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Dr S K Gupta

E-mail: medicolegalupdate@gmail.com

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Narcotics and Juvenile Delinquency: Exploring The Role of Drugs in The Formation of Youth Criminals in India

Tharanidaran I

II M.Sc. Criminology and Criminal Justice Department of Political Science School of Social Sciences Madurai Kamaraj University, Madurai Tamil Nadu, India

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Abstract

The alarming rise in Juvenile Delinquency in India has become a significant concern for policymakers, legal authorities, and social institutions. Among the various contributing factors, narcotic drug abuse has emerged as a critical yet underexplored dimension influencing the behavioral and criminal trajectories of youth. This study investigates the correlation between narcotics consumption and the development of delinquent tendencies among juveniles in India. Employing a multidisciplinary framework that integrates criminology, psychology, and legal analysis, the research examines socio-economic, familial, and psychological conditions that predispose adolescents to drug use and subsequent criminal behavior. The study also evaluates the effectiveness of existing legal mechanisms, including the Juvenile Justice (Care and Protection of Children) Act, 2015, and the Narcotic Drugs and Psychotropic Substances (NDPS) Act, 1985, in addressing the nexus between substance abuse and juvenile crime. The findings underscore the necessity of comprehensive intervention strategies encompassing education, community support, and mental health care to mitigate narcotic-induced delinquency. This research contributes to the broader discourse on juvenile justice by offering actionable insights for legal reform, public policy, and youth rehabilitation in India.

Keywords: Juvenile Delinquency, Narcotics, Drug Abuse, NDPS Act, Juvenile Justice Act

Introduction

Juvenile delinquency has long been a subject of concern for lawmakers, social workers, and educators across the globe. In India, the issue has gained increasing urgency due to the growing number of crimes committed by minors. Among the many contributing factors to this troubling trend, the influence of narcotic substances stands out as both a cause and a consequence of youth criminal behavior. The easy availability of drugs, combined with peer pressure, broken families, lack of guidance, and socio-economic instability, creates

an environment where young individuals are more likely to engage in deviant acts¹.

India's geographical position, with its proximity to the Golden Crescent and the Golden Triangle two of the world's major drug-producing regions further complicates the matter. Narcotics often find their way into urban slums, rural pockets, and even educational institutions, where adolescents are exposed to these substances at an alarmingly young age. The initial use may begin as experimentation but soon spirals into addiction, often pushing juveniles into theft, assault, trafficking, and even violent crimes

to sustain their habits. This paper delves into the critical intersection between drug abuse and juvenile delinquency in India. It seeks to understand how narcotics not only impair judgment and self-control but also entangle young individuals in criminal networks. Through an analysis of recent crime trends, socio-legal frameworks, and case studies, this study aims to unravel the deeper roots of youth crime influenced by drugs and highlight the urgent need for holistic interventions².

Objectives

- To identify the socio-economic, psychological, and environmental factors that lead juveniles to substance abuse.
- To examine how drug addiction influences the involvement of youth in criminal activities.
- To study the role of drug trafficking networks in recruiting and exploiting vulnerable juveniles.
- To explore the preventive and rehabilitative measures adopted by government and non-governmental organizations.
- To propose evidence-based recommendations for reducing drug influence and delinquency among Indian youth.

Methodology

This study adopts a qualitative method and descriptive research design to analyze Narcotics and Juvenile Delinquency. It examines existing Reports, Case studies, Legal and Policy, Policy Recommendations. The research relies on secondary data sources to provide a comprehensive overview of the issue.

Scope of Narcotics and Juvenile Delinquency

The increasing prevalence of juvenile involvement in narcotic abuse in India is a deeply disturbing trend with far-reaching social, psychological, and criminal implications. Juvenile drug abuse is not merely a health issue it is intrinsically linked with the rising tide of juvenile delinquency, where addiction

often serves as a gateway to criminal behaviour. The scope of juvenile drug abuse in India today reflects a growing crisis that threatens the nation's youth, security, and developmental progress.

India is home to the world's largest youth population, with more than 253 million adolescents aged 10-19 years, making up a significant portion of the country's demographic dividend. However, this population is increasingly exposed to vulnerabilities such as poverty, peer pressure, broken family systems, and urban stress, making them susceptible to narcotics. Recent studies and reports by the National Crime Records Bureau (NCRB) and Narcotics Control Bureau (NCB) have shown that a growing number of juveniles are being caught not only consuming drugs but also engaging in trafficking, peddling, and other crimes under the influence of substance abuse. This trend is particularly alarming in urban slums, border areas, and transit cities like Mumbai, Delhi, Chennai, and Amritsar. The types of substances abused by juveniles range from soft drugs like marijuana and prescription opioids to hard drugs such as heroin, cocaine, and methamphetamines. Inhalants such as correction fluids and industrial adhesives are also commonly misused, especially among street children and slum dwellers due to their easy availability and low cost. The age group most affected appears to be between 14 and 18 years. This age bracket is marked by intense psychological, emotional, and social transformation. Adolescents experimenting with identity and social belonging are especially vulnerable to the influence of deviant peer groups³.

The scope of juvenile drug abuse also extends to educational institutions. Several studies, including those by the Ministry of Social Justice and Empowerment and All India Institute of Medical Sciences (AIIMS), have documented the growing incidence of substance abuse among school and college students. Hostels, coaching centres, and private accommodations in major educational cities like Kota, Bengaluru, and Hyderabad are increasingly becoming breeding grounds for drug culture. This is often exacerbated by easy online access to drugs, the glamorization of substance uses in media, and

the absence of effective counselling or mental health support within institutions⁴.

Causes of Drug Abuse among Juveniles

Drug abuse among juveniles in India is a multifaceted issue influenced by psychological, social, economic, and environmental factors. Adolescents, due to their developmental stage, are emotionally vulnerable and often face identity confusion, peer pressure, and exposure to external influences. These factors combine to push them toward experimenting with, and potentially becoming dependent on, narcotics paving the way for juvenile delinquency. Peer pressure is one of the most significant contributors. The desire for social acceptance during adolescence often compels juveniles to conform to group norms, even if it involves drug use. In settings where drug use is seen as trendy or rebellious, refusing can result in ridicule or exclusion, driving many into substance use. The family environment also plays a critical role. Children from broken homes, abusive households, or those with parents who abuse substances are more vulnerable.

Psychological distress is another core cause. Many juveniles deal with anxiety, depression, trauma, or low self-worth, which can drive them to self-medicate with drugs. In India, where mental health support for youth is inadequate and stigma is prevalent, many adolescents go untreated, increasing their vulnerability.

Media and internet influence has grown significantly in recent years. Exposure to films, music videos, and influencers that glamorize drug use can desensitize juveniles to its dangers. Moreover, the internet provides easy access to information about substances and where to acquire them, making drugs more accessible than ever before. A lack of drug education and counseling in schools is another major concern. Most Indian schools do not have dedicated programs to educate students about substance abuse. Easy availability of drugs, especially in border and urban areas, also contributes to rising juvenile drug abuse. Moreover, lack of recreational facilities and youth engagement programs leaves

adolescents with idle time and little positive engagement. This can lead to experimentation with drugs out of boredom or curiosity⁵.

Consequences: From Addiction to Crime

Drug abuse among juveniles in India has far-reaching consequences, extending beyond physical health deterioration and social isolation. The use of narcotics among youth disrupts mental stability, education, family life, and eventually leads many down a path of criminal behavior. Addiction significantly alters adolescent behavior and decision-making. Young individuals experience emotional instability, poor impulse control, and reduced judgment. Academic disengagement is typically the first observable consequence students skip school, perform poorly, and may drop out. This educational disruption diminishes future job prospects and increases the chances of deviant lifestyle adoption.

Drug peddling is a frequent result, particularly in lower-income communities. Traffickers recruit juveniles to distribute drugs in schools and neighborhoods, offering narcotics or money in return. Exploiting the legal protections afforded under the Juvenile Justice Act, such networks groom youth into deeper criminal participation, making recovery increasingly difficult. Gang involvement and violence also stem from addiction. In high-risk neighborhoods, gangs offer protection, money, or a sense of belonging especially to drug-using youth. These gangs often normalize drug use and crime, immersing juveniles in environments where unlawful behavior becomes routine. The psychological toll is equally severe. Drug abuse increases risks of depression, anxiety, suicidal thoughts, and psychosis. Many commit crimes during drug-induced mental episodes or emotional distress. As addiction worsens, it also damages family relationships, often pushing juveniles out of their homes and into the streets making them easy prey for traffickers or criminal groups⁶.

India's lack of adequate rehabilitation facilities and child-friendly support systems exacerbates these issues. De-addiction centers are scarce, especially in rural areas. Stigma, ignorance, and

insufficient public resources mean that early interventions are rare, and juveniles often cycle through the justice system without proper reform or care. Legally, although juvenile offenders are supposed to be treated with reformatory approaches, in practice they often face social exclusion. Labeled as delinquents, many are denied education and employment, leaving crime as the only path forward. Alarming, drug abuse is also linked to sexual exploitation.

Drug abuse among young people significantly elevates their risk of both physical health deterioration and infectious diseases. Substance misuse impairs immune function— affecting natural killer cells, T-cells, macrophages, and cytokine response—making users more vulnerable to infections like HIV, hepatitis B and C, and other opportunistic illnesses. Intravenous drug use, in particular, is a critical pathway for transmission: sharing needles or injection equipment dramatically increases the risk for blood-borne infections, including HIV, HBV, and HCV. Beyond direct transmission, drug-induced intoxication often compromises judgment, increasing engagement in high-risk sexual behaviors that further elevate infection risk.

Moreover, prolonged substance abuse imposes additional burdens on vital organ systems and overall health. Long-term use is linked to cardiovascular disease, respiratory illnesses, cognitive impairment, organ damage, and heightened susceptibility to infections beyond those directly tied to drug use. These health consequences are especially concerning for young individuals, whose developing bodies and immune systems are more susceptible to both immediate and long-term damage⁷.

Why Traffickers Target Juveniles

Juveniles are attractive to drug traffickers for several reasons. First and foremost, under Indian law specifically the Juvenile Justice (Care and Protection of Children) Act, 2015 minors are generally treated more leniently than adults, even for serious offenses. This legal shield creates an opportunity for traffickers to use children as pawns, knowing they

are less likely to face severe penalties. Moreover, the perception that juveniles evoke less suspicion from law enforcement makes them ideal couriers and street-level dealers.

Many juveniles who are recruited into the drug trade come from vulnerable environments poverty-stricken families, dysfunctional households, or conflict-prone regions. For children growing up in slums, border areas, or communities with high unemployment and limited educational opportunities, drug networks often provide what appears to be a viable escape when compared to the children coming from high economic status. The promise of quick money, protection, status, or a sense of belonging draws them in⁸.

Types of Exploitation Involved

The exploitation of juveniles in drug trafficking goes beyond just using them as couriers or dealers. Many are forced into dangerous environments, exposed to violence, and denied access to education and healthcare. They may be used as “mules” to carry drugs across borders or state lines, often hiding substances in their bodies or belongings placing their lives at physical risk. Some girls recruited into drug networks face an additional layer of exploitation, including sexual abuse or being forced into sex work as part of their entrapment. In such cases, the lines between drug trafficking and human trafficking blur, further complicating rescue and rehabilitation efforts⁹.

Legal Framework: The Juvenile Justice Act and NDPS Act

Addressing the issue of juvenile drug abuse and its link to delinquency requires a strong legal foundation that both prevents such behavior and rehabilitates the affected youth. In India, two major legislations address the problems of narcotics and juvenile crime the Juvenile Justice (Care and Protection of Children) Act, 2015 (JJ Act) and the Narcotic Drugs and Psychotropic Substances Act, 1985 (NDPS Act). Together, these laws attempt to strike a balance between penal action and rehabilitation, particularly for minors involved in drug-related offenses. The

JJ Act, 2015 is the central law that governs children in conflict with the law and children in need of care and protection. It acknowledges that juvenile's individuals below 18 years are in a formative phase of emotional and psychological development. Hence, their involvement in crimes, especially due to addiction, social pressure, or poverty, should be treated through a reformatory rather than purely punitive lens. Under this Act, children caught consuming or even peddling narcotics are classified as Children in Conflict with the Law. Their cases are brought before Juvenile Justice Boards (JJBs) instead of regular courts. These boards include a judicial magistrate and two social workers (one of whom must be a woman), ensuring a supportive and sensitive environment for hearing cases. The JJB evaluates the child's mental state, background, and circumstances to decide on suitable interventions.

A notable feature of the JJ Act is that juveniles found guilty of minor or first-time offenses, such as drug use or small possession, are not sent to jail. Instead, they are referred to rehabilitation centers, counseling, or community service, depending on the case. The law encourages the development of individual care plans, psychological assessments, and vocational training to promote reintegration into society. Since many juvenile drug users are also victims of addiction, this approach emphasizes medical and emotional support over imprisonment. However, a controversial clause in the JJ Act allows children aged 16 to 18 years, involved in heinous crimes, to be tried as adults after a proper assessment by the JJB. This may include serious drug trafficking offenses linked to organized crime. This provision aims to prevent the misuse of legal protection by repeat or violent offenders, but critics argue it could undermine rehabilitation opportunities for children in complex circumstances¹⁰.

The NDPS Act, 1985, is India's primary law for controlling and regulating narcotic drugs and psychotropic substances. It criminalizes the manufacture, possession, sale, and use of banned substances and enforces strict penalties, particularly for large-scale operations. However, when juveniles are involved, the NDPS Act is applied in conjunction

with the JJ Act. This means a child under 18 found in possession of drugs or involved in trafficking is not immediately subjected to the harsh penal provisions of the NDPS Act. Instead, their case is brought before the JJB to determine a rehabilitative course of action. However, this protection is limited in serious or repeated offenses. A crucial addition to the NDPS Act was Section 64A, introduced through an amendment in 2001. This section offers immunity from prosecution to individuals, including juveniles, who voluntarily seek treatment for addiction.

Prevention and Rehabilitation Measures

The rising nexus between narcotics and juvenile delinquency in India is a matter of grave concern. As the youth increasingly fall prey to drug addiction, there is an urgent need to not only curb access to narcotics but also implement holistic prevention and rehabilitation strategies. Tackling the problem requires a multipronged approach involving government institutions, civil society, educational systems, healthcare services, and community participation. Prevention is the first line of defense, while effective rehabilitation ensures that young offenders are reintegrated into society as productive individuals rather than being stigmatized or criminalized.

Early detection of emotional distress, peer pressure, or isolation can allow for timely intervention. Schools, on the other hand, must integrate substance abuse awareness programs into their curricula. Life skills education that includes decision-making, conflict resolution, and emotional intelligence helps students build resilience against harmful influences. Many students are either misinformed or unaware of the long-term physiological, psychological, and legal consequences of drug use. Regular workshops, seminars, and interactive sessions conducted by trained professionals, psychologists, and recovering addicts can help dispel myths and create awareness. Peer mentoring systems can also be introduced where older or trained students counsel juniors, making the message more relatable and credible.

Community engagement and outreach programs are equally essential. Local NGOs, youth clubs, religious organizations, and community leaders can create awareness campaigns, promote healthy lifestyles, and organize recreational activities that offer alternatives to drug experimentation. Sports events, arts, music, and volunteerism not only keep youth engaged but also instill discipline and purpose¹¹.

Psychological counseling and therapy are central components of rehabilitation. Cognitive Behavioral Therapy (CBT), group therapy, family therapy, and motivational interviewing have proven effective in treating addiction. These services should be accessible within juvenile homes, observation centers, and even schools. Family involvement in the recovery process is also crucial. When families participate in counseling, they better understand the child's struggle and are more equipped to support long-term recovery. Vocational training and skill development play a vital role in rehabilitation. Many juveniles engage in drug use and crime due to a lack of opportunities or financial hardship.

By equipping them with practical skills—such as tailoring, carpentry, graphic design, or culinary arts—they gain self-confidence and a sense of purpose. Government schemes like the Skill India Mission should be linked with juvenile justice institutions to promote economic reintegration. Post-rehabilitation support systems are equally important. Without continued guidance, counseling, and employment support, there is a risk of relapse. Juveniles returning to the same toxic environments or peer groups may fall back into substance use and criminal behavior. Therefore, structured after-care programs, halfway homes, and mentorship schemes must be in place. Probation officers, child welfare committees, and NGOs must collaborate to monitor progress and provide emotional and social support¹².

Challenges in Controlling Drug-Related Juvenile Delinquency

Controlling drug-related juvenile delinquency in India presents numerous challenges, rooted in legal, social, institutional, and infrastructural limitations. Despite the existence of robust legislation such as

the Juvenile Justice (Care and Protection of Children) Act, 2015 and the Narcotic Drugs and Psychotropic Substances Act, 1985, the practical implementation of these laws often falls short. One of the primary challenges lies in the early identification of at-risk youth. Many children and adolescents begin drug use due to peer pressure, broken family structures, exposure to crime, or lack of proper guidance. However, the absence of effective monitoring in schools, communities, and families often allows this behavior to go undetected until it escalates into criminal activity.

Lack of awareness and stigma further complicate the issue. Both juveniles and their families often hesitate to seek help due to the fear of social rejection or legal consequences. Drug addiction is still seen more as a moral failing than a medical or psychological issue, leading to isolation rather than support. This stigma discourages open conversations and delays access to treatment and rehabilitation services. Moreover, the shortage of juvenile-specific de-addiction centers and mental health professionals means that even when help is sought, it may not be available or suitable for adolescents. Many existing centers cater to adults and lack the sensitive, age-appropriate care that juvenile patients require. Another significant hurdle is the inadequate training of law enforcement personnel and juvenile justice stakeholders. Police officers, probation officers, and even juvenile justice board members sometimes lack the specialized knowledge and empathy needed to handle drug-involved juveniles appropriately. As a result, many children are subjected to criminal trials, detention, or harsh treatment instead of being guided through a reformatory and rehabilitative path. Cases have been documented where juveniles are charged under the stringent provisions of the NDPS Act without considering the more reform-oriented protections of the JJ Act¹³.

The expansion of organized drug networks also poses a major threat. Drug traffickers often exploit vulnerable minors to transport or sell narcotics, knowing that juveniles are likely to receive lighter legal penalties. These children are easily manipulated due to financial needs or emotional instability, and

once caught in this web, find it extremely difficult to escape. Law enforcement agencies struggle to dismantle such networks due to jurisdictional issues, lack of intelligence, and limited technological resources.

Case Studies

One notable case comes from Delhi in 2022, where a 16-year-old boy was arrested for his involvement in a series of mobile snatching incidents. Upon investigation, it was found that the juvenile was addicted to synthetic drugs, including MDMA and LSD, which he sourced through social media platforms and local peddlers. The boy confessed that his criminal activities were aimed at financing his addiction. His parents, both daily wage laborers, were unaware of his drug use and assumed his behavior changes were due to adolescence. The Juvenile Justice Board referred him to a rehabilitation center after determining he had no prior criminal record. This case highlights how substance abuse can push juveniles toward property crimes to sustain their drug habits and shows how social media has become a tool for drug procurement.

Another case involved a group of three minors in Mumbai in 2021, aged between 15 and 17, who were caught trafficking cannabis and tramadol tablets. They were acting as couriers for a local drug syndicate operating near railway stations and bus terminals. The syndicate had intentionally recruited school dropouts from low-income families, exploiting their need for money and lack of awareness about the legal consequences. The Juvenile Justice Board, after a detailed social background report, found that the boys had been victims of exploitation and recommended rehabilitation rather than punishment. This case underlines how juveniles are often used as pawns in larger drug operations, with organized criminals targeting the most vulnerable for high-risk tasks.

In Punjab's border areas, a region plagued by drug smuggling due to its proximity to international borders, several reports have emerged of juveniles acting as "mules" to transport heroin and other substances across districts. One such case involved

a 17-year-old boy from Amritsar who was caught with heroin worth ₹10 lakh. Interrogation revealed that he had been recruited by a known smuggler in the village, and was promised a motorcycle in return for completing the job. With little understanding of the legal risk and desperate for social validation, he agreed. Unfortunately, in this case, the Juvenile Justice Board recommended that he be tried as an adult under the heinous offences clause of the JJ Act due to the commercial value of the drugs. This case showcases the tension between reformatory and punitive measures in situations involving serious drug crimes.

Another tragic case from Bengaluru in 2020 involved a 14-year-old boy who died from a drug overdose in a government school toilet. The post-mortem revealed traces of inhalants and sedatives. A subsequent investigation exposed a local paan shop selling narcotic-laced candies and cough syrups to students. The child's parents had approached school authorities earlier about their son's strange behavior, but no action was taken. This case drew attention to the lack of vigilance in educational institutions and the ease with which minors can access harmful substances, even in controlled environments.

Discussion

The intersection of narcotics and juvenile delinquency is a pressing concern in India's criminal justice and public health landscape. With a growing number of youth falling victim to drug addiction, the consequences are visible not only in deteriorating mental and physical health but also in increased involvement in criminal activities. Adolescents often face peer pressure, emotional instability, family neglect, academic stress, and exposure to toxic environments, making them easy targets for drug use. In many cases, drug consumption begins as an experiment or an escape mechanism, but it gradually leads to dependence. As addiction progresses, it often pushes juveniles toward illegal activities such as theft, drug peddling, assault, or even organized crime to fund their substance use. This establishes a clear link between drug abuse and criminal behavior

among youth. The normalization of drug use in some social circles and the influence of media also contribute to increased narcotic consumption. Online platforms often glamorize substance use, making it appear trendy or rebellious.

From a socio-economic perspective, poverty, unemployment, and lack of educational or recreational facilities are major risk factors. In urban slums and rural areas, where access to quality education and structured support systems is limited, children often grow up in environments marked by crime, addiction, and neglect. Drug mafias and traffickers exploit this vulnerability by recruiting minors for drug transportation and sale, knowing the legal system treats juveniles with leniency. This exploitation transforms young victims into offenders, perpetuating a dangerous cycle of crime and substance abuse. While the Juvenile Justice (Care and Protection of Children) Act, 2015 and the Narcotic Drugs and Psychotropic Substances Act, 1985 provide the legislative framework, there is often a lack of clarity in implementation¹⁴.

Juveniles involved in drug cases may be treated either as victims in need of rehabilitation or as offenders deserving punishment, depending on the discretion of authorities. Another major gap is the lack of specialized rehabilitation facilities for juveniles. Most de-addiction centers are designed for adults and are ill-equipped to handle the unique psychological and developmental needs of adolescents. The scarcity of vocational training, aftercare services, and community reintegration programs further exacerbates the problem. Moreover, societal stigma associated with addiction and criminal records discourages families and communities from seeking help.

Drug abuse among juveniles in India is not merely a legal issue but a complex socio-psychological and economic crisis. Effective intervention requires a shift from punitive measures to preventive, rehabilitative, and inclusive strategies. Early identification of at-risk youth, school and community-based awareness programs, trained counselors, and youth-friendly de-addiction facilities are essential. Moreover, a coordinated effort among

law enforcement, healthcare, education, and child welfare agencies is vital to break the cycle of drugs and delinquency. Only then can India truly address the roots of this alarming issue and secure a healthier, safer future for its youth¹⁵.

Conclusion

The growing link between narcotics and juvenile delinquency in India is a critical issue that demands immediate and sustained attention. Drug abuse among adolescents is not merely an individual behavioral issue but a broader social. Adolescents, being in a crucial stage of psychological development, are highly impressionable. The use of narcotics for emotional escape or social acceptance can impair their judgment and increase the chances of becoming involved in criminal activities like theft, drug peddling, or gang violence.

Despite having legal frameworks such as the Juvenile Justice Act and the NDPS Act, India struggles with effective implementation. Juveniles are often treated punitively rather than rehabilitative, and the lack of dedicated counseling, de-addiction centers, and reintegration programs hampers long-term recovery. Addressing this issue requires a multi-dimensional approach, involving early education, better mental health services, community-based interventions, and legal reforms that focus on rehabilitation over punishment. Collaboration between schools, law enforcement, NGOs, and families is crucial.

Recommendations

While existing laws like the Juvenile Justice Act (2015) and the Narcotic Drugs and Psychotropic Substances Act (1985) provide a foundational framework, the rising involvement of youth in drug-related crimes suggests the need for a more integrated and proactive approach. The following recommendations aim to strengthen prevention, intervention, enforcement, and rehabilitation mechanisms:

- Comprehensive Drug Education in Schools
- Early Intervention through School and Community-Based Programs

- Strengthening Family Support Systems
- Legal Reforms and Enforcement Sensitization
- Improving Access to De-Addiction and Rehabilitation Centers
- Socio-Economic Development Programs
- Strengthening Coordination Between Stakeholders
- Youth Engagement Through Constructive Platforms

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Psychological Autopsy: A Tool for Resolving Equivocal Deaths

Sindhu Sudha Sahu

Associate Professor Department of Forensic Medicine and Toxicology, All India Institute of Medical Sciences, Bhubaneswar, Odisha

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Abstract

In equivocal deaths especially in cases of suicide where the reason for suicide is not evident, and in cases where manner of death is doubtful as in accidental fall or homicidal fall, or suicidal or accidental hanging, a retrospective & thorough investigation about the victims Intention, State of mind and Individual's actions before death called Psychological Autopsy, provides a significant clue to the investigating team. The further investigation through psychological autopsy requires a detailed interviewing of the close family members, relatives, friends and colleagues or acquaintances of the deceased regarding the various contributing factors which may get revealed subsequently which can help in delineating the manner and reason for the instant death.

This article brings about the objective of psychological autopsy, the interviewing areas which need to be explored while conducting this procedure, its legal acceptance and limitations. Also discussing here various cases where psychological autopsy supported the investigative team and forensic experts to appease the queries in the mind of the relatives, friends, and family members to the sudden unexpected loss of their dear ones.

Keywords: Psychological Autopsy; Equivocal deaths; Manner; Suicide; fall; Forensic medicine

Introduction

Analysis in the case of Equivocal deaths especially in Suicide cases where the reason for suicide is not evident, and in cases where manner of death is doubtful as in accidental fall or homicidal fall, a retrospective & thorough investigation about the victims Intention, State of mind and Individual's actions before death provides a significant clue. This process of evaluation is Psychological Autopsy¹. The various types of psychological autopsy are: a) Suicide Psychological autopsy- Psychology of the victim prior to the death in case of suicide is evaluated. B) Equivocal Psychological autopsy- where actual cause

or manner of death is evaluated. C) Social media psychological autopsy- evaluation associated with the social mobile and media^{2,3}.

Objectives

The main objectives of psychological autopsy are:

- Circumstances surrounding the death
- To arrive at a conclusion as to the manner of death
- Contribute to the research on suicide.
- Refine suicide assessment and prevention techniques reducing the number of suicides.

Procedure

- This involves extensive interviews and relevant document and record collection. The interview is done in following steps
- Face to face interview: Done right in front without any interface on Family members, friends, relatives, next of kin.
- Health personal interview: All the personnel who were associated with the victim's health are interviewed
- Last contact personal interview: Person who was last in contact with the victim prior to his/her death.

There are various criteria's for the interview:

1958: Shneidman's 14 criteria to review when conducting a Psychological autopsy.

1980: Centers for Disease Control, List of 22 criteria k/a Operational Criteria for the Determination of Suicide (OCDS).

1991: Suicide researcher's Empirical Criteria for the Determination of Suicide (ECDS) 16 criteria.

2002: Department of Defense (DoD), Proposed model for psychological autopsies, including a model curriculum, report format, and peer review.

2007: Standard protocol -Expert consensus as well as evidenced-based research and clinical findings⁴.

Psychological autopsy protocol considers areas as

- Emotional reactivity: History of violence toward others, Impulsive behaviors, Excessive rage or aggression.
- Physical health: Recent visit to physician (reasons), Chronic pain, Chronic, fatal, or debilitating disease, Recent reduction in physical/functional capabilities, Current medications (compliance, recent changes).
- Family history: Suicide or attempted suicide, Non-natural deaths, Level of support in family, Physical, sexual, or emotional abuse, Substance abuse, Affective or other psychiatric disorders.

- Substance /alcohol abuse: History and pattern of alcohol, drug abuse, Recent attempts to discontinue use, Recent increase in pattern of use, History of "accidental overdose" (when, type of drug).
- Social support: Ability to create and maintain close personal relationships, Relative success in personal relationships, getting support from family, Attachment to hobbies, interests, religion, Recent talk about feeling unsupported, uncared for, and unimportant.
- Personality and life style: Typical coping patterns, pattern of reaction to stress, Self-destructive behaviors (e.g., self-mutilation, deliberate self-harm, driving while intoxicated), Victimization behaviors (bullied, abused)
- Previous suicidal attempts: methods use, reason for those attempts.
- Factors associated with suicide risk reduction: Evidence of future-oriented thinking or behaviors, Absence of suicidal ideas or intent, Hopefulness, Willingness to accept help and/or treatment for psychiatric conditions, Good therapeutic alliance with mental health professional and Stable, supportive marriage or spouse.
- Psychiatric history: Prior suicidal behaviors, Prescribed psychiatric medications, Compliance with psychiatric medications, Psychiatric hospitalization (reasons, dates, diagnosis, treatment), Outpatient treatment (psychiatrist, psychologist, therapist).
- Precipitants: Significant losses (relationships, job, finances, prestige, family member, etc., Traumatic events, Family member or loved one completed or attempted suicide, Anniversary of important death or loss, Expressed wish to reunite with a deceased loved one or to be "reborn".
- Symptoms and behaviour: Appeared Depressed, sad or moody
High-risk depressive symptoms- insomnia, wt. loss, appetite loss, feeling of worthlessness,

Expressed suicidal ideation, agitated, behaved in an impulsive manner, Appears confused, disoriented, or psychotic, Engaged in excessive risk-taking behaviors.

- Demographics: Age/gender/race/height/weight, Educational status, Socioeconomic status, Employment status, Financial status, Marital status, Adopted versus biological family status, Immigrant status—acculturation issues.
- Death scene: Photos of Death scene and site visit, Decedent's relationship to site, Evidence of planning and/or rehearsal.
- Records and documents: Medical records, Mental health records, Police records, Legal records, Criminal records.
- Other areas: Personal interests, hobbies, Gambling history, Description of activities/behaviors in last days before death
- Collateral interviews-Relationship to deceased, Time interval between death and interview, Reactions to the death (surprise, acceptance, beliefs), Attitudes about suicide

With the rampant use of social mobile and media a subdiscipline of Psychological Autopsy (PA) is Social media Psychological Autopsy, with analyzing messages exchanged on social networks - Forensic investigation on suicide. The reconstruction of conversations or posts published - assess the psychological context of the victim^{5,6}.

The psychological autopsy plays a major role to determine Cause of death, Manner of death and also determine the Motive- Try to understand the reasons or events that prompted the individual to act, If suppose, manner of death - suicide, why the decedent committed suicide?, the Intent - Represents the resolve of an individual in carrying out consciously or unconsciously their death, how and to whom people communicated their suicidal ideations behavior, and also the Lethality - Probability/risk-taking behavior that an individual successfully kills themselves, Degree of lethality into high, medium, low, and absent⁷.

Case Discussion

- **Case 1:** A 30 years woman was brought dead to the casualty of a tertiary care centre with history of fall from 15th floor of her apartment, subsequently the body was sent for postmortem examination with a suspicion regarding manner of death i.e., whether it is a case of accidental/ suicidal/ homicidal fall?. On Post mortem examination, deceased was of average built, fair complexion with facial hair. Rigor mortis fully developed and post-mortem lividity fixed at the back. The external injuries were open fractures at lower ends of both femur and tibia bilaterally, multiple grazed abrasions at front of abdomen, directed from right side abdomen starting medially, passes obliquely upwards to left lateral aspect of abdomen. Two linear, parallelly placed hypopigmented old scars over flexor aspect of left wrist. On internal dissection, there were no fracture of the skull, two and half litres of fluid blood in peritoneal cavity with extensive liver laceration, base of heart was contused, bilateral lungs were collapsed with multiple punctures on anterior surface. There was complete transection of vertebrae & spinal cord at level between T2 & T3. Genital and other organs were intact without any abnormality. A crime scene visit was also done with the investigative team. The postmortem findings and the crime scene confirmed the case to be a fall from height.

A psychological autopsy evaluation was done to obtain the deceased profile, which revealed she was highly educated, and also was a working professional, belonging to a family of high Socioeconomic status, she was married, and then separated since last 5 years as her in-laws didn't accept her. When we interview her brothers with whom she was staying since separation revealed that many a times she had talked of feeling of worthlessness, hopelessness and had expressed her thoughts of committing suicide and also had left the job. Also, she had tried to commit suicide by cutting the wrist hand one year ago, they had also consulted

to psychiatrist and was on medication since then. On the day prior to her death, she had appeared in interview, where she was rejected. On interviewing her friends and colleagues, she was bullied in school when she was 16 years of age and was diagnosed as a case of Schizophrenia and later with Bipolar disorder. She was not regular in her medications and there was no primary caregiver at home to look after her condition, also she had lost her mother two years back. This psychological autopsy revealed the mental illness of the deceased she was suffering from and the mental condition just prior to the event, the triggering factors which could delineate the reason and manner of her death, confirming it to be a case of suicide⁸.

- **Case 2:** A 13 year old boy, was brought dead to the mortuary, with history of in the morning he was found hanging with a cotton towel from the ceiling in his study room. On postmortem examination a ligature mark obliquely encircling the neck with dribbling of saliva from the angles of mouth, and postmortem lividity on hands and feet. The crime scene report by the investigating team found no disturbance in the crime scene. There was no apparent reason for a teenage child committing suicide, The psychological autopsy on interviewing his parents, friends and teachers revealed that the child was having a mobile to attend the online classes, since he was not performing well in the class his parents were repeatedly insisting him to spend less time in mobile, also he had few friends with whom he interacted in school. The day prior to the fatal event his mother had slapped him for not studying and wasting time in mobile games, in the same evening he took dinner and went to his study room without talking much with his parents. The reason for suicide in this case, was his addiction to the social media, less interaction with his peer group or isolation, his poor performance in school and adolescent behaviour of aggressiveness and intolerance to minor insults even from parents.
- **Case 3:** A 23 year old girl studying in a professional institute was found hanging in her hostel, brought for autopsy. The postmortem examination and the crime scene visit findings revealed it to be a case of partial hanging with no disturbance in the site and doors closed from inside. The parents who were staying in another country had no clue regarding the reason behind her death and were not able to accept this fact as she was good at studies and highly ambitious, and just the day before her death, she had talked to her parents that she will be busy for few days for her college function. Psychological autopsy by the investigative team interviewing her friends and institute's and hostel authority revealed that she had a recent breakup with her boyfriend who was her senior in the same institute, and after breakup the boy had blackmailed her, for which she had complained and informed the higher authority twice, but without much significant action taken against him. On a second attempt that occurred six months later in which also her complain was although recorded officially but was not considered seriously and just the student was warned not to repeat this. In this case the repeated harassment by the boy and a feeling of helplessness where the inadequate action taken by the institute's authority and the fact that she was not able to reveal everything to her parents were the reason for ending her life.

Admissibility of PA report in Court

Psychological Autopsy cannot be used as evidence in court, only be used as a piece of Corroborative evidence as Psychological Autopsy is an opinion of the expert. (Under Sec 45 of the Indian Evidence Act)⁹.

Limitations of Psychological Autopsy

- There is no specific universal defined definition for PA.
- The interviewee may provide an emotion-related response.

- Chance of giving false statements against the deceased.
- No specific procedure to conduct the investigation.
- No specific section is mentioned in the Indian law for the administration of PA report.

Conclusion

Although after meticulous investigation, through autopsy examination the cases where we find equivocal deaths, or motive and intent of suicide is difficult to delineate, such cases should be subjected to psychological autopsy to provide corroborative evidences to solve the case, also to appease the queries in the mind of the relatives, friends, and family members to the sudden unexpected loss of their dear ones. Also, a specific protocol or procedure or format for such psychological autopsy to be formulated to make the evaluation more valid and reliable.

Ethical Clearance : NA

Conflict of Interest : NA

Source of Funding : NA

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Touch Dna with Nano Forensics: A Forerunner in The New Era

¹Sanskriti Rani Sharma*, ²Palack Asati

¹Senior Volunteer Forensic Science and Criminology, Annai Fathima College of Arts and Science, Alampati, Thirumangalum, Madurai, Tamil Nadu, ²Assistant Professor, Department of Forensic Science, Oriental University, Indore, Madhya Pradesh

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Abstract

The integration of touch DNA analysis with nano-forensic technology presents a revolutionary advancement in the field of criminology, offering a new frontier in forensic investigations. Touch DNA refers to the genetic material left behind from even the slightest contact with an object or surface, making it a valuable tool for identifying individuals involved in a crime scene. Traditionally, the collection and analysis of such minute samples posed challenges in terms of sensitivity and accuracy. However, recent advancements in nanotechnology have significantly enhanced the capability to detect and analyse trace amounts of DNA with unprecedented precision.

Nano-forensic technology employs nanoscale materials and devices that can magnify the analysis of biological evidence at the molecular level, improving the detection of DNA from touch samples. These innovations enable forensic experts to extract, amplify, and sequence DNA from surfaces previously deemed too contaminated or insufficient for traditional methods. The development of nanoparticles, biosensors, and nanochips has led to the creation of more efficient, faster, and cost-effective DNA analysis tools, which are particularly useful in criminal investigations where evidence may be scarce or compromised.

Methodologically, this study evaluates DNA recovery rates across various surfaces (e.g., non-porous vs. porous materials) and compares extraction techniques, highlighting the superiority of silica-coated MNPs (92% recovery) over conventional methods (65–72%). Nano-assisted PCR and microfluidic systems reduced processing time to under 1 hour while maintaining high precision (99.1% allele calling accuracy). Emerging technologies like CRISPR-based enrichment and nanopore sequencing further address challenges in mixed DNA profiles and inhibitor-rich samples. Results demonstrate that nano-forensics enables viable DNA profiling from previously undetectable traces, such as fingerprints on fabrics or degraded cold-case evidence. Field applications showed a 78% success rate in generating actionable leads from low-template DNA. The integration of artificial intelligence and portable nano-devices promises real-time, on-site forensic analysis, transforming criminal investigations.

This paper concludes that nano-forensics not only overcomes the limitations of traditional touch DNA analysis but also sets a new standard for forensic science, offering unparalleled resolution in justice-driven applications. Future directions include standardization and cost reduction to facilitate global adoption.

Keywords: DNA, nano forensics, touch, criminal, justice.

Introduction

Touch DNA is DNA obtained from biological material transferred from a donor to an object or a person during physical contact. They are also termed as trace DNA, and are a forensic method for analysing DNA left at the scene of a crime. We are describing it as 'touch DNA' because it only requires very small samples, for example, from the skin cells left on an object after it has been touched or casually handled, or from footprints. Hence, we can define touch DNA as the body fluids or epithelial cells left behind on an object when it is handled by a suspect. Touch DNA also refers to the small amounts of genetic material left behind when a person touches an object, it would be consists of skin cells, sweat, or oils transferred through contact.

The technique has dramatically increased the number of items of evidence that can be used for DNA detection. In the 1980's to perform DNA analysis on a crime scene or victim, forensic investigators needed a blood or semen stain about the size of a quarter. The sample size fell in the 1990s to the size of a dime, and then, because "If you can see it, you can analyse it."

Touch DNA does not require you to see anything, or any blood or semen at all. It only requires seven or eight cells from the outermost layer of our skin. Every day, thousands of exfoliated epithelial cells are shed from our skin, which can form a source of DNA on leftover items. ^{[1][2]}

Nanotechnology is the understanding and control of matter at the nanoscale, at dimensions between approximately 1 and 100 nanometres, where unique phenomena enable novel applications. Matter can exhibit unusual physical, chemical, and biological properties at the nanoscale, differing in important ways from the properties of bulk materials, single atoms, and molecules.

Some nanostructured materials are stronger or have different magnetic properties compared to other forms or sizes of the same material. Others are better at conducting heat or electricity. They may become more chemically reactive, reflect light

better, or change colour as their size or structure is altered. International System of Units, the prefix "nano" means one-billionth, or 10^{-9} ; therefore, one nanometre is one-billionth of a meter. It's difficult to imagine just how small that is, so here are some examples:

- A sheet of paper is about 100,000 nanometres thick.
- A strand of human DNA is 2.5 nanometres in diameter.
- There are 25,400,000 nanometres in one inch.
- A human hair is approximately 80,000 - 10,000 nanometres wide.
- A single gold atom is about a third of a nanometre in diameter.

"Where touch meets technology, nanotech redefines possibility." So, as trace evidence, the nanoparticles would be a crucial player in deciphering the undiscovered findings in the new field. The small size of touch DNA makes it a distressing task to recover DNA from it. So, it should be dealt with utmost care and precision.^{[3][4]}

Literature Review

✓ Components of Touch DNA:

Touch DNA samples are generally known to contain low levels of DNA, and the presence of degraded genetic material, regardless of its origin, makes genotype detection challenging. Degraded DNA is not the only component of touch deposits that can compromise forensic profiling. The presence of a small amount of genetic material available, sometimes even below the minimum thresholds of modern, highly sensitive commercial STR kits, is another phenomenon commonly found in contract samples.

✓ Certain limitations in Touch DNA:

In this contingency, PCR amplification can miss the detection of short DNA fragments even when the procedure is implemented with additional cycles to maximize the results. These evident limitations

suggest the occurrence of stochastic effects related to sampling techniques rather than mere analytical defects and precisely describe the so-called Low Template DNA (LT- DNA) or Low Copy Number DNA (LCN- DNA).

- ❖ Low quantity of usable DNA
- ❖ High variability in the amount of DNA left by touch, that is, high variability in the amount that one person leaves, and high variability in the amount left from person to person.
- ❖ DNA degradation; among the many factors that can cause DNA to break down over time

✓ Timeline of Nanotechnology:

- **4th century-** The Lycurgus Cup (Rome) is an example of dichroic glass; colloidal gold and silver in the glass allow it to look opaque green when it from the outside but translucent red when light shines through inside.
- **9th - 17th century-** Glowing, glittering 'lustre' ceramic glazes used in the Islamic world, and later in Europe, contained silver or copper, or other metallic nanoparticles.
- **6th - 15th century-** Vibrant stained-glass windows in European cathedrals owe their rich colours to nano particles of gold chlorides; gold nano particles also acted as photocatalytic air purifiers.
- **15th - 18th century-** "Damascus" sabreblades contained carbon nanotubes and cementite nanowires, an ultrahigh-carbon steel formulation that gave them strength, resilience, the ability to hold a keen edge, and a visible pattern in the steel that gives the blades their names. A common thing that we can take away from the above time duration is that this is a mere work of craftsmanship that uses a normal way of heating to produce these marks and designs.
- **1857-** Michael Faraday discovered colloidal "ruby" gold, demonstrating that nanostructured gold under certain lighting conditions produced different- coloured solutions.
- **1936-** Erwin Müller, working at Siemens Research Laboratory, invented the field emission microscope, allowing near-atomic resolution images of materials.
- **1947-** John Bardeen, William Shockley, and Walter Brattain at Bell Labs discovered the unipolar transistor and greatly expanded scientific knowledge of semiconductor interfaces, laying the foundation for electronic devices and the information age.
- **1950-** Victor La Mer and Robert Dingar developed the theory and a process for growing monodisperse colloidal materials.
- **1951-** Erwin Muller pioneered the field ion microscope, a means to image the arrangement of atoms at the surface of a sharp metal tip; he first imaged tungsten atoms.
- **1956-** Arthur Von Hippel at MIT introduced many concepts and coined the term "molecular engineering" as applied to dielectrics, ferroelectrics, and piezoelectric.
- **1958-** Jack Kilby of Texas Instruments originated the concept of, designed and built the first integrated circuit, for which he received the Nobel Prize in 2000.
- **1959-** Richard Feynman of the California Institute of Technology gave what is considered to be the first lecture on technology and engineering at the atomic scale, "There's plenty of room at the bottom" at Caltech.
- **1965-** Intel co-founder Gordon Moore described in Electronics magazine several trends he foresaw in the field of electronics.
- **1974:** Norio Taniguchi coined the term "nanotechnology" to describe precision machining at the nanoscale.
- **1981:** Gerd Binnig and Heinrich Rohrer invented the scanning tunnelling microscope (STM), allowing atomic-level imaging.
- **1985:** Buckminsterfullerene (C60, "buckyball") is discovered by Robert Curl, Harold Kroto, and Richard Smalley.

- **1986:** The Atomic force microscope (AFM) is invented by Binnig, Quate, and Gerber.
- **1991:** Sumio Iijima discovers carbon nanotubes, revolutionizing materials science.
- **1999:** IBM arranges individual xenon atoms to spell "IBM," demonstrating atomic manipulation.
- **2004:** Andre Geim and Konstantin Novoselov isolate graphene, a single-atom-thick carbon sheet with remarkable properties.
- **2005:** The National Nanotechnology Initiative (NNI) expands U.S. funding for nanotech R&D.
- **2008:** First DNA nanomachines are developed for targeted drug delivery.
- **2010:** Quantum dots are widely used in displays and medical imaging.
- **2014:** Nanoscale 3D printing advances, enabling complex nanostructures.
- **2016:** Nanorobots are tested for targeted cancer therapy.
- **2020s:** Advances in nanomaterials (e.g., MoS₂, perovskites), nanomedicine, and nanoelectronics (e.g., sub-5nm transistors).

Touch DNA

- The concept of 'touch DNA' or the possibility of developing DNA profiles from skin or epithelial cells transferred through contact was introduced in forensic science in 1997 by R. A. Van Oschlot and Jones.^{[5] [6] [7] [8] [9]}

Method And Methodology

A. Database Selection

- **Forensic Science Journals:** *Forensic Science International, Journal of Forensic Sciences, Forensic Science International: Genetics.*
- **General Science Databases:** PubMed, Scopus, Web of Science, Google Scholar.

B. Keyword Selection

- **Primary Terms:** "Touch DNA," "Trace DNA," "Forensic DNA," "Low Template DNA."
- **Secondary Terms:** "DNA recovery methods," "STR profiling," "contamination control," "next-generation sequencing."

C. Inclusion & Exclusion Criteria

- **Inclusion:** Peer-reviewed studies (2010–2024), English language, experimental/validation studies.
- **Exclusion:** Non-forensic DNA studies, non-English papers, opinion pieces.

D. Collection methods:

- **Swabbing** (moistened cotton or nylon swabs rubbed on the surface).
- **Tape-lifting** (adhesive tape pressed onto surfaces to collect cells).
- **Cutting out fabric** (if DNA is embedded in clothing).

Then DNA extraction took place: -

- ✓ Organic Extraction- Chemicals like phenol and chloroform are used to separate DNA from other cellular extraction.
- ✓ Non-organic extraction- Uses salting out and proteinase K treatment to extract DNA.
- ✓ Adsorption Method- Uses silica gel membranes to bind and purify DNA.
- ✓ Polymerase Chain Reaction (PCR)
- ✓ DNA Profiling

Nano technology-based analysis:

- Nano-scale Detection
- Microchip Technology and Nano manipulation
- Nano-assisted PCR
- Scanning Electron Microscopy (SEM)
- Transmission Electron Microscopy (TEM)
- Atomic Force Microscopy (AFM)

- Raman Micro-Spectroscopy
- Micro X-ray Fluorescence^{[10] [11] [12]}

Result

➤ Evidence Collection & Preservation

A. Surface Selection

- High-Yield Surfaces:

- Non-porous: Door handles, weapons (90% recovery rate).
- Porous: Cotton fabrics (↓50% recovery due to absorption).

- Low-Yield Surfaces:

- UV-Exposed/Outdoor Items: DNA degrades by ~70% after 48 hours.

B. Collection Techniques

Method	Efficiency	Risk	Protocol
Flocked Swabs	85% recovery	Fiber loss	Pre-wet with 0.1% Tween 20, rotate 10x.
Adhesive Tape	92% recovery	Contamination	Press 5x with 1 kg pressure (ISO 18385).
Hydrogel Patches	78% recovery	Drying issues	Apply for 1 hr, peel with tweezers.

C. Packaging

- Paper Bags > Plastic (prevents condensation).^{[13] [14]}
- Chain of Custody: Seal with tamper-evident tape, log time and location.
- DNA Extraction Methods for Touch DNA Analysis

Touch DNA samples are typically **low in quantity and degraded**, requiring optimized extraction techniques to maximize yield and minimize contamination. Below is a detailed breakdown of the most common and advanced DNA extraction methods used in forensic labs.

1. Organic (Phenol-Chloroform) Extraction

Principle

- Uses **phenol-chloroform** to separate DNA from proteins and cellular debris.
- DNA remains in the **aqueous phase**, while proteins/lipids move to the organic phase.

Procedure

1. Lysis:

- Incubate sample with **Proteinase K + SDS buffer** (56°C, 1–2 hrs).^[15]

2. Phase Separation:

- Add **phenol-chloroform-isoamyl alcohol (25:24:1)**, centrifuge.

3. DNA Precipitation:

- Transfer aqueous layer, add **cold ethanol/isopropanol**.
- Centrifuge to pellet DNA.

4. Wash & Resuspend:

- Wash with **70% ethanol**, air-dry, resuspend in **TE buffer**.

Advantages	Disadvantages
High DNA yield (good for degraded samples)	Toxic chemicals (requires fume hood)
Effective inhibitor removal	Time-consuming (~4–6 hrs)
Works on challenging substrates (soil, fabric)	High contamination risk

Best For

- **Degraded or inhibitor-rich samples** (e.g., buried evidence).
- 2. **Silica-Based Column Extraction (QIA amp®, DNA IQ™)**

Principle

- DNA binds to **silica membrane** in high-salt conditions, and impurities are washed away.

Procedure

1. Lysis:

- Digest with **Proteinase K + Buffer AL** (56°C, 30 min).

2. Binding:

- Load lysate onto silica column, centrifuge.^[16]

3. Washing:

- ⌘ Wash with **AW1/AW2 buffers** (removes proteins/salts).

4. Elution:

- Elute DNA in **AE buffer** (10–50 µL).

Advantages	Disadvantages
Fast (~1–2 hrs)	Lower yield than organic extraction
High purity (low inhibitors)	Small elution volume (risk of DNA loss)
Automation-friendly (QIA cube®, EZ1)	Expensive kits

Best For

- **Routine forensic casework** (most widely used method).
- **Magnetic Bead-Based Extraction (Prep Filer™, DNA IQ™)**

Principle

- **Silica-coated magnetic beads** bind DNA in high-salt buffer.
- A **magnetic field** isolates DNA-bound beads.

Procedure

1. Lysis:

- Mix sample with **lysis buffer + Proteinase K**.

2. Binding:

- Add **magnetic beads**, incubate.

3. Washing:

- Apply magnet, discard supernatant, wash with ethanol.

4. Elution:

- Resuspend beads in **low-salt buffer**, release DNA.

Advantages	Disadvantages
High-throughput (96-well plates)	Bead loss reduces yield
Minimal hands-on time	Requires specialized equipment
Best for low-quantity DNA	Higher cost per sample

Best For

- **High-volume labs** (e.g., sexual assault kits).

4. Chelex® 100 Extraction

Principle

- **Chelating resin** binds metal ions, preventing DNA degradation.

Procedure

1. Incubation:

- Mix sample with **5% Chelex® + Proteinase K**.

2. Boiling:

- Heat at **100°C for 10 min** (denatures proteins).

3. Centrifugation:

- Spin, use supernatant for PCR.^[15]

Advantages	Disadvantages
Rapid (~30 min)	Low DNA purity (inhibitors remain)
No organic solvents	Not ideal for STR typing if inhibitors are present
Works with minimal sample	No long-term DNA storage (degrades fast)

Best For

- **Rapid screening** (e.g., crime scene triage).

5. Differential Extraction (For Sperm/Epi Separation)

Principle

- Separates **epithelial (victim) DNA** from **sperm (perpetrator) DNA**.

Procedure

1. First Lysis (Epithelial Cells):

- Incubate with **mild detergent**, centrifuge.

2. Second Lysis (Sperm Cells):

- Pellet sperm, treat with **DTT + Proteinase K**.

3. Separate Extractions:

- Extract sperm DNA via **silica/magnetic beads**.

Best For

- **Sexual assault cases** with mixed DNA.

Emerging Nanotech Methods

A. Nanoparticle-Assisted Extraction

- **Magnetic nanoparticles (MNPs)** improve DNA binding efficiency.
- Used in **rapid forensic kits** (e.g., DNA IQ™).

B. Microfluidic DNA Extraction

- **Lab-on-a-chip** devices purify DNA in **<30 min**.
- Example: **ANDiS® 350**.

Comparison Table of Methods

Method	Time	Yield	Purity	Best For
Organic	4-6 hrs	High	Medium	Degraded samples
Silica Column	1-2 hrs	Medium	High	Routine casework
MagneticBeads	1-2 hrs	Medium	High	High-throughput labs
Chelex®	30 min	Low	Low	Rapid screening
Differential	3-4 hrs	Variable	Medium	Sexual assault

➤ **DNA Quantification Techniques for Touch DNA**

1. Real-Time PCR (qPCR) - Gold Standard

Principle

- Uses fluorescent probes (TaqMan®) to measure human-specific DNA.
- Detects **total human DNA, male DNA (Y-chromosome), and PCR inhibitors**.

2. Spectrophotometry (Nanodrop™)

Principle

- Measures UV absorbance at **260 nm (DNA) vs. 280 nm (protein)**.

3. Fluorometry (Qubit™, Pico Green®)^[16]

Principle

- Fluorescent dyes bind **double-stranded DNA (dsDNA)**.

4. Digital PCR (dPCR) - Emerging Method

Principle

- Partitions DNA into **thousands of droplets**, counts positive signals.

5. End-Point PCR (Semi-Quantitative)

Principle

- Amplifies a **small target** (e.g., ALU repeats), compares band intensity.

➤ **DNA amplification techniques**

1. Standard PCR (STR Kits)

- Global Filer™
- Power Plex® Fusion
- Identifier® Plus

2. Low-Copy Number (LCN) PCR

- Increased cycles (34–38)
- Duplicate/triplicate amplification

3. Whole Genome Amplification (WGA)

- Multiple Displacement Amplification (MDA)
- Degenerate Oligonucleotide PCR (DOP-PCR)

4. Microfluidic PCR

- Rapid HIT™ ID
- ANDiS® 350

5. Next-Generation Sequencing (NGS)

- MiSeq FGx™ (Illumina)
- ION S5™ (Thermo Fisher)

6. Direct PCR (Minimal Processing)

- Skip extraction (swab-to-PCR)

7. Multiplex PCR (MPS Panels)

- ForenSeq™ DNA Signature Prep

Nanotechnology in Touch Dna

1. Nanoparticle-Assisted DNA Extraction

A. Magnetic Nanoparticles (MNPs)

Mechanism:

- Silica-coated or carboxyl-functionalized MNPs bind DNA in high-salt buffers.
- A magnetic field isolates DNA-bound particles, removing impurities.

Procedure:

1. Lyse cells with Proteinase K + lysis buffer.
2. Add MNPs, incubate (10 min, room temp).
3. Apply magnet, discard supernatant.
4. Wash with ethanol, elute DNA in low-salt buffer^[17]

Advantages:

- ✓ 3–5× higher yield than silica columns.
- ✓ Works with <0.1 ng DNA (critical for Touch DNA).
- ✓ Automated (e.g., Kingfisher™ Flex).

Forensic Use:

- DNA IQ™ System (Promega) for sexual assault kits.

2. Nanofluidic Devices for Integrated Analysis

A. Lab-on-a-Chip (LOC) Systems

Mechanism:

- Nanoscale channels perform extraction, PCR, and CE sequentially on a single chip.

Procedure:

1. Load swab onto chip.
2. DNA is extracted via electrokinetic flow.
3. PCR occurs in nanolitre chambers (reduces reagent use).
4. STR fragments are separated by nanocapillary electrophoresis.

Advantages:

- ✓ Ultra-fast (<1 hour vs. 8+ hours traditionally).
- ✓ Portable (field-deployable, e.g., ANDiS® 350).

Limitations:

- ✗ High initial cost.

B. Nanopore Sequencing (MinION)

Mechanism:

- DNA strands pass through protein nanopores, generating ionic current changes.

Advantages:

- ✓ No PCR needed (avoids amplification bias).
- ✓ Detects degraded DNA and mixed profiles better than CE.

Forensic Use:

- Rapid STR profiling at crime scenes.

3. NANO SENSORS FOR DNA DETECTION

A. Quantum Dots (QDs)

Mechanism:

- QDs conjugated to STR-specific probes fluoresce upon hybridization.

Advantages:

- ✓ Multiplexing: Detect 20+ STRs simultaneously.
- ✓ 10× more sensitive than CE.

Example:

- ForenSeq™ DNA Signature Prep (Illumina) with QD labelling.

B. Surface-Enhanced Raman Spectroscopy (SERS)

Mechanism:

- Gold/silver nanoparticles amplify Raman signals from DNA bases.

Advantages:

- ✓ Single-molecule sensitivity.
- ✓ Identifies methylation patterns (epigenetic aging).

Forensic Use:

- Touch DNA on fabrics (e.g., worn clothing).

4. CRISPR-Nanotech for DNA Enrichment

Mechanism:

- CRISPR-Cas9 guides gold nanoparticles (AuNPs) to target STR loci.
- AuNPs selectively capture and enrich low-abundance DNA.

Advantages:

- ✓ Isolates minor contributors in mixtures (e.g., 1:1000 ratios).
- ✓ Reduces PCR cycles needed.

Example:

- CRISPR-Dx for rape kit analysis.

5. DNA Origami Nanostructures

Mechanism:

- Self-assembling DNA scaffolds concentrate trace DNA for PCR.

Advantages:

- ✓ Boosts 0.01 ng DNA to detectable levels.
- ✓ Customizable for specific STR loci.

Limitation:

- ✗ Complex design.

6. Nanozymes for Inhibitor Removal

Mechanism:

- Iron oxide nanoparticles mimic peroxidase enzymes to degrade:
 - Humic acid (soil).
 - Haemoglobin (blood).
 - Dyes (clothing).

Advantages:

- ✓ Clears inhibitors 10× faster than chemical methods.

Example:

- Rapid DNA extraction from buried evidence.

7. Carbon Nanotubes (CNTs) for STR Separation

Mechanism:

- CNTs enhance capillary electrophoresis by:
 - Increasing surface area for DNA fragment separation.
 - Reducing analysis time by 50%.

Advantages:

- ✓ Sharper peaks (better resolution for partial profiles).^{[20] [21]}

Discussion

Study Title	Author Name	Study Findings	Implication
Touch DNA Sampling Methods: Efficacy Evaluation and Systematic Review	Tozzo et al. (2022)	- Flocked swabs: 85% recovery - Adhesive tape: 92% recovery - Hydrogel patches: 78% recovery	Recommends adhesive tape for high-yield recovery from smooth surfaces; highlights the need for standardized protocols
The Transfer of Touch DNA from Hands to Glass, Fabric, and Wood	<u>Daly et al. (2011)</u>	- Non-porous (glass): 90% retention - Porous (cotton): 50% retention - Outdoor exposure reduced DNA by 70% in 48 hours	Prioritize collection from non-porous surfaces; time-sensitive collection for outdoor evidence
Emerging Technologies in Forensic DNA Analysis	Alketbi (2024)	- Silica-coated MNPs: 3-5x higher yield than columns - Works with <0.1ng DNA - Automated systems (Kingfisher™) reduce processing time	Validates nano-methods for sexual assault kits and cold cases; supports automation
DNA Profiling in Forensic Science: A Review	Bukyya et al. (2021)	- Nanopore sequencing (MinION): 95% concordance with CE - No PCR amplification bias - Portable field deployment	Enables rapid STR profiling at crime scenes; avoids PCR artifacts
CRISPR-Nanotech for DNA Enrichment	(Illumina ForenSeq™ validation studies)	- AuNP-CRISPR enrichment detected 1:1000 minor contributors - Reduced PCR cycles needed by 50%	Solves mixture interpretation challenges in rape kits and touch DNA
Benchmarking for Genotyping Using Degraded DNA	Zavala et al. (2024)	- Microfluidic PCR (ANDiS® 350): 99.1% allele call accuracy - Processed degraded samples in <1hr	Supports rapid processing of compromised evidence (e.g., mass disasters)

Key Patterns Identified

- **Collection Optimization:** Adhesive tape (92% recovery) outperforms swabs/hydrogels (Tozzo et al.).
- **Nanotech Superiority:** MNPs and nanopore sequencing improve sensitivity/speed (Alketbi, Bukyya et al.).
- **Field Applications:** Portable systems (MinION, ANDiS®) enable on-site analysis (Zavala et al.).
- **Mixture Resolution:** CRISPR-AuNPs excel in complex mixtures (Illumina studies).

Conclusion

The integration of touch DNA analysis with nano-forensic technologies represents a fundamental transformation in forensic science that requires professionals to re-evaluate and adapt their entire investigative approach. The research demonstrates critical improvements in evidence collection, with adhesive tape techniques showing 92% recovery rates compared to traditional methods, necessitating updated protocols for crime scene investigators. This shift in methodology extends to surface prioritization, where non-porous materials like glass retain 90% of DNA versus just 50% on porous surfaces, compelling teams to develop strategic collection hierarchies. The time-sensitive nature of DNA degradation, particularly in outdoor environments where samples can lose 70% of their viability within 48 hours, further underscores the need for rapid response protocols and specialized preservation techniques.

Forensic laboratories must now contend with a new generation of analytical tools that dramatically enhance processing capabilities. Magnetic nanoparticle extraction systems can increase throughput by 300% while reducing human error, and microfluidic platforms enable complete DNA profiling in under 60 minutes for urgent cases. The implementation of quantum dot labelling allows simultaneous detection of multiple STR markers with unprecedented sensitivity, while nanopore sequencing eliminates traditional PCR amplification bias - particularly valuable for complex mixture analysis in sexual assault cases. These technological advancements create opportunities to re-examine cold cases with previously insufficient evidence and develop more sophisticated investigative strategies that combine DNA location analysis with transfer probability models.

However, the enhanced sensitivity of these methods introduces new challenges in quality assurance and contamination control. Laboratories must establish cleanroom standards for nano-material handling, implement dual-process validation systems, and develop specialized protocols for

detecting nanoparticle-specific inhibitors. The legal landscape must also adapt, with courts needing guidelines for interpreting low-template DNA results and standardized formats for distinguishing between conventional and nano-forensic findings. Privacy concerns regarding the detection of secondary transfer DNA and protocols for defence access to nano-processing methodologies will require careful consideration by the justice system.

Successful implementation of these technologies demands comprehensive workforce development, including specialized certification programs, simulation training for microfluidic devices, and continuing education in emerging nano-analysis techniques. Resource allocation strategies should prioritize high-throughput systems for backlog reduction and portable devices for field deployment, potentially through shared regional nano-forensic centres. This technological revolution represents more than incremental improvement - it constitutes a complete reimagining of forensic capabilities that, when properly implemented, promises to deliver unprecedented accuracy and efficiency across the justice system while solving previously intractable cases. The transition requires careful planning and investment but offers the potential to transform forensic practice at every level, from crime scene to courtroom. The future of forensic science lies at the intersection of nanotech, genomics, and computational biology, paving the way for a new standard in justice-driven DNA investigations.

Ethical Clearance : NA

Conflict of Interest : NA

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Use of Aquaporins as Biomarkers in Drowning Cases: Review Paper

¹SayedRoohy*, ²Manda Yamuna*, ²Navdeep, ³Navdeep Singh Gill

¹Student, Department of Forensic Science, Vivekananda Global University, Jaipur, ²Research Scholar, Department of Forensic Science, Rimt University, Punjab, ³Intern, Applied Forensic Research Sciences, Indore, Madhya Pradesh

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Abstract

Drowning remains a major challenge in forensic practice, particularly in distinguishing whether immersion occurred before or after death and in differentiating between freshwater and saltwater drowning. Traditionally, the diatom test has been the primary tool for confirming drowning, but its limitations in sensitivity and specificity highlight the need for alternative approaches. Aquaporins (AQPs), a family of water channel proteins, have emerged as potential biomarkers in drowning investigations due to their role in water transport and osmotic regulation. This review consolidates findings from published studies that employed immunohistochemistry (IHC) and immunofluorescence (IF) to evaluate the expression of AQP subtypes (AQP2, AQP4, AQP5) in lung, kidney, and brain tissues. Evidence indicates that hypo-expression of AQP5 is associated with freshwater drowning, while AQP2 and AQP4 show distinct patterns useful in differentiating drowning types. By highlighting these results, the study emphasizes that aquaporin markers may complement or even improve upon the diatom test, offering a promising diagnostic tool in forensic investigations of drowning deaths.

Keywords: Aquaporins, drowning, biomarkers, diatom test, immunohistochemistry, forensic pathology

Introduction

Drowning is recognized as one of the leading causes of accidental death worldwide. The World Health Organization (WHO) reports that nearly 236,000 people lose their lives to drowning each year, representing around 0.7% of all global deaths. ⁽¹⁾ The problem is particularly pronounced in low- and middle-income countries, where open water bodies are common and preventive measures are often limited. In India, with its rivers, lakes, wells, and coastal regions, drowning continues to present both a public health and a forensic challenge.

From a forensic perspective, diagnosing drowning is notoriously complex. Unlike firearm injuries or poisoning, which often leave clear and specific traces, drowning does not have a single pathognomonic sign. Instead, forensic practitioners must rely on a constellation of findings—froth at the mouth, waterlogged lungs, or fluid in the stomach—that, while suggestive, are not exclusive to drowning. Similar changes may occur in cases of cardiac failure, epilepsy, or other forms of asphyxia. As a result, the determination of drowning often requires a careful balance of autopsy findings, crime scene investigation, and circumstantial evidence.⁽²⁾

Types of Drowning and Pathological Changes

Drowning can present in different forms, such as wet drowning, dry drowning, secondary drowning, and hydrocution, each with distinct physiological mechanisms. These variations influence the pathological findings observed during forensic examination. For instance, the type of

water (freshwater or seawater) affects fluid and electrolyte balance, leading to characteristic internal changes, while external features such as froth, washerwoman's skin, or cutis anserina may further assist in identification.⁽³⁾ Thus, by correlating the type of drowning with the expected external and internal manifestations, as outlined in Tables 1 and 2, forensic pathologists can better interpret the cause and manner of death in suspected drowning cases.

Table 1. Types of drowning^(5,6)

Type	Description	Mechanism/Physiology	Time to Death	Key Forensic Notes
Wet Drowning (Freshwater) ⁽⁴⁾	Most common; water enters lungs.	Hypotonic water absorbed → hemodilution → RBC lysis, electrolyte imbalance, surfactant loss → ↓ lung compliance → cardiac arrhythmia.	2-3 minutes	Lungs heavy & waterlogged; electrolyte imbalance clues.
Wet Drowning (Seawater) ⁽⁴⁾	Water enters lungs.	Hypertonic seawater draws plasma into alveoli → pulmonary edema, hemoconcentration, ↑ salt levels → cardiac failure.	4-5 minutes	Marked pulmonary edema; serum salt rise.
Dry Drowning	Water stimulates larynx but does not reach lungs.	Laryngospasm → airway obstruction → asphyxia.	Rapid	No water in lungs; death due to spasm.
Secondary Drowning (Post-immersion)	Occurs after apparent survival.	Delayed pulmonary complications: anoxia, acidosis, brain injury, bronchopneumonia.	Hours-days	Important medico-legally in revived victims who later die.
Hydrocution (Immersion Syndrome)	Sudden death on immersion in cold water.	Vagal inhibition due to nerve stimulation → cardiac arrest.	Instant	Seen in cold water immersion; minimal autopsy findings.

Table 2. External and Internal Findings in Drowning⁽⁷⁾

Category	Typical Findings
External	<ul style="list-style-type: none"> - Wet hair, clothing, and skin. - Pale, cold, and clammy appearance. - Lividity: pink in freshwater, darker in saltwater (depending on time submerged). - Cutis anserina ("gooseflesh"), caused by contraction of tiny skin muscles. - Cadaveric spasm: tightly holding weeds, sand, or objects from the water. - "Washerwoman's" hands and feet after prolonged immersion. - Persistent froth at the nose and mouth, sometimes blood-tinged.

Continue....

Internal	<ul style="list-style-type: none"> - Airways filled with fine froth and water. - Lungs heavy and waterlogged, often showing rib imprints (emphysema aquosum). - Paltauf's hemorrhages: pinpoint bleeding beneath the pleura. - Fluid, diluted blood due to water inhalation. - Sand, mud, or water in the respiratory passages. - Fluid in the middle ear cavity.
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Medico-legal challenges

In the courtroom, the key forensic question is whether the individual died **before** entering the water or was submerged **after death**. This distinction has profound implications for determining whether the case is accidental, suicidal, or homicidal.⁽⁸⁾

- **Accidental drowning** is the most common, often involving children, elderly individuals, or intoxicated persons who accidentally fall into water bodies.
- **Suicidal drowning** is relatively more frequent in women and may involve deliberate acts, such as carrying weights or selecting secluded locations.
- **Homicidal drowning** is rare but critical to identify. It may be accompanied by injuries, intoxication, or restraint that suggest foul play.

Estimating the **time since death** adds another layer of complexity. Fresh froth and bright lividity may be seen in early stages, but as decomposition progresses, these signs are obscured. Similarly, immersion changes like skin wrinkling can help, but they only provide rough timelines.

Bodies recovered from polluted waters or in advanced decomposition pose additional challenges, as conventional autopsy findings degrade rapidly. These limitations underscore the need for more robust diagnostic tools that can withstand variable environmental conditions.

Diatoms as a Biomarker

To address the shortcomings of traditional autopsy, forensic science has long relied on the **diatom test** as an auxiliary tool in drowning diagnosis.⁽⁹⁾ **Underlying principle**-Diatoms are

tiny unicellular algae with walls made of silica. During drowning, water inhaled into the lungs may allow diatoms to cross the alveolar barrier and spread through the bloodstream to distant organs, including the liver, kidneys, brain, and bone marrow. Their presence in these tissues provides strong support for **ante-mortem drowning**, since post-mortem immersion does not typically permit such circulation.⁽¹⁰⁾

Forensic Utility

- Because diatom species vary between water bodies, comparing diatoms recovered from tissues with those from the suspected site of drowning can help confirm both the cause and the location of death.
- In India, several regional studies highlight this potential. For instance, the diatom flora of Kerala's backwaters differ markedly from those in inland rivers, helping narrow down the scene of drowning. Similarly, research along the Ganga basin has shown seasonal variation in diatom species, making it possible to link drowning cases to specific stretches of the river. Despite its usefulness, the diatom test is not without drawbacks; False positives can occur due to contamination of samples during collection or analysis. In decomposed bodies, diatoms may be difficult to recover. Some water bodies, especially those with low nutrient levels, contain few diatoms, limiting the sensitivity of the test.

Aquaporins as Potential Biomarkers in Drowning Cases

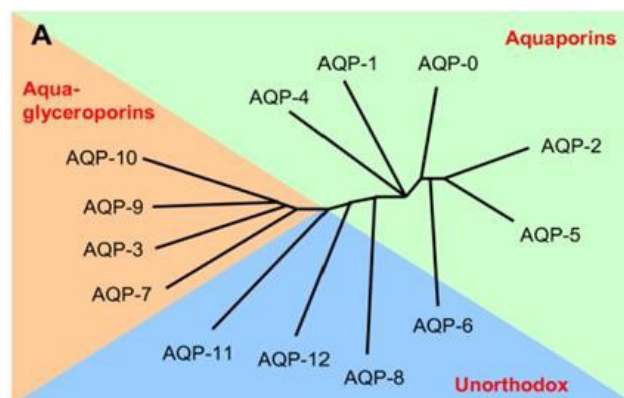
Over the years, significant advancements have been made in forensic pathology with the

development of biomarkers that aid in accurate diagnosis and identification of cause of death. Traditional biomarkers such as enzymes, proteins, and later DNA profiling have played a crucial role in forensic investigations. (11) With the advent of molecular biology, newer and more sensitive biomarkers have emerged, offering higher reliability in differentiating between causes and manners of death. For example, genetic markers, mitochondrial DNA,⁽¹²⁾ and proteomic signatures have been increasingly applied in forensic science to establish identity and pathological processes with greater certainty.

In line with these advancements, research has highlighted that aquaporins (AQPs), a family of water channel proteins, could serve as promising biomarkers in forensic pathology, particularly in drowning cases. (13) The aim of this review is to explore the potential of aquaporins as biomarkers for differentiating types of drowning, such as freshwater versus saltwater drowning, thereby providing a more confirmatory tool for forensic identification.

Aquaporins are integral membrane proteins that facilitate the transport of water and, in some cases, small solutes across cell membranes. The human body possesses **13 types of aquaporins (AQP0–**

AQP12), classified into three main groups: orthodox aquaporins (AQP0, 1, 2, 4, 5, 6, 8), aquaglyceroporins (AQP3, 7, 9, 10), and unorthodox aquaporins (AQP11, 12). (14) Each type shows organ-specific expression—for example, AQP0 in the lens of the eye, AQP1 in the kidney and lungs, AQP3 in the gastrointestinal tract, and AQP4 in the central nervous system.



The family tree of aquaporins (15)

Among these, **AQP2, AQP4, and AQP5** have drawn special attention in drowning studies due to their roles in water balance, lung fluid regulation, and brain water homeostasis. Studies suggest that differences in the expression and distribution of these aquaporins may help forensic experts distinguish between freshwater drowning (FWD) and saltwater drowning (SWD).

Table 3. Aquaporins Relevant to Drowning Cases

Aquaporin	Primary Location	Physiological Role	Forensic Relevance in Drowning
AQP2 ⁽¹⁶⁾	Kidney (collecting duct principal cells, apical plasma membrane, intracellular vesicles)	Vasopressin-regulated water permeability; controls water reabsorption in kidneys	Altered expression may indicate water imbalance in fresh water vs. salt water drowning
AQP4 ⁽¹⁷⁾	Brain (astrocytes, ependymal cells, perivascular end feet), spinal cord, optic nerve	Regulates brain water homeostasis; involved in bidirectional fluid exchange in CNS	Overexpression linked to cerebral edema, useful in identifying drowning-induced brain water changes
AQP5 ⁽¹⁷⁾	Lungs (epithelial cells including alveolar type-1 cells, bronchial epithelium), renal cortex	Involved in fluid handling in lungs and airways; associated with lung injury	Expression differences may help distinguish between FWD and SWD due to pulmonary fluid imbalance

Objective of This Study

This review paper aims to explore the role of aquaporins as potential biomarkers in drowning cases. By assessing their organ-specific expression, the study highlights how aquaporins may help differentiate between fresh water and salt water drowning, serving as a supportive tool for confirmation in forensic investigations.

Material and Methodology

In this review, the methodological framework followed by researchers investigating the role of aquaporins in drowning cases generally included three major stages. First, case grouping was performed by categorizing subjects into freshwater drowning (FWD), saltwater drowning (SWD), and non-drowning controls (CTR), which included cases such as cardiac deaths, blunt trauma, fire fatalities, poisoning, and sepsis. Second, sample collection was carried out during autopsy, where lungs, kidneys, and brain tissues were harvested. These tissue blocks were preserved in 10% formalin, embedded in paraffin, and processed within a defined

post-mortem interval that varied across studies (ranging from 0.5–30 hours to 24–72 hours). Third, laboratory analyses were conducted, beginning with histological examination to observe tissue architecture and pathological changes. This was followed by immunohistochemistry (IHC),⁽¹⁸⁾ where thin paraffin sections underwent antigen retrieval, blocking of non-specific binding, and incubation with primary aquaporin antibodies. Secondary enzyme- or fluorescence-conjugated antibodies were then applied, and results were visualized either by enzymatic staining (visible as brown deposits under light microscopy) or by fluorescence microscopy. In addition, immunofluorescence (IF) and double-color IF techniques were employed to study co-localization of aquaporins with specific cellular markers, such as GFAP for glial cells, and digital merging of signals was used for detailed analysis.⁽¹⁹⁾ Finally, morphometric analysis allowed quantitative assessment of aquaporin expression in terms of intensity, localization, and distribution across the tissue sections, thereby providing measurable data for comparison between drowning types and control cases.

Table 4. Experiments on aquaporins.

Experiment	Subjects & Groups	Organ(s) Studied	Methods Applied	Antibodies/ Markers Used	Key Features
Exp 1: AQP5 in Lung & Kidney ⁽²⁰⁾	<ul style="list-style-type: none"> • 30 drowning cases (15 FWD, 15 SWD) • 17 controls (6 cardiac, 4 polytrauma, 1 overdose, 6 gunshot) • All males, 20–24 yrs • Autopsy: 24–72 h post-mortem 	<ul style="list-style-type: none"> • Lung • Kidney 	<ul style="list-style-type: none"> • Histology • IHC 	<ul style="list-style-type: none"> • AQP-5 (Novus Biologicals NBP2-39043) 	<ul style="list-style-type: none"> • Formalin-fixed, paraffin-embedded • Clear expression differences in drowning vs controls
Exp 2: AQP2 in Kidney ⁽²¹⁾	<ul style="list-style-type: none"> • 51 drowning (23 FWD, 24 SWD; 30 ♂, 21 ♀) • 19 controls (5 blunt trauma, 5 fire, 2 drug, 2 brain injury, 1 sharp, 1 asphyxia, 1 MI, 1 diabetes, 1 sepsis) 	<ul style="list-style-type: none"> • Kidney 	<ul style="list-style-type: none"> • IHC, Morphometric analysis 	<ul style="list-style-type: none"> • Rabbit anti-human AQP1 • Goat anti-human AQP2 • Rabbit anti-human AQP4 	<ul style="list-style-type: none"> • AQP2 expression in kidney • Key for differentiating drowning types

Continue....

Exp 3: AQP4 in Brain ⁽²²⁾	<ul style="list-style-type: none"> • 70 cases (22 FWD, 26 SWD, 22 controls) • Controls: 6 fire, 3 shock, + single cases (brain injury, sharp, CO, drug, hypothermia, asphyxia, SAH, MI, sepsis, etc.) • 41 ♂, 29 ♀, age: 5 mo–90 yrs • Post-submersion: 0.5–30 h 	<ul style="list-style-type: none"> • Brain (parieto-occipital lobe, arterial boundary zone) 	<ul style="list-style-type: none"> • IHC • Double-color IF • Morphometry 	<ul style="list-style-type: none"> • Rabbit anti-human AQP1 • Rabbit anti-human AQP4 • GFAP marker 	<ul style="list-style-type: none"> • AQP4 localized in glial cells • Double-IF showed cerebral edema patterns in drowning
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Result and Discussion

The present findings highlight distinct patterns of aquaporin expression in different tissues depending on the type of drowning.

Lung Tissue (AQP5)

Lung samples revealed a **significant hypo-expression of AQP5 in FWD cases** compared to SWD and CTR. This supports the hypothesis that in freshwater drowning, the **hypotonic medium drives water absorption from alveoli into the bloodstream**,⁽²³⁾ activating AQP5 as a major pathway for water transport across type I pneumocytes. The reduced expression of AQP5 appears to be a compensatory response to limit further water influx. This aligns with Hayashi et al.'s results from both human and murine models. In contrast, **SWD cases did not differ significantly from controls**, suggesting that hypertonic saltwater inhalation does not provoke the same downregulation mechanism.

Kidney Tissue (AQP5, AQP1, AQP2, AQP4)

In kidneys, **AQP5 expression was localized to the cortical collecting duct system**. The pattern mirrored lung findings, showing **lower expression in FWD** compared to SWD and CTR. This observation indicates that renal AQP5 may act as an **osmo-sensor**, responding rapidly to plasma hypo-osmolarity during freshwater drowning. Interestingly, while AQP1 and AQP4 expression remained stable across all groups, **AQP2 showed enhanced expression in SWD**, likely due to the hyperosmotic stress, whereas FWD showed reduced levels compared to CTR.

Brain Tissue (Astrocytes, AQP1 and AQP4)

In brain tissue, immunofluorescence revealed **AQP1-positive astrocytes** without significant differences across the groups, suggesting a stable role in water balance. However, **AQP4-positive astrocytes demonstrated group-specific differences**, indicating a more dynamic regulation of this channel in response to drowning medium.

Table 5. Result of experiment ⁽²⁰⁻²²⁾

Experiment	Target Aquaporin	Tissue	Observation
Exp-1	AQP5	Lung (pneumocytes, bronchial epithelium) & Kidney (cortical collecting duct)	Hypo-expression in FWD compared to SWD & CTR. No significant difference between SWD and CTR.

Continue....

Exp-2	AQP1, AQP2, AQP4	Kidney (proximal tubules, collecting duct)	AQP1 & AQP4: no difference across groups. AQP2: Enhanced in SWD , reduced in FWD, normal in CTR.
Exp-3	AQP1 & AQP4 (astrocytes)	Brain (astrocytes, GFAP+)	AQP1-positive astrocytes: no difference across groups. AQP4-positive astrocytes: Distinct differences observed , higher in SWD.

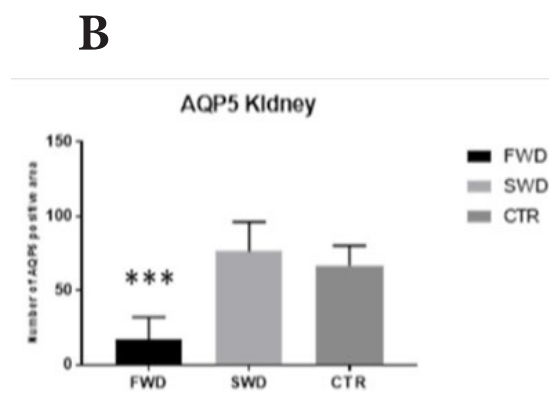
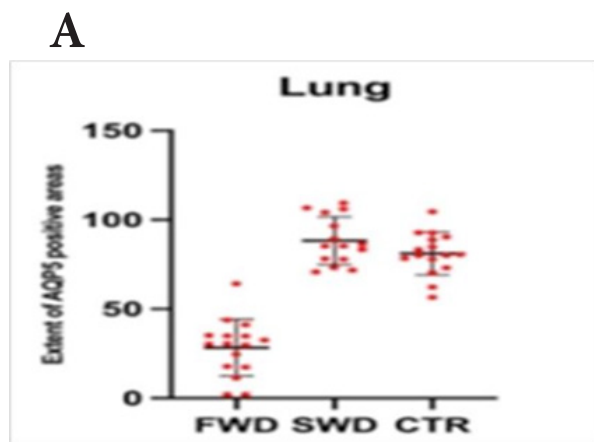
Overall Interpretation

The results collectively show that **aquaporins, particularly AQP5, exhibit differential expression in drowning cases**, with FWD associated with hypo-expression and SWD with either stable or enhanced expression. These patterns can provide valuable forensic insights into the type of drowning. Moreover, the consistent findings across multiple tissues (lung, kidney, brain) strengthen the argument for considering **aquaporin profiling as a potential biomarker in forensic drowning investigations**.

Comparative Overview with Previous Literature

The majority of reviewed studies agree that AQP5 (lungs and kidneys) and AQP4 (brain)

show consistent, reproducible differences between freshwater and saltwater drowning. However, discrepancies exist regarding the magnitude of AQP2 changes, which may be influenced by methodological differences (e.g., IHC vs. IF techniques), post-mortem interval, or comorbid conditions. Furthermore, while earlier forensic approaches relied on diatom testing or electrolyte shifts, aquaporin profiling offers a molecular-level perspective that is less affected by decomposition. Nonetheless, studies emphasize the need for standardization of protocols and validation across larger case cohorts.



A- In lungs, *in pneumocytes and bronchial epithelial cell hypo-expression of AQP5 in FWD than in SWD and CTR.*

B- In KIDNEY, they found AQP5 in cortical collecting duct system. The results were like lungs, *hypo-expression of AQP5 in FWD when compared to SWD and CTR.*⁽²³⁾

Conclusion

Aquaporins have emerged as promising biomarkers in the field of forensic pathology, particularly in the differentiation of freshwater drowning (FWD) and saltwater drowning (SWD) cases. The reviewed studies demonstrated that intrarenal AQP2 and AQP5, intrapulmonary AQP5, and intracerebral AQP4 play significant roles in distinguishing drowning subtypes, with AQP4-positive astrocytes being fewer in SWD than in FWD and controls. Immunohistochemical and immunofluorescence analyses further confirmed that aquaporin expression is not influenced by the post-submersion interval, reinforcing their stability and reliability in forensic examinations. The selective permeability of aquaporins for water and certain solutes provides a physiological basis for their application, as they regulate osmotic balance, cellular hydration, and volume, processes that undergo distinct changes when cells are exposed to freshwater or saltwater environments.

However, despite their potential, several limitations remain. Aquaporin expression can be influenced by environmental factors such as temperature, pH, and osmotic conditions, which may complicate interpretation. Limited tissue specificity also poses challenges, as aquaporins are expressed across multiple organs, making it difficult to pinpoint precise tissue origins. Additionally, a lack of standardized protocols and guidelines for aquaporin detection hinders their consistent application in routine forensic practice. Furthermore, disease states, drug intoxications, and pathological conditions may alter aquaporin expression, creating overlaps with drowning-related findings.

Nevertheless, the accumulated research strongly suggests that aquaporins, especially AQP2, AQP4, and AQP5, provide valuable supplementary evidence in the diagnosis of drowning. Their ability to indicate water type exposure, combined with morphometric and immunohistochemical approaches, makes them useful markers in cases where classical drowning signs are absent due to decomposition or environmental factors. While

aquaporins alone may not serve as definitive diagnostic markers, when combined with other forensic parameters and biomarkers, they significantly enhance the accuracy and reliability of drowning investigations. Thus, aquaporins hold great potential as emerging forensic tools, warranting further research and standardization for their broader adoption in forensic science.

Ethical Clearance : NA

Conflict of Interest : NA

Source of Funding : NA

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Qualitative Detection of milk Adulterants A Qualitative study on milk Adulterants in Capital of Jharkhand, Ranchi India

Muskan kumari

Student of MSC Forensic Science Delhi University Department of Anthropology

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Abstract

Introduction: Food is essential for sustenance of life. Adulteration of food cheats the consumer and can pose serious risk to health in some cases. major food adulteration and contamination events seems to occur with some regularities, such as the widely publicized adulteration of milk products with melamine and the recent microbial contamination of vegetables across Europe for example. With globalization and rapid distribution systems, these can have international impacts with far-reaching and sometimes lethal consequences. These events, through potentially global in the modern era, are in fact far from contemporary and deliberate adulteration of food products is probably as old as the food processing and production system themselves. Food forensic is a sub field of forensic chemistry which is used to detect when it is contaminated or adulterant, it helps to check the purity of the food item. Food adulteration is the most leading fraud which is growing rapidly.

Methodology: The study was conducted in several stages to assess the presence of adulterants in the milk samples collected from different local areas of Ranchi. the methodology involved the following steps such as sample collection, presumptive testing, documentation and data recording, Analysis, interpretation using forensic chemistry principles.

Results: The qualitative analysis of 11 different brands of milk samples collected from different regions of Ranchi revealed a significant presence of hydrogen peroxide, starch, sodium chloride etc. the presumptive tests detected multiple adulterants that pose considerable health risk to consumers.

Conclusion: The study demonstrate that milk adulteration is a prevalent and alarming issue in the city of Ranchi. It also emphasizes the urgent need for regulatory bodies, public health authorities, and forensic science professionals to work collaboratively to ensure the availability of safe and unadulterated milk for all consumers.

Keywords: Adulteration of food, globalization, and rapid distribution system, adulterants, forensic chemistry presumptive

Introduction

Forensic chemistry is a branch of forensic science which deals with the principle of chemical techniques to aid in investigation agencies and law Enforcement, in one such application food forensic is a field of study under the forensic chemistry division and deals with the detection and identification of

illegalities in relation to food products or items of local consumptions. Forensic studies with regards to food have not been widely popular especially in India, were most of the cases of adulteration and testing are carried out by the food and drug Administration. The study is carried to analyse the purity of milk available in market both branded and local¹.

Milk

Milk is nutrient rich, white liquid food produced by the mammary glands of mammals. Milk is considered as nearly perfect which is readily digested and absorbed.

Milk is very important due to its special nutritive value and important role for human and animal health. It is chemically very complex system containing more than one lakh substances. physically milk is heterogenous mixture of various components, present either in solution, suspension or emulsion in water. The major constituents of milk include fat, casein and proteins the major proteins of milk, lactose- milk sugar and ash - minerals matter of milk².

Main constituent	Range	Mean (%)
Water	85.5-89.5	87.0
Total Solids	10.5-14.5	13.0
Fat	2.5-6.0	4.0
Protein	2.9-5.0	3.4
Lactose	3.6-5.5	4.8
Minerals	06-0.9	0.8

Adulteration

In general adulteration may be defined as any addition of the legally prohibited substances into or from a more valuable genuine product. whereas adulteration of milk is defined as "any change caused in the normal levels of milk constituents is known as adulteration of milk. The change may be brought about by addition of some foreign matter to milk or by removing some more valuable ingredients out of it." Some of the cause of adulteration in milk are

demand and supply gap, physical nature of milk, degraded moral of the society, spoiled economic structure, perishable nature of milk, this unorganized condition of dairy industry, lack of strict and effective regulatory system and lack of suitable, rapid and sure tests³.

One of the important reasons for wide spread adulteration of milk sits physical and chemical nature, due to which it can hide many things when added to it. Thus, milk can be easily and many ways that affect the quality of dairy products manufactured out of it. In the milk mainly two groups of substances are added adulterating agents, whose purpose is to increase the economical yield and preservatives, which delay or inhibit the spoilage of the milk. addition of cheaper material like water and skimmed milk powder is one of the most common ways of adulteration. water is added for increasing volume of milk while the skim milk powder is added to increase the SNF content of milk. starch, glucose, gelatine, glucose, malt dextrin and cellulose are added in milk as thickening agent as well as to increase the SNF content of milk. some miscellaneous adulterants are also added in milk and milk product such as vegetable oil, soybean protein, antibiotics detergents etc⁴.

Adulterants is one of the major problems that stands against the progress of dairying in India and has detrimental effect on export of our dairy products. A worrying 79% of branded or loose milk available in the market is adulterated, the latest annual report by the consumer guidance society of India (CGSI) has found. The report said of all the milk samples 73 were branded packets and only 11 of them (15%) adhered to the standard mark. The remaining 85% of the branded milk currently sold in the market was adulterated⁵.

Milk standards as per FSSAI (current as of 2025)²

S. no	Parameters	Standards limit	
		FSSR 2011	FSSR2011, AMENDED 2018
1	fat	0.5-6.0%	0.5-6.0%
2	SNF	8.5-9.0%	6.0-9.0%
3	Vegetable oil/fat	Negative	negative

Continue....

4	sugar	negative	negative
5	glucose	negative	Negative
6	urea	700mg/kg	Negative
7	starch	negative	Negative
8	detergent	negative	negative
9	Hydrogen peroxide	negative	negative
10	Maltodextrin	negative	negative

Adulteration of Milk in India

In 2019, the national survey on adulteration confirmed that about 68% of milk did not match the standards of food safety and FSSAI different surveys are carried out to detect the adulteration of milk in different cities of India .as a result about 80 to 90 of milk is found to be adulterated. So, there is a need for strict analytical control for detection of adulteration in milk to safeguards the citizens health. There are

number of instrumental methods, test kits, physical and chemical method for detecting the adulterants present in milk. For an example ammonium salt can be detected by various tests like Nessler’s reagent test, phenol test and turmeric test. For the detection of detergent methylene blue test is commonly done⁶. Detection of sulphate can be done by barium chloride test. Some of the quantitative tests for detecting the adulterants present in milk are described in methodology section.

Key Recent FSSAI Updates (2023–2025)

Initiative	Highlights
“Food Safety on Wheels” (March 2023)	FSSAI deployed 168 mobile testing vans equipped with Milk-o-Screen systems to conduct on-the-spot testing of milk samples for constituents like fats, SNF, protein, and adulterants such as water, urea, sucrose, maltodextrin, ammonium sulphate.
Festive Season Surveillance Intensified	During major festivals, FSSAI has repeatedly directed States/UTs to ramp up surveillance of dairy products – including sweets, milk, paneer, ghee, etc.
SOPs for Dairy Producers (Oct 2024)	New standard operating procedures for cleanliness, animal health, milking hygiene, and milk handling/storage (e.g., transportation within 3–4 hours or refrigerated at 4–6 °C) were introduced to reduce contamination risk.
Clarifications on Additives (Oct 2023)	FSSAI clarified that protein binders are not permitted in milk and milk products – only additives listed in Appendix A of FSS (Food Products Standards and Food Additives)
Labelling and Testing Protocols	Drafts for enhanced labelling (e.g., mandatory milk logos, prominent nutritional info), faster lab timelines (within 14 days), and adoption of international methods (AOAC, ISO, Codex) have been issued.
Ongoing Surveys	A large-scale survey across 766 districts (over 10,000 samples) was launched by FSSAI in 2023 to assess compliance of milk and dairy products, with findings expected in reports to the Health Ministry.

Methodology

The study is planned to determine the qualitative test used for the detection of adulterants in milk. The materials and methodology used in this detection technique are mentioned as follows:

List of Chemicals

Phenol, Phenolphthalein, Sodiumbicarbonate, Sodiumhydroxide, sulphuricacid, Chloroform, Diethylether, Iodine solution Nessler'sreagent, Nitric acid methylene blue etc.

Apparatus and Glassware

Beaker, Conicalflask, Measuringcylinder, Testtube, Glassrod, Funnel, Pipettes, Spiritlamp, Waterbath, Centrifuge Refrigerators etc.

Analytical Techniques for Detection of Adulteration in Milk

Various measures are taken to contain the menace of adulteration; including regulatory monitoring, enhancing awareness in the communities, extensive testing/analysis of raw milk, etc. Analysis is one of the essential parts of overall quality assurance system operated by dairy plants. Various analytical methods are used for the purpose of checking adulteration of milk including physical methods, instrumental methods and chemical methods.

Physical Methods

Methods based on physical properties of milk are density (lactometer reading), freezing point, refractive index, etc, which are easy to perform, but can be very easily manipulated due to natural variations in milk composition. Physical methods are simple, fast, easy, cheap and convenient. However, sensitivity of these tests is less in comparison to chemical and instrumental methods⁷.

Freezing point can be significantly affected by seasonal and regional factors. Thus, considering geographical vastness of India and consequent seasonal and regional variation it cannot be a reliable means of adulteration detection.

The density (or specific gravity) depends on composition, temperature and temperature history of milk. As Indian dairy sector is still predominantly unorganized in nature; it is difficult to control most parameters affecting density. Therefore, density measurement cannot be a useful tool for adulteration detection. Thus, physical methods suffer from some of the general limitations due to large natural variations, lower sensitivity, poor specificity, proneness to manipulation⁸.

Instrumental methods are one of the good options for quality control of milk and milk products. Though, it possesses several advantages like higher sensitivity, high specificity and reliability, it also suffers from several limitations as described below.

- Very limited adoptability for practical applications.
- Requires high initial investment, operational cost and expensive maintenance.
- Most methods are time consuming as it necessitates isolation, purification, concentration &/or derivatization of the target analyte.
- Impractical for routine analysis and field applications.

As these methods require skilled manpower it is difficult to use as routine methods. Indian dairy industry is still characterised by small scale farming and small cooperative societies which may not be able to afford capital requirements for sophisticated instruments as well as its maintenance.

Chemical Methods

The chemical methods are simple, fast, easy, cheap, convenient and have better specificity for adulterant/ chemical compounds being tested. However numerous qualitative tests are reported for detection of adulterants in milk with wide variation in procedure for a given test. There is lack of information regarding sensitivity between various reported qualitative tests. To overcome these limitations various reported tests and procedural variations were evaluated⁹.

Qualitative Tests for Detection of Adulterants in Milk

Upon review and evaluation of qualitative tests available for detection of common adulterants reported in milk, it was observed that there is wide variation related to several test performance parameters like sensitivity, convenience, cost etc. This variation in performance was mainly attributable to variations in the procedures of the test. Further, it also appeared from the survey of literature that scant attention has been paid on systematic work for improving performance of the qualitative tests for detection of adulterants in the milk. Thus, it was envisaged to undertake work for improving some qualitative tests suggested for detection of common adulterants encountered in milk. The qualitative tests were optimized considering three different aspects of the test procedure.

1. To select suitable medium for performing the tests in detection of adulterants.
2. To standardize various chemicals/reagents used in the tests.
3. To optimize different conditions involved in performing the tests.

Considering the requirements for improving the performance of existing qualitative tests reported for common adulterants, including detergent, urea, ammonium salts, glucose, sucrose, maltodextrin, starch, hydrogen peroxide, salt, nitrate, sulphate, formaldehyde and neutralizers were modified¹⁰.

Detection of Detergents by Methylene Blue Test

Methylene blue is cationic dye which forms complex with anionic detergents. It is normally water-soluble compound; however, it shows affinity for anionic detergents, if they are present. In this method, detergent is first extracted in chloroform and then methylene blue solution is added. In presence of detergent blue colour is developed in chloroform layer of the sample, whereas blue colour is observed in milk layer in control (pure milk). Chloroform is heavier settles at the bottom.

This implies that observation of blue colour in the bottom layer indicates presence of detergents¹¹.

Reagents

1. Methanol (AR)
2. Methylene blue solution: 12.5 mg methylene blue (AR) is dissolved in 100 ml of distilled water. Protect the solution from direct sunlight.
3. Chloroform (AR):

Precaution: Inflammable and toxic on inhalation. Mouth pipetting is not recommended

Procedure

1. Take 2.5 ml of suspected milk sample in a test tube and add 7.5 methanol.
2. Filter the content through Whatman No. 1 filter paper.
3. Take 2 ml filtrate in a test tube.
4. Add 2 ml of methylene blue solution and shake well.
5. Subsequently add 4 ml chloroform and shake well again.
6. Allow the chloroform layer to separate.
7. Compare the colour extracted in the chloroform layer in suspected milk with that for pure milk.

Interpretation: If the methylene blue colour extracted from a suspected sample into the chloroform layer is darker than that extracted from pure milk sample, it indicates the presence of detergent in milk.

Limit of detection: 0.02 g/ 100 ml milk

(**Note:** The method reported by Varadkaret *al.* (2000b) is modified. Methanol was used in place of ethanol. Methylene blue concentration was reduced from 25 mg/100 ml to 12.5 mg/100 ml.) **Benefits of modified method over reported method**

- Better differentiation between adulterated and pure sample
- Eliminates use of ethanol (a regulated chemical)

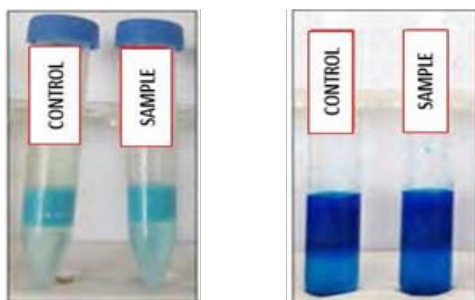


Figure 1: Test of Detergents

Detection of Urea By Dmab Test

A yellow-coloured complex is formed between urea and *p*-dimethyl amino benzaldehyde (DMAB) reagent in low acidic alcoholic solution at room temperature. The intensity of colour can be measured at 440 nm (Leafier, 1996). The colour developed is in proportion to urea content in the sample

Reagents

Dimethylaminobenzaldehyde reagent (DMAB): The reagent is prepared by dissolving 1.6 g of *p*-dimethylamine benzaldehyde (AR) in methanol (AR) subsequently adding 10 ml of concentrated HCl (AR) and making volume to 100 ml with methanol.

Procedure

1. Take 5 ml milk in a test tube.
2. Add 5 ml DMAB reagent.
3. Mix the content and observe the colour.

Interpretation: Formation of distinct yellow colour indicates the presence of added urea in milk sample. Pure milk shows light yellow colour due to natural urea.

Limit of detection: 0.2 g/ 100 ml milk

Benefits of modified method over reported method

- Better differentiation between adulterated and pure sample.
- Eliminates use of ethanol (a regulated chemical).



Figure 2: Test of Urea

Detection of Ammonium Salts By Nessler's Test

Nessler's test is one of the classical methods for qualitative and quantitative analysis of ammonia and ammonium ions. Nessler's reagent is an alkaline solution of potassium mercuric iodide (K_2HgI_4). On reaction with ammonium ion Nessler's reagent produces a yellowish-brown colour. The intensity of the colour is directly proportional to the amount of ammonia/ammonium ion present¹².

Reagents

- 1) Nessler's reagent: Dissolve the following chemicals separately.
 - a) 8.0 g of mercuric chloride (AR) in 150 ml distilled water.
 - b) 60.0 g of sodium hydroxide (AR) in 150 ml distilled water.
 - c) 16.0 g of potassium iodide (AR) in 150 ml distilled water.

Add reagent „a“ to reagent „b“ and mix well. To this mixture, add reagent „c“, mix and dilute the contents to 500 ml with distilled water. Leave this solution undisturbed and decant the clear upper layer of the solution. Store in a stoppered amber glass bottle.

Note: Alternatively, commercially available (readymade) Nessler's reagent can also be used.

- 2) Citric acid solution (5%): Dissolve 5 g citric acid monohydrate (AR) in distilled water and make up the volume to 100 ml with distilled water.

Procedure

- 1) Take 20 ml milk in a conical flask.
- 2) Warm the milk to 70-80 °C either on direct flame or water bath.
- 3) Add 5% citric acid solution drop wise in the milk with gentle stirring till visible coagulation occurs. Flask if stirred vigorously will result in fine curd particles and which may impact colour observation.
- 4) Filter the content using Whatman No. 1 filter paper.
- 5) Take 5 ml of filtrate into a test tube.
- 6) Add 0.4 ml of Nessler's reagent.
- 7) Observe the colour without shaking the test tube.

Interpretation: Carefully observe instant development of orange colour in milk adulterated with ammonium salts. Whereas pale yellow colour indicates unadulterated milk.

Limit of detection: 0.02 g/ 100 ml milk

Benefits of modified method over reported method.

- Improved sensitivity over the reported method.
- Better differentiation between adulterated and pure sample.

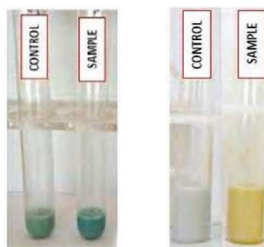


Figure 3: Test for Ammonium Salts

Detection of Sucrose by Seliwanoff's Test

Sucrose is a disaccharide containing glucose and fructose (a ketose sugar). Seliwanoff test is used for detection of ketoses. The dilute hydrochloric acid used in Seliwanoff reagent along with heat leads to hydrolysis of sucrose and subsequent dehydration of fructose. Further keto group more actively attacks

resorcinol in comparison to aldehyde group. The dehydration product 5-hydroxymethylfurfural condenses with resorcinol forming cherry red colour. Ketoses react rapidly in comparison to aldoses because dehydration of aldoses to 5-hydroxymethylfurfural proceeds in a much slower way than the reaction of ketoses¹³.

Reagents

Resorcinol solution (0.05%): The reagent is prepared by dissolving 0.05 g of resorcinol (AR) in 100 ml hydrochloric acid (The acid is prepared by taking 30 ml conc. HCl and diluting to 100 ml with distilled water.)

Procedure (using milk as medium)

- 1) Take 3 ml milk and 5 ml resorcinol solution in a test tube.
- 2) Keep the content in boiling water bath for 6 min.
- 3) Cool the tubes immediately after heating under tap water to retard the rate of reaction, which if not done would narrow the colour difference between negative and positive samples.
- 4) Observe for colour development.

Procedure (using whey as medium):

- 1) Take 3 ml milk and 5 ml resorcinol solution in a test tube. (Quantity of both milk and reagent can be doubled proportionately to get sufficient filtrate.)
- 2) Mix and filter the content using Whatman No. 1 filter paper.
- 3) Keep the filtrate in boiling water bath for 4 min.
- 4) Cool the tubes immediately after heating under tap water to retard the progress of reaction, which if not done would narrow the colour difference between negative and positive samples.
- 5) Observe for colour development.

Interpretation: Development of red colour indicates adulteration of sucrose in milk. The intensity of red colour increases with increase in the sucrose content in the milk. Pure milk remains light in colour.

Limit of detection:

0.1g/ 100 ml milk (When test is performed in milk)

0.06 g/100 ml milk (When test is performed in whey)

Benefits of modified method over reported method

- Better differentiation between adulterated and pure sample.

Different concentration of hydrochloric acid (30 ml conc. hydrochloric acid diluted to 100 ml) was used instead of reported concentration (1-part HCl: 2 parts distilled water). The test was also modified using whey as medium.



Figure 4: Test for Sucrose

Detection of Glucose by Barfoed Test

Barford's test is routinely used for detection of extraneous glucose in milk. By means of the Barford's reaction it is possible to differentiate reducing monosaccharides from reducing disaccharides as monosaccharides can reduce copper fast enough in comparison to disaccharides. The formation of green, red, or yellow precipitate is a positive test for reducing monosaccharides¹⁴.

Reagents

Barfoed reagent: Dissolve 13.3 g of copper acetate (AR) in distilled water, subsequently add 2.0 ml of lactic acid (AR) and make up the total volume to 200 ml.

Procedure (using milk as medium)

- 1) Take 1 ml milk in a test tube.
- 2) Add 2 ml Barfoed reagent.
- 3) Keep the test tube in boiling water bath for 6 min.
- 4) Cool the test tube to room temperature under tap water.
- 5) Observe for colour development.

Procedure (using whey as medium)

- 1) Take 1 ml milk and 2 ml Barfoed reagent in a test tube (quantity of both milk and reagent can be doubled proportionately to get sufficient filtrate).
- 2) Mix the content and filter using Whatman No. 1 filter paper.
- 3) Keep test tube containing filtrate in water bath for 4 min.
- 4) Cool the test tube to room temperature using water.
- 5) Observe for colour development.

Interpretation: Development of green colour indicates presence of glucose in milk.

Limit of detection:

0.1 g/100 ml of milk (milk as medium)

0.15 g /100 ml of milk (whey as medium)

Benefits of modified method over reported method -better differentiation between adulterated and pure sample.

- Better clarity in whey as compared to milk.
- Eliminates large number of costly chemicals.
- One step procedure, improved convenience.

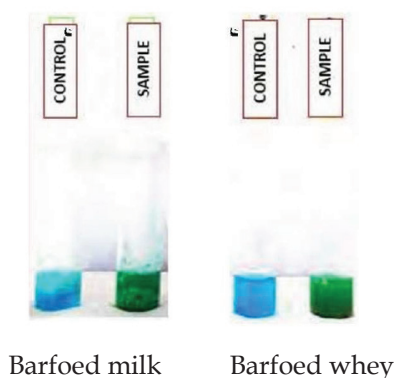


Figure 5: Test for Glucose

Detection of Maltodextrin by Iodine Test

Similar to starch maltodextrin also forms complex with iodine. However, instead of blue colour, red to brown colour is observed. This is because the complex formed is dependent on chain length of dextrin's. The chain length more than 45 DP (degree of polymerization) gives blue colour, whereas lesser DP gives red to brown colour complex, as in the case of maltodextrin¹⁵.

Reagents: Iodine solution (1%): Dissolve 2.5 g potassium iodide (AR) in 100 ml distilled water. Then, add 1 g pure iodine crystals (AR). Prepare iodine solution at least a day before as iodine dissolves slowly.

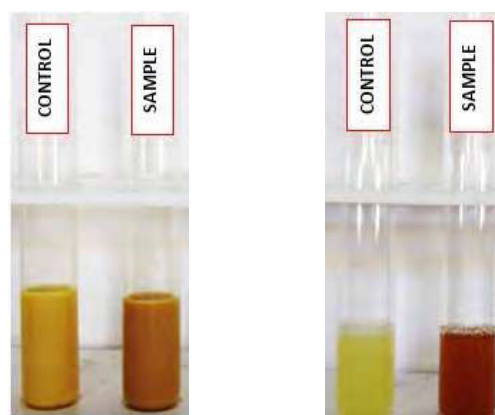
Procedure

- 1) Take 20 ml milk in a conical flask.
- 2) Warm the milk to 70-80
- 3) Add 5% citric acid solution drop wise in the milk with gentle stirring until clear coagulation occurs (Approximate consumption of citric acid required would be 1.5-2 ml.). Flask, if stirred vigorously will result in fine curd particles and which may impact colour observation.
- 4) Filter the content using Whatman No. 1 filter paper.
- 5) Take 5 ml filtrate in a test tube.
- 6) Add 0.25 ml of 1% iodine solution.
- 7) Mix the content and observe the colour.

Interpretation: Development of red-brown colour indicates adulteration of milk with maltodextrin. Pure milk remains yellow in colour. **Limit of detection:** 0.1 g / 100 ml milk.

Benefits of modified method over reported method

- Better differentiation between adulterated and pure sample.
- Significant improvement in sensitivity.



Iodine Test (Whey)

Figure 6: Test for Maltodextrin

Detection of Starch by Iodine Test

The development of blue colour on addition of iodine solution in starch containing milk is due to complex formation between iodine and amylose component of starch. The other component, amylopectin, gives a red-purple colour which is much less intense than the amylose. The acidic condition in the reagent mixture accentuates the blue colour, whereas alkali reduces its intensity, the blue colour disappears above a pH of about 9.5. Heating the solution containing starch-iodine complex also destroys the colour although reversibly.

Reagents

- 1) Iodine solution: Dissolve 2.5 g potassium iodide (AR) and 1 g of pure iodine crystals (AR) in 100 ml distilled water. Prepare iodine solution at least a day before as iodine dissolves slowly.

- 2) Acetic acid (10%): Dissolve 10 ml glacial acetic acid (AR) in distilled water and make up the volume to 100 ml.
- 3) Citric acid solution (5%): Dissolve 5 g citric acid monohydrate (AR) in distilled water and make up the volume to 100 ml with distilled water.

Procedure (using milk as medium)

- 1) Take 3 ml milk in test tube.
- 2) Bring the milk to boil on a direct flame or on a boiling water bath.
- 3) Cool the test tube to room temperature under tap water.
- 4) Add a drop of 10% acetic acid in the test tube.
- 5) Add 0.2 ml of iodine solution.
- 6) Mix the content and observe colour.

Procedure (using whey as medium)

- 1) Take 20 ml milk in a conical flask.
- 2) Bring the milk to boil on a direct flame or on a boiling water bath.
- 3) Add 5% citric drop wise till visible coagulation. (Approximate consumption of citric acid would be 1.5-2 ml.)
- 4) Filter the content using Whatman No. 1 filter paper. Let the filtrate cool down to room temperature.
- 5) Take 3 ml filtrate in another test tube and add 0.1 ml of iodine solution.
- 6) Mix the content and observe colour.

Interpretation: Blue/Dark blue colour formation indicates adulteration of milk with starch. Whereas pure milk remains yellow due to colour of iodine.

Limit of detection:

0.02 g/ 100 ml milk (When test is performed in milk)

0.01 g /100 ml milk (When test is performed in whey)

Benefits of modified method over reported method

- Better differentiation between adulterated and pure sample.
- Better sensitivity
- Reduces the chances of interference of neutralizers on the detection of starch.

Detection of Sodium Chloride by Silver Nitrate Test

The chloride ion (Cl) from sodium chloride reacts with silver ion of silver nitrate forming white precipitates of silver chloride. Simultaneously water-soluble sodium nitrate is also formed. After the Ag⁺ from silver nitrate has complexed with all the available chloride in the sample, the Ag reacts with chromate from silver chromate added in the reaction mixture; forming an orange-coloured precipitates of silver chromate.

Reagents

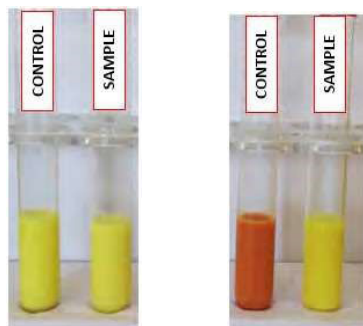
1. Silver nitrate solution (0.1N): The reagent is prepared by dissolving 16.987 g silver nitrate (AR) in 1000 ml distilled water.
2. Potassium chromate solution (5%): The reagent is prepared by dissolving 5 g potassium chromate (AR) in 100 ml distilled water.
Procedure:
3. Take 5 ml milk in test tube.
4. Add 0.5 ml of 5% potassium chromate solution.
5. Add 2 ml 0.1 N silver nitrate and mix the contents.
6. Observe for colour change.

Interpretation: Yellow colour indicates adulteration of milk with common salt (sodium chloride). Unadulterated milk gives chocolate or reddish-brown colour.

Limit of detection: 0.04g /100 ml of milk

Benefits of modified method over reported method

Better differentiation between adulterated and pure sample.



Chromate Test (Milk)

Figure 7: Test for Salt (Sodium Chloride)

Detection of Nitrate by Diphenylamine Test

The test consists of adding a solution of diphenylamine in sulphuric acid to milk. Nitrates are considered as oxidising agent. Under the conditions of test, diphenylamine is oxidized by nitrate to the intensely blue quinone-ammonium salt *by* diphenyl benzidine.

Reagents

Diphenylamine solution: The reagent is prepared by dissolving 0.085 g diphenylamine in 50 ml distilled water and gradually 450 ml of concentrated sulphuric acid is added with constant stirring. During preparation of reagent the content is kept cool by dipping in cold water. Diphenylamine solution should be prepared freshly and shall be colourless.

Acetic acid (10%): Take 10 ml glacial acetic acid (AR) in 100 ml volumetric flask and make up the volume with distilled water.

Procedure

- 1) Take 20 ml milk in a conical flask.
- 2) Warm the milk to 70-80 C either on direct flame or boiling water bath.

- 3) Add 10% acetic acid drop wise in the milk with gentle stirring till visible coagulation.
- 4) Filter the content using Whatman No. 1 filter paper.
- 5) Take 2 ml diphenylamine solution in a test tube.
- 6) Add 1 ml filtrate in test tube containing diphenylamine solution.
- 7) Observe for ring formation at the junction of two solutions.

Interpretation: Formation of blue ring at the junction of two solutions indicates adulteration of milk with nitrate or surface water.

Limit of detection: 0.002g / 100 ml milk

Benefits of modified method over reported method

- Better differentiation between adulterated and pure sample.
- Elimination of toxic chemicals from precipitating reagent.

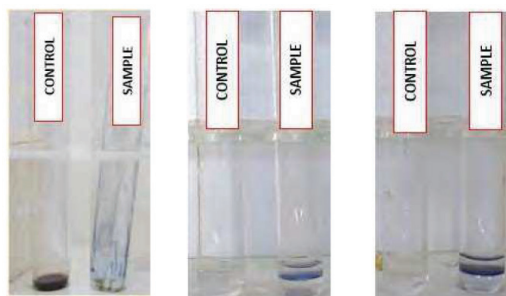


Figure 8: Test for Nitrate

Detection of Sulphate by Barium Chloride Test

Barium chloride test is one of the commonly used methods for detection of sulphate adulteration in milk. The barium ion (Ba^{2+}) reacts with sulphate ion (SO_4^{2-}) to give white precipitates of barium sulphate ($BaSO_4$). Using this test, presence of added sulphate like ammonium sulphate, sodium sulphate, zinc sulphate, magnesium sulphate, etc. to milk can be confirmed by observing milky-white precipitates.

Reagents

- 1) Barium chloride solution (5%): Dissolve 5.0 g barium chloride (AR) in distilled water and make up the final volume to 100 ml.
- 2) Lactic acid solution (5%): Take 5 ml lactic acid (AR) in 100 ml volumetric flask and make up the volume with distilled water.

Procedure

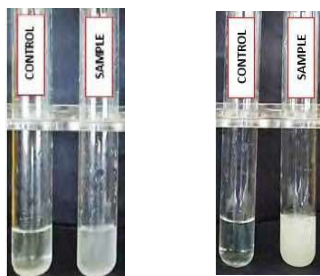
- 1) Take 20 ml milk in a conical flask/test tube bring it to boil on direct flame.
- 2) Add 2 ml of 5% lactic acid in hot milk and filter the content using Whatman No. 42 filter paper.
- 3) Take 2 ml filtrate in a separate test tube.
- 4) Add 0.2 ml of 5% barium chloride and observe for turbidity development.

Interpretation: Formation of turbidity after barium chloride solution addition indicates adulteration of milk with sulphate.

Limit of detection: 0.015 g /100 ml milk

Benefits of modified method over reported method -better differentiation between adulterated and pure sample.

- Improved sensitivity.
- Elimination of TCA (hazardous chemical).



Barium Chloride Test Barium Chloride Test

Figure 9: Test for Sulphate

Detection of Hydrogen Peroxide by Iodine Test

Hydrogen peroxide presence can be detected iodometric test. Hydrogen peroxide oxidises iodide

to iodine. Starch forms a deep, dark blue complex with minute amounts of triiodide ions that are formed only in the presence of both iodine and iodide in solution. Thus, formation of blue colour indicates presence of hydrogen peroxide.

Reagents

1. Potassium iodide solution (20%): Weigh 20 g of potassium iodide (AR) and dissolve it in distilled water to obtain 100 ml solution. The solution should be prepared fresh before every use.
2. Starch solution (1%): Take 1 g of soluble starch and make paste using cold water. Transfer the paste to 100 ml volumetric flask and make the volume to 100 ml using boiling distilled water. Cool and decant the clear solution. The solution should be prepared fresh before every use.
3. Starch-potassium iodide reagent: The reagent is prepared by mixing equal volumes of 20 per cent potassium iodide solution and 1 per cent starch solution. The solution should be prepared fresh before every use.
4. Acetic acid (10%): Dissolve 10 ml glacial acetic acid in water and make up the volume to 100 ml.

Procedure

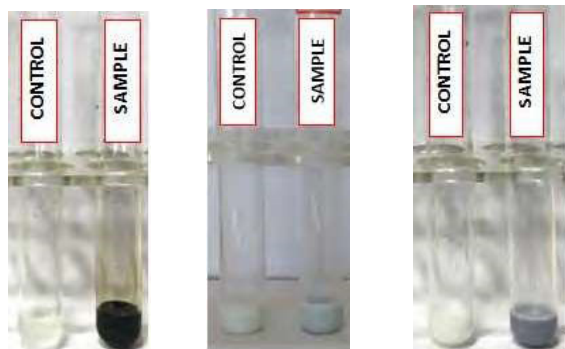
1. Take 20 ml milk in a conical flask.
2. Add 2 ml 10% acetic acid.
3. Mix the content and filter using Whatman No. 1 filter paper.
4. Take 1 ml filtrate and add 1 ml starch-potassium iodine reagent.
5. Observe for colour development.

Interpretation: Appearance of bluish black colour indicates the presence of hydrogen peroxide in the milk sample whereas control milk sample remains colourless.

Limit of detection: Modified test: 0.015 g /100 ml of milk

Benefits of modified method over reported method Better differentiation between adulterated and pure sample.

- Improved sensitivity.
- Method uses relatively benign chemicals in place of potentially harmful chemical paraphenylenediamine.



phenylenediamine Iodometric Iodometric Test

Figure 10: Test for Hydrogen Peroxide

Detection of Formaldehyde by Hehner Test

In the Hehner test for detection of formaldehyde in milk, concentrated sulphuric acid and ferric chloride is used. The test is an aldehyde-oxidation reaction of an aromatic amine. Ferric chloride acts as oxidising agent for formaldehyde. The formaldehyde reaction depends on the presence of the tryptophan in the protein molecule. The violet colour develops as a result of the reaction of oxidised formaldehyde with tryptophan. The intensity of the reaction with different proteins varies in direct proportion to the amount of tryptophan present in the protein molecule.

Reagents

- Ferric chloride (10%): Take 10 g of ferric chloride (AR) in 100 ml volumetric flask and mark the volume with distilled water.
- Sulphuric acid (80%): Add 80 ml concentrated sulphuric acid (AR) into 20 ml distilled water.

Procedure

- 1) Take 5 ml milk sample in a test tube.
- 2) Add 5 ml distilled water and 0.1 ml of 10% FeCl_3 solution.
- 3) Mix the content and add 10 ml H_2SO_4 (80%) from the side of the test tube.

Interpretation: Violet ring at the junction of two layers indicates presence of formaldehyde.

Limit of detection: 0.0005 ml formalin/ 100 ml milk

Benefits of modified method over reported method - better differentiation between adulterated and pure sample.

Improved sensitivity.

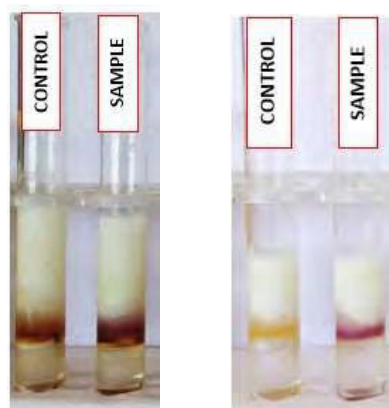


Figure 11: Test for Formaldehyde

Detection of Neutralizers by Phenol Red Test

Phenol red is used as a pH indicator and shows colour transition from yellow to red over the pH range 6.8 to 8.2. However, at pH value greater than 8.2 phenol red turns bright pink. As neutralization increases the pH of milk (beyond 6.8), it can be detected using phenol red indicator.

Reagents

Phenol red solution: 0.05 g of phenol red is dissolved in 20 ml ethanol and volume is made up to 100 ml using distilled water.

Procedure

- 1) Take 4 ml milk in a test tube.
- 2) Add 1 ml phenol red solution.
- 3) Mix the content and observe the colour.

Interpretation: Development of pink colour indicates presence of sodium hydroxide (NaOH), whereas orange colour indicates sodium carbonate (Na_2CO_3) or sodium bicarbonate (NaHCO_3) in milk. Milk without added neutralizers shows yellow colour.

Limit of Detection:

NaOH: 0.04 g/ 100 ml milk

Na_2CO_3 : 0.08 g/ 100 ml milk

NaHCO_3 : 0.2 g/ 100 ml milk

Benefits of developed method over reported method -better differentiation between adulterated and pure sample.

- Improved sensitivity.
- Elimination of ethanol (a regulated chemical)

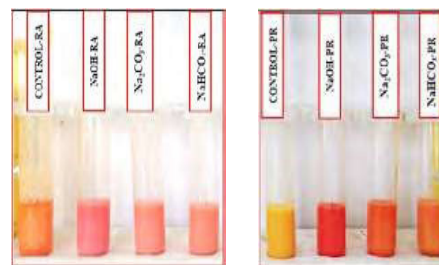


Figure 12: Test for Neutralizers

Result

S. NO	MILK BRAND SAMPLE	ADULTERANTS					
		detergent	urea	Sodium chloride	Hydrogen peroxide	starch	sucrose
Positive spot result		Blue	Intense yellow	yellow	Bluish black	Dark blue	red
1.	MILKY KOOL	X	X	X	✓	X	X
2.	OSEM DIARY	X	X	X	X	X	X
3.	BERRY DIARY	X	X	✓	X	X	X
4.	SUDHA MILK	X	X	X	X	X	X
5.	MEDHA	X	X	X	X	✓	X
6.	AMUL	X	X	X	X	X	X
7.	W-ORGANIC	X	X	X	✓	X	X
8.	MOTHER DAIRY	X	X	X	X	X	X
9.	ORGANIC	X	X	X	X	✓	X
10.	DAIRYBEST	✓	X	X	X	X	X
11.	OMFED	X	X	X	X	X	X

Discussion

There are numerous variations are observed in particular qualitative tests used for detection of specific adulterants. therefore, it is important

from the public health view point to evaluate the detection of adulterants in milk. moreover, the presence of adulterant in milk is a major challenge for the country. The supplies of milk appear to have found three ways to increase their margin or profit

from the sale of milk ;(a) dilution (b) extraction of valuable components i.e. milk fat removed as cream (c) a combination of cheaper bulking additives and some chemicals, adulterants and malpractices results in public health concern and malnutrition.

Based on research, it was found that many of the milk samples show the presence of adulterants, which has been shown on the above. For the confirmatory conclusive we can proceed for quantitative techniques like SDS-PAGE, HPLC, IMMUNODIFFUSION METHOD, NIR SPECTROSCOPY.

DAIRY BEST milk sample indicates the presence of detergent, which can lead to gastrointestinal issues, tissue damages, & food poisoning.

BERRY DIARY indicates the presence of sodium chloride (NaCl), excessive amount of NaCl may cause vomiting, hypernatremia, respiratory distress, physio-chemical changes like PH Disbalance.

MILKY KOOL & W-ORGANIC shows the presence of hydrogen peroxide (H₂O₂), it is milk preservative used to inhibit microbial growth and spoilage. Higher exposures may cause a build-up of fluid in the lungs (pulmonary oedema) with severe shortness of breath, nausea and vomiting.

MEDHA & ORGANIC milk sample shows the presence of starch. A higher amount of starch in milk can cause Diarrhoea and other gastrointestinal problems such as gastric and bloating.

After surveying and testing the milk sample, the result was quite shocking. People expected water to be present as an adulterant in their consumed milk but for some cases test indicated the presence of other adulterants that are harmful for health. Therefore, this contradictory information shows that consumers are unaware of these adulteration practices. For tackling this problem government should periodically collect samples and do some test to make sure quality of milk supplied and consumed by people.

Detection of adulteration is complicated as the indicators of adulterants in milk can vary in quality due to various biological, climatic and agronomic factor. Moreover, processing can also dramatically

change the composition of minor constituents, which can be problematic in setting the specification ranges for various inspection methods. Thus, the challenge is to develop simple and cost-effective techniques for detecting adulteration in milk, which could be used with high degree of responsibility.

Conclusion

The findings of this study underscore the widespread and alarming issue of milk adulteration in Ranchi, with multiple samples showing the presence of harmful substances such as hydrogen peroxide, starch, and sodium chloride. These adulterants pose significant health risks to consumers, highlighting the urgent need for effective monitoring and strict regulatory measures. The application of forensic chemistry has proven to be a valuable tool in identifying food adulterants and ensuring food safety. Collaborative efforts between public health authorities, regulatory bodies, and forensic experts are crucial to curb the growing threat of food fraud and to guarantee the distribution of safe and unadulterated milk to the public.

Ethical Clearance : NA

Conflict of Interest : NA

Source of Funding : NA

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Digital Footprint of Crime: Exploring The Relationship between Social Media and Criminal Behaviour

¹Navin T S, ²Vaishnavi Vivek Sawant, ²Anisha Kudaskar and ³Theju Kumar C

¹Student, Department of Forensic Science Acharya Institute of Graduate Studies, Bengaluru, Karnataka, ²Assistant Professor, Department of Forensic Science & Criminology, Acharya Institute of Graduate Studies, Bengaluru, Karnataka, ³HoD, Department of Forensic Science & Criminology, Acharya Institute of Graduate Studies, Bengaluru- Karnataka

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Abstract

Social media has laid a strong foundation in everyone's daily life. It not only reshaped our communication style but also our thoughts and routine activities. This research paper explores the relationship between social media and criminal behaviour. The digital footprints can include messages, shared media, location check-ins and interactions that can be traced and analysed for behavioural pattern. These digital traces have emerged as crucial tools in both committing and solving crimes. Digital platforms provided new pathway to engage in criminal activities. The accessibility and reach of platforms like Facebook, Instagram, and Snapchat allow criminals to operate anonymously, target victims globally, and coordinate illegal activities discreetly. Social media can also play a role in the radicalization process, where individuals are exposed to extremist content, misinformation, or ideological grooming within online communities. By the results obtained through survey and research articles we found that social media represents a powerful double-edged sword (around 83%), it plays a major role in both construction and destruction of criminal behaviours. The digital footprint behaviour is the vital resource for crime detection and prevention. By knowing this relationship it will be an effective tool for developing criminal justice responses, digital literacy education, and ethical guidelines in the digital era. As technology continues to evolve, so too must our approaches to managing its impact on criminal behaviour and law enforcement practices.

Keywords: Footprint, crime, criminal, social, media, Forensic, Science.

Introduction

It is a digital era, social media is a platform to communicate, share information and express views of the people. This has both positive and negative impact on the environment. This platform is used to disseminate important information and facilitates global communication. The news shared in social media is not restricted for people of certain age groups, most of the platforms are open for all. Platforms such as Facebook, Instagram, Twitter,

and TikTok have not only reshaped social dynamics but also influenced patterns of criminal behaviour. It created opportunities for criminal activity and antisocial conduct. Social media platforms, designed to facilitate social interaction and community building, have become breeding grounds for illicit behaviour, including cyberbullying, harassment and exploitation. Social media allows to documenting and broadcasting criminal activities, either to assert dominance or gain social validation. As social media continues to permeate every aspect of our lives, it

is essential to understand its impact on criminal behaviour and implications for law enforcement, policymakers and individuals. The link between social media and criminal activity is both complex and layered. On one hand, social media can be a valuable tool for law enforcement agencies, enabling them to gather intelligence, track suspects, and disseminate information to the public. On the other hand, social media can also facilitate criminal activity, providing a platform for offenders to connect with potential victims, share illicit content, and coordinate illegal activities¹.

Theories Relating Media and Criminology

There are several theories and perspectives that connect media and criminology, analyzing how crime is portrayed, perceived and influenced through media channels. Moral panic theory is a sociological concept that explains how public's fear and anxiety can be disproportionately aroused over an issue perceived to threaten the social order. The term was popularized by Stanley Cohen. The core idea of this theory that relates media and crime is that media exaggerates and sensationalizes events and creates public fear or anxiety about certain groups or behaviour. The late 1960s saw the emergence of cultivation theory, formulated by George Gerbner, shaping the way mass communication is studied. The core idea of this theory is Heavy exposure to television especially crime dramas or news cultivates a distorted view of reality, making viewers believe the world is more dangerous than it actually is. It is related to criminology as it influences the public support for harsh criminal justice policies due to fear of crime. Social Learning Theory was developed by Albert Bandura (1977) it argues that people learn behaviors—including criminal ones—through interaction, observation, and imitation of others, especially when those behaviors appear to be rewarded or go unpunished. When relating this theory to media and crime, users may mimic the criminal behaviour seen in viral content or from the influencers. Normalization of Deviant behaviour via memes, videos, or group chats can encourage participation in criminal acts. Self-Control Theory also known as General Theory of Crime

was developed by Michael Gottfredson and Travis Hirschi (1990). The theory proposes that people who lack self-control have a higher tendency to participate in impulsive and potentially criminal activities. The instant gratification and impulsive nature of social media can trigger criminal behaviours like extortion, cyberbullying, or online fraud. Platforms reward risky behavior with attention especially among youth².

Media Facilitating Criminal Activity

Cybercrime

The crime done using computer system or digital devices is referred as cybercrime. It encompasses a broad range of offenses that target individuals, organisations, and government, often resulting in financial loss or damage of reputation of an individual or organization. As people run towards technology there is emerging threat about cybercrime. Illicit activities in cyberspace often include breaching computer systems, engaging in cyberbullying, committing identity-related offenses, and conducting fraudulent schemes online. Cybercriminals may act as an individual or a group of people work together. They sometimes span across international borders which makes tracking and prosecuting them more difficult. The influence of cybercrime extends across multiple levels, causing substantial harm. Even governments may face threats to national security and individual can experience emotional distress and privacy violations. The rise of the dark web has further fuelled cybercrime by offering a hidden marketplace for illegal activities. Drugs, weapons, fake documents, and stolen data are bought and sold anonymously through encrypted networks. Cryptocurrency is often used in these transactions, further complicating traceability and enforcement³.

Gang Activities

Social media is used by gangs to gather members, plan activities and promote their criminal ventures even without having any direct link between them. It has paved different ways for the criminals to communicate. Social media allows gangs to construct and promote their image by showcasing symbols of

power-such as weapons, drugs, money, and luxury goods-in ways that can attract young, impressionable users seeking identity, respect, or protection. These online conflicts then merge into real world and cause severe damage to both the parties. In sum, social media has become an integral extension of gang life, influencing how gangs recruit, communicate, and commit crimes. Its role in shaping contemporary gang culture underscores the urgent need for innovative, tech-informed strategies in both prevention and policing⁴.

Objective

- To examine how social media contributes to the criminal behavior.
- To create awareness among individual about the risk factors of social media.
- To understand how social media can be used to spread extremist ideologies and develop strategies to counter this.
- To examine the relationship between social media and specific types of crime.
- To inform prevention strategies and interventions to mitigate negative impacts.

Methodology

Methodology refers to the systematic plan, process, or approach used to carry out research. It explains how the research was conducted, including the tools, techniques, procedures, and methods used to collect, analyze, and interpret data.

Aim: To study the relationship between social media usage and criminal behaviour by analysing public perceptions, experiences, and opinions across different age groups, and to assess how social media influences crime rates and criminal activities.

Procedure

1. Research Design

The research was conducted using a survey-based approach combined with a literature review to analyze the relationship between social media usage and criminal behaviour.

2. Primary Data Collection

- A structured questionnaire was designed using **Google Forms** to collect data efficiently and securely from participants across different age groups.
- The questionnaire consisted of both closed-ended and open-ended questions aimed at understanding individuals' social media usage patterns, exposure to crime-related content, and their perceptions regarding the influence of social media on criminal behaviour.

3. Survey Process

- The Google Form link was circulated via social media platforms such as WhatsApp, Instagram, and Email to reach a diverse audience.
- The participants were selected randomly and included individuals from various age groups and backgrounds to ensure a broad perspective.
- Participation in the survey was entirely voluntary, and anonymity and confidentiality were maintained throughout the study.
- Respondents were informed about the purpose of the research, and consent was implied upon submission of the form.

4. Survey Questions Included

- Age
- Frequency of social media usage
- Mental health impact of not using social media for a day
- Preferred social media platforms
- Experience of witnessing crimes shared on social media
- Types of crimes influenced by social media
- Opinion on social media companies' role in preventing crime
- Perception of media influence on crime rates
- Willingness to report criminal activity seen on social media

- Personal influence by criminal activities viewed on social media
- Suggested measures to reduce social media-related crimes

Data Analysis

From the survey conducted through google forms, 187 people responded to it, the bar graph and pie chart results of 1-10 questions are as follows:

1. Age

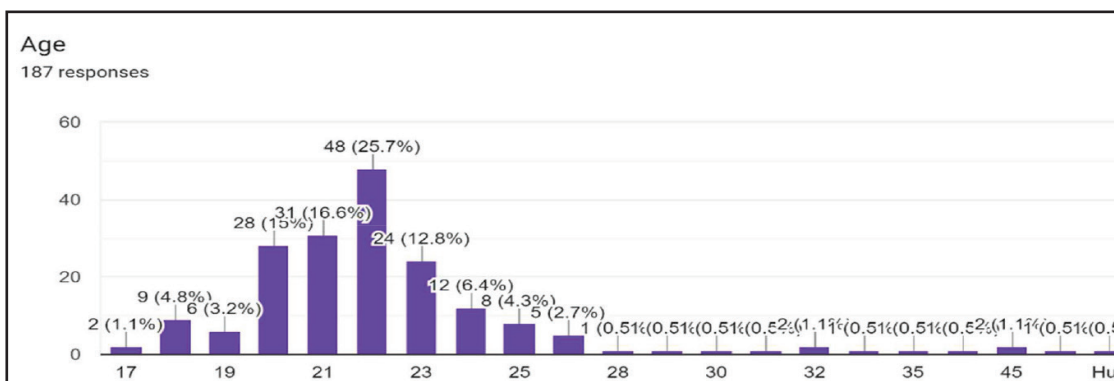


Figure 1: Age of the respondents

The people who attended the survey are under the age group of 17-45.

2. How often do you use social media?

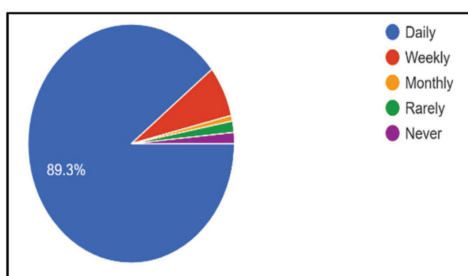


Figure 2: Usage of social media by the respondents

According to study, 89.3% people use social media daily.

3. Does a day without social media affect your mental health?

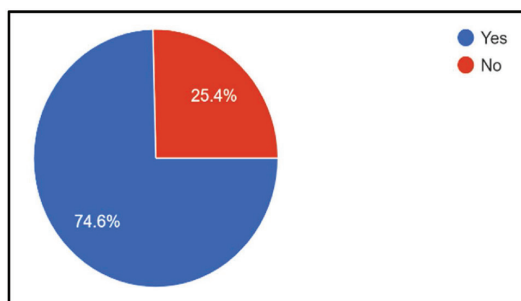


Figure 3: Effects of social media on mental health of the respondents

According to study, 74.6% people's mental health get affected without social media, while 25.4% reported that their mental health remains unaffected.

4. Which social media platform you use?

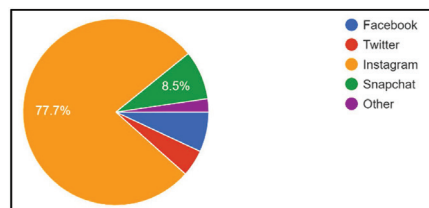


Figure 4: Frequently used social media platform by respondents

According to study, 77.7% of people use Instagram, followed by 8.5% who use Snapchat, and a small percentage use Facebook, twitter, and other platforms.

5. Have you ever witnessed crime being shared on social media?

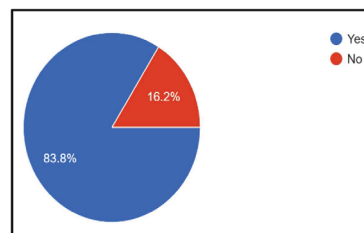


Figure 5: Crimes witnessed on social media by the respondents

According to study, 83.8% of people have witnessed crime related content being shared on social media, while 16.2% have not.

6. What types of crime do you think are most influence by social media?

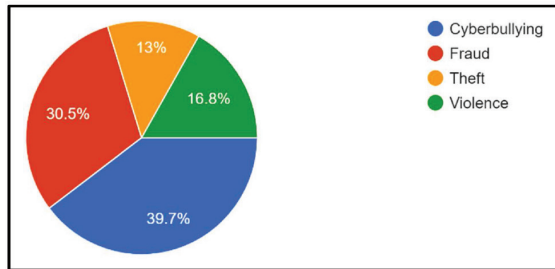


Figure 6: Types of crimes witnessed on social media by the respondents

According to the study, 39.7% of participants believe cyberbullying is the most influenced crime type by social media, followed by fraud (30.5%), violence (16.8%), and theft (13%).

7. Do you think social media companies should do more to prevent crime?

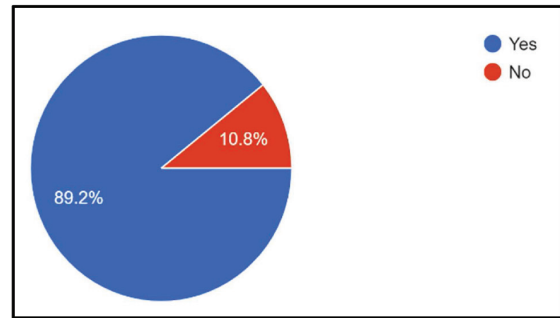


Figure 7: Prevention of crime on social media by the respondents

According to study, 89.2% of participants believe that social media companies should take more responsibility to prevent crime on their platforms and 10.8% of participants do not think social media companies need to do more.

8. In your opinion how does media influence crime rates?

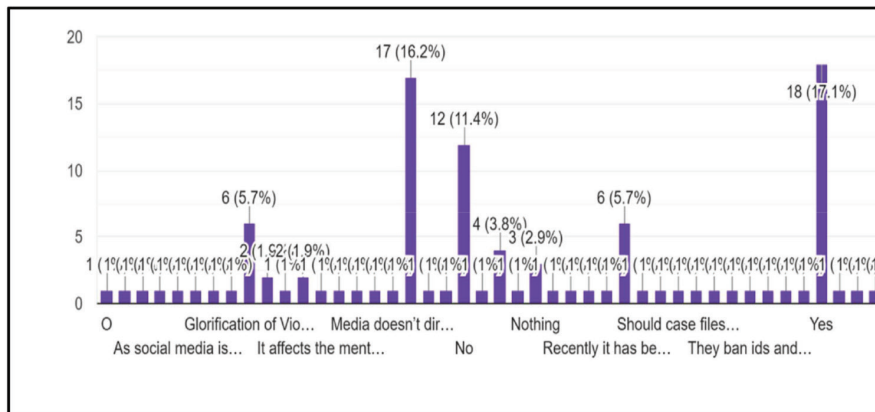


Figure 8: Social media influence on crime rates by the respondents

According to study, people response to how media influences crime rates, 5.7% said media glorifies violence, 16.2% felt media's influence has increased recently, 3.8% said media normalizes crime or spreads disturbing content, 74.3% gave unique individual opinions on media's role in crime.

9. Would you report criminal activity or suspicious behaviour in social media?

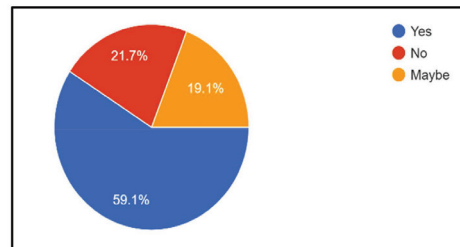


Figure 9: Reporting of criminal activities on social media by the respondents

According to study, out of 115 respondents, 59.1% said they would report criminal activity or suspicious behaviour on social media. Meanwhile, 21.7% said they would not report it, and 19.1% were unsure.

10. Have you ever felt influenced by the criminal activity you saw on social media?

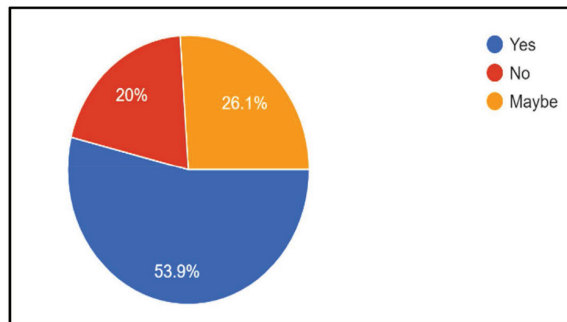


Figure 10: Influence of criminal activities on social media

From the survey it is found that among the 115 responses, 53.9% are influenced, 26.1% are unsure about it and 20% of respondents are not influenced by the criminal activity they saw on social media.

Results and Discussion

The survey was done on total of 187 individuals. From the responses obtained among 100% of people 89.3% people use socialmedia onthe daily basis. Around 74.6% people are almost addicted to it, without social media there mental health gets affected and there is a major chance of getting influenced by the activities they see in social media. And the criminal activity faced by people via social media is fraud, theft, cyberbullying and the violent contents shared throughthis platform influences the minds of people who view it this can be explained by Social learning theory. Thereare emerging trendsand challenges suchas Anonymity, cyberbullying, cybersecurity risks, jurisdictional issues, digital literacy, policy gaps, etc. These can be overcome by creating awareness among people regarding the crime happening via social media. Social media can be used by law enforcement to investigate crimes,gather evidence and even identify suspects.The positive side of social media has to

be focused. The activities in social media has to be monitored closelyto avoid the crimes.

Our results reflect broader research in the field. For instance, global prevalence of cyberbullying victimization among adolescents generally ranges between 14% and 57%, with perpetration rates from 6% to 46%¹¹. In India, one community-based study in the Gurugram district found that 1 in 4 adolescents experienced cyberbullying, particularly via WhatsApp, Instagram, and Snapchat. Additionally, a scoping review of social media studies reported a median cyberbullying prevalence of 23%, and confirmed consistent associations with depression— even though links with anxiety and self-harm were mixed¹². Compared to these figures, our findings of high exposure to crime-related content and elevated perceptions of influence and willingness to report align well with established prevalence data, offering a current, perception-based backdrop to prior empirical and review literature¹³.

Conclusion

The intersection of social media and criminal behaviour also raises important ethical and legal concerns. Issues related to privacy, surveillance, and the rights of individuals must be carefully balanced with the need for public safety and effective crime prevention. Law enforcement must navigate the delicate line between lawful investigation and infringement on personal freedoms, often in a legal and scape that is struggling to keep pace with technological change. The interaction between crime and social media presents several challenging dimensions. Social media can both facilitate crime and aid in its detection, making it a critical area of study in contemporary criminology and digital forensics. The ongoing growth of technology will influence how crimes are committed and how they are addressed by law enforcement. It is imperative for society, policymaker, and law enforcement to understand this dynamic and to develop adaptive strategies that address the challenge of the digital age while protecting civil liberties. The digital foot print of crime, once considered peripheral, is now central to the understanding and combating of modern criminal behaviour.

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