Cell Wall to Cell: Microscopic Forensic Botany

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Forensic botany includes a wide variety of subdisciplines, from ecology and limnology to palynology and molecular biology. Plant anatomy is especially pertinent to trace evidence. For our purposes, botanical materials will include any portion of higher and lower plants, algaes (including diatoms), and fungi. Trace evidence is the small amounts of materials transferred when two objects come in contact with each other. This concept is Locard's Exchange Principle [1], and it can be used to make associations between the suspect, victim, crime scenes, and weapons. In addition to making associations, trace evidence may be used to corroborate statements. The importance, or probative value, of a particular type of evidence is dependent on the circumstances of the case. What is the case scenario? How common is the type of botanical material? Are there any unusual acquired characteristics? How could it have been transferred? Was it related to the crime or possible non-criminal activity such as wind, animals, insects, water, or gravity?

Botanical materials are found in their natural as well as processed states – from botanical debris in the soil, to gastric contents from food, textile fibers in our clothing, and the fibers in paper, wood, and engineered wood products. The cell wall is particularly important for identification of botanicals because the wall is not easily destroyed by processing or digestion by most organisms. Therefore, the cell wall persists when other plant features are destroyed. All plant cells are surrounded by a thin primary cell wall which is comprised of randomly deposited cellulose microfibrils. Schlerenchyma cells also have a secondary wall comprised or oriented cellulose microfibrils. This secondary cell wall is deposited in specific patterns or thickenings. The cell wall morphology may be used to identify natural fibers made from plants, wood and wood products such as paper, as well as botanical microfragments found in debris and gastric contents.

Natural fibers from plants come from the non-woody tissues. Such fibers are called vegetable fibers, and they may be derived from stems, leaves, or seed tissues. Identification of these different cell types is based upon the morphology of the cell wall. Also, there are only a limited number of plants that are used to produce commercial natural fibers [2]. Natural fibers derived from seed tissues are called seed fibers. They are comprised of ultimate fibers and may be attached directly to the seed, such as cotton, or they may be part of the seed pod, such as kapok. Natural fibers derived from stem tissues are called bast fibers. These fibers are comprised of technical fibers and originate from the vascular bundles found towards the outer side of stems from dicots. Vascular bundles are comprised of transportation tissues called xylem and phloem. The xylem tissues transport water up from the roots, through the stem, and into the leaves. The phloem tissues transport sugars and other important products in the opposite direction, from the leaves through the stems to the roots. These transportation tissues need different types of cells, some for the actual transport of materials and others to support the tissue. Technical fibers originate from the vascular bundles of different plant organs; therefore, these types of fibers are comprised of different cell types. Ultimate fibers originate from seed tissues and provide only structural support.

Identification of natural fibers begins with polarized light microscopy. The morphology of the cell wall and the degree of extinction can be used to narrow down the possibilities. Technical fibers may be separated by a variety of maceration techniques. Herzberg's stain, phloroglucinol, and a modified Billinghame's test may be used to assess the relative amounts of cellulose and lignin in ultimate fibers. The drying-twist test may also be used to differentiate vegetable fibers based on a clockwise or counter-clockwise spin that occurs as the fibers dry. This spin is based on the chiral nature of the cell wall, which is based on the secondary wall being deposited in layers in opposite directions.

Wood identification is the oldest area of forensic botany. Wood cells are arranged in a three dimensional triaxial form, radiating outward from a central axis. Therefore, if you have a weathered piece of wood bearing unrecognizable surface orientation, you can expose sectional views of the wood showing the three principal orientations for correct species identification. If you make a cut perpendicular to the stem it is called a transverse or cross-section. This section will expose the growth rings and show the transition from early wood the late wood. If the rays are large enough they will be seen in this orientation. Another view, called the radial plane is exposed by making a cut parallel to the stem and through the pith. If you stand a cut section of tree on its end and chopped it through the center you will have exposed two radial planes. The last principal plane to examine is the tangential. This section is exposed by making a cut perpendicular to the rays which do not pass through the pith. The characteristics of the cell types observed in these three sections may be used to identify the type of wood. The same types of cells may be observed in paper products; however, with paper, the cells must be analyzed without the directional information provided by sections.

Identification of botanical microfragments is limited because of the wide variety of sources. However, a few substances may still be present in addition to the cell wall depending on the source of the botanical microfragment. Such substances include starch, lipids, tannins, crystals or phytoliths, protein bodies, and mucilage. These substances may be used for comparisons even if an identification of the source is not possible. Botanical microfragments may be identified in gastric contents by the morphologies of the cell walls and starches. The different combinations of starch types and grain source make comparison of cereals, breads, and other grain products feasible. Another group of very important substances are crystals or phytoliths found in some plants. Phytoliths are made of the inorganic substances silica or calcium oxalate; therefore, these crystals don't decay when the rest of the plant decays over time and can survive in conditions that would destroy organic residues. Phytoliths can provide evidence of both economically important plants and those that are indicative of the environment at a particular time period.

References
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