Instituto Nacional
de Ciências e Tecnologia de Timor-Leste

Relatório final de
Investigação Científica INCT 2023

Formalin (CH_2O) Contamination in Seafood and Frozen Meat Imported into Timor-Leste

Rui Daniel de Carvalho

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Área de Conhecimento: SAÚDE E BEM-ESTAR

Estudo Elaborado por:

Investigador Responsável : Rui Daniel de Carvalho

Investigador 1 : Acacio Cardoso Amaral

Investigador 2 : Stefany S.A. Fernandez

Investigador 3 : Graciano Soares Gomes

Investigador 4 : Lourenca Mendonça

Dili, 12 December 2023

Declaração

Nome do Investigador Responsável	:	Rui Daniel de Carvalho
Endereço Eletrónico	:	danielruicarvalho@gmail.com
Telefone/Telemóvel		+670 78539140
Número de Bilhete de Identidade	:	
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Abstract

Formalin Contamination in Seafood and Frozen Meat Imported into Timor-Leste

Studies were carried out to detect formalin contamination and the levels of formalin in imported seafood, frozen meatand a few locally produced food. Imported seafood and frozen meat samples from supermarkets situated within Dili city were purchased and tested using Colorimetric determination with color card and sliding comparator. Formalin were detected in all the supermarket surveyed and also in the locally produced seafood. Meat and fish are contaminated at different level of contamination. The highest contamination (50%) was found in fish with 13 samples positive (2 samples were contaminated at the level of 2.25 mg/kg; 1 sample at the level of 7.2 mg/kg, 2 samples at the level of 9 mg/kg, 5 samples at the level of 13.5 mg/kg, one sample at the level of 17 mg/kg and 2 sample at the level of 25.5 mg/kg). The second highest (23.1%) level of contamination of formalin was found in chicken with 6 samples positive with different level of contamination (one sample at the level of 0.9 mg/kg, 2 samples at the level of 3.6 mg/kg, one sample at the level of 5.40, one sample at the level of 7.2 mg/kg, and one sample at the level of 9 mg/kg). The third most contaminated of formalin was detected in prawn/shrimp with 3 samples postive at different level of contamination (one sample at 2.25 mg/kg and 2 samples at the level of 13.50 mg/kg). Whereas pork – 0.4 and 5.4 mg/kg, sausage - 9 mg/kg and nuget – 3.6 mg/kg). The level of contamination mostly above the maximum limit of 5 mg/kg and pose a threat to public health in Timor-Leste.

Keywords: Formaldehide, formalin, fish, meat

Resumo

Contaminação com formalina em marisco e carne congelada importados para Timor-Leste

Foram realizados estudos para detectar a contaminação por formaldeído e os níveis de formaldeído em marisco importado, carne congelada e alguns alimentos produzidos localmente. Amostras de frutos do mar importados e carne congelada de supermercados situados na cidade de Díli foram adquiridas e testadas usando determinação colorimétrica com cartão colorido e comparador deslizante. A formalina foi detectada em todos os supermercados pesquisados e também nos frutos do mar produzidos localmente. Carne e peixe estão contaminados em diferentes níveis de contaminação. A maior contaminação (50%) foi encontrada em peixes com 13 amostras positivas (2 amostras foram contaminadas ao nível de 2,25 Mg/Kg; 1 amostra ao nível de 7,2 mg/kg, 2 amostras ao nível de 9 mg/kg, 5 amostras ao nível de 13,5 mg/kg, uma amostra ao nível de 17 mg/kg e 2 amostras ao nível de 25,5 mg/kg). O segundo maior nível (23,1%) de contaminação por formaldeído foi encontrado em frango com 6 amostras positivas com diferentes níveis de contaminação (uma amostra no nível de 0,9 mg/kg, 2 amostras no nível de 3,6 mg/kg, uma amostra no nível de 5.40, uma amostra ao nível de 7,2 mg/kg e uma amostra ao nível de 9 mg/kg). O terceiro mais contaminado de formaldeído foi detectado em camarão/camarão com 3 amostras positivas em diferentes níveis de contaminação (uma amostra a 2,25 mg/kg e 2 amostras a nível de 13,50 mg/kg). Enquanto carne suína – 0,4 e 5,4 mg/kg, salsicha – 9 mg/kg e pepita/nuget – 3,6 mg/kg). O nível de contaminação ultrapassa principalmente o limite máximo de 5 mg/kg e representa uma ameaça para a saúde pública em Timor-Leste.

Palavra save: Formaldehyde, formalina, Carne, Peixe

List abreviation

Df	:	Dilution factor
Gr	:	Gram
H-CHO/ CH_2O	:	Formaldehyde/Formalin
INCT	:	Instituto Nacional de Ciências e Tecnologia
INFORDEPE	:	Instituto Nacional de Formação de Docentes e Profissionais da Educação de Timor-Leste
IPB	:	Instituto Politecnico de Betano
Mg	:	Mili gram
mL	:	Mili liter
PPM	:	Part per million
POLTEKES	:	Politeknik Kesehatan
TL	:	Timor-Leste
UNTL	:	Universidade Nacional Timor Lorosa’e

I. Introduction

1.1 Contextualization

Any material that an organism consumes for sustenance is considered food. Food often comes from plants, animals, or fungi and is packed with vital elements like vitamins, minerals, lipids, and carbohydrates. Even though it frequently goes unnoticed, maintaining food safety is crucial for sustainability and the expansion of every country's economy. Every person who handles food has a responsibility to practice the finest food safety practices. Knowing the common sources of food safety hazards is one of the essential components of this daily goal.

As the level of contamination in food products rises, there is a greater demand for study on food safety worldwide. In order to guarantee safety in the case of food production, distribution, and preparation, it is imperative to preserve human health. Food contamination, a major issue in many parts of the world, can cause a variety of harmful health outcomes, such as developmental defects, chronic diseases, or even death. There have been rumors in Timor-Leste for years that imported meat and fish were preserved in formalin, but these suspicions have never been tested to determine whether the information was true or just rumors. Therefore, this study aims to detect whether imported meat and fish are preserved in formalin. The results of this study will be useful to consumers, researchers, stakeholders and legal authorities as evidence to strengthen food safety policies in the context of ensuring public health.

1.2 Literature revew

The quality of the meat, whether beef, lamb, pork, poultry or fish, must correspond to consumer expectations regarding sanitary, nutritional and organoleptic quality attributes, to have a fair price. When purchasing one of these meats, the consumer must be well informed and assume that the meat comes from healthy animals, slaughtered and processed hygienically, and that this condition has been subject to rigorous examination, is rich in necessary nutrients, has an appearance typical of the species to which it belongs, very palatable, and these premises are synonymous with obvious quality. Another category of characteristics is called attractive quality and free from any

contamination. By definition, attractive quality includes attributes that can surprise the consumer by offering something more that other competitors are not able to offer (Felício 1998). In general, what is an attractive quality today will soon be an obvious quality, and therefore anyone who wants to stay ahead of the competition needs to always be innovating.

Although meat is often present in the daily diet of the Timorese, the majority of the meat consumed comes from market products whose nutrient quality is obviously unknown, including some contamination in the packaging process. Timor-Leste (TL) is one of the countries with the highest import rate of beef, pork, chicken and fish. Brazil is the largest supplier, in addition to Australia and China among others. An important aspect to be observed when selling meat products of animal origin is maintaining the appropriate temperature for each food (Lundgren et al. 2009). Meat, fish, milk and dairy products, when exposed to inappropriate temperatures, can change quickly, especially in tropical regions where temperatures are high during the summer, requiring rigorous control to guarantee the quality of these products.

Statistics from the World Health Organization (2001) prove that food-borne diseases are considered the biggest public health problem worldwide, with food handlers being considered one of the main vehicles of contamination, given that their participation reaches up to 26% of contaminating sources. In 2011, Timor-Leste (TL) imported 2,937 MT of frozen whole chicken and 2,764 MT of fresh and frozen meat (Young 2013). The 174th largest importer of processed meat in the world, and grossed $1.2M in 2020. The top five countries from which TL imports other processed/prepared meats are: China ($606k), Brazil ($243k), Singapore ($ 124 thousand), Malaysia ($88.4 thousand) and Indonesia ($64.1 thousand)[1].

Meat is a high-quality source of protein and is essential for human survival. Nutrient-rich meats are essential for maintaining human health. Meats, however, are easily spoiled or damaged. This damage is caused by microorganisms found in food or when meat is contaminated. As meat is perishable, preservation methods are necessary. The preservation of meat through the use of chemicals is an example. It is inevitable that people will continue to use formalin to preserve meat and other food products in the

[1] https://oec.world/en/profile/bilateral-product/other-prepared-meat/reporter/tls

future. Commonly formaldehyde aqueous solution in the market sales as formalin (Zhang et al. 2017). Formaldehyde is highly soluble in water, as well as most organic solvents, and is a highly reactive molecule that can irritate tissues upon direct contact. Formaldehyde can be absorbed after inhalation, dermal and oral exposure and the amount of absorption depends on the route of exposure.

Formalin from corpses or meat products is a substance that has been used to preserve food. As formalin is not categorized as a food additive, using it in food can have negative health effects. Acute effects of formalin are rare to occur, but the body typically experiences severe abdominal discomfort, vomiting, coma, nephritic damage, and even death from an excessive amount of formalin (Suwanaruang 2018). Research using test on animals also shows that there is congestion of blood vessels, degeneration of collagen fibers and alveolar matrix around the alveolar bone were significantly different between the control and formaldehyde groups (Laçin et al., 2019).

Formalin or formaldehyde can be used as pesticides, fungicides, preservatives and decolorizers in the food industry (Zhang et al. 2017). Formalin also used in household products, glues and as an industrial fungicide, germicide and disinfectant. It is commonly used as a preservative in morgues and tissue fixation in medical laboratories (Laçin et al., 2019). Some studies have indicated that formalin contamination in foods is common. For example, a study in Bangladesh indicated that almost 50% of fish samples contain formalin (Uddin et al. 2011). Formalin contamination in food is also a national problem in Indonesia (Adlim et al. 2011). No more than 0.2 mg/kg of body weight of formalin per day is recommended by the U.S. Environmental Protection Agency (Zhang et al. 2017).

1.3 Formulation of problems

There is still formalin found in many types of food, according to evidence from other nations. The effects of eating food contaminated with formalin include nausea, vomiting, diarrhea, and even death. In Timor-Leste there has not been a research to prove if our meat (imported and locals) are really free from formalin contamination.

1.4 Formulation of hypothesis

The hypothesis for this research is that “seafood and meat imported into Timor-Leste is contaminated with formalin.

1.5 Objetives

The general purpose of this work is to evaluate formalin contamination of imported seafood and meat stored under refrigeration. The specific objective of this research is to examine formalin contamination in the following products: Chicken meat, Beef, pork, meat, Fish and prawns. The objective of testing formalin in food can be summarized as follows:

- Safety Assurance: The primary objective is to ensure that food products are free from excessive levels of formalin, which is a toxic substance. The testing helps identify any potential health risks associated with the consumption of formalin-contaminated food and ensures consumer safety.
- Compliance with Regulations: Many countries have established legal limits or regulations regarding the acceptable levels of formalin in food. Testing allows food producers and regulatory authorities to verify compliance with these regulations, ensuring that food products meet the necessary standards.\n\n3. Quality Control: Formalin can be added fraudulently to food products as a preservative or to enhance their appearance. Testing helps detect and prevent such adulteration, ensuring that consumers receive the quality of food they expect and preventing potential deception.
- International Trade Facilitation: Testing formalin in food products aids in complying with international standards and regulations for export and import purposes. It ensures that food items meet the requirements of different countries, promoting trade and facilitating the smooth movement of food products across borders.\n\n5. Industry Accountability: Formalin testing establishes accountability within the food industry. By identifying any presence of formalin and taking appropriate measures, businesses can demonstrate their commitment to consumer health and safety. It helps maintain trust and credibility in the industry as a whole
- In summary, the objective of testing formalin in food is to safeguard consumer health, ensure regulatory compliance, maintain food quality, facilitate international trade, and foster accountability within the food industry.

1.6 The importance of the investigation

Formalin testing in food is important for several reasons:

- Consumer Safety:
    - Formalin is a toxic substance that can have harmful effects on human health, such as irritations to the eyes, nose, throat, and respiratory system, as well as potential carcinogenic properties.
    - By conducting formalin testing, food authorities can ensure that food products are free from excessive levels of formalin and are safe for consumption.
- Quality Assurance:
    - Formalin can be added to certain food products as a preservative or as a deceptive practice to enhance visual appearance.
    - By implementing formalin testing, regulatory bodies can verify the accuracy of food labeling claims and ensure that consumers are not misled by fraudulent practices.
- International Trade and Regulations:
    - Many countries have set maximum limits for formalin levels in food products. Conducting formalin testing helps food producers comply with international standards and regulations, facilitating the import and export of food products.
- Industry Accountability:
    - Regular formalin testing helps in monitoring adherence to safety and quality guidelines within the food industry.
    - By holding businesses accountable for the presence of formalin in their products, consumer trust is fostered, and the reputation of the industry as a whole is safeguarded.
- Overall, formalin testing in food is crucial for protecting consumer health, ensuring quality standards, complying with regulations, and promoting transparency within the food industry.

1.7 Organization of the research

First proposal was elaborated and submitted to INCT for approval. Upon its approval, an agreement was signed between IPB and Politeknik Kesehatan Kupang for sample testing. While waiting for the approval of trip of the staff of POLTEKES, interviews

were conducted with the help of animal health students of UNTL. This, then followed by purchasing the sample from respective supermarket for laboratory testing.

1.8 Local Geográfico (Local/locais de realização do estudo)

This research was conducted in major supermarket in capital city, Dili.

2 Methodology

2.1 Methodology of research

2.1.1 Site selection method

Selection of supermarkets based on the results of the team's preliminary observation in the city of Dili. Therefore, the largest supermarkets that sell and distribute these products in Timor-Leste will likely be selected as sampling sites. Therefore, the meat and fish to be collected as samples in this study will be main supermarkets or distributors in Dili

2.1.2 Materials and equipments Used

The materials used for the test were frozen meats, such as chicken (sausages, whole chickens, nugets), beef, pork (sausages, pork belly, skin), prawns and fish (dried and frozen). Samples were tested in the Chemistry Laboratory of INFORDEPE.

The equipment used include: tubes, tubes tray, scale to weigh samples, knife to cut samples, filter, paper filter, MQuant pH-Indicator strips (merck), color card scale of formalin, camera, mortar, and sliding comparator

2.1.3 Sample Testing/ Formalin Test method.

All samples were tested for contamination and formalin level using the Merck MColortest Formalin Test Kit. The method was imunocronomatography or colorimetric with a color chart and sliding comparator. This test is equipped with a series of colors in a concentration of 0.1 - 1.5 mg/l.

Colorimetric determination with color card and sliding comparator. Formaldehyde reacts with 4-amino-3-hydrazino-5-mercapto-1,2,4-triazole to form a purple-red tetrazine. The formalin concentration is measured semiquantitatively by visual comparison of the color of the measure- ment solution with the color fields of a color card. Measuring range / color-scale graduation 0.10 - 0.25 - 0.4 - 0.6 - 0.8 - 1.0 - 1.5 mg/l CH_2O.

The procedures of testing are as follows:

1. 5 gram of samples taken from all sites were weighed and placed in a mortar and then grounded. The grounded samples were then extracted in 40mL of distilled water.
2. Calculation of the dilution factor (df) for extraction was calculated using the formula:

   $$df = \frac{\text{sample (gram)} + \text{Aquadest (mL)}}{\text{Sample (gram)}}$$

3. The sample was then filtered to remove large pieces of the sample, then filtered again using filter paper to prevent residue and cloudy filtrate results which can make reading difficult. Then 5 mL of each sample was put into 2 different tubes to be used as a blank solution for comparison or control and a sample solution for testing;
4. 5 mL of the filtrate from each sample was taken and put into 2 different test tubes (tube A for the test solution and tube B for the blank solution). The sample temperature was maintained at the temperature between 15 - 25 °C;
5. The pH of the samples were checked using the MQuant pH-Indicator strips provided. The sample pH must be above 13. If not, then adjust the pH of each sample by adding 5 drops of FO-1 reagent (sodium hydroxide solution) then homogenize;
6. One measuring spoon of FO-2 reagent was added to the test solution;
7. The tube was closed tightly and shaked vigorously for 1 minute;
8. The sample test was then left for 15-90 minutes;
9. The blank tube and sample were then inserted into the sliding comparator. Tube B containing the blank was set at the top of the tube. Tube A containing the sample was set at the bottom then the comparator was placed on the color card provided;
10. The comparator was moved along the color scale until the closest color match was achieved between the two open tubes when viewed from above. The results were indicated by the pointed end of the comparator;

11. If the color of the measurement solution was stronger than the darkest color on the scale, the measurement was repeated using a fresh, diluted sample until a value of less than 1.5 mg/L CH_2O was obtained.

2.1.4 Data analysis method

SPSS version 26 and excell 2010 software were used to input all laboratory test results to perform statistical analysis. The statistics used was descriptive statistics to know the level of formalin contamination in the samples collected.

2.1.5 Research population and sampling

There are about eight major supermarkets in the country. We target those 8 supermarkets plus one extra supermarket to purchase seafood and meat sold independently and purposively without telling the owner of the supermarket (we visit the supermarkets as ordinary consumers). However prior to purchasing we interviewed managers of those supermarket and ask if they are willing to provide information neccesary in relation to this research.

The variabels measured in this research include: formalin contamination according to sites (supermarkets), type of seafoods and meats and country of origin.

2.1.6 Techniques (means) and Data Collection Instruments

Instrument used for this research include: questionnaires. The questions were an open questions. Questionnaire was used to interview respondent of supermarket managers. Respondents were asked only if they want to participate by signing the concern froms. At the same time we also observed their storage system in the containers and also in the refrigerators (if we were allowed).

2.1.7 Data Collection, Data Analysis and Data Transcription

Data collection in this research were conducted as follows:

- First supermarkets owners or managers were interviewed. They were asked if they were willing to participate in the research. If they agreed to participate then a concern forms were signed by the interviewees;
- Afters the interviews, samples were purchased from supermarkets. Researchers went to each supermarket and purchased the products (seafood and meat) that are sold in respective supermarkets without telling the owners;
- After purchasing the products, some portions (5 gr) were tested according to the procedure described in the sampling testing sections;

- After testing all data are entered in excell and imported to SPSS 26 versions to be analysed using descriptive statistics.

3 Result and discussion

3.1 Analysis of Results/Presentation of Data

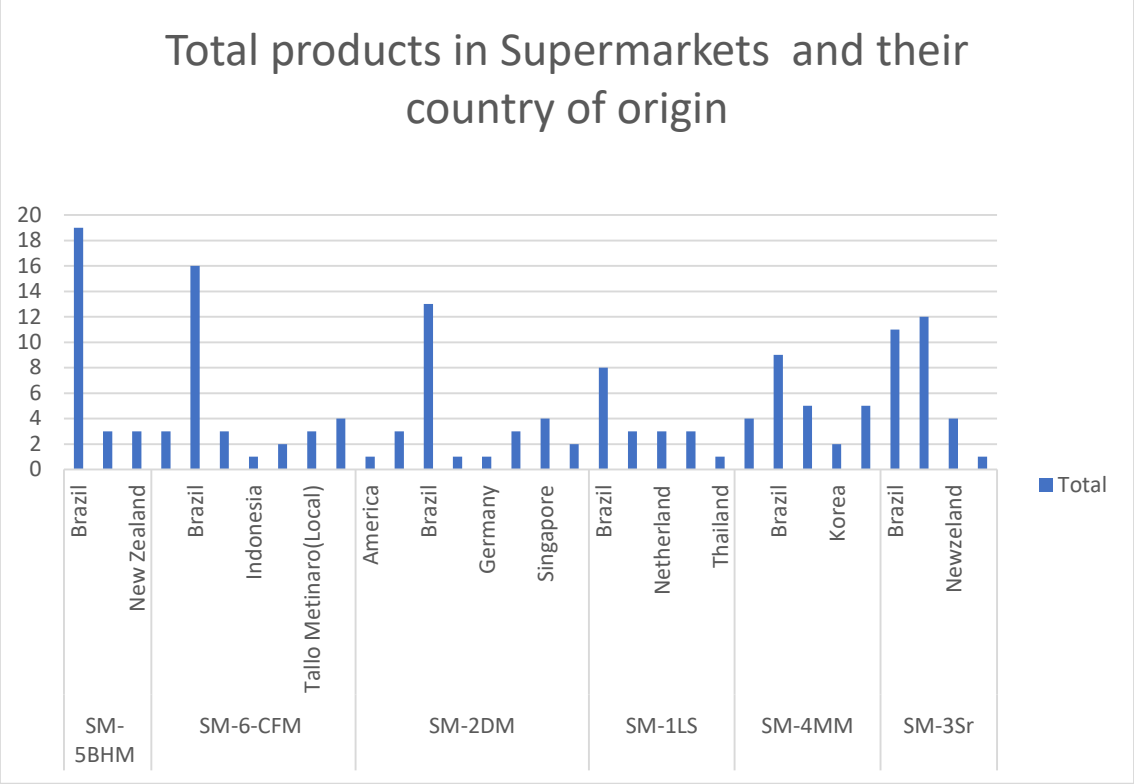


Figure 1. Total products listed during interview, their orogin and super market where they are sold

From our interview with managers of some major supermarkets in Dili, we discovered that these supermarkets imported seafood and meats from several countries. These include Brazil, New Zealand, Indonesia, USA, Germany, Singapore, Netherland, Thailand, and Korea (Figure 1 and Table 1). There are various type of seafood and meats such as: beef (for example tenderloin, striploin, omasum, Fresca minced beef, beef liver, beef cube, etc), pork (pork belly, belly strip, lomboid, palbali, handfeet,etc), chicken (sadia, perdigao, languiro, etc), fish (Mackarel, Tilapia, fish filler, etc), prawns/shrimp (Black tiger), sausage (Borella, perdix, frango soul, etc). For complete list please see appendix 1 and Table 2. The team listed at least one to 19 types of product of seafood and meat from different countries imported into Timor-Leste (Table 1 and Appendix 1).

Table 1. Supermarket visisted and interviewed for the product they sell and their country of origin

Sellers	Products						Exporter
	Porks	Fish	Beefs	Chickens	Sousage	Prawns	
	☑	☑	☑	☑	☑	☐	Brazil
SM-5BHM	☐	☐	☐	☐	☐	☑	Malaysia
	☐	☐	☑	☐	☐	☐	New Zealand
SM-6CFM	☐	☐	☑	☐	☐	☐	Australia
	☑	☐	☐	☑	☑	☐	Brazil
	☐	☑	☐	☐	☐	☐	Chile
	☐	☐	☐	☑	☐	☐	Indonesia
	☐	☐	☐	☐	☐	☑	Singapore
	☐	☐	☑	☐	☐	☐	Tallo Metinaro(Local)
	☐	☑	☐	☐	☐	☐	Vietnam
SM-1LS	☑	☐	☐	☑	☑	☐	Brazil
	☐	☑	☐	☐	☐	☐	China
	☑	☐	☐	☐	☐	☐	Netherland
	☐	☐	☑	☐	☐	☐	New Zealand
	☐	☐	☐	☐	☐	☑	Thailand
SM-4MM	☐	☐	☐	☑	☐	☐	America
	☑	☐	☐	☐	☑	☐	Brazil
	☐	☐	☐	☐	☐	☑	China
	☐	☑	☐	☐	☐	☐	Korea
	☐	☐	☑	☐	☐	☐	Newzeland
	☐	☐	☐	☐	☐	☐	
SM-3SR	☑	☐	☑	☑	☑	☐	Brazil
	☐	☑	☐	☑	☐	☑	Malaysia
	☐	☐	☑	☐	☐	☐	Newzeland
	☑	☐	☐	☐	☐	☐	Portugal
SM-2DM	☑	☐	☐	☑	☐	☐	America
	☐	☐	☑	☐	☐	☐	Australia
	☑	☐	☐	☑	☑	☐	Brazil
	☐	☑	☐	☐	☐	☐	Chile
	☐	☑	☐	☐	☐	☐	Germany
	☐	☐	☑	☐	☐	☐	New Zealand
	☐	☐	☐	☐	☑	☑	Singapore
	☐	☑	☐	☐	☐	☐	Vietnam
SM-9LH	☐	☐	☑	☐	☑	☐	Portugal
	☑	☐	☑	☐	☐	☐	New Zealand
	☑	☐	☑	☑	☑	☐	Brazil

The above Table shows that Super markets imported seafood and meat from various countries. Most of the product (seafood and meat) are imported from Brazil. It shows also diferrent products of seafood and meat that are available from different supermarkets.

Table 2. Length of transportation of imported seafood and meat, storage, distribution and Product handling from surveyed super markets

Description	Supermarket					
	SM-1LS	SM-2DM	SM-4MM	SM-3SR	SM-6CFM	SM-5BHM
Time between country origin to port	No information	2-3 Months	No information	1-2 Weeks	No information	No information
Time between port to supermarket	3 Days	3-4 Days	No information	3 Weeks	No information	No information
Container Temperature	No information	No information	No information	minus 16	No information	No information
Temperature Monitoring	No information	No information	No information	1 Weak	1 Week	No information
Storage system	inside container	Freezer	Freezer	Freezer	Freezer -19° C	Freezer
Time of distribution	No information	Depend on Customer	Depend on Customer	Depend on Customer	1 Month	Depend on Customer
Distributed to	All shops within Dili	All shops within Dili	All shops within Dili	All shops within Dili	only customer	only customer
Distribution data	No information	No information	No	No	No	No information
What is the target Market outside Dili?	All Municipalities	Bobonaro Municipality	All Municipalities	All Municipalities	No information	No information
What did you do when Power off?	Generator	Utilized Freezer	Utilized Freezer	Generator	Utilized Freezer	Utilized Freezer
What happen to the product when power off?	No information	Less than 1 Hour	Less than 1 Hour	No information	Less than 1 Hour	Less than 1 Hour
Do you have refrigerator?	Yes	Yes	Yes	Yes	Yes	Yes
Did your product get demage?	No information	Yes	Yes	Yes	No	No
Reasons for getting damage	No information	Power off too often	Never got demaged	Power off too often	Never got demaged	Never got demaged

From the Table above, it is shown that, not all super market provide data on shipping time. From those who provided, the shipping time from the country of origin varies between one week to 3 months. The time needed from port to supermarket was between 3 days to 3 weeks. The containers tempearture is 16^{o}C. The time of temparture monitoring mostly conducted once a week (Table 2).

Most products are stored in the freezer with the temparature of minus 19^{o}C. Time of distribution depend on the customer, can be up to one month. Most (67%) of imported seafood and meats are distributed to all municpalities.

Most of supermarket depend on electricity provided by government but all super market surveyed reported that they have generators to anticipate power off. Even so, some supermarket owners reported that sometimes their product got damaged due to power outaged (see Table 2)

All supermarket surveyed with the exception of SM-9LH, samples of meat and seafood were positive of formalin contamination. There were 19 samples purchased from SM-1LS of which 42.1% positive. In SM-2DM and SM-3Sr, there were 4 out 11 samples (36.4%) collected were positive of formalin. The rest were 30%, 7.1%, 14.3%, 12.5% and 18.2% for SM-4MM, SM-5BHM, SM-6CFM, SM-7NM and SM-8W respectively. SM-9LH, which is part of SM-1LS was the one without positive samples (see Table 3). From the local market, dried fish from LCM-2TM was the only one tested positive

(20%), whereas Tofu and dried fish from a local shop in Bairopite were negative of formalin contamination (see Table 3).

Table 3. Formalin testing of meat and seafood sold in the local market and supermarket in Dili

Supermarket Code	Products	Negative	Positive	Total	%Positive
SM-1LS	Pork	4	1	5	20.0%
	Chicken	4	2	6	33.3%
	Fish	0	3	3	100.0%
	Prawn/Shrimp	0	1	1	100.0%
	Sausage	3	0	3	0.0%
	Nuget	0	1	1	100.0%
	Total	11	8	19	42.1%
SM-2DM	Pork	2	1	3	33.3%
	Chicken	2	1	3	33.3%
	Fish	0	1	1	100.0%
	Prawn/Shrimp	0	1	1	100.0%
	Beef	3	0	3	0.0%
	Total	7	4	11	36.4%
SM-3SR	Pork	1	0	1	0.0%
	Chicken	2	2	4	50.0%
	Fish	2	2	4	50.0%
	Prawn/Shrimp	1	0	1	0.0%
	Sausage	1	0	1	0.0%
	Total	7	4	11	36.4%
SM-4MM	Pork	2	0	2	0.0%
	Chicken	1	0	1	0.0%
	Fish	3	2	5	40.0%
	Sausage	0	1	1	100.0%
	Beef	1	0	1	0.0%
	Total	7	3	10	30.0%
SM-5BHM	Pork	2	0	2	0.0%
	Chicken	2	0	2	0.0%
	Fish	3	1	4	25.0%
	Prawn/Shrimp	2	0	2	0.0%
	Beef	1	0	1	0.0%
	Squid	1	0	1	0.0%
	Crabs	1	0	1	0.0%
	Eel	1	0	1	0.0%
	Total	13	1	14	7.1%
SM-6CFM	Pork	1	0	1	0.0%
	Chicken	1	0	1	0.0%
	Fish	1	1	2	50.0%
	Sausage	3	0	3	0.0%
	Total	6	1	7	14.3%
SM-7NM	Pork	1	0	1	0.0%

Supermarket Code	Products	Negative	Positive	Total	%Positive
	Chicken	2	0	2	0.0%
	Prawn/Shrimp	0	1	1	100.0%
	Sausage	2	0	2	0.0%
	Nuget	1	0	1	0.0%
	Squid	1	0	1	0.0%
	Total	7	1	8	12.5%
SM-8W	Chicken	5	1	6	16.7%
	Fish	1	1	2	50.0%
	Sausage	1	0	1	0.0%
	Beef	1	0	1	0.0%
	Noodles	1	0	1	0.0%
	Total	9	2	11	18.2%
SM-9LH	Pork	5		5	0.0%
	Fish	1		1	0.0%
	Prawn/Shrimp	3		3	0.0%
	Beef	2		2	0.0%
	Total	11		11	0.0%
LCM-1LFFTL	Fish	7		7	0.0%
	Total	7		7	0.0%
LCM-2TM	Fish	4	2	6	33.3%
	Tofu	4	0	4	0.0%
	Total	8	2	10	20.0%
LCM-3BPM	Fish	1		1	0.0%
	Tofu	1		1	0.0%
	Total	2		2	0.0%
Grand total		95	26	121	21.5%

In this research, the colorimetric determination with color card indicates that 26 samples (21.5%) of our samples (n=121) were positive. Of those that are positive (n=26), most of them have more than 1.5 mg/L HCHO as shown in the following Figure.

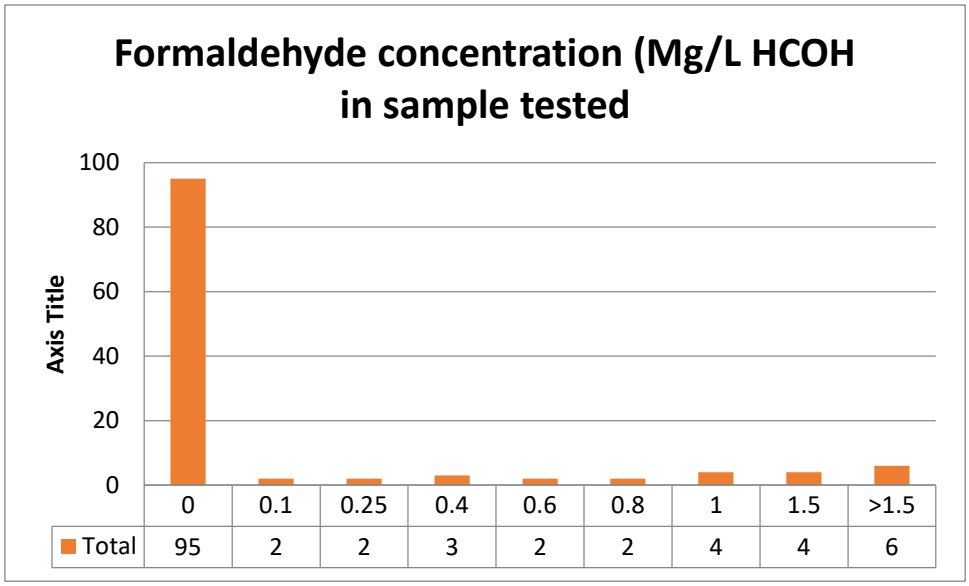


Figure 2. Formalin contamination in meat and seafood in local market and main supermarket in Dili

The concentration of formalin as shown in the above Figure, when it is multiplied with the dilution factor (df) to get part per milion (ppm) or mg/kg of formalin, it was discovered that the contamination of meat and seafood were between 0.4 to 25.5 mg/kg (see the following Figure).

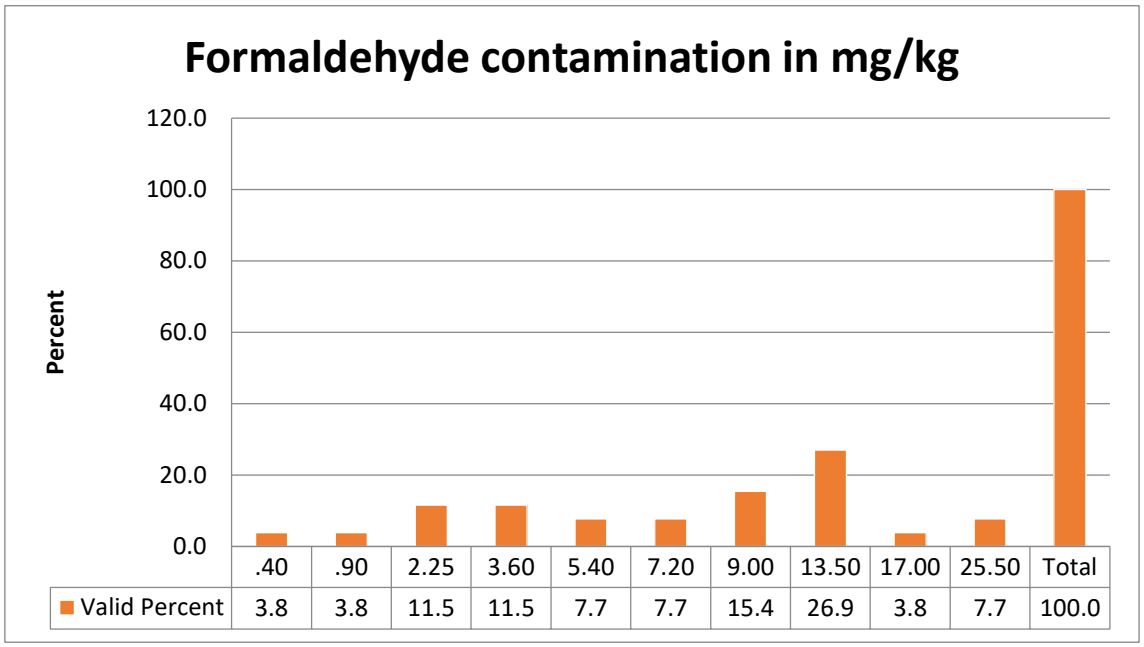
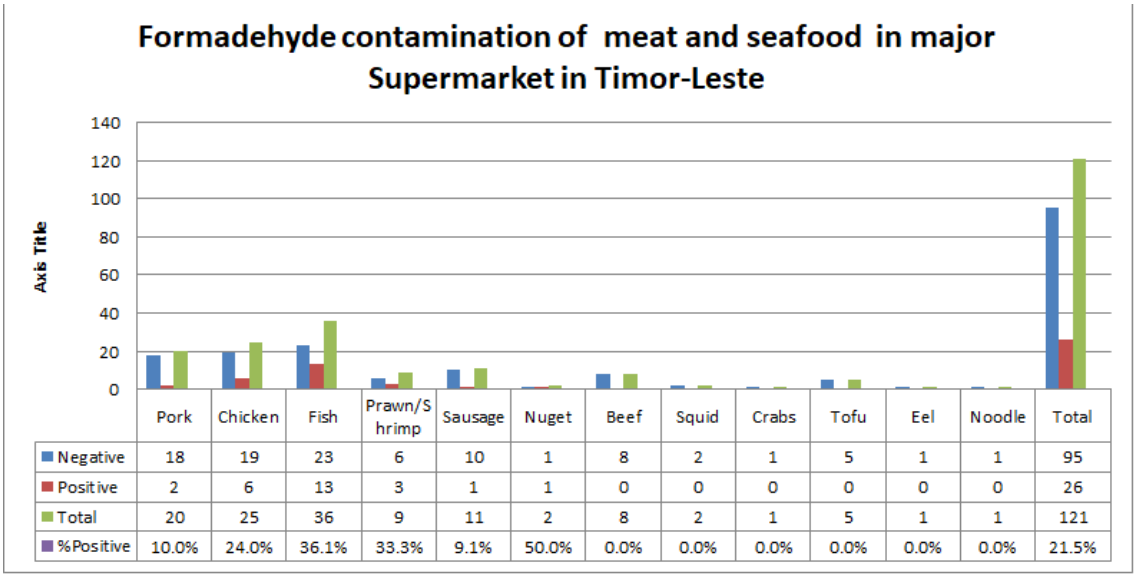


Figure 3. Percentage of formalin contamination of meat and seafood in lokal market and major supermarket in Timor-Leste (posotive samples 21.5%, n 121

Note that if we use the maximum limit of 5mg/kg or 5 ppm, there 69.2% were above the limit and only 30.8% were bellow the maximum limit (see Figure 3).

From the total of 121 samples tested, there are 26 postive samples (21.5%). From the 26 postive samples most (26%) positive with the concentration of 13.50 mg/kg, followed by 15.4% with 9 mg/kg and the least percentage (3.8%) with the concentration of 0.40 mg/kg (see Figure 3).



	Pork	Chicken	Fish	Prawn/Shrimp	Sausage	Nuget	Beef	Squid	Crabs	Tofu	Eel	Noodle	Total
Negative	18	19	23	6	10	1	8	2	1	5	1	1	95
Positive	2	6	13	3	1	1	0	0	0	0	0	0	26
Total	20	25	36	9	11	2	8	2	1	5	1	1	121
%Positive	10.0%	24.0%	36.1%	33.3%	9.1%	50.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	21.5%

Figure 4. Formalin contamination of meat and seafood in mahor supermarket and local market in Dili

The Figure above indicates that the positive samples were from various seafood and meats. The product that are positive include pork (10.0%), chicken (24.0%), fish (36.1%), sausages (9.1%), and nuget (50%). The rest (beef, squid, crabs, tofu, eel and noodles) are negative (see Figure 4). Based on the orgin of the seafood and meat , it was identified in this study that there are 12 countries (see the following table), some of their products (seafood and meat) were positive of formalin except the products that are imported from the nation of C-6Au and C-7Nz

Table 4. Level of contamination in different meat and fish of those products that were tested positive

Level of contamination	Pork	Chicken	Fish	Prawn/ Shrimp	Sausage	Nuget	Total	%Positive
0.4	1	0	0	0	0	0	1	3.8%
0.9	0	1	0	0	0	0	1	3.8%
2.25	0	0	2	1	0	0	3	11.5%
3.60	0	2	0	0	0	1	3	11.5%
5.40	1	1	0	0	0	0	2	7.7%
7.20	0	1	1	0	0	0	2	7.7%
9.00	0	1	2	0	1	0	4	15.4%
13.50	0	0	5	2	0	0	7	26.9%
17.00	0	0	1	0	0	0	1	3.8%
25.50	0	0	2	0	0	0	2	7.7%

Level of contamination	Pork	Chicken	Fish	Prawn/ Shrimp	Sausage	Nuget	Total	%Positive
Total	2	6	13	3	1	1	26	100.0%
%Positive	7.7%	23.1%	50.0%	11.5%	3.8%	3.8%	100.0%	

Meat and fish were contaminated at different level of contamination. The most contaminated product (50%) was found in fish with 13 samples positive (2 samples were contaminated at the level of 2.25 mg/kg; 1 sample at the level of 7.2 mg/kg, 2 samples at the level of 9 mg/kg, 5 samples at the level of 13.5 mg/kg, one sample at the level of 17 mg/kg and 2 sample at the level of 25.5 mg/kg). Example of positive sample of fish is shown in the following Figure).

Figure 5. Fish sample tested positive with 25.5 mg/kg

The second most common (23.1%) level of contamination of formalin was found in chickens with 6 samples positive with different level of contamination (one sample at the level of 0.9 mg/kg, 2 samples at the level of 3.6 mg/kg, one sample at the level of 5.40, one sample at the level of 7.2 mg/kg, and one sample at the level of 9). Example of positive sample of chicken is shown in the Folowing Figure.

Figure 6. Whole chicken sample positive of formalin at the level of 9 mg/kg

The third most contaminated of formalin was detected in prawn/shrimp with 3 samples postive at different level of contamination (one sample at 2.25 mg/kg and 2 samples at the level of 13.50 mg/kg) (see positive sample in Figure 7). Whereas pork – 0.4 and 5.4 mg/kg, sausage - 9 mg/kg and nuget – 3.6 mg/kg) (see Table 4). An example of prawn tested positive of formalin contamination is shown in the following Figure (Figure 7) and an example positive samples of pork is shown in Fugure 8 .

Figure 7. A sample from prawn postive of formalin at the level of 13.5 mg/kg

Figure 8. Pork sausage positive of formadehyde at the level of 9 mg/kg

The following Table shows various product of seafood and meat imported from various countries that are tested. It reveals formalin contamination in some of the seafood and meat imported into Timor-Leste.

Table 5. Formalin tes result of meat and seafood sold in local market and Supermarket in Dili

Country	Products	Negative	Positive	Total	%Positive
1. C-1Br	Pork	12	1	13	7.7%
	Chicken	9	5	14	35.7%
	Sausage	6	0	6	0.0%
	Beef	1	0	1	0.0%
	Eel	1	0	1	0.0%
	Total	29	6	35	17.1%
2. C-2Nt	Pork	2	1	3	33.3%
	Total	2	1	3	33.3%
3. C-3Vt	Fish	2	5	7	71.4%
	Prawn/Shrimp	1	0	1	0.0%
	Total	3	5	8	62.5%
4. C-4Pr	Fish	0	1	1	100.0%
	Prawn/Shrimp	1	0	1	0.0%
	Sausage	2	1	3	33.3%
	Total	3	2	5	40.0%
5. C-5Tr	Chicken	1	0	1	0.0%
	Sausage	1	0	1	0.0%
	Nuget	0	1	1	100.0%
	Total	2	1	3	33.3%
6. C-6Au	Beef	1		1	0.0%
	Total	1		1	0.0%

Country	Products	Negative	Positive	Total	%Positive
7. C-7Nz	Beef	5		5	0.0%
	Total	5		5	0.0%
8. C-8Ml	Chicken	1	0	1	0.0%
	Fish	2	2	4	50.0%
	Total	3	2	5	40.0%
9. C-9In	Chicken	4	0	4	0.0%
	Fish	7	2	9	22.2%
	Prawn/Shrimp	1	0	1	0.0%
	Squid	1	0	1	0.0%
	Noodles	1	0	1	0.0%
	Total	14	2	16	12.5%
10. C-10Ch	Pork	2	0	2	0.0%
	Chicken	1	0	1	0.0%
	Fish	1	1	2	50.0%
	Prawn/Shrimp	0	1	1	100.0%
	Sausage	1	0	1	0.0%
	Nuget	1	0	1	0.0%
	Beef	1	0	1	0.0%
	Squid	1	0	1	0.0%
	Crabs	1	0	1	0.0%
	Total	9	2	11	18.2%
11. C-11Cl	Fish		1	1	100.0%
	Total		1	1	100.0%
12. C-12Tl	Fish	8	0	8	0.0%
	Prawn/Shrimp	0	1	1	100.0%
	Tofu	5	0	5	0.0%
	Total	13	1	14	7.1%
13. C-13Unknown	Pork	2	0	2	0.0%
	Chicken	3	1	4	25.0%
	Fish	3	1	4	25.0%
	Prawn/Shrimp	3	0	3	0.0%
	Total	11	2	13	15.4%
14. C-14-Sg	Prawn/Shrimp		1	1	100.0%
	Total		1	1	100.0%

There are 12 countries that export their meat and seafood to Timor-Leste. These are Brazil, Netherland, Vietnam, Portugal, Turkey, Australia, New Zealnd, Malaysia, Indoensia, China, Chile, and Singapore. There are some samples that are unknown, i.e. not identified by countries, some local products (Table 3 and 5). Based on country of origin, total postive samples of meat and seafood varies from 0 to 100%. However the total samples collected among exporter countries were not the same, so we cannot really compare.

3.2 Discussion of results

Samples of seafood and meat from every supermarket investigated, with the exception of SM-9LH, which is part of SM-1LS, tested positive for formalin contamination. 42.1% of the 19 samples that were purchased from SM-1LS were positive. Four out of the eleven samples (36.4%) that were obtained in SM-2DM and SM-3SR were positive for formalin. For SM-4MM, SM-5BHM, SM-6CFM, SM-7NM, and SM-8W, the remaining percentages were 30%, 7.1%, 14.3%, 12.5%, and 18.2%, respectively, (see Table 3). This is a concern since these supermarkets are the main suplier to municipalities (see Table 1).

We discovered that samples tested from different supermarkets shows the the formalin contamination range from 0.4 mg/kg in pork to 25.5 mg/kg in fish. The fish with this level of formalin contamination (25.5 mg/kg) was imported from the country of C-4Pr. Contamination at this level indicates how serious the formalin contamination of imported seafood and meat into Timor-Leste.

The second most contaminated of formalin was Mackarel fish from the country of C-3Vt with the formalin concentration of 17 mg/kg (see Table 2). This level of contamination is categorised as dangaerous but if we compare to other countries the level of contamination is lower. For example in Thailand, it it was found that Mackarel fish was 288 mg/L (Suwanaruang 2018). But even with this lower level of contamination it is far above the acceptable level of contamination.

Beside fish, frozen chicken was also identified with formalin contamination. Chicken was contaminated ranges from 0.9 mg/kg to 9 mg/kg. There are 3 chicken samples that go beyond 5 mg/kg (two whole chickens from SM-1LS (7.2 & 9 mg/kg) and a chicken leg from SM-3SR (5.4 mg/kg). The finding of formalin contamination by Suwanaruang (2018) in Thailand was much higher (293 mg/L).

There are 3 samples of prawns that were positive with different level of formalin contaminations. The range of contaminations ranges from 2.25 mg/kg to 13.5 mg/kg. From the 3 positive samples, one sample with the contamination level of 2.25 (from SM-2DM, this is a local prawn) and the other two with the contamination level of 13.5 mg/kg from SM-1LS and from SM-7NM supermarket, imported from the country of C-10Ch. In Thailand a researcher identified that the level of formalin contamination of shrimp was 294 – 295 mg/L (Suwanaruang 2018).

There were two pork samples that are positive (0.4 mg/kg from C-2Nt & 5.4 mg/kg from C-1Br) (see Table 2). There is one pork sample that had the formalin contamination more than 5 mg/kg.

There was one sausage sample positive with the formalin contamination of 9 mg/kg (from SM-4MM, imported from C-4Pr) and there was also nuget which has the level of contamination of 3. 6 mg/kg (from SM-1LS imported from the country of C-5Tr).

We discovered that most of our positive tested samples were over 5 mg/kg of formalin. It was reported that, humans who were exposed to high levels of formalin (5–30 mg/kg) may experience or develop bronchitis, chest pain, coughing, wheezing, and asthma symptoms (Islam 2020).

What does it mean for us in Timor-Leste? Do we have some standards of the maximum limit of formalin in food? As far as we know there is none. Let us see what are the maximum limit in some countries for our refrences.

There are some differences in setting the maximum limit of formalin in some countries. According to the U.S. Environmental Protection Agency, formalin should not be consumed in excess of 0.2 mg/kg of body weight each day (Zhang et al. 2017). In the Malaysian Food Regulation of 1985 sets a maximum limit of 5 mg/kg for formalin in fish and fish products (Sahu et al. 2018). In Timor-Leste we do not have any regulation yet in relation to maximum limit of formalin in food. It is in our opinion that the presence of formalin should not be tolerated because it is a carcinogenic agent (Sahu et al. 2018) but if we were to use the maximum limit of 5 ppm or 5 mg/kg, of the 26 postive samples in our research there are 69.2% were above the limit and only 30.8% were bellow the maximum limit (see Table 4, Figure 4). Our data demonstrated that most samples tested have formalin levels higher than the 5 mg/kg recognized safety level.

It is worth noted that, the monitoring of illegally added formalin is challenging since different food sources naturally contain varied amounts of formalin due to post-mortem enzymatic reactions, including seafood (Laly et al. 2018).

Food contamination with formalin is a concerning issue as formalin is a toxic substance that can have adverse health effects on consumers. There are several ways in which food can become contaminated with formalin.

- One potential source of formalin contamination is the intentional addition of formalin as a preservative. In some cases, this may be done to extend the shelf life of certain food products or to enhance their appearance. However, the use of formalin as a preservative is strictly regulated in many countries due to its potential health risks. This could be done in countries of origin or could be done within the country. Whichever it is, an investigation must be done by the relevant authorities;
- Another source of contamination can be attributed to improper handling and processing of food. Formalin can be formed as a result of the breakdown of certain compounds present in food when exposed to heat, such as during cooking or processing. This can be an issue particularly in the case of seafood, as it has higher susceptibility to spoilage compared to other food items;
- Furthermore, formalin can also be present as a result of environmental contamination (Sidoretno et al. 2018). It may come from various sources such as polluted air, water, or contaminated storage facilities. Cross-contamination during transportation, handling, or storage processes can introduce formalin into food products;
- Detecting and preventing formalin contamination is crucial for maintaining food safety. Regular testing for formalin in food products is necessary to ensure compliance with regulatory standards and to protect consumers. Proper handling, storage, and processing techniques are vital in preventing contamination during various stages of the food supply chain;
- Collaboration between food producers, regulatory authorities, and suppliers is essential in addressing formalin contamination. Stakeholders need to work together to establish and enforce strict quality control measures, implement testing protocols, and educate employees on safe food handling practices. Continuous monitoring and surveillance of food products can help identify and prevent potential instances of contamination;
- Ultimately, raising awareness among consumers about the risks associated with formalin contamination is important. Clear and transparent communication is necessary to inform consumers about any incidents, recalls, or precautionary measures taken to ensure their safety.

4 Conclusions and Recommendations

4.1 Conclusions

From the above discusions, it can be conclude that:

a. It is evident that meat and seafood sold in major supermarkets and local market contain formalin. It signifies a potential health hazard and a breach of food safety standards. However we do not know if the contamination is from the countries of origin or it is done in our country;
b. Public health is seriously threatened by the illegal addition of the cancer-causing chemical formalin to food products;
c. Seafood and meat contaminated with formalin is a serious issue that requires continuous vigilance. By implementing strict quality control measures, conducting regular testing, and promoting proper handling practices, the food industry can mitigate the risk of formalin contamination and provide consumers with safe and high-quality food products.

4.2 Recomendations

From this research is was known that some seafood and meat sold in the local market and supermarkers are contaminated with formalin. The following are some reccomendations to relevant Ministries and authoties:

1. Recall and Removal: Promptly initiate a recall of the affected food products from the market to prevent further distribution and consumption. Remove the contaminated products from shelves and dispose of them properly;
2. Investigation and Source Identification: Conduct a thorough investigation to identify the source of formalin contamination (because we do not know if the contamination is from the country of origin or it was done in our country). This may involve examining the production processes, storage conditions, and raw materials used. Identify any potential points of contamination to prevent future occurrences;
3. Communication and Notification: Notify relevant authorities, such as food safety regulatory agencies, about the positive formalin test results. Cooperate fully with their investigations and provide all necessary information.

Communicate transparently with consumers, informing them of the issue, the actions taken, and any potential health risks;

4. Quality Control and Testing Protocols: Strengthen quality control measures to prevent future instances of formalin contamination. Implement stricter testing protocols and conduct regular monitoring of raw materials, production processes, and finished products to ensure compliance with safety standards;
5. Training and Education: Provide training to employees on proper food handling, storage, and processing techniques to minimize the risk of formalin contamination. Emphasize the importance of quality control and adherence to regulations;
6. Collaboration with Suppliers: Collaborate closely with suppliers to ensure the quality and safety of raw materials. Establish strict guidelines for suppliers to follow and conduct regular audits to verify compliance.\n\nConclusion: The presence of formalin in food is a serious issue that requires immediate attention and action. By implementing the recommendations mentioned above, steps can be taken to rectify the situation, prevent any potential harm to consumers, and improve overall food safety practices. Continuous monitoring, transparency, and adherence to regulations are essential to maintain consumer trust in the food industry.

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Appendixes

Appendix 1. Sample testing activities

1

1-Group photo of researchers and Deprtment of Animal Health Student from UNTL who participate in the formalin testing on Septembet 2023

2

2- Sample extraction for formalin testing at Chemistry laboratory of INFORDEPE on September 2023

3

3-Formalin testing- each students was asigned different tasks of formalin testing at Chemistry Laboratory of INFORDEPE

4

4-Data entry or data recording of test result

Appendix 2. Examples of positive samples and their reading measurement

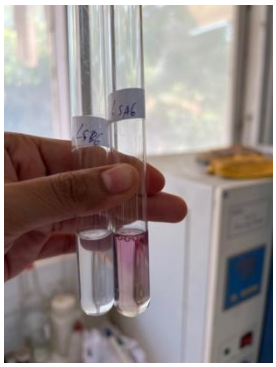
1

1-An example sausage positive of formalin contamination

2

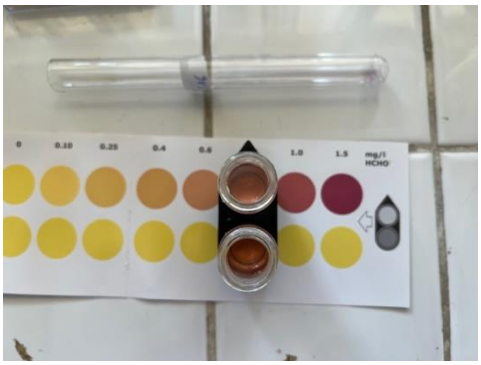
2-An example measurmenr reading of positive sample of sausage

3



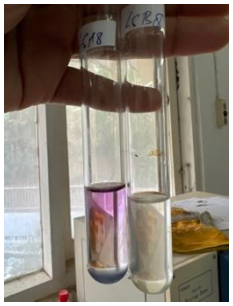
3-An example of frozen chicken sample positive of formalin contamination

4



4- An example measurmenr reading of formalin positive sample of frozen chicken

5



5- An example of fish sample positive of formalin contamination

6

6- An example measurement reading of formalin positive sample of frozen fish

Appendix 3. Preliminary result of research presentation and Presentation to commemorate day of higher education at INFORDEPE

1

1- Preliminary result presentation at INFORDEPE, DILI

2

2- Group photo after preliminary result presentation at INFORDEPE

3

3-Research presentation at Salão Ian Martin at INFORDEPE on the 30th of November 2023

4

4-Group photo with H.E. Ministry of Higher Education after the research presentation on the day of higher education at Salão Ian Martin INFORDEPE on 30th of November 2023