What Causes Buddy Syrup and What Can Be Done to Prevent it?

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Ontario has more than 3,000 farms where maple syrup production is more than 50% of cash receipts. The Ontario Maple Syrup Producers' Association (OMSPA) has invested in basic research in a number areas including the diversity of molds that are found in maple syrup and the factors that increase risk for post-consumer mold damage (Int J Food Microbiol 207:66). The next big problem we began to tackle was to see if a way could be found to detect the changes in maple sap that leads to "buddy syrup" before the sap was even boiled.

Buddy off-flavour is an annual, natural occurrence that has been well recognized since the dawn of commercial maple production in the late 19th century. The sap is collected and processed, consuming fuel and other resources, but is ultimately not suitable for sale due to an off flavor. For individual producers, as much as 10% of annual income can be lost in some years as a result of some stopping too early in order to avoid producing buddy syrup, and others producing unsalable syrup. Currently, producers rely on guesswork to try to determine when to stop collecting and processing sap. Common responses of producers of strategies to avoid buddy syrup range from noting the height of wild leeks in the bush, the sounds of the spring peepers, or when the maple buds have started to swell and show some green. As seasonal winter-into-spring weather patterns are changing, dealing with this problem is expected to become more challenging in the future.

Sixty years ago, USDA researchers suggested the mobilization of amino acids into sap before budding was the reason for the appearance of buddy syrup. To that end, an early USDA research used paper chromatography of maple syrup stained with ninhydrin reagent, which reacts with amino acids. This never made it into practice as testing for buddy flavours after processing does not offer an economic benefit to producers who are capable of identifying the off-flavour by palate alone.

As we began our investigation there were two basic ideas for the sudden appearance of buddy syrup. The first was that heating sap containing elevated levels of particular amino acids produced compounds (pyrazines) that contributed to buddy off flavour. A more recent idea has been that yeasts in the sap convert sulfur-containing amino acids into compounds that explain the off flavours.

The project was undertaken in two phases. First, OMSPA arranged for two maple producers in the 2018 season to collect samples of sap over a period up to the point that buddy syrup was de-

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tected. Antimicrobials were added to the samples which were stored cold. The samples were analyzed with a sophisticated liquid chromatography-mass spectrometer (Thermo Q-Exactive Orbitrap mass spectrometer coupled to an Agilent 1290 HPLC system). This was used to look for potential chemical markers in the sap that coincided with the development of buddy flavour in the syrup. The focus was on changes in the concentrations of 18 amino acids and a suite of sugars over the course of the growing season. This initial work suggested that a number of nitrogen rich amino acids and some sulfur containing amino acids showed marked increases in concentration up to the budding stage.

The second phase involved a systematic study from more stands across the 11 OMPSA districts. We knew that fundamental characteristics of the syrup vary considerably between stands in Ontario. These include the yeast mycoflora, ions and pH, all of which vary considerably according to soil chemistry and site history.

The chemical composition of maple syrup and, to a lesser extent, maple sap has been investigated for decades. In previous studies of sugar maple sap, known chemicals were targeted for analysis. The decrease of sucrose content in maple sap in late season sap has previously been observed. The causative agent of this decrease at the end of the maple production season appears to be the result of microbial activity. In 1947, Holgate reported that the sugar content diminished in the late season, conversely, when the sap was collected aseptically, the total sugar percent remained above 2% at the end of the season. Holgate found that the nitrogen content of sap increased towards the end of the season regardless of sterility.

We found that sucrose was present in much higher concentrations at the beginning of the sap run and in much lower concentrations in late spring. In addition to sucrose, and the monosaccharides fructose and glucose, two major tri-saccharides were also detected (see figure). A number of more complex sugars formed the remaining part of the picture.

In contrast, amino acids, particularly asparagine and methionine increased in late season sap. Some researchers have proposed that the small sulphur-containing compounds (similar to a skunk's spray) may be responsible for the late season, 'buddy' off-flavor. We detected a sulfur-containing amino acid in late season sap called Methionine. If the sap was heated to high temperatures, the methionine can decompose into a chemical some of which can be converted to dimethyl disulfide, which

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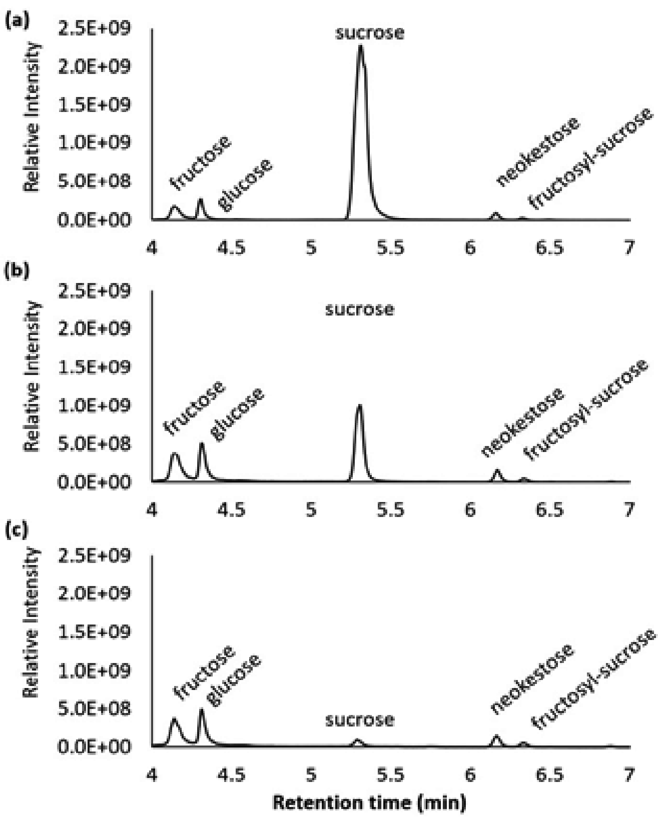

Figure 1: (a) In early season sap, sucrose was the major saccharide detected, decreased in mid season (b), and was present at low concentrations at the end of the season (c) [PLoS One

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is volatile at room temperatures, and smells bad.

The classes of compounds which most likely contribute to the unfavorable aftertaste in maple syrup made from late season are alkyl pyrazines and sulfides. Pyrazines such as those reported in late season or buddy sap have an aftertaste characterized as 'malty' and 'astringent.' Related compounds are found in raw potatoes that have been stored for a long time. Compounds such as some of the proposed sulfur containing compounds are described as 'peppery' and 'brassica' flavours (similar to Brussels sprouts). Similar to work from Vermont researchers, this basic research supports pyrazine alkaloids as the chemistry of the off flavour. Regardless, as noted methionine and asparagine tended towards greater concentrations later in the season compared to early season values. Asparagine has been shown to be most efficiently converted to pyrazines compared to the other amino acids detected in sap. In contrast, in foods, methionine is typically most important in producing the sulfides. These two amino acids represent strong candidates for the development of poor after taste and thus targets for sap based in situ tests.

Consistent with many previous studies, the nitrogen content was higher quantities in the late season samples. The amino acids asparagine and methionine, both known precursors of off-flavours in food increased considerably in late versus early season sap. One or both of these compounds might be useful markers for sap that will not be salable.

At present we are investigating whether aptamer-based methods for the detection of these two amino acids in sap. Aptamers are like synthetic antibodies that can be used to produce tests like pregnancy test kits. However, they are much less expensive. These can be made into tests on strips of paper like a litmus test for pH. These potentially would be useful to maple producers in the field to monitor the transition to late season sap.

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The full study can be accessed on line:

Garcia EJ, McDowell T, Ketola C, Jennings M, Miller JD, Renaud JB (2020) Metabolomics reveals chemical changes in Acer saccharum sap over a maple syrup production season. PLoS One 15(8):e0235787

https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0235787