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Effect of commercial starter cultures and native yeasts on Ochratoxin A production in meat products

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Introduction: Ochratoxin A (OTA) is a secondary metabolite produced by *Penicillium* and *Aspergillus* genera and is considered one of the most important mycotoxins occurring in animal and human food chains. It is teratogenic, immunotoxic, neurotoxic and mostly nephrotoxic. In dry-cured and fermented meat products, OTA is mostly associated with *Penicillium nordicum*, but *Aspergillus westerdijkiae* has also been found to be responsible for high OTA levels in cured meat products.

Methods:

1. Inocula:

Yeasts: Candida zeylanoides (Cz) and Rhodotorula mucilaginosa (Rh) in PDB

- Starter culture (St) in MRS broth
- OTA-producing fungi: Penicillium nordicum (Pn) and Aspergillus westerdijkiae (Aw)

2. Meat-based media and inoculation:

- Ham (3% ham extract, 3% NaCl, 2% glycerol, 2% agar), traditional dry-sausage (Trad) and industrial dry-sausage (Chour) (3% sausage extract, 3% NaCl, 10% glycerol, 2% agar)
- media were inoculated by incorporation with 10⁵ cells/mL of Cz, Rh, Mix (Cz+Rh) and St. Fungal spores (2x10⁴) were co-inoculated by three-point inoculation. Petri dishes were incubated at 20 °C for 15 days.



Figure 1: A: cooking the meat for the medium (extract). B: Spore inoculation on medium with incorporated cells

3. OTA extraction and quantification

> OTA was extracted from agar plugs with methanol and quantified by HPLC-FLD (λ ex 330 nm and λ em 463 nm) with a RP-C18 column (100 x 4.6 mm) in water: acetonitrile: acetic acid (29.5:70:0.5) at 0.8 mL/min.



Figure 2: A: OTA extraction by the three agar plug method. B: Chromatogram of OTA (Top: OTA standard; bottom: OTA produced in ham sample)

Conclusions: This study highlights the need to account for all mycotoxigenic fungi potentially present in food products. Studies are currently being developed to try to understand the mechanism behind these unexpected results.

Fungal growth and OTA production in meat products are strongly influenced by environmental conditions, physico-chemical characteristics of the matrix, and its endogenous flora.

Aims: This work aimed to evaluate the role of meat native yeasts and one commercial starter culture, on the growth of OTA-producing fungi as well as on OTA production, by using meat-based culture media as model systems.



1. Co-inoculation with P. nordicum:



Figure 3: Growth and OTA production by Pn under different conditions after 15 d incubation. A: Pn on ham (control); B: Pn with Cz on ham; C: Pn growth (colony diameter, in cm); D: OTA production (ng/g agar). NOTE: Rh showed limited growth, thus limited effect, in ham medium

- ✓ Yeasts inhibited Pn growth, but not St
- ✓ OTA production by Pn was only detected in ham medium.
- ✓ In ham medium, all co-inoculants were able to inhibit OTA production

2. Co-inoculation with A. westerdijkiae:



Figure 4: Growth and OTA production by Aw under different conditions after 15 d incubation. A: Aw on ham (control); B: Aw with Cz on ham; C: Aw growth (colony diameter, in cm); D: OTA production (ng/g agar). NOTE: Rh showed limited growth, thus limited effect, in ham medium

- ✓ Yeasts inhibited Aw growth, but not St
- ✓ OTA production by Aw (alone) was low in all media.
- ✓ All co-inoculants highly stimulated OTA production in all media

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