

# **Tobacco aside, e-cigarette flavorings may harm blood vessels**

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Flavor additives used in electronic cigarettes and related tobacco products could impair blood vessel function and may be an early indicator of heart damage, according to new laboratory research in

*Arteriosclerosis, Thrombosis and Vascular Biology*, an American Heart Association journal.

The health effects of "combustible" tobacco products including traditional cigarettes and hookah are well-established, but the potential dangers of e-cigarettes have not yet been extensively studied. E-cigarettes are battery-powered devices that heat a liquid—including tobacco-derived nicotine, flavoring and other additives—and produce an aerosol that is inhaled.

Nine chemical flavorings—menthol (mint), acetylpyridine (burnt flavor), vanillin (vanilla), cinnamaldehyde (cinnamon), eugenol (clove), diacetyl (butter), dimethylpyrazine (strawberry), isoamyl acetate (banana) and eucalyptol (spicy cooling) - which are widely used in e-cigarettes, hookah, little cigars and cigarillos were tested for their short-term effects on endothelial cells, the cells which line the blood vessels and the inside of the heart.

Researchers found all nine flavors were dangerous to cells in the laboratory at the highest levels tested and all the flavorings impaired nitric oxide production in endothelial cells in culture (outside of the body). Several of the flavorings—menthol, clove, vanillin, cinnamon and burnt flavoring—resulted in higher levels of an inflammatory marker and lower levels of nitric oxide, a molecule that inhibits inflammation and clotting, and regulates vessels' ability to widen in response to greater blood flow.

"Increased inflammation and a loss of nitric oxide are some of the first changes to occur leading up to cardiovascular disease and events like heart attacks and stroke, so they are considered early predictors of heart disease," said lead study author Jessica L. Fetterman, Ph.D., assistant professor of medicine at Boston University School of Medicine in Massachusetts. "Our findings suggest that these flavoring additives may

have serious health consequences."

Endothelial cells were collected from volunteers (nine non-smokers/non-e-cigarette users; six non-menthol and six menthol cigarette smokers) and tested in the lab. Researchers found that both groups of smokers had a similar deficit in nitric oxide production when stimulated by a chemical called A23187. Nonsmokers' cells that were treated with menthol or a clove flavoring also had impaired nitric oxide production, suggesting those flavorings cause damage like that found in active smokers.

The team also exposed commercially-available human aortic endothelial cells to the flavorings. Burnt flavor, vanilla, cinnamon and clove flavors impaired nitric oxide production and boosted an inflammatory chemical called interleukin-6 (IL-6) at all concentrations tested, suggesting the endothelium is particularly sensitive to these flavors.

Menthol applied to the cells increased IL-6 at high concentrations and reduced nitric oxide even at low doses. In smokers, scientists don't see differences in heart disease between menthol and non-menthol users—probably because cigarette smoke is overwhelmingly toxic, Fetterman said. "But menthol is certainly not a benign player, based upon our work."

At the highest levels tested, all nine chemicals caused cell death, while at lower levels cinnamon, clove, strawberry, banana and spicy cooling flavor did. Dimethylpyrazine/strawberry flavor had that effect even at very low levels, suggesting endothelial cells are especially sensitive to it. Vanillin and eugenol also increased oxidative stress in the cells.

Three flavorings were tested when heated, to mimic what happens in e-cigarettes. Nitric oxide production was impaired with vanillin and eugenol, but not with menthol.

"Our work and prior research have provided evidence that flavorings induce toxicity in the lung and cardiovascular systems. Flavorings are also a driver of youth tobacco use and sustained tobacco use among smokers," Fetterman said.

A key strength of the new research was that it directly tested effects of just the flavorings, at levels likely to be reached in the body. Limitations include the fact that testing did not heat all the flavorings or include other chemicals used in e-cigarettes. Also, the study gauged just the flavorings' short-term effects and captured these with cells outside the body, not inside.

"We still don't know what concentrations of the flavorings make it inside the body," Fetterman said.

Most adult e-cigarette users are current or former combustible cigarette smokers who may use e-cigarettes as an aid in smoking cessation or as a harm-reduction tool. In addition, e-cigarette use by youth is rising rapidly with 37 percent of high schoolers reporting they have had an e-cigarette in 2015. Flavored tobacco products are a major driver of experimentation among youth.

The American Heart Association cautions against the use of e-cigarettes, stating that e-cigarettes containing nicotine are tobacco products that should be subject to all laws that apply to these products. The Association also calls for strong new regulations to prevent access, sales and marketing of e-cigarettes to youth, and for more research into the product's health impact.

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