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INSIDE

Science
FINDINGS

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"Science affects the way we think together."
Lewis Thomas

Alluring Signals: The Role of Ethanol in Attracting Bark Beetles to Heat-Stressed Trees

Janet Kelsey

Retired Forest Service scientist Rick Kelsey collects bark beetles captured in funnel traps following a controlled burn in Oregon. Forest managers design fuel-reduction treatments to achieve specific outcomes. Understanding the interplay between tree response to heat stress and certain insects can help ensure the treatments are successful.

"There is probably a beetle view of things of which a man is entirely ignorant."
— G.K. Chesterton

Healthy trees have natural physical and chemical defense systems against bark beetle attacks. The flow of tree resin, for example, can physically trap and kill beetles after they bore through the bark, and some resin compounds are directly toxic to the beetles. But, when trees are stressed by drought, disease, physical damage, or wildfire, their defenses are impaired and they become more vulnerable to beetle colonization.

Rick Kelsey, a research forester with the USDA Forest Service Pacific Northwest

IN SUMMARY

Foresters have long known that trees under stress from fire injury are vulnerable to bark beetle attacks. Rick Kelsey and Doug Westlind, researchers with the Pacific Northwest Research Station, have developed a new mechanistic model to explain how physiological changes cause heat stress in woody tissues exposed to sublethal fire temperatures and produce a chemical signal that attracts some bark beetles.

When heat disrupts normal cell functions and energy production, ethanol is synthesized as a short-term survival strategy. If enough ethanol accumulates and is released to the atmosphere (mixed with resin volatiles), it becomes a strong attractant for red turpentine beetles, which target tree locations with high ethanol concentrations.

Kelsey and Westlind have shown that ethanol interacts synergistically with 3-carene, a dominant ponderosa pine resin monoterpene. In a trapping study, red turpentine beetles were more attracted to lures combining ethanol and 3-carene than lures with ethanol or 3-carene alone.

Understanding ecosystem responses to fire can help managers characterize forest health and plan for postfire management. The results also hold promise for developing simple ethanol detection methods for monitoring tree stress. Real-time feedback on ethanol levels could help forest managers quickly assess which trees to leave following a fire and which to cull.

Doug Westlind

The red turpentine beetle is the largest bark beetle in western North America and widely distributed in pine forests.

KEY FINDINGS

- A new mechanistic model proposes that the stress woody tissues experience when exposed to sublethal temperatures between 86 °F to 140 °F (30°C to 60°C) during a fire is a decrease in normal energy production by aerobic respiration. The decrease in aerobic respiration, and induction of ethanol synthesis, is a result of one or more physiological impairments to: (1) the cell's oxygen supply, (2) membrane functions, or (3) enzyme activities.
- Ethanol production is a key indicator of tree stress.
- Red turpentine beetles are attracted to fire-injured trees and attack specific locations on trees with elevated ethanol concentrations.
- In a trapping study, red turpentine beetles were weakly attracted to ethanol lures and strongly attracted to lures releasing 3-carene, a dominant pine monoterpene. But, they responded in greatest numbers to ethanol plus 3-carene lures.

(PNW) Research Station, has been studying the chemical ecology of trees since the late 1980s. His research has been instrumental in revealing how some bark beetles identify which trees are stressed and thus most susceptible to an attack.

Kelsey was the first to describe increased ethanol concentrations in trees injured by fire or taxed by drought and then link the elevated ethanol levels to attacks from different bark beetle species. Some bark beetles, including red turpentine beetles (*Dendroctonus valens*), are able to detect locations on tree trunks with elevated ethanol concentrations.

Red turpentine beetles are widely distributed in U.S. pine forests, and are especially attracted to fire-injured trees. Foresters have even reported seeing the beetles attack trees while there are still flames on the ground. Although red turpentine beetle attacks rarely kill the tree directly, some do carry spores of fungal pathogens that can cause disease in the tree. Red turpentine beetle attacks why also further weaken a tree, making it more vulnerable to other stressors.

Recently, working with his colleague Doug Westlind at the PNW Research Station, the researchers expanded on Kelsey's earlier studies and developed a much more complete picture of the triangular relationship between fire and heat stress, ethanol production in trees, and red turpentine beetle attacks.

"My work is typically an iterative process," Kelsey says. "We do an experiment, interpret the results, and generate more questions—how do we explain this or what does this mean? And then we design some more experiments to kind of poke the system again to see how it responds—that's really how this developed."

Their latest work has expanded our understanding of red turpentine beetle attacks, showing that they are extraordinarily precise in targeting specific locations on the tree with elevated ethanol concentrations. Their findings reveal that ethanol doesn't work alone to signal insects. Ethanol interacts synergistically with 3-carene, a chemical compound often found in the resin of ponderosa pine. This has practical implications for forest managers wanting to know which trees beetles are likely to select as hosts for colonization.

Building the Foundation

In 2003, Kelsey conducted a study that measured ethanol levels in the phloem and sapwood of ponderosa pines damaged in a wildfire two weeks earlier. The degree of fire injury was categorized as zero (control), moderate, heavy, or severe, based on the observed level of crown scorch. The researchers found that ethanol levels were 15 to 53 times higher in the severely scorched pines compared to those that weren't burned.

During this study, Kelsey also observed that bark beetles landed on the fire-damaged trees in greater numbers than on the nonburned trees, and that spring landing rates were related to ethanol concentrations in the sapwood of trees burned the previous fall. This was an early indication that ethanol was influencing beetle behavior. Still, Kelsey wanted to know how to quantify the behavior they were observing.

"I suspected a strong relationship between ethanol and red turpentine beetle attacks, but initially I wasn't sure how to set up the study design to prove it," says Kelsey.

In their first attempt, they didn't sample close enough to the beetle's entrance hole.

"Ethanol production and release in woody tissue is highly variable," Kelsey says. "If you sample two locations 1 or 2 inches apart on the tree trunk, you can get quite different results."

So, Kelsey and Westlind designed another study, sampling as quickly as possible after the fire, and very close to the point where the beetles were boring into the bark. The same experiments were conducted after two fires, a June wildfire and an October prescribed fire near Bend, Oregon. They sampled the phloem and sapwood right above the point of

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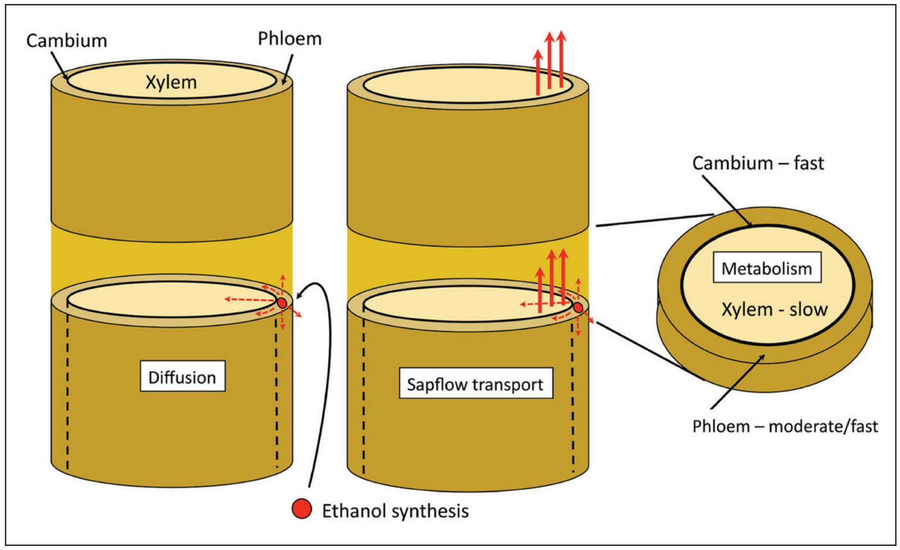
Doug Westlind

Pitch tubes showing sites of red turpentine beetle attacks. Attacks are typically confined to the lower 6 feet of the tree.



Kelsey and Westlind 2017c

As ethanol accumulates in the tree, it immediately begins to dissipate via (1) diffusion, (2) sapflow, and (3) metabolism. Each process is affected differently by the heat-stress mechanism the tissues and whole tree experience.

attack, on the opposite side of the tree where there were no attacks, and on an adjacent tree with a similar level of injury that had not been attacked.

The results were quite clear.

Ethanol concentrations were much higher above the point where the beetle had bored into the tree, compared to adjacent nonattacked trees in both types of fire. They also found that ethanol concentrations were not uniform throughout a burned tree, and beetles selected and attacked trees—and specific positions on the tree—where ethanol levels were highest.

"They were very precise in selecting the spot to attack on the stem," says Westlind.

Kelsey and Westlind followed that work with a study investigating the role of 3-carene in attracting red turpentine beetles. 3-carene is a compound found in the oleoresin of many pine species. Other studies had shown that 3-carene attracts red turpentine beetles more than other terpenes, but no one had ever examined beetle attraction to a combination of ethanol and 3-carene.

The researchers set up a relatively simple study in a pine forest in Oregon with funnel traps containing a variety of lures: ethanol, 3-carene, ethanol plus 3-carene, and traps with no lures as a control. They found that red turpentine beetles were weakly attracted to lures with just ethanol and strongly attracted to lures releasing 3-carene. However, the beetles responded in greatest numbers to the lures releasing a mixture of both ethanol and 3-carene. In fact, Kelsey says the attraction results met the criteria for a synergistic response. In other words, the catch was larger than you would expect from ethanol separately and 3-carene separately, if added together.

The Mechanics of Heat Stress at Sublethal Temperatures

Interpretation of the ethanol concentrations measured from trees burned in June and October, combined with Kelsey's work on other tree stressors inducing ethanol production, as well as related research results in the literature, led to the development of a new, detailed mechanistic explanation of the physiological processes causing heat stress in woody tissues when subjected to sublethal temperatures (≤ 140 °F; 60 °C).

All normal physiological processes in living plant or animal cells are dependent on the chemical energy stored in ATP, or adenosine triphosphate, which is produced during aerobic respiration in the mitochondria. Any disruption limiting this energy supply will stress the cells's functions.

The lethal temperature limit for most plant tissues is recognized as 1 minute at 140 °F (60 °C). Exposure to lower temperatures may result in heat stress. Kelsey and Westlind theorize that when woody tissues are subjected to temperatures between 86 °F to 140 °F (30 °C to 60 °C), changes can occur in affect tissue oxygen supplies, membrane functions, or enzyme activities; if these changes impair normal mitochondrial functions, aerobic respiration decreases. When aerobic ATP production declines or stops, ethanol synthesis begins with a very limited production of ATP, but just enough to keep the cells alive until the stress has passed, and normal aerobic respiration has resumed.

"Loss of energy is the stress," Kelsey says. "But there are multiple physiological processes influenced by heat that can ultimately impair mitochondrial energy production and subsequent ethanol synthesis."

When ethanol is present in woody tissue, it's an indicator that there's been a limit on respiration, or normal cell functions, Kelsey explains.

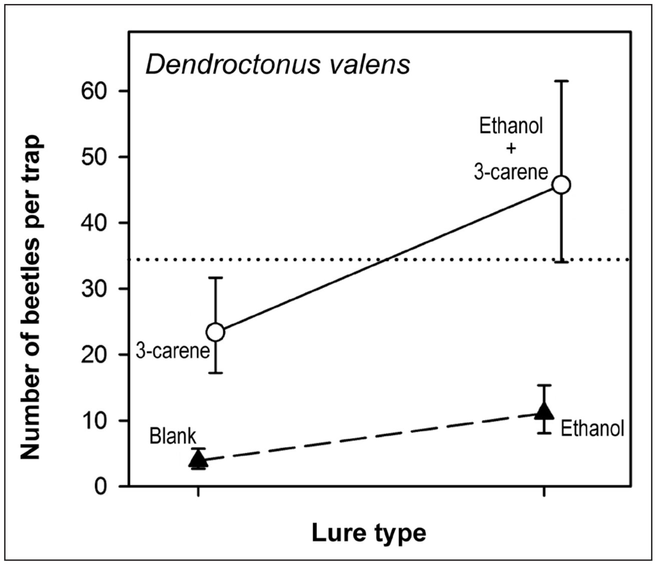
Understanding the mechanics of heat stress provides land managers with a more thorough understanding of fire effects. Fuel reduction treatments can be adjusted or designed to ensure desired outcomes are more likely achieved. And after a fire, the identification of heavily stressed trees can be used to help determine which to leave and which to cull.

New Clues for Management

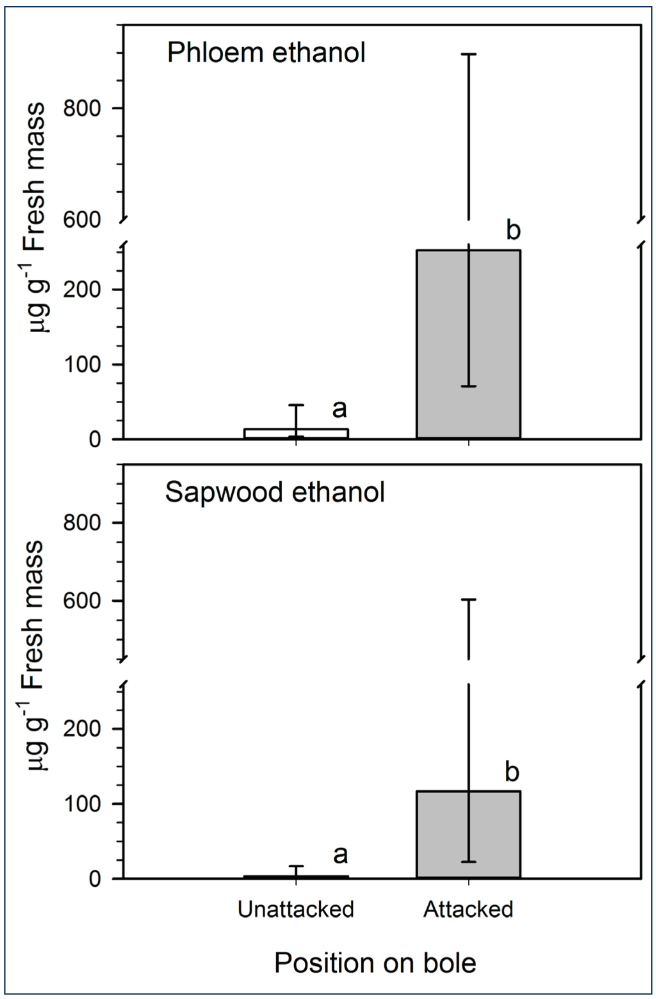
Dave Shaw is a forest health specialist with forest and natural resources extension at Oregon State University who works with county forestry extension agents, forestry professionals, and small woodland owners and tree farmers. He says the work by Kelsey and Westlind on fire, stress, and beetles will be welcomed by forest owners and professionals

Janet Kelsey

Funnel traps were set up with lures of ethanol, 3-carene, and a combination of ethanol and 3-carene to measure beetle attraction to the compounds. More beetles were caught in the ethanol plus 3-carene lures than in the combined catches of the only ethanol or only 3-carene lures



Mean number of red turpentine beetles trapped by lure type. Dotted line indicates the sum of beetle catch from 3-carene and ethanol lures. Adapted from Kelsey and Westlind 2017a.



The same wildfire-stressed tree, differing ethanol concentrations on either side of the trunk (bole). The red turpentine beetles targeted the side of the tree with the higher concentration of ethanol. Adapted from Kelsey and Westlind 2017b.

who are always looking for guidance in making forest management decisions.

"This research provides landowners with one more piece of information in characterizing the health of their forests," Shaw says. "They might want to do some management to reduce stress in an overstock situation, or to remove trees that are more likely to die after a fire. This helps them identify 'leave' trees and 'take' trees."

The research could also spur further development of simple ethanol detection methods for use in early recognition of tree stress. Shaw half-jokingly envisions a future handheld device, much like the tricorder on Star Trek, which could measure tree stress.

"It could give readings of chemicals released in the air, so a forester could say, 'okay, these trees are really stressed," Shaw says. "That would be fascinating. Short of that, I think understanding some of these fundamentals will help landowners understand why they might need to thin or salvage."

The confirmation that ethanol is a primary attractant for red turpentine beetle, especially when mixed with 3-carene, can be used to create a valuable tool for protecting North American forests from foreign beetle-borne tree pathogens. Red turpentine beetles are native to North America, but have become invasive in China, attacking their native pines. In China, red turpentine beetles are associated with pine mortality over large areas and have picked up the spores of tree fungal pathogens not found here. There is concern that beetles carrying these pathogens could be reintroduced to North America.

"If nonnative fungi are brought back here and picked up by a native beetle—oh my, that could be a complete disaster," says Shaw.

Kelsey and Westlind suggest that the combined ethanol plus 3-carene lures used in their study could help in early detection trapping surveys at ports of entry where red turpentine beetles from Chinese wood products are most likely to arrive.

Overall, research on the keystone role of ethanol in understanding physiological processes underlying heat stress in trees, as well as in host selection by red turpentine beetles, makes valuable basic scientific contributions while also having valuable management applications.

"It's that old story you probably heard, we need the foundation to build a house on it," says Shaw. "And the fundamental work that Rick and Doug have been doing is the foundation. We'll be building everything on this important ethanol link."

"Why do we study insects? Because, together with man, hummingbirds and the bristlecone pine, they are among the great achievements of organic evolution."

—Edward O. Wilson

LAND MANAGEMENT IMPLICATIONS

- Understanding heat stress physiology in tree woody tissues can contribute to successful forest management both pre- and postfire.
- By understanding how bark beetles select fire-injured trees for colonization, land managers can make more informed treatment decisions.
- Red turpentine beetles have become an invasive species in China, where they carry a new community of fungal spores, including forest pathogens. There is concern for potential reintroduction of these beetles carrying any new invasive pathogens to North American pine forests. Traps with lures containing ethanol and 3-carene could help in early detection and management of reintroduced beetles.

Further Reading

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Scientist Profiles

RICK KELSEY is a recently retired research forester with the Pacific Northwest Research Station. As an emeritus scientist, his research continues to focus on understanding the complex interactions between trees and the biotic (insects, pathogens) and abiotic (fire, drought, etc.) agents that cause them stress. His other research is directed toward discovering compounds in tree tissues with biological activity and understanding their ecological importance. Kelsey received his Ph.D. in forestry from the University of Montana.

DOUG WESTLIND has been a forestry technician with the Pacific Northwest Research Station since 1991. His early work with the station focused on laminated root rot in Douglas-fir, but since 1997 he has worked on the interactions of fire, insects, and diseases, including bark beetles and black stain root disease in ponderosa pine. He combines extensive field experience with a background in experimental design and statistics. He has a degree in geology from Oregon State University.

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