

IAFP'S EUROPEAN SYMPOSIUM ON FOOD SAFETY

PROGRAMME

Held at La Cité des Congrès de Nantes

ORGANIZED BY



www.foodprotection.org

P2-34 Exploring Yeast Biodiversity from Dry-Salted Naturally Black Olives with Culture Dependent and Independent Molecular Methods

Zoe Gounari¹, Stamatoula Bonatou¹, GEORGE-JOHN NYCHIAS¹ and Efthalia Paragou¹
¹Agricultural University of Athens, Athens, Greece

Introduction: Dry-salted olives are cultivated mainly on the island of Thasos and they are traditionally prepared by placing the fully mature olives in layers with coarse salt. Despite the low-water activity of the final product due to high NaCl concentration, the olives are susceptible to spoilage microorganisms such as yeasts and fungi. **Purpose:** To explore the yeast biodiversity of dry-salted table olives from Greek retail outlets with culture-dependent and independent molecular methods.

Methods: Nine samples of dry-salted olives were analyzed. One hundred eighty isolates were clustered after PCR amplification of repetitive DNA with the oligo-nucleotide primer (GTG)_n and profiles with identity percentage higher than 90% were considered to belong to the same group. The typical isolates of each group were further clustered after the amplification of ITS1 region using ITS1 and ITS2 as primers and the implementation of RFLP analysis with the restriction endonucleases Hinf I, Hae III and Cfo I. A total of 20 isolates with discriminant differences were subjected to sequencing of the ITS1 region of rDNA gene and the results obtained were aligned with BLAST to determine the closest known relatives. Next generation sequencing was performed after the amplification of 26S rDNA gene using LS27 and NS483 as primers.

Results: *Pichia membranifaciens*, *Candida arborvictrix*, *Campylopus caryosensis*, *Candida etchellsii*, *Molecularomyces subspicuosus*, *Candida apicola*, *Wickerhamomyces anomalus*, *Tortoniopsis delimitata*, and *Wickerhamomyces variabilis* were the dominant species among the samples according to ITS1 sequencing (culture-dependent method), while the dominant species showed by NGS (culture-independent method) were *Candida etchellsii*, *Pichia integrans*, *Pichia membranifaciens* and *Candida reuskei*.

Significance: Dry-salted olives are an economically important product for the Greek table olive industry and the biodiversity of yeasts in the final product is revealed for the first time by means of molecular techniques.

P2-35 Control of Undesirable Microbial Growth in Table Olive Fermentation Using Selected Yeast Strains with Multifunctional Potential

Stamatoula Bonatou¹, GEORGE-JOHN NYCHIAS¹ and Efthalia Paragou¹
¹Agricultural University of Athens, Athens, Greece

Introduction: The implementation of microbial starters related to table olives with multifunctional features during the fermentation process can reduce the risk of spoilage and the growth of undesirable microorganisms.

Purpose: To assess the survival of five autochthonous yeast strains belonging to *Candida boidinii* and *Saccharomyces cerevisiae* inoculated in the brines of Kalamata natural black table olives during fermentation and monitor the changes in the microbiological and physicochemical profile.

Methods: Olives were fermented in seven percent v/v NaCl and inoculated in monoculture with four yeast strains belonging to *C. boidinii* (Y27, Y28, Y30, and Y31) species and one strain of *S. cerevisiae* (Y34) during the first day of the fermentation to a final concentration of 3×10^7 CFU/ml. Changes in microbiological counts and pH were analyzed for a period of 150 days. The survival of the inoculated strains has been estimated by gel electrophoresis from PCR amplification of repetitive DNA elements with the oligonucleotide primer (GTG)_n at different fermentation points.

Results: At the end of the fermentation, yeasts' populations ranged from 3.8 to 5.3 log CFU/g in olives, while they reached a population between 3.8 and 5.4 log CFU/ml in brines. Moreover, pH values ranged from 3.7 to 4.9 among the treatments for both the olives and the brines. The results obtained from rep-PCR for the first day of fermentation showed 100% survival of the starters. At the end of the process three out of five starters namely, Y27, Y30, and Y31 (yeast isolates, one isolate and two isolates, respectively) showed survival on the olive flesh.

Significance: The survival of the strains used as starters during fermentation is very important as table olives are considered to be a good matrix for several microorganisms with probiotic potential.

P2-38 Prevalence and Relevance of *Salmonella* spp. Testing in Camellia Siversis Tea

Dimitra Wachniska¹, Gary Mycock² and ALEJANDRO AMEZQUITA¹
¹R&D Unilever, Sharnbrook, United Kingdom, ²Unilever, Sharnbrook, United Kingdom

Introduction: Tea is the second most commonly consumed beverage in the world. Its forecasted growing consumption is driven by consumer demand for new flavours and recognition of the health benefits, which results in the emergence of innovative herbal tea and tea blends with the addition of herbs and spices. Currently, no legislation specifies microbiological criteria for tea in Europe. However, available guidelines require the absence of pathogenic *Salmonella* spp. in finished products.

Purpose: This study evaluates the prevalence of *Salmonella* spp. in leaf and herbal teas to mitigate the risk associated with this pathogen. Findings are used to assess the relevance of finished product *Salmonella* testing for these commodities.

Methods: Information relating to the prevalence of *Salmonella* spp. in Camellia siversis was collected following searches of electronic databases, scientific journals, books, technical reports and company internal data. The search strategy included keywords "microbiology", "tea", "prevalence", "quality", "*Salmonella*".

Results: An exhaustive search of the literature (over 2,100 publications screened) and internal data yielded 17 publications. The overall analysis indicated low prevalence (46 positive samples of total 2,338 identified) of *Salmonella* at the level of 2%, with no study reporting pathogens in black and green tea. All positive samples were in herbal teas (e.g. jasmine, lemongrass, rooibos, peppermint or lemon verbena tea).

Significance: Findings of this review align with a history of safe use of tea over thousands of years and equally confirm that control of *Salmonella* spp. within factory facilities manufacturing black and green tea should rely on preventative approaches, e.g. ingredient quality programmes, Good Manufacturing Practices, Hazard Analysis Critical Control Point and environmental monitoring programmes, rather than finished product testing. Considering that the pathogen has been sporadically isolated from herbal teas, robust sampling plans aligned to finished product testing based on statistical methods are suitable to control the risk of *Salmonella* spp. associated with tea blends containing herbs, dried flowers, fruit pieces, and spices.

P2-37 Exploring the Virulence Gene Expression of *Acrobacter butzleri* during Simulated Infection of Human Gut Models

DAVIDE BUZZANCA, Valentina Alessandra, Cristian Bota and Kalliopi Pantelou
 University of Torino-DISAFA, Grugliasco, Italy

Introduction: *Acrobacter butzleri* is an emerging foodborne pathogen often isolated from pork, chicken and beef meat, which causes different gastrointestinal diseases in human due to its invasive behaviour. However, the pathogenicity of *A. butzleri* is still underestimated due to a substantial lack of information on its virulence mechanisms, metabolic and genomic features.

Purpose: This study aims to explore, in simulated host-pathogen interactions, the expression of nine genes that are currently correlated with virulence traits of *A. butzleri*.

Methods: After the *de novo* design of primers for the nine genes (*cadF*, *cadB*, *qT26*, *lpg4*, *hcaA*, *hcaB*, *mvh1*, *pda*, *tyrA*) on an *A. butzleri* reference genome (type strain LMG 12623^T), their relative expression was quantified by quantitative RT-PCR under simulated host-pathogen interaction conditions. Briefly, in *in vitro* gut models of mucus-producing (HT29-hTDC) and non-producing (HT29, Caco-2) human cells were co-incubated with the pathogen and total bacterial RNA was recovered at different time points. At the same time, bacterial counts were performed to describe the colonization and translocation capabilities of *A. butzleri* by using two-dimensional and three-dimensional gut models, respectively.

Results: As the first outcome, an RT-qPCR protocol, suitable to quantify the relative expression of the nine virulence genes of *A. butzleri* in the presence of human cells, was optimized. Applying this protocol, an upregulation of part of those genes along the co-incubation time was observed. In addition, a favourable role of the mucus for the pathogen colonization was observed whereas the data from the three-dimensional models suggested an intracellular passage of *A. butzleri* through the epithelial barrier.

Significance: The results of this study represent a first step in the understanding of *A. butzleri* pathogenicity and are important to explain its role in gastrointestinal diseases.

P2-38 Survival of *Listeria monocytogenes* In Food Residue on Packaging Materials for Dairy Products

Filipi Di Cicco, FRANCESCO CHIESA, Maria Auxilia Grassi, Daniela Rubiola and Tiziana Chivers
 Dipartimento di Scienze Veterinarie, University of Turin, Torino, Italy

Introduction: *Listeria monocytogenes* is known for causing foodborne infections often associated with a high mortality rate. Survival under adverse conditions of *L. monocytogenes* for extended periods of time has been reported. *L. monocytogenes* contamination

P2-37 Exploring the Virulence Gene Expression of *Arcobacter butzleri* during Simulated Infection of Human Gut Models

DAVIDE BUZZANCA, Valentina Alessandria, Cristian Botta and Kalliopi Rantsiou

University of Torino-DISAFA, Grugliasco, Italy

Introduction: *Arcobacter butzleri* is an emerging foodborne pathogen often isolated from pork, chicken and beef meat, which causes different gastrointestinal diseases in human due to its invasive behaviour. However, the pathogenicity of *A. butzleri* is still underestimated due to a substantial lack of information on its virulence mechanisms, metabolic and genomic features.

Purpose: This study aims to explore, in simulated host-pathogen interactions, the expression of nine genes that are currently correlated with virulence traits of *A. butzleri*.

Methods: After the *ex novo* design of primers for the nine genes (*cadF*, *ciaB*, *cj1349*, *irgA*, *hecA*, *hecB*, *mviN*, *pldA*, *tlyA*) on an *A. butzleri* reference genome (type strain LMG 10828^T), their relative expression was quantified by quantitative RT-PCR under simulated host-pathogen interaction conditions. Briefly, In Vitro gut models of mucus-producing (HT29–MTX) and non-producing (HT29, Caco-2) human cells were co-incubated with the pathogen and total bacterial RNA was recovered at different time points. At the same time, bacterial counts were performed to describe the colonization and translocation capabilities of *A. butzleri*, by using two dimensional and three dimensional gut models, respectively.

Results: As the first outcome, an RT-qPCR protocol, suitable to quantify the relative expression of the nine virulence genes of *A. butzleri* in the presence of human cells, was optimized. Applying this protocol, an upregulation of part of those genes along the co-incubation time was observed. In addition, a favourable role of the mucus for the pathogen colonization was observed whereas the data from the three-dimensional models suggested an intracellular passage of *A. butzleri* through the epithelial barrier.

Significance: The results of this study represent a first step in the understanding of *A. butzleri* pathogenicity and are important to explain its role in gastrointestinal diseases.

AUTHORS AND PRESENTERS

Authors and Presenters

Presenter

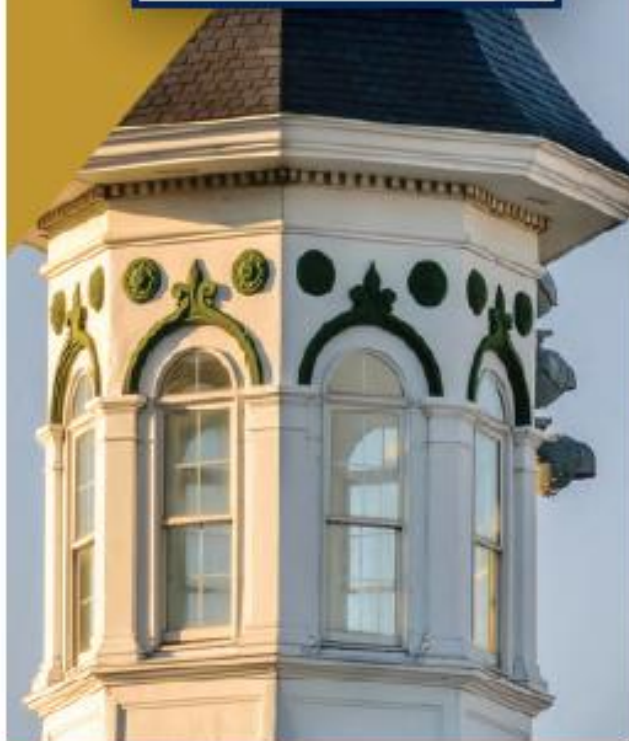
- Abram, Florence, *National University of Ireland Galway* (P2-02)
Adreu de Asaunqas, Ricardo, *INSA* (P1-09)
Ackermann, Elliana, *Centre for Food Safety Stellenbosch University* (P2-03)
Acuff, Gary, *Texas A&M University* (S21*)
Alexandridis, Georgios, *Süßler AG* (T9-04)
Alessandria, Valentina, *University of Torino-OrSAFA* (P2-07)
Allard, Sarah, *Maryland Institute for Applied Environmental Health, University of Maryland* (P2-04)
Allende, Ana, *CSBAG-CSIQ* (S8*)
Almuyah, Basim, *University of Kufa* (P2-04*)
Alitto, Paola, *INSA* (T4-04, P1-09)
Alwan, Nisreen, *Modern University for Business & Science* (P1-08, P1-01)
Amecopita, Alejandro, *Unilever* (P2-09)
Amin, Aya, *The Ohio State University* (P1-01*)
Anastasio, Aniello, *University of Naples "Federico II"* (P2-08)
Anderson, Martin, *IN-Water Agri* (S17*)
Anderson, Nathan, *U.S. Food and Drug Administration* (P2-03, T8-06)
Anderson, Wayne, *Food Safety Authority of Ireland* (S23*)
Anderson-Coughlin, Brianna, *University of Delaware* (P2-04)
Andrighetto, Christian, *Univero-Agriculture, Istituto per la Qualità e le Tecnologie Agromolecolari* (P2-10)
Antoni, Laura, *Università di Torino* (P1-10)
Atriusa, Eduard, *University of Barcelona* (S9)
Armas, Andrea, *Universidad Nacional de Aconcagua DGCYT CSMT* (P2-05*)
Armas, Pablo, *CID* (P2-05)
Assaf, Ali, *University of Nantes* (P2-14*)
Assunção, Ricardo, *INSA* (T4-04)
Atchley, Julie, *SaleTrace* (P2-04)
Athanasaki, Konstantina, *Agricultural University of Athens* (P2-02)
Auvray, Frédéric, *Université Paris-Est, ANSES* (P1-09)
Avery, Simon, *University of Nottingham* (S18*)
Azeiteiro, Sarah, *International Iberian Nanotechnology Laboratory* (T2-02, P1-06)
Azar, Arantxa, *Universidad Politécnica de Cartagena* (T1-02)
Baeumlidberger, Mathias, *Herc Foods, Sigma-Aldrich/Production GmbH* (P1-01*)
Baguet, Justine, *ADRIA Food Technology Institute* (T2-06, P1-11)
Bakir, Gyung-Jin, *Kuans National University* (P1-08, P1-01)
Bala, Harish, *University of Delaware* (P2-11)
Balogai, Anastasia, *AUA* (T9-04)
Balestrieri, Marco, *Institute of Biotechnologies and BioResources – UO3 Naples, National Research Council* (P2-08)
Baloyi, Thintswalo, *DST/NRF Centre of Excellence in Food Security, University of Pretoria* (P1-02)
Baranyi, József, *University of Debrecen* (S10)
Barnes, Jerry, *Newcastle University* (T8-01)
Barreto, Caroline, *NeST Research* (T2-01)
Barrin, Benjamin, *Q Laboratories, Inc.* (P1-03, P1-02, P2-41)
Barlow, Zoe, *Cardiff Metropolitan University* (P1-03)
Barrington, Matt, *Viasable Solution AB* (P1-02)
Benoit, Fabienne, *LABFO* (P1-02)
Berger, Thomas, *Agricoop* (T8-08)
Berghof-Jaeger, Kornelia, *Biotecor Diagnostics* (P1-02)
Bergis, Hélène, *ANSES* (S18*, P1-17)
Bernard, Marie, *ADRIA Food Technology Institute* (P1-06, P1-04, P1-03, T7-01)
Bemas, Cécile, *ADRIA Food Technology Institute* (T2-06, P1-11)
Bernstein, Chris, *U.S. Department of Agriculture – FSIS* (P1-02)
Bertasi, Barbara, *CSLGR* (P2-02)
Betta, Gill, *Camden SR* (P1-07, P2-08, P1-17)
Betta, Roy, *Camden SR* (P11*)
Binet, Rachel, *U.S. Food and Drug Administration* (P1-17)
Biolcati, Federica, *Università di Torino* (P2-10*)
Bird, Patrick, *Q Laboratories, Inc.* (P1-07, P1-02, P2-41)
Blaker, Stine Loewenrup, *Wlan AG* (P2-00*)
Blawie, Przemyslaw, *Allegor Corporation* (T7-01)
Black, Glenn, *U.S. Food and Drug Administration* (T8-08)
Blasi, Michele, *Associazione Italiana Allevatori* (S24*)
Bleichner, Laura, *Bioflex Genelecian Technologies GmbH* (P1-10)
Bonatou, Stamatoula, *Agricultural University of Athens* (P2-04, P2-05)
Bonifati, Loretta, *ANSES* (P2-09)
Bortolozzi Ricardo de Carvalho, Ana Carolina, *University of Campinas* (P2-02)
Boch, Albert, *University of Barcelona* (S9)
Botta, Cristian, *University of Torino-OrSAFA* (P2-07, T1-01*)
Bottero, Mariela, *University of Bari* (S9)
Bottero, Maria Teresa, *Università di Torino* (P2-10)
Boué, Géraldine, *Seccalm, INRA/Oxite* (T4-04, T8-02, P1-09)
Bouju-Albert, Agnès, *SECALM, INRA, Oxite, Université Bretagne Loire* (T1-02, T1-08)
Bouju-Albert, Agnès, *UMR 1014 Seccalm, IRL, INRA, Oxite* (P1-02*)
Bours, Marcia, *University of Reading* (T7-08)
Bourdichon, François, *International Dairy Federation – Standing Committee on Microbiological Hygiene (RT1*)*
Bower-Cid, Sara, *IRTA, Food Safety Programme* (P2-03, P1-17)
Bovo Campagnolo, Fernanda, *University of Campinas* (P2-01*, P2-02*, P1-08*)
Bozman, John P., *University of Tennessee* (P1-41)
Bozkurt, Haytiye, *The University of Sydney* (T8-02*)
Bradshaw, Rhodri, *U.S. Department of Agriculture – ARS, Environmental Microbial and Food Safety Laboratory* (P2-04)
Brahma, Pia, *Central Institute of Technology Kolarhiser* (P1-42)
Brage, Thomas, *ANSES, Laboratory for food safety* (T3-01)
Brazil, Roberto, *INSA* (T4-04)
Bridier, Arnaud, *Anesi* (P2-08, P2-07)
Briolat, Jean-Pierre, *Méts College of Arts, Science & Technology* (P1-08)
Brodkorb, Andre, *Teagasc Food Research Centre* (P2-17)
Brulenberg, Paul, *Teagasc Nutrition* (P2-02)
Bruschi, Carolina, *University of Reading* (T7-05*)
Bugari, Maria, *Teagasc University* (T7-02*)
Bramson, Catherine, *Cardiff Metropolitan University* (P1-02)
Burgess, Catherine M., *Teagasc Food Research Centre, Ashford, Teagasc* (P2-04, P1-43, P2-02*, P1-07*, P1-01*)
Burgess, Kaye, *Teagasc* (T8-01)
Burfesio, Johnathan, *SaleTrace* (P2-04, T2-02)
Buziniani, Melissa, *FoodCheck Laboratories Inc.* (P1-06)
Buzzaon, Davide, *University of Torino-OrSAFA* (P2-07*)
Cadavez, Vasco A. R., *Polytechnic Institute of Setúbal* (T8-06, P2-06, P2-01)
Calderson, Della, *Hygiene* (P1-12, P1-13)
Callahan, Mary Theresa, *University of Maryland* (P2-04)
Callahan, Michael, *Cork Institute of Technology* (P2-17*)
Calvez, Sébastien, *INOGENIX, INRA, Oxite* (T1-02)
Campagnoli, Matteo, *NeST Research Center* (P2-08, P1-03, P2-08, P1-02*)
Campos, Gabriela Zanpieri, *University of São Paulo* (P2-18*)
Cantargiani, Frederique, *NeST Research Center* (P1-17)
Cardoso, Maria João, *Universidade Católica Portuguesa* (P2-08)
Carmona, Paulo, *ASAIE* (T4-04)
Carvalho, Antonio Fernandes, *Universidade Federal de Minas* (P2-18)
Carvalho, Joana, *International Iberian Nanotechnology Laboratory* (T2-02, P1-06)
Carvalho, Marta, *Universidade Católica Portuguesa* (S9)
Castelljo, Greetje, *Netherlands Food and Consumer Product Safety Authority (VWA)* (T2-04*)
Cauvin, Bode, *LABFO* (P1-02*)
Cavallero, Maria Chiara, *M.I.A.C. S.r.l. - Polo AGRIFOOD* (T1-01)
Cazal, Cinthia, *Universidad Nacional de Aconcagua DGCYT CSMT* (P2-05)
Cerillo, Lucia, *SaleTrace* (P2-04, T2-02)
Chandry, P. Scott, *CSIRO Agriculture & Food* (P1-41)
Chantapikul, Bowwornan, *University of Guelph, ORFS* (P2-04*)
Chapman, Benjamin, *North Carolina State University* (P1-02)
Charrier, Thomas, *Bioflex* (P1-08, S11*)
Chatzizola, Christina, *University of Meis* (T9-02)



Louisville
KENTUCKY



IAFP 2019
ANNUAL MEETING - JULY 21-24



It's a Sure Bet!

Join more than 3,600 food safety professionals at the world's leading food safety conference, and take part in hundreds of informative symposia, roundtables, and technical presentations throughout four days. IAFP's Professional Development Group on-site meetings provide additional opportunities to share, learn and network with your peers about today's food safety challenges.

REACH FOR THE FINISH LINE WITH IAFP!

Our commitment to *Advancing Food Safety Worldwide®* is second to none.

Go the distance by attending IAFP 2019!