Available online on 15.02.2025 at http://jddtonline.info

Journal of Drug Delivery and Therapeutics

Open Access to Pharmaceutical and Medical Research

Open Access Full Text Article

Research Article

Effect of Short-Term Exposure to Formalin on the Prostate Health of Medical Students in Okofia, Nnewi

Augustine Chinedu Ihim[1]*, Patricia Chisom Obiezekpazu [1], Patrick Chinedu Obi[2], Collins Uchechukwu Obi[1], Romanus Ogai Ogalagu [3], Tochukwu Anthony Ikwelle [1]

[1]. Department of Clinical Chemistry, Nnamdi Azikiwe University, Nnewi Campus, Anambra State, Nigeria.

[2]. Department of Internal Medicine, Federal University Teaching Hospital Owerri, Imo State, Nigeria

[3]. Department of Biochemistry, Tansian University, Umunya, Anambra State, Nigeria

Article Info:

Article History:

Received 13 Nov 2024
Reviewed 20 Dec 2024
Accepted 16 Jan 2025
Published 15 Feb 2025

Cite this article as:

Ihim AC, Obiezekpazu PC, Obi PC, Obi CU, Ogalagu RO, Ikwelle TA, Effect of Short-Term Exposure to Formalin on the Prostate Health of Medical Students in Okofia, Nnewi, Journal of Drug Delivery and Therapeutics. 2025; 15(2):7-12 *DOI: http://dx.doi.org/10.22270/jddt.v15i2.6974*

*Address for Correspondence:

Dr Ihim Augustine Chinedu, Department of Clinical Chemistry, Faculty of Medical Laboratory Science, Nnamdi Azikiwe University, Awka, Nigeria.

Abstract

Medical students exposed to formalin, a preservative used for cadavers, may face health risks during dissection lessons. This study assessed the levels of free prostate-specific antigen (fPSA), total prostate-specific antigen (tPSA), and the percentage of free prostate-specific antigen (%fPSA) in students at Nnamdi Azikiwe University's College of Health Sciences after three hours of exposure to formalin. Forty-five participants aged 18 to 30 were recruited for this study. Baseline samples taken before a 3-hour exposure served as control (pre-exposure), while samples collected afterward were classified as post-exposure (test). The levels of tPSA and fPSA were measured using the Enzyme-Linked Immunosorbent Assay (ELISA) method. For this analysis, the AccuBind Total and Free Prostate-Specific Antigen ELISA kit was utilized.The analysis using a paired samples t-test and Pearson's correlation coefficient revealed a significant decrease in total PSA (tPSA) after formalin exposure, with mean values dropping from 0.88 ± 0.60 ng/ml to 0.67 ± 0.50 ng/ml ($p < 0.05$). However, no significant changes were found in free PSA (fPSA) levels or participants' blood pressure post-exposure ($p > 0.05$). There was no relationship between the Total PSA, Free PSA, % Free PSA and the Body Mass Index (BMI), Systolic and Diastolic Blood Pressure of the participants post-exposure to short-term formalin. It was concluded that a three-hour formalin exposure may at a certain dosage be a therapeutic agent to decrease raised total PSA levels which is sometimes indicative of prostate dysfunction.

Keywords: short-term, formalin, exposure, prostate, health, medical, students, Nnewi

INTRODUCTION

Formalin is a saturated solution of formaldehyde gas in water. It has a tiny quantity of stabilizer and about 10–12% methanol is used as the general stabilizer to prevent formaldehyde polymerization [1] (Cammalleri et al., 2022). According to [2] the other names for formalin include; oxomethane, methylaldehyde, oxymethyline, methanal, and methylene oxide. Formalin is usually sold commercially as water soluble containing 37% formaldehyde. It is miscible in water at 25ºC, soluble in ethanol and chloroform. Formaldehyde is a volatile organic molecule, highly soluble in water. It is a colorless, flammable gas with a pungent odour, at room temperature and ordinary atmospheric pressure [1](Cammalleri et al., 2022). With a molecular weight of 30.03, formaldehyde dissolves in water (400 g/Lat 20°C), ethers, and alcohols [3](Foti et al., 2021). At temperatures above 150°C, formaldehyde metabolises into methanol and carbon monoxide. In ambient settings, formaldehyde is photo-oxidized in sunlight to produce carbon dioxide [4](Asare-Donkor et al., 2019). Silver crystals function as a catalyst during the partial oxidation and dehydrogenation of methanol with air to create formaldehyde, produced at 600°C to 650°C [4] (Asare-Donkor et al., 2019). As a byproduct of oxidative metabolism, formaldehyde exists naturally in biological systems, the environment, some foods, and the bodies of animals, including humans [5] (Yang et al., 2017). Formaldehyde is a well-known occupational carcinogen (this is dependent on length and concentration of exposure) and a recognized sensory irritating chemical, particularly for sensitive persons, that is prevalent in a wide range of working environments and industries such as construction, textile, furniture, medical, chemical, and pharmaceutical industries. As a result of its chemical-physical properties and a broad range of microbicide action, formaldehyde is frequently employed in various manufacturing processes and sanitary applications [1](Cammalleri et al., 2022). The

primary sources of formaldehyde exposure in humans are anthropogenic; some are found in interior surroundings, such as insulating materials, resins, glues, chipboard, plywood, textiles, and so on [6](Nielsen et al., 2017). Additional sources of exposure include operations involving combustion processes, tobacco, electronic cigarettes, cooking, particularly frying, and smoking both actively and passively. Formaldehyde is frequently used in the medical profession for sterilizing purposes in autopsy rooms, pathology or histology departments, as well as a dehydrating agent or preservation (formalin) during the production of mixtures, tissue processing, and staining [7](Corradi et al., 2012). Formaldehyde is dangerous for medical professionals such as nurses, clinicians, medical laboratory scientists and doctors. This is especially true for those working in the healthcare sector, since they may be exposed to formaldehyde and other genotoxic compounds [8](Elshaer and Mahmoud, 2017). Using formaldehyde-containing solutions to cure and preserve biological tissues and prepare cadavers is a common task done in operating rooms, gross anatomy, pathology, and histology labs. Medical schools, research labs, and hospital settings all have an occupational exposure risk due to formaldehyde vapour pollution of the indoor air. Furthermore, research shows that formaldehyde exposure can happen not only while handling formaldehyde and materials treated with it, but also when storing this material or treated items improperly and when a local exhaust ventilation system isn't working well [9](Higashikubo et al., 2017).

The importance of routinely inspecting formalin air-level concentrations to ensure they remain below allowable thresholds is often overlooked in safety protocols. This negligence can be attributed to various factors, including a lack of understanding, poor scheduling, staffing issues, inadequate supervision by management, or simple negligence [10](Kamruzzaman, 2016). Exposure to formalin can have harmful effects on the human body. Common short-term symptoms of formalin poisoning include inflammation of the skin, eyes, and respiratory tract. Long-term effects can be more severe, leading to permanent metaplastic changes in the nasopharynx and oropharynx, potentially resulting in lung or nasal cancer [10](Kamruzzaman, 2016). A study by [8] Elshaer and Mahmoud (2017) focused on mortuary attendants exposed to formalin during embalming procedures. It revealed that exposure affected their eyes, respiratory system, skin, and appetite, causing symptoms such as itchy eyes, eye pain, and excessive tearing.Moreso, a longitudinal study by [11] Ihim et al 2017 on the effect of short term exposure to formalin on male reproductive hormones, recruited medical students exposed for three hours during their cadaver dissections. The result showed a significant decrease in the mean serum testosterone level post formalin exposure compared with pre-formalin exposure in the male participants. The prostate gland, located in the true pelvis, plays a crucial role in the male reproductive system. Its primary function is to produce an alkaline solution that protects sperm from the vagina's acidic environment. By balancing the vaginal acidity, this fluid extends the lifespan of sperm, increasing their chances of successfully fertilizing an egg [12](David and Leslie, 2022). The prostate contains two ejaculatory ducts and the proximal urethra, as well as various tubular structures. After exiting the seminal vesicles, the ejaculatory ducts enter the prostate, running from inferior and medial positions to posterior and lateral ones. They converge in the seminal colliculus, a region of the prostate near the urethra[13] (Singh and Bolla, 2023). Situated just below the bladder and pelvic floor muscles, the prostate is adjacent to the rectum. This proximity allows for the prostate gland to be felt during a Direct Rectal Examination, a procedure used to check for prostate cancer [14](Das et al., 2019). The prostate contributes to the production of semen, which consists of fluid from the prostate, sperm cells from the testicles, seminal vesicle fluid, and secretions from the bulbourethral gland. All these fluids combine in the urethra, and the secretion from the prostate is essential for the healthy functioning of sperm and, consequently, male fertility[15] (Sunder and Leslie, 2020). Prostate-specific antigen (PSA) is found in the thin, milky fluid known as prostatic secretion. PSA is a glycoprotein produced by the prostate gland, the lining of the urethra, and the bulbourethral gland. It is a serine protease enzyme generated by the columnar epithelial cells of the prostatic ducts and acini [16] (Leslie et al., 2019). PSA's role is to break down large proteins in sperm into smaller components. This process reduces the viscosity of semen over time, improving sperm function and fertility [12](David and Leslie, 2022). Typically, the blood secretes only small amounts of PSA. Serum PSA serves as a biomarker and is commonly used to monitor the progression of hormone-naïve prostate cancer after hormonal treatment [16](Prensner et al., 2012). PSA levels in the blood may increase due to benign prostatic hypertrophy, prostatitis, trauma, inflammation, urogenital procedures, biopsies, or prostate cancer-related tissue damage [17](Mediu et al., 2021).There are two types of prostate-specific antigen (PSA) in serum: unbound (free PSA), which makes up 10–30% of total PSA, and complexed PSA, bound to alpha-1-anti-chymotrypsin, comprising 70–90% of total PSA. Assessing the fraction of PSA complexed with alpha-2-macroglobulin requires breaking down the complex to access the PSA epitopes [17](Mediu et al., 2021). Cancer alters tissue architecture, raising levels of complexed PSA (cPSA) and pro-PSA while lowering free PSA (fPSA). Consequently, a low free/total PSA (f/tPSA) ratio has long been used to suggest prostate cancer in individuals with ambiguous PSA results [18](Ferro et al., 2020). The prostate-specific gene kallikrein 3 (KLK3) on chromosome 19q13.4 encodes PSA. Inactive precursors pre-proPSA and pro-PSA undergo cleavages to form mature PSA [19](Prensner et al., 2012). Measuring serum-free PSA levels can improve the specificity of prostate cancer detection in patients with total PSA ≥ 4.0 ng/mL [17](Mediu et al., 2021). [17]Mediu et al. (2021) found that in men with total PSA levels over 4.0 ng/mL, measuring serum-free PSA alongside total PSA enhances diagnostic accuracy and reduces unnecessary biopsies. For total PSA levels between 4 and 10 ng/mL and a negative digital rectal examination (DRE), the %fPSA can

differentiate between prostate cancer and benign prostatic hyperplasia (BPH) [12] (David and Leslie, 2022). To calculate the % free PSA, multiply the free PSA level by 100 and divide by the total PSA level [16](Leslie et al., 2019). A lower free PSA/total PSA ratio can increase prostate cancer diagnosis sensitivity [12](David and Leslie, 2022). This research aims to evaluate the effects of short-term formalin exposure during anatomy classes on the prostate health of medical students at the College of Health Sciences, Okofia Nnewi.

MATERIAL AND METHODS

Study Area

The study was conducted at the Anatomy Laboratory of the Department of Human Anatomy, Faculty of Basic Medical Sciences, College of Health Sciences, Nnamdi Azikiwe University, Okofia, Nnewi Campus, Anambra State.

Study Design

This study was a longitudinal study that was designed to assess the effects of short-term (3 hours) exposure to formalin in the dissection room on prostate health of male medical students who attend dissection classes and have already been exposed in the Anatomy Laboratory in College of Health Sciences, Okofia, Nnewi Campus, Anambra State. A total of 45 students within the age range of 18-30 years were recruited. The baseline samples (before 3 hours exposure) which was considered as control was collected first from the students before exposure to formalin. A questionnaire was prepared and administered to the subjects to ascertain their socio-demographic parameters and medical history.

Sample Size

The sample size was calculated using G*Power software version 3.1.9.4 (Universitat Dusseldorf Germany). Analysis for difference between two paired means (two groups) was conducted in G*Power to determine the sufficient sample size using an alpha of 0.05, a power of 0.85 and an effect size of 0.45. Based on this, the calculated sample size of 45, has a power of 85% to detect difference of 0.45 at a significant level of 0.05. A total sample size of 45 was used for this study.

Inclusion Criteria

Participants within the age range of 18-30 years.Participants that have been exposed to formalin. Participants that attended anatomy dissection class.

Exclusion Criteria

Participants who were sick. Participants who were non-students of College of Health Sciences, Nnewi. Participants that were not interested in being included in the research.

Informed Consent

Informed consent of the participants was obtained prior to the study. Subjects were free to exit from the study without anything affecting them.

Anthropometric Measurements

Weight and height were measured in clothing without shoes and body mass index (BMI) was calculated as BMI= Weight (kg) / Height (m^2). The height was obtained with a measuring tape while the weight was measured using a manual weighing scale. Overweight and generalized obesity was defined as Body Mass Index (BMI) $\geq$ 25 and 30 kg/m^2, respectively.

Blood Pressure Measurements

Systemic blood pressure was obtained using OMRON digital blood pressure monitor on the left arm after a 10 minutes rest using a cuff of appropriate size with the subject comfortably seated. Blood pressure was expressed as Systolic and Diastolic.

Sample Collection and Storage

Five millilitre(5ml) of venous blood samples was collected asceptically by venipuncture from each participant through the antecubital vein using a plastic syringe. It was dispensed in a lithium heparin container. Afterwards, it was centrifuged at 4000rpm for 5 minutes using a table top centrifuge. The plasma was used for the evaluation of Total PSA and Free PSA levels in the sample. The plasma was stored at a temperature of -20ºC.

LABORATORY METHODS

All the reagents were commercially obtained and the manufacturers' standard operating procedures were strictly observed.

free prostate-specific antigen (fPSA), total prostate-specific antigen (tPSA), and the percentage of free prostate-specific antigen (%fPSA) were determined by enzyme-linked immunosorbent assay using ACCUBIND ELISA kits as described by[20] Manafa et al., 2015

Evaluation of Total PSA

Serum Total PSA level was estimated using microplate immunoassay (ELISA sandwich method), colorimetric method.

Evaluation of Free PSA

Serum Free PSA levels was estimated using microplate immunoassay (ELISA sandwich method).

Calculation of %Free PSA

To determine the percentage free PSA, multiply the free PSA level by 100 and divide it by the total PSA number [16](Leslie *et al*., 2019) Free PSA/ Total PSA *100

Statistical Analysis

The statistical analysis was done using Pearson's correlation coefficient test and Paired samples t-test. Statistical Package for Social Sciences (SPSS) version 23.0 was used for the analysis of the results. Data was presented as mean +/- standard deviation. The level of significance was set at $p<0.05$. Values were considered significant at $p<0.05$.

RESULTS

Table 4.1. The mean values of the Total Prostate Specific Antigen (PSA) (ng/ml), Free PSA (ng/ml) and %Free PSA of the participants pre and post exposure to short-term (3 hours) formalin (Mean ± SD).

Variables	Pre-formalin Exposed (n=45)	Post-formalin Exposed (n=45)	t-value	p-value
Total PSA (ng/ml)	0.88 ± 0.60	0.67 ± 0.50	2.770	0.008
Free PSA (ng/ml)	0.17 ± 0.13	0.14 ± 0.09	1.656	0.105
% Free PSA	20.51 ± 14.84	21.04 ± 13.54	-0.224	0.824

**Statistically significant at (p < 0.05).*

There was a significant decrease in the mean values of the serum Total PSA levels of the participants post exposure to short-term formalin (0.67 ± 0.50) (ng/ml) compared with the baseline (0.88 ± 0.60) (ng/ml) (p < 0.05).

However, no significant difference was observed in the mean values of the serum Free PSA levels of the participants post exposure to short-term formalin (0.14 ± 0.09) (ng/ml) compared with the baseline (0.17 ± 0.13) (ng/ml) (p > 0.05).

However, a significant increase was observed in the mean values of the serum % Free PSA levels of the participants post exposure to short-term formalin (21.04 ± 13.54) (ng/ml) compared with the baseline (20.51 ± 14.84) (ng/ml) (p > 0.05).

Table 4.2. The mean values of the Systolic and Diastolic Blood Pressure of the participants pre and post exposure to short-term (3 hours) formalin (Mean ± SD).

Variables	Pre-formalin Exposed (n=45)	Post-formalin Exposed (n=45)	t-value	p-value
Systolic Blood Pressure	126.96 ± 14.22	123.00 ± 18.36	1.705	0.095
Diastolic Blood Pressure	77.91 ± 11.91	77.47 ± 9.36	0.222	0.825

**Statistically significant at (p < 0.05).*

There was no significant difference in the mean values of the Systolic and Diastolic Blood Pressure of the participants pre (126.96 ± 14.22, 77.91 ± 11.91) (mmHg) and post (123.00 ± 18.36, 77.47 ± 9.36) (mmHg) exposure to short-term formalin (p > 0.05).

Table 4.3. Relationship of the levels of Total PSA (ng/ml), Free PSA (ng/ml), % Free PSA with the Body Mass Index (BMI), Systolic and Diastolic Blood Pressure of the participants post exposure to short term (3hours) formalin.

Parameters		BMI (kg/m²)	SBP (mmHg)	DBP (mmHg)
Total PSA (ng/ml)	r	0.004	0.002	-0.085
	p	0.978	0.991	0.580
Free PSA (ng/ml)	r	-0.095	0.060	-0.033
	p	0.537	0.697	0.828
% Free PSA	r	-0.154	0.025	-0.061
	p	0.313	0.868	0.690

**Statistically significant at (p <0.05).*

There was no relationship between Total PSA, Free PSA, % Free PSA, and Body Mass Index (BMI), as well as Systolic and Diastolic Blood Pressure among the participants after short-term exposure to formalin.

DISCUSSION

Medical students are exposed to formaldehyde in the dissection hall making them among those who are at high risk of formaldehyde intoxication due to its poisonous effects[21] (Tiruneh, 2021). According to current molecular principles of cancer, all malignancies come from both heredity and the environment, which means that a combination of internal genetic alterations and various external influences can cause cancer in humans[22] (Wu *et al*., 2018). For this reason, it is crucial to investigate how environmental contaminants like formaldehyde affect prostate health indicators like prostate specific antigen (PSA). In this study, the levels of Total PSA, Free PSA and % Free PSA were evaluated in students who, although already exposed to formalin prior to the research, were exposed to the formalin for a three-hour duration in Nnewi Campus to determine the impact of the three hours exposure on their serum Total PSA, Free PSA and % Free PSA.It was observed that the mean values of serum Total PSA was significantly decreased in the participants after short-term (3 hours) exposure to formalin compared with the baseline ($p<0.05$). This finding is in disagreement with a study by [23]Mohamed El Far *et al*. (2006), where industrial workers were grouped according to the industrial hazard they were exposed to and their serum PSA level was evaluated along with other parameters. It was observed that the serum PSA levels of the workers who had been exposed to formalin was not significantly decreased compared to the control group ($p>0.05$). This could be as a result of the difference in duration of exposure. On the other hand, the mean values of serum Free PSA and % Free PSA did not differ significantly in the participants after short-term (3 hours) exposure to formalin compared to the baseline ($p>0.05$). Although there is limited research on the link between formaldehyde exposure and prostate health, certain studies suggest that exposure to some toxic chemicals can affect serum PSA levels. Therefore, as a result of the somewhat novelty of this research, this study explored the effect of other toxic chemicals on the prostate health. As mentioned earlier, it was observed that the mean value of serum Total PSA was significantly decreased in the participants after formalin exposure compared with the baseline ($p<0.05$). This is in contrast with a study carried out on quarry site workers who had been exposed to quarry pollutants for a long duration. No significant difference in the mean Total PSA, Free PSA and % Free PSA was observed between the exposed group and the unexposed control ($p>0.05$) [24](Ewenighi *et al*., 2016). The disparity in the result could be due to difference in chemical toxins to which the participants were exposed to as well as the duration of exposure to these toxins. However, existing research by [11]has explored the effects of formalin exposure on male reproductive hormones. For instance, a study titled "Effect of Short-Term Exposure to Formalin on Male Reproductive Hormones of Students in Nnewi" found that serum testosterone levels significantly decreased after subjects were exposed to formalin during dissection in a cadaver room, compared to their levels before exposure. Additionally, the "Toxicological Profile for Formaldehyde" by [25] discusses various health effects of formaldehyde exposure, including potential impacts on reproductive health, while these studies provide insights into the effects of formalin/formaldehyde exposure on male reproductive hormones, they do not specifically address changes in total PSA levels following short-term exposure. Further research would be necessary to establish a direct link between formalin exposure and alterations in PSA levels. This study also revealed no significant difference in the mean Systolic and Diastolic blood pressure following short term (3 hours) exposure to formalin ($p>0.05$). This is in concert with a study carried out by[26] Neginhal *et al*. (2013) which also showed no significance in Systolic and Diastolic Blood pressure after acute exposure (2 hours) to formalin. This could be as a result of the disparity in duration of exposure to formalin.

CONCLUSION

It was concluded that after short-term (3 hours) exposure to formalin, there was a notable decrease in total prostate-specific antigen (PSA) compared to the baseline. These findings suggest that at a certain dosage, formalin may be used as a therapeutic agent for the decrease of raised total PSA level which is often an indication of prostate dysfunction.

Conflicts of Interest: The authors declare that they have no conflicts of interest.

Contributors: ACI, PCO, and PCO conceived and designed the research proposal. CUO, ROO, OAI, and ACI performed sample collection, experiments, and data analysis. CUO, ACI, ROO, and OAI contributed to the final version of the manuscript. All authors have read and approved the final manuscript.

Acknowledgments: The authors would like to pay their most profound gratitude to the management and staff of Nnamdi Azikiwe University Teaching Hospital Nnewi, and Reene Medical Diagnostic Laboratory, Awada, Anambra State, for all laboratory analyses of all biochemical parameters

Data availability: The data used to support the findings of this study are available from the corresponding author upon reasonable request.

Funding: No funding sources.

Conflict of interest: None declared.

Ethical approval:

The study sought and obtained ethical approval from the Ethics Committee of the Faculty of Health Sciences and Technology College of Health Sciences Nnamdi Azikiwe University with reference no. FHST/REC/023/566

REFERENCES

1. Cammalleri V, Pocino RN, Marotta D, Protano C, Sinibaldi F, Simonazzi S, Petyx M, Iavicoli S, Vitali M, "Occupational scenarios and exposure assessment to formaldehyde: A systematic review." Indoor air, 2022; *32*(1):12949. DOI: https://doi.org/10.1111/ina.12949
2. Centers for Disease Control and Prevention (CDC). (n.d.). *Formaldehyde*. NIOSH Pocket Guide to Chemical Hazards.

Retrieved January 1, 2025, from https://www.cdc.gov/niosh/npg/npgd0294.html

3. Foti C, Bonamonte D, Romita P, Guarneri F, Patruno C, Angelini G, "Common Allergens: Clinical Contact Dermatitis." A Practical Approach, 2021; 437-497.

4. Asare-Donkor NK, Boakye-Agyemang D, Voegborlo RB, Adimado AA, "Assessment of formaldehyde levels and its health effects in some selected residential homes and offices in Ashanti region, Ghana." Journal of Multidisciplinary Engineering Science Study, 2019; 5(8):2748-2755.

5. Yang M, Ospina M, Tse C, Toth S, Caudill SP, Vesper HW, "Ultraperformance Liquid Chromatography Tandem Mass Spectrometry Method to Determine Formaldehyde Hemoglobin Adducts in Humans as Biomarkers for Formaldehyde Exposure" Chemical Research in Toxicology, 2017; 30(8):1592–1598. DOI: https://doi.org/10.1021/acs.chemrestox.7b00114

6. Nielsen GD, Larsen ST, Wolkoff P, "Re-evaluation of the WHO (2010) formaldehyde indoor air quality guideline for cancer risk assessment." Archives of Toxicology, 2017; *91*(1):35–61. DOI: https://doi.org/10.1007/s00204-016-1733-8

7. Corradi M, Gergelova P, Di Pilato E, Folesani G, Goldoni M, Andreoli R, Selis L, Mutti A, "Effect of exposure to detergents and other chemicals on biomarkers of pulmonary response in exhaled breath from hospital cleaners: a pilot study." International Archives of Occupational and Environmental Health, 2012; *85*(4):389–396. DOI: https://doi.org/10.1007/s00420-011-0686-8

8. Elshaer NSM, Mahmoud MAE, "Toxic effects of formalin-treated cadaver on medical students, staff members, and workers in the Alexandria Faculty of Medicine" Alexandria Journal of Medicine, 2017; *53*(4):337-343. DOI: https://doi.org/10.1016/j.ajme.2016.11.006

9. Higashikubo I, Miyauchi H, Yoshida S, Tanaka S, Matsuoka M, Arito H, Araki A, Shimizu H, Sakurai H, "Assessment of workplace air concentrations of formaldehyde during and before working hours in medical facilities" Industrial Health, 2017; *55*(2):192–198. DOI: https://doi.org/10.2486/indhealth.2016-0147

10. Kamruzzaman M, "Formalin crime in Bangladesh: a case study" European Journal of Clinical and Biomedical Sciences, 2016; *2*(5):39-44. DOI: https://doi.org/10.11648/j.ejcbs.20160205.12

11. Ihim Augustine Chinedu, "Effect of Short-Term Exposure to Formalin on Male Reproductive Hormones of Students in Nnewi." IOSR JDMS (IOSR-JDMS), 2017; 33-36. DOI: https://doi.org/10.9790/0853-1611063336

12. David MK, Leslie SW, Prostate Specific Antigen (PSA). PubMed Available at: https://www.ncbi.nlm.nih.gov/books/NBK557495/ (Accessed May 10, 2024).

13. Singh O, Bolla SR. Anatomy, Abdomen and Pelvis, Prostate. In: StatPearls. Treasure Island (FL): StatPearls Publishing; 2023 Jul 17. Available at: https://pubmed.ncbi.nlm.nih.gov/32644334/

14. Das CJ, Razik A, Sharma S, Verma S, "Prostate biopsy: when and how to perform." Clinical Radiology, 2019; 74(11):853–864. DOI: https://doi.org/10.1016/j.crad.2019.03.016

15. Sunder M, Leslie SW, Semen Analysis. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2020. Available from: https://www.ncbi.nlm.nih.gov/books/NBK564369/

16. Leslie SW, Soon-Sutton TL, Sajjad H, Siref LE, Prostate Cancer. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2019. Available from: https://www.ncbi.nlm.nih.gov/books/NBK470550/

17. Mediu R, Rama A, Mediu N, "Screening for prostate cancer: a study on the free and total prostate-specific antigen." Discoveries (Craiova, Romania), 2021; *9*(4):139. DOI: https://doi.org/10.15190/d.2021.18

18. Ferro M, De Cobelli O, Lucarelli G, Porreca A, Busetto GM, Cantiello F, Damiano R, Autorino R, Musi G, Vartolomei MD, Muto M, Terracciano D, "Beyond PSA: The Role of Prostate Health Index (phi)." International Journal of Molecular Sciences, 2020; 21(4):1184. DOI: https://doi.org/10.3390/ijms21041184

19. Prensner JR, Rubin MA, Wei JT, Chinnaiyan AM, "Beyond PSA: the next generation of prostate cancer biomarkers." Science Translational Medicine, 2012; *4*(127):1273. DOI: https://doi.org/10.1126/scitranslmed.3003180

20. Manafa PO, Ihim AC, Ekwueme CI, Oluboyo AO, Chukwuma G, Chukwuanukwu B, Onyemelukwe A, "Assessment of the risk of prostate cancer in adult smokers in Nnewi, Nigeria using prostate-specific antigen as a biomarker." IJBAMR, 2015; 4(2):172-183.

21. Tiruneh C, "Acute Adverse Effects of Formaldehyde Treated Cadaver on New Innovative Medical Students and Anatomy Staff Members in the Dissection Hall at Wollo University, Northeast Ethiopia." Advances in Medical Education and Practice, 2021; (12):41–47. DOI: https://doi.org/10.2147/amep.s291755

22. Wu ZY, Yang C, Luo J, Deng SL, Wu B, Chen M, "Establishment of reference intervals for serum [−2]proPSA (p2PSA), %p2PSA and prostate health index in healthy men." OncoTargets and Therapy, 2019; 12:6453–6460. DOI: https://doi.org/10.2147/ott.s212340

23. Mohamed El-Far M, El Naggar, Elkhawaga AY, Yahya R, Allam A, Khalifa A, "Carcinoembryonic Antigen, Alpha-fetoprotein, and Prostate-Specific Antigen in the Sera of Industrial Workers Exposed to Phenol, Formaldehyde, Urea, and Mixed Vapors." Inhalation Toxicology, 2006; 18(13):1041–1046. DOI: https://doi.org/10.1080/08958370600904603

24. Ewenighi CO, Dimkpa U, Onyeanusi JC, Babtunde A, Onoh LUM, Onoh GO, Ezeugwu, U, "Prostate-specific antigen and its derivatives in young adults occupationally exposed to quarry pollutants in southeastern Nigeria." Archives of Environmental & Occupational Health, 2016; 72(5):258–263. DOI: https://doi.org/10.1080/19338244.2016.1207593

25. Agency for Toxic Substances and Disease Registry (ATSDR). Toxicological Profile for Formaldehyde. Atlanta (GA): U.S. Department of Health and Human Services, Public Health Service; 2022. Available at: https://www.ncbi.nlm.nih.gov/books/NBK597627/

26. Neginhal R, Herur A, Chinagudi S, Rairam G, Kolagi S, Ambi U, "Cardiorespiratory effects of acute exposure to formaldehyde in gross anatomy laboratory in medical students- A comparative study." Medica Innovatica, 2013; 2(1):32–35.