This article addresses the feasibility of using an electronic nose to monitor the production of aromas during a Muscatel fermentation.

- **What is an electronic nose?** Developed in the early 1980s, the operating principle consists of an array of chemical sensors that are coupled to an appropriate pattern recognition program that emulates the human olfactory system. (Easy to say!) The individual sensors consist of conductive polymers which have defined adsorptive surfaces that, when interacting with volatile chemicals, display a change of electrical resistance that can be recorded. Even though each individual sensor responds more selectively to a certain group of chemicals, they all show an overlapping response (this is called *cross-selectivity*). How the electronic nose actually works is that, for each complex aroma, the array of sensors produces a unique response pattern -called a “fingerprint”- which reflects the aroma complexity of that sample. An electronic nose, therefore, acts more like a human nose in that it does not measure the amount of an individual aroma compound, but rather, gives a global and qualitative idea of the whole aroma profile.

- **The authors inoculated a 50 L fermentor containing a Muscatel must with *S. cerevisiae* and monitored the fermentation process for 20 days. They used 2 different sampling methods for the electronic nose monitoring: 1) on-line, directly from the fermentor headspace, and 2) off-line, using a static-headspace multisampler, that took samples in sequence and equilibrated them for 1 hour prior to analysis. Each analysis consisted of a cycle of 8 minutes, in which the sensors were exposed to a sequence of different backgrounds.

- **The data collected by the electronic nose was then processed through PCA (principal component analysis).** The way PCA works is it reduces the high multidimensionality of a complex data set into two or three variables —the *principal components*- which contain the majority of the information necessary to discriminate the samples. The result is a 2- or 3-dimensional chart where similar samples can be seen as “clusters”. Finally, the authors also monitored density, redox potential, number of viable cells, glucose and fructose consumption, organic acids production, and —very importantly, as we will see- ethanol production.

- **Monitoring the stages of fermentation.** The PCA results showed different clusters of aroma fingerprints which represented the different days of fermentation, and which allowed the identification of 3 main fermentation stages: 1) an *initial stage*, with must densities from 1080 to 1073 g/L; 2) a *rapid fermentation stage*, with corresponded to a rapid increase in yeast cell numbers, a rapid conversion of sugars into alcohol (causing a fast decline in density from 1073 to 1034 g/L), and the fastest rate of aroma production; and 3) a *final stage*, when the must/wine reached a final density of 992 g/L, and aroma production started to slow down.
• **Interference of ethanol.** But did those clusters truly represent different aromas, or could they actually represent different responses to different levels of ethanol? That is, how accurately could the electronic nose be monitoring the evolution of aromas given the presence of increasing levels of ethanol in the samples? To address that issue, the authors prepared 2 sets of model solutions: one set had increasing levels of ethanol only (Set E, for Ethanol); the other set had the same increasing ethanol levels but also increasing amounts of aroma standards (Set A, for Aroma). The authors were then able to see that the electronic nose was unable to distinguish a given E sample cluster from the equivalent A sample, that is, the high ethanol content was masking the contribution of the aromas. So the clusters the nose had detected up to this point were actually a simple reflection of different ethanol levels. The authors could have derived that information with a simple hydrometer!

• What could be happening? Perhaps the aroma compounds were below the sensor detection limit. The authors thought this explanation could be plausible for aroma compounds known to be produced in small concentrations (linalool, ethyl-hexanone), but not for those expected to be abundant (isoamyl alcohol). They thought it more likely that there was competition between both types of compounds at the surface of the sensor. As the authors explain, because ethanol can act as a co-solvent in the aqueous must solution, it can lower the effective concentration of the aroma compounds, which tend to be hydrophobic. The end result would be that the electronic nose would be detecting less of a given aroma compound as the fermentation proceeds, and ethanol increases, even if the concentration of that compound would have remained the same. This observation pointed out to the authors that they needed to come up with some sort of sample preparation prior to analysis.

• **Sample preparation.** Preparing the sample would have to involve either removing the ethanol, or enriching the aromas. Because the former might involve loss of aroma compounds also, the authors chose the latter method. To enrich their samples in aroma compounds they used a selective membrane separation process called **pervaporation.** This separation process enriches the aroma compounds relative to the ethanol in the permeate, so they can then be more accurately measured by the nose. The permeate obtained for each sample was then diluted back to adjust the ethanol and analyzed using the nose. The result was that the enriched samples could then be clearly discriminated from the corresponding A (aroma+ethanol) and E (just ethanol) samples. That is, combining the pervaporation with the electronic nose, it was now possible to follow the evolution of the aroma compounds through the fermentation.

• When the authors looked at which individual sensors showed the best response to the aroma compounds, it was those with a low sensitivity to ethanol (4 sensors out of 32). According to the manufacturer, these specific sensors are characterized as being most responsive to long-chain compounds, which happens to be the nature of the most relevant aroma compounds.

In conclusion, without sample preparation, the electronic nose could only perceive the evolution of ethanol. But incorporating pervaporation as a selective enrichment step, the electronic nose was able to detect the aroma compounds produced throughout a Muscatel fermentation. The authors believe that this combination of the two methods can also have application in other phases of wine production, such as monitoring wine aging compounds, or the development of off-flavors for quality control. Even though “electronic noses” may still have a long way to go, the sample preparation presented here may help bring them closer to that magnificent thing that is the human nose.

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