COLOR TESTS FOR DIFFERENTIATING
HEARTWOOD AND SAPWOOD IN
CERTAIN SOFTWOOD TREE SPECIES

May 1962

No. 2246
COLOR TESTS FOR DIFFERENTIATING HEARTWOOD AND SAPWOOD IN CERTAIN SOFTWOOD TREE SPECIES

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Summary

In certain softwood tree species, very little color difference exists between the sapwood and heartwood. Being able to differentiate the sapwood-heartwood boundary in such species is of definite value to the research forester and particularly the wood preservation industry for determining depth of penetration in the sapwood. This report brings together the published information on sapwood-heartwood indicators for certain softwood species and also describes tests made to determine the effectiveness of 21 chemicals and dyes on 23 different species.

Introduction

In many woods, heartwood can be readily distinguished from sapwood by heartwood's darker color. In such woods, however, as hemlock, most spruces, the true firs, aspen, and cottonwood, there is very little color difference between the heartwood and the sapwood (13). 2 Trees with this characteristic are sometimes referred to as "sapwood trees" (3).

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1 Maintained at Madison, Wis., in cooperation with the University of Wisconsin.
2 Underlined numbers in parentheses refer to the Literature Cited at the end of this report.

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There are many situations in which it is necessary to distinguish the heartwood-sapwood boundary in sapwood trees. For example, when using these woods under conditions that are favorable to decay or insect attack, it is often valuable to determine the proportion of heartwood present since heartwood is frequently less susceptible to decay than sapwood. This greater resistance is due to the presence of infiltration products, which are more or less toxic to fungi and obnoxious to insects. In the penetration of preservatives, which is mostly confined to the sapwood, suitable means for determining the sapwood boundaries are necessary in establishing how much of the sapwood is treated and whether or not the treated portion is thick enough to give adequate protection. Undoubtedly, knowledge of whether heartwood or sapwood tissue is being dealt with is of value in the use of glues and paints since both types of tissue are different in chemical makeup and therefore produce different bonding properties. This chemical difference would also affect the preparation of pulp from these two types of tissues. Determining percentage of sapwood to heartwood is also useful as an indicator of site or exposure conditions. Width of sapwood may also be useful as an indicator of the crown class of the growing tree.

The purpose of this report is to bring together published information on sapwood-heartwood indicators, carry out necessary tests to validate this information, and to rate these indicators as to effectiveness. This information was compiled for particular use in the density survey of western woods that is being carried out by the U.S. Forest Products Laboratory. Part of this survey requires measuring the width of the sapwood on a large number of increment cores. The measurements that are taken from the cores are being used to correlate width of sapwood with various growth and environmental factors. The measurements will also provide the information that has been requested by the wood preservation industry on sapwood width for particular species and diameter classes.

The criteria for an ideal sapwood-heartwood indicator (2) are: Is easy to apply, either in the laboratory or in the field; gives sharp color differentiation with clear-cut contrast; and gives uniform results even if the specimen is decayed, weathered, or impregnated with a preservative.

The five types of tests that fulfill these criteria most closely and that were used in this study on increment cores from 23 different tree species are:

(a) Starch tests--sapwood may contain more starch than heartwood.

(b) Sugar tests--sapwood may contain more sugar than heartwood.

(c) Organic stains--heartwood and sapwood may be different in chemical composition; for example, when benzidine is applied, a red color is produced if pinosylvin phenols are present in the heartwood (7).
(d) Inorganic stains—when ferric chloride is applied to the sapwood, a green color will result if catechols are present, or a dark gray-blue will result if pyrogallos are present (10).

(e) Hydrogen-ion activity—when bromcresol green, bromphenol blue, or methyl orange are applied to species such as Douglas-fir, contrasting colors are produced between the heartwood and sapwood because of a difference in pH (5).

A number of other types of tests and techniques based on various chemical and physical properties can be used to differentiate sapwood from heartwood when no visible color difference exists. These include:

(a) Ultraviolet light and fluorescent pH indicators—the difference in acidity might be made visible by using a fluorescent pH indicator that fluoresces different colors depending on the pH of the specimen.

(b) Microscopic examination—living cells may be present in the sapwood and lacking in the heartwood.

(c) Differential electrical conductivity—a higher moisture content in the sapwood may allow a greater flow of current than the drier heartwood.

(d) Differential heat transfer—a higher moisture content in the sapwood may allow a more rapid flow of heat than the drier heartwood.

(e) Differential absorption rate of water, dyes, or carbon tetrachloride—heartwood may be less permeable than sapwood because tannins, gums, resins, or salts infiltrate the cell walls or accumulate in the cell cavities during heartwood formation.

(f) Differential diffusion rate of soap solution—when soap solution is forced through the specimen, soap foam forms on opposite side of the more permeable area showing a difference in permeability (12).

(g) Differential diffraction by soft X-rays—heartwood may contain certain chemical constituents not found in the sapwood, such as gums, resins, and salts of organic acids, which therefore could have a different effect on the passage of X-rays.

(h) Difference in photographic image appearance of heartwood and sapwood when specimen is placed in contact with film (Russell Effect). This is a chemical phenomenon based on the presence or absence of unknown reducing volatile materials in the specimen (9).
Most all of these tests were tried on a preliminary basis on specimens with known sapwood-heartwood boundaries before it was decided to utilize only the first five types as noted above.

A partial list of United States suppliers of dyes and stains is listed in Appendix I.

**Materials Tested**

Between 10 and 15 increment cores of 23 different species were obtained through the cooperation of three U.S. Forest Service Experiment Stations. These cores were all untreated and arrived at the Laboratory at approximately 12 percent equilibrium moisture content (E.M.C.). Cores were obtained from the Pacific Northwest Forest and Range Experiment Station, Intermountain Forest and Range Experiment Station, and Pacific Southwest Forest and Range Experiment Station.

**Methods of Investigation**

The cores were placed in trays made of single-faced, corrugated, white paper for easy handling and to provide a white background for the comparison of the color reactions. The trays were 15 inches long and 30 grooves wide, which provided a space of 1 groove between each of the 15 cores. A 1-by 1/4-inch strip of wood was glued across the top and bottom of each tray onto the corrugated side to give added support to the trays and provide a means of numbering the cores.

Each tray contained cores from only one species, and the cores were spot-glued into the grooves with rubber cement. All trays were labeled as to species, and the cores in each tray were numbered. The trays were placed in a room in which the temperature was 80° F. and the relative humidity was 65 percent to condition the specimens to 12 percent E.M.C. The cores that are being tested in the density survey are being received in an air-dry condition (approximately 12 percent E.M.C.). Thus, it was desirable to have all the cores at the same moisture content since this tended to eliminate any variation in reactions caused by different amounts of moisture in different cores of the same species that were collected from different trees on different sites.
A 4- by 6-inch card was made out for each core. On this card was recorded the species, tray number, core number, chemical and date on which it was applied, and color of the sapwood and heartwood immediately after application, after 1 hour, and after 24 hours.

Each of 21 solutions was prepared and placed in a glass-stoppered, 60-millimeter eyedropper bottle. The solutions were applied in a series of continuous drops to the cores.

Solution 1

Alizarine - Iodine

\[
\begin{align*}
69.4 \text{ milliliters methanol} \\
0.3 \text{ gram iodine (crystals)} \\
0.1 \text{ gram Alizarine Red S Indicator} \\
0.5 \text{ milliliter sulfuric acid (concentrated)} \\
29.7 \text{ milliliters water}
\end{align*}
\]

Mix iodine with methanol, add acid to this mixture; mix alizarine with water, add iodine-methanol-acid mixture to alizarine-water mixture; resultant solution is stable.

Solution 2

Alizarine Red S 0.75 percent (14)

\[
\begin{align*}
99.25 \text{ milliliters water} \\
0.75 \text{ gram Alizarine Red S Indicator}
\end{align*}
\]

Solution 3

Ammonium Bichromate 6 percent (5)

\[
\begin{align*}
6 \text{ grams ammonium bichromate} \\
94 \text{ milliliters water}
\end{align*}
\]

Solution 4

Benedict's Solution (5)

\[
\begin{align*}
10.00 \text{ grams sodium carbonate} \\
17.38 \text{ grams sodium citrate} \\
1.73 \text{ grams copper sulfate - } \text{CuSO}_4 \cdot 5\text{H}_2\text{O} \\
\text{Enough water to make 100 milliliters}
\end{align*}
\]

\(^{3}\)Described in a personal communication to R. M. Lindgren, U.S. Forest Products Laboratory, from L. J. Wildes, T&TI, Atlantic Coast Line Railroad Company, Jacksonville, Fla.

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With aid of heat dissolve 17.38 grams of sodium citrate and 10 grams of sodium carbonate in 80 milliliters of water. Filter, if necessary, and dilute to 85 milliliters. Dissolve 1.73 grams of copper sulfate in 10 milliliters of water. Pour this solution, with constant stirring, into the carbonate-citrate solution, and make up to 100 milliliters total volume.

Solution 5

Benzidine (14)
Solution A
1 gram Benzidine
5 milliliters of 25 percent hydrochloric acid
(5 milliliters of concentrated acid plus 2.45 milliliters of water makes 25 percent hydrochloric acid)
194 milliliters water

Solution B
20 grams sodium nitrite
180 grams water

Solutions A and B must be stored in separate bottles; A gradually deteriorates and forms a precipitate. When ready to make test, mix solutions together in equal amounts. Mixture is stable for only 2 or 3 hours, after which reaction to it becomes slower.

Solution 6

Bromcresol Green 0.0413 percent (8)
0.0413 gram Bromcresol Green
100 milliliters water

Solution 7

Bromphenol Blue 0.0413 percent (5)
0.0413 gram Bromphenol Blue
100 milliliters water

Solution 8

Fehling's Solution (5)
3.5 grams copper sulphate - CuSO$_4$.5H$_2$O
17.3 grams potassium sodium tartrate
6.0 grams sodium hydroxide
100.0 milliliters water
Solution 9

Ferric Chloride 10 percent (2)
10 grams ferric chloride
90 milliliters water

Solution 10

Ferric Nitrate 0.1 N (5)
1.346 grams ferric nitrate--Fe(NO₃)₃ · 9H₂O
100 milliliters water

Solution 11

Ferrous Ammonium Sulfate 0.1 N (5)
1.96 grams ferrous ammonium sulfate --Fe(NH₄SO₄)₂ · 6H₂O
0.10 milliliters sulfuric acid
100 milliliters water

Solution 12

Ferrous Sulfate 0.1 N (5)
1.39 grams ferrous sulfate--FeSO₄ · 7H₂O
0.10 milliliter sulfuric acid
100 milliliters water

Solution 13

Harlco Indicator--Hartman-Liddon Company (Appendix I)
Universal Wide Range pH Indicator

Solution 14

Hydrochloric Acid - Methanol (6)
2.5 milliliters hydrochloric acid
100 milliliters methyl alcohol

Solution 15

Iodine 2-1/2 percent (4)
1.25 grams iodine
48.75 milliliters ethyl alcohol
Solution 16
Methyl Orange 0.1 percent (5)
99.9 milliliters water
0.1 gram methyl orange

Solution 17
Perchloric Acid 40 percent (5)
100 milliliters of 70 percent perchloric acid
75 milliliters water
Very caustic, degrades wood

Solution 18
Per-Osmic Acid 1 percent (15)
1 gram Per-Osmic Acid
99 milliliters water
Very caustic, degrades wood

Solution 19
Phenol-HCl-Ethanol + U.V. (1)
10 milliliters phenol
5 milliliters hydrochloric acid
45 milliliters 95-percent ethanol

Melt phenol by heating in a water bath. Add mixture of acid and ethanol to phenol. After application of chemicals to wood, the wood must be exposed to direct sunlight or to rays of an ultraviolet lamp for 2 to 4 minutes to bring about reaction.

Solution 20
Potassium Iodide - Iodine (5)
0.5 gram potassium iodide
24.5 milliliters water
0.5 gram iodine
24.5 milliliters ethanol

Dissolve potassium iodide in water, dissolve iodine in ethanol, mix both solutions together.
Results and Conclusions

The various chemicals tested were rated for each particular species as follows: A, effective; B, fairly good; and C, not useful for differentiating sapwood from heartwood (table 1). N indicates that according to the literature the chemical is not useful and therefore no test was made. The colors given in table 2 are only approximate since they refer to all the species tested. For some tests, the colors in the sapwood and heartwood were actually reversed from those given in the tables but in every one of these cases the sapwood-heartwood boundary was still delineated (5).

In utilizing the results given in table 1, one should keep in mind that in many cases only one test was made with a particular chemical on a single core of a species. Also, very few of the tests, even those rated as A, are infallible, and it is recommended that more than one be used on different areas of the same specimen to determine most positively the sapwood-heartwood boundary.

Very different results might be obtained if the cores that were tested had not been untreated and at 12 percent E.M.C. Some of the factors which can affect the results of similar tests on specimens that do not meet these conditions include (2):

(a) Presence of a preservative.

(b) Immersion in seawater--may result in the deposit of minute quantities of salts, which could give a false reaction.

(c) Presence of extractives in the bark--extractives may diffuse into the wood and interfere with the reaction given by the sapwood.

(d) Green wood from the living tree--may react differently than seasoned wood because of differences in moisture content or chemical changes caused by seasoning.
(e) Freshly exposed surface—may be more effective than an aged or weathered surface.

(f) Abnormal environmental conditions—soil conditions, for example, may affect pH conditions in the growing tree.

(g) Time of felling—presence of sugars and starches in the sapwood is dependent on whether the tree is felled during the growing period or dormant period.

(h) Wet wood—may give a weaker reaction due to a dilution effect.

(i) Presence of wound heartwood, false heartwood, or included sapwood may confuse the test results.

(j) Presence of decay—chemical changes brought about by decay may affect the results.
Literature Cited

(1) American Wood-Preservers' Association

(2) Brown, E. A.

(3) Brown, H. P., Panshin, A. J., and Forsaith, C. C.

(4) Eades, H. W.


(6) Isenberg, I. H., and Buchanan, M. A.

(7) Lindstedt, G.

(8) Lund, A. E., and Sciascia, M.

(9) Nambiyar, V. P. N.

(10) Roberts, E. A. H., and Wood, D. J.

(11) Samuels, R. M., and Glennie, D. W.
(12) Tiemann, H. D.

(13) U.S. Forest Products Laboratory


(15) Wahlberg, H. E.
Appendix I

Partial List of United States Suppliers of Dyes and Stains

Allied Chemical Corporation, National Aniline Division, 40 Rector Street, New York 6, N.Y.

Chroma (formerly G. Grübler & Company), Roboz Surgical Instrument Company, 2500 Wisconsin Avenue, Northwest, Washington 7, D.C.

Difco Laboratories, Inc., 920 Henry Street, Detroit 1, Mich.

Eastman Kodak Company, Chemical Sales Division, Rochester, N.Y.


Hellige, Inc., 877 Stewart Avenue, Garden City, N.Y.

Matheson Coleman & Bell, Division of the Matheson Company, Norwood, O.

Merck & Company, Inc., Rahway, N. J.

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1This list was prepared for informational purposes and the inclusion of names in it implies no endorsement as to quality and prices. The list is known to be incomplete and, upon request, other names will be added at the time of the next revision.
Table 1.—Rating of indicator solutions used for heartwood-sapwood differentiation in some softwood species

<table>
<thead>
<tr>
<th>Species</th>
<th>Solution numbers²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cedars</td>
<td></td>
</tr>
<tr>
<td>Alaska-cedar</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td>Incense-cedar</td>
<td></td>
</tr>
<tr>
<td>Port-Orford-cedar</td>
<td></td>
</tr>
<tr>
<td>Western redcedar</td>
<td></td>
</tr>
<tr>
<td>Firs</td>
<td></td>
</tr>
<tr>
<td>Subalpine fir</td>
<td></td>
</tr>
<tr>
<td>Grand fir</td>
<td></td>
</tr>
<tr>
<td>Noble fir</td>
<td></td>
</tr>
<tr>
<td>Pacific silver fir</td>
<td></td>
</tr>
<tr>
<td>California red fir</td>
<td></td>
</tr>
<tr>
<td>Shasta red fir</td>
<td></td>
</tr>
<tr>
<td>White fir</td>
<td></td>
</tr>
<tr>
<td>Hemlocks</td>
<td></td>
</tr>
<tr>
<td>Mountain hemlock</td>
<td></td>
</tr>
<tr>
<td>Western hemlock</td>
<td></td>
</tr>
<tr>
<td>Pines</td>
<td></td>
</tr>
<tr>
<td>Jeffrey pine</td>
<td></td>
</tr>
<tr>
<td>Lodgepole pine</td>
<td></td>
</tr>
<tr>
<td>Ponderosa pine</td>
<td></td>
</tr>
<tr>
<td>Sugar pine</td>
<td></td>
</tr>
<tr>
<td>Western white pine</td>
<td></td>
</tr>
<tr>
<td>Spruces</td>
<td></td>
</tr>
<tr>
<td>Engelmann spruce</td>
<td></td>
</tr>
<tr>
<td>Sitka spruce</td>
<td></td>
</tr>
<tr>
<td>Douglas-fir</td>
<td></td>
</tr>
</tbody>
</table>

(Sheet 1 of 2)
Table 1.--Ratios of indicator solutions used for heartwood-sapwood differentiation in some softwood species--Con.

<table>
<thead>
<tr>
<th>Species</th>
<th>Solution numbers 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Redwood</td>
<td>B : A : B : B : B : A : B : A : B : C : C : A : A : A : B : A</td>
</tr>
</tbody>
</table>

1 A means effective; B, fairly effective; C, not useful; and N, not useful, according to literature.

2 1 - Alizarine - Iodine
   2 - Alizarine Red S .75 percent
   3 - Ammonium Bichromate 6 percent
   4 - Benedict's Solution
   5 - Benzidine
   6 - Bromcresol Green .0413 percent
   7 - Bromphenol Blue .0413 percent
   8 - Fehling's Solution
   9 - Ferric Chloride 10 percent
   10 - Ferric Nitrate .1 N
   11 - Ferrous Ammonium Sulfate .1 N
   12 - Ferrous Sulfate .1 N
   13 - Harleco Indicator
   14 - Hydrochloric Acid - Methanol
   15 - Iodine 2-1/2 percent
   16 - Methyl Orange .1 percent
   17 - Perchloric Acid 40 percent
   18 - Per-Osmic Acid 1 percent
   19 - Phenol-HCl-Ethanol + U.V.
   20 - Potassium Iodide - Iodine
   21 - Triplex Soil Indicator (Sheet 2 of 2)
Table 2.—Color reactions of sapwood and heartwood of some softwoods in response to various chemical indicators

<table>
<thead>
<tr>
<th>No.</th>
<th>Chemical</th>
<th>Sapwood</th>
<th>Heartwood</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alizarine - Iodine</td>
<td>Green to yellow</td>
<td>Brown</td>
</tr>
<tr>
<td>2</td>
<td>Alizarine Red S .75 percent</td>
<td>Red</td>
<td>Yellow to orange</td>
</tr>
<tr>
<td>3</td>
<td>Ammonium Bichromate 6 percent</td>
<td>Orange to yellow</td>
<td>Brown</td>
</tr>
<tr>
<td>4</td>
<td>Benedict's Solution</td>
<td>Green</td>
<td>Brown</td>
</tr>
<tr>
<td>5</td>
<td>Benzidine</td>
<td>Yellow to brown</td>
<td>Red</td>
</tr>
<tr>
<td>6</td>
<td>Bromcresol Green .0413 percent</td>
<td>Yellow to green</td>
<td>Green to brown</td>
</tr>
<tr>
<td>7</td>
<td>Bromphenol Blue .0413 percent</td>
<td>Brown</td>
<td>Blue</td>
</tr>
<tr>
<td>8</td>
<td>Fehling's Solution</td>
<td>Green</td>
<td>Brown</td>
</tr>
<tr>
<td>9</td>
<td>Ferric Chloride 10 percent</td>
<td>Green</td>
<td>Brown to black</td>
</tr>
<tr>
<td>10</td>
<td>Ferric Nitrate .1 N</td>
<td>Blue to green</td>
<td>Brown to gray</td>
</tr>
<tr>
<td>11</td>
<td>Ferrous Ammonium Sulfate .1 N</td>
<td>Blue to green</td>
<td>Gray</td>
</tr>
<tr>
<td>12</td>
<td>Ferrous Sulfate .1 N</td>
<td>Blue to green</td>
<td>Blue to gray</td>
</tr>
<tr>
<td>13</td>
<td>Harleco Indicator</td>
<td>Red to orange</td>
<td>Pink to brown</td>
</tr>
<tr>
<td>14</td>
<td>Hydrochloric Acid - Methanol</td>
<td>Yellow</td>
<td>Purple to brown</td>
</tr>
<tr>
<td>15</td>
<td>Iodine 2-1/2 percent</td>
<td>Yellow to green</td>
<td>Orange to brown</td>
</tr>
<tr>
<td>16</td>
<td>Methyl Orange .1 percent</td>
<td>Orange to yellow</td>
<td>Red to orange</td>
</tr>
<tr>
<td>17</td>
<td>Perchloric Acid 40 percent</td>
<td>Green</td>
<td>Dark green to brown</td>
</tr>
<tr>
<td>18</td>
<td>Per-Osmic Acid 1 percent</td>
<td>Black</td>
<td>Brown</td>
</tr>
<tr>
<td>19</td>
<td>Phenol-HCl-Ethanol + U.V.</td>
<td>Red</td>
<td>Brown</td>
</tr>
<tr>
<td>20</td>
<td>Potassium Iodide - Iodine</td>
<td>Brown to green</td>
<td>No change</td>
</tr>
<tr>
<td>21</td>
<td>Triplex Soil Indicator</td>
<td>Green to yellow</td>
<td>Green to yellow</td>
</tr>
</tbody>
</table>