

# Pros and Cons of Methods Used to Test the Antimicrobial Behavior of Wood

Created by: 🚱 Muhammad Tanveer Munir 1

1, mtmunir@outlook.fr

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Some wood species have antimicrobial properties and a variety of methods are used to investigate them. For testing the effect of antimicrobial compounds, direct and indirect methods are used. Meanwhile, the survival of microbes is also tested on surfaces (or material in contact with microbes) to validate the safety of surfaces.

Some wood materials show antimicrobial properties <sup>[1][2]</sup>. This potential can be employed for hygienic purposes<sup>[3][4][5]</sup>. Therefore, researchers and field experts study the wood material to investigate its hygienic safety and search for new natural antimicrobial compounds. Many factors influence the choice of method selection to study the antimicrobial properties of wood materials<sup>[6]</sup>. These factors are related to the availability of experimental material, test samples, the purpose of study, and skills of handlers. The advantages and disadvantages of the methods are summarized in Table 1.

Table 1. Pros and cons of the methods used to study the antimicrobial behavior of wood material.

	Method Name	Procedure	Advantage	Disadvantage
Direct methods	Direct diffusion method (Well and disc)	The wood material is directly placed on microbe-inoculated agar or in a well and incubated for recommended time Presence of the zone of inhibition is considered a positive result	<ol> <li>Rapid and time saving</li> <li>Applicable for low amount of material</li> <li>Adapted for screening</li> </ol>	<ol> <li>Disc preparation time</li> <li>High variability for quantitative</li> <li>applications</li> <li>Studies only the</li> <li>effect of agar-diffused</li> <li>chemicals</li> <li>May require the</li> <li>sterilization of wood</li> <li>samples</li> </ol>
	Culture-based microbial survival test	Initial microbial quantity is inoculated on wood samples and after the incubation time, the microbes are recovered, cultured, and viable cells are counted	<ol> <li>Can study the structural and chemical role of wood components</li> <li>Qualitative and quantitative results</li> <li>Applicable for low amount of material</li> </ol>	<ol> <li>Difficulty in recovering all microbes present in pores</li> <li>Microbial quantification is an extra step needed</li> <li>Only viable cells are identified, while there can be still non-viable infectious cells present</li> </ol>
	Microscopy	The behavior and distribution of inoculated microbes on wooden structures is observed via microscopy	<ol> <li>Rapid and time saving</li> <li>Applicable for low amount of material</li> <li>Adapted for screening</li> </ol>	<ol> <li>May require the fixation of samples</li> <li>Difficult to differentiate microbial structures from wooden structures</li> <li>May require competencies of image analysis</li> </ol>

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	ATP luminescence	The ATP of microbes on wood is measured	<ol> <li>Rapid and easy</li> <li>Applicable for low amount of material</li> <li>Adapted for screening</li> </ol>	<ol> <li>Difficult to differentiate the microbial ATP from other organic debris</li> <li>Adapted only for solid surfaces</li> </ol>
	Molecular biology methods	The quantity and viability of microbes is tested via nucleic acid amplification	Accurately measures the microbial survival	<ol> <li>Expensive</li> <li>Require sophisticated handling</li> </ol>
Extractive based methods	Extractive- based diffusion and dilution method	Extractives are placed on agar or in agar wells, or in broth, after loading on filter paper discs or directly	<ol> <li>Adapted for qualitative and quantitative antimicrobial studies</li> <li>Specific chemicals can be extracted depending upon the solvent used</li> </ol>	<ol> <li>Involves chemical handling</li> <li>Extra step of extraction</li> <li>One solvent cannot extract all active components</li> <li>Does not study the role of structure of wood</li> </ol>
	Bioautography	Extractives are loaded on a chromatographic layer, and then the diffusion of active chemicals is studied for their antimicrobial properties	<ol> <li>Adapted for qualitative antimicrobial studies</li> <li>Specific chemicals can be extracted depending upon the solvent used and identified on the basis of their diffusion on the chromatographic layer</li> </ol>	<ol> <li>Involves chemical handling and extraction</li> <li>One solvent cannot extract all active components</li> <li>Does not study the role of structure of wood</li> <li>Not a quantitative method</li> </ol>
	Mass spectrometry	The total profile of microbes is measured	<ol> <li>Applicable for a low amount of material</li> <li>Accurately measure the content of the active ingredient</li> </ol>	For more specific results, the identified compounds are supposed to be tested by other culture-based methods

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#### Keywords

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Natural material; wood; antimicrobials; AMR; Extraction; Microbial survival; Natural compounds; Diagnostics; Discovery



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