ADDIS ABABA UNIVERSITY
COLLEGE OF HEALTH SCIENCES
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Risk of Nephrotoxicity among Dry Cleaning Workers Exposed to Perchloroethylene in Addis Ababa, Ethiopia

By: Philemon Mohammed (BSc)

Advisor's name

Teferi Abegaz (PhD)
Meaza Gezu (PhD)

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Name of investigator	Philemon Mohammed
Name of Advisor(s)	1. Teferi Abegaz (PhD) 2. Meaza Gezu (PhD)
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Address of investigator	Tel**:+251933807885** Mail:**philozel734@gmail.com**

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Risk of Nephrotoxicity among Dry Cleaning Workers Exposed to Perchloroethylene in Addis Ababa, Ethiopia: A comparative cross-sectional study

By: Philemon Mohammed (BSc)

Signature _____Date _____

Approved by the examining board

	Signature	**Date**
Chairperson, Department Graduate Committee	_____	_____
Advisors:		
Teferi Abegaz (PhD)	_____	_____
Meaza Gezu (PhD)	_____	_____
Internal examiner:		
Samson Wakuma (PhD)	_____	_____
External Examiner:		
Akeza Awealom (PhD)	_____	_____

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Abbreviations and Acronyms

ACGIH	American Conference of Governmental and Industrial Hygienists
AOR	Adjusted Odds Ratio
BUN	Blood Urea Nitrogen
EPA	Environmental Protection Agency
EU	European Union
GGT	Gamma-Glutamyl Transferase
GSH	Glutathione
GST	Glutathione S-Transferase
LOAEL	Lowest Observed Adverse Effect Level
NOAEL	No Observed Adverse Effect Level
NSAIDs	Non-Steroidal Anti-Inflammatory Drugs
OSHA	Occupational Safety and Health Administration
PCE	Perchloroethylene
PERC	Perchloroethylene
PPE	Personal Protective Equipment
PPM	Parts Per Million
RFD	Reference Dose
TCA	Trichloro acetic Acid
TURI	Toxics Use Reduction Institute
TWA	Time-Weighted Average
USA	United States of America

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Abstract

Background: Perchloroethylene, a solvent commonly used in dry cleaning, have an impact on kidney's health. As the dry-cleaning industry grows in Addis Ababa, it presents an occupational hazard. Limited global research and a lack of African studies on this issue highlight this study to inform policymakers and promote worker health and safety.

Objective: The objective of this study is to measure selected urinary and blood nephrotoxic biomarkers among dry cleaners in Addis Ababa and compare them with hotel laundry workers to see the impact of perchloroethylene on kidney health.

Method: A multi-center comparative cross-sectional study was carried out in 17 dry-cleaning shops and 21 hotel laundries in Addis Ababa, involving 70 participants from each group. The study assessed nephrotoxic biomarkers such as urinary protein, creatinine, calcium, sodium, blood urea nitrogen, and serum creatinine. Data collection involved both biological samples and structured questionnaires administered via Open Data Kit (ODK), with data analysis conducted using SPSS. Statistical methods included t-tests, chi-square tests, correlation and multivariable regression with significance determined at $p < 0.05$.

Result: The result showed that majority of the employees were females in both groups and the risk of nephrotoxicity was higher in dry cleaners as compared to hotel laundry workers, where 88.2% of the abnormal proteinuria, 76.9% of the abnormal creatinine and 38.5% and 62.5% of the abnormal urinary calcium and sodium values were found in dry cleaners respectively. We found a significant mean difference in three biomarkers namely, Total Protein (TPU) with a Median & IQR value of (102 mg/dl &70.75 mg/dl) and (54.5 mg/dl &27.25 mg/dl), Urinary creatinine with a Median & IQR value of (193 mg/dl & 111.06 mg/dl) and (142.93 mg/dl & 78.17 mg/dl) and Urinary Calcium with a Median & IQR value of (2.60 mmol/l & 2.94 mmol/l) and (0.835mmol/l & 0.79 mmol/l) for the exposed and the control groups respectively. However, a significant difference was not found in urinary sodium, Blood urea nitrogen and S. creatinine between the two groups, but higher value of sodium above range and higher BUN within range was observed in dry

cleaners. The Log_{10} transformed TPU and raw BUN showed a significant association with experience of PCE spillage, r (140) = 0.201, **p = 0.017** and r (24) = 0.536, **p = 0.007,** respectively.

Conclusion: The study concluded that dry cleaners faced the highest risk and abnormalities in kidney biomarkers, with factors such as duration of employment, PCE spillage, sex, frequency of handling PCE, and poor ventilation positively correlating with immediate symptoms. Although the absence of precise exposure measurements limited definitive conclusions, the findings indicated that dry cleaners are the most vulnerable group. Therefore, strong safety protocols and regular medical checkups are recommended for workers frequently exposed to Perchloroethylene

Keywords: Perchloroethylene, PCE, Nephrotoxicity, Dry cleaners, Hotel laundry workers, Urinary biomarkers, Serum biomarkers

1. Introduction

1.1 Background

Perchloroethylene (PCE), also known as tetrachloroethylene, which is a colorless, non-flammable liquid solvent primarily used in the dry cleaning industry, has been known to cause significant public health problems (1). This chlorinated hydrocarbon is an important metal-degreasing solvent and an intermediate for other chemical production (2). It is slightly soluble in water, having a boiling point of 121.4 °C, and a higher density than air, which makes good ventilation in an occupational setting mandatory (3).

The application of PCE as a cleaning solvent started in the 20th century (3). Before this, people tended to clean their clothes using non-aqueous solvents like kerosene, gasoline, and benzene, but due to the flammable nature of those solvents, people evolved into using safer alternatives like carbon tetrachloride, trichloroethylene, and then finally arrived at perchloroethylene (4).

Dry cleaners are the most occupationally exposed group to PCE (5). Inhalation is the major route of entry (6). It takes 3 days for half of it to be removed from a human body (1). During its application, a significant amount of PCE is released into the environment, contaminating soil, surface water, and possibly groundwater, as well as the air. It has a half-life of 100 days in the atmosphere (1).

PCE has been identified by the International Agency for Research on Cancer as a probable carcinogen, with a very limited study on humans (4). PCE has both acute and chronic health impacts, causing many health issues affecting the nervous system, kidneys, and liver (1). It is believed to be a nephrotoxic, skin, and respiratory irritant (5).

Kidney toxicity is one of the critical concerns of PCE health effects (6). Chronic exposure to it has been associated with end-stage renal disease (7). Few studies have tried to study the renal effects of PCE among dry cleaners and found inconsistent results (1). Since the practice of dry cleaning is increasing in Ethiopia, it is crucial to understand the extent of its health impacts among dry cleaners to adopt and implement safer working practices.

1.2 Statement of the problem

During the 1990s, the use of perchloroethylene (PCE) increased rapidly, particularly in dry-cleaning shops where it became a commonly used solvent. This trend drew the attention of public health researchers, prompting efforts to assess occupational exposure. Consequently, numerous exposure assessment studies were conducted during this period (12,4).

Many exposure and some health effect studies have been conducted in Western and Asian countries suggesting a link between perchloroethylene (PCE) exposure and kidney toxicity (7, 10, 12). Nephrotoxicity, including both acute kidney injury (AKI) and chronic kidney disease (CKD), is a major global health concern, affecting an estimated 10% of the world's population (13). Research indicates that workers exposed to perchloroethylene (PCE), such as those in the dry-cleaning industry, are at increased risk of developing kidney-related health problems (6). Unfortunately, consistent and conclusive evidence linking PCE exposure to kidney toxicity remains limited. While some studies suggest a possible association, the findings are often inconsistent, highlighting the need for more robust and region-specific research, especially in underrepresented areas like Africa (1,14).

Despite acknowledging limitations in our search coverage, we found no notable studies on PCE exposure and its health effects in Africa or Ethiopia. This highlights a significant knowledge gap, especially as the dry-cleaning sector continues to grow in the region. Even in developed countries, research on the human health impacts of PCE remains limited, often underestimating its potential effects on workers. (12).

In Ethiopia, the increasing number of dry-cleaning shops has raised concerns about occupational exposure to perchloroethylene (PCE), making it an emerging public health issue. This study aims to investigate the relationship between perchloroethylene (PCE) exposure and nephrotoxicity in Ethiopia, where no prior data on this association currently exists. The Westerns claim that they have put the exposure below the recommended level. But here in our case, we don’t have any data, which makes the mitigation impossible without clear evidence. So, to improve the health of dry cleaners by developing tailored solutions in the Ethiopian context, this study will play a crucial role in providing evidence of its first kind and be a landmark for future PCE-related studies.

1.3 Rationale and Significance of the Study

1.3.1 Rationale of the study

The dry cleaning industry has a higher risk of PCE exposure, and the employees there are at risk of developing kidney-related issues (9, 16–19). The lack of documented and reported data about PCE makes it difficult to consider its health impact, which makes it challenging to intervene in the problem.

The American Conference of Governmental and Industrial Hygienists (ACGIH) suggests conducting further research regarding the health effects of PCE (18). A study conducted in Italy highlights the need to do research on the health effects of PCE at even low occupational doses (19).

In Ethiopia, where the dry cleaning industry is expanding, workers are at a high risk due to limited awareness and insufficient protective measures. The lack of comprehensive studies on PCE exposure and its health effects in Ethiopia undermines the need for targeted interventions, which violates the right to safety of employees. So, investigating the health effects of PCE is a crucial step to protect the health and well-being of workers.

1.3.2 Significance of the study

Occupational chemical risk assessment is one of the mandatory tools to protect and promote the health and safety of employees. Particularly in a resource-limited setting like Ethiopia, where managing the consequences of exposure can be challenging, proactive measures are essential. Considering this, carrying out research focusing on exposure-related and actual health impacts of chemicals is the first step to the solution.

This research will investigate PCE exposure risk factors among dry cleaners in Ethiopia and its nephrotoxic effects. And upon its completion, this project hopes to fill the existing knowledge gap and provide evidence-based recommendations for improving occupational health standards in the Ethiopian dry-cleaning industry, additionally, the findings will be crucial for policymakers, health practitioners, and industry stakeholders in implementing effective protective measures.

2. Literature review

2.1 Occupational Sources of Perchloroethylene

Perchloroethylene, usually abbreviated as PCE or PERC, is used in many different sectors, including dry cleaning, coal testing, film cleaning, textile, and several metal degreasing activities (4, 13, 19). The dry cleaning sector is a major occupational source of PCE and is also responsible for discharging the solvent into the atmosphere (1). Workers in dry-cleaning shops, such as machine operators, receptionists, ironers, and spot cleaners face significant inhalation risks, with the level of exposure varying by job role (7). An occupational exposure study conducted in Iran from 69 dry-cleaning shops reported an 8-hr-TWA PCE concentration of 11.5 ppm value in machine operators, 9.6 ppm in pressers and 7.2 ppm in counter clerks (21).

In the USA in 2017, the number of dry-cleaning shops was estimated to surpass 20,000 and 60 % of them used PCE as their primary solvent. (8). An exposure study conducted in there from 44 dry cleaning establishments found a concentration range of 4-149 PPM for machine operators, with a geometric mean of 22 ppm, however, the mean exposure concentration for pressers was relatively low from the Iran study with a value of 3.3 ppm (22). A similar but more advanced exposure study conducted among dry cleaners in Italy, which included biological exposure measurements, reported a time-weighted average (TWA) concentration of 8.4 ppm for PCE from environmental sampling. Additionally, urine samples revealed PCE concentrations of 17.7 mg/L and trichloroacetic acid (TCA) concentrations of 1.03 mg/L (23).

A preliminary exposure study conducted in the UAE found the highest PCE exposure value of 510 ppm while transferring dry-cleaned fabrics from machines to trays before finishing the full cycle of washing. The result exceeded ACGIH Short-Term Exposure Limit (STEL) which is 100 ppm (24).

Although detailed studies have not been conducted, the toxicological profile of perchloroethylene (PCE) identifies several high-risk occupational sources of exposure. These include open tank degreasing and manual cleaning of tanks in metal manufacturing industries, manual spot cleaning, spraying PCE on old textile fabrics, and its use in automotive repair (25,26).

Few studies have mentioned a high amount of PCE exposure in houses and buildings found close to dry cleaning shops. They have also highlighted indoor air pollution of PCE from the slow release of dry cleaned fabrics inside residential houses, many kilometers away from the dry cleaning shops (21, 22)As a result, there have been consecutive updates to minimize the amount of PCE released from the dry-cleaning machines. The aim was to upgrade it from manually transferred wash fabrics to closed-loop ones, which substantially decreased the amount of PCE released (19, 20). Currently, many EU countries and the USA have prohibited using dry cleaning machines below the 4th generation (12, 21).

2.2 Occupational Perchloroethylene Exposure Assessments

2.2.1 Route of Exposure

The most common route of entry for PCE exposure is inhalation. It is readily absorbed in the lungs. The odor of perchloroethylene (PCE) can typically be detected in the air at concentrations ranging from 5 to 50 ppm, which may help identify acute high-dose exposures. However, because of olfactory fatigue and variability in individual sensitivity, smell is not a reliable indicator for chronic or low-level exposure, limiting its usefulness as a long-term safety measure.(27–29).

Ingestion of perchloroethylene (PCE) leads to rapid absorption and can cause systemic toxicity similar to that from inhalation, making it a serious health concern. Currently, there is no specific antidote available for PCE poisoning in clinical management. Dermal contact is another route of exposure; while systemic toxicity through the skin is less likely, secondary contamination can occur if PCE is spilled onto clothing or skin, posing additional health risks.(28).

2.2.2 Air Sampling

Different studies have conducted personal and environmental air sampling to quantify PCE exposure(5, 29, 30). A study conducted in Italy used personal diffusive samplers (Radiello) for collection and gas chromatography for analysis to conduct personal PCE monitoring among 71 dry cleaners and found a mean value of 52.32 mg/m^3 (30). A similar study conducted in the country in a specific district called Modena found a PCE mean value of 10.6 mg/m3 using a personal sampling SKC 575 series for a full work shift (19).

In 2008, research was conducted in Ohio as a pilot study to see the exposure level of PCE in 18 female dry cleaners using personal monitoring with a charcoal tube for 2 days on Wednesday and Thursday coming up with a result of 2.41 PPM of PCE in consideration to the fact that all shops use 4th generation machines (31). Directing instantaneous monitoring has also been applied as a subordinate measurement in one study using a colorimetric gas tube to identify peak exposures in the shop (30).

2.2.3 Biological Monitoring

Several studies have tried to measure the exposure status of PCE in dry cleaners using biological indicators and found them reliable and effective (33). ACGHI recommends exhaled air and blood as biological indices to measure PCE exposure (34). However, different noninvasive exposure assessment studies have recommended to use of urine samples as an alternative for PCE exposure(5, 16, 36, 42). The public health statement of PCE tells us that the detection of PCE and its metabolites in urine indicates acute exposure (1).

A study conducted in Italy (Padua and province) tried to assess the most reliable bio-indicator of PCE exposure by correlating them with personal air sampling, as a result, PCE in blood, PCE, and TCA in urine were evaluated on Thursday evening and Friday morning. And found a mean value of 0.024 PCE in urine (mg/l) end-shift (Thursday evening) and 0.304 mg/l in blood samples. The study noted that the smaller the dry-cleaning shop, the greater the pollution, and a statistically significant association was found between environmental PCE exposure and urine end shift exposure with r=0.68, but found no significant association with TCA. Accordingly, we can conclude that PCE in urine at the end of the shift and PCE in blood at the beginning and end of the shift are the most dependable markers for biological surveillance (30).

In 2019, another study conducted in Italy among dry cleaners to see if the trend of occupational PCE exposure is decreasing found a positive result with a urinary PCE of 0.0084 mg/l of mean value and a correlation of 0.81 with environmental exposure assessment (19). Another biological exposure assessment study highlights that exhaled air is a good bio-indicator of PCE. It has the highest correlation with environmental monitoring exposure, giving r value of 0.931 (35).

A study in Ohio used blood, urine, and exhaled air specimens to assess the exposure of PCE in dry cleaners and suggested that although blood is a good indicator, it is also the most challenging one to collect since it is invasive (31).

In Indonesia, different biological monitoring methods were used related to PCE. The researchers conducted an epithelial cell collection in buccal cells to see if PCE, being genotoxic, affects the frequency of micronuclei in mucosal epithelium (36).

2.3 Health Effects of Perchloroethylene

Epidemiological health research regarding PCE started in 1977 when the US National Institute of Cancer identified liver tumors in mice caused by PCE (37).

The health impact of PCE is adverse, especially for those exposed in occupational settings, however, its impact on the community and the environment is not negligible. Years and even months of exposure have been associated with neuropsychological and color vision problems (14). Even at low doses, neurotoxicity is considered the most common and adverse non-cancer effect (38). Exposure to extremely high amounts of tetrachloroethylene can result in hematuria, proteinuria, oliguria renal failure (28).

Acute PCE exposure can cause headache, dizziness, and, in severe cases, unconsciousness, highlighting its neurotoxic risks.(39). In the past, PCE was used as a human anesthetic at doses above 1500 ppm, though this practice is now obsolete due to its toxic effects (3). Prolonged low-dose exposure to PCE is associated with chronic effects such as vision problems, mood swings, and memory loss (1).

Some occupational surveys and research have investigated the potential kidney and liver effects of PCE but found no clear and consistent results. Acute exposure of PCE was estimated to be 3000 mg/kg for oral LD_{50}, 10,000 mg/kg for dermal $LD_{50,}$ and > 3000 ppm for inhalation LC_{50} (3).
A positive association between low PCE exposure in dry cleaners and bladder and kidney cancer has been found, although those results were also inconsistent (40).

In animal experiments, high dosages of PCE (usually 300–1000 mg/kg/d) administered over an extended period of time resulted in renal disease (41). Nevertheless, a study conducted on dry cleaners exposed to PCE was unable to show a consistent correlation between exposure and kidney dysfunction markers such as albumin, urine protein, and N-acetyl-glucosaminidase (NAG) levels (16).

Even though the impact of PCE in a community is not well studied, prenatal and childhood exposure to it is believed to cause cognitive effects, bipolar disorder, PTSD, along with cancer (42–44).

2.4 Metabolism of Perchloroethylene

The impact of PCE on human health is mainly related to the period of exposure and the concentration of it in the environment (7). The metabolism of PCE in the human body takes two pathways (14). The first one is oxidative metabolism, which involves P_{450} to convert it into TCA(trichloroacetic acid) and others, causing liver and kidney toxicity (45). The second metabolism is GSH (glutathione conjugation), which results in nephrotoxic metabolites and causes acute proximal tubal injury and kidney damage (46). but the effect of GSH metabolites on kidney toxicity was not assured, but rather hypothesized (14).

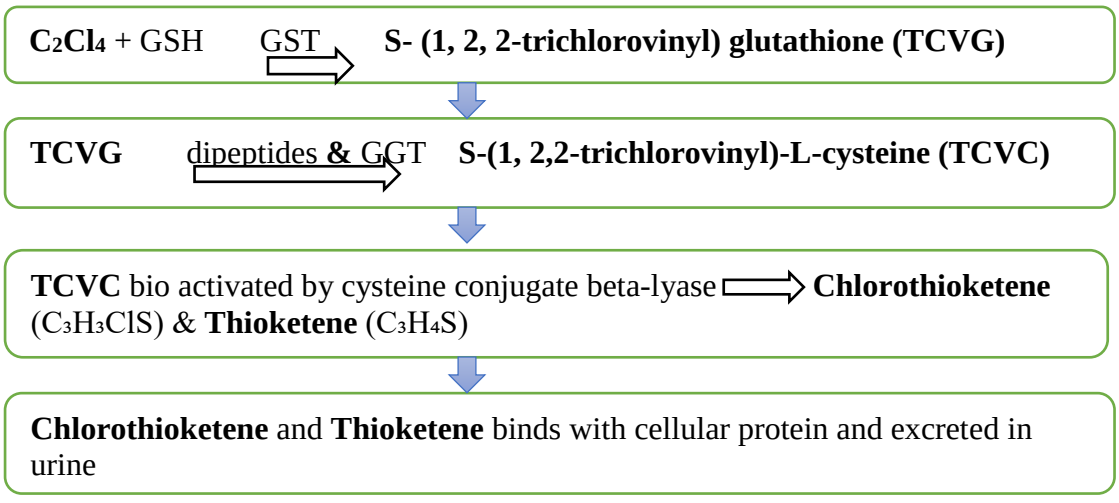

Figure 1Perchloroethylene Glutathione conjugation metabolism pathway adopted from (47,48)

The main route of both absorption and elimination of PCE in an occupational setting is through the respiratory system. 80-95% of inhaled air is removed through expiration without going through any metabolism, and 3% of it is excreted in urine (29). PCE metabolism is affected by age, sex, body adipose tissue, and energy expenditure (40). For this reason, it is better to assess biological exposure measurements.

2.5 Nephrotoxic Effect of Perchloroethylene

Kidney toxicity due to PCE exposure has been identified and suggested in some clinical research(35, 38). But remains unclear and requires extensive research (49). Some studies have found signs of kidney toxicity in workers exposed to PCE for a long time. As the dose of PCE exposure increases, the time required to show the effect decreases(44, 45). On the contrary, there was this study on dry cleaners that assessed a 21 ppm PCE exposure and its effect in 6 years and found no renal toxicity (52).

Biomarkers are essential in understanding the nephrotoxic effects of PCE. Proteinuria serves as a key biomarker for detecting PCE-induced nephrotoxicity. Some studies have shown elevated total protein (TPU), indicating glomerular injury from PCE toxicity in both animals and humans (47, 51). However, other research has found no significant or consistent association, highlighting inconsistent evidence on this effect (16).

According to a case report in South Korea, urinary calcium levels are elevated due to calcium crystals in the renal tubules caused by PCE exposure (54). Similar to this, low urinary sodium excretion often precedes the diagnosis of AKI, especially in critically ill patients (55). So, it is believed in theory that exposure to PCE may compromise kidney function, potentially resulting in decreased urinary sodium excretion. And also, electrolyte imbalance was hypothesized as a result of solvent exposure (53). BUN and S. creatinine, which are standard kidney function biomarkers (56), would show impaired urea excretion and reduced GFR, respectively, as a result of PCE exposure toxicity (47, 51, 55).

Creatinine level is used to evaluate overall kidney condition, which will help in detecting any impairment caused by toxic substances. Altered urinary calcium and sodium levels show a significant kidney dysfunction, which could lead to renal tubular injury, when the kidney tubules responsible for filtering waste are damaged (16, 21, 51, 52). By addressing these conditions and incorporating serum biomarkers, a more comprehensive understanding of PCE-induced nephrotoxicity could be gained.

2.6 Factors Influencing Perchloroethylene-Induced Nephrotoxicity

Renal diseases are multifactorial, which can be caused and aggravated by the interaction of many elements (58). For instance, sociodemographic variability plays a crucial role in understanding nephrotoxic effects by assuming and considering older individuals as more vulnerable due to their age-related compromised immunity. Similarly, due to their distinguished physiologic characteristics, males and females might show different susceptibility to PCE (1). Additionally, longer employment means longer exposure, which in turn affects their kidney (7). Furthermore, educational status and availability of nearby health care could also correlate with reduced or increased protective measures and medical checkups regarding PCE-induced nephrotoxicity.

Behavioral factors such as smoking and alcohol consumption also increase the overall risk of kidney damage. They could interact negatively with PCE or increase the susceptibility of one's kidney by affecting renal function. Moreover, Pre-existing medical conditions such as diabetes, hypertension, and currently used medications could influence renal toxicity (21).

Occupational and organizational factors also play their role in identifying PCE-induced nephrotoxicity. PCE exposure varies depending on specific tasks, which will influence individual risk. Additionally, the size of the establishment, availability of PPE, ventilation system, safety training and chemical handling procedures, and awareness could mitigate the exposure, which will significantly alter the health outcome (30,32).

Nephrotoxicity is one form of renal disease mainly caused when the kidney encounters a toxic substance (59). However, there are a lot of diseases that could influence renal function. For instance, the kidneys' blood vessels could be strained by high blood pressure, as a result, nephrosclerosis will occur (60). Diabetes is also another disease that affects the kidneys' filtration system and leads to end-stage renal disease (61). Diabetes and hypertension could support each

other in causing chronic kidney disease (62). Last but not least, heart disease reduces renal perfusion due to increased vascular resistance (63). Kidney impairment could also result in cardiovascular disease as a result of fluid overload, which makes both a risk factor for each other (60).

Certain medications carry a high risk of nephrotoxicity, including Ibuprofen, Aspirin, and most Non-steroidal Anti-Inflammatory Drugs (NSAIDs) (64). Prolonged or high-dose use of antibiotics like aminoglycosides, antiviral drugs, and mood stabilizers such as lithium can also contribute to kidney damage (65, 66).

Obstruction in urinary flow, possibly caused by a kidney stone or an enlarged prostate, along with kidney infection, was found to affect urinary biomarkers for nephrotoxicity (62).

People who are usually above 60 years old and obese individuals are susceptible to kidney toxicity due to decreased renal reserve and high blood pressure (58). Exposure to heavy metals such as lead could also affect urinary biomarkers by damaging the kidneys' filtration system (62).

The aforementioned and many other factors could affect the kidneys' function, as a result, understanding their interplay and relationship with nephrotoxicity is an important aspect for capturing the holistic nature of the problem.

2.7 Regulatory Standards and Guidelines

In Italy, exposure assessment regarding PCE in dry cleaners is conducted almost every year, as stated by their legislation. They use TCE in urine as a biological marker, but the problem is that TCE doesn't have a good correlation with environmental exposure assessments (19). In the Netherlands, a 15-minute TWA PCE concentration was found to be greater than 35 ppm, but after implementing a certification program, they were able to reduce all of it below 35 ppm, even though certain health effects were still found (14).

A Literature review of occupational exposure to PCE found the average concentration to be 0.399 micrograms per cubic meter (9). It has been suggested that the biological equivalent value for urinary PCE be 56 µg/l (0.056 mg/l), which equates to a TLV TWA of 25 ppm (67).

According to OSHA regulations, the exposure limit for a full work shift in the US is 100 ppm and 200 ppm for ceiling points (68). However, the EU shows a more serious concern about PCE and has regulated the exposure standard much below that of OSHA, with 20 ppm for 8 hours and 40 ppm for 15-minute exposure (69).

Perchloroethylene Reference Dose (RFD) is estimated to be 0.006 mg/kg/d, based on adult occupational exposure neurotoxicity. The RFD is an estimate of a daily oral dose to the human population, with uncertainty possibly ranging over an order of magnitude. Since this number was calculated from LOAEL rather than NOAEL, the EPA has medium confidence in it (10).

The Toxics Use Reduction Institute (TURI) evaluated alternatives to perchloroethylene (PCE) based on various criteria, including technical, economic, environmental, regulatory, and human health factors. They ranked these alternatives into five groups, where Professional wet cleaning (water) is considered the best, and n-propyl bromide (n-PB) is a listed alternative (70).

In conclusion, although the lack of references in the context of Africa and Ethiopia poses a certain challenge, it clearly shows the need to conduct future studies since the title hasn't been given prior focus, yet the chemical has been acknowledged as having many health effects for both workers and the general population. Finally, even though Western researches have sociodemographic and environmental variability, it will play a crucial role in developing methodological steps to conduct health effect studies.

Conceptual Framework

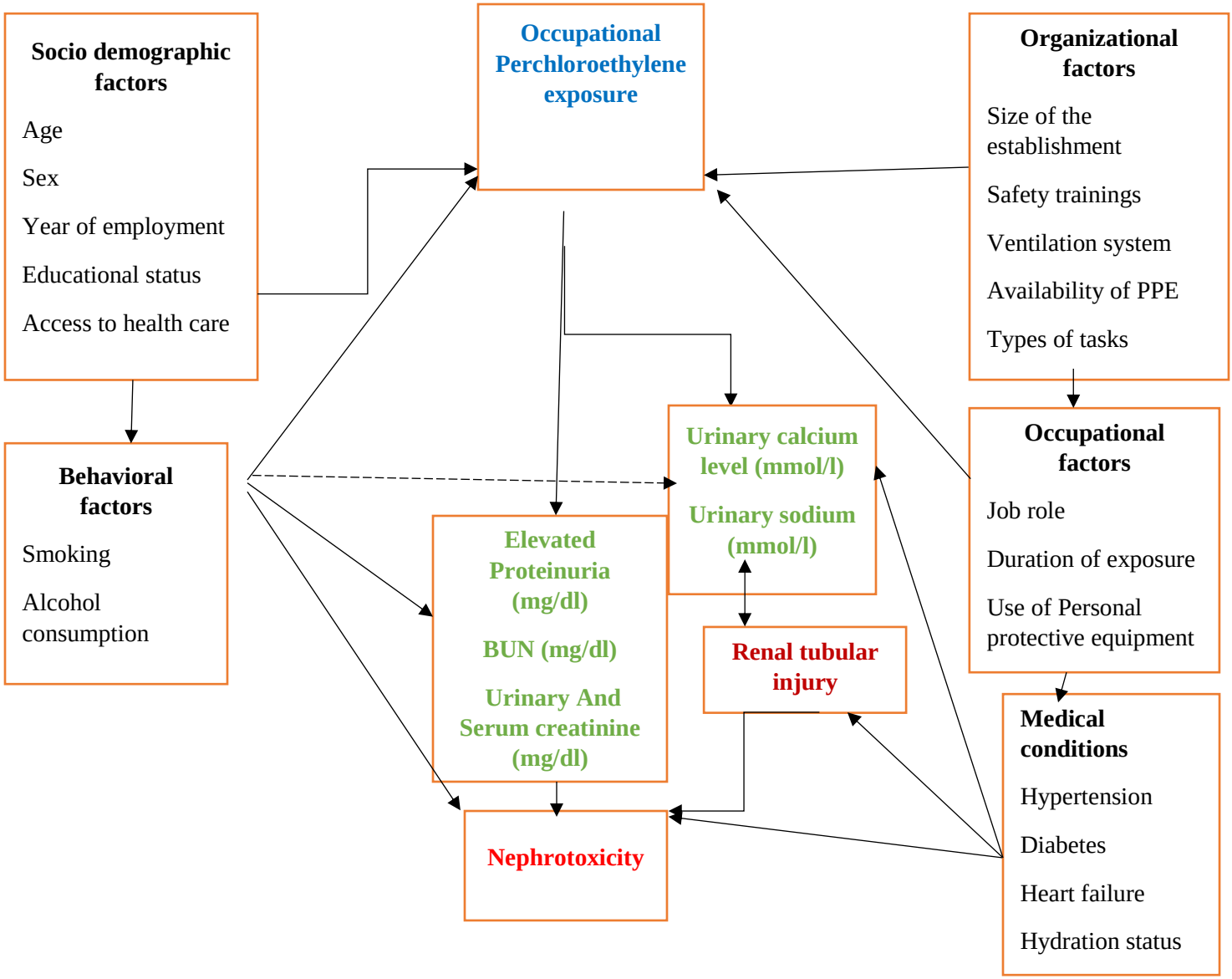

Figure 2 Conceptual frame work for Risk of Nephrotoxicity among Dry Cleaning Workers Exposed to Perchloroethylene in Addis Ababa, Ethiopia (adopted from (3,7,16,21,32,71) *)*

3. Objective

3.1 General objective

- ✓ To evaluate and compare the effect of occupational exposure to perchloroethylene on kidney health among selected dry-cleaning workers and hospitality laundry workers in Addis Ababa, Ethiopia, 2025

3.2 Specific objectives

- ✓ To measure and compare specific urinary biomarkers of nephrotoxicity, namely protein, creatinine, calcium, and sodium, between dry cleaning workers exposed to perchloroethylene and non-exposed hospitality laundry workers in Addis Ababa, 2025.

- ✓ To assess factors associated with nephrotoxicity urinary biomarkers among dry cleaning workers in Addis Ababa, 2025.

- ✓ To measure and compare specific serum biomarkers, namely Blood Urea Nitrogen and Serum creatinine, between dry cleaners and non-exposed hotel laundry workers in Addis Ababa, 2025

4. Method

4.1 Study Setting

This study was conducted in Addis Ababa, the capital and largest city of Ethiopia, with approximately 5.7 million people (72). There are currently 11 sub-cities in Addis Ababa, which serves as the political, cultural, and economic heart of Ethiopia, housing numerous international organizations, including the African Union and the United Nations Economic Commission for Africa.

According to "Addisbiz" an Ethiopian business directory and product catalog there are currently 62 registered laundry establishments, that also provide dry cleaning under their services in Addis Ababa and according to Addis Ababa ministry of culture and tourism, a total number of 547 hotels and 136 stared hotels are found in the hospitality industry, without including cafes, restaurants, lounges, resorts, and bars (73).

Addis Ababa was and still is experiencing rapid population growth and urbanization. This growth has led to an increase in service industries, making dry cleaning one of them. The diverse socio-economic background of the city's residents, including a temporary and permanent stay of foreign citizens, contributes to a high demand for dry cleaning shop establishments. Multiple dry-cleaning establishments and hotel facilities were chosen randomly from both groups.

4.2 Study Design

A multicenter comparative cross-sectional study was conducted from June 08, 2024, to April 07, 2025, for all the three specific objectives of the study, which allowed us to assess the urinary biomarkers, and serum biomarkers and associated factors by taking snapshot urine and blood samples of the study participants along with the questionnaires.

4.3 Population

4.3.1 Source Population

The source population included all dry-cleaning workers in Addis Ababa, which was taken from the number of registered dry-cleaning businesses and their employees in the "addisbiz" catalog,

and for the control group all laundry workers who didn't use PCE in the hospitality industry were selected from the registry we got from ministry of culture and tourism.

4.3.2 Study Population

The study population consists of dry-cleaning workers from randomly selected establishments, and participants were recruited from a variety of job roles within the dry-cleaning process, including machine operators, ironers, receptionists, and managers. A control group was taken from randomly selected hotel laundry workers who use non-PCE solvents.

4.4 Inclusion and Exclusion Criteria

4.4.1 Inclusion Criteria

Workers aged 18 years and older currently employed in the dry-cleaning industry for at least six months and above and who were able to provide informed consent were eligible for the exposed group.

Workers in the hospitality laundry service who have no known PCE exposure in their current occupational setting, fulfilling the same inclusion criteria listed above as the exposed group, were eligible for the control group.

4.4.2 Exclusion Criteria

Workers who had renal disease before joining any dry-cleaning establishments, workers who were using temporary medication for other non-kidney related diseases, pregnant or lactating women, and also women who were in their menstrual cycle during the data collection period were excluded due to the potential physiologic changes of metabolites.

4.5 Sample Size Determination for the First and the Second Objectives

The sample size was calculated using the formula for comparative cross-sectional studies. However, because we could not identify definitive values for the effect size, mean, or standard deviation for both groups and the available data only pertained to workers' exposure status, which would have greatly limited our sample size for assessing nephrotoxicity, we opted to use Cohen's

d. We assumed a medium effect size of 0.5, which is commonly considered appropriate for detecting meaningful differences in public health studies with a 1:1 allocation ratio. (74).

$$n = \frac{(Z\alpha/_2 + Z\beta)^2 * (\sigma 1^2 + \sigma 2^2)}{(M1 - M2)^2}$$

Cohen's d is calculated as:

$$D = \frac{M1 - M2}{SD}$$

n: Sample size required for the study.

Zα/2: Z-score corresponding to the desired level of statistical significance (α).

Zβ: Z-score corresponding to the desired power of the test (1 - β).

σ_1^2: Variance of the first group.

σ_2^2: Variance of the second group.

M_1: Mean of the first group.

M_2: Mean of the second group.

D: Cohen's d, a measure of effect size.

SD: Standard deviation of the difference between groups.

Assuming equal variances for both groups, we simplified the formula by assuming a common standard deviation (SD), which we can denote as S*D*.

$$n = \frac{(Z\alpha/2 + Z\beta)\ 2 * (2SD^2)}{(0.5 * SD)^2}$$

$$n = \frac{(1.96+0.84)^2 * (2 \cdot SD^2)}{(0.5 * SD)^2}$$

n= 63

Anticipating a dropout rate of 10%, the sample size was;

New Sample Size = n / (1−Dropout Rate)

= 63 / 1-0.1

n = 70 for each group

A total of 140 participants were included.

4.5.1 Sample size calculation for the third objective

Taking the mean and standard deviation values for serum creatinine from a health impact assessment conducted in Iran (57). Our sample size was calculated as follows

$$n = \frac{(Z\alpha/2 + Z\beta)\ 2 * (2SD^2)}{(M1 - M2)^2}$$

$$n = \frac{(1.96+0.84)^2 * (0.2^2 + 0.04^2)}{(1.1-0.92)^2}$$

N = 10 + (10% drop out) = 12

- **Assuming a one-to-one allocation ratio, we had a total of 24 participants: 12 dry cleaners and 12 hotel laundry workers.**

4.6 Sampling Procedure

A simple random sampling method was used to ensure representative selection across various dry-cleaning establishments and hotels operating in Addis Ababa. The sampling frame was first constructed from "addisbiz" business directory catalog, as we could not get detailed distribution by sub-city for the dry-cleaning establishments. We noticed an average of 4-5 workers per establishment during the preliminary site observation. As a result, we applied a lottery method to select 17 dry-cleaning establishments from the 62 registered shops and reached 70 dry cleaners as study participants.

The control group sampling procedure was also executed by using the lottery method, since we observed an average of 3-4 workers in each hotel laundry during the preliminary site observation, we randomly selected 21 hotels from the registered 547 hotels to achieve 70 non-exposed individuals.

Finally, for the third objective, four dry-cleaning shops were randomly selected from the previously unselected shops. Similarly, 5 hotels were also selected from the previously unselected 526 hotels and achieved 12 participants in each group.

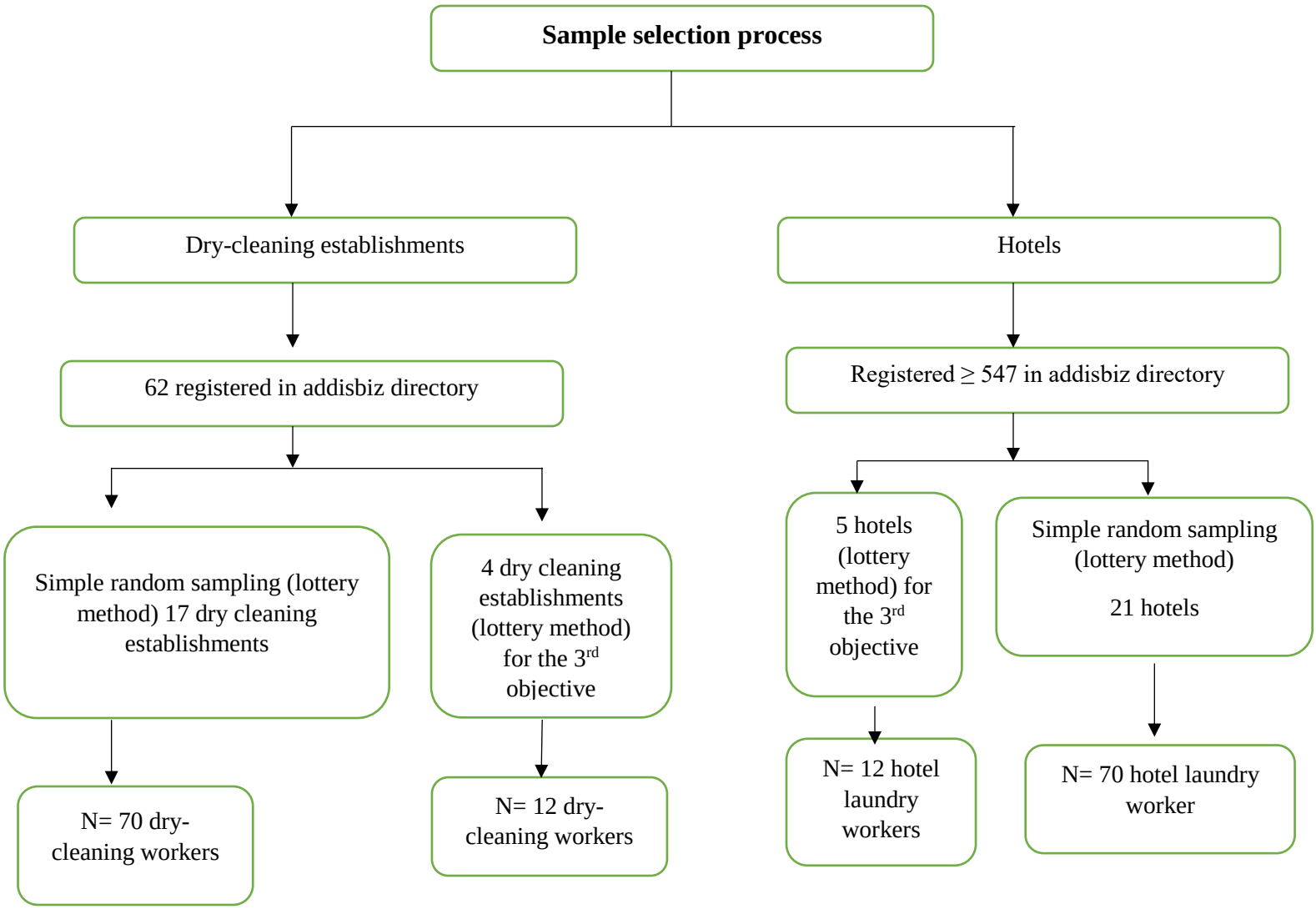

Figure 3 Schematic representation of sampling procedure

4.7 Data Collection Procedure

Data were collected through a combination of standard questionnaires and biological sampling.

1. **Questionnaires:** Participants completed structured questionnaires consisting of both open- and closed-ended questions. Open-ended questions explored individual experiences such as PPE usage, diet, and chronic illness, while close-ended questions gathered quantifiable data on factors like ventilation availability, organizational training, PPE provision, job roles, and task frequency. Trained data collectors administered the questionnaires and assisted as needed.

2. **Biological Sampling:** participants were instructed by a trained data collector on how to collect clean catch mid-stream urine samples to minimize contamination as explained by EPHI urine collection standard operation procedure, then urine samples were collected in sterile cups labeled with an identification code afterward to measure biomarkers of nephrotoxicity, including proteinuria, creatinine, urinary calcium, and sodium. Finally, trained health professional data collectors took blood samples from 24 study participants in order to measure BUN and Serum creatinine.

4.7.1 Measurements

4.7.1.1 Independent Variables

- Demographic factors of the workers
- Job role within the dry-cleaning establishment
- Size of the working shop
- Duration of employment
- Availability of a good ventilation system
- Use of personal protective equipment
- Chronic diseases like diabetes, hypertension, and heart disease

4.7.1.2 Dependent Variables

- Urinary Protein (mg/dl)
- Urinary Creatinine (mg/dl)
- Urinary Calcium (mmol/l)
- Urinary Sodium (mmol/l)
- Blood Urea Nitrogen (mg/dl)
- Serum Creatinine (mg/dl)

4.8 Data Collection

Four data collectors and two supervisors, each holding a bachelor's degree in medical laboratory science or nursing and possessing prior experience in biological sample collection, were hired to conduct urine and blood sample collection, as well as structured interviews with the study participants.

Data collection took place from March 8 to April 7, 2025. Blood samples were collected on Wednesdays and Thursdays at the end of the shift, based on previous studies that suggest these samples are representative of PCE blood exposure over the working days. In contrast, urine samples were collected on all working days.

The data collector explained the procedure to the participants and instructed them to collect midstream clean-catch urine samples based on their sex. Where for females the procedure started by cleaning the urethral area with a cleansing wipe and then holding the labia apart while collecting urine, and for males it started by wiping the tip of the penis first, and then catching the urine midstream in the cup, while urinating in the toilet. A blood sample was also drawn from 24 study participants. After this, each sample cup was covered. And finally, the samples were labeled and transported after preserving them in a cold box at 2-8°C, then finally the samples were transferred to Nunc tube to be stored in EPHI laboratory refrigerator for later analysis, using automated urine biochemistry analyzers for all biomarkers.

4.9 Operational Definitions

- Perchloroethylene Exposure: Defined as the concentration of PCE in biological samples, indicative of occupational exposure levels. Having a biological equivalent value of 56 µg/l (0.056 mg/l), which equates to a TLV TWA of 25 ppm (67). However, we were not able to quantify PCE exposure as a result, those who do work in the dry-cleaning shops were categorized as exposed.
- Nephrotoxicity refers to kidney damage or dysfunction, assessed through urinary and blood biomarkers. A significant increase above the standards in any of the urinary biomarkers could indicate an early sign of nephrotoxicity.
- Proteinuria: Increased total protein levels in urine indicate a glomerular injury and the kidney's reduced capacity to filter proteins. It's regarded as one of the initial indicators of nephrotoxicity. With a normal value of <150 mg/dl (75).
- Urinary creatinine: a normal range for women is approximately 29 – 226 mg/dl, and for men is approximately 40 – 275 mg/dl. Deviation from this range shows abnormality (76).
- A urinary calcium normal range from 2.5-7.5 mmol/l for individuals on a normal diet is considered healthy (77).
- Urinary sodium between 27 – 287 mmol/L/ for females and 40-220 mmol/l for men shows a healthy range for both sexes (77).
- Blood urea nitrogen: an increase in BUN level suggests the kidneys' ability to filter urea was damaged as a result of PCE exposure, with a normal range of 7-20 mg/dl (53).
- Serum creatinine: normal range for men is 0.6-1.2 mg/dl and for women, 0.5-1.1; elevated levels suggest reduced GFR function or tubular dysfunction, suggesting nephrotoxicity (53).

4.10 Data Management

The principal investigator extracted the obtained data and performed data cleaning to check for mistakes and inconsistencies. The supervisors addressed any confusion by accompanying the data collectors during the actual data collection process. Any issues beyond their capacity were addressed by the principal investigator.

4.11 Data Analysis

Statistical analysis was performed using IBM SPSS Statistics version 25 software. The analysis encompassed two main components: descriptive statistics and inferential statistics, after checking for normality.

The laboratory analysis performed in EPHI which is ISO 15189:2012 accredited for its National Clinical Chemistry Reference Laboratory has quantified the four selected kidney function urinary biomarkers results for all 140 participants, and the two serum biomarkers for all 24 samples. Normality check for the raw data was performed by combining the Shapiro-Wilk Statistic, Kolmogorov-Smirnov Statistic, kurtosis, skewness, and histogram. Even though the first two tests are sensitive to small deviations when the sample size is above 50, we included them for better confirmation. All four urinary biomarkers showed non-normal distribution, with $p<0.05$ for both Shapiro and Kolmogorov tests, where total protein showed extreme abnormality while positively skewed. Log10 transformation was applied to all biomarkers, where protein significantly improved to be normal, urinary calcium and creatinine also fall under the normality range of kurtosis and skewness where parametric test is valid with caution, the only abnormal result after transformation was seen in urinary sodium, square root transformation also yielded non-normal distribution, so non-parametric test was used for this specific biomarker.

Descriptive statistics provided a comprehensive summary of the demographic, health, and exposure-related data collected from the study participants. Box plot visualization was used to show the distribution of the four urinary biomarkers across work workplace. In the same fashion bar graph was also used to show the percentage of abnormal urinary biomarkers distribution.

Following the descriptive analysis, inferential statistics were done, where an independent t-test and Mann-Whitney test for normally distributed and non-normally distributed data, respectively, were used to see if there was a significant difference in the means of the biomarkers. Following this, categorical variables comparison was done using the Chi-square test to check if there was a significant association between PCE exposure-related variables and health symptom-related variables. Point-Biserial and Spearman correlation analysis was also done to see which risk factors significantly influence each specific biomarker. Then, finally, multiple linear regression was performed to see the effect of different predictors on each dependent variable.

4.12 Data Quality Assurance

In order to assure the quality of the data, appropriate training for the data collectors was given so that they could get familiar with the data collection tools. To validate the collection tool and to see the fieldwork challenges, pre-testing was done on 5% of the sample size for the questionnaires only in another establishments that was not included in the study for both group pf workers. During the laboratory analysis, standard operating procedures and guidelines were followed. Finally, before data analysis, the principal investigator conducted data cross-checking and cleaning.

4.13 Ethical Consideration

The major ethical concern that was raised in this research was getting informed consent to collect biological samples, which was essential before starting the data collection. Individuals were fully informed about the goals, risks, and advantages of the study. Confidentiality was kept in order to safeguard participants' private health and personal data and avoid any possibility of prejudice. We informed the participants that the study would not exacerbate any pre-existing health risks and assured them that they would be promptly notified of any unfavorable results. Participants were informed of their health status and the potential PCE risk to their kidneys. They were informed when the result showed positive nephrotoxic risk. The result was used to enhance both the workers' and their employer’s safety awareness, where safety and chemical handling education and training were advocated in order to promote the employees’ health, and in the wider aspect Ministry of Labor would consider setting occupational health policies and guidelines for dry cleaners. Finally, the research began after getting Ethical approval from the Addis Ababa University School of Public Health institutional review board and the Addis Ababa Health Bureau.

4.14 Dissemination of the Result

The findings of this research were submitted to Addis Ababa University, College of Health Sciences, School of Public Health, as partial fulfillment of a Master of Public Health degree, and also shared with the Addis Ababa Health Bureau. The results will be communicated to researchers in the field, presented at conferences when possible, and ultimately published in a scientific journal.

5. Result

5.1 Demographic characteristics of the study participants

The following result section shows the basic demographic characteristics of both the exposed and control group workers from the selected 17 and 21 dry cleaning establishments and hotels, respectively. Participants' sex, job role, working days, hours, and years of employment are shown in the table along with their behavioral characteristics of smoking and drinking. The majority of the workers in both laundries were females, covering 61.4% of study participants in dry cleaning and 65.7% in hotel laundry. 40 % of the participants in the dry cleaning have only been in the job for less than 3 years, whereas 40% of the workers in hotel laundry have been on the job between 4 to 6 years. According to the result of our study, although rotation is very common, job roles within the laundry service in Ethiopia is classified into four major categories, namely, machine operator, ironer, receptionist, and manager, based on the workers' first line of engagement.

Table 1Demographic characteristics of the study participants (N=140)

Category	Description	Dry Cleaning Laundry	Hospitality Laundry	Significance (p)
Age	Mean ± SD	30.66 ± 6.255	27.93 ± 4.261	<0.016
Sex	Female	43 (61.4%)	46 (65.7%)	<0.598
	Male	27 (38.6%)	24 (34.3%)	
Education level	Bachelor's Degree	9 (12.9%)	14 (20.0%)	<0.428
	Diploma/Certificate	22 (31.4%)	18 (25.7%)	
	Elementary	9 (12.9%)	5 (7.1%)	
	High School	30 (42.9%)	33 (47.1%)	
Years of employment	Less than 1 year	9 (12.9%)	12 (17.1%)	<0.091
	1 to 3 years	28 (40.0%)	16 (22.9%)	
	4 to 6 years	21 (30.0%)	28 (40.0%)	
	7 to 10 years	10 (14.3%)	7 (10.0%)	
	11 to 15 years	2 (2.9%)	7 (10.0%)	
Job Role	Ironer	21 (30.0%)	14 (20.0%)	<0.546
	Machine Operator	25 (35.7%)	27 (38.6%)	

	Manager/Supervisor	11 (15.7%)	15 (21.4%)	
	Receptionist	13 (18.6%)	14 (20.0%)	
Working Days per Week	<3 days	1 (1.4%)	4 (5.7%)	<0.002
	4 days	3 (4.3%)	5 (7.1%)	
	5 days	9 (12.9%)	26 (37.1%)	
	6 days	55 (78.6%)	35 (50.0%)	
	7 days	2 (2.9%)	0 (0%)	
Work Hours per Day	4-6 hours	5 (7.1%)	11 (15.7%)	<0.298
	7-8 hours	53 (75.7%)	48 (68.6%)	
	>8 hours	12 (17.1%)	10 (14.3%)	
Smoking Status	Current Smoker	1 (1.4%)	4 (5.7%)	<0.392
	Former Smoker	4 (5.7%)	4 (5.7%)	
	Never Smoked	65 (92.9%)	62 (88.6%)	
Alcohol Consumption	No	19 (27.1%)	25 (35.7%)	<0.298
	Occasionally	45 (64.3%)	36 (51.4%)	
	Yes, frequently	6 (8.6%)	9 (12.9%)	

5.2 Descriptive statistics of the four urinary biomarkers

The table presents a comparison of key urinary biomarkers between dry cleaning workers (exposed group) and hospitality laundry workers (control group). The data are expressed as median values with interquartile ranges (IQR) to illustrate the central tendency and variability within each group. This comparison highlights differences in total protein, creatinine, calcium, and sodium levels, providing insight into potential occupational exposure effects on renal function.

Table 2 Descriptive summary of the four urinary biomarkers between exposed and control groups (N=140)

Variables	**Exposed Group (Dry Cleaning workers)**	**Control Group (Hospitality laundry workers)**
	Median & IQR	Median & IQR
Total Protein (mg/dl)	102.00 & 70.75	54.50 & 27.25
Urinary Creatinine (mg/dl)	193.78 & 111.06	142.93 & 78.17
Urinary Calcium (mmol/l)	2.60 & 2.94	0.835 & 0.79
Urinary Sodium (mmol/l)	167.00 & 67.00	158.00 & 73.75

Table 3 give additional information by comparing biomarker levels between dry cleaning workers and hospitality workers, presenting means and standard deviations (SD) for log-transformed values. Variables include Log10 Total Protein, Log10 Creatinine, Log10 Calcium, and Log10 Sodium. The exposed group shows higher mean values for all biomarkers compared to the control group, with varying SDs indicating differences in data dispersion.

Table 3 Descriptive summary of Log10 transformed urinary biomarkers comparing means of the exposed and control groups (N=140)

Variables	**Exposed Group (Dry Cleaning) Mean ± SD**	**Control Group (Hospitality) Mean ± SD**
Log10 Total Protein	1.9878 ± 0.27536	1.7604 ± 0.19209
Log10 Creatinine	2.2169 ± 0.25417	2.0819 ± 0.24159
Log10 Calcium	0.3738 ± 0.35870	-0.1139 ± 0.40680
Log10 Sodium	2.1586 ± 0.19896	2.1208 ± 0.22444

Table 4 compares blood urea nitrogen (BUN) and serum creatinine levels between an exposed group and a control group, presenting means and standard deviations (SD). Variables include BUN (mg/dl) and S. Creatinine (mg/dl). The exposed group exhibits higher mean values for both biomarkers compared to the control group, with larger SDs indicating greater variability in the data. From the total of 12 samples in each group the prevalence of abnormal BUN was 33.3% (4 workers) and 16.7% (2 workers) in dry cleaners and hotel laundry workers respectively, while 3 workers in each group showed abnormal serum creatinine level with a prevalence of 25%.

Table 4Descriptive summary of serum biomarkers comparing means of exposed and control groups (N=24)

Variables	**Exposed Group (Dry Cleaning) Mean ± SD**	**Control Group (Hospitality) Mean ± SD**
BUN (mg/dl)	15.5 ± 6.08	12.5 ± 4.15
S. Creatinine (mg/dl)	0.8658 ± 0.36694	0.8262 ± 0.28804

5.3 Visual Boxplot representation of Urinary biomarkers distribution

The median of total protein distribution in the box plot showed a higher value in dry cleaners compared to hotel laundry workers, with an approximate median of 102mg/dl and 70mg/dl, respectively. The majority of the distribution in both groups was found between the median and Q3. The exposed group showed around 3 outliers above the maximum value, whereas 6 outliers were observed in the control group above the maximum value. The upper whisker goes beyond 200, and the lower whisker falls below 20 approximately for the exposed group; the control group has an upper whisker slightly above 100 and close lower whisker value with the exposed group. Overall all the control group is the least dispersed one

Figure 4Simple Boxplot representation showing distribution of total protein among exposed and control groups (N=140)

The urinary creatinine and workplace boxplot show a higher median value around 190 mg/dl for the exposed group compared to a 140 mg/dl median value for the control group. The dry-cleaning plot shows that the majority of the distribution falls below the median line however, the difference is not that much large with the upper dispersion. In case of the hospitality laundry workers majority of the values are clustered below the median line. The lower whisker for both groups somehow falls in the same value approximately, showing both groups have similar minimum creatinine values. The maximum value for the exposed is around 350 mg/dl, and above 150 for the control group. The number of outliers noticed in dry cleaners is one, four outliers showed a higher creatinine value in hotel laundry workers.

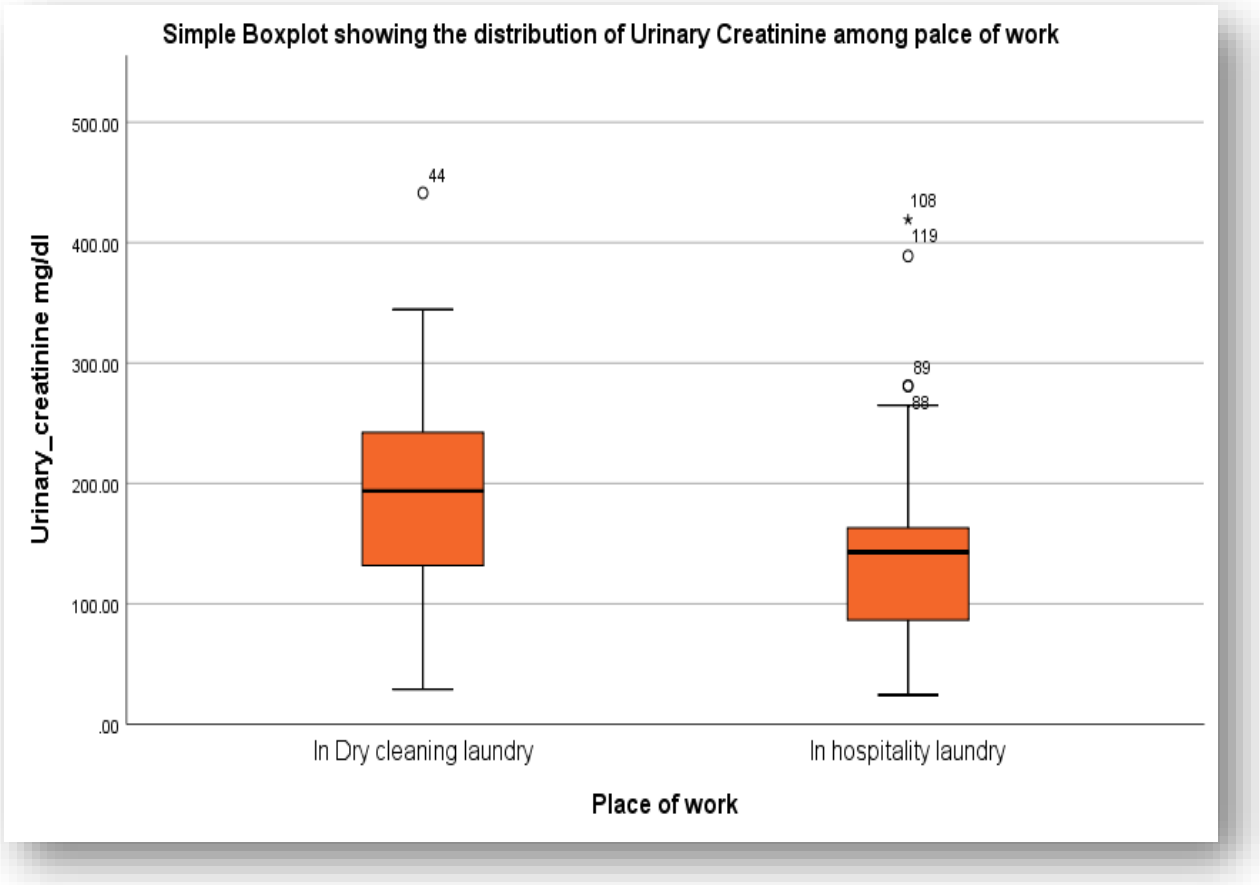

Figure 5 Simple Boxplot representation showing distribution of urinary creatinine among exposed and control groups (N=140)

The visual representation shows a higher urinary calcium value in dry cleaners compared to hotel laundry workers, with 2.6 mmol/l and 0.84 mmol/l, respectively. The exposed groups are more dispersed and have a long inter-quartile range with only one outlier above the upper whisker. The control group is clustered around a small range with many outliers. The lower whisker in the control group touches the x-axis, which is the result of less than the test value capacity in the EPHI machine monitor, as reported by the analysis expert.

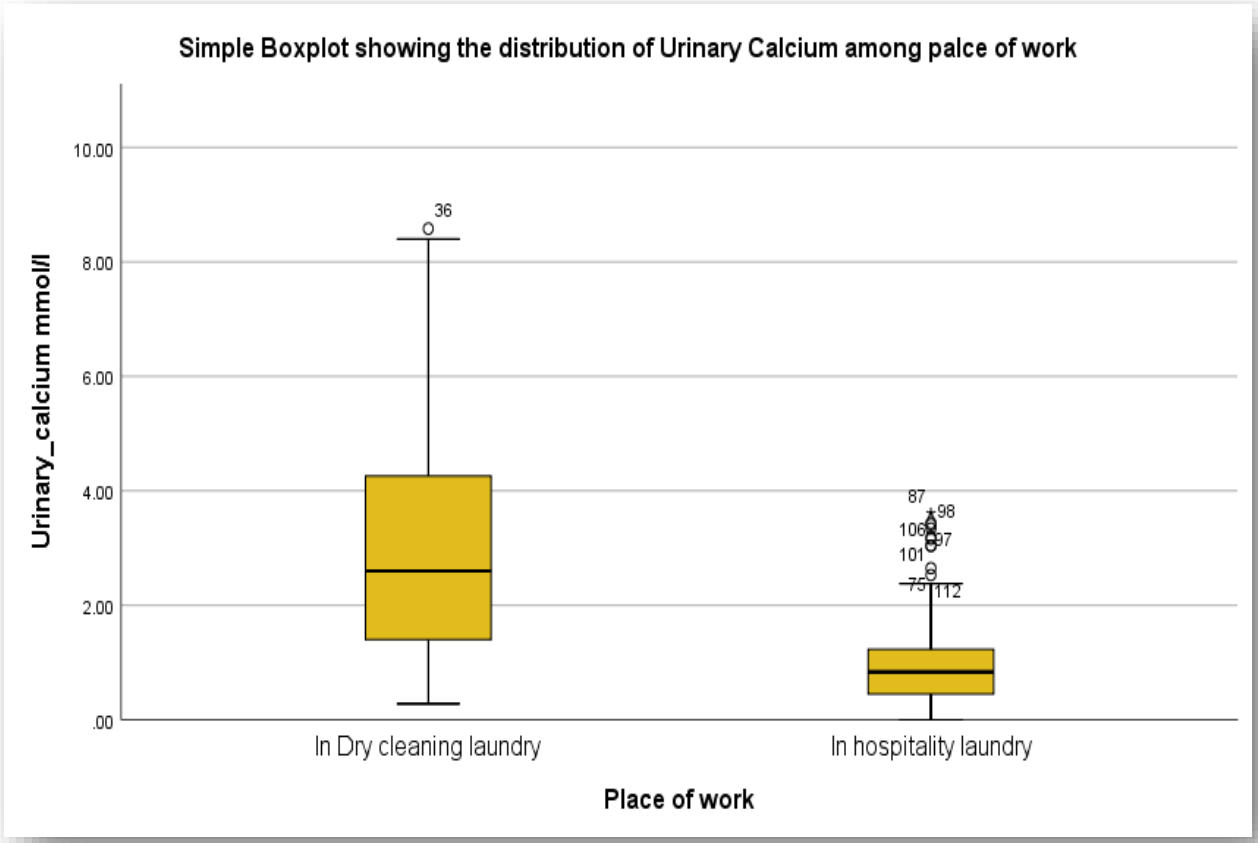

Figure 6 Simple Boxplot representation showing distribution of urinary calcium among exposed and control groups (N=140)

The visual boxplot representation of urinary sodium in both groups shows a similar overall dispersion and a mean value of 167 mmol/l and 158 mmol/l for the exposed and control groups, respectively. Both groups show a majority cluster below the median value. Both groups have a close upper whisker value; the lower whisker value of the hotel laundry workers seems to be lower than the dry cleaners, although lower outliers are found in the dry cleaners.

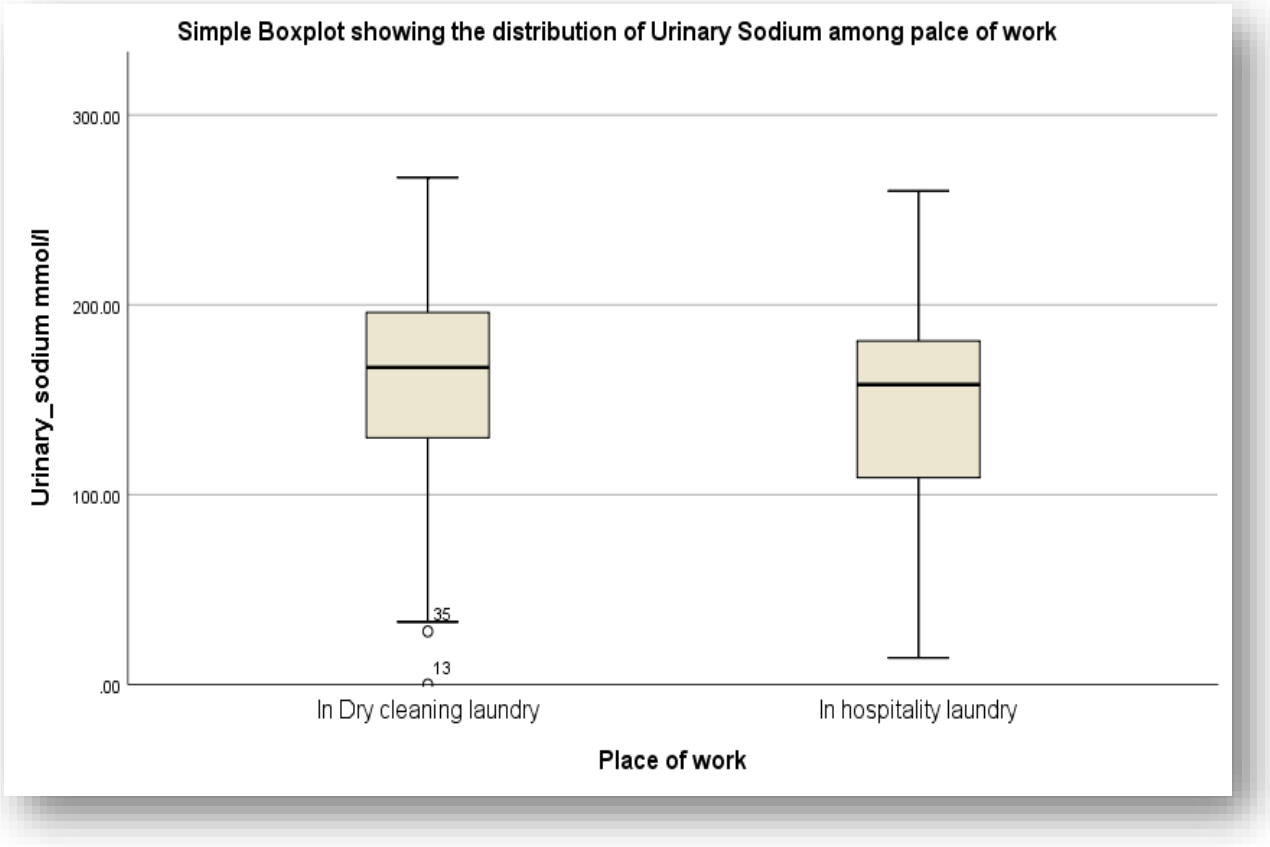

Figure 7 Simple Boxplot representation showing distribution of urinary sodium among exposed and control groups (N=140)

Table 5 compares health symptoms and training experiences between dry cleaning laundry workers and hotel laundry workers. The dry cleaning group reports higher percentages of decreased urine output, foamy/discolored urine, and dizziness/headache, while the hotel laundry group shows a higher prevalence of no changes in urination and no training on PCE handling. Fatigue/weakness and nausea/loss of appetite frequencies are relatively similar between groups, with slight variations in specific symptom prevalence.

Table 5Disease symptoms and changes noticed in both dry cleaners and hotel laundry workers (N=140)

Category	Description	Dry cleaning laundry	Hotel laundry
Urination patterns	Decreased urine output	22 (31.4%)	8 (11.4%)
	Foamy/discolored urine	16 (22.9%)	12 (17.1%)
	Increased frequency	5 (7.1%)	3 (4.3%)
	No changes	27 (38.6%)	47 (67.1%)
Fatigue/weakness	Always	13 (18.6%)	5 (7.1%)
	Never	7 (10.0%)	2 (2.9%)
	Often	31 (44.3%)	31 (44.3%)
	Sometimes	19 (27.1%)	32 (45.7%)
Nausea/loss of Appetite	Neither	29 (41.4%)	33 (47.1%)
	Both symptoms	13 (18.6%)	8 (11.4%)
	Loss of appetite only	14 (20.0%)	18 (25.7%)
	Nausea only	14 (20.0%)	11 (15.7%)
Training on PCE handling	No training	33 (47.1%)	53 (75.7%)
	Trained once	35 (50.0%)	15 (21.4%)
	Regularly trained	2 (2.9%)	2 (2.9%)
Dizziness and headache	No	12 (17.1%)	24 (34.3%)
	Yes	58 (82.9%)	46 (65.7%)

Our data revealed that 39 (55.7%) employees in the dry-cleaning laundry perform their job in a work setting that utilizes natural ventilation only, while 9 participants (12.9%) work in an establishment that uses forced ventilation only. Additionally, 22 participants (31.4%) work in establishments that use a combination of both ventilation methods. In the hospitality laundry sector, the findings showed that 53 participants (75.7%) work in an establishment that rely on natural ventilation only, 16 participants (22.9%) work in hotel laundries that use a combination of forced and natural ventilation, and finally only 1.4% of workers reported working in a laundry that have no ventilation system at all.

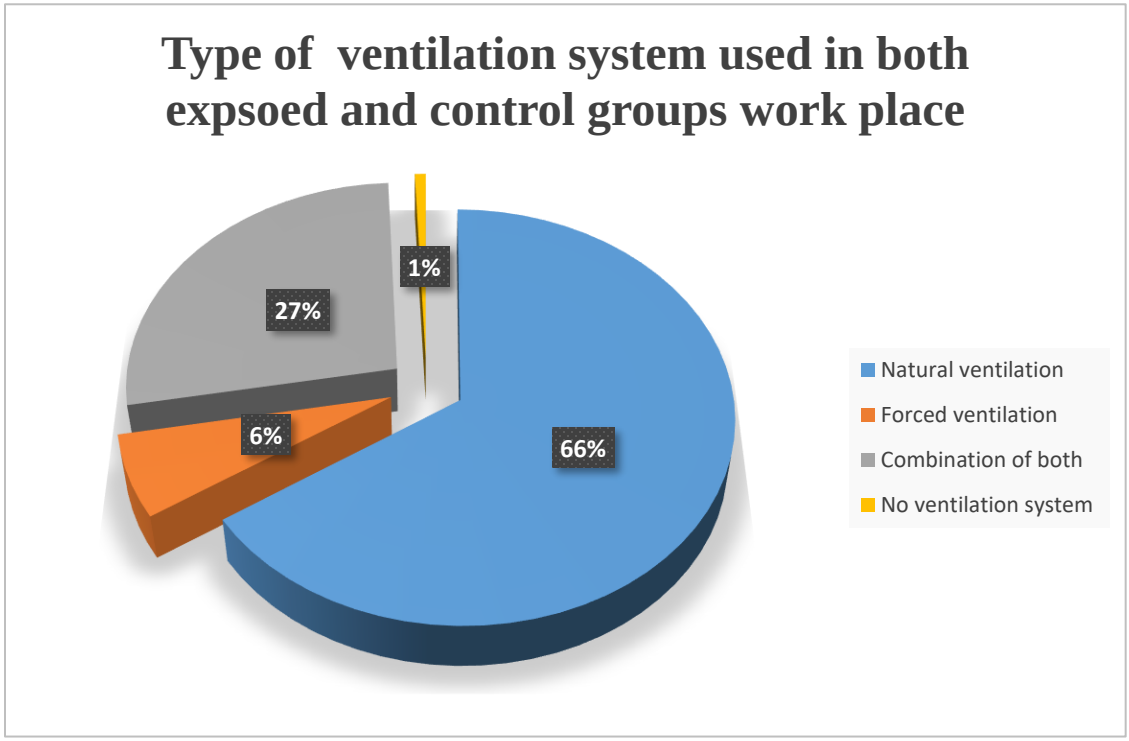

Figure 8 Pie chart visualizing the type of ventilation system used in the selected 17 dry cleaning establishments and 21 hotel laundries in Addis Ababa, 2025 (N=140)

5.4 Inferential statistics

5.4.1 Independent sample t-test for urinary biomarkers

An independent t-test was performed to see if there was a significant, meaningful difference in urinary and serum biomarkers distribution between the exposed and the control groups. The result showed that Log_{10} transformed total protein, urinary creatinine and calcium had a significant association with $p<0.05$ for all three urinary biomarkers, However, BUN and S. creatinine didn't show a significant association. As a result meaningful difference was not detected between the two means.

Table 6 Independent t-Test result of the log-transformed urinary biomarkers and serum biomarkers between exposed and control groups

Urinary Biomarker	Dry Cleaning Workers (Mean ± SD)	Hospitality Laundry Workers (Mean ± SD)	T	df	P
Log10Total Protein(TPU)	1.988 ± 0.275	1.760 ± 0.192	5.67	123.3	P<0.001**
Log10 Creatinine	2.217 ± 0.254	2.082 ± 0.242	3.22	137.6	P<0.002**
Log10 Calcium	0.374 ± 0.359	-0.114 ± 0.407	7.49	134.4	P<0.001**
BUN	15.5 ± 6.08	12.5 ± 4.15	-1.412	22	P<0.172
S. creatinine	0.8658 ± 0.36694	0.8262 ± 0.28804	-0.295	22	P<0.771

(** = shows significant association)

5.4.2 Mann-Whitney U result for urinary biomarkers

Non- parametric Mann-Whitney U test was conducted to see if there was a significant meaningful urinary sodium distribution difference between the exposed and the control groups, and the result showed a non-significant distribution with a p value of 0.176, which makes the null hypothesis true.

An additional non-parametric test was conducted on the non-log-transformed, non-normally distributed variables (TPU, urinary creatinine, and calcium) to ensure caution with the log-transformed independent t-test. The results revealed a similar significant difference between all three biomarkers, consistent with the t-test, with a p-value less than 0.05.

Table 7 Mann-Whitney U test result of the log10 transformed urinary biomarkers and serum biomarkers between exposed and control groups

Biomarker	Mann-Whitney U	Wilcoxon W	Z-score	p-value	Significant Difference
Total Protein	1151.000	3636.000	-5.415	<0.001**	Yes
Urinary Creatinine	1529.000	4014.000	-3.838	<0.001**	Yes
Urinary Calcium	901.500	3386.500	-6.454	<0.001**	Yes
Urinary Sodium	2125.000	4610.000	-1.355	<0.176	No

(** = shows significant association)

5.5 Perchloroethylene exposure and health symptom-related categorical variables comparison using Fisher's Exact Test

Fisher's Exact Test was performed to assess associations between exposure-related and symptom-related variables collected through questionnaires. The results showed several significant links between exposure related and safety practices with symptoms such as leg swelling, urine changes, and itching. Due to many expected values being less than five, Fisher's exact test was used instead of chi-square to strengthen the analysis and support the conclusions. An analysis between PPP usage and the immediate onset of symptoms while at work showed a significant association, with a chi-square of $x^2(4, N=140) = 12.631$. p=0.013 and a Fisher's test p-value of 0.004

A similar test was conducted for frequent handling of PCE and immediate health symptoms while at work, the result showed a significant association with the value of $x^2(4, N=140) = 41.092$. p<0.001 and a Fisher's test p-value of <0.001

Table 8 Fisher's exact test result between exposure and health-related symptoms (N=140)

Variable	Pearson value	Fisher's Exact Test p-value	% cells with low expected counts
PPE vs. Immediate Symptoms	Pearson: 12.631, df = 4	0.004**	33.3%
Frequency of Handling PCE vs Immediate Symptoms	Pearson: 41.092, df = 6	<0.001**	50%
Duration of PCE Exposure vs Immediate Symptoms	Pearson: 35.155, df = 6	<0.001**	33.3%
Duration of PCE Exposure vs Urination Changes	Pearson: 15.529, df = 9	<0.055	37.5%
Frequency of Handling PCE vs Urination Changes	Pearson: 19.359, df = 9	<0.024**	50%
Ventilation Adequacy vs Swelling	Pearson: 23.606, df = 6	<0.004**	41.7%
Type of Protective Equipment vs Nausea/Loss of Appetite	Pearson: 25.063, df = 18	<0.022**	67.9%
Type of Protective Equipment vs Persistent Itching	Pearson: 25.502, df = 18	<0.047**	75%
Training on Safe Handling of PCE vs Immediate Symptoms	Pearson: 24.868, df = 4	<0.001**	44.4%

(** = shows significant association)

5.6 Assessing the relationship between risk factors and urinary biomarkers using Correlation analysis

A correlation analysis was conducted to see which risk factors significantly influence each specific biomarker. Point-Biserial correlation was conducted between risk factors and log-transformed urinary biomarkers of kidney function, and the result showed the strength and direction of the significantly associated results.

Log_{10} transformed TPU and raw BUN showed a significant association with experience of PCE spillage, r (140) = 0.201, p = 0.017 and r (24) = 0.536, p = 0.007, respectively, suggesting workers who had experience of PCE spillage tend to have more proteinuria and BUN. Similarly, being a dry-cleaning worker tends to result in more protein in urine than being a hotel laundry worker, r (140) = 0.434, p< 0.001. However, the workplace was not significantly correlated with BUN and S. Creatinine.

Log_{10} transformed urinary creatinine showed a significant positive association with being a dry cleaner, r (140) = 0.264, p = 0.002. However, having a family member who had been affected by kidney disease and being an individual who had past kidney disease negatively correlates with urinary creatinine. With values, r (140) = -0.178, p = 0.035 and r (140) = -0.179, p = 0.034, respectively. This suggests that the kidney could potentially be compromised due to a reduced glomerular filtration rate (GFR), which would lead to a decrease in urine creatinine release.

Log_{10} transformed calcium showed a positive correlation with being a dry cleaner, experience of spillage, previous job experience involving PCE, and age. But similar to urinary creatinine, it showed a negative correlation with having a family member with kidney disease. r (140) = -0.166, p= 0.05

Table 9 Point-Biserial correlation result between log10 transformed urinary biomarkers and risk factors

Variable Pearson	**Log10 Total Protein (r, p)**	**Log10 Creatinine (r, p)**	**Log10 Calcium (r, p)**	**Log10 Sodium (r, p)**
PCE spillage	0.201, <**0.017**		0.220, <**0.009**	NS
Workplace	0.434, <**0.001**	0.264, <**0.002**	0.539, <**0.001**	NS
Family kidney history	NS	-0.178, <**0.035**	-0.166, <**0.050**	NS
Individual kidney history	NS	-0.179, <**0.034**		NS
Age in years	NS	NS	0.224, <**0.008**	NS
Previous job PCE exposure	NS	NS	0.267,<**0.001**	NS

5.6.1 Assessing the relationship between Risk factors with Ordinal response and Urinary Biomarkers using Spearman correlation analysis

Spearman correlation analysis was conducted for urinary sodium and for the three urinary biomarkers to see if there is an association with variables that have an ordinal nature.

Number of days worked per week is positively associated with TPU and urinary creatinine with a value of p (140) = 0.378, p< 0.001and p (140) = 0.354, p< 0.001, respectively, which tells us that as the number of working days increases, TPU and urinary creatinine increase.

Only TPU showed a significant association with total working hours (p(140) = 0.183, p = 0.03), suggesting that as the number of working hours increases, urinary protein levels also rise.

TPU, urinary creatinine, and calcium showed a significant positive association with the frequency and duration of PCE handling. As the number of handling days per week and hours of daily exposure increased, the levels of these biomarkers also rose in urine.

TPU and urinary sodium showed a negative correlation with handwashing frequency after handling PCE, with values of r (140) = -0.188, p = 0.026 and r(140) = -0.164, p = 0.054, respectively. This suggests that increased handwashing is associated with decreased urinary protein and sodium Level of education and safe handling training were both found to play a role in reducing certain urinary biomarkers. Higher education levels showed a negative correlation with TPU and urinary creatinine, indicating that as education increases, these biomarker levels tend to decrease. Similarly, frequent attendance in safe handling training was associated with lower levels of TPU and urinary calcium. Safe handling training correlation also showed that as a workers attend training frequently, TPU and urinary calcium decrease.

Last but not list urinary sodium correlation with previous job PCE exposure and frequency of spillage yielded a positive significant relation with values of p (140) = 0.173, p=0.041 and p (140) = 0.214, p=0.011 respectively, suggesting as those variables increase, meaning more exposure would result in more sodium output in urine of workers.

Table 10 Spearman correlation result between ordinal risk factors and urinary biomarkers

Variable spearman	Log10 Total Protein (ρ, p)	Log10 Creatinine (ρ, p)	Log10 Calcium (ρ, p)	Log10 Sodium (ρ, p)
Days worked	0.378, <0.001	0.354, <0.001	NS	NS
Total hours worked in a day	0.183, <0.030	NS	NS	NS
How often participants handle PCE	0.324, <0.001	0.183, <0.031	0.318, <0.001	NS
How long participants are exposed to PCE in a day	0.272, <0.001	0.254, <0.002	0.378, <0.001	NS
Hand washing after PCE handling	-0.188, <0.026	NS	NS	-0.164, <0.054
Education level	-0.208, <0.014	-0.170, <0.045	NS	NS
Training received	-0.161, <0.058	NS	-0.196, <0.021	NS
Size of the laundry in m^2	-	-	-	0.175, <0.039
Experience of PCE spillage	-	-	-	0.214, <0.011
Previous job PCE exposure	-	-	-	0.173, <0.041

5.7 Abnormal urinary biomarkers distribution among exposed and control groups.

The following clustered bar graph compares proteinuria (mg/dl), urinary creatinine (mg/dl), urinary calcium (mmol/l), and urinary sodium (mmol/l), respectively, between hotel laundry works and dry cleaners. The place of work is placed on the x-axis, while the abnormal TPU distribution percentage is found on the y-axis. Total urinary protein less than 150 mg/dl is considered normal according to the EPHI guiding reference. From a total of 140 samples, 17 results showed deviation from the normal range, where most of the deviations are found in the dry-cleaning sector. 26 samples showed abnormal urinary creatinine results, where 29-226mg/dl for females and 40-275mg/dl for males were considered as a normal range. 97 tests showed abnormal urinary calcium results, where 2.5-7.5 mmol/l is considered normal. From the urinary sodium test, only 8 samples showed abnormal range, where 27-287mmol/l for females and 40-220mmol/l for males were considered in the normal range.

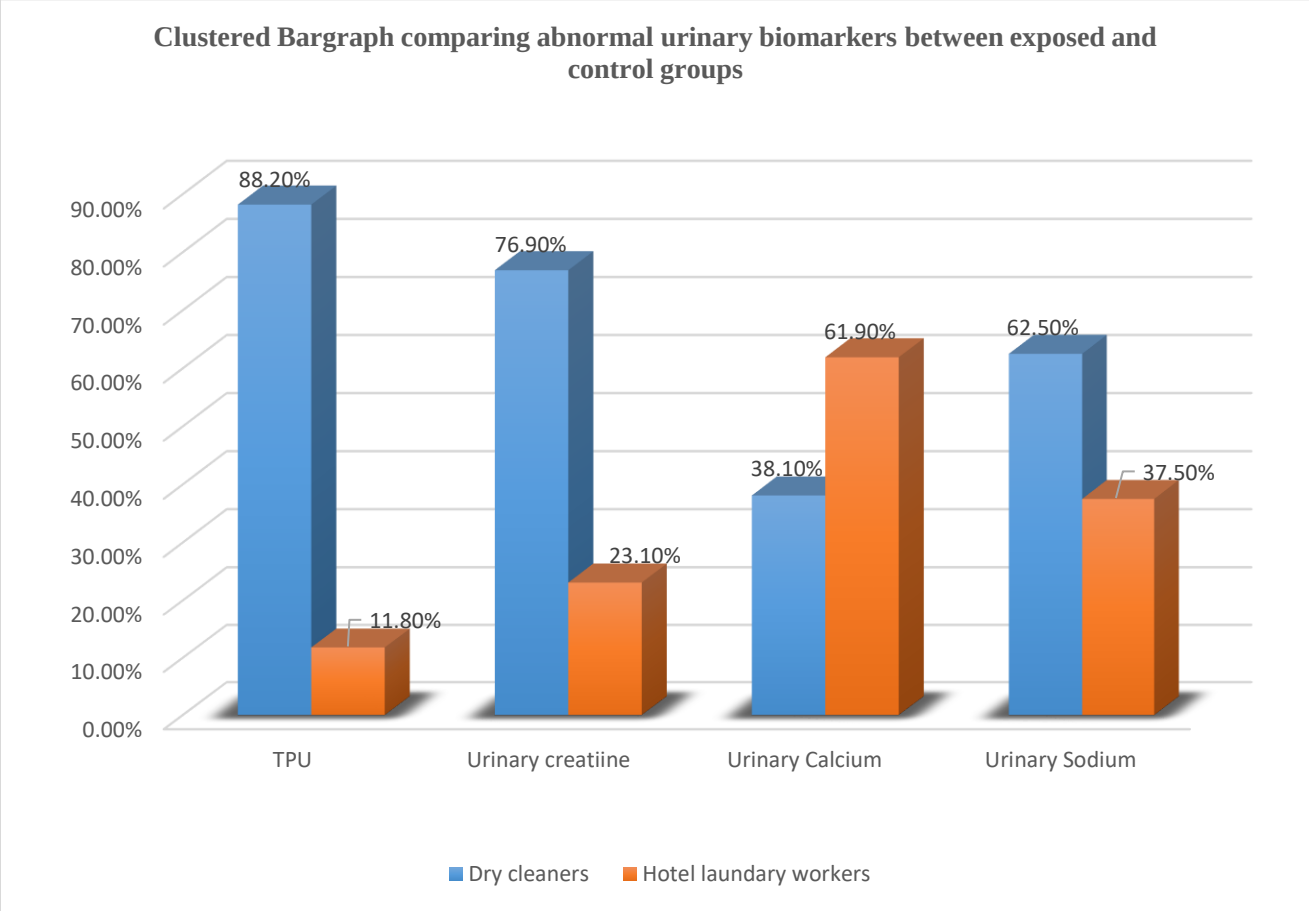

Figure 9 Clustered Bar graph comparing abnormal urinary biomarkers between dry cleaners and hotel laundry workers (N=17 (TPU), N=26 (Urinary creatinine), N=97 (calcium), N=8 (sodium))

5.8 Multiple linear regression analysis of predictors influencing urinary biomarkers

5.8.1 Regression model analysis result for urinary protein

A multiple linear regression analysis was conducted to examine the effects of PCE exposure predictors, demographic variables, health-related factors, and workplace safety practices on urinary total protein (mg/d). The model was statistically significant (F (13, 126) = 4.42, $p < 0.001$), and explaining 31.3% of the variance in proteinuria (R^2=0.313 and adjusted R^2=0.242). A histogram plot of the residuals showed a bell-shaped distribution, a residual verses fitted value plot showed a random scatter of points, all predictors Variance Inflation Factor (VIF) was below 10 and tolerance value for all factors were above 0.1, by which normality, homoscedasticity and absence of multi collinearity assumptions were met for the model.

Thirteen predictors were used in this model including, where they work, how often they handle PCE, how long they are exposed, experience of spillage, age, sex, years of employment, ventilation adequacy, PPE use, hand washing after handling, water intake, smoking status and history of kidney disease, justifying the rule of thumb 10-15 observations per indicator

Among the thirteen predictors, three showed a significant association with urinary total protein levels. The first was the workplace, which had a significant positive effect ($\beta = 0.308$, $p < 0.001$), indicating that being a dry-cleaning worker is associated with a $10^{0.308}$ increase in proteinuria, which is 2.03 times higher than for hotel laundry workers, based on log10 transformed data. The second predictor was years worked in laundry establishments, with a significant association ($\beta = 0.083$, $p < 0.001$), suggesting that each additional year of work is linked to a 1.21 times increase in proteinuria. Lastly, age also showed a significant association with proteinuria, where each additional year of age resulted in a 1.04 increase in proteinuria ($\beta = 0.015$, $p = 0.005$).

Table 11 multiple linear regression model showing significantly associated indicators for total protein

Variable	B	Std. Error	T	Sig.	95% Confidence Interval for B (Lower, Upper)	Tolerance	VIF
Constant	1.951	0.281	6.946	<0.0001	(1.395, 2.507)		
Years of work	0.083	0.024	3.507	<0.001**	(0.036, 0.131)	0.568	1.761
Age in years	0.015	0.005	3.011	<0.003**	(-0.024, -0.005)	0.518	1.930
Work place	0.308	0.065	4.696	<0.001**	(0.178, 0.437)	0.348	2.872

(** = shows significant association)

5.8.2 Regression model analysis result for urinary creatinine

A multiple linear regression was also conducted to see the effect of predictors to urinary creatinine level (mg/dl) after checking the assumptions in the same fashion done above for TPU. 10 predictors were included in the model, age, sex, BMI (Body Mass Index), work place, year of employment, working hour, water intake, smoking, alcohol consumption, history of kidney disease. The model was statistically significant (F (10, 129) = 2.412, p < 0.012), and explained 15.7% of the variance in urinary creatinine (R^2=0.157 and adjusted R^2=0.092).

Only two predictors were significant. The first one is a place of work, which had a significant positive effect (β = 0.152, p =0.009), indicating that being a dry-cleaning worker is associated with 1.42 times increase in urinary creatinine, holding other variables constant. And the second one is years of employment (β = 0.051, p =0.038), indicating that a one-year increase in laundry establishment results in 1.12 times increase in urinary creatinine, holding other variables constant.

Table 12 multiple linear regression model showing selected significantly associated and near significant indicators for Urinary Creatinine

Variable	B	Std. Error	T	Sig.	95% Confidence Interval for B (Lower, Upper)	Tolerance	VIF
Constant	1.817	0.280	6.490	<0.0001	(1.263, 2.371)		
Place of work	0.152	0.058	2.644	<0.009**	(0.038, 0.266)	0.514	1.947
Years of experience	0.051	0.024	2.094	<0.038**	(0.003, 0.098)	0.628	1.593
History of Kidney disease	-0.249	0.149	-1.672	<0.097**	(-0.543, 0.046)	0.918	1.090

(** = shows significant association)

5.8.3 Regression analysis result for urinary calcium and sodium

Separate regression analysis was done for this two urinary biomarkers by adding approximated categorical dietary intake of sodium and calcium predictors from the recent meal they had before sample collection along with predictors like age, sex, BMI, place of work, working hour, water intake, smoking status, alcohol consumption, past history of kidney disease, experience of PCE spillage and availability of adequate ventilation. the model for urinary calcium was statistically significant (F (14, 124) = 4.21, $p < 0.001$), and explaining 32.9% of the variance in urinary calcium (R^2=0.322 and adjusted R^2=0.246), however the only significant predictor was place of work which had a significant positive effect (β = 0.438, $p < 0.001$), indicating being a dry cleaning worker is associated with a 2.74 times increase in urinary calcium, holding other variables constant. Regression analysis for urinary sodium showed a non-significant result in both the model and in all of the predictors, as a result, we excluded it from the regression report.

Table 13 multiple linear regression model showing selected significantly associated and near significant indicators for Urinary Calcium

Variable	B	Std. Error	T	Sig.	95% Confidence Interval for B (Lower, Upper)	Tolerance	VIF
(Constant)	-0.755	0.518	-1.458	0.147	(-1.779, 0.270)	—	—
Place of work	0.438	0.108	4.050	<0.001**	(0.224, 0.652)	0.382	2.618
Age	0.013	0.008	1.500	0.136	(-0.004, 0.029)	0.512	1.951
Work hours	0.047	0.055	0.862	0.391	(-0.061, 0.156)	0.534	1.873
Sex	0.053	0.079	0.672	0.503	(-0.103, 0.210)	0.768	1.302
Alcohol use	-0.053	0.061	-0.865	0.389	(-0.173, 0.068)	0.794	1.259
Years of experience	-0.018	0.040	-0.456	0.649	(-0.097, 0.061)	0.606	1.651
Calcium intake	0.052	0.053	0.991	0.324	(-0.052, 0.156)	0.846	1.182

(** = shows significant association)

6. Discussion

The result of our study revealed the mean and median values of the six biomarkers along with workers disease symptom while at work. A higher median value of TPU (102 mg/d) in dry-cleaners was observed as compared to hotel laundry workers (54.5 mg/d). In the same way the median value of urinary creatinine in the exposed group (193.78 mg/dl) was higher compared to the control group (142.93 mg/dl). Urinary calcium also showed a higher mean value in dry-cleaning workers (2.6 mmol/l) than hotel laundry workers (0.835 mmol/l). However there was no much meaningful difference in S. Creatinine between the two groups just like urinary sodium. Last but not list, even though significant association was not found between the two groups BUN mean value, a small increase in dry-cleaners (15.5 mg/dl) was observed than the control group (12.5 mg/dl). The prevalence of nausea while at work was 20.0% and 15.7% for the exposed and control groups respectively showing a slight increase in the exposed group. The occurrence of dizziness and headache while on the job was also higher in dry-cleaners, with 82.9% and 65.7% prevalence in exposed and control groups respectively.

From the four urinary and two serum biomarkers, TPU was the most crucial and directly linked to Perchloroethylene induced nephrotoxicity (53). Different studies have identified urinary protein levels as a sensitive indicator of kidney damage caused by toxic substances (77, 80). Our result showed a significant increase in proteinuria in dry cleaners as compared to hotel laundry workers, with a P-value of 0.001 in both parametric and non-parametric tests. Similar to our findings, two studies conducted in Germany and the Netherlands to assess the renal effects of PCE in dry cleaners showed a significant increase in proteinuria in the exposed groups compared to the control groups(47, 76). Dry cleaners accounted for 88.2% of observed abnormal TPU (>150mg/d) in our study, while 87% of subjects in the Italy study showed an increased high molecular weight protein in urine, which was associated with tubular alteration (49). On the other hand, some studies contradicted our finding. A study conducted in the US found no direct association between PCE exposure and urinary biomarkers from 192 dry cleaning workers, even though protein/creatinine showed a weak positive association (16). In contrast to the previous studies, a study conducted in the Czech Republic found no significant difference in total urinary protein between the exposed

and control groups, countering the general claim of a clear association between PCE exposure and proteinuria (83). Prevalence of abnormal value of protein showed no significance between the exposed and control groups in another study conducted in the Netherlands (51), whereas abnormal TPU in our findings showed a significant association with a 0.007 p-value, by which hotel laundry workers had 88.2% lower odds of TPU abnormality compared to dry cleaners.

According to the agreed-upon pathophysiological study of Proteinuria, PCE exposure results in higher protein in urine by compromising the glomerulus, which is the key filtration unit within in nephron that allows the passage of tiny water and electrolyte molecules, while blocking large molecules like protein (84). When this filtration unit is compromised by PCE (controlling for other factors like diabetes, hypertension), protein output in urine increases. (14, 81). According to the National Kidney Foundation, proteinuria levels greater than 150 mg/d are considered clinically abnormal and may indicate kidney dysfunction(61). Our finding aligns with the pathophysiological reasoning and the abnormal proteinuria level to determine PCE effect in TPU output and provide evidence for PCE effect on kidney health.

Urinary creatinine was used to show the effect of PCE on kidney filtration as a result of toxic-induced renal damage, resulting in GFR reduction (48). It is a key indicator of overall kidney function, since it is a result of creatine degradation from muscle (86). However, most of the time, it is used as a normalizer for other biomarkers (87). PCE exposure is believed to decrease GFR (48), which in turn decreases urinary creatinine and indicates acute and chronic kidney disease (88).

Some studies used S. creatinine only as a biomarker to detect nephrotoxicity, but found no significant association, which is similar to our S. creatinine finding. The results of a study in Iran found no significant difference in Cr levels between the two groups (57). Similarly, a study conducted in China to see the effects of the exposure on liver and kidney functions yielded negative results as judged by the emission enzyme activities (89). Complimentarily a study in Italy found that chronic exposure to organic solvents did not significantly alter urinary creatinine level, although it stated that urinary creatinine can't be used to detect renal dysfunction alone (17).

Our research found a significant urinary creatinine difference between the exposed and the control group with a log 10 transformed p-value of (0.002), and 76.9% of the abnormal creatinine results being found in the dry cleaners, however majority of them showed an increased urinary creatinine

above the range (>226 mg/dl for female and >275 mg/dl for males). Which might come from renal tubular damage caused by PCE metabolites like S-(1, 2, 2-trichlorovinyl)-L-cysteine (TCVC), which induce oxidative stress and affect tubular integrity (47, 86). As a result kidneys' ability to filter will decrease, and GFR would fall, so to compensate for this, the proximal tubules would increase creatinine secretion and another possibility is tubular epithelial barrier might be compromised by PCE metabolites, resulting in passive leakage of Cr to urine (91). From this, we can hypothesize that the non-significant result of creatinine from the above research could be due to the compensation secretion of creatinine by proximal tubules or due to leakage of Cr through compromised tubular epithelial barrier during the early stages of PCE Impact on kidney.

Based on a few PCE-induced tubular damage studies, we formulated a scientifically relevant hypothesis related to urinary calcium and sodium and tested both of them.

Urinary calcium is an important indicator of tubular function, which could detect early kidney damage caused by PCE, since the GSH metabolite results are believed to cause tubular toxicity (53). Kidney reabsorbs calcium in order to balance body's Ca level. But if anything happens to the kidney, such as being compromised by PCE exposure, hypercalcemia could be seen in urine (81). As calcium output increases in urine, the chance of crystallization with other substances increases, leading to kidney stone or calcium phosphate formation (78).

Reabsorption of urinary calcium occurs in the proximal tubule, which would indicate a secondary change in ion handling as a result of PCE-induced toxicity, since GSH metabolites causes tubular damage. Urinary sodium would also show PCE toxicity in the same fashion as calcium, in addition to its ability to indicate electrolyte imbalance caused by the solvent (51, 52, 77–79).

Our research findings showed a median value of 2.6 mmol/l and 0.835 mmol/l for exposed and control groups, respectively. Just like our hypothesis, a higher value of calcium in urine output was seen in dry cleaners. The difference between the two groups was significant with p-value <0.001 for both independent t-test and Mann-Whitney test; however, dry cleaners account only 38.1% of the observed abnormal urinary calcium. Most of the abnormal results were below the normal range, which is less than 2.5 mmol/l. Most of the study participants have a calcium level below the normal range, but those who work in the dry cleaners have more urinary release as compared to hotel laundry workers.

Urinary sodium is an important biomarker to regulate the kidneys' ability to maintain electrolyte balance. In normal conditions, 99% of Na is reabsorbed, where 70% of it is reabsorbed by the proximal tubule, 25% by the Loop of Henle in the thick ascending limb, and the rest 5 % is absorbed by the distal tubule and collecting duct (92).

PCE metabolites could impair the thick ascending limb, which is a site for sodium reabsorption. This, in turn, results in disruption in the sodium-potassium-2-chloride (NKCC2) cotransporter, which results in increased urinary sodium (48), our study didn't find any significant difference between the exposed and control groups.

Although no significant difference in BUN was found between the exposed and control groups, the data showed higher values in dry cleaners, though still within the normal range. A similar result was observed in a study conducted in Iraq, where dry cleaners also exhibited higher BUN values within the normal range(57). Most of the results are normal in our finding, but the higher value in dry cleaners indicates early renal stress caused by PCE exposure as a result early detection and routine monitoring would be valuable in order to prevent kidney toxicity, since long employment, PPE utilization and other factors could influence it even if we haven't found significant correlation in our study for BUN.

The result from the socio-demographic factors showed that 44.3% of participants had 4-10 years of work experience, with only 2.9% working for more than 10 years. Additionally, 67.1% of workers used PPE, such as gloves and masks. In comparison, a study in Iraq reported 62.5% of workers had 5-10 years of experience, and 37.5% worked for more than 10 years. However, their study found that 28.1% of workers did not use PPE. This discrepancy between the two studies may be attributed to the smaller sample size of 32 in the Iraqi study, as well as the relatively recent emergence of dry-cleaning establishments in our country, which has led to a narrower range of employment experience. These factors could influence the percentages observed(79).

Due to the nature of PCE, daily exposure to the solvent results in headache and dizziness, 82.9 % (n=58) of the dry cleaners felt headache and dizziness frequently in the workplace. Dry cleaners in Iraq also showed symptoms like dizziness 9(28.1%) and Headache 12(37.5%) (79). Similarly, a subjective symptom assessment related to PCE study conducted in Asia showed a 44.6% and

23.2% prevalence in dizziness and headache respectively, with 15% of the workers experiencing appetite loss, which is close enough to our study where 20% of dry cleaners showed loss of appetite, however 18.6 % of participants reported both nausea and loss of appetite, which suggests a high prevalence in Ethiopian dry cleaners.

A study conducted in US documented renal and related symptoms like dizziness, altered urination in dry cleaners with high PCE exposure (16). Supporting this claim our study also found high prevalence of dizziness (82.9%) and altered urination (61.4%). This result could decrease significantly if individual PPE utilization is improved beyond supply.

Another study about PCE in US identified inadequate safety training as a risk amplifier in solvent exposed cohorts (93). Similar to this, our study also observed the need for improvement in safety training in dry cleaners, only 2.9% of them receives regular training, while 47.1% of dry cleaners have never taken training in their current work place.

The role of perchloroethylene exposure on general kidney health has been a debatable issue in the past, where some researchers confirmed kidney effects on animals but concluded by saying'' we don't know what those findings mean to humans''(1).Another article also supported this claim by saying we haven't found evidence of PCE on kidney health effect (3). On the contrary, some research confirmed urinary and serum biomarkers alteration as a result of PCE exposure (49, 73–76). The biomarkers used in each study are different, which shows that there are no agreed-upon specific biomarkers; however, urinary protein is the most commonly used biomarker, along with other indicators like BUN, creatinine, enzymes, and electrolytes (47,51). The absence of specific biomarkers, the absence of consistent findings, and the developing investigation of PCE metabolism and toxicity are believed to be some of the reasons why ACGIH suggested further PCE health effect studies (12).

In conclusion, although we haven't quantified the exposure status in our study, different exposure related factors did influence the nephrotoxic biomarkers, as a result preventive measures are needed to be taken in occupation setting in order to keep the health and safety of dry cleaners.

7. Strengths and limitations of the study

7.1 Strength of the study

This study is the first to explore occupational PCE health effects in Ethiopia, specifically nephrotoxicity. The study assessed four urinary biomarkers and two serum biomarkers; this multiple biomarker approach helped to increase the chance of detecting nephrotoxicity as a result of PCE exposure. By comparing dry cleaners with hotel laundry workers, this study tried to minimize other detergent chemicals in nephrotoxicity, since dry cleaning shops also provide normal wet cleaning like hotel laundries. We exclude participants who used temporal medication to control unwanted changes in biomarkers. BMI, immediate previous meal, individual and family kidney history, water intake, and other factors were included to control for cofounders.

The study was a multi-center cross-sectional study, which managed to address multiple dry-cleaning establishments and hotel laundries, which decreased specific location factors by incorporating diverse workplace cultures. The cross-sectional nature of the study allowed us to capture both exposure-related questions and the biomarkers at the same time, which generates helpful data for future cohort studies.

7.2 Limitations of the Study

The study didn't measure and quantify specific exposure assessment due to unavailability of direct reading devices and high cost of biological assessment, as a result, dose response linkage was not performed and since we used cross-sectional study, cause and effect relationship couldn't be assured, by which the observed biomarker difference is unclear whether it was the result of cumulative exposure or recent exposure. Enzyme biomarkers such as NAG were not used, since we didn't have the analytical machines for them.

Data collection and handling differences due to individual data collectors' variability could also influence the sample, although training was given to reduce this error. Lack of control over the confounding variables is also another limitation. And finally the study didn't relate the result of the biomarkers with clinical outcomes and imaging's, so it only assed if there was a risk or not.

8. Conclusion

The finding showed a significant difference between dry cleaners and hotel laundry workers in TPU, urinary creatinine and calcium, which suggests PCE role on those biomarkers. Although the study failed to demonstrate a significant difference in urinary sodium, BUN and serum creatinine, a higher prevalence of abnormality was seen in dry cleaners, which aligns with the biological plausibility of PCE-induced toxicity in dry cleaners. The cross-sectional nature of the study and the absence of direct exposure assessment hindered us from concluding definitively, but still the biomarkers showed a possible physiologic change and risk of nephrotoxicity as a result of the solvent. In conclusion, the result of this study suggests that dry cleaners need to be more cautious as they are more susceptible to kidney toxicity however, a longitudinal study is needed to confirm the cause-and-effect relation.

9. Recommendations

Based on the findings of this study, recommendations are directed toward the Ministry of Labor, dry-cleaning businesses and employers, the Addis Ababa Health Office, and future researchers. These stakeholders are urged to recognize the potential health risks of PCE, take active measures to minimize its impact, and contribute to identifying and promoting safer alternatives. For the Ministry of Labor and Skills in Ethiopia, to study, decide, limit, and enforce national PCE occupational exposure limit in order to prevent its health impact. Additional workplace follow-up inspection, training campaign for dry cleaners on how to handle toxic chemicals and detergents, preparation of guiding manuals, and promotion of safe working practice are crucial steps that need to be done by the ministry to prevent the adverse health impact of PCE chemical.

For the Addis Ababa health office, to conduct and implement an occupational health surveillance system in those groups of workers who are highly exposed to PCE, to collaborate with different stakeholders and support related research initiatives and training opportunities.

For dry cleaning employers, to improve and adopt safer alternatives, like other chemicals or procedures, in order to minimize workers' health risk. Providing the necessary PPE like long sleeve coats, respirators, gloves and googles, improving the ventilation system, encouraging workers to do regular medical checkups, and arranging health-related trainings, developing guiding procedures, and making sure that the workers are following them.

For future researchers, to include exposure assessment and use wide panel of biomarkers like NAG and other enzymes along with clinical outcomes, to conducted longitudinal study to confirm cause and effect association, last but not list to widen and explore different health impact of PCE such as, cancer, liver toxicity, neurological problems and so on.

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11. ANNEX

ANNEX 1- Questioners in English format

1. Demographic questions			
No.	Variable	Response	Skips
101	Sex	1. Male 2. Female	
102	Age in years	_____	
103	What is your highest level of education?	A. Elementary B. High School C. Diploma/Certificate D. Bachelor's Degree E. Postgraduate Degree and above	
104	Where do you work?	A. In Dry cleaning laundry B. In hospitality laundry	
105	How many years have you worked in the dry cleaning industry? / In hotel laundry?	A. Less than 1 year B. 1 to 3 years C. 4 to 6 years D. 7 to 10 years E. 11 to 15 years F. More than 15 years	
106	What is your job role?	A. Machine operator B. Manager/Supervisor C. Ironer D. Receptionist	
107	How many days do you work in a given week?	A. <3 days B. 4 days C. 5 days D. 6 days E. 7 days	

108	How many hours per day do you work in the cleaning service?	A. <3 hours B. 4-6 hours C. 7-8 hours D. > 8hours	
2. Perchloroethylene exposure related questions			
201	How often do you handle perchloroethylene (PCE)?	A. Daily B. Weekly C. Monthly D. Never	
202	How long are you exposed to PCE during a typical workday?	A. Less than 1 hour B. 1-4 hours C. More than 4 hours D. Not exposed	
203	Have you worked in any other jobs that involved exposure to PCE or similar chemicals?	A. Yes (please specify) B. No	
204	Have you ever experienced a spillage of PCE while at work?	A. Yes B. No	
3. Safety related questions			
301	Is there adequate ventilation in your workplace when using PCE?	A. Yes, very good B. Moderate C. Poor	
302	What type of ventilation system is available in your shop?	A. Natural ventilation only B. Forced ventilation only C. Combination of both D. No ventilation system	

303	What type of protective equipment do you use when handling PCE?	A. Gloves B. Masks C. Respirators D. Goggles E. None	
304	Have you received training on safe handling of PCE?	A. Yes, regularly trained B. Yes, once C. No, not trained	
305	Do you wash your hands after handling PCE?	A. Always B. Sometimes C. Never	
306	Are there emergency procedures in place for PCE spills at your workplace?	A. Yes, well-defined B. Some procedures exist C. No procedures	
307	Are you provided with personal protective equipment (PPE)?	A. Yes, regularly provided B. Yes, occasionally provided C. No, not provided	
308	Are there regular inspections of the dry cleaning equipment for leaks?	A. Yes, regularly inspected B. Occasionally inspected C. Not inspected	
309	How often is the equipment maintained?	A. Monthly B. Quarterly C. Annually D. Rarely E. Never	

4. Health related questions			
401	Do you notice any immediate symptoms after handling PCE?	A. Yes, frequently B. Yes, occasionally C. No symptoms	
402	Have you noticed any changes in your health since starting work in dry cleaning?	A. Yes, significant changes B. Yes, minor changes C. No changes	
403	Have you noticed any changes in your urination patterns? such as:	A. Increased frequency of urination B. Decreased urine output C. Foamy or discolored urine D. No changes	Select all that apply
404	Have you experienced any swelling in your legs, ankles, or feet?	A. Yes, frequently B. Occasionally C. Rarely D. No swelling at all	
405	How often do you feel unusually tired or weak?	A. Always B. Often C. Sometimes D. Neve	
406	Have you experienced nausea or a loss of appetite recently?	A. Yes, both nausea and loss of appetite B. Yes, nausea only C. Yes, loss of appetite only D. No, neither	
407	Have you experienced persistent itching without an apparent cause?	A. Yes, frequently B. Occasionally C. Rarely D. No itching	

408	Do u think the health issues got something to do with PCE exposure?	A. Yes B. No	
409	Do you have any pre-existing medical conditions	A. Yes B. No	
410	Have you experienced any symptoms such as dizziness, headaches, or nausea more frequently while at work than other places?	A. Yes B. No	
411	Do you have any history of kidney problems?	A. Yes B. No	
412	Have you undergone any kidney function tests (e.g., total protein, albumin, urine analysis) in the past year?	A. Yes B. No	
413	If your answer for the above question is yes what were the results?		
414	Is there a family history of kidney disease or related conditions?	A. Yes B. No	
415	How many glasses of water do you drink per day?	A. Less than 2 B. 2-4 C. 5-7 D. More than 7	

416	Do you have any other health conditions that could affect kidney function? (Select all that apply)	A. Diabetes B. Hypertension C. Liver disease D. None	
417	Have you ever been diagnosed with any chronic illnesses?	A. Yes B. No	If yes
418	If your answer for the above question is yes mention what they are?		
419	Do you smoke or have a history of smoking?	A. Current smoker B. Former smoker C. Never smoked	
420	Do you consume alcohol regularly?	A. Yes, frequently B. Occasionally C. No	
421	What does your typical diet consist of?		
5. **Work environment related questions**			
501	What is the size of your dry cleaning shop? In meter/kilometer square	_____	
502	How many employees work at your dry cleaning shop? / In the hotel	A) 1-4 employees B) 5-10 employees C) 11-20 employees	

	laundry service?		
503	How are chemicals stored in your shop?	A) In a designated, locked area with proper labeling B) On shelves without specific safety measures C) In open containers or areas	

ANNEX 2: Questioners in Amharic format

የስነሕዝብ ጥያቄዎች	ምላሽ
101. *ፆታ*	ሀ. ወንድ ለ. ሴት
102. እድሜወት በአመት	……
103. ከፍተኛ የትምህርት ደረጃወት	ሀ.አንደኛ ደረጃ ለ.ሁለተኛ ደረጃ ትምህርት ቤት ሐ.ዲፕሎማ/ሰርቲፊኬት መ.የባችለር ዲግሪ ሠ.የድህረ ምረቃ ዲግሪ እና ከዚያ በላይ
104. የምትሰሩበት ቦታ	A. ደረክ ላዉንደሪ ማጠቢያ ቦታ B. በሆትል ላንድሪ
105. በደረቅ ላዉንደሪ ጽዳት ውስጥ ስንት ዓመት ሰርተዋል? /በሆቴል ልብስ ማጠቢያ ውስጥ?	ሀ.ከ 1 ዓመት በታች ለ 1 እስከ 3 ዓመታት ሐ. ከ 4 እስከ 6 ዓመታት መ. ከ 7 እስከ 10 ዓመታት ሠ. ከ 11 እስከ 15 ዓመታት ረ.ከ 15 ዓመታት በላይ
106. የስራ ሚናዋት ምንድን ነው?	ሀ.ደረቅ ጽዳት ሠራተኛ ለ.አስተዳዳሪ/ተቆጣጣሪ ሐ.ካዉያ መተኮሰ መ.እንግዳ ተቀባይ ሠ.የሆቴል የልብስ ማጠቢያ ሰራተኞች
107. በአንድ ሳምንት ስንት ቀን ይሰራሉ?	ሀ. ከሶስት ቀን በታች ለ. 4 ቀን ሐ. 5 ቀን መ. 6 ቀን

የስነሕዝብ ጥያቄዎች	ምላሽ
	ሠ. 7 ቀን
108. በአንድ ቀን ስንት ሰዓት ይሰራሉ?	A. ከ3 ሰዓት በታች B. 4-6 ሰዓት C. 7-8 ሰዓት D. ከ8 ሰዓት በላይ

ቁጥር	**ከፐርክሎሮኢታይን መጋለጥ ጋር የተያያዙ ጥያቄዎች**	**ምርጫ**
201	በፔርክሎሮኤተለን (PCE) ምን ያህል ጊዜ ያገኛሉ?	ሀ. በየቀኑ ለ. በሳምንት አንድ ጊዜ ሐ. በወር መ። ፈጽሞ
202	በተለመደው የስራ ቀን ለ PCE ለምን ያህል ጊዜ ተጋልጠዋል?	ሀ.ከ 1 ሰዓት ያነሰ ለ. 1-4 ሰዓታት ሐ. ከ 4 ሰዓታት በላይ መ. አልጋለጠም
203	ለ PCE ወይም ለተመሳሳይ ኬሚካሎች መጋለጥን በሚያካትቱ ሌሎች ስራዎች ላይ ሰርተሃል?	ሀ. አዎ (እባክዎት ይገልጹ) ለ. አይ
204	በስራ ላይ እያሉ የ PCE መፍሰስ አጋጥሟቸዋል?	ሀ. አዎ ለ. አይ
ተ.ቁ	ከደህንነት ጋር የተያያዙ ጥያቄዎች	ምርጫ
301	PCE ሲጠቀሙ በስራ ቦታዎ በቂ የአየር ማናፈሻ አለ?	ሀ. እጅግ ጥሩ ለ. መካከለኛ ሐ. ደካማ
302	በሱቅዎ ውስጥ ምን አይነት የአየር ማናፈሻ ስርዓት አለ?	ሀ. ተፈጥሯዊ አየር ማናፈሻ ብቻ ለ. የመሳሪያ አየር ማናፈሻ ብቻ።

ቁጥር	ከፐርክሎሮኢታይን መጋለጥ ጋር የተያያዙ ጥያቄዎች	ምርጫ
		ሐ. የሁለቱም ጥምረት መ. የአየር ማናፈሻ ስርዓት የለም
303	PCEን በሚይዙበት ጊዜ ምን ዓይነት የመከላከያ መሳሪያዎችን ይጠቀማሉ?	ሀ.ጓንቶች ለ.ጭምብሎች ሐ.የመተንፈሻ አካላት መ.መነጽር ሠ.ምንም
304	የ PCE ደህንነቱ የተጠበቀ አያያዝ ላይ ስልጠና ወስደዋል?	ሀ.አዎ፣ በመደበኛነት የሰለጠኑ ለ.አዎ, አንድ ጊዜ ሐ.አይ፣ አልሰለጠነም።
305	PCEን ከያዙ በኋላ እጆችዎን ይታጠባሉ?	ሀ.ሁሌም ለ.አንዳንድ ጊዜ ሐ.በጭራሽ
306	በስራ ቦታዎ ላይ ለ PCE መፍሰስ የአደጋ ጊዜ ሂደቶች አሉ?	ሀ.አዎ, በደንብ የተገለጸ ለ.አንዳንድ ሂደቶች አሉ። ሐ.ምንም ሂደቶች የሉም
307	የግል መከላከያ መሳሪያዎች (PPE) ተሰጥቷቸዋል?	ሀ.አዎ፣ በመደበኛነት የቀረበ። ለ.አዎ፣ አልፎ ይቀርባል። ሐ.አይ፣ አልቀረበም።
308	ለደረቅ ማጽጃ መሳሪያዎች በየጊዜው ምርመራዎች አሉ?	አ.አዎ, በመደበኛነት ይመረመራሉ ለ.አልፎ አልፎ ይመረመራል ሐ.አልተመረመረም።
309	መሳሪያዎቹ ስንት ጊዜ እንደተጠበቁ ታውቃላችሁ?	ሀ.ወርሃዊ

ቁጥር	ከፐርክሎሮኢታይን መጋለጥ ጋር የተያያዙ ጥያቄዎች	ምርጫ
		ለ.በየሩብ ሐ.በየዓመቱ መ.አልፎ አልፎ ሠ.በጭራሽ

ተ.ቁ	ከጤና ጋር የተያያዙ ጥያቄዎች	ምርጫ
401	PCEን ከያዙ በኋላ ወዲያውኑ ምልክቶችን ያስተውላሉ?	ሀ.አዎ ፣ ብዙ ጊዜ። ለ.አዎ, አልፎ ሐ.ምንም ምልክቶች የሉም
402	ከደረቅ ማጠቢያ ሥራ ከጀመርህ ጀምሮ በጤና ላይ ምንም አይነት ለውጥ አስተውላለህ?	ሀ.አዎ, ጉልህ ለውጦች ለ.አዎ, ጥቃቅን ለውጦች ሐ.ምንም ለውጦች የሉም።
403	በሽንትዎ ላይ ምንም አይነት ለውጥ አስተውለሃል?	ሀ.የሽንት ድግግሞሽ መጨመር ለ. የሽንት ውጤት መቀነስ ሐ. አረፋ ወይም ቀለም ያለው ሽንት መ. ምንም ለውጦች የሉም።
404	በእግሮችዎ፣ በቁርጭምጭሚቶችዎ ወይም በእግሮችዎ ላይ ምንም አይነት እብጠት አጋጥሞዎታል?	ሀ. በተደጋጋሚ ለ. አንዳንድ ጊዜ ሐ. እጅግ አነስተኛ መ. ፈጽሞ አልተከሰተም
405	ባልተለመደ ሁኔታ ድካም ምን ያህል ጊዜ ይሰማዎታል?	ሀ. ሁልጊዜ ለ. ብዙ ጊዜ ሐ. አንዳንድ ጊዜ መ. ፈጽሞ
406	በቅርብ ጊዜ የማቅለሽለሽ ወይም የምግብ ፍላጎት ማጣት አጋጥሞዎታል?	አ. አዎ, ሁለቱም ማቅለሽለሽ እና የምግብ ፍላጎት ማጣት ለ. አዎ, ማቅለሽለሽ ብቻ ሐ. አዎ የምግብ ፍላጎት ማጣት ብቻ። መ. አይ፣ ሁለቱም
407	ያለ ግልጽ ምክንያት የማያቋርጥ ማሳከክ አጋጥሞዎታል?	ሀ.አዎ ፣ ብዙ ጊዜ። ለ. አልፎ ሐ. አልፎ መ. ማሳከክ የለም
408	የጤና ጉዳዮች ከ PCE መጋለጥ ጋር ግንኙነት ያላቸው ይመስላችኋል?	አ.አዎ

		ለ.አይ
409	ቀደም ሲል የነበሩ ህመሞች አሉዎት	ሀ.አዎ ለ.የለም
410	በስራ ላይ እያሉ እንደ ማዞር፣ ራስ ምታት ወይም ማቅለሽለሽ ያሉ ምልክቶች ከሌሎች ቦታዎች በበለጠ በተደጋጋሚ አጋጥሟቸዋል?	ሀ.አዎ ለ.የለም
411	የኩላሊት ችግር ታሪክ አለቦ?	ሀ.አዎ ለ.የለም
412	ባለፈው አመት ምንም አይነት የኩላሊት ተግባር ምርመራ (ለምሳሌ አጠቃላይ ፕሮቲን፣ አልቡሚን፣ የሽንት ትንተና) ወስደዋል?	ሀ.አዎ ለ.የለም
413	ከላይ ለተጠቀሰው ጥያቄ መልስህ አዎ ከሆነ ውጤቱ ምን ነበር?	
414	የኩላሊት በሽታ ወይም ተዛማጅ ሁኔታዎች የቤተሰብ ታሪክ አለ?	ሀ.አዎ ለ.የለም
415	በቀን ስንት ብርጭቆ ውሃ ይጠጣሉ?	ሀ.ከ 2 በታች። ለ.2-4 ሐ.5-7 መ.ከ 7 በላይ
416	የኩላሊት ሥራን ሊጎዱ የሚችሉ ሌሎች የጤና ችግሮች አሉዎት? (የሚተገበሩትን ሁሉ ይምረጡ)	ሀ.የስኳር በሽታ ለ.የደም ግፊት ሐ.የጉበት በሽታ መ.ምንም
417	ሥር የሰደደ ሕመም አለቦት	ሀ.አዎ ለ.የለም
418	ከላይ ለተጠቀሰው ጥያቄ መልስህ አዎ ከሆነ? ምን እንደሆኑ ይጥቀሱ	
419	ያጨሳለሉ ወይስ የማጨስ ታሪክ አለዋት?	አ.የአሁኑ አጫሽ ለ.የቀድሞ አጫሽ ሐ.በጭራሽ አታጨስም።
420	አልኮል አዘውትረው ይጠጣሉ?	አ.አዎ ፣ ብዙ ጊዜ። ለ.አልፎ አልፎ ሐ.የለም

421	የተለመደው አመጋገብዎ ምን ያካትታል?	

ተ.ቁ	ከስራ አካባቢ ጋር የተያያዙ ጥያቄዎች	ምርጫ
501	የደረቅ ማጽጃ ሱቅዎ መጠን ምንድን ነው? በሜትር/ኪሎሜትር ካሬ	
502	በደረቅ ማጽጃ ሱቅዎ ውስጥ ስንት ሰራተኞች ይሰራሉ? /በሆቴል የልብስ ማጠቢያ አገልግሎት?	1. 1-4 ሰራተኞች 2. 5-10 ሰራተኞች 3. 11-20 ሰራተኞች
503	ኬሚካሎች በሱቅዎ ውስጥ እንዴት ይቀመጣሉ?	ሀ.በተሰየመ፣ የተቆለፈ ቦታ ከትክክለኛ መለያ ጋር። ለ) ልዩ የደህንነት እርምጃዎች ሳይወሰዱ በመደርደሪያዎች ላይ ሐ) በክፍት ኮንቴይነሮች ወይም ቦታዎች

Annex 3: Consent Form for study participants

I have read and understood the information provided in this consent form, and I voluntarily agree to participate in the research study on Nephrotoxicity Risk among Dry cleaners Exposed to Perchloroethylene in Addis Ababa, 2025.I understand that my participation is entirely voluntary, and I am free to withdraw at any time without any other consequences. I also understand that my identity will be kept confidential, and my data will be handled securely.

Participant's
Name: _____
Participant's
Signature: _____ Date: ___

Principal Investigator's
Name: _____ Principal
Investigator's Signature: _____ Date: ___

ANNEX 4: Consent form Amharic version

ለጥናት ተሳታፊዎች የስምምነት ቅጽ

በዚህ የስምምነት ቅጽ ላይ የቀረበውን መረጃ አንብቤ ተረድቻለሁ፣ እና በአዲስ አበባ፣ 2025 ለፐርክሎሮኢታይሊን በተጋለጡ ደረቅ ማጽጃዎች መካከል በኔፍሮቶክሲክነት ስጋት ላይ በተደረገው የምርምር ጥናት ላይ ለመሳተፍ በፈቃደኝነት ተስማምቻለሁ። የእኔ ተሳትፎ ሙሉ በሙሉ በፈቃደኝነት ላይ የተመሰረተ እንደሆነ ተረድቻለሁ, እና ምንም አይነት መዘዝ ሳያስከትል በማንኛውም ጊዜ ለመውጣት ነፃ ነኝ. እንዲሁም ማንነቴ በሚስጥር እንደሚጠበቅ ተረድቻለሁ፣ እና የእኔ መረጃ ደህንነቱ በተጠበቀ ሁኔታ ይያዛል።

የተሳታፊው ስም፡

የተሳታፊው ፊርማ፡ ቀን፡-

የዋና መርማሪ ስም፡

የዋና መርማሪ ፊርማ፡

ቀን፡-

ANNEX 5- Ethiopian Public Health Institute (EPHI) Specimen Collection Reference Manual

Before Collecting the Specimen

1. Prepare equipment and supplies. Ensure labels available.
2. Explain the procedure to the participant and/or their significant other if present
3. Complete requisition form with type of specimen, time of collection, site of collection, you name (name of person collecting specimen), patient's location, etc.
4. Verify the patient's identification by using two patient identifiers per policy.
5. Remember to always label the specimen with the required information:
   - Collection Date and Time
   - Patient Identification (Name, DOB, etc.)
   - Collection Method. The requisition must be filled out and the exact source of the specimen Indicated e.g. "midstream urine".
   - Test required.
6. Wash hands thoroughly with soap and water and dry the hands with a paper towel.
7. Follow Standard Precautions for all patients. Use appropriate protective equipment such as gloves, masks, and/or face shields.

Midstream Urine Collection

From Females

The data collector should instruct the participants on proper collection and how to minimize contamination to the specimen and the collection container.

The following protocol should be followed:

1. Wash hands thoroughly with soap and water and dry the hands with a paper towel.
2. with one hand spread the labia and hold them apart and use sterile wipe/cleansing towel to clean the meatus from front to back.
3. Start voiding urine, after the first portion of the urine is passed collect apportion of the urine by voiding into the sterile container.
4. Avoid contact between the container and the legs, vulva, or clothing. Do not touch the inside of the container or lid.

5. Stop collection when container is about half-full.

6. Screw cap on container.

7. Wash hands thoroughly with soap and water, and dry with a clean towel.

8. Give specimen to personnel.

From Males

The healthcare provider should instruct the patient on proper collection and how to minimize contamination to the specimen and the collection container.

The following protocol should be followed:

1. Wash hands thoroughly with soap and water and then dry hands with a clean towel.

2. If the patient is uncircumcised, instruct him to retract the fore skin, holding it back during the entire procedure.

3. If possible, instruct the patient to clean the area around the penis opening (glans penis) by starting at the tip of the penis and cleaning downward (using a towel) and also cleaning directly across the meatus with a different towel.

4. Pass the initial portion of urine into the toilet bowl.

5. Pass some of the remaining urine into the sterile, screw-cap plastic cup provided. Do not touch the inside of the container or lid.

6. Stop collection when container is about half-full.

7. Carefully screw cap on container.

8. Wash hands thoroughly with soap and water, and dry with a clean towel.

9. Give sample to personnel.

ANNEX 6: Curriculum Vita

Philemon Mohammed

+251933807885

philozel734@gmail.com

Addis Ababa, Ethiopia

EDUCATION

October 2023- present Addis Ababa, Ethiopia	**Master of Public Health in Environmental and Occupational health and safety** Addis Ababa University, college of health sciences, school of public health
September 2018- September 2022 Gondar, Ethiopia	**Bachelor of Science in Environmental and Occupational Health and Safety** University of Gondar, college of medicine and health science **GPA**: **3.97**- Very Great Distinction
September 2016- July 2018 Addis Ababa, Ethiopia	**Dessie catholic kidanmehret school**, Preparatory school Grade 11- ranked second Grade 12- ranked third
September 2014- July 2016 Addis Ababa, Ethiopia	High school Top scorer in both Grade 9 and 10 **CGPA 4** of national matric result

RESEARCH

2024 – Present	**Thesis**: Nephrotoxicity Risk among Dry Cleaning Workers Exposed to Perchloroethylene in Addis Ababa

2021 -2022	**Thesis**: Assessment of Knowledge Attitude, and Practice towards waste segregation and utilization of trash bags among Lideta sub-city, Gondar town North West, Ethiopia 2022

CERTIFICATES

November , 2021	**World health organization (WHO)** 1. Basic microbiology 2. Prevention, identification and management of infection in health workers in the context of covid-19 3. Podoconiosis : training of health workers at national and district levels on skin-NTDs
October , 2024	4. Water and Sanitation for Health Facility Improvement tool (WASH FIT) for improved quality of care

ACTIVITIES

2021-2022	**President** of Environmental Health Students' Association at University of Gondar, college of medicine and health sciences. • President of the association • Chaired meetings • Organized blood donation campaigns
2019-2022	**Executive Committee** at Ethiopian Health Professional Students' Association- Gondar branch • Worked as a General program director • Published brochures to help raise awareness in early days of the COVID-19 pandemic • Organized volunteer works
2021/2022	**Students' representative** during community health attachment in Gondar printing enterprise , felege abiot primary school and in east African bottling industry Bahir dar ,Ethiopia • Led the team of internship assigned for the site. • Conducted a risk assessment • Drafted and implemented OSHA management • Provided health education for workers and students.

2022	**Team leader** in team training program in Dembiya Woreda, Kola Deba, North Gondar, Ethiopia • Group leader • Conducted community home visits • Prepared weekly reports to team training program office • Implemented project on maintenance of latrine for The Town • Provided health education on communicable diseases for prisoners.

LICENSE

Health Professional Licensing Certificate	Registered and Licensed as, Junior environmental Health Officer Issued on: April 30, 2023

AWARDS

Educational sponsorship recipient (2024) (Awarded by wollo university)	A fully funded scholarship to study master of Public Health in Environmental and Occupational Health
Certificate of achievement (2022) (Awarded by Gondar university)	I stood first from my batch class of 2022 Gondar university, college of health science students.

ANNEX 7: Curriculum Vita of Primary Advisor

Name: Teferi Abegaz Shifaw (PhD)

Address: Addis Ababa University School of Public Health

E-Mail: Teferi.abegaz@aau.edu.et

Phone: +251 911361607

Nationality: Ethiopian

Education:

- **PhD in Public Health:** Addis Continental Institute of Public Health and Hawassa University September 2011-June 2014.
- **Master's degree** in Public Health: Addis Ababa University, 2005-2007 GC.
- **BSc degree in Environmental Health Sciences**: Jimma University, 2002-2005 GC.
- **Diploma in Sanitary Science:** Jimma Institute of Health Sciences, 1995-1997 GC

Work experience:

- March 2018 till now as an Assistant Professor of Public Health, Addis Ababa University, School of Public Health (Currently MPH Program Coordinator)
- June 2014 GC to February 2018 as an assistant professor of Public Health and Postgraduate coordinator of the School of Public and Environmental Health, Hawassa University.

Key Skills

- Proficient in statistical analysis and software (SPSS, STATA and Epi-Data)
- Manuscript writing, reviewing, and editing
- Monitoring and evaluation of public health programs
- Teaching/mentoring and advising public health students

Research Publications

- Occupational health and safety status of selected public hospitals in Ethiopia. Minister of Health-sponsored project
- Semegnew Takele, Yifokire Tefera, Teferi Abegaz, and Hailemichael Mulugeta. Use of Seatbelts and Observable Factors among Public Transport Drivers in Addis Ababa, Ethiopia. Journal of Advanced Transportation, 2022; Volume 2022 (doi:10.1155/2022/3256727)
- Damen Hailemariam, Abera Kumie*, Samson Wakuma, Yifoker Tefera, Teferi Abegaz,
- Worku Tefera, Wondimu Ayele, Mulugeta Tamire, Shibabaw Yirsaw. Trends in non-pharmaceutical intervention (NPI) related community practice for the prevention of COVID-19 in Addis Ababa, Ethiopia. PLoS ONE, 2021; 16(11): e0259229. (doi:10.1371/journal.pone.0259229)
- Zuriyash Mengistu*, Ahmed Ali, Teferi Abegaz. The pattern of orthopedic fractures and visceral injury in road traffic crash victims, Addis Ababa, Ethiopia. PLoS ONE, 2021; 16(9): e0253690. (doi:10.1371/journal.pone.0253690)
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References

1. Prof. Damen Hailemariam, Dean, School of Public Health, Addis Ababa University hcfp.aau@ethionet.et

2. Prof. Wakgari Deressa, School of Public Health, deressew@gmail.com
3. Prof. Abera Kumie, School of Public Health aberakumie2@yahoo.com

ANNEX 8: Declaration

I, the undersigned declare that this thesis is my original work for in partial fulfillment of the requirement for the degree of Master of Public Health with specialty in Environmental and Occupational health. I also declare that it has never been presented in this or any other university and that all resources and materials used in the thesis have been duly acknowledged.

Name of the student: Philemon Mohammed
Date:
Signature:

Approval of the primary Advisor

Name of the primary advisor: Teferi Abegaz (PhD)
Date:
Signature: