**Evaluation of Enology**

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**Introduction**

In most cases, an enological evaluation is instigated due to a flaw in the appearance of the wine (cloudiness, deposits…) or an abnormal taste or aroma. Among these problems, a "moldy" or "musty" flavor or aroma is the most unpleasant defect. All types of wines are affected by this problem and it seems to be ***occurring more frequently these past ten years***.

Corks have been singled out as the guilty element, but it is doubtful that they could be the only cause. As a matter of fact, ***this problem has appeared in numerous cases before wine has been bottled***—while it was stored or ageing in tanks or barrels. In such cases, the classic cause of "storage" is evoked as the explanation for the damage caused to the wine. However, this is not thoroughly convincing when we are talking about new wine cellars with perfectly maintained tanks and irreproachable hygiene. An expert must, therefore, determine the exact nature and origin of the defect because ***any one of a number of components***—all quite different in chemical make-up—***could be the source of the "moldy" aroma***.

Chloroanisoles have been known for a long time to cause intense moldy flavors in wine, cork, Cognac, and many other products. A large variety of derivatives come from a chlorine aromatic ring. Although 2,3,6 Trichloroanisole is surely one of the strongest smelling chloranisoles (sensory threshold perception = 0,1 pg/l in water), 2,4,6 trichloroanisole and 2,3,4,6 tetrachloroanisole have the strongest odors but the weakest sensory threshold perception (0,03 and 4 ng/l in water). Pentachloroanisole is much less odoriferous than the other chloroanisoles (sensory threshold perception = 4 ug/l in water).

Chloroanisoles come from methylation and related chlorophenols. This biochemical reaction comes from many micro-organisms, and most notably mold. In an opposite reaction to that of chloroanisoles, chlorophenols are not odoriferous molecules since they are generally proton related to the wine pH. Chlorophenols can come from several sources. In the past they have been mostly used as insecticides for treating wood, but they could also have other uses.

Many cleaning agents that are recommended for use in the domain of enology contain chlorine. This element in the form of C12, HCIO or CIO according the to pH, can react with phenols in order to create polychlorophenols through a reaction of substitute electrophile. On the other hand, chlorine reduced in the form of chloride ion CI is inactive. Some molds such as penicillium sp is capable of synthesizing 2,4,6-trichlorophénol through shikimic acid with free chlorine or incorporated in an amine acid. This reaction will not work with simple chlorine. This chlorophenol can in turn be transformed into 2,4,6-trichloroanisole by the same micro-organism.

The appearance of cholorphenols has sometimes been interpreted as evidence of organochlorinated pesticide decomposition. The problems encountered with the accumulation of organochlorinated insecticides has resulted in the majority of the molecules of this family being banned and are now replaced by organophosphorous. However, hexachlorocyclohexane isomers (HCH), and Lindane in particular (¡-hexachlorocyclohexane), are still used for certain insecticidal treatments. Several polychlorophenols have been identified as metabolic substances in mammals, but we do not know what the decomposed products are that come from cellar micro-flora. Certain fungicides such as Chloranil (tetrachloro-para-benzo-quinone), (no longer marketed in France), could lead to the development of chlorophenols under certain conditions. Prochlororaz or 1-{N-propyl-N-2 (2,4,6-trichlorophenoxy)-ethyl carbamoyle]—an active material of the fungicide Spotak, which us used widely in agriculture, can decompose to 2,4,6-trichlorophenol then to 2,4,6-trichloroanisole through bacterial methylation and irreparably alter the quality of the products. This molecule has never been allowed for use in vineyards and as far as we know there is no similar structure catalogued in the phytosanitary index. Among those herbicides—non-commercialized molecules in France—derivatives from trichloro-2,3,6-benzoic (TCBA) acid can also decompose into substances that can be metabolized by micro-flora into chloroanisoles, but they have never been allowed in vineyard use. Several derivatives of dithio or trithio-phosphates from chlorophenols, and notably from 2,4,6-trichlorophenol are said to have been authorized in Japan for use as an insecticide for agriculture. As far as we know, no currently used material which make up the composition of those phytosanitary products authorized in viticulture is liable to release chloroanisoles or chlorophenols.

***The defect can appear before or after bottling but detecting it happens most often after bottling***. In order to find the contamination, research needs to go back to the origin. Contaminants can be identified and quantified in even their weakest concentrations (ppb) through the use of high-resolution gas-phase chromatography coupled with highly sensitive mass spectrometry. However, an enological evaluation must begin with a sensory examination.

**Role and Utilization of a Sensory Analysis**

Tasting is always the most essential method of evaluation in enology. In fact, an alleged organoleptic defect is the basis for a complaint. First, the defect needs to be confirmed. Sometimes a claim of an imaginary defect is made in order to apply for some compensation. Once the flaw has been confirmed, the cause must be researched. According to tasting results, an effective research effort can be directed towards certain compounds through instrumental analyses. Despite powerful modern analysis systems, it is not always easy (and sometimes too costly) to establish an undeniable relationship between the presence of certain compounds and tasting results. In this case, sensory analysis and an enologist's opinion will always take precedence.

When the guilty substance is identified and quantified, and before making any conclusion, it is essential to compare the concentrations found through analysis to the value of the sensory threshold perception of the substance in order to assure that the determined elements can indeed be those responsible for the flaw.

**Moldy Flavor Linked to Cellar Contamination**

Chloroanisoles are not normal wine components. Their presence, therefore, constitutes a contamination. From several valid examples of pollution taken from cellar storage, we have researched the exact origin of this wine contamination.

a) Contamination of wine in vats through the air

Through the analysis of wine contaminated during storage in static containers, important amounts of 2,3,4,6-tetrachloroanisole, pentachloroanisole and chlorophenols were detected in those samples deemed "moldy" to the taste. Atmospheric tests of the installations (by trapping air through an absorbent matter) shows evidence of variable contamination by chlorophenols (pentachlorophenol, 2,3,4,6-tetrachlorophenol and sometimes traces of 2,4,6-trichlorophenol) and corresponding chloroanisoles—molecules that are responsible for the "moldy" flavor in wine.

Chloroanisoles have a high boiling temperature but weak steam pressure. This explains why it is easy to find high quantities of contaminants trapped statically or dynamically from the cellar atmosphere on an inert absorbent material (a porous polymer such as substituted di-vinyl benzene, bentonite type clay or activated carbon. Once these compounds are in a gaseous phase, not only can they easily infiltrate the entire atmosphere of the location where the source of the contamination resides, but they can also be diffused in the most distant parts of the cellar thanks to air currents, thereby polluting the areas that were not previously contaminated.

In a gaseous phase, the contaminants can easily dissolve in the wine during a pumping stage, due to the inevitable gaseous emulsion that is inherent in this operation. The fluctuation of volume of stored wine along with the temperature variations of the cellar can also affect the dissipation of the tainted air in the vats, tanks or barrels, thus contaminating the wine. The alteration of the wine is significant when considering concentrations levels of 10ng/l for 2,4,6-trichloroanisole and around 100 ng/l for 2,3,4,6-tetrachloroanisole. Pentachloroanisole has much less smell but we don't know what the eventual additive effects are, or what synergy there could be with other anisoles.

One of the main difficulties in identifying whether or not the defect comes from the cellar resides in the fact that it is almost impossible to discern the odor when one is immersed or surrounded by a contaminated atmosphere. In fact, the levels of chlorophenols and chloroanisoles are sometimes so high that olfactory senses are rapidly saturated. This phenomenon is particularly intense with 2,4,6-trichloranisole and 2,3,4,6- tetrachloroanisole. Consequently, the moldy odor is really only perceptible when wine is tasted outside of the contaminated location.

Different samples taken from wood materials located in a variety of wine cellars reveals that much of the wood has been treated with chlorophenol-based products. The most active ingredient used is generally pentachlorophenol (or one of its salts), containing impure 2,3,4,6-tetrachlorophenol and sometimes some 2,4,6 -trichlorophenol, often associated with derivatives of hexachlorocyclohexane (Lindane, Diéldrine, υ –HCH, b –HCH). The wood cases and palettes widely used for storing bottled wine, or the wood slats used for ceiling finishes, are just some examples of where these contaminants are largely used. Cardboard boxes that have been manufactured from chlorine-whitened paper pulp or from recycled wood that was treated somewhere along the line with chlorophenols may also contain these components and generate chloroanisoles, but the quantities may be too minimal to be the source of a cellar contamination. On the other hand, cardboard can easily incorporate atmospheric smells and could just as easily strongly emit a disagreeable moldy odor if it had been stored in a contaminated location. Natural stone cellars or caves could also be contaminated in the same way since stone can effectively incorporate odoriferous substances. ***We have also discovered that stone can be contaminated by paint or fixing agents that contain chlorophenols and are used for white washing***. These can rapidly degrade within the humid environment of a cellar.

b) Contamination of storage containers through the air

Cellars for red wine barrels with ceilings ***insulated with fiberglass and covered with wooden slats or particle boards*** can be a source of a polluted atmospheric environment for wines that have been more or less singled out with a moldy character.

High levels of chloroanisoles in barrel-aged wines have been noted in samples taken from those batches considered moldy. High levels of chlorophenols and chloroanisoles have been noted in samples taken from wood paneling and particle boards. Since cellar environments need to be humid, this also facilitates the development of mold that slowly transforms chlorophenols from the bad-smelling chloroanisoles in the wood. These components diffuse from the ceiling to the surrounding atmosphere, from the atmosphere to the wood of the barrels, and then from the wood to the wine. Samples of the wood from used barrels show the presence of contaminants in the entire piece of wood with gradients ranging from the exterior to the interior of the barrel.

***Samples of cement tanks and masonry cellar walls show evidence of occasional weak levels of contamination***. Depending upon the level of pollution of the items contained in the cellar, the tank walls can eventually contaminate the wine that it would contain.

c) Contamination of wine by dry materials stored in contaminated containers.

Products that are used for the wine production process that have been stored in a contaminated atmosphere could have a direct influence on the pollution of the wine. Bentonite, which is used to stabilize the protein structure of white wine, has a microscopic layered structured that enables organic molecules to be caught between the layers. These same molecules can be released into the wine during certain stages of the process. Filtration compounds based on diatoms are able to set variable chlorophenols and chloroanisoles but always on a weaker level than those of betonites. In cases where wine is conserved for long periods of time, it would seem that these elements would also contaminate wine during the filtration process. Filtration plaques composed of cellulose and kieseghur fibers are also sensitive to setting bad odors. Rinsing abundantly before use should normally eliminate the major part of the contaminants.

The cellular structure of corks and the abundance of lipids in their composition renders them particularly sensitive to the chlorophenol and chloroanisole contaminants that are polluting the atmosphere where they are stored. In the case of long-term storage in unsealed packaging, these corks could eventually pass on enough chloroanisoles to the wine that gives it a corky flaw.

**Cork Flavor**

The cork flavor has existed since cork material has been used for bottling. However, in the 1930s to the '50s, the rate of deterioration in wines from Champagne varied between .14 and .45 per 1000. Today, this rate has risen worldwide, sometimes arriving at several percent or even several tens of percent in the worst cases.

By alluding to the cork flavor (a material that is never really inert but only has a negative effect when it becomes too intense with a more-or-less aromatic corky character), it means that the cork can release various bad flavors and aromas. If one does not attach too much of a philosophical or rhetorical distinction between the meanings of "real" and "false" cork tastes, we quickly realize that the major portion of the bad taste (90 to 95 percent of the surveyed corky flavors) are actually moldy tastes. In fact, the deterioration of wine due to corks is due to the same family of elements as in the preceding cases: chloroanisoles.

***A mixed contamination (from both the cellar and from the cork) can always be a possibility***. First we proceeded with a sensory analysis from samples taken from a wide selection of bottles. However, given the size of the sampling lot, it is not always possible to respect the rules for sample testing (NF X 06-22) due to the large number of samples to examine (several hundred or even hundreds of thousands).

Tasting is a powerful investigative tool when employed by a competent panel of professionals, especially trained for sensory analysis. The expert should always do a blind tasting, without any prior knowledge about the possible presence or nature of a defect. Furthermore, he should never taste alone at the site in question, this in order to eliminate any preconceived ideas or other factors that could be unduly influential.

Evidence of a high level of deterioration (>30%) is often indicative of contamination outside of the cork, but only a precise analysis of the cork and wine can allow for certain identification of the origin of the pollution. In fact, the difference between the moldy cellar flavor—mainly caused by 2,3,4,6-tetrachloroanisole and pentachloroanisole—and "typical cork flavor" is most often linked to the presence of a minor 2,4,6-trichloroanisole molecule, and most often absent in most cases. However, we also find cork flavors attributed to 2,3,4,6-tetrachloranisole. We can only make a final objective conclusion once a final tally has been made of all the contaminants present in the wine and the cork.

**Conclusions**

Wine tasting experts must be extremely competent in all aspects of sensory analysis in order to evaluate any organoleptic flaws such as moldy flavors in wine. For a long time, tasting has been the only viable tool. Today it has become imperative to confirm the presence of any flaw detected through sensory methods by scientific methods in order to have irrefutable objective results.

An isolated scientific approach is often subject to failure. One must avoid coming to a conclusion based only on scientific results. Only by comparing these results with the sensory threshold perception can one conclude where the real responsibility lies in regards to the flaw in question.