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A REVIEW ON INSTRUMENTAL ANALYSIS OF DATURA AND RICIN POISONING.

Toxicology

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ABSTRACT

Both ricin and datura are strong poisons that come from natural sources. Rich in tropane alkaloids including atropine, scopolamine, and hyoscyamine, as well as the poisonous protein ricin from Ricinus communis (castor beans), Datura species are dangerous because of their high toxicity and potential for abuse. In biological and environmental matrices, analytical equipment is essential for detecting, measuring, and tracking these poisons. Several instrumental methods for the identification and description of ricin and datura poisoning are examined in this review, including mass spectrometry, chromatography, spectroscopy, and immunoassays. The report also gives forensic analysts useful information. The advanced approaches are more dependable and use less time than the traditional methods.

KEYWORDS

Datura Poisoning, Ricing Poisoning, Instrumental Analysis, Chromatography, Mass Spectroscopy, Forensic Toxicology, Poison Detection, Alkaloids, Protein Toxins.

1. INTRODUCTION

The study of plant poisons is called Phyto-toxicology. The most common poisonous higher plants are angiosperms, or the flowering plants, but it is only an incredibly small percentage that is regarded as being toxic [1]. Plant poisons can be divided into four major classes: neurotoxic, cardiotoxic, cytotoxic, and hepatotoxic. Mostly plant poisons are classified into 4 distinct class- neurotoxic, cardiotoxic, cyto-toxic and hepato-toxic. Few neurotoxic plants are deadly nightshade (*Atropa belladonna*), *Nerium oleander*, Castor beans (*Ricinus communis*), water hemlock (*Cicuta maculata*). Examples of cardiotoxic plants are foxglove (*Digitalis purpurea*), nicotine, etc. examples of cytotoxic plants are *Catharanthus roseus* (Madagascar Periwinkle), *Camptotheca accuminata* (happy tree), *Podophyllum peltatum* (American mandrake), etc. examples of hepatotoxic plants are Comfrey (*Symphytum officinale*), Senecio (Ragwort), Crotalaria (Rattlebox), etc.

1.1 Overview of Datura and Ricin poisoning

Datura stramonium is also called devil's trumpet or thorn apple. This genus of plants is composed of 9 to 15 known species and is native to different temperate and warm areas of the Old as well as New World. Datura plants are characterized by their large trumpet-shaped flowers, which can be of various colours, including white and purple, as well as spiny round seedpods (often referred to as thornapples). It is a small shrub that, although it is smelly and not too nice, is a member of the family Solanaceae which is rampant all-over rural India. It is a plant that stands 3–5 feet tall with thick leaves that are dark green and ovate, and flowers are trumpet shape in white or purple colour. The spherical capsule like fruit, has spines which contains up to 200 small seeds that are black dark or brown, kidney-shaped, and thus look like they are chilled seeds. The seeds of the plant are the most dangerous but all parts of the plant can be poisonous [2]. The active component of datura includes scopolamine or hyoscine, atropine, and hyoscyamine. These herbs have been used for a variety of purposes, including their use as poisons, medicines, and hallucinogens, as rituals of intoxication. Their highly potent tropane alkaloids, such as scopolamine and atropine, make them both alluring and harmful. The plant is highly toxic even a tiny quantity of any of its the parts[3].

Ricinus communis is the scientific name of castor beans. This seed contains ricin, a very special poison which is available from it. Ricin is the toxic component in castor oil. Main controversy in that status is between ricin and ricinolic acid. The biological factor stepping in the body to prevent production of vital proteins and harm the organism as a whole is ricin. The thing that is needed to kill half of the mice is around 22 mg/kg of body weight, which is one of the most dangerous substances for life. In contrast, the oral intake of ricin appears to be much less poisonous, with an estimated lethal oral dose in human being approximately 1 mg/kg of body weight [4][5].

1.2 Need of analytical methods

Forensic toxicology is a sector of forensic science focused on examining the chemical impacts of drugs and toxins on the human body. Common criminal cases include driving while intoxicated, poisoning of wildlife, and investigating causes of death. Analytical methods are effective in identifying the composition of the sample and quantifying its levels. This provides evidence that is undeniably reliable [6]. Analytical techniques consist of a series of guidelines and processes that facilitate the identification (qualitative analysis) and potentially the measurement (quantitative analysis) of a sample being examined[7].

2. Toxicological Aspects of Datura and Ricin

In Datura plants, there are tropane alkaloids such as hyoscyamine found in roots, leaves and seeds; atropine dl-hyoscyamine, hyoscine and scopolamine l-hyoscine found in roots. They are antagonists acting competitively to muscarine, which is structural acetylcholine or in other words, derivative of acetylcholine in both peripheral and central which causes a blockade of general functioning of all the parasympathetic innervated organs [8]. Dryness of mouth, delirium, dilated pupils, slurry speech, swallowing difficulty, hallucinations, difficulty in urination, muscle incoordination, perspiration, increase in pulse rate, and even death that is headed by tachycardia, arrhythmias, coma, respiratory depression, are clinical features of datura poisoning.

It is extremely potent to mammals, being able to kill 1500 cells per minute with small doses [9]. Ricin has toxicity similar to the neurotoxic agent sarin and can be easily extracted from the castor beans [10]. The ricin toxin consist of both A and B polypeptide chains which are covalently linked by a disulfide bond. Ricin toxicity can be explained by the action of the ricin toxin A chain, which inhibits protein synthesis. This is accomplished by the action of a specific ricin toxin A chain which destroys ribosomes through the hydrolysis of the N-glycosidic bond of an adenosine residue in the 28 S ribosomal RNA [11]. Clinical symptoms are uraemia, dehydration, cramps, seizures, tremors, burning sensation in gastral-intestinal track and few postmortem changes are haemorrhage in brain, intestine, myocardium, pleura, etc.

3. Samples preparation techniques

3.1 Sample matrixes

Anti-mortem Sample matrices include keratinous samples (like hair and nails) and bio-fluids (like urine, blood, plasma, saliva, gastric lavage, etc.). Samples taken after death include visceral material (live, kidney, brain, stomach, and intestine) and biofluids (blood, urine, gastric clavage, vitreous humor, CSF, bile, etc.).Unconventional matrices include things like cream cookies, tea, sweets, ointments, water, veggies, pharmaceutical pills, and capsules [12].

3.2 Pre-treatment methods

It includes modern extractions techniques like liquid-liquid extraction, solid-phase extraction and Protein precipitation for ricin samples.

3.2.1 Solid-phase extraction

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Using sorption onto a solid phase and solvent elution, solid phase extraction (SPE) is one of the analytical methods for sample preparation that concentrates and purifies analytes from solution [13]. Principle of SFE, to extract a solid phase, it is usually necessary to directly contact sorbent or solid phases and allow the resulting compound to selectively absorb its surface. The solid phase sorbent is commonly found in small tubes or cartridges. When the sorbent is selected with care, it should retain the organic compounds and avoid using extra materials that may be present in the sample. A suitable solvent can pass through the sorbent to remove this extraneous material. After that, it is possible to use a suitable solvent to elute the compounds of interest from the sorbent. This solution is then gathered for examination [13].

3.2.2 Liquid-liquid extraction

Liquid-liquid extraction, also known as solvent extraction or partitioning, is a method used to separate chemicals or metal complexes based on their differing solubilities in two immiscible liquids. Typically, one of these liquids is water (polar), while the other is a non-polar organic solvent. There is a transfer of one or more molecules from aqueous to organic [14]. Microfluidic devices can dramatically accelerate liquid-liquid extraction, reducing extraction and separation times from minutes or hours to a matter of seconds when compared to conventional extractors [15].

3.2.3 Protein precipitation technique (for ricin sample)

The process of protein precipitation technique (PPT) has been widely employed to prepare biofluid samples for chromatography and spectroscopy analysis. There are different types of protein precipitation techniques such as Polyethyleneimine (PEI) precipitation, organic solvent precipitation, ammonium sulphate precipitation and solvent precipitation.

4. Instrumental Techniques for Datura Poisoning Analysis

4.1 Chromatography techniques

These methods are employed to separate and refine in very small amounts of metabolic products [16]. In chromatography, the components of a mixture are separated based on their different physicochemical interaction with the stationary and mobile phases. This process involves introducing a suitable flowing mobile phase or eluent to the mixture, or initiating the flow of the eluent in another manner. The stationary phase can be either an immiscible or non-volatile liquid on a solid support (as in partition chromatography) or a porous solid such as silica gel (used in adsorption chromatography).

4.1.1 Gas Chromatography (GC) coupled with Mass Spectrometry (MS) or Flame Ionization Detector (FID)

It is the chromatography technique used to detect the volatile evidences or samples. It separates the compounds according to the strength of their molecular interactions using a packed column as the stationary phase and gas as the mobile phase. At the VIT-SIF Lab, SAS, Chemistry Division for GC-MS analysis, Vellore, Tamil Nadu, chemical profile of Datura metel L. seeds were completed. A Clarus 600 (E.I.) mass spectroscopy detector (GC-MS) in conjunction with a Perkin Elmer Clarus 680 type gas chromatograph was employed [17]. GC-MS can identify metabolites (byproducts) of drugs or poisons, providing evidence of exposure or ingestion [18]. Flame ionization detector (FID) may destruct the analyte and it is unable to detect organic compounds, as MS has more advantages over FID so it is more commonly used.

4.1.2 High-Performance Liquid Chromatography (HPLC)

HPLC commonly used with diode-array detection (DAD) or UV. The Agilent (1200 series) HPLC system with a manual sample injector was used to quantify the marker component in the plant extract. It had a variable wavelength detector (VWD), a quaternary pump, and a degasser unit with a diode array detector (DAD). The data was generated using Chemstation32 software. To measure the proportion of the marker component in the plant's seed, a calibration graph was created using reference standards [17].

4.2 Mass spectrometry

Mass spectrometer is a detector attached to the instrument such as GC-MS (as mentioned above) or LC-MS/MS for high sensitivity and specificity. A potent method for identifying harmful alkaloids in Datura plants, such as atropine and scopolamine, is mass spectrometry. It is utilized in archaeology to determine the historical use of Datura and in forensic science to validate poisoning cases.

4.2.1 Liquid Chromatography-Mass Spectrometer (LC-MS)

The Interdisciplinary Institute of Indian System of Medicine (IIISM) at SRM University in Kattankulathur, Chennai, conducted the examination of Datura metel seeds. The single quadruple mass spectrometer (LC-MS) was employed in conjunction with a Shimadzu 2020L.C. system [17].

4.3 Spectroscopy techniques

Spectroscopy techniques are crucial, especially when it comes to detecting and measuring the plant's alkaloids (including atropine, scopolamine, and hyoscyamine) and other chemical components. These techniques use the interplay of light and matter to provide important details about the concentration and molecular makeup of substances.

4.3.1 Fourier Transform Infrared (FTIR)

In contrast to traditional approaches, Fourier transform infrared spectroscopy, a vibrational spectroscopy technique, is notable for its speed and accuracy [19]. The first step in preparing and detecting a sample for Datura poisoning using FTIR is to ground the plant material into a fine powder if it is solid, or use a liquid extract. When working with solid materials, combine the powder and potassium bromide (KBr) and form the mixture into a pellet. To spread a drop of a liquid sample equally, lay it on a KBr plate and cover it with another plate. After preparation, the sample is put inside the FTIR spectrometer, which allows infrared light to flow through it. The sample produces a distinct spectrum by absorbing particular wavelengths. To determine the presence of particular substances, like the poisonous alkaloids in Datura, this spectrum is compared with reference spectra [20].

5. Instrumental Techniques for Ricin Poisoning Analysis

Detecting ricin poisoning involves several instrumental techniques such as mentioned below:

5.1 Immunoassays- Methods such as electrochemiluminescence (ECL) and enzyme-linked immunosorbent assay (ELISA) are frequently used. These techniques detect ricin with high sensitivity, frequently at the ng/mL level, by using particular antibody-antigen binding [21].

5.2 Chromatography- To improve detection, mass spectrometry (MS) is frequently used with liquid chromatography (LC). By separating ricin from intricate biological matrices, LC facilitates its identification and quantification [21].

5.3 Mass Spectrometry (MS)- LC-MS/MS and other MS-based techniques are very good at identifying and distinguishing ricin from other proteins. By recognizing the release of adenine from nucleic acid substrates, these techniques can track the activity of ricin and identify it based on its distinct amino acid sequence [22][21].

5.4 Molecular Techniques- The genetic material of ricin can be found using methods such as polymerase chain reaction (PCR). Because of their high specificity, these techniques can detect even trace levels of ricin DNA or RNA in samples [23].

6. Challenges in Instrumental Analysis

Identifying Datura and ricin poisoning involves several difficulties in instrumental analysis:

6.1 Complex Mixtures: Biological samples, including blood or urine, have many substances that can hinder the detection of poisons like those found in Datura and ricin [24][25].

6.2 Sensitivity and Precision: Methods must be very sensitive and precise to accurately identify low amounts of these toxins. This is especially tough for ricin because of its brief half-life in the body [25].

6.3 Sample Preparation: Getting samples ready for testing can take a lot of time and needs careful handling to prevent breakdown or loss of the toxin [24].

6.4 Availability of Testing Methods: Not every lab has access to advanced methods like mass spectrometry or immunoassays, which are crucial for accurate detection [24].

6.5 Timelines of Results: Quick detection is vital in poisoning situations, but the time needed for sample preparation and testing can slow down diagnosis and treatment [24].

7. Forensic significance-

Datura poisoning, which results from tropane alkaloids such as atropine, hyoscyamine, and scopolamine can be verified using instrumental testing. Methods like GC-MS are important for finding and measuring these poisons in biological samples (like blood, urine, or stomach material), recognizing the origin of the poisoning, and offering scientific proof in legal situations related to accidental, intentional, or harmful intake.Ricin, a strong poison taken from Ricinus communis (castor beans), is frequently involved in deliberate poisoning due to its deadly nature. Instrumental analysis is essential for spotting ricin in biological samples (like blood, urine, or tissues) and environmental clues (such as food or powders). Methods like Immunoassays and Liquid Chromatography-Mass Spectrometry (LC-MS) assist in verifying ricin exposure, tracking its origin, and determining its involvement in poisoning incidents. This analysis gives vital scientific proof for investigations and legal actions, especially in cases of bioterrorism or deliberate poisoning.

8. Future Directions-

8.1 Exploration of Novel Biomarkers for Early Detection

One promising direction is identification and validation of novel biomarkers for early detection of Ricin and Datura poisoning. Biomarkers can provide specific and sensitive indicators of exposure, allowing for quicker diagnosis and treatment. Research is focusing on discovering unique metabolic or protein signatures that can be detected in biological samples using advanced techniques like mass spectrometry and molecular assays [2].

8.2 Development of More Robust, Multiplexed Platforms

Finding and confirming novel biomarkers for the early identification of ricin and Datura poisoning is a promising strategy. Faster diagnosis and treatment are made possible by these biomarkers, which provide sensitive and accurate exposure indicators. The goal of current research is to use sophisticated techniques like mass spectrometry and molecular assays to identify unique metabolic or protein signatures that can be found in biological samples [26].

8.3 Improved Cross-Disciplinary Approaches

Bringing together knowledge from chemistry, biology, and data science is crucial for improving the identification of Datura and ricin poisoning. Chemists may create novel reagents and techniques for extracting and analysing toxins, biologists can examine the biological impacts and interactions of these toxins, and data scientists can design algorithms for analysing complex data sets. This cooperative approach can result in creative solutions and enhanced diagnostic tools [27].

9. CONCLUSION-

The detection and analysis of datura and ricin poisoning require sophisticated instrumentation for accuracy and reliability. Advances in analytical chemistry continue to enhance our capabilities, but challenges remain, especially in field applications and rapid diagnostics. Continued research and development are essential to meet the growing demand for effective toxin analysis.

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