% reported daily consumption, all of whom were female. Interestingly, only 2% of male participants stated that they never consumed UPFs. These results suggest that UPFs are a regular part of students' diets, with females showing a higher frequency of intake compared to males.

The timing of consumption further reflects lifestyle-related eating behaviors. Evening consumption was predominant (63%), particularly among females (47%), followed by afternoon consumption (26%). Morning (5%) and late-night (6%) consumption were reported less frequently, though late-night eating was more common among females (5%) compared to males (1%). This pattern indicates that UPFs are often consumed as snacks or convenient alternatives to main meals, particularly in the evening (Monteiro CA et al.,2019).

Overall, the results demonstrate that students are widely aware of UPFs but primarily informed through non-academic sources. Consumption is frequent, with the majority eating UPFs at least two to three times per week, and females consistently reporting higher levels of awareness and intake compared to males. The preference for evening consumption suggests that UPFs may serve as accessible and convenient food choices during students' daily routines.

Table 2. Student's consumption habit of UPFs

		Female (%)	Male (%)	Total (%)
Heard of ultra-processed foods	Yes	49%	21%	70%
	No	15%	15%	30%
Source get the information of ultra-processed food	Family	4%	2%	6%
	Friends	6%	2%	8%
	Social media/ internet	30%	15%	45%
	University/ literature	9%	2%	11%
	Other source	0%	1%	1%
Eat ultra-processed food in a week	1 time	17%	11%	28%
	2-3 times	45%	16%	61%
	4-daily	9%	0%	9%
	Never	0%	2%	2%
Usually, eat ultra-processed foods	Morning	3%	2%	5%
	Afternoon	14%	12%	26%
	Evening	47%	16%	63%
	Late night	5%	1%	6%

3.3 Types of UPFs Consumed

Figure 1. showed the frequency of consumption of different types of ultra-processed foods (UPFs) among participants. The findings indicate that **instant noodles (26 respondents, 26%)** and **snacks (25 respondents, 25%)** were the most frequently consumed UPFs, followed closely by **fast food (20 respondents, 20%)** and **sweetened drinks (18 respondents, 18%).** In contrast, fewer participants reported consuming **fried food (6 respondents, 6%), meatballs/hot dogs (10 respondents, 10%),** and items categorized as **other (6 respondents, 6%).** Only **2 respondents (2%)** reported not consuming any UPFs.

Overall, the results highlight that the majority of participants regularly consumed UPFs, with a clear preference for instant noodles, snacks, and fast food, while healthier patterns such as abstaining from UPFs were rare. This suggests that UPF consumption is prevalent and may represent a significant dietary concern for the studied group.

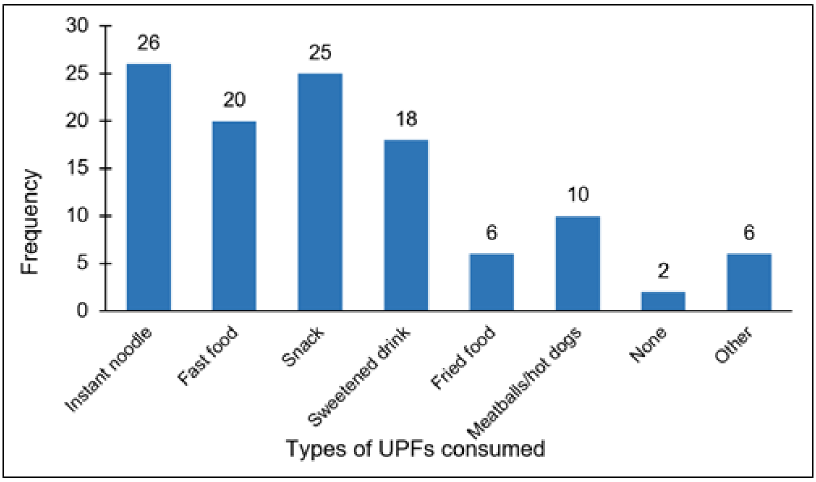

Figure 1. The frequency of UPFs commonly consumed by students

3.4 Student's knowledge and health effect perception of UPFs

Table 3. The relation between students who had heard or not about UPFs and the number of correct answers in the questionnaire

Have you heard of UPFs?		N of correct answers in questionnaires	Total
Yes	No	MCQs/ True or false	
0	1	1	1
3	1	2	4
5	2	3	7
14	5	4	19
23	11	5	34
21	4	6	25
4	1	7	5

By using Pearson's correlation coefficient, the relation between students who had heard or not about UPFs and the number of correct answers were a low-level correlation (r=0.091) with (*p*-value = 0.368). This shows that even though students who reported that they knew about UPFs, their score doesn't match their knowledge as show in Table 3. (Sarmiento-Santos et al., 2022). In addition, the perception from the students about the health effects of consuming UPFs, most of the students mentioned that consuming UPFs would lead into diabetes (n=32), noticeably overweight (n=16), heart disease (n=15), obesity (n=13), high blood pressure (n=11) and cancer (n=10) as show in Figure 2.

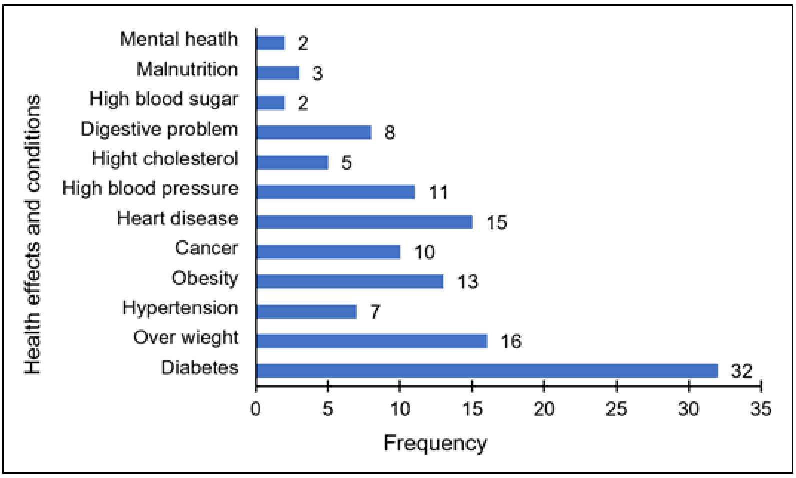

Figure 2. Health effect of consuming UPFs

3.5 Factors and Perception of Participants on the Effects of UPFsConsumption

Reasons of consuming UPFs: Among those who have known the health effects of UPFs but still chosen to eat was 66% (Female 48% and male 18%). The main reason of choosing UPFs is due to its good in taste with 35% individuals (Female 21% and male 9%). Convenience and quickness were the second factor of consuming the UPFs with 22% individuals (Female 16% and male 6%). Moreover, easiness to access was another factor including 5% individuals (female 4% and male 1%). Along with that, 2% individuals (Female 0% and male 2%) chose UPFs because they were cheap in price and also 2% individuals (Female 2% and male 0%) chose UPFs because they could be stored for long periods (**Table 4**).

Table 4. Factors of choosing UPFs for consumption

		Female (%)	Male (%)	Total (%)
Know about the effects of consuming UPFs	Yes	48%	18%	66%
	No	21%	13%	34%
Prefer to eat ultra-processed foods if knew the health effect	Taste good	26%	9%	35%
	Convenient and quick	16%	6%	22%
	Can store for long periods	2%	0%	2%
	Cheap price	0%	2%	2%
	Easy to access	4%	1%	5%

4. Conclusion

This study revealed that while the majority of students demonstrated awareness of ultra-processed foods (UPFs) (70%) and their potential health effects (66%), a considerable proportion still lacked sufficient knowledge. Despite this awareness, high consumption patterns were observed, with 61% of students consuming UPFs 2–3 times per week and 9% consuming them daily. Taste (35%) and convenience (22%) emerged as the primary factors influencing UPF consumption, followed by affordability and accessibility. These findings highlight a critical gap between knowledge and behavior, underscoring the need for effective health promotion strategies to encourage healthier dietary choices. Raising awareness of the risks associated with UPFs can play a vital role in guiding students toward more nutritious and balanced eating habits.

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3

Leveraging Hyperspectral Imagery Data for Vegetation Analysis in Advancing Agriculture Practices

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Highlight

- Hyperspectral imaging (HSI) is a cutting-edge technology that captures detailed spectral information across a wide range of wavelengths, offering insights into plant health, nutrient status, and crop stress.
- Vegetation Indices derived from hyperspectral data, such as NDVI, ARI, PRI, and WI are critical tools for monitoring crop health, growth stages, and stress, enabling early detection of problems and better farm management.

Leveraging Hyperspectral Imagery Data for Vegetation Analysis in Advancing Agriculture Practices

1. Introduction

HSI was considered as a fast and nondestructive sensor for early diagnosis plant health, which has revolutionized agricultural monitoring by providing crucial data for assessing vegetation health and productivity (Wan et al., 2022). The improving of crop monitor, detection potential issues early, and plant pest managements on time are crucial for minimizing crop damage and yield lose and increasing production (Lucas, 2011). Regarding to Lu et al. (2020), the remote sensing provides an effective approach to monitor variations of changes in crop morphology and physiology, offering valuable support for precision farming practices. This review article exposes the significance of vegetation indices, various sources of imagery data, and their applications in modern agriculture, along with the challenges and future perspectives of this innovative approach.

2. HSI Procedures

The procedures of HSI involves capturing hyperspectral images across the entire crop and subsequently processing these images to retrieve hundreds of spectral bands. This workflow generates a substantial and highly informative dataset that enables comprehensive assessment of crop health and supports advanced analytical applications (Figure 1).

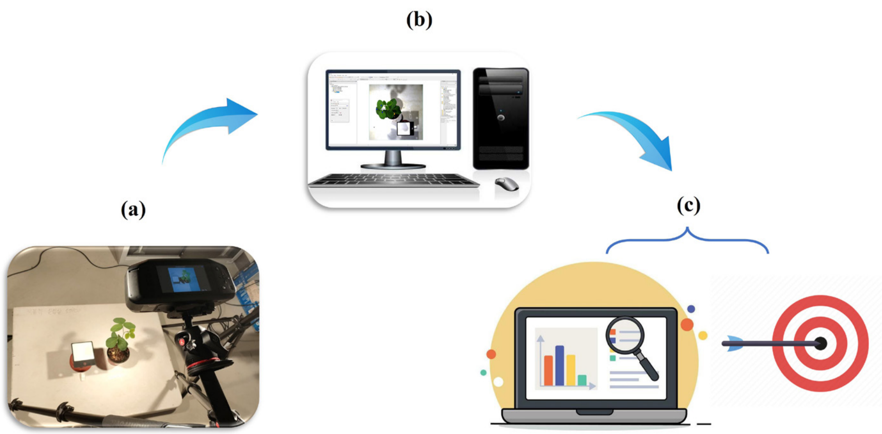

Figure 1. Working flow (a) image capture by hyperspectral camera, (b) image annotation and data extraction in ENVI software, and (c) data processing and decision management

2.1. Hyperspectral Imaging

The disease diagnosis was performed by using hyperspectral cameras/sensors based on Specim's push-broom technology. The cameras have the capability to take 2D images with a contiguous spectral wavelength range of 400 – 1000 nm, allowing detailed hyperspectral data collection from the plants (Figure 2). It offers detailed invisible information on plant physiology, chlorophyll content, and nutrient status. Compared to multispectral imaging, hyperspectral data provides enhanced differentiation between healthy and stressed crops, enabling precise decision making in agricultural practices.

Figure 2. Using hyperspectral camera to capture leaf area

2.2. Image Processing in ENVI Software

The key statistical hyperspectral image features were computed for analysis: 204 different spectral bands and the entire wavelength range from 400 to 1000 nm. These features were computed using ENVI V.5.5.3 software from Research System, Inc. in the USA.

2.2.1. Image Annotation in ENVI Software

To ensure the accuracy of each plant's assessment, a region of interest (ROI) was carefully annotated within every hyperspectral image of plant leaves. Essentially, this ROI selection process pinpointed specific hyperspectral wavelength data required to establish reference information for disease detection (Figure 3).

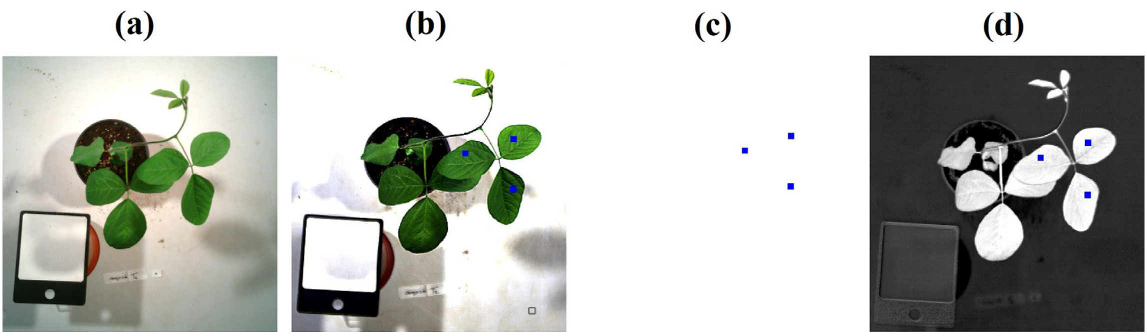

Figure 3. Annotation of the regions of interest (ROI). (a) Original image, (b) selection region of interest (ROI) annotation using ENVI software, (c) ROI image, and (d) image after analysis

2.2.2. Vegetation Indices Measurement in ENVI Software

Vegetation indices (VIs) are mathematical combinations of spectral bands derived from remote sensing data, used to quantify various characteristics of vegetation and assess plant health (Skendži et al., 2023). Some applicable formulas used hyperspectral vegetation indices in ENVI software include in (table 1).

Normalized difference vegetation index (NDVI) is a widely used in vegetation index that measures the greenness and health of vegetation (Chung et al., 2018). It's calculated using the reflectance of red and near-infrared (NIR) bands from remote sensing data. The range value for NIR is the reflectance in the near-infrared wavelength 859 and Red is the reflectance in wavelength 649.

Anthocyanin reflectance index (ARI) is used to detect the presence of anthocyanin pigments in vegetation, which are associated with red or purple coloring in plants (Feng et al., 2022 & Gitelson et al., 2001). The wavelength range for R700 is the reflectance in the 700nm (red-edge) and R550 is the reflectance in the 550nm (yellow). ARI values can help identify the presence of anthocyanin-rich vegetation, which is often associated with stress or specific stages in the plant's lifecycle.

The Photochemical reflectance index (PRI) is an index that provides information about the physiological status of plants, particularly the efficiency of photosynthesis (Mulero et al., 2023). It's based on reflectance in the green and red wavelengths. The wavelength range for R531 is the reflectance in the 531nm (green) and R570 is the reflectance in the 570nm (red). PRI values can vary and are typically interpreted in the context of the specific study and the type of vegetation being observed.

The Water index (WI) is used to evaluate water content, which exposes of plant hydration status and stress level (Sun et al., 2019). The wavelength range for R900 is the reflectance at 900 nm (near-infrared, sensitive to vegetation biomass and structure), while R970 is the reflectance at 970 nm (a strong water absorption band).

Table 1. The applicable formulas of vegetation indices calculation

No	VIs	Formula	Stressed Value	Application
1	NDVI	(NIR - Red) / (NIR + Red)	< 0.50	Drought stress Nutrient deficiency Pest detection Disease detection
2	ARI	(1/R550) - (1/R700)	> 0.06	Disease detection
3	PRI	(R531 - R570) / (R531 + R570)	< 0.00	Heat stress Disease detection
4	WI	(R900) / (R970)	< 0.95	Water stress Water content Drought stress

The data obtained from the applicable formulas in ENVI software were used to conduct a comparison between stressed value and average of vegetation indices parameters. Based on Figure 4, the sample of NDVI of selected ROI on detected plant showed the average lower than stress value in Day 2 to Day 4. Regarding to this data extraction and analysis process, hyperspectral imagery provides researchers and farmers with actionable insights for monitoring crop status, detecting stress early, and optimizing management practices.

3. Concluding Remarks and Future Perspective in Cambodia

Hyperspectral imaging (HSI) provides a promising approach for modernizing agriculture in Cambodia by detecting early changes in crop physiology, timely diagnosis of pest outbreaks, nutrient deficiencies, and stress conditions. However, some challenges remain in this advancement including the initial cost of hyperspectral camera/sensor, accessible software, and affordable satellite data in increasing attainable for local applications. Encouraging collaboration among policymakers, university levels, stakeholders, and farmers accelerate adoption technology for improving sustainable agriculture and optimizing food security in Cambodia.

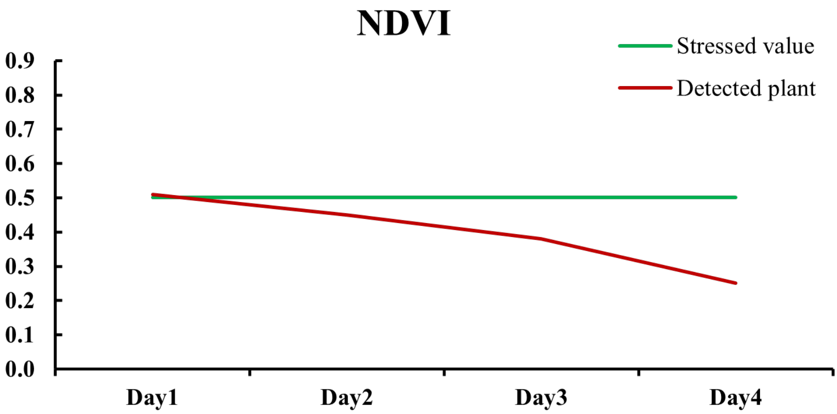

Figure 4. The effect of stress on NDVI from Day 1 to Day 4 of detected plant

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4

Organic Waste as a Source of Bioactive Compounds: Extraction and Evaluation of Their Capacity by Vitro Test

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Highlight

- Citrus peel and lemongrass leaves are sustainable sources of bioactive compounds with antioxidant activity.
- Extraction of bioactive compounds adds economic value and supports sustainable waste management.
- Orange peel oil had a sweet, fruity aroma, while lemongrass oil exhibited a fresh fragrance with pinene and terpinene.
- Lemongrass leaves and orange peels exhibited strong antioxidant activity, with DPPH scavenging above 80%.

Organic Waste as a Source of Bioactive Compounds: Extraction and Evaluation of Their Capacity by Vitro Test

1. Introduction

The growth of food processing and agricultural businesses has created a large volume of organic waste, such as fruit peels, seeds, vegetable residues, and fermentation by-products. Among these residues, citrus fruit waste and lemongrass leaves stand out as promising, sustainable sources of bioactive compounds with diverse biological activities. Bioactive compounds are mostly specific secondary metabolites with antioxidant, inflammatory, immunomodulative potential, antimicrobial properties, etc (Sytar & Smetanska, 2022).

Oranges are among the most widely cultivated fruit crops in the world. Approximately 70% of the oranges produced are generated into juice, marmalade and other foods, resulting in citrus peel waste that accounts for roughly 50–60% of the fruit being processed (Zema et al., 2018). Orange peel waste was found to contain a higher amount of essential oil d-limonene compared to other lignocellulose feedstocks (Tahir et al., 2023). Lemongrass leaves, on the other hand, is an another aromatic plant. Its leaves contain phenolic compounds that exhibit antioxidant activity, along with alkaloids, terpenoids, flavonoids, saponins, and tannins. Citral is identified as the main active compound(Falah et al., 2015). The positive sensory effects of flavor, aroma, and color make peel and lemongrass oil products a desirable ingredient in various industries, such as food, perfume, and pharmaceuticals.

Extracting bioactive compounds from organic waste not only added economic value but also supports sustainable waste management and environmental protection (Dable-Tupas et al., 2022). Moreover, research of their antioxidant abilities in the laboratory provide key a deep understanding of their potential uses for preserving food and creating natural health products and supplements (Generalić Mekinić & Šimat, 2025; Sorrenti et al., 2023). Therefore, objectives of this study were to (1) extract the essential oils and bioactive compounds from orange peels and lemongrass leave and (2) evaluate the antioxidant activity of the extracted bioactive compounds.

2. Material and Method

2.1 Sample Collection and Preparation

Fresh lemongrass leaves at a maturity stage of 4–6 months were collected from local farmers in Areykhsat, Kandal Province. Citrus peels (specifically from Pursat and Krasaing oranges) were obtained from local street vendors located along the main roads near Kramounh Market, Orussey Market, and Phsar Chas in Phnom Penh. The samples were thoroughly washed with clean tap water to remove dirt, cut into small pieces, and dried for further analysis.

2.2 Essential Oil Extraction

Two hundred grams (200 g) of citrus peels and lemongrass leaves were placed into a round-bottom flask and mixed with 400 mL of distilled water. The samples were then extracted at 80 ℃ for 80 minutes. The solution obtained from the extraction was separated using a separatory funnel to remove the excess distilled water. The extracted oil was wrapped in aluminum foil to prevent deterioration and stored in a refrigerator at 4 ℃

until further study (Golmohammadi et al., 2018).

2.3 Determination of Total Phenolic Content

The determination of total phenolic content was carried out by following the method from Xu et al. (2007). Citrus peel and lemongrass leaves were dried using a freeze dryer. The dried samples were then ground into a fine powder using a grinder, followed by sieving through a 500 µm mesh to ensure uniform particle size. Ten grams of the dried powder were mixed with 100 mL of 80% ethanol. The mixture was stirred using an IKA magnetic stirrer at 500 rpm for 6 hours at ambient temperature. After stirring, the mixture was centrifuged at 4000 rpm for 15 min. The supernatant was collected and evaporated to remove ethanol. After that, the sample was dried using freeze dryer.

First, 300 µL of a sample was mixed with 900 µL of distilled water, followed by the addition of 300 µL of 1 N Folin–Ciocalteu reagent and then mixing. After 5 min, 1500 µL of 8% (w/v) sodium carbonate (Na_2CO_3) and 1500 µL of distilled water were added, and the mixture was mixed well. The reaction mixture was then kept in the dark for 30 minutes and measured using a UV–visible spectrophotometer at 765 nm. The total phenolic content was expressed as gallic acid equivalents (mg GAE/g dry mass).

2.4. Antioxidant Activity of Citrus Peel and Lemongrass Samples

2.4.1 Radical DPPH Scavenging Activity (2,2-diphenyl-1-picrylhydrazyl assay)

Forty-five milligrams of the extract were dissolved in 1 mL of distilled water (DW). Then, 1 mL of this solution was mixed with 2 mL of DPPH radical scavenger solution (0.1 mM in methanol), thoroughly mixed, and incubated in the dark at room temperature for 30 minutes. The absorbance of the reacted solution was then measured using a UV–visible spectrophotometer at 517 nm. Trolox was used as a standard equivalent.

Percentage of inhibitory sample (PI%) = $\frac{(\text{Absorbance of blank} - \text{absorbance of sample}) \times 100}{\text{Absorbance of blank}}$

2.5 Statistical Analysis

Experiments were performed in triplicate and results were expressed as mean ± standard deviation (Mean ± SD). The combined data were analyzed using one-way analysis of variance (ANOVA) to determine significant differences among sample means at $p < 0.05$, with Tukey's post-hoc test applied for multiple comparisons.

3. Result and Discussion

3.1 Yied of Essential Oil

After extraction, orange peel essential oil exhibited a sweet and fruity aroma, whereas lemongrass oil showed a fresh as shown in Figure 1, pleasant fragrance with distinct pinene and terpinene notes. Citrus peels from Pursat and Krassaing yielded 4.94 g (2.47%) and 3.56 g (1.78%) of oil, respectively, while lemongrass leaves produced 0.69 g (0.35%). The higher yield from orange peel likely reflects the greater concentration of essential oils in the peel compared to the leaves and stems of lemongrass (Mukarram et al., 2021; Vasquez-Gomez et al., 2024). (Coulibaly et al., 2024)reported that lemongrass oil yields range from 0.032% to 0.50%, whereas orange peel yields are approximately 4.23% using distillation. Differences in essential oil concentrations between studies may result from factors such as harvesting period, fruit ripeness, climate conditions, extraction methods, and equipment quality.

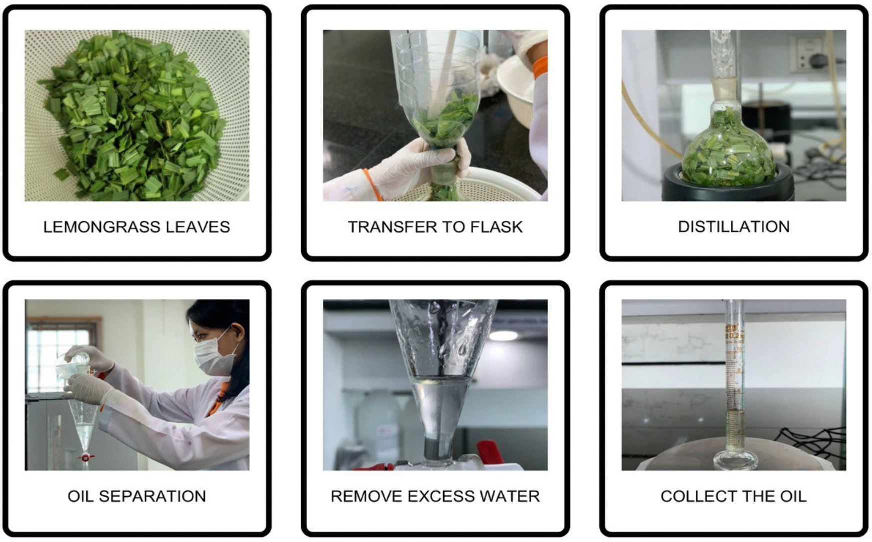

Figure 1. Process of essential oil extraction

3.2 Total Phenolic Content and Antioxidant Activity

The concentration of total phenolic has been calculated from the equation as shown in Figure 2 and 3. The results of this study showed that the total phenolic content in the samples was 1.4, 1.2, and 0.6 mg/100 g (dry weight) for Pursat orange peel, Krasaing orange peel, and lemongrass leaves, respectively. In particular, the total phenolic content of orange peels and lemongrass can also vary depending on the extraction method, and the maturity stage of the selected samples. The percentage of free radical scavenging activity was determined using the DPPH assay. The antioxidant activities of Pursat orange peel, Krasaing orange peel, and lemongrass leave were 88.89%, 83.72%, and 93.00%, respectively. Based on the benefit of bioactive compounds, the leave of lemongrass and peel of orange could be helpful in the contribution food security and fighting foodborne diseases (Ashaq et al., 2024; Viñas-Ospino et al., 2023).

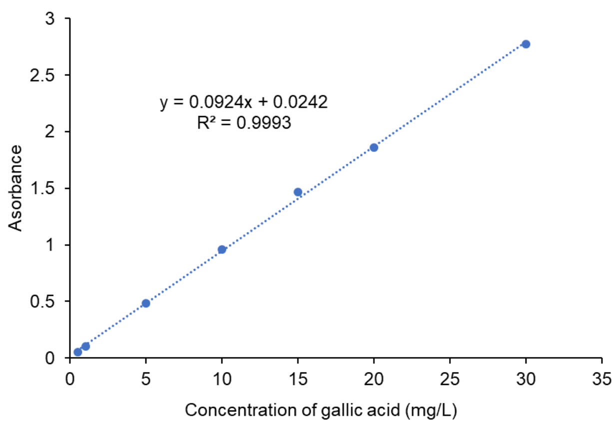

Figure 2. Calibration curve for total phenolic determination

Figure 3. Preparation of the calibration curve for total phenolic content

4. Conclusion

This study demonstrated the extraction of essential oils from organic wastes, such as orange peels and lemongrass leaves. The yield of essential oil from orange peels was higher than that from lemongrass. Similarly, the total phenolic content of orange peels was greater than that of lemongrass. All samples exhibited strong antioxidant activity, with a minimum of 88%. This research also emphasizes the recycling and extraction of valuable bioactive compounds from fruit waste or other parts of the samples, highlighting their importance for researchers, consumers, and the economic sector. Figure 1 Process of essential oil extraction

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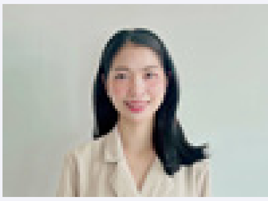

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5

Evaluation of Cherry Tomato Shelf Life Stored in Different Types of Packaging

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Highlight

- Cherry tomatoes is a type of climacteric fruit with a short shelf life that led to freshness and quality loss during storage.
- Plastic packaging materials has benefit in prolonging the shelf life of food product including post-harvest due to its property as barrier protection from the external environment.
- Application of different packaging materials on cherry tomatoes fruit has shown the result of quality improvement in term of physicochemical, microbiological (decay rate) and sensory evaluation during 14 days storage.

Evaluation of Cherry Tomato Shelf Life Stored in Different Types of Packaging

1. Introduction

Cherry tomatoes are a popular and widely grown vegetable crop in the world. They are a popular and commercially available vegetable and contain many nutrients (Gharezi et al., 2012). The main causes of post-harvest losses or spoilage are fruit rot, external injuries during harvest, handling and storage. In addition, environmental factors such as soil type, temperature and harvest during the rainy season can also affect the storage and quality of tomatoes (Haile and Safawo, 2018). Generally, after harvest, tomatoes should be stored at 10°C or higher to avoid injury, which can also affect their taste (Jimenez and Cantwell, 1996; Keri et al., 2002; Maul et al., 2000).

The quality of fruits and vegetables can also be affected by water loss during storage, depending on the temperature and humidity of the environment. Despite the increasing choice of packaging types for use in food industry, most four basic polymer materials as packaging materials such as polyvinyl chloride (PVC), polyethylene terephthalate (PET), polypropylene (PP) and polyethylene (PE) for fruit and vegetable (Ahvenainen, 2003; Exama et al., 1993; Kader and Watkins, 2000; Kader et al., 1989; Marsh and Bugusu, 2007; Prasad, 1995; Willige et al., 2002). The methods for extending the shelf life of cherry tomatoes are very important, including the packaging application and storage at a suitable temperature, which makes the cherry tomatoes last longer, which is the reason this study was conducted. The main objective of this research was to study the quality of cherry tomatoes using different types of packaging materials at different storage temperatures.

2. Materials and Methods

2.1 Sample Preparation

Cherry tomato samples were purchased from Doem Kor Market in Phnom Penh, Cambodia. After washing, the samples were divided into 6 groups. Group 1 was prepared and packaged in polypropylene bags (PP), group 2 was prepared and packaged in high-density polyethylene bags (HDPE), group 3 was prepared and packaged in low-density polyethylene bags (LDPE), group 4 was prepared and packaged in plastic clamshell (PCS), group 5 was prepared and packaged in foam plates with PVC film wrapped on top (FT), and group 6 was used as a control (without packaging). The samples were stored at the same temperature of 23±15°C until the experiment was performed.

2.2 Physico-Chemical Determination

Cherry tomatoes packed in different packaging were monitored and recorded for changes in physicochemical properties during storage. Each experimental parameter was monitored every 3 days for 14 days. The parameters to be tested were:

a. pH

The pH level was measured using a pH meter (HANNA HI2211),10ml of sample was taken. Each sample was measured until the pH value was constant and the data was recorded. For each sample, three measurements were taken.

b. Weight Loss

Weight loss is measured every 3 days and calculated as the percentage lost until the last 14 days of storage.

$$\text{Weight loss (\%)} = \frac{(Initial\ weight - Final\ weight)}{Initial\ weight} \times 100 \qquad \text{(Eq.1)}$$

c. Total Soluble Solid

The tomatoes were blended; the juice was extracted and then tested with a refractometer.

d. Total Acidity

The measurement was done according to (Ayele et al., 2012). The sample was shaken and filtered to obtain 10 ml, then diluted with 100 ml of distilled water and added with 3 drops of phenolphthalein indicator, finally titrated with 0.1N sodium hydroxide solution until the color turned to a light pink and recorded the amount of sodium hydroxide used. The formula was used to calculate:

$$\text{(\%) Citric acid} = \frac{Titrat\ value \times Normality \times m.eq.wt.of\ acid}{Volume\ of\ sample} \times 100 \qquad \text{(Eq.2)}$$

e. Moisture Content

3g of sample was weighed and placed on the plate that already dried. Then put in the oven to dry at 105 ° C for 3 hours. Frequently weigh the sample until it is stable and record the value to calculate the percentage of moisture loss.

$$\text{Moisture (\%)} = \frac{W1 - W2}{W1} \times 100 \qquad \text{(Eq.3)}$$

f. Vitamin C (Ascorbic Acid)

Vitamin C analysis was conducted according to (Ismail et al., 2014). Take 20ml of sample and put it in an Erlenmeyer flask (250ml), add 2ml of oxalic acid, 150ml of distilled water and 1ml of starch, then treat with iodine until the color changes to dark blue. Then record the amount of iodine used.

3. Determination of Decay Rate

To monitor the rot of cherry tomatoes, it was determined by observing the evolution of various spots on the skin of the tomato, both softening and decay were recorded accordingly (Sharma and Singh, 2010).

Formula: $DP = ND / TS \times 100$ (Eq.5)

DP = Percentage of decay

ND = Number of spot/decays

TS = Total sample

4. Sensory Evaluation

Sensory tests to monitor the quality of the cherry tomatoes were evaluated based on 9-point hedonic scale on days 0, 3, 6, 9, 12 and 14 by 12 panelist, food chemistry students, including men and women aged between 18 and 25. The questionnaire required consumers to rate each type of sample on the attribute such as color, smell, texture and overall acceptability. The score was in range from 1-9, with 1 being very disliked and 9 being very liked.

5. Statistical Analysis

The data were analyzed by using SPSS, (One-way ANOVA) to see the significant difference at the level ($p < 0.05$).

3. Results and Discussion

3.1 Physico-Chemical Properties

a. pH

The study found that pH values of cherry tomatoes varied with packaging and storage time. The control sample increased to 4.48 and 4.44 on days 3 and 6, while the sample packed with HDPE showed a drop to 4.33 on day 9 before rising to 4.51 on day 12. The sample packed with FT packaging decreased to 4.41 and 4.15 on days 12 and 14. Significant differences ($P<0.05$) were observed between unwrapped and packaged tomatoes under the same conditions. These results agree with previous studies showing that pH changes are influenced by packaging, temperature, storage time, and microbial activity, with tomatoes typically ranging from 4.2–4.6 as ripening stage progresses. The higher the barrier to the gases through the packaging film, the lower was the pH (Proudel and Basnet., 2022).

b. Weight Loss

The weight loss study showed that unpacked tomatoes (control) had the highest weight loss, consistent with Sammi and Masud (2007). The sample packed with PP bag have performed best, with the lowest weight loss (2.93% on day 14). Significant differences ($P<0.05$) were found between packaged and unpackaged tomatoes under the same storage conditions (23±1.5°C). The higher weight loss in controls is linked to climacteric fruits ripening, which increases ethylene production, respiration, and water loss (Sabir et al., 2004). In contrast, packaging like PP reduces weight loss by allowing limited moisture exchange, lowering respiration, and minimizing water loss (Jenkins et al., 2001). The postharvest water loss of fruits and vegetables results from respiration and diffusion through the surface in horticultural products, leading to a deterioration in quality (Kabir et al., 2020).

Another research have shown that this could be attribute to the maintenance of high humidity in the microatmosphere within the package by the respiring fruits and due to low water vapor transmission loss due to temperature effects on vapor pressure difference and increased water retention (Gharezi and Sadeghian, 2012).

c. Total Soluble Solid

The study found that control cherry tomatoes had the highest total sugar, while all the packaged tomatoes showed the lowest total sugar during storage. Significant differences ($P<0.05$) were observed between packaged and unpackaged tomatoes under the same storage conditions. This was attributed to retard the respiration and metabolic activity in packaged tomatoes, which delays ripening (Gharezi et al., 2012). Total sugars increase during ripening as polysaccharides break down into simple sugars, paralleling color development, with typical values ranging from 5.0 to 5.45 ºBrix at the red stage (Shahnawaz et al., 2012). The minimum change in TSS vales of tomatoes in controlled atmosphere storage increased from 5.2 to 5.59 over 40 days of storage. The rapid initial increase in TSS content which then decreased during the storage of cherry tomatoes at ambient and cold storage condition (Kabir et al., 2020).

d. Total Acidity

The study showed that total acidity varied with packaging type and storage time. Cherry tomatoes packed with LDPE decrease from day 3 to day 9 (0.33–0.25), despite the control was the lowest on day 3 (0.31). The sample packed with PCS and FT showed the highest acidity on day 14 (0.53 and 0.57). Significant differences ($P<0.05$) were found between packaged and unpackaged cherry tomatoes which show the effectiveness of packaging materials application. This decrease is linked to the ripening process and microbial used of citric acid as a carbon source, with malic acid loss followed by citric acid breakdown during ripening (Mathew et al., 2007; Anyasi et al., 2016). The changes in organic acids during ripening have been attribute to a rise in citrate and fall in malate, indicating a change in metabolism of citrate and reduction on the level of citric acid (Gharezi and Sadeghian, 2012).

e. Moisture Content

The study showed significant differences ($P<0.05$) in moisture content of packaged and unpackaged cherry tomatoes under the same storage conditions. The control sample have shown the moisture loss gradually until day 9. The sample packed with PCS packaging had the highest increase on day 3 (94.61±0.13), while FT showed a decline on days 9–12 (91.76–90.8). PP, LDPE, and HDPE maintained higher moisture, with slight increases on days 6 and 14 (around 93.3–94.6). Most packaging material helped to prevent the moisture loss from the inside package also it could inhibit the penetration of water vapor or oxygen from the outside package, that could affect the respiration process of cherry.

f. Vitamin C

The study showed that vitamin C content in cherry tomatoes decreased over storage, with significant differences ($P<0.05$) between packaged and unpackaged samples. The sample packed with PP, LDPE, and HDPE had similar declining trends, while the sample packed with PCS and FT retained higher vitamin C values, decreasing more slowly from day 3 to 14. An increase in ascorbic acid content in fruit is though to be an indication that the fruit is still in the ripening stage, while a decrease indicates a senescent fruit. Hence, the reduction is consistent with earlier findings (Gharezi et al., 2012), as vitamin C is highly sensitive to oxidation, temperature, light, ripening, and storage duration (Tigist et al., 2013). The oxidative degradation of ascorbic acid can be catalyzed by enzymes such as ascorbate oxidase and peroxidase, which are more active at higher temperatures and in the presence of oxygen (Bapary et al., 2024).

3.2. Decay Rate

The study found that control tomatoes had the highest percentage rot in range of 33% to100% from day 9 to day 14, while the sample with packaging could reduce spoilage eventually. The sample with HDPE and PCS showed the percentage increase in rage of 13% to 66% rot and 27% to 46% from day 12 to day 14 respectively. Despite PP, LDPE, and FT could extended shelf life up to 14 days. Packaging reduced moisture loss and external contamination but did not fully prevent microbial growth due to the moisture content maintaining. Spoilage was mainly caused by Alternaria alterna, consistent with previous studies (Mir & Beaudry, 2004; Feng et al., 2008).

Table 3.1 Decay of small tomatoes from day 0 to day 14

Packaging	Decay rate of small tomatoes for 14 days					
	Day0	Day3	Day6	Day9	Day12	Day14
Control	0±0.00	0±0.00	0±0.00	33±0.02	67±0.01	100±0.00
PP	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	20±0.05
LDPE	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	40±0.02
HDPE	0±0.00	0±0.00	0±0.00	0±0.00	13±0.04	66±0.01
PCS	0±0.00	0±0.00	0±0.00	0±0.00	27±0.02	46±0.03
FT	0±0.00	0±0.00	0±0.00	0±0.00	0±0.00	53±0.07

3.3 Sensory Evaluation

According to the sensory evaluation of cherry tomatoes, the result was found that the sample packaged in PP, LEPE, HDPE bags were effective in extending the shelf life and maintaining the freshness of cherry tomatoes up to 9 days, with unacceptably low scores on the 12th and 14th days. The cherry tomatoes without packaging were only kept for 3 days because there was no barrier, which made them susceptible to air reaction, which was prone to water loss and could not protect against microorganisms in the environment. Through this study, PP, LDPE, PCS and HDPE bags were able to maintain the appearance, texture, aroma, color and freshness of tomatoes up to 9 days when stored at a temperature of 23±15ºC.

4. Conclusion

This study focused on the type of packaging used on tomatoes stored at 23 ± 15 ° C for 14 days. According to the physicochemical characteristics, the pH, total sugars, and total acids varied slightly in the five types of packaging. For weight loss and decay in PP, HDPE and LDPE packaging, there is less loss and decay than other packaging. Therefore, PP, HDPE and LDPE packaging is more effective than other types of packaging, which can prevent dehydration of cherry tomatoes during storage, compared with cherry tomatoes without packaging that lose freshness within 3 days. In terms of color, odor, texture, appearance and general characteristics, cherry tomatoes packed with PP HDPE and LDPE bags were more popular with customers, in contrast to PCS and FT packaging. As a result of this study, it is worthwhile paying attention to the packaging of fruits and vegetables, using effective packaging, because the right packaging can help keep fresh fruits and vegetables longer. In the future, the use of this technique should be widely used in the packaging of fruits and vegetables to help reduce post-harvest damage and promote the technique of storing fresh produce for a longer period.

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6

Base Formula Development of Syrup from Tomatoes and Jam from Tomato

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Highlight

- Developed a base formula for producing syrup from tomato juice and jam from tomato residues (peels and seeds), reducing food waste.
- Physico-chemical analysis showed pH 3.64–3.76 for syrup and 3.51–3.76 for jam, comparable to commercial standards.
- Microbiological tests confirmed safe levels of yeast and mold (<100 CFU/mL) and absence of harmful bacteria.
- Sensory evaluation identified SR012 syrup and JM040 jam as the most preferred formulas for taste and texture.
- Demonstrated that tomato residues can be transformed into value-added, sustainable food products, contributing to food innovation and waste reduction.

Base Formula Development of Syrup from Tomatoes and Jam from Tomato

Tomatoes (*Roma variety*)

↓

Processing

Wash → Blanch → Peel → Separate juice, pulp, seeds & skin

↓

Product Development

Tomato Syrup (Formulations SR011, SR012, SR013)

Tomato Jam (Formulations JM030, JM040, JM050)

↓

Physicochemical Analysis

(pH, Titratable acidity, Total soluble solids (TSS))

↓

Microbiological Analysis

(Yeast & mold, *E. coli*, *Salmonella spp.*, *Staphylococcus spp.)*

↓

Sensory Evaluation

Color, Texture, Taste, Smell, Overall acceptability

↓

Best Formulas Identified

SR012 (Tomato Syrup)

JM040 (Tomato Jam)

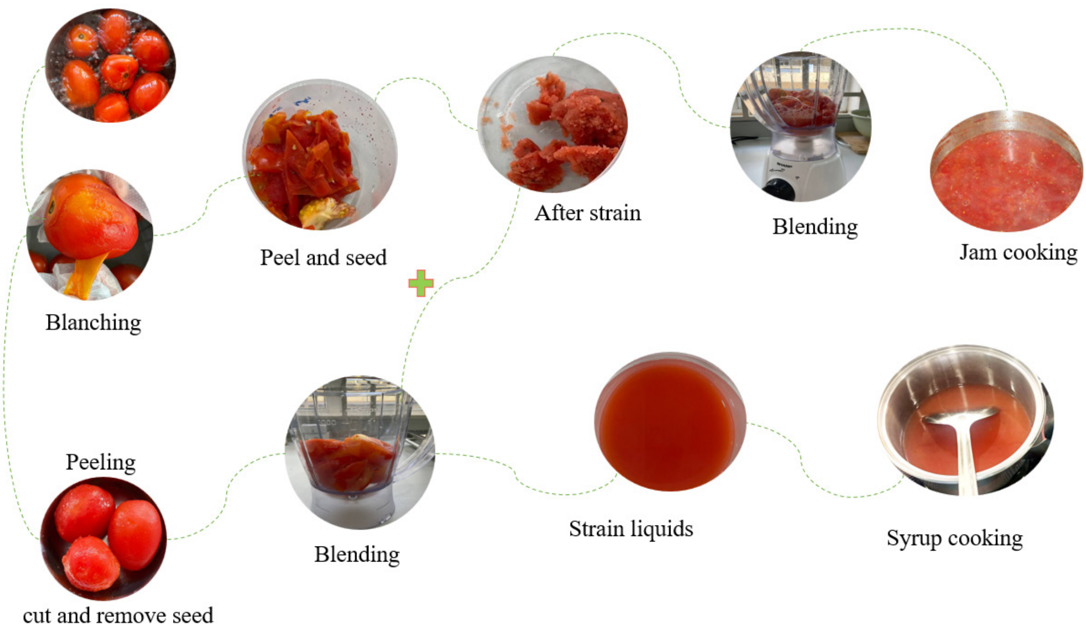

Figure 1. Flow chart of research design, Jam and syrup processing

1. Introduction

Tomatoes (Solanum lycopersicum) are among the most widely consumed vegetables and an important dietary source of nutrients and bioactive compounds. They contain carotenoids, fiber, protein, amino acids, vitamins, and minerals, contributing to health benefits such as improved circulation, blood pressure regulation, and protection against chronic diseases (Elbadrawy & Sello, 2016; Li et al., 2021). Lycopene, the major carotenoid in tomatoes, has been associated with antioxidant, anti-inflammatory, and cardioprotective properties.

Despite these benefits, tomato residues including skins and seeds are often discarded during processing, contributing to food waste (Ray et al., 2016). Developing value-added products such as syrups and jams from Roma tomatoes and their by-products may reduce waste while enhancing nutritional and economic value. Roma tomatoes are particularly suitable due to their high lycopene content and firm flesh, which provide desirable sensory and functional qualities (Frusciante et al., 2007).

This study aimed to develop base formulations for tomato syrup and jam from tomato residues, and to evaluate their physicochemical, microbiological, and sensory properties.

2. Materials and Methodology

2.1 Raw Materials and Processing

Roma tomatoes were purchased from Dermkor market in Phnom Penh, Cambodia. Fresh tomatoes were washed, blanched, and peeled. Tomato juice was used for syrup preparation, while seeds and skins were combined with pulp for jam production.

2.2 Tomato Syrup Preparation

Tomato juice was cooked at 400 W for 30 min until the raw flavor was reduced. Three syrup formulations (SR011, SR012, SR013) were prepared by varying tomato-to-sugar ratios (1:1, 2:1, 3:1) with water and 1% citric acid. Syrups were pasteurized at 80 °C and stored in sterilized bottles.

Table 1. Tomato syrup formulation with different sugar concentrations.

Ingredient	Sample		
	SR011	SR012	SR013
Tomato juice (g)	300	300	300
Sugar (g)	300	150	100
Water (ml)	300	300	300
Citric acid (g)	3	3	3

2.3 Tomato Jam Preparation

Tomato pulp (peel + seeds) residue from tomato syrup production was combined with sugar (30%, 40%, and 50%), pectin (1.2%), and citric acid (0.6%). Three formulations (JM030, JM040, JM050) were boiled until gel consistency (60 °Brix), cooled, pasteurized, and stored at 4 °C (Fellows, 2013).

Table 2. Tomato jam formulation with different sugar concentrations.

Ingredient	Sample		
	JM030	JM040	JM050
Tomatoes pulp (seed + skin) (g)	400	400	400
Sugar (g)	200	160	120
Water (ml)	200	200	200
Citric acid (g)	4.8	4.8	4.8
Pectin (g)	2.4	2.4	2.4

2.4 Physicochemical Analysis

Total Soluble Solids (TSS) were measured using a refractometer (ATAGO, Japan) following standard methods (Kaur et al., 2022), while pH value of jam and syrup were determined using a calibrated pH meter (Nguyen et al., 2014). The acidity of both products was measured by titration with 0.1 N NaOH (Olugbenga Awolu, 2018). Moisture and Ash Content were analyzed using oven-drying (105 °C) and Muffle furnace (500 °C) following by AOAC methods (Liu et al., 2020).

2.5 Microbiological Analysis

Spread plate techniques were performed on PDA, MacConkey, XLD, and MSA agar to detect yeast/mold, E. coli, Salmonella, and Staphylococcus spp. (Khan et al., 2015).

Table 3. Type of medium and conditions use for microbial testing

Media	Selective Bacteria	Technique	Incubation
PDA	*Yeast and Molds*	Spread plate	37 °C for 5day
MSA agar	*Staphylococcus spp.*	Spread plate	37 °C for 24h
MacConkey agar	*Escherichia coli*	Spread plate	37 °C for 24h
XLD agar	*Salmonella spp.*	Spread plate	37 °C for 24h

2.6 Sensory Evaluation

A panel of 15 untrained judges assessed color, texture, taste, smell, and overall acceptability using a 7-point hedonic scale (Galyuoni, 2022).

3. Results and Discussion

3.1 Physicochemical Properties

According to Table 4, the pH values of tomato syrup (3.64–3.76) and jam (3.51–3.76) remained within the recommended safe range of below 4.0, which supports microbial stability and product safety. The titratable acidity results presented in Table 5 show that syrup samples contained 1.18–2.14% acidity, while jam samples ranged between 3.51–3.76%, contributing both to the characteristic sourness and the preservation potential of the products. In terms of soluble solids, Table 6 indicates that syrup had TSS levels of 63.33–70.58 °Brix, whereas jam contained 56.52–64.97 °Brix. These values align with commercial fruit-based syrups and jams, suggesting desirable sweetness and product quality.

The proximate composition results provided in Table 7 reveal that moisture content of jams was notably lower (5.13–7.51%) compared to fresh tomatoes (27.18%), confirming effective concentration during processing. Ash content ranged from 0.5–3.5%, which reflects the mineral contribution from tomato peels and seeds incorporated into the product.

Finally, the microbial evaluation results (Table 8) confirmed that both syrup and jam were within acceptable safety limits, with low counts of yeast and mold, and no harmful microorganisms detected, ensuring the microbiological stability of the products during storage.

Table 4. pH of tomato syrup and jam during storage

Sample	Timeline				
	Day 0	Day 5	Day 10	Day 15	Day 20
SR011	3.64 ± 0.01^b	3.68 ± 0.01^a	3.71 ± 0.01^a	3.73 ± 0.01^a	3.76 ± 0.02^a
SR012	3.64 ± 0.01^b	3.65 ± 0.01^b	3.68 ± 0.01^b	3.69 ± 0.01^b	3.70 ± 0.01^b
SR013	3.68 ± 0.02^a	3.62 ± 0.01^c	3.65 ± 0.01^c	3.65 ± 0.03^c	3.69 ± 0.01^c
JM010	3.55 ± 0.01^a	3.58 ± 0.02^a	3.64 ± 0.02^a	3.64 ± 0.01^a	3.76 ± 0.01^a
JM020	3.52 ± 0.02^{ab}	3.55 ± 0.02^b	3.59 ± 0.01^b	3.62 ± 0.02^a	3.74 ± 0.01^b
JM030	3.51 ± 0.01^b	3.54 ± 0.01^c	3.57 ± 0.01^c	3.63 ± 0.01^a	3.68 ± 0.01^c

Table 5. Titratable acidity of tomato syrup and jam during storage

Sample	Timeline				
	Day 0	Day 5	Day 10	Day 15	Day 20
SR011	2.14 ± 0.01^a	2.09 ± 0.02^a	2.07 ± 0.02^a	2.12 ± 0.01^a	2.13 ± 0.01^a
SR012	2.03 ± 0.02^b	1.97 ± 0.01^b	1.94 ± 0.02^b	1.94 ± 0.01^c	1.93 ± 0.01^b
SR013	1.18 ± 0.01^c	1.25 ± 0.02^c	1.19 ± 0.02^c	1.24 ± 0.02^c	1.24 ± 0.01^c
JM010	1.03 ± 0.01^c	0.88 ± 0.03^c	0.77 ± 0.01^c	0.68 ± 0.01^c	0.43 ± 0.02^c
JM020	1.13 ± 0.02^b	0.95 ± 0.00^b	0.86 ± 0.02^b	0.77 ± 0.01^b	0.67 ± 0.01^b
JM030	1.22 ± 0.03^a	0.97 ± 0.01^b	0.88 ± 0.01^c	0.77 ± 0.01^b	0.71 ± 0.02^c

* Superscripts (a–c) in the same row indicate significant differences ($p < 0.05$)

Table 6. TSS of tomato syrup and jam during storage

Sample	Timeline				
	Day 0	Day 5	Day 10	Day 15	Day 20
SR011	69.11 ± 0.01^a	70.03 ± 0.01^a	70.31 ± 0.01^a	70.54 ± 0.02^a	70.58 ± 0.01^a
SR012	67.97 ± 1.15^b	68.92 ± 0.03^b	69.31 ± 0.03^b	69.43 ± 0.03^b	69.55 ± 0.01^b
SR013	63.33 ± 0.01^c	63.52 ± 0.02^c	63.86 ± 0.02^c	63.87 ± 0.02^c	63.85 ± 0.01^c
JM010	63.51 ± 0.03^a	64.03 ± 0.01^a	64.47 ± 0.02^a	64.97 ± 0.02^a	65.46 ± 0.03^a
JM020	58.53 ± 0.01^b	60.06 ± 0.03^b	60.56 ± 0.02^b	60.92 ± 0.01^b	61.43 ± 0.02^b
JM030	56.52 ± 0.55^c	57.61 ± 1.31^c	58.08 ± 0.01^c	58.55 ± 0.02^c	59.07 ± 0.05^c

Table 7. Moisture and Ash content of tomato and tomato products

Sample	Moisture content (%)	Ash content (%)
Tomatoes	27.18 ± 0.02^{a}	0.5 ± 0.03^{a}
SR011	27.21 ± 0.03^{b}	1.0 ± 0.04^{c}
SR012	30.04 ± 0.01^{a}	2.0 ± 0.06^{b}
SR013	27.01 ± 0.04^{b}	2.50 ± 0.07^{c}
JM030	6.61 ± 0.03^{b}	3.50 ± 0.09^{a}
JM040	7.51 ± 0.02^{a}	0.50 ± 0.05^{c}
JM050	5.13 ± 0.01^{c}	1.50 ± 0.04^{b}

* Superscripts (a–c) in the same row indicate significant differences ($p < 0.05$)

3.2 Microbiological Quality

No E. coli, Salmonella, or Staphylococcus spp. were detected for both SR012 and JM040. Yeast and mold counts (40 CFU/mL in syrup; absent in jam) were found below international safety thresholds (<100 CFU/mL) (Khan et al., 2015). This indicates the pasteurization process effectively maintained microbial safety.

Table 8. Microbial of tomato syrup and jam tomato product

Bacteria	SR012 (CFU/ml)	JM040 (CFU/ml)	Acceptable
Yeast and Molds	40	Not detected	< 100
Staphylococcus spp.	Not detected	Not detected	Not detect
Escherichia coli	Not detected	Not detected	Not detect
Salmonella spp.	Not detected	Not detected	Absent 25g

3.3 Sensory Evaluation

Although ANOVA showed no significant differences among formulations ($p>0.05$), SR012 syrup and JM040 jam received the highest scores for taste and texture. Panelists described these formulations as more balanced in sweetness and acidity. Previous studies confirm that sugar-to-acid ratio strongly affects acceptability in fruit-based products (Uribe-Wandurraga et al., 2021).

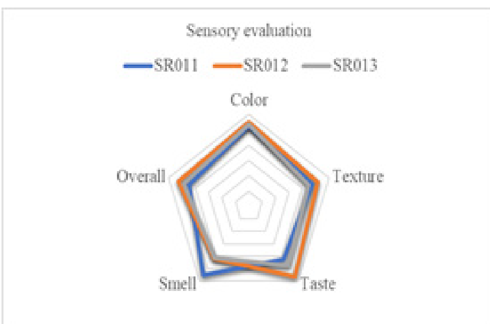

Figure 2. Sensory scores of tomato syrup with different sugar concentrations.

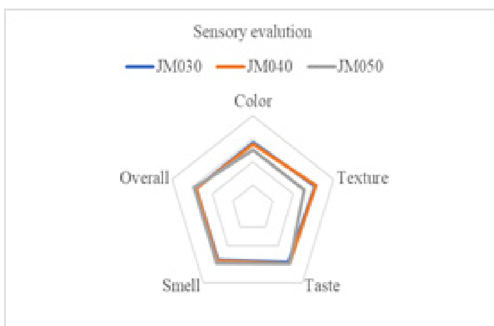

Figure 3. Sensory scores of tomato jam with different sugar concentrations.

3.4 Implications

The study demonstrates that tomato residues can be transformed into acceptable, safe, and nutrient-rich value-added products. Similar to other fruit-based innovations, such products could reduce postharvest losses, diversify tomato product lines, and support sustainable food systems (Ray et al., 2016; Salehi et al., 2019).

4. Conclusion

This study successfully developed base formulations for tomato syrup and jam using Roma tomatoes and processing residues. The products met acceptable physicochemical and microbiological standards, and sensory evaluation suggested SR012 (syrup) and JM040 (jam) as optimal formulas. These findings highlight the potential of tomato by-products as raw materials for value-added food production, reducing waste while creating nutritionally beneficial and economically viable products. Future research should focus on proximate composition, calorie-free sugar alternatives, and consumer testing to optimize market readiness.

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TECHNOLOGY TRENDS

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7

Converting Agricultural Waste into High-Quality Compost Using Lactic Acid Bacteria

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Highlight

- Lactic acid bacteria (LAB) reduced composting time to 30 days by boosting heat and decomposition.
- LAB produces dark, stable, odor-free compost with optimal moisture and pH.
- Ae66 LAB strain improves compost quality and supports high BSFL growth.
- LAB offers a sustainable solution for converting Cambodia’s agro-waste.

Converting Agricultural Waste into High-Quality Compost Using Lactic Acid Bacteria

1. Introduction

Cambodia's expanding agricultural industry produces large quantities of organic waste, including crop residues and food leftovers (National Institute of Statistics (NIS), Mar 17, 2025). The improper waste management leads to environmental issues, including greenhouse gas emissions, contamination of soil and water, and threats to public health (Hoklis & Sharp, 2014). In Phnom Penh, food and organic materials make up the majority of the city's waste (55%), but only 2% undergoes composting (Kaiser, 20 January 2025; Pheakdey et al., 2022).

This highlights a significant lost opportunity to convert biodegradable waste into a useful agricultural input (Farhidi et al., 2022). A recent study indicates that lactic acid bacteria (LAB) show strong potential for fermenting carbohydrates into lactic acid, which reduces compost pH and suppresses the growth of harmful pathogens (Ayivi et al., 2020). LAB also aids in breaking down complex organic materials, minimizing nutrient loss, and retaining key elements such as nitrogen and phosphorus, which together enhance compost quality (Waqas et al., 2023; Zainudin et al., 2022).

In addition, LAB can be successfully integrated into Black Soldier Fly Larvae (BSFL) composting systems, where they pre-digest the substrate, lower pH levels, and inhibit harmful microorganisms. This process increases nutrient availability for BSFL and accelerates waste decomposition (Osama Elhag et al., 2022; Gold et al., 2018; Zhang et al., 2021). BSFL further supports pathogen reduction by hosting beneficial gut bacteria, such as Bacillus subtilis (O Elhag et al., 2022; Gold et al., 2018; Surendra et al., 2016). However, despite these advantages, the application of LAB in composting remains limited in Cambodia. Therefore, this study aims to evaluate the effectiveness of LAB in composting agricultural waste within the Cambodian context, aiming to encourage broader adoption of sustainable composting practices nationwide.

2. Materials and Methods

2.1 Materials

Pineapple peels, green leaves, and ground coffee were utilized along with the inoculation of Lactiplantibacillus pentosus strain Ae48 and Ae66, and *Lactobacillus delbrueckii* subsp. *bulgaricus* NBRC 13953, a commercial strain, was used as the control.

2.2. Methods

2.2.1 Feedstock Preparation

Pineapple peels and green leafy vegetables were collected from Doeurm Kor Market in Phnom Penh, Cambodia, while ground coffee was obtained from a coffee shop. The feedstock was prepared by mixing pineapple peels, green leaves, and ground coffee at a ratio of 5:5:6 (w/w, kg), chopping them into small pieces, mixing thoroughly, and treating with nystatin (5,000 U/mL) to inhibit fungal growth. The mixture was then left at room temperature for 12 hours before inoculation.

2.2.2 Lactic Acid Bacteria Inoculation to Feedstock

LAB were streaked onto MRS agar (HIMEDIA, India) and incubated at 37°C for 24 hours. A single colony was transferred into 3 mL MRS broth and cultured for 18 hours. The LAB suspension (10^8 CFU/ml) was inoculated into a molasses solution diluted with distilled water (1:4, v/v). The feedstock was subjected to five treatments: (1) Inoculated with NBRC, (2) without any LAB inoculation, (3) Inoculated with Ae48, (4) Inoculated with Ae66, and (5) a co-culture (Ae48 and Ae66). The mixtures were thoroughly blended in round plastic baskets for storage and underwent solid-state fermentation at room temperature, with weekly monitoring for 30 days, as illustrated in Figure. 1.

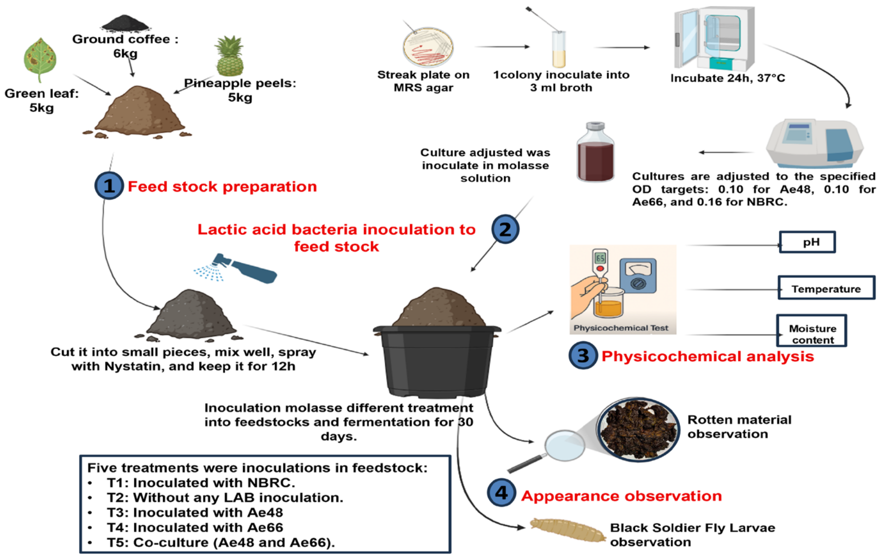


Figure 1. Experimental workflow for lactic acid bacteria inoculation and composting process.

2.3. Physicochemical Analysis

Ten grams of compost were collected at various time intervals (day 0, 7, 14, 21, and 30) from different locations within the pile: top, middle, bottom, left, and right, to examine.

2.3.1 pH

The pH of the fermented feedstock was determined with a calibrated pH meter (Hanna Instruments Edge® pH Meter, HI2001 series). For the measurement, a 1:9 (w/v) slurry of the material in distilled water was prepared.

2.3.2 Temperature

Temperature was measured daily using a digital thermometer (Model TA612C, TASI Instruments, China) from the middle of each compost pile.

2.3.3 Moisture Content

Moisture content was determined using the gravimetric (oven-drying) method (Barbosa-Cánovas et al., 2017). Approximately 10 g of fresh compost was dried in an oven at 60°C until a constant weight was achieved.

2.4 Appearance Observation

The compost was visually inspected on day 0, 7, 14, and 30 to check for the presence of rotten material and the number of BSFL.

3. Results and Discussion

3.1 Physicochemical Analysis

3.1.1 pH Analysis

Figure 2. presents the pH value over days 0-30. The pH decreased during the first 14 days and then increased toward day 30. LAB-treated samples exhibited a stable U-shaped trend, whereas the control (without LAB) continued to decline, indicating that LAB inoculation promotes microbial balance and accelerates compost stabilization. During the early stages of composing, the pH dropped slightly to around 4.8, aligning with previous findings (Tran et al., 2015).

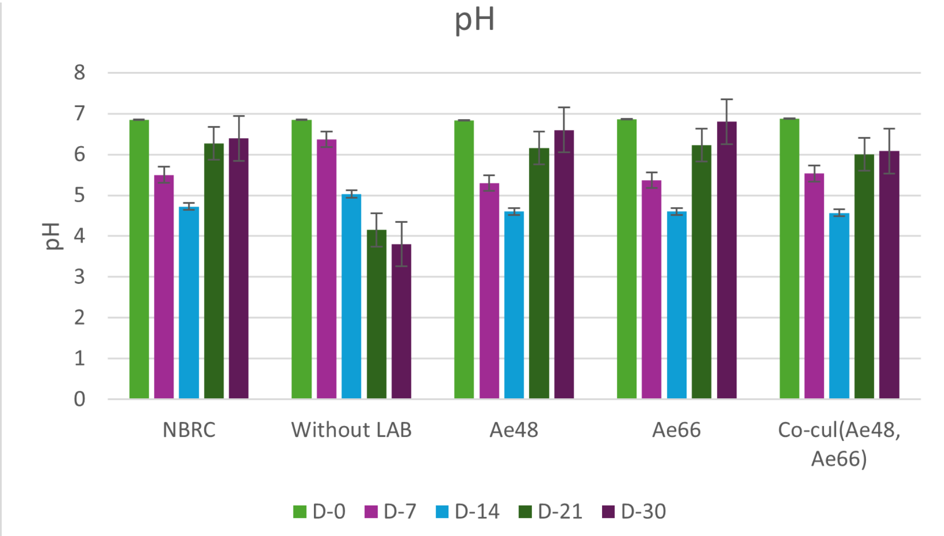


Figure 2. Effect of LAB on compost pH

3.1.2 Temperature

Figure 3. shows that temperature increased rapidly in LAB-inoculated treatments, reaching thermophilic levels (64–68°C) by day 7, whereas the control peaked at only 37°C. This indicates that LAB enhanced microbial activity, creating optimal thermophilic conditions for faster and more effective composting. Consistent with Liu et al. (2023) and Tran et al. (2015), the degradation phase (days 7–15) was characterized by compost temperatures of 60–65°C.

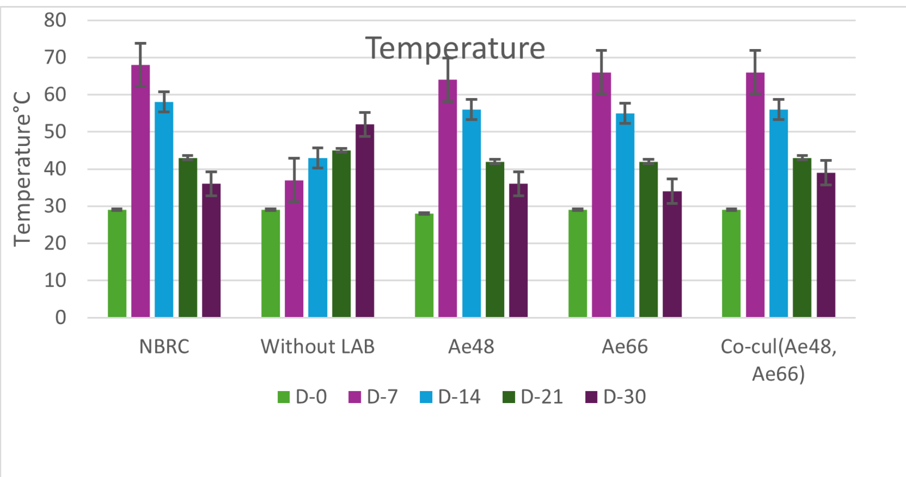


Figure 3. Effect of LAB on Compost Temperature

3.1.3 Moisture Content

Figure 4. illustrates that by day 30, moisture content in the LAB-treated compost decreased to 38%-39%, whereas the untreated control remained high at 67%. These results indicate efficient compost drying, driven by active microbial metabolism and heat production that promotes moisture evaporation. This aligns with the findings of Liu et al. (2023), who reported a final compost moisture content of 31.7%, showing a similar trend in moisture reduction.

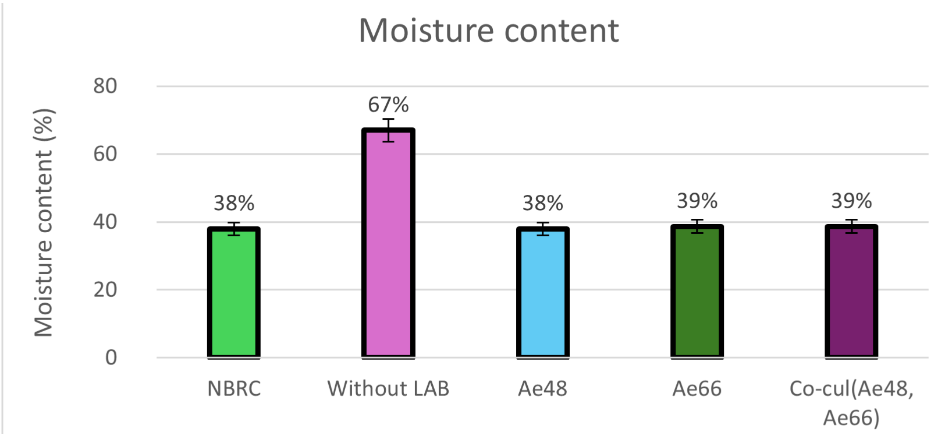


Figure 4. Final moisture content of the compost after fermentation.

3.2 Appearance Observation

3.2.1 Rotten Material Observation

Figure 5. shows the composting progression over 30 days for treatments inoculated with LAB strains (NBRC, without LAB, Ae48, Ae66, and a co-culture) compared to a non-LAB control. On day 0, all treatments consisted of fresh, undecomposed residues. By day 7, compost exhibited faster tissue breakdown and darker coloration, while the control still contained intact plant material. By day 14, LAB composts exhibited advanced decomposition with a crumbly texture, whereas the control contained undegraded fragments. At day 30, LAB-inoculated composts reached maturity—uniform dark brown, crumbly, odor-free, and stabilized—while the non-LAB control remained partially decomposed and required ~65 days to mature.

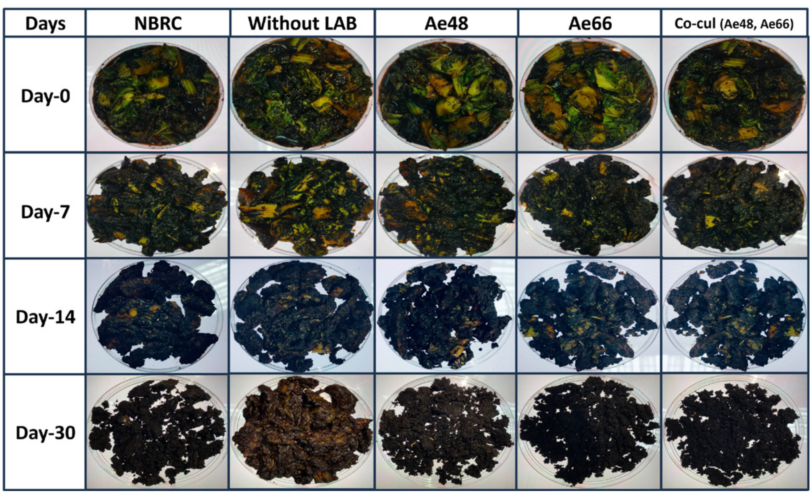


Figure 5. Composting progress in different compost treatments over 30 Days.

3.2.2 Black Soldier Fly Larvae Observation

The BSFL population differed across compost treatments. Compost inoculated with Ae66 supported the highest number of larvae (370), followed by the NBRC positive control (320) and the Ae48- Ae66 co-culture (287). In contrast, Ae48 inoculated compost showed the lowest count (106), and the non-inoculated control produced no measurable BSFL. These findings suggest that LAB inoculation affects compost suitability as a BSFL substrate, with Ae66 showing the most pronounced positive impact.

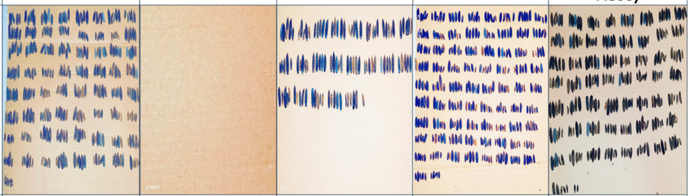
N°	NBRC	Without LAB	Ae48	Ae66	Co-cul (Ae48, Ae66)
BSFL					
Total	320	0	106	370	287

Figure 6. Number of Black Soldier Fly Larvae (BSFL) in different compost treatments.
Conclusion

4. Conclusion

This study demonstrated that lactic acid bacteria (LAB) notably improved the composting process compared to the non-inoculated control, which required about 65 days to mature. Inoculation with *Lactiplantibacillus pentosus* strain Ae48 and Ae66, particularly with strain Ae66, accelerated decomposition, stabilized pH at 6.75, reduced moisture to optimal levels, and supported the largest BSFL population. These findings emphasize the promise of LAB as a bio-enhancer for generating nutrient-rich compost and suggest a sustainable approach to managing agricultural waste in Cambodia.

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8

CRISPR/Cas9 Technology in Biomedical Science

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Highlight

- CRISPR/Cas9 is the Nobel Prize-winning gene editing technology.
- Cellular/animal model for studying the untreated disease.
- Genetic mutations/genetic disorders are challenging diseases for patients and the medical field.
- Gene therapy is the new insight for the genetic diseases.

CRISPR/Cas9 Technology in Biomedical Science

1. Introduction

Clustered regularly interspaced short palindromic repeats-associated protein 9 (CRISPR/Cas9) is a novel gene editing technology. It could target specific sites and multiple sites of the genome sequencing at the same time. The process is precise and simpler compared to the previous technique, such as Zinc-Finger Nucleases and Transcription Activator-Like Effector Nucleases (Rocha-Martins et al., 2015). In 2020, Jennifer Doudna and Emmanuelle Charpentier won the Nobel Prize in Chemistry for discovering this cutting-edge technology. With this technology, biomedical engineering could incorporate this with the principles of biology, physics, and engineering to study the human genetic diseases, develop novel analysis tools and new therapies. The novel analysis tool such as evaluating the inflammatory response of biomaterials in vitro (outside the body), that previously only available in vivo (inside the body). The new therapy for untreated diseases such as cancer, genetic single/multiple mutations, and highlighted in this review Figure 1.

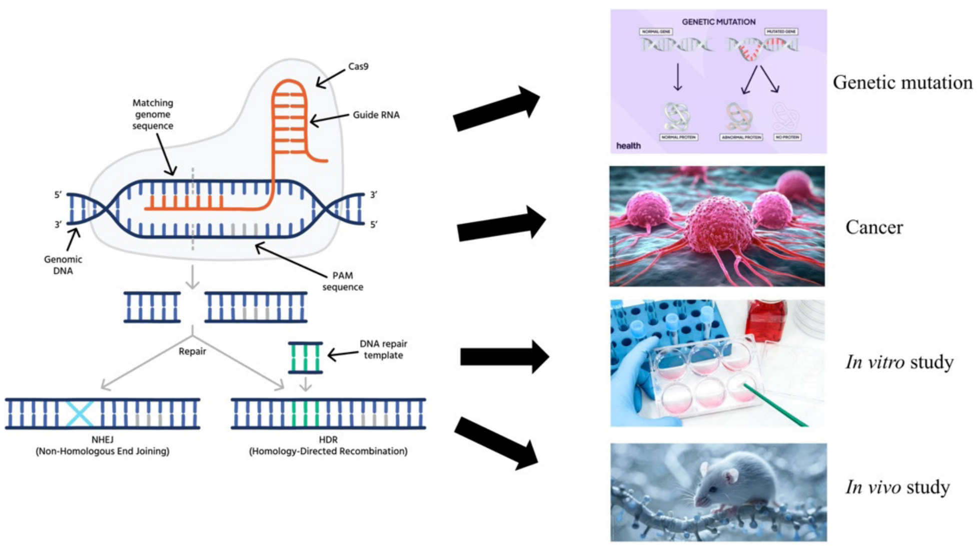

Figure 1. CRISPR/Cas9 technology in the development of the research model

2. Mechanism of the CRISPR/Cas9

The CRISPR/Cas9 system consists of two components, such as single-guide RNA (sgRNA) and Cas9 nuclease. The sgRNA is engineered from two RNA sequences, namely CRISPR RNA (crRNA) and trans-activating crRNA (tracrRNA) (Mali et al., 2013). The mechanism of the CRISPR/Cas9 consists of 2 steps, which are the DNA double-strand break and the DNA repair process. sgRNA, known as guide RNA, binds to Cas9 and forms a Cas9–sgRNA ribonucleoprotein complex. These complex enzymes screen the DNA sequence to find the matching site and then unwind those locations. The sgRNA pairs with the unwinding DNA, and then the Cas9 enzyme is activated, resulting double-strand break (Sternberg et al., 2014).

The gap between the DNA strands occurs, leading to DNA repair by non-homologous end joining (NHEJ) or homology-directed repair (HDR). Non-homologous end joining is a process of binding the ends of broken

DNA together, resulting in the deletion of the DNA target. Homology-directed repair is a process for inserting the new DNA sequence (Figure 2). With this technology, the study of gene sequence function and developing the disease model is easier.

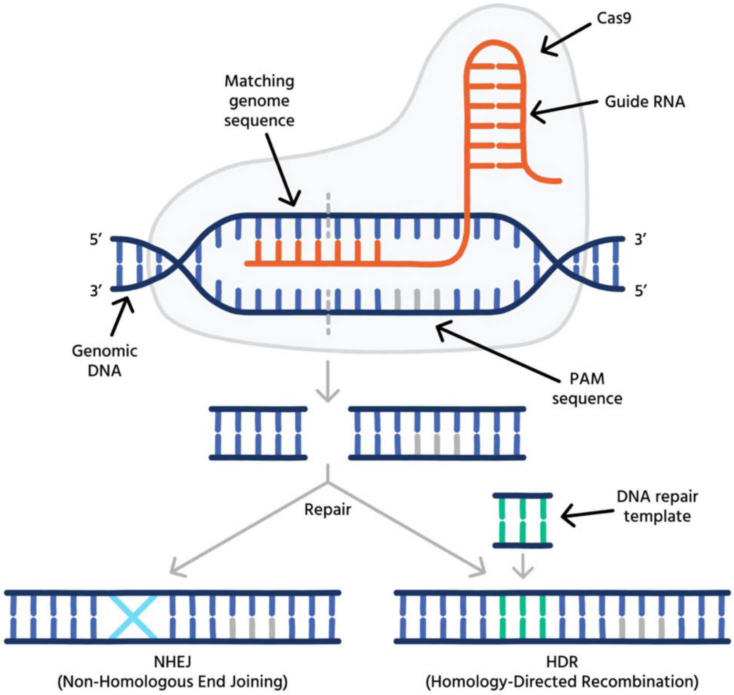

Figure 2. Principles of CRISPR-Cas 9

3. Develop from the Gene to Model (Cell/Animal)

For developing the model, the genome sequencing was first undergone editing with the CRISPR/Cas9 technique at the specific sequence by removing that sequence or adding the new sequence as described in the Figure 2. Then, the edited gene is microinjected into the cells and allowed to undergo cell division until the pure cell with the gene editing model is achieved Figure 3a. The type of cells depends on the function and purpose of the study. In case of the animal model, the edited gene is microinjected into the zygote and allows cell division into the blastocyst, then transferred it into the embryo of an animal to produce the gene-edited animal (eg., mice, rat, monkey, pig) Figure 3b.

4. Application of the CRISPR/Cas9 in the Biomedical Sciences

With the cells and animal model, many disease models are developed to support the research. Some examples of the model are summarized in the section below, and Table 1.

Multigenic diseases, genetic disorders on the Tet1 and Tet 2 gene, causes unnormal in regulating stem cells, forming blood cell, controlling the brain function, and preventing cancer (Gu et al., 2011). In this case, mouse embryonic stem cells of BDF2 mice were mutant at the Tet1 and Tet2 gene sequences and the difference expression was evaluated (H. Wang et al., 2013). Under the process of the Figure 3 , effects of the mutant condition of the animal, the complex organ, and each tissue (brain, testis, thymus, liver, and tail) are studied (H. Wang et al., 2013; L. Wang et al., 2017).

The aged mice model was also developed for studying age-related diseases and considering for reversing the aging of humans. In this model, as shown in Figure 4, 2 mice are siblings, but one is 150% older due to a gene editing technique (Yang et al., 2023).

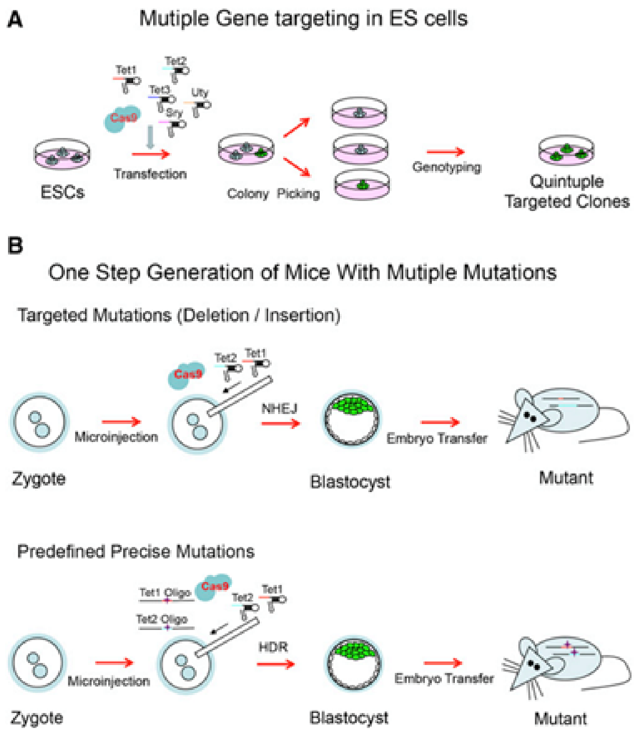

Figure 3. From gene to the cell and animal developing process. Adapted from (Wang et al., 2013)

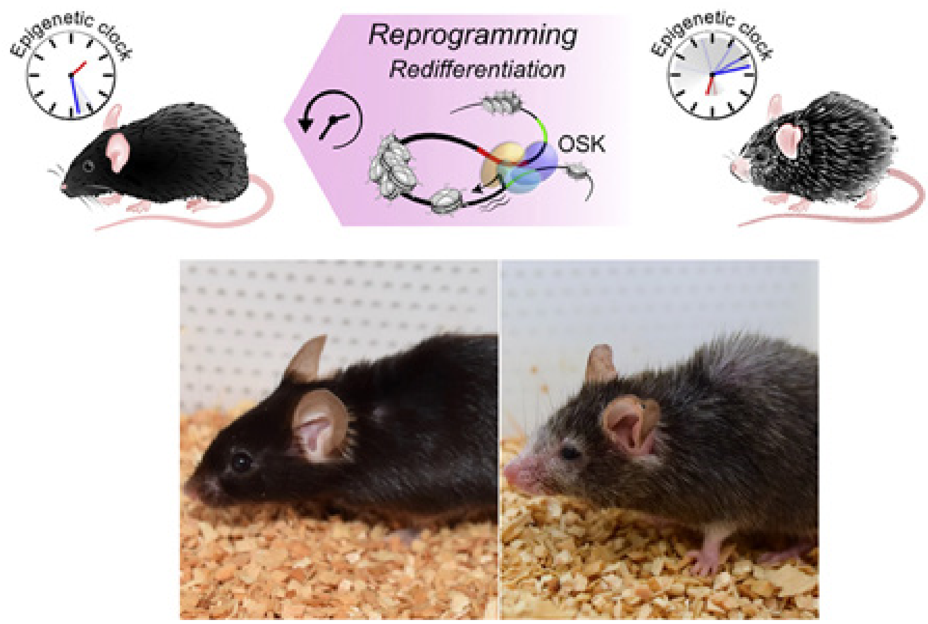

Figure 4. 150% aged mice compared to its siblings (Yang et al., 2023)

The inflammation of the biomaterial (decellularization tissue, hydrogel) is a concern for the researcher and surgeon because to causes the material to loosen, fail, and require replacement. Prioritizing the evaluation of the materials is necessary. To address this issue, an inflammation evaluation model was developed by editing the human leukemia monocytic cell line (THP-1 cells) with 1L-1β tagged HiBiT (Figure 5) (Say et al., 2024b, 2024a; Yabuuchi et al., 2023).

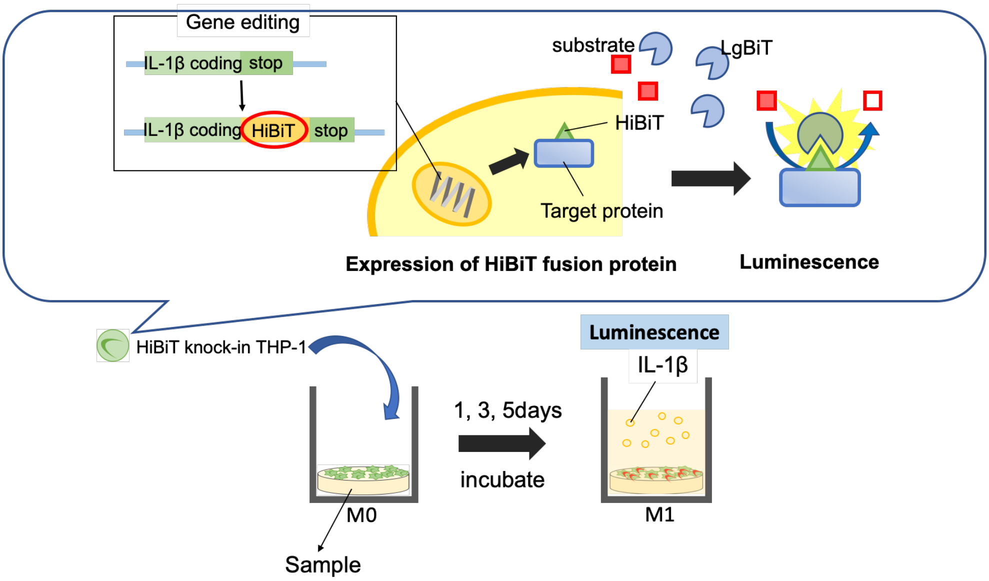

Figure 5. Evaluation of the inflammatory response level of the sample by using the gene editing THP-1 cells (Say et al., 2024b, 2024a; Yabuuchi et al., 2023)

Unusual shapes of the red blood cells (sickle cell disease) are common monogenic diseases causing blockage the blood vessels. The researchers have developed the hematopoietic stem and progenitor cells (HSPCs) by editing the erythroid-specific enhancer region of BCL11A to reduce BCL11A expression, restore γ-globin synthesis, and reactivate production of fetal hemoglobin (Canver et al., 2015; Frangoul et al., 2021; Wu et al., 2019).

Table 1. Summary list of the cell/animal model/ therapy developed under the CRISPR/Cas9 technology

Model	Application	Reference
Human leukemia monocytic cell line (THP-1 cells)	Inflammatory response	(Say et al., 2024b, 2024a; Yabuuchi et al., 2023)
Hematopoietic stem and progenitor cells (HSPCs)	Sickle Cell Disease and β-Thalassemia	(Canver et al., 2015; Frangoul et al., 2021; Wu et al., 2019)
Mice	Liver metabolic disorder	(Pankowicz et al., 2016)
Mouse embryonic stem (ES) cells/ BDF2 Mice	Multigenic diseases	(Wang et al., 2013;Wang et al., 2017)
C57BL/6N mice	Lung adenocarcinoma (Lung cancer)	(Platt et al., 2014)

The metabolic disorders of the liver caused the unnormal in triglyceride metabolism, glucose homeostasis, cholesterol synthesis, or the urea cycle. Patients require drug therapy or orthotopic liver transplantation. Reprogramming the metabolic pathways is another solution. In this case con HT-I (Fah–/–) gene is converted into HT-IIIa (Pankowicz et al., 2016). It is also used in the gene therapy technology by reprogramming the hematopoietic stem cells (HSC cells) and T cells (T lymphocytes) Figure 6.
Embryonic stem cells of C57BL/6N mice were mutated p53, Lkb1, resulting in the development of macroscopic tumor of adenocarcinoma (Platt et al., 2014)

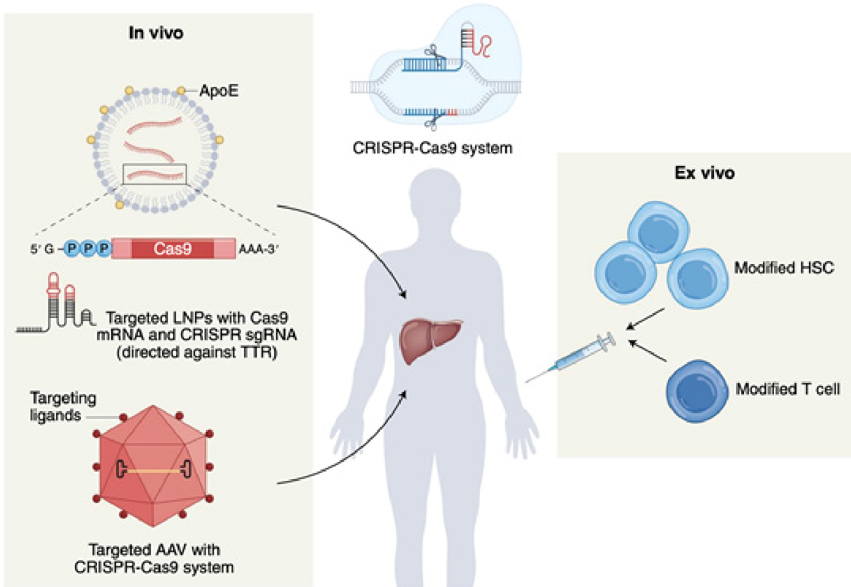

Figure 6. In vivo and ex vivo of CRISPR–Cas9-based gene editing for gene therapy (Büning & Schambach, 2021)

5. Summary of the Application of the CRISPR/Cas9

With the summary above, the CRISPR/Cas9 technology is being studied by researchers in various fields and applications. Some research has already reached the clinical trial, especially for the gene mutation. With this insight, the novel analysis tool and the therapy are expected to be discovered in the near future.

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