

CORRESPONDENCE/REBUTTAL

Comments on Truffle Aroma Analysis by Headspace Solid Phase Microextraction [Is Butylated Hydroxytoluene (BHT) a “Natural” Volatile Constituent of Truffles?]

Sir: In recent years, the solid-phase microextraction (SPME) technique has undoubtedly gained widespread success in the analytical field, due to its mild and solvent-free conditions. In particular, it now represents the method of choice for the analysis of volatile organic compounds (VOCs) and has found countless applications in the characterization of flavors and aromas in foodstuffs (1), as readers of *Journal of Agricultural and Food Chemistry* will have noticed. When coupled to GC-MS, it allows a rapid and reliable analysis of the flavor profile, either qualitatively or quantitatively. In fact, although some questionable identification may occur, usually the majority of volatile compounds can be identified unequivocally on the basis of their MS fragmentation and subsequent comparison with database libraries. Undesired contaminations, on the contrary, always occur, especially in the case of headspace volatiles, but the most common contaminants can be easily recognized in a GC-MS chromatogram by the expert eye and checked off from the output list accordingly. Regrettably, however, this is not always the case, and we have stumbled upon a couple of these examples, one of which appeared recently in this Journal.

In a paper dealing with the analysis of truffle aroma by means of headspace SPME (2), a Spanish group has nicely described the characterization of the odor profile of *Tuber aestivum*, and most of their results are well in agreement with the literature data, including our own (3). An astonishing statement at the end of the paper, however, caught immediately our attention. The authors, in fact, reported that “The compound **2,6-bis(1,1-dimethylethyl)-4-methylphenol** (boldface ours) has been cited as a volatile component of the mycelium of *T. borchii* Vitt., but this is the first time that it has been detected in a different *Tuber* species”, thereby quoting (and thus confirming) the results of another paper where 1,3-di-*tert*-butylbenzene was claimed, surprisingly, to represent the main volatile compound in mycelial cultures of *T. borchii* Vitt., along with 2,4-bis(1,1-dimethylethyl)phenol and a number of other “unusual” compounds (4).

These researchers should have recalled that 2,6-bis(1,1-dimethylethyl)-4-methylphenol, which they claim to contribute to the aroma of their truffles, is just but the IUPAC name for butylated hydroxytoluene (BHT), a widespread synthetic antioxidant that is added to plastics, elastomers, solvents, and food items as well. It can be easily released from plastic vials or coatings and contaminate analytical samples, headspace included, giving rise to an extra peak. As many chemists working with GC-MS have experienced, BHT (this is how it is usually referred to also in MS fragmentation databases) comes out in almost every GC run, as a recognizable impurity with a diagnostic base peak at *m/z* 205 due to the loss of a methyl group. Definitely, BHT is not a fungal metabolite, and in our opinion it would be somewhat misleading to include such a compound among the volatile components of truffle flavor, as if it was produced by the fungus itself. In fact, because the paper deals with the analysis of truffle aroma, one would simply expect

to find a list of the *natural* compounds responsible for the flavor. BHT is obviously just a contaminant, and it could have been easily taken out of the list, or at least it should have been labeled as such in the text. The same applies for 2,4-bis(1,1-dimethylethyl)phenol and 1,3-di-*tert*-butylbenzene, which are known to occur as volatile contaminants arising from tris(2,4-di-*tert*-butylphenyl) phosphite, a common plastic stabilizer (5, 6).

Undoubtedly, GC-MS represents a powerful analytical technique that allows a quick and easy determination of volatile compounds, even in trace quantities, but puts out a great deal of structural data that should be examined critically in order to avoid mistakes such as the above-reported ones. When GC-MS output lists are processed, *the identity of any compound must be always consistent with the nature of the matrix under examination*, regardless of the matching value with the MS database. If structures that look somewhat unrelated to the sample are suggested by the database library on the basis of the MS fragmentation, some doubt should arise and the possibility of sample contamination should be considered seriously, unless a reasonable pathway to account for their formation is proposed. In this respect, authors as well as referees are strongly recommended to examine carefully and pedantically GC-MS data, especially if dealing with the characterization of natural flavors, and check for the consistency of any compound that is claimed.

LITERATURE CITED

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