Identification of some \( o \)-aminophenones as secondary metabolites of *Saccharomyces cerevisiae*

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**Summary:** During fermentation of a peculiar model medium a strain of *Saccharomyces cerevisiae* var. *cerevisiae* (SIC) yeast from our collection was able to produce \( o \)-aminoacetophenone as well as other metabolites tentatively identified as \( o \)-aminopropiophenone and 3-(\( o \)-aminophenyl)-prop-1-en-3-one.

**Key words:** \( o \)-aminophenones, *Saccharomyces cerevisiae*, fermentation by-products.

**Introduction:** The identification of some \( o \)-aminophenones as volatile by-products of *Saccharomyces cerevisiae* var. *cerevisiae* is reported in this note.

\( o \)-aminoacetophenone which usually occurs in musts and wines of *V. vinifera* x *V. labrusca* hybrids (ACREE *et al.* 1989) has been recently identified in *V. vinifera* wines (RAPP *et al.* 1993) and considered as a contributor to the "hybrid-naphthalene" off-flavour.

**Materials and methods:**

**Composition of raw model nutrient medium:** (NH\(_4\))\(_2\)SO\(_4\) 0.9 g/l; (NH\(_4\))\(_2\)HPO\(_4\) 0.9 g/l; saccharose 200 g/l; tartaric acid 4 g/l; NaOH for reaching pH=3; CaCl\(_2\) 0.1 g/l; NaCl 0.1 g/l; KH\(_2\)PO\(_4\) 1 g/l; MgSO\(_4\) \( \cdot \) 7 H\(_2\)O 0.5 g/l; NaMoO\(_4\) \( \cdot \) 7 H\(_2\)O 200 \( \mu \)g/l; ZnSO\(_4\) \( \cdot \) 7 H\(_2\)O 400 \( \mu \)g/l; H\(_2\)BO\(_3\) 500 \( \mu \)g/l; KI 100 \( \mu \)g/l; FeCl\(_3\) \( \cdot \) 6 H\(_2\)O 400 \( \mu \)g/l; MnSO\(_4\) \( \cdot \) H\(_2\)O 400 \( \mu \)g/l.

The two media were prepared by adding to the above raw medium: (1) vitamin B6 (100 \( \mu \)g/l) and tryptophan (100 mg/l), (2) vitamin B6 (100 \( \mu \)g/l). All the media were sterilized by filtration with 0.2 \( \mu \)m membrane prior to the yeast cells inoculum. 10\(^3\) cells/ml of a *Saccharomyces cerevisiae* var. *cerevisiae* strain (SIC) from our collection were inoculated and the temperature was kept at 20 °C during the fermentation.

**Sample preparation for GC-MS analysis:** 200 ml of each fermenting medium after addition of 1-heptanol as internal standard (67.4 \( \mu \)g) were eluted through a 10 g C18 reversed phase cartridge (Millipore), in order to absorb lipophylic compounds (from DI STEFANO 1991). After washing with 50 ml of water, the adsorbed compounds were eluted with 50 ml of CH\(_2\)Cl\(_2\), the extract was dried on CaSO\(_4\), and reduced to a volume of about 0.5 ml, before GC-MS analysis.

A GC 5890 with 5970 mass selective detector Hewlett Packard, equipped with J&W DB-WAX (30 m; 0.25 mm; 1 column) was used for the gas chromatographic separation. The corresponding mass spectra of the different \( o \)-aminophenones are shown in the figure below.

**Figure:** Mass spectra of \( o \)-aminophenones identified in medium (1) fermented by a *Saccharomyces cerevisiae* var. *cerevisiae* strain: a) \( o \)-aminoacetophenone, b) \( o \)-aminopropiophenone, c) 3-(\( o \)-aminophenyl)-prop-1-en-3-one.

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0.32 mm i.d.; d.f. = 0.32 μm) WCOT capillary column was used. Mass spectra were recorded at 70 eV with a total ion acquisition programme. The response factor for semiquantitative analysis is 1.

Results: The mass spectra of the o-aminoacetophenone as well as the two other compounds of the same chemical class identified in the medium (1) after fermentation are reported in the Figure. They were tentatively assigned to o-aminopropiophenone and 3-(o-aminophenyl)-prop-1-en-3-one. At higher retention time other unidentified metabolites of the same class of these compounds were detected.

The amounts of the o-aminophenones in the medium (1) are the following (μg/l): o-aminoacetophenone = 58; o-aminopropiophenone = 107; 3-(o-aminophenyl)-prop-1-en-3-one = 141.

Under the reported analytical conditions, only in the medium (1) it was possible to identify the above o-aminophenones.

