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Determination of Formalin in Food Products Collected in Bamako and Surrounding Area by High Performance Liquid Chromatography at the National Health Laboratory of Mali

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ABSTRACT

Objectives: Food contamination and adulteration are significant problems in Mali. The lack of strict regulatory controls, weak transport, storage and refrigeration infrastructure and increasing consumer demand for fresh produce have led to an increase in fraudulent practices aimed at increasing the shelf life of food products. Formalin, illegally and commonly added to extend the shelf life of food, is carcinogenic and is detrimental to public health.Ingestion of formalin leads to immediate inflammation of the lining of the mouth, throat and gastrointestinal tract and, sometimes, ulceration and necrosis of the mucosal epithelium of the gastrointestinal tract, with possible renal damage and cardiovascular collapse leading to death.

Methods: All meat samples used in this study were sampled in the Bamako district. A total of 26 samples were analyzed out of a total of 78 samples collected.

Results: Among the 26 samples analyzed, 6 revealed the presence of Formalin at varying concentrations. These samples are mostly made up of meat (E5, E13, E14, E60 and E66). Only one sample of smoked fish (E67) could be analyzed before deterioration and revealed the presence of formalin.

Conclusion: This work allowed us to develop a reproducible method for measuring formaldehyde in food (meat and fish). These results clearly raise the issue of food contamination and adulteration in Mali and the need to strengthen regulatory controls, transport, storage and refrigeration infrastructures in the face of growing consumer demand for fresh produce.

Keywords: Formalin,Food products, HPLC, Bamako.

I. INTRODUCTION

Food contamination and adulteration are significant problems in Mali. The lack of strict regulatory controls, weak transportation, storage and refrigeration infrastructure, and increasing consumer demand for fresh produce have led to an increase in fraudulent practices aimed at increasing the shelf life of food products. Food adulteration can have a negative impact on the health of a population, as adulterants can lead to developmental defects, chronic diseases or death. Children, in particular, are more vulnerable to unsafe food and are a major cause of infant mortality [1].

Formaldehyde is carcinogenic and is harmful to public health (International Agency for Research on Cancer (IARC), 2004). The illegal addition of formalin (37% formaldehyde and 14% methanol) to foods to extend their shelf life is considered a common practice [2].

Ingestion of formalin results in immediate inflammation of the lining of the mouth, throat and gastrointestinal tract and, occasionally, ulceration and necrosis of the mucosal epithelium of the gastrointestinal tract, with possible renal damage and cardiovascular collapse leading to death (WHO 1989). Pneumonia, haemorrhagic nephritis and abortion are other effects associated with formalin ingestion [3].

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In 2001, the World Health Organization (WHO) estimated that the presence of formaldehyde should not reach 20 mg/kg for fish and meat (WHO IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2006).

The lack of accurate methods and the ubiquitous presence of formaldehyde in foods make the detection of illegally added formalin difficult. Several analytical techniques have been proposed for the determination of formaldehyde in various food products and water, including HPLC, GC, TLC, spectrophotometry, and other colorimetric and electrochemical techniques. In order to assist regulatory authorities, a sensitive high-performance liquid chromatography method was applied for the quantitative determination of formaldehyde by chemical derivatization with 2,4-dinitrophenylhydrazine (DNPH) in meat, fish, and vegetables at the National Health Laboratory.

II. MATERIAL AND METHOD

1. Samples

All samples used in this study were collected in Bamako district. The collected samples were stored in a freezer at a temperature of -24 °C in the National Health Laboratory. A total of 26 samples were analyzed.

2. Analysis

2.1. Reagents, chemicals and solvents.

Analytical reagents were used throughout the experiment. The reagents used were ultrapure water, 37% formalin, 2,4-dinitrophenylhydrazine (DNPH), HPLC grade acetonitrile.

2.2. Instruments

HPLC INFINITY 1260, OHAUS analytical balance, BRANSON 5510 ultrasonic water bath, EBA 2 centrifuge and SUS 304 grinder.

2.3. Analyticalprocedure

2.3.1. Preparation of the derivatization agent (2, 4-dinitrophenyl hydrazine)

Dissolve 1.5 g of 2,4-dinitrophenyl hydrazine crystals in 50 mL of a 20% solution of sulfuric acid (98%). This solution was prepared immediately before use.

2.3.2. Sample preparation

Grind and homogenize the samples in a blender. Five grams (5g) of the chopped sample were weighed and placed in a conical tube. The homogenized sample was added to 5 mL of ultrapure water. The vial was then capped and sonicated for 40 minutes at room temperature before centrifugation at 6000 rpm for fifteen minutes. The solution was filtered using a 0.45 µm filter, then 2 mL of the filtrate was taken into another conical tube to which 1 mL of DNPH (2,4 dinitrophenyl hydrazine) was added. Then, these flasks were placed in a dark place for 6 hours at room temperature to complete the reaction between formaldehyde and 2,4-dinitrophenylhydrazine to form a yellow or orange precipitate, and then filtered by a 0.45 µm membrane filter. The precipitate was dissolved in 2 ml of acetonitrile. Then filtered through a 0.45 µm filter membrane. [4], [5], [6], [7].

Chromatographic conditions

Parameter	Conditions
Column	Extension C-18 (150 mm x 3.9mm, 5 µm)
Mobilephase	Acetonitrile : Water (60:40)
Detector	DAD
Detectionwavelength	365 nm
Speed	1 ml
Columntemperature	40ºC
Sample volume	20µl

III. RESULTS AND DISCUSSION

A total of 26 samples were analyzed among the 78 samples taken. This is explained by the fact that many samples were degraded due to poor preservation for smoked fish and a break in the cold chain for vegetable samples (lettuce and tomato). Among the 26 samples analyzed, 6 revealed the presence of Formalin at varying concentrations (Table I). These samples mostly consisted of meat (E5, E13, E14, E60 and E66). Only one sample of smoked fish (E67) could be analyzed before deterioration and revealed the presence of Formalin.

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Table I: Description of the samples analyzed.

Samplenumber	Absence or presence of Formalin	Concentration (mg/kg)
Blank	Absence	NA
Samplespikedwith Formol 1	Presence	33.73
Samplespikedwith Formol 2	Presence	38.57
Samplespikedwith Formol 3	Presence	40.83
E1	Absence	NA
E2	Absence	NA
E5	Presence	21.56
E6	Absence	NA
E13	Presence	0.08
E14	Presence	25.74
E20	Absence	NA
E27	Absence	NA
E28	Absence	NA
E29	Absence	NA
E30	Absence	NA
E35	Absence	NA
E42	Absence	NA
E43	Absence	NA
E48	Absence	NA
E50	Absence	NA
E51	Absence	NA
E52	Absence	NA
E53	Absence	NA
E60	Presence	10.11
E61	Absence	NA
E66	Presence	0.14
E67	Presence	22.6
E73	Absence	NA
E75	Absence	NA
E76	Absence	NA

NA : Not Applicable

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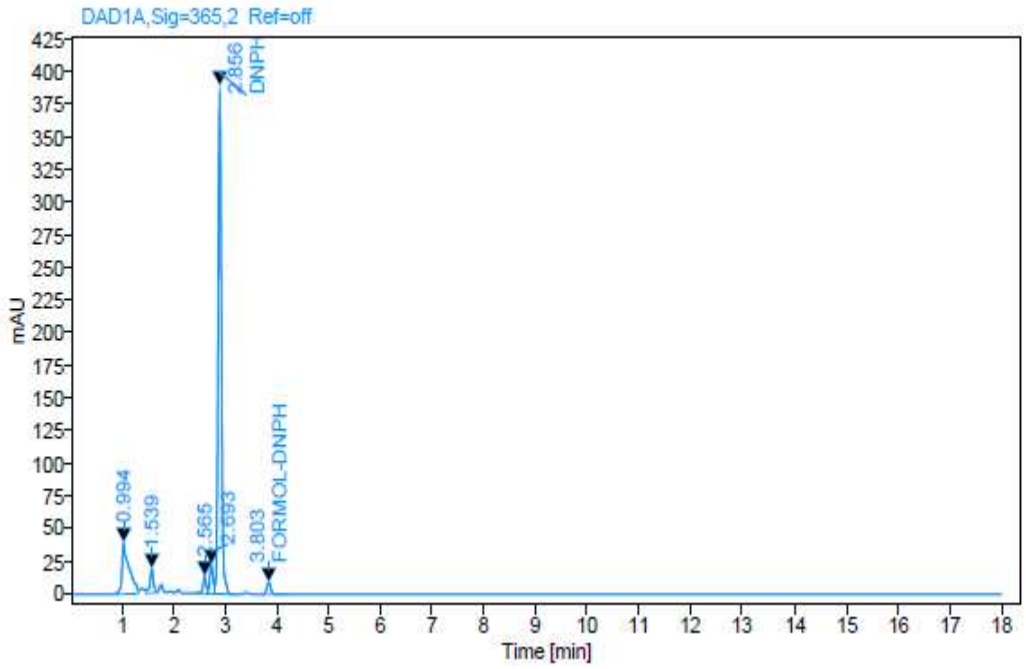

Figure 1 :Chromatogram of a Sample spiked with Formalin identified at a retention time of 3.803 min.

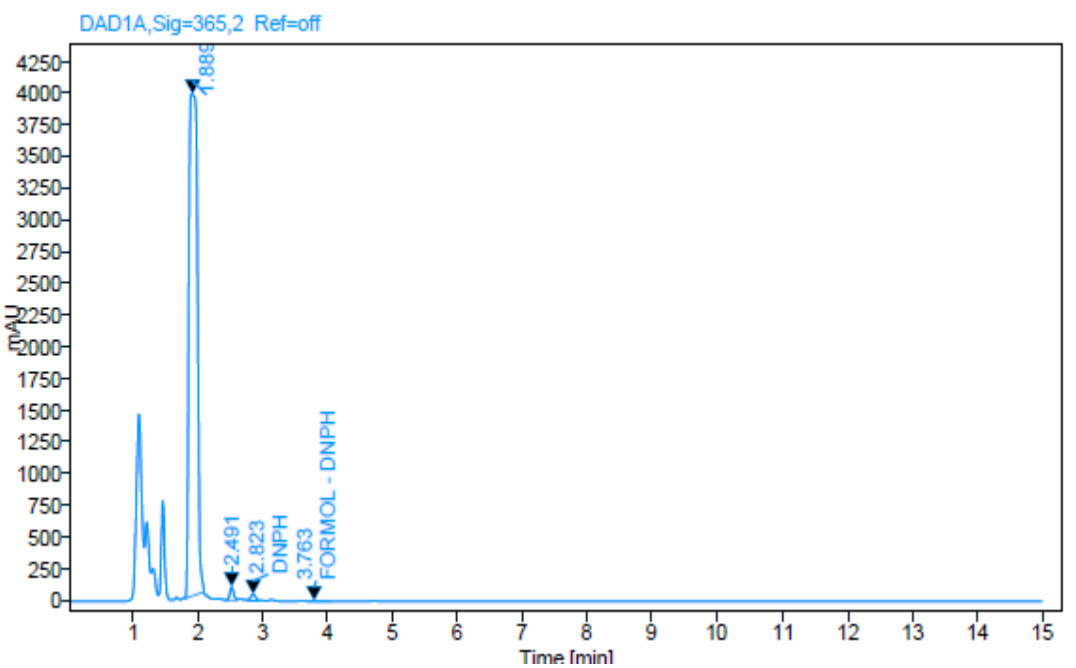

Figure 1: Chromatogram of Sample (E5) identified at a retention time of 3.763 min.

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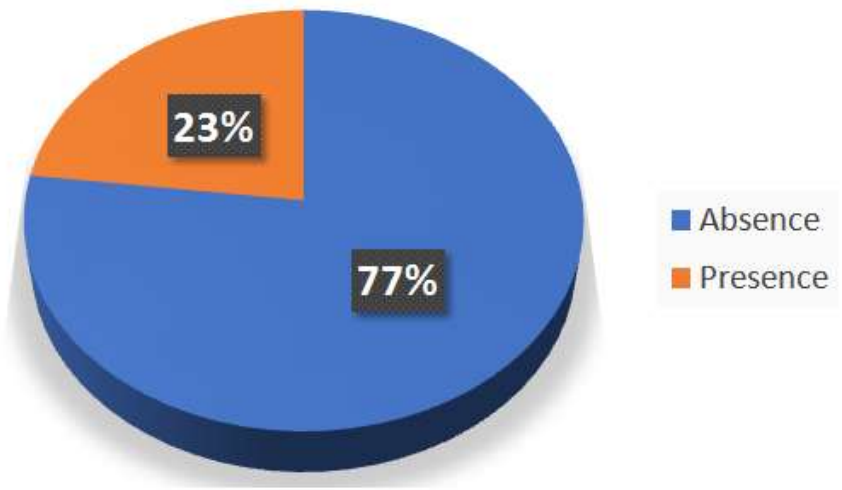

Figure 1: Global result of the analyzed samples

Our results reveal formalin contamination of meat and smoked fish consumed in Bamako, with 23% (6/26) of the samples analyzed, some of which had formalin concentrations above the WHO upper limit of 20 mg/kg for naturally occurring formalin in fish flesh (WHO IARC Working Group on the Evaluation of Carcinogenic Risks to Humans 2006). We further reveal that the highest contamination is associated with samples from Commune I (E5, E14) and N'GabakoroDroit (E67) with (21.56, 25.74 and 22.6 mg/kg) respectively, with a high probability that these samples are contaminated above the permitted limit. On the other hand, we reveal low contamination (0.08, 10.11 and 0.14 mg/kg), associated with samples (E13, E60 and E66) from Commune I, Kati and N'Gabakoro respectively. In addition, the high contamination of the samples obtained could also be linked to the deliberate addition of formalin, among other harmful chemicals, as a preservative to stored foods.

IV. CONCLUSION

A total of 78 samples were taken, of which 26 samples were analyzed. This difference is explained by the deterioration of many samples, including smoked fish, lettuce and tomatoes during storage. Among the samples analyzed, some revealed the presence of formaldehyde contamination to varying degrees.

This work allowed us to develop a reproducible method for measuring formaldehyde in food (meat and fish). These results clearly raise the issue of food contamination and adulteration in Mali and the need to strengthen regulatory controls, transport, storage and refrigeration infrastructure in the face of growing consumer demand for fresh produce.

CONFLICTS OF INTEREST

None, it was in the context of public health and for the well-being of the Malian population in accordance with the mission of the LNS.

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