Pollen in Forensic Palynology: An Exploration into a Crime Solving Tool

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Abstract— Palynology is the study of pollen grain and spores. The term forensic when inserted into the picture marks the usage of pollen into a crime-solving and suspect finding tool. Pollen grain has an outer layer made up of sporopollenin which gives high durability and resistance due to which pollen can survive at adverse and extreme conditions. The samples collected from the crime scene are analysed, studied and investigated along with the samples obtained from the suspects. A relation is derived which acts as evidence during the judicial custody. Pollen analysis is highly useful if a crime that has happened long back, and now needs to be opened up. Advancement of this technique in India is still awaited.

Keywords— Forensic palynology, crime, pollen spores, evidence, sporopollenin.

I. INTRODUCTION

Forensic palynology is a branch in forensic sciences that deals with the practices of not merely the usage of pollen grains and spores but also its identification. Moreover, it is as well concerned with the application to investigation, and thereby, solution of civil and criminal cases. It is a light yellow powdery substance found on the top surface of most flowers. It is the sexual reproductive part from which a plant produces its off-springs. On the other hand, spores are for asexual reproduction. Pollen and spores are too microscopic to be seen by the naked eye as it ranges from 7–200micrometre.[6] They can adhere or stick to most of the surfaces and can deeply get struck in clothing, ensuring that they do not get washed-off by detergents. Since the mode of pollination includes wind, water, and animals, it is easily deposited onto people or other items – like clothes, shoes and nostrils. In short it sticks like glue to the parts of a person’s body.

Pollen grains date back to 450 million years ago, such is its resistance and durability. It is hence known as a remarkable geographic locator. [5] Pollen and spore analysis is not a new concept. It all began in 1916, when it was first examined by Lennart Von Post. Since 1950’s, the spores and pollen grains are being used frequently in criminal investigations like theft, rape, terrorism, bomb blasts, drug cases, and murder mystery solving. Often the case is that the criminals do very rarely realizethat they have collected the spores on themselves from a crime scene as they are in a hurry to escape the location, which later acts as boon for the investigators. Pollen and spores are inbuilt with several characteristics that make them functional for forensic analysis. Both are produced in large quantities, and are relatively resistant as compared to any other substance on Earth. It is due to the presence of sporopollenin in the exine layer, to prevent destruction.[13] This means that they can be used as evidence even after many years of a criminal incident having taken place. [9] On addition they have such unique appearances that allow distinguished species to be identified with a microscope.

Etymologically, the term ‘palynology’ is fetched from the Greek language meaning ‘the study of powder and dust.’ Forensic palynology is a branch that is dedicated to the study of the usage of pollen grains and spores along with its identification. It is also an analysis to investigate and solve civil as well as criminal cases.[14] Diversifying its area of application, the stream does include all the legal information that are obtained from the analysis of a range of microscopic organisms- such as dinoflagellates and chitinozoans- that are a common inhabitant of fresh and
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marine habitat. However, this is not mostly required as the search of a palynologist starts and ends at fossils. In the backdrop of this brief introduction regarding pollen, spores and palynology, this paper talks about how palynology can be useful in investigating crimes, and how the smallest particle can aid to unleash the biggest murder cases, and thereby, the study attempts to establish the relation between the suspect and the crime scene findings.[7]

The main objective of this paper is to highlight on the following aspects.

1. It establishes the relation between an item and the crime scene.
2. It traces back to history of travelling.
3. It races environmental information in the primary crime scene.
4. It verifies the presence of victim and the murderer at the crime scene.
5. It attempts helping in cutting down the suspect list.
6. It clarifies whether the crime scene is primary or secondary.

II. ANALYTICAL ISSUES

There are two significant questions which are to be tackled at the very outset. To begin with, there are around 1.5 to 2 million different varieties of plants that has the ability to produce pollen or spores to satisfy its purpose of reproduction, best part out of which is that they are quite unique in their own way, be it varying on terms of size, shape and roughness to its ability to travel long distances and germinate on flowers.[13] This is to be noted that all region’s native plants have their own unique and distant pollen type which is termed as pollen fingerprints. Nowadays palynology findings have become important in investigation of criminal cases, due to its requirement before the judge in the court of law as an evidence. Therefore, they are frequently used by the countries like Australia, United Kingdom and New Zealand.[8] From the expert’s view, fossil or the pre-existing pollen are also being used for investigation purpose along with the modern pollen, as it’s been useful in determining, what series of events happened and more importantly where it happened, hence, revealing the crime location.

In exploration, investigation and analysis, there are eight vital aspects.

- First, an essential concept of tracing pollen is based on an important ideology known as “Locard’s principle of exchange”.
- Second, the idea of production and dispersal pattern of each pollen is important to determine the pollen print meaning and in order to locate the location of its existence.
- Third, the very basic components involved in crime scene investigation that include soil, water, dust are present almost in every case, the careful examination of which can help discover the pollen samples that can possibly be used up as evidence for the crime.
- Fourth, the sampling can be done in lab and can be matched with the pollen or spore samples obtained from the items fetched from the suspect cloth, skin, shoes, of the victim or the suspect thereby indicating the presence of the same person at the crime scene, hence stating some relation between the two.[11][14]
- Fifth, in a similar scenario, imagine getting mud from the tire of the vehicle, followed by the inspection of the mud found in the crime scene, - it will clearly state that there is a relation between the two as mud might contain pollen grains which is native and unique in its domain, thus, stating the fact.
- Sixth, similarly, if a pollen found on the cloth worn by the suspect matches with the pollen collected at the crime scene, then it directs the investigation towards the suspect being present at that location at some point of time. This method can also help us in listing out and differentiate between a primary and secondary crime scene.
- Seventh is the use of trace evidence.[12]
- Eighth is the new fact that can come to light at any instance during investigation.
Differentiation criteria:

Before an investigation and corresponding analysis begins, an expert need to scan the angiospermic pollen, and develop a differentiation criterion based on size, shape, membrane composition and amount (abundance and dispersal).

First, each pollen has its own identification mark with respect to its specific size. Careful microscopic examination of which can reveal the small minute differences, if any. Second, the shape of every pollen is different to one another, when compared. Some are in the form of triangle shape; some are in round while others are in oval and even irregular in shape. Owing to their mode of dispersal, some of the pollen have smooth surfaces while others are covered with rough ones. Third, the biochemical analysis of membrane also known as cysts reveal the secret that no two pollen or spores share the same composition in their membranes. Fourth, the amount of abundance is based on the mode of dispersal, either through wind, water or animals. Example, 95% of anemophilous {animal as mode} plants are found within a range of 2km to 100m from the parent plant. In the case of water and wind mode, pollens have the capability to reach a range of even 100 km.

III. MATERIALS AND METHODOLOGY

Sample Types: There are three major samples - sediments, hair and clothes and illegal drugs.
Sediment: Soil, dirt, and dust are commonly found elements at almost every crime scene, which needs to be properly handled as they can contain abundant pollen and spores. Samples of dirt collected from the clothing, skin, hair, shoes, or car of a victim might prove to be useful in linking the victim to the location where the crime has occurred. Mud that is found on a stolen vehicle, or a vehicle which has been used in committing the crime, could link the vehicle with the crime scene or to the place from which it was initially stolen from. Examples of where these types of soil, dirt, or dust samples should be collected are many. In addition, primarily, steps must be taken to ensure that samples do not become contaminated otherwise it will lose its originality.[1][12]

Hair and Cloth: Woven cloth, woolen blankets, ropes, and stuff like that make excellent traps for pollen and spores. Woven materials are made of tiny interwoven fibers, hence, when air comes in contact with these materials, the fibers here become filters that can retain these solid like spores. Woolen garments that primarily includes the blankets, skirts, and sweaters, act up as the best pollen and spore trap. Apart from that, hair whether human or animal, remains one of the very best pollen and trap for spores. When wind blows through hair strands, pollen gets stuck in between. In humans, the usage of various types of hair serums, natural oils, makes hair surfaces sticky and becomes an even better trap for pollen and spores to stick on. [6]

Hair from a victim, or suspect, can be taken for sampling by carefully washing it with detergents followed by rinsing it with warm distilled water. This process will loosen the trapped pollen in the strands. Once collected, the washed water can be stored before analysis in a sterile container that is either tightly closed or frozen, or a small amount of alcohol can be added to reduce the microbial growth. Fur found at the crime scene is generally used for wiping shoes and hence are rich in pollen and spores. Domestic pets such as cat, dog, sheep can be found on the crime scene and pollen and spores can be found on their hair or fur. This can embark a new story for the crime. Hence, this should be considered as a potential use for its forensic samples.

Illegal drugs: Marijuana plants come in two sexes- plants that are male and some that are female. Only the male ones have the caliper to produce pollen. Male plants are often weeded out because pollination and seed production are not demanded. In such cases, sampling would reveal very little amounts, a factor that is considered to be important and can possibly be used in court as significant evidence.[6]

Miscellaneous Samples: In addition, there are miscellaneous samples as well. For example, people can make out the origin of the shipment of the crude oil or petroleum. While packaging the pollen spores can get attached to it and be carried along. Honey carrying spores can be processed and purified for identifying a geographical location. Food found in the gut and intestines of the suspect or the murderer can reveal a lot too.

Methods of pollen dispersal
Four points are significant to note in investigation and analysis:

One, the methods follow up by which plants disperse their pollen or spores. The aquatic angiosperms live completely submerged under water and release their pollen there, thereby, relying on water currents to transport the pollen from the anther to the female stigma of a same species flower. However, these does not have a very high success rate as this medium of transport, like the wind, is a hit and miss method of pollen dispersal. Owing to this reason, these plants produce pollen types that comprise of only of a single-layered cellulose wall, the pollen is almost never preserved here in the form of sediments and usually oxidizes rapidly as soon as it gets removed from water.

Two, another minor group of plants are called "autogamous" referring that they are self-pollinating in nature and are so efficient that only less amounts are needed. Pollen from these plants gets rarely dispersed into the atmosphere even though their pollen preserves well and has a comparatively highly durable outer wall, called as "exine," which is made up of a stable chemical compound called "sporopollenin".[7]

Three, in a larger group of plants, known as zoogamous plants, pollination depends upon the transport of pollen by some type of insect or animals. Because of its efficiency, pollen productivity is low, yet it is not as low as the ones that are found in autogamous plants. The potential yield of zoogamous pollen in forensic work is excellent due to two reasons. The zoogamous pollen grains comprise of some of the most durable version of its exine. Meaning that their pollen will remain preserved and undisturbed in deposits for longer periods of time and hence they are less susceptible to destruction, the zoogamous pollen is produced in low amounts, thus, this feature makes it to not normally being found at all places. This point is both good and bad. It is good because if this grain of pollen is matched in a forensic sample, there is a confirmation that the pollen is from a particular atmospheric region. It is bad as such little traces of pollen is produced by the plant that its chance of getting mixed is drastically reduced.

Fourth, the last category is the wind-pollinated or the anemophilous type. This group includes a wide range of producers like the gymnosperms and angiosperms. Spore-producing plants such as fungi, ferns, and mosses are also...
named in the list. Listing wind pollution as the most inefficient way of dispersal, the anemophilous plants must release large amounts of these low weight grains. “Sinking speed”, an important factor to take a note of means the rate at which pollen grain fall on Earth, such as marijuana or birch, which are light has an average rate of around 2cm per second whereas that of maize tree is 15 times more faster than the lighter ones. Listing out these two examples it can be understood that small dispersion areas point to greater precision in identifying the source region.[1] [15]

Security is yet another essential concern. The palynologist must state under oath that the samples were kept at a locked chamber which was out of reach by people other than the designated ones. Ensuring that there was no contamination is a serious issue to be looked upon. One final view of concern is the amount of material recovered for the analysis. Not having enough to sample will keep off from the various techniques to experiment with. If something goes wrong, samples cannot be retested. Before a destructive pollen test is done, other tests can take up the little pollen that was extracted, hence, sufficiency is not achieved.

![Fig.4: Pollen grains when viewed under a microscope.](image)

Analysis of pollen is usually carried out using transmission electron microscopy (TEM), which simply is referred to any type of microscopy in which a source of light is transmitted through the sample, allowing the sample to be viewed through a lens. In addition to this, scanning electron microscopy (SEM) may also be utilised. This technique is widely used alongside systems like QEMSCAN (Quantitative Evaluation of Minerals by Scanning Electron Microscopy), which allows for the automated analysis of minerals and other substances that are put to viewed. The palynologist will then do the follow up, and compare the pollen grains using their own expertise as well as taking insights from the pollen reference collections, if available. [3]

There are three methods for obtaining useful pollen grains images, namely transmitted-light microscopy (TLM), the Widefield fluorescent method and the structured illumination (Apotome) method. All these analysed products depend on the role of dispersal.

HTS methods are also used, the other name of which is next-generation sequencing (NGS) method or simply DNA sequencing. These technologies allow for the genome sequencing of DNA and RNA much more easily and inexpensively, hence widely used in molecular biology.

IV. RESULT AND DISCUSSION

Pollen can also be picked up and transferred owing to the dispersal mechanisms. Pollen have the ability to stick on surfaces be it human body as well, hence people can pick up these spores from the crime scene. Keeping these specific usages in mind, an essential use of palynology in crime investigation is to build up a connection between two regions, objects or people. For instance, it is possible to link a suspect to an object found on the crime scene or, a vehicle to a place. If a suspect was present at a particular crime scene at which pollen can be found, there is a possibility that, they may have picked up pollen on their clothing or in their hair. Pollen has a characteristic feature of its high resilience and so, it can often stick to other objects even after it has been washed. The pollen found on the suspect, if matches with the pollen spores extracted from the crime scene can depict that the person was present at the crime scene at some point of time. However, it must be noted that despite, the presence of pollen may establish a link, the lack of pollen does not necessarily signify that there is not a link.[3] Palynology may hence be able to determine the location of a crime scene if it is not known. The study of pollen can also be used to find about the travel history of an item. In some cases it may be essential while solving the crime, to ascertain about the origin, especially illicit drugs, money, antiques and sometimes even food.

Although forensic palynology may not be able to derive at the exact location, it may at least be able to drive the investigation towards the correct way. It may sometimes even be possible to estimate the time of year at which the crime has taken place. It is quite obvious that despite the links palynology as a subject can establish, further evidences may be needed to support any conclusions drawn in the judicial custody.[2]
Future of forensic palynology in India

The secret life of pollen is that it can make us sneeze, itch our eyes but can solve crimes find, find the convict and solve the murder.[5] A single grain of spore can tell us nothing, but pollens in mass can unleash a lot of secrets.[4] The full zest of forensic palynology remains untapped and undiscovered in India and most parts of the country leaving New Zealand which has taken the lead in its use of forensic palynology as a crime solving method as well as in acceptance of pollen evidence in courts. All these and still the Forensic palynology is in its infant state. Forensic palynology has become a widely discussed topic as it provides us with insights related to microscopic pollen and spores that get trapped and are used to resolve criminal and civil cases, including cases of rape, bomb blasts, homicide, theft.[10]

Hopefully, as the benefits of pollen analyses are realized, forensic palynology will eventually become a valuable tool which would be used each time a crime is committed and justice is to be served.

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