

RICERCHE EFFETTUATE

IGIENE DEGLI ALIMENTI AD USO UMANO

Acciari VA, Torresi M, Iannetti L, Scattolini S, Pomilio F, Decastelli L, Colmegna° S, Muliari R, Bossù T, Proroga Y, Montagna C, Cardamone C, Cogoni P, Prencipe VA, Migliorati G

Listeria monocytogenes in smoked salmon and other smoked fish at retail in Italy : frequency of contamination and strain characterization in products from different manufacturers

J Food Prot. - Vol. 80 no 2 (2017). - p 271-278. - 41 bib ref [Nr. Estr. 7538]

Seven hundred seventy-eight samples of packaged smoked fish (774 smoked salmon and 4 smoked swordfish) on sale in Italy, from 50 different manufacturers located in 12 European Union countries, were purchased from the Italian market between May and December 2011. The surface temperatures of the samples on sale ranged from 0 to 13°C ($3.4 \pm 1.5^\circ\text{C}$, mean \pm SD). Six hundred eighty (87.4%) of 778 samples were stored at $=4^\circ\text{C}$. One hundred fifty-seven samples (20.2%, 95% confidence interval 17.5 to 23.1%) were contaminated by *Listeria monocytogenes*, with 26 samples (3.3%, 95% confidence interval 2.3 to 4.8%) at levels >100 CFU/g. The maximum level of contamination was 1.3×10^6 CFU/g. The differences in the level of contamination of smoked fish between countries ($\chi^2 = 91.54$, $P < 0.05$) and manufacturers ($\chi^2 = 193.22$, $P < 0.05$) were significant. The frequency of detection for products from different manufacturing premises ranged from 0 to 76.9%. Serotyping by serological agglutination revealed that the main serotypes detected were 1/2a (65.3%) and 1/2b (22.4%). Pulsed-field gel electrophoresis typing with restriction enzymes *Ascl* and *Apal* yielded 36 pulsotypes from 144 isolates, clustering into 17 groups. Eight main pulsotypes accounted for 70.8% of the isolates. Three of the main pulsotypes were exclusively from products of a single manufacturer. In general, products from the same manufacturer showed genetic homogeneity, with one strongly prevalent pulsotype. Different manufacturers usually showed very different levels of contamination of the final product, confirming the importance of the management of process hygiene for controlling *L. monocytogenes* contamination.

Amato E, Filipello V, Gori M, Lomonaco S, Losio° MN, Parisi A, Huedo P, Knabel SJ, Pontello M

Identification of a major Listeria monocytogenes outbreak clone linked to soft cheese in Northern Italy – 2009-2011

BMC Infect Dis. - Vol. 17 (2017). - no 342 (7 p) . - 28 bib ref (ultimo accesso 27/07/2018 <https://bmcinfectdis.biomedcentral.com/track/pdf/10.1186/s12879-017-2441-6>) [Nr. Estr. 7909]

Background: Molecular subtyping and enhanced surveillance in Lombardy region identified a cluster of possibly related listeriosis cases from 2006 to 2010. This cluster grouped 31 isolates that belonged to serotype 1/2a and Sequence Type 38 (ST38) as defined by Multilocus Sequence Typing (MLST). Methods: Our study expanded the previous investigation to include cases from 2011 to 2014 and used MultiVirulence-Locus Sequence Typing (MVLST) on all ST38 isolates to better understand their epidemiology and possibly identify a common source outbreak. Results: Out of 306 *L. monocytogenes* clinical isolates collected, 43 (14.1%) belonged to ST38 with cases occurring in nine out of twelve Lombardy provinces. The ST38 isolates were split by MVLST into two Virulence Types (VTs): VT80 ($n = 12$) and VT104 ($n = 31$). VT104 cases were concentrated between 2009 and 2011 in two provinces, Bergamo and Milan. An epidemiologic investigation was performed and in one case, a matching VT104 isolate was retrieved from a soft cheese sample from a patient's refrigerator. Conclusions: Our findings revealed a major listeriosis outbreak in Northern Italy linked to soft cheese in 2009– 2011, which went undetected by local health authorities. Our study shows that integrating subtyping methods with conventional epidemiology can help identify the source of *L. monocytogenes* outbreak clones.

Andreoli° G, Merla° C, Dalla_Valle C, Corpus F, Morganti M, D'incau° M, Colmegna° S, Marone P, Fabbi° M, Barco L, Carra° E

Foodborne salmonellosis in Italy : characterization of Salmonella enterica serovar Typhimurium and monophasic variant 4,[5],12:i:- isolated from salami and human patients

J Food Prot. - Vol. 80 no 4 (2017). - p 632-639. - 25 bib ref [Nr. Estr. 7539]

Salmonella enterica serovar Typhimurium (STm) and its monophasic variant 4,[5],12:i:- (VMSTm) have been responsible for an increased number of foodborne infections in humans in Europe in recent years. The aim of this study was to investigate the origin of three foodborne salmonellosis outbreaks that occurred in Pavia Province (Lombardy region, northern Italy) in 2010. Phenotypic and genetic characteristics of the STm and VMSTm isolates from patients and from food that were recovered in the framework of the three outbreaks were evaluated through serotyping, phage typing, antimicrobial susceptibility testing, pulsed-field gel electrophoresis (PFGE), and multiple-locus variable-number tandem repeat analysis (MLVA). Salami from three artisan producers, which had all purchased meat from the same slaughterhouse, was the food source of infection in outbreak I. STm isolates were recovered from salami and patients with symptoms of gastroenteritis. These isolates had the same PFGE type and the same rare MLVA profile (3-18-9-NA-211). The same molecular profiles were found in an STm isolate from a salami, which likely was the source of another family outbreak (II). A VMSTm strain with common phenotypic and molecular profiles was isolated from three hospitalized patients and identified as the cause of another putative outbreak (III). During the following 3 years (2011 through 2013), 360 salami produced in Pavia Province were monitored for the presence of *S. enterica*. In 2011, no STm and VMSTm isolates were recovered from 159 salami tested. During 2012 and 2013, 13.9% of 201 tested salami harbored *S. enterica*, and half of the isolates were VMSTm, mainly in salami from those artisan producers involved in the previous outbreaks. These isolates were genetically variable, especially in terms of MLVA profiles. The data collected suggest that from 2012, VMSTm has replaced STm in the environments of the salami producers monitored in this study, and these data confirm the dominance of this emergent serovar along the pork supply chain.

Anesi A, Panceri ML, Asticcioli S, Baroni D, Rognoni V, Marazza G, Rossetti E, Labbadini S, Archenti A, Luini° M; Bertasi° B, Pontello M, Belloni A

The importance of an early alert from the Microbiology Laboratory and multidisciplinary collaboration during a suspected salmonellosis outbreak

Microbiol Med. - Vol. 32 no 1 (2017). - no 6275 (5 p). - 7 bib ref (ultimo accesso 02/11/2017 <http://pagepressjournals.org/index.php/mm/article/view/6275>) [Nr. Estr. 7672]

Salmonellosis Outbreak Alert Food safety Food-borne diseases.

Background and aims. Salmonellosis is one of the most common and widely distributed food-borne diseases. The increasing complexity and globalization of the food industry are causing an increase of some of these large-scale food-borne illnesses, thus there is a need for improvements in public health signal detection and communication streams between laboratories and regulatory agencies. The aim of this study is to show how the early reporting of salmonellosis cases directly from the Laboratory of Microbiology to the Local Health Service Infectious Diseases Office along with the prompt response of the ASL, and the rapid involvement of the Local Veterinary Prevention Department resulted in an improved individuation and investigation of a suspected food-borne outbreak with anomalous manifestation. Materials and methods. From August to November 2014 the early warning from the Laboratory of Microbiology regarding Salmonella spp. isolates with the identical serogroup and antibiotic resistance phenotype, allowed for prompt identification of a food-borne infection. Results and conclusions. The genotyping analysis suggested that over the period considered there was more than a single monophasic Salmonella typhimurium isolate: one responsible for the sporadic cases that occurred in September and October, and another in

November.

Antoci S, Pomilio F, Daminelli° P, Romanelli° C, Ciorba AB, Santini N, Castoldi F, Pierantoni M, Iannetti L, Migliorati G

Monitoraggio dei requisiti igienico-sanitari negli stabilimenti autorizzati all'esportazione di prodotti di origine animale verso Paesi Terzi

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locale e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia, 13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 35-36 (Poster P026-78) [Nr. Estr. 7643]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

L'esportazione di prodotti di Origine Animale verso Paesi Terzi avviene attualmente secondo specifici accordi bilaterali basati sugli standard del Codex Alimentarius e sulle regole determinate dall'SPS Agreement, particolarmente il principio di Equivalenza. In reazione all'export verso Paesi Terzi diversi dagli Stati Uniti d'America, la Direzione Generale per l'Igiene e la Sicurezza degli Alimenti del Ministero della Salute, ha ritenuto opportuno progettare uno studio finalizzato alla valutazione dell'efficacia dei controlli ufficiali eseguiti dai Servizi Veterinari delle ASL, con l'obiettivo di verificare l'omogeneità e il livello di applicazione della legislazione Europea. Sono stati selezionati 50 stabilimenti che lavorano prodotti a base di carne, latte e ittici, dalla lista che include le aziende autorizzate all'esportazione l'attività di monitoraggio è stata svolta da personale degli Istituti Zooprofilattici Sperimentali dell'Abruzzo e del Molise e della Lombardia e Emilia Romagna nel periodo compreso tra ottobre 2016 e aprile 2017. In ciascun stabilimento selezionato è stato eseguito un audit per la verifica dell'applicazione della normativa dell'Unione Europea [Regolamento (CE) n. 882/2004 relativo ai controlli ufficiali, in particolare dell'applicazione delle procedure SSOP (Sanitation Standard Operating Procedures), e di eventuali requisiti specifici per l'esportazione stabiliti negli accordi bilaterali tra Paesi Terzi e Unione Europea. È stato, quindi, effettuato un sopralluogo nello stabilimento dove sono stati prelevati campioni durante la lavorazione in una sola linea operativa, comprendenti 15 tamponi ambientali (10 da superfici a contatto con l'alimento e 5 non a contatto) per la ricerca di *Listeria monocytogenes*. Contestualmente sono stati prelevati 5 campioni di prodotto, per la ricerca di *L. monocytogenes* e *Salmonella* spp. I campioni sono stati analizzati dagli IZSS competenti per territorio, mediante l'applicazione delle norme ISO. *L. monocytogenes* è stata isolata in 59 superfici (33 a contatto e 26 non a contatto) su 750 esaminate e in 7 campioni di alimento su 250. In conclusione, sono risultati contaminati da *L. monocytogenes* gli ambienti di 25 stabilimenti su 50 (50%), mentre nessuna positività è stata rilevata per *Salmonella* spp. È in corso la valutazione della presenza di eventuali correlazioni tra l'effettiva applicazione delle SSOP e la presenza di contaminazioni.

Bardasi° L, Taddei° R, Fiocchi° I, Pelliconi° MF, Ramini° M, Toschi° E, Meriardi° G

Shiga toxin-producing Escherichia coli in slaughtered pigs and pork products

Ital J Food Safety. - Vol. 6 no 2 (2017). - no 6584 (p 79-82). - 30 bib ref (ultimo accesso 06/06/2018 <https://www.pagepressjournals.org/index.php/ijfs/article/view/6584>) [Nr. Estr. 7711]

During the years 2015-2016, 83 faecal samples were collected at slaughter from pigs reared in farms located in Central- Northern Italy. During the years 2014-2016 a total of 562 pork products [465 not-ready-to-eat (NRTE) and 97 ready-to-eat (RTE) products] were collected from retail outlets, large retailers and processing plants. The samples were analysed according to ISO TS 13136:2012. Out of 83 swine faecal samples, 77 (92.8%) resulted stx-positive by real time polymerase chain reaction (PCR), 5 stx2+ and 1 stx1+ Shiga toxin-producing *Escherichia coli* (STEC) strains were isolated. Among the 465 NRTE samples, 65 (14.0%) resulted stx-positive by real time PCR and 7 stx2+ STEC strains were isolated. The stx2 gene was detected more frequently than the stx1 gene both in faecal samples (90.4 vs 8.4%) and in NRTE pork products (13.3 vs 1.3%). All the RTE

samples included in the analysis resulted stx negative. Among the samples resulted positive for stx and eae genes, serogroup-associated genes were detected at high frequency: O26 resulted the most frequent in faecal samples (81.3%) and O145 in pork products (88.1%). The O157 serogroup resulted positive in 83.3 and 78.1% of pork products and faecal samples, respectively. Despite the frequent detection by real time PCR of genes indicating the possible presence of STEC strains belonging to the six serogroups, the bacteriological step did not confirm the isolation of any such strains.

Bassanetti I, Carcelli M, Buschini A, Montalbano S, Leonardi G, Pelagatti P, Tosi° G, Massi° G, Fiorentini° L, Rogolino D

Investigation of antibacterial activity of new classes of essential oils derivatives

Food Control. - Vol. 73 (2017). - p 606-612. - 36 bib ref [Nr. Estr. 7471]

Essential oils (EOs) have deserved much attention in the past decades for their antimicrobial activity, since many of them have demonstrated efficacy against food-borne pathogenic and spoilage microorganisms. Moreover, they have potential application in animal nutrition as multifunctional feed supplements, avoiding or diminishing the use of antibiotics in livestock. However, low solubility and bioavailability as well as volatility and marked aromatic note are important limitations in food and feed applications. In this study we present the synthesis, characterization and evaluation of the antibacterial activity of new thymol, carvacrol and menthol derivatives. The new compounds have been designed to overcome the limitations of the precursors, such as poor water solubility and volatility, still maintaining a good antimicrobial profile. We evaluated the activity of the synthesized compounds against pathogens causing important foodborne diseases, i.e. *Clostridium perfringens*, *Salmonella typhimurium*, *Salmonella enteritidis* and *Escherichia coli*. The low MICs and MBCs values for some of the studied compounds, combined with water solubility and negligible cytotoxicity towards HT-29 human cells, confirmed the potential use for EOs derivatives in the food industry on *Sarcocystis* explain neurological clinical symptoms of the animals, they can be considered the start for further studies on *Sarcocystis*.

Benevenia° R, Mangeri° L, Tilola° M, Delibato E, Scaramagli S, Bertasi° B

Digital polymerase chain reaction : approccio innovativo nell'identificazione di specie

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locale e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia, 13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 36-37 (Poster P028-90). - 2 bib ref [Nr. Estr. 7646]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

L'emergenza carne equine del 2013 ha contribuito al crescente interesse di Autorità Competenti, OSA e laboratori d'analisi, nell'ambito dell'identificazione di specie. La necessità di rilevare quantità di ingredienti molto basse e inoltre legata alla verifica di conformità rispetto all'etichetta come da indicazioni del Reg. UE 1169/2011. Per l'individuazione di elementi in tracce sono tuttora applicate metodiche di biologia molecolare dotate di ottima sensibilità, basate però su identificazione qualitativa del target ricercato. Molti laboratori hanno avviato studi sull'utilizzo della tecnologia quantitativa digital per l'identificazione di specie poiché molteplici possono essere le applicazioni come l'identificazione della percentuale di un ingrediente sul totale del prodotto (per evitare possibili frodi) e la verifica della composizione del prodotto in relazione a particolari trattamenti (ad esempio torrefazione del caffè). Nel presente lavoro è stata valutata l'applicabilità del sistema digital in due differenti aree di interesse: i) l'identificazione di specie domestiche in prodotti a base di carne; ii) l'identificazione delle varietà di *Coffea arabica* (caffè Arabica) e *Coffea canephora* (caffè Robusta) Nel primo caso è stata ottenuta la reazione con un target specie specifico ed un gene housekeeping

(miostatina) e valutata la linearità di risposta a differenti concentrazioni di target. Inoltre è stata controllata l'uniformità della presenza della miostatina in differenti target e costruito un rapporto percentuale DNA target/DNA totale basato su misurazioni strumentali e calcolo in peso (ng). Le sperimentazioni hanno suggerito l'impossibilità di utilizzare la miostatina, come gene housekeeping per tutte le specie; l'approccio del calcolo delle percentuali in peso sembra essere la strada maggiormente perseguibile. Nel secondo caso sono state identificate le specie in oggetto mediante PCR, rendendo possibile successiva analisi in digital PCR per la valutazione dei livelli di contaminazione (Fioren et al., 2015; Laube et al., 2007).

Bolzoni° G, Bettoni° S, Buffoli° E, Baiguera° C, Marcolini° A, Zanardi° G

Stima dell'effetto pascolo sul profilo in acidi grassi del latte bovino in allevamenti di montagna = Pasture influence on fatty acids profile of mountain farms bovine milk

Large Anim Rev. - Vol. 23 no 5 (2017). - p 163-168. - 17 bib ref [Nr. Estr. 7767]

La composizione in acidi grassi del latte è stata oggetto negli ultimi decenni di numerosi studi, sia per aspetti tecnologici e organolettici dei prodotti derivati, sia per quelli nutrizionali e di salubrità. La quantità, così come la composizione lipidica del latte, sono in parte caratteri ereditari, ma in gran parte sono determinati dall'alimentazione degli animali. Lo scopo del presente lavoro è quello di caratterizzare il contenuto in acidi grassi saturi ed insaturi del latte bovino prodotto in 37 allevamenti tipici per alimentazione e stabulazione di un'area di montagna del nord Italia (Valle Camonica), tramite una tecnica analitica di stima indiretta basata sulla spettroscopia in infrarosso. Attraverso l'analisi di 189 campioni di latte di massa raccolti tra la primavera e l'estate 2015, si è potuto verificare che il periodo di pascolo influisce in misura statisticamente significativa sul rapporto tra acidi grassi insaturi e saturi del latte: 0,42 (DS 0,025) rapporto medio nelle stalle a fondovalle vs 0,57 (DS 0,048) rapporto medio delle stesse mandrie al pascolo. Il presente lavoro fornisce inoltre un quadro complessivo delle caratteristiche del latte di montagna evidenziando alcune peculiarità indotte dal periodo di pascolo. Considerato che la tecnica analitica di screening utilizzata, seppur con un'accuratezza relativamente limitata, garantisce analisi semplici, rapide, a costo molto contenuto ed eseguibili su elevati numeri di campioni anche per periodi prolungati, riteniamo che questo tipo di attività possa concorrere a supportare la produzione di latte e derivati nelle aree di montagna, valorizzandone le componenti di genuinità e salubrità.

Introduction - Milk and dairy products ducts are considered an important source of fatty acids in the diet that varies in quantity according to the methods of bovine rearing and feeding. In the last decades, evaluation of the fatty acid composition in bovine milk has been the subject of several studies for the beneficial effects on human health. The aim of this study is to quantify saturated and unsaturated fatty acids by indirect estimation with Infrared spectroscopy in mountain farms, in particular to confirm the effect of pasture period on the milk composition. Materials and methods - The 189 milk samples were collected from 37 dairy farms representative of the typical "farm" in a mountainous area of North Italy (Valle Camonica): small (12 heads/farm on average), prevalence of Brown Alpine and Red Piebald, low production level (about 16 Lt/head/die), mainly fed with local hay, integration of feed-stuffs under 30% of the feed ration, without maize silage. The samples were collected directly from farms (first period, April-May 2015) or from pasture areas (second period, June-July 2015). Results and discussion - Our observations confirm the change in milk composition related to the grazing period as well with the increased content in fat and unsaturated fatty acids and, in particular, the "grazing effect" was statistically confirmed by a significant increase in the unsaturated/saturated fatty acids ratio: 0.42 (SD 0.025) in farms vs 0.57 (SD 0.048) in pasture areas, as already highlighted by other studies made with reference method. Conclusions - The Fourier transform infrared spectroscopy analytical method, easy to use and cheap, although the indirect evaluation of the milk composition, produces results and information useful to support the quality of milk and milk products in mountain areas, also for larger projects in the future.

Bolzoni° G, Buffoli° E

Il controllo della pastorizzazione : indicazioni pratiche per situazioni "poco definite"

Alimenti & Bevande. - Vol. 19 no 2 (2017). - p 46 - 50 [Nr. Estr. 7568]

Bolzoni° G, Marcolini° A, Fontana A, Monaco L, Ferrini AM

BactoScan FC : conversion system for results at the national level in Italy and reproducibility of total bacterial count testing four years after implementation

Euroreference. - Vol. no 2 (2017). - p 46-54. - 16 bib ref (ultimo accesso 12/01/2018 <http://euroreference.mag.anses.fr/en/issue/2%20EUROREFERENCE>) [Nr. Estr. 7717]

BactoScan and BactoCount are automated instruments for the determination of the total bacterial count (TBC) in milk through an alternative routine method. Results are given in impulses, but the TBC is officially expressed in colony-forming units per ml (CFU/ml), making a conversion system necessary in order to transfer results onto the official scale. In Italy, these instruments were introduced at the beginning of the 1980s, and today amount to more than 50 units. The initial huge number of conversion lines was gradually reduced over the years until 2012, when a single conversion relationship, developed by a joint NRCBMQ — NRLMMP project, was finally made available to Italian laboratories. In fact, it has been adopted by almost all the laboratories that routinely use these instruments. This article examines the results of about 50 proficiency tests (PTs) organised by the Italian Breeders Association (AIA) on a national scale in the period 2003-2016, for which laboratories were asked to provide results in impulses and in CFU, according to their own current conversion system. A retrospective statistical analysis of the results enabled us to assess the changes in the reproducibility of the results expressed in both units of measurement over time: that is, in impulses (mainly dependent on instrumental performance) and in CFU (also dependent on the conversion line used). In particular, we demonstrate the effect of applying the national conversion system developed via the 2008-2012 harmonisation project.

Bonardi S, Bruini I, Bolzoni° L, Cozzolino P, Pierantoni M, Brindani F, Bellotti° P, Renzi M, Pongolini° S

Assessment of Salmonella survival in dry-cured Italian salami

Int J Food Microbiol. - Vol. 262 (2017). - p 99-106. - 53 bib ref [Nr. Estr. 7665]

The inactivation of Salmonella during curing of Italian traditional pork salami was investigated. A total of 150 batches of ground raw meat (GRM) used for salami manufacturing by four producers were tested for Salmonella by real-time PCR followed by ISO 6579 cultural confirmation and MPN enumeration. Salami produced with Salmonella positive GRMs were re-tested at the end of their curing period. Aw, pH and NaCl content were also measured. Detection of Salmonella was performed testing both 25 and 50 g of the samples. By Real-Time PCR 37% of the GRMs resulted positive, but cultural detection of Salmonella was obtained in 14% of the samples only. Salmonella enumeration ranged from 31 MPN/g to < 1.3 MPN/g. The difference between testing 50 g and 25 g of the samples was statistically significant (p value = 0.01). In particular, ISO-50 g detected Salmonella in 100% of all positive samples, vs. 62% of ISO-25 g. Salami made of the contaminated GRMs were 29% Salmonella-positive, as most batches of salami produced with Salmonella-positive GRMs resulted negative after regular curing (20–48 days). Overall, 13% of salami produced with Salmonella-contaminated GRMs were positive. They belonged to six batches, which turned out negative after prolonged curing ranging between 49 and 86 days. Salmonella enumeration in salami ranged from 8.7 MPN/g to < 1.3 MPN/g. Unlike GRMs, no significant difference was observed between the ISO-50 g and the ISO-25 g in detecting Salmonella in cured salami (p value: > 0.05). The most common Salmonella serovars in GRMs were Derby (52%), Typhimurium monophasic variant 4, (Barbuti et al., 1993), 12:i:- (19%) and Stanley (10%). Salmonella Derby (56%), London, Branderup, Panama (13%, respectively) and Goldcoast (6%) were most frequent in cured salami. The study showed negative correlation between real-time CT values and cultural confirmation of Salmonella, as well as the importance of sample size for Salmonella detection. Among considered factors with possible effect on the occurrence of Salmonella in salami, statistical analysis revealed a role for aw in salami and for Salmonella load in GRMs, while pH and NaCl content did not

significantly affect the probability of finding Salmonella in dry-cured salami in the context of this study. In particular the lower aw values due to longer curing were associated with lower Salmonella presence in traditional dry-cured salami.

Buffoli° E, Bolzoni° G, Biancardi° A, Vitali A, Zanardi° G

Aflatoxin M1 : an extraordinary surveillance plan in Lombardy Region, Italy

18th International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD), June 7-10, 2017 : Sorrento, Italy : abstract book / [s.l. : s.n., 2017]. - p 274 (Poster 133). - 4 bib ref [Nr. Estr. 7598]

International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD) (18th : Sorrento : June 7-10, 2017)

Colagiorgi A, Filipello° V, Di_Ciccio P, Tilola° M, Finazzi° G, Ianieri A

Molecular characterization and biofilm formation of Staphylococcus aureus isolated from dairy products

71° Convegno Nazionale della Societa' Italiana delle Scienze Veterinarie (SISVET), XVII Convegno S.I.C.V, XV Convegno S.I.R.A, XIV Convegno AIPVet, XII Giornata Studio So.Fi.Vet, IV Convegno RNIV : I Convegno ANIV 28 Giugno - 1 Luglio 2017, Napoli / [s.l. : s.n., 2017]. - p 184 . - 5 bib ref [Nr. Estr. 7613]

Convegno Nazionale della Societa' Italiana delle Scienze Veterinarie (SISVET) : 71 Convegno SICV : 17 Convegno SIRA : 15 Convegno AIPVet : 14 Convegno So.Fi.Vet : 12 Convegno RNIV : 4 Convegno ANIV : 1 : Napoli : 28 Giugno - 1 Luglio 2017)

Staphylococcus aureus (*S. aureus*) is a foodborne pathogen considered to be the world's third most important causative agent of food borne illnesses [1]. Besides the production of enterotoxins, the formation of biofilm is increasingly being recognized as an important virulence factor in *S. aureus* [4]. The present study focused on the molecular characterization of *S. aureus* strains isolated from dairy products analyzing the biofilm-forming ability of the isolates. Forty strains were characterized by spa typing [2] and screened by PCR for the presence of enterotoxin encoding genes, and some genetic markers associated to the biofilm production [3]. Furthermore, the strains were tested for the biofilm production on polystyrene at 37°C [4]. A total of 19 spa types were found, the most frequent were t524 (7/40) and t2953 (5/40), which have been frequently reported also in other European countries. Majority of isolates (83%) showed similar distribution of adhesion genes (*icaA*, *icaD*, *cna*, *fnbA* and *fnbB*), toxin genes (*hla* and *hlb*), and staphylococcal regulators (*sarA*). Biofilm formation was observed in the 48% (19/40) of the isolates, of which 11% (2/19) strains formed biofilms strongly, 42% (8/19) moderately, and 47% (8/19) weakly. Interestingly, all strains carrying agr type III (5/40) were found to be biofilm producer, including the two strong biofilm producers. Furthermore, 62.5% of the isolates (25/40) were found to be potentially able to produce enterotoxins, carrying at least one gene encoding for staphylococcal enterotoxins. In conclusion, this study underlines the ability of *S. aureus* strains from dairy products to form biofilm. The biofilm formed by *S. aureus* on milk equipment surfaces can lead to significant food safety issues. The majority of strains (13/19) classified as biofilm producers, in fact, were found to be potentially able to produce enterotoxins. Currently, a regular cleaning and disinfecting is the first and most important step to get rid of raw milk residues and prevent biofilm formation by *S. aureus* in the dairy industry.

Comandatore F, Corbella M, Andreoli° G, Scaltriti° E, Aguzzi M, Gaiarsa S, Mariani B, Morganti° M, Bandi C, Fabbi° M, Marone P, Pongolini° S, Sasserà D

Genomic characterization helps dissecting an outbreak of listeriosis in Northern Italy

Plos Current Outbreaks. - Vol. 2017). - 23 p . - 33 bib ref (ultimo accesso 07/12/2017)

<http://currents.plos.org/outbreaks/article/genomic-characterization-helps-dissecting-an-outbreak-of-listeriosis-in-northern-italy/>) [Nr. Estr. 7710]

Listeria monocytogenes (Lm) is a bacterium widely distributed in nature and able to contaminate food processing environments, including those of dairy products. Lm is a primary public health issue, due to the very low infectious dose and the ability to produce severe outcomes, in particular in elderly, newborns, pregnant women and immunocompromised patients. **Methods** In the period between April and July 2015, an increased number of cases of listeriosis was observed in the area of Pavia, Northern Italy. An epidemiological investigation identified a cheesemaking small organic farm as the possible origin of the outbreak. In this work we present the results of the retrospective epidemiological study that we performed using molecular biology and genomic epidemiology methods. The strains sampled from patients and those from the target farm's cheese were analyzed using PFGE and whole genome sequencing (WGS) based methods. The performed WGS based analyses included: a) in-silico MLST typing; b) SNPs calling and genetic distance evaluation; c) determination of the resistance and virulence genes profiles; d) SNPs based phylogenetic reconstruction. **Results** Three of the patient strains and all the cheese strains resulted to belong to the same phylogenetic cluster, in Sequence Type 29. A further accurate SNPs analysis revealed that two of the three patient strains and all the cheese strains were highly similar (0.8 SNPs of average distance) and exhibited a higher distance from the third patient isolate (9.4 SNPs of average distance). **Discussion** Despite the global agreement among the results of the PFGE and WGS epidemiological studies, the latter approach agree with epidemiological data in indicating that one the patient strains could have originated from a different source. This result highlights that WGS methods can allow to better.

Cosciani_Cunico° E, Dalzini° E, Monastero° P, Daminelli° P, Losio° MN

Use of predictive tools to rank the safety of raw milk cheeses processes vs Verocytotoxic Escherichia coli

10th International Conference on Predictive Modelling in Food (ICPMF) : 26-29 September, 2017 Cordoba, Spain / [s.l. : s.n., 2017]. - 1 p (P. 80) [Nr. Estr. 7723]

International Conference on Predictive Modelling in Food (ICPMF) (10th : Cordoba, Spain : 26-29 September, 2017)

Verocytotoxin-(VT) producing *Escherichia coli* (EC) is a foodborne pathogen that can cause severe illness and death. Cattle is a natural reservoir of VTEC, consequently they may be isolated from raw cow milk and non-pasteurized dairy products, typically produced in many Italian regions. During the cheese making, the temperature is highly the most important control variable affecting the kinetics of the micro-organisms present in the milk and possibly surviving the process. The aims of the study were i) validate a predictive model available in literature with VTEC data obtained in broth at dynamic conditions (temperature and pH) that mimic the conditions of different cheese making processes and ii) rank the cheese processes regarding the probability to find VTEC at the beginning of seasoning. **MATERIAL AND METHODS** VTEC ATCC 35150 data was obtained in Brain Heart Infusion broth. A bio-reactor was used in this study and programmed with 8 temperature/pH dynamic profiles in growth region (15-20-25-30°C) and 16 profiles in survival region (55-52-50-48°C), pH (5.2, 6.5). The pathogen concentration was measured by plate count (ISO 4833-2:2013). The predictive model "E. coil safe ferment" proposed by Quinto et al. (2014) was validated by observed data obtained in this study. The accuracy factor (Ai) and Bias (B) were calculated according to Baranyi et al. (1999). Cheese making processes of Parnnigiano Reggiano (PR), Grana Padano (Gp), Silter (S), Formai de Mut (FM), Formaggella della Valsassina (FV), Formaggella Luinese (FL) were taken from literature. **RESULTS** In favorable condition, VTEC initial inoculum was 2.45+/-0.1 log CFU/ml, the observed log increase was 5.02 +1-0.63 log CFU/ml (ranging from 2.5 log CFU/ml to 6.03 log CFU/ml). In hostile environment, VTEC initial inoculunn was 5.65 +/- 0.67 log CFU/ml, the observed log reduction was 2.92+/-1.1 log CFU/ml (ranging from 5.42 to 1.19 log CFU/ml). 232 observed data, log VTEC CFU/ml, were compared with predicted values, Afand B between data were 1.36 and 0.86 respectively. During the cheese making, the model predict for PR and GP cheeses 4.1 log CFU/g and 3.02 log CFU/g VTEC reduction respectively; in cheeses S and FM the concentration remains

almost constant, with a 1.19 and 0.89 log CFU/g increase, while in cheeses FV and FL the VTEC concentration increases of 2.5 and 3.26 log CFU/g. **CONCLUSIONS** This study can be used to help the risk assessors, which have to judge the safety of the process of cheeses made by raw milk.

Costa MC, Goumperis T, Andersson W, Badiola J, Ooms W, Pongolini^o S, Saegerman C, Jurkovic M, Tuominen P, Tsigarida E, Steinwider J, Holzl C, Mikushinska N, Gross-Boskovic A, Kanari P, Christodoulidou M, Babicka L, Korsgaard H, Pesonen S, Fillet AM, Foures F, Lohman M, Lubert P, Szabo M, Cseh J, Noteborn HPJM, Faerden K, Fulke A, Trnovec T, Ilbaeck NG, Andersson T, Donohoe T, Merten C, Robinson T

Risk identification in food safety : strategy and outcomes of the EFSA emerging risks exchange network (EREN), 2010–2014

Food Control. - Vol. 73 (2017). - p 255-264. - 46 bib ref [Nr. Estr. 7517]

The European Food Safety Authority (EFSA) established an Emerging Risks Exchange Network (EREN) to exchange information between EFSA and the Member states (MSs) on possible emerging risks for food and feed safety in 2010. The Network is composed of delegates from MSs and Norway designated through the Advisory Forum of EFSA and observers from the European Commission, EU pre-accession countries, the Food and Drug Administration of the USA and the Food and Agricultural Organisation of the United Nations. Through 2010 to 2014, the EREN met 12 times. The EREN discussed a total of 63 signals of potential emerging issues that were presented and assessed using a standard template developed by the Emerging Risks unit of EFSA (EMRISK). Out of these signals, 39 originated from EFSA, 24 from MSs. The issues discussed were mainly microbiological and chemical hazards, but also food safety issues as result of illegal activity, new consumer consumption trends, biotoxins, new technologies and processes, allergens, animal health, environmental pollution, new analytical methods, new food packaging technology and unknown hazards were on the agenda. Based on the available evidence, EREN recommended whether an issue should be considered emerging or not, and if it merited further consideration, such as generating data on the issue, starting a full risk assessment and/or consultation of other bodies. According to the emerging risks identification process set in place at EFSA, the issues discussed and found of relevance by EREN were sent to the EFSA's Scientific Committee Standing Working Group on Emerging Risks for final evaluation. With four case studies, i.e the zoonotic potential of Usutu virus, risk of ciguatera fish poisoning in EU, zoonotic aspects of illegally imported wildlife products and benefits and risks of 3D food printing, the method developed to preliminary assess signals of potential emerging issues is presented and discussed.

Cristoni S, Dusi^o G, Brambilla P, Albini A, Conti M, Brambilla M, Bruno A, Di_Gaudio F, Ferlin L, Tazzari V, Mengozzi S, Barera S, Sialer C, Trenti T, Cantu M, Rossi_Bernardi L, Noonan DM

SANIST : optimization of a technology for compound identification based on the European Union directive with applications in forensic, pharmaceutical and food analyses

J Mass Spectrom. - Vol. 52 no 1 (2017). - p 16-21. - 18 bib ref [Nr. Estr. 7510]

Electrospray Ionization and collision induced dissociation tandem mass spectrometry are usually employed to obtain compound identification through a mass spectra match. Different algorithms have been developed for this purpose (for example the Nisr match algorithm). These approaches compare the tandem mass spectra of the unknown analyte with the tandem mass spectra spectra of known compounds inserted in a database. The compounds are usually identified on the basis of spectral match value associated with a probability of recognition. However, this approach is not usually applied to multiple reaction monitoring transition spectra achieved by means of triple

quadrupole apparatus, mainly due to the lack of a transition spectra database. The Surface Activated Chemical Ionization-Electrospray-NIST Bayesian model database search (SANIST) platform has been recently developed for new potential metabolite biomarker discovery, to confirm their identity and to use them for clinical and diagnostic applications. Here, we present an improved version of the SANIST platform that extends its application to forensic, pharmaceutical, and food analysis studies, where the compound identification rules are strict. The European Union (EU) has set directives for compound identification (EU directive 2002/657/EC). We have applied the SANIST method to identification of 11-nor-9- carboxytetrahydro-cannabinol in urine samples (an example of a forensic application), circulating levels of the immunosuppressive drug tacrolimus in blood (an example of a pharmaceutical application) and glyphosate in fruit juice (an example of a food analysis application) that meet the EU directive requirements. Copyright © 2016 John Wiley & Sons, Ltd.

Dalipi R, Borgese L, Margui E, Sangiorgi^o E, Depero LE

Total reflection X-ray fluorescence technique for multi-elemental analysis of food

Spectrosc Europe. - Vol. 29 no 1 (2017). - p 12-15. - 3 bib ref [Nr. Estr. 7529]

Dalipi^o R, Sangiorgi^o E, Berneri^o R

Exploring the possibility to distinguish MSM from non-MSM in chicken meat by means of TXRF

17th International Conference on Total Reflection X-ray Fluorescence Analysis and Related Methods : September 19-22, 2017, Brescia, Italy : program and book of abstracts / editor, Annalisa Zacco. - [Brescia : Smart Solutions srl, 2017]. - 1 p (Poster P104) [Nr. Estr. 7702]

International Conference on Total Reflection X-ray Fluorescence Analysis and Related Methods (TXRF) (17th : Brescia, Italy : September 19-22, 2017)

Meat products are of great importance in food since they are a rich source of nutrients, essential for growth and development of the living beings. However, the wide consumption of meat products it is not recommended by IARC, the cancer agency of the World Health Organization (WHO) due to cancer risks on humans [1]. Moreover, in the last years, the attention of scientific community has been addressed to a specific aspect of meat production, mechanically separated meat (MSM). According to EC Regulation 853/2004, MSM is a term for "the product obtained by removing meat from flesh-bearing bones after boning or from poultry carcasses, using mechanical means resulting in the loss or modification of the muscle fiber structure" [2]: MSM is produced from poultry and swine carcasses from which whole muscles have been cut to obtain as much meat as possible. The microbial risk grows with the muscle fiber degradation and under this aspect food with MSM could be carefully monitored and identification of a meat product obtained from MSM is very important from food quality and safety point of view. As reported from the European Food Safety Authority (EFSA) an important chemical parameter that could be used to distinguish MSM from non-MSM products is the content of calcium [3]. Consequently, the development of reliable and cost-effective analytical methods is very important to monitor the chemical composition of these foods. In this study, we have developed an analytical method for elemental analysis of meat samples that uses a low power benchtop total reflection X-ray fluorescence (TXRF) system. Sample preparation consisted in suspending a previously grinded (<100 µm) meat sample in an adequate disperser agent only. This procedure is fast, easy, and does not implicate the use of dangerous reagents. Fresh chicken meat, chicken meat with different percentage of MSM, pure MSM and meat products prepared with MSM were analyzed. MSM was obtained with low and high pressure mode. Statistical tests based on the use of principal component analysis (PCA) were performed for differentiation purposes. Evaluation of method accuracy was performed by comparing the obtained results with those obtained after acidic digestion and ICP-MS analysis..

Dalzini° E, Cosciani_Cunico° E, Monastero° P, Bernini V, Neviani E, Bellio A, Decastelli L, Losio° MN, Daminelli° P, Varisco° G

Listeria monocytogenes in Gorgonzola cheese : study of the behaviour throughout the process and growth prediction during shelf life

Int J Food Microbiol. - Vol. 262 (2017). - p 71-79. - 64 bib ref [Nr. Estr. 7655]

As reported on RASFF's portal, in the first 9months of 2016, a total of 13 "alerts/information for attention" were issued concerning the presence of *Listeria monocytogenes* in mould cheeses throughout Europe. This study analyzes the behaviour of *L. monocytogenes* in Gorgonzola cheese, a typical Italian soft blue-veined cheese, when contaminated at different time points. In the first challenge test, the pasteurized milk was contaminated and the complete cheese manufacture (cheesemaking, ripening) and shelf life was simulated. After a decrease during the first days of the cheesemaking, the pH remained constant for 35days (5weeks) and then it increased rapidly reaching the final values of 6.8 ± 0.02 in the core and 5.8 ± 0.4 on the rind. At the same time, the pathogen concentration decreased (about $2\log\text{CFU/g}$), although during the last week a rapid pathogen growth was observed after the rise in pH values. When the cheese was stored at thermal abuse condition ($8-12^{\circ}\text{C}$), the pathogen concentration on the rind was $4.8\pm 0.3 \log \text{CFU/g}$ and after 66days (about 9weeks) no significant difference ($p>0.05$) was observed; whereas, a growth from 5.4 ± 0.4 to $7.1\pm 0.5\log\text{CFU/g}$ was observed in the core. A second challenge test was performed using three batches of commercial slices of Gorgonzola cheese inoculated by *L. monocytogenes* and stored at 8°C . The maximum specific growth rates (μ_{max} , 1/h) of *L. monocytogenes* estimated ranged from 0.007 to 0.061. The square root model was used to predict the μ_{max} at others temperature and to establish the time necessary to reach the European critical legal limit of $2\log\text{CFU/g}$, in different storage scenarios. The predictions obtained in this study can be applied to any time-temperature profile, and in particular to the conditions to which the product is most likely to be subject in normal use, up to its final consumption. This study can be considered a valuable contribution also aimed at supporting the monitoring surveys carried out by officers of the Regional Veterinary Authority.

Dalzini° E, Cosciani-Cunico° E, Monastero° P, Varisco° G, Losio° MN, Daminelli° P

Modelling the behaviour of Listeria monocytogenes during the making of cheese from raw milk

European Symposium on Food Safety : 29-31 May 2017 Brussels, Belgium / [s.l. : s.n., 2017]. - p 110-111 (poster P2-25) [Nr. Estr. 7574]

European Symposium on Food Safety : Brussels, Belgium : 29-31 May 2017)

Introduction: The presence of *Listeria monocytogenes*, even if at low levels, in raw milk used for to produce raw milk cheeses represents a safety issue. Purpose: The aims of this study were i) to study the effect of the inoculum level of lactic acid bacteria (LAB) on the behaviour of *L. monocytogenes* during cheese making and ii) to develop of a predictive model to describe this effect, as well as the growth of LAB as a function of temperature. Methods: Raw milk was inoculated with three different LAB concentrations (4, 6, and $8 \log \text{CFU g}^{-1}$) and, also, contaminated with *L. monocytogenes* registered strain ATCC 19115. The contaminated milk was used to produce soft cheeses. The obtained data were used to validate a predictive model, which in turn was based on data, partly produced by IZSLER and partly available from the ComBase database (www.combase.cc). The model of Baranyi and Roberts was used to calculate the primary growth parameters, while the model of Ratkowsky was used to describe the growth rate as a function of the temperature. To take the inhibitory effect of the LAB concentration into account, we assumed that no cell division took place after LAB reached a concentration level (Jameson effect). Results: The concentration of *Listeria* did not exceed the level of $8 \log \text{CFU g}^{-1}$, which is normally its maximum population density, but stopped at around $5 \log \text{CFU g}^{-1}$ when the LAB reached their threshold concentration. The predicted data were in good agreement with independent observations in cheese, showing a discrepancy between 2.18% and 4.64%. Significance: This model may be a very useful tool to support the monitoring surveys carried out by officers of the Regional Veterinary

Authority.

Daminelli° P, Marchini D, Cosciani_Cunico° E, Dalzini° E, Monastero° P, Montanari R, Losio° MN

Sviluppo di un software che riproduca il comportamento termico di un banco frigorifero nella grande distribuzione organizzata

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locali e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia, 13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 19-20 (C040-44). - 2 bib ref [Nr. Estr. 7640]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

La temperatura (T) è uno dei fattori più importanti in grado di influenzare la capacità di moltiplicazione e sopravvivenza dei microrganismi negli alimenti. Una T di stoccaggio inappropriata durante la vita commerciale può sottostimare la capacità di crescita dei microrganismi, sovrastimando la shelf life degli alimenti pronti al consumo (RTE) e quindi ponendo a rischio la salute del consumatore. Obiettivo del lavoro è stato lo sviluppo di uno strumento software che aiuti a comprendere l'andamento termico dell'ambiente interno di un banco frigo di stoccaggio prodotti nell'ambito della grande distribuzione organizzata (GDO). Il progetto è articolato in due fasi: i) rilievo delle T all'interno di un banco frigo in diverse condizioni di funzionamento; ii) elaborazione dei dati e creazione di modelli empirico-matematici che riproduca la distribuzione di T all'interno del banco frigo utilizzato. La fase i) ha permesso di valutare le temperature nel banco in fase di quiete e l'impatto di fattori quali: cicli di sbrinamento, livello di carico del banco, affluenza clientela, T dell'ambiente nel quale è inserito il banco e T di set-up del banco. I dati raccolti hanno permesso di sviluppare un modello analitico-matematico per prevedere le temperature in tutte le zone del banco al variare delle condizioni di funzionamento dello stesso. Il modello predittivo è stato ricavato tramite regressione lineare e analisi della varianza. È stato poi tradotto in uno strumento software sviluppato ad hoc, basato su linguaggio VBA sfruttando l'ambiente Excel. Lo strumento è dotato di un'interfaccia grafica riassuntiva delle T previste in funzione delle condizioni al contorno inserite. Il progetto ha consentito di dedurre altre importanti indicazioni: le temperature interne dell'espositore sono fortemente influenzate da quella dell'ambiente nel quale si trova e da flussi d'aria prolungati direzionati contro il banco frigo; le perturbazioni introdotte dall'afflusso di persone non hanno evidenziato alcun impatto sul funzionamento del banco. Il progetto prevederà in futuro l'estensione dello strumento di previsione termica ai prodotti alimentari. Sarà in grado di prevedere l'andamento della T interna e superficiale dei prodotti al variare di quella dell'ambiente in cui si trovano e quindi approfondire lo studio degli effetti di deterioramento che avvengono nei prodotti in funzione della loro T reale (FDA/USDA/CDC, 2003; FAO/WHO, 2004).

Delibato E, Bilei S, Pucci E, Capuano F, Bertasi° B, Finazzi° G, Ferrari° M, Bossu T, Lovari S, Losio° MN, De_Medici D, Proroga YTR

Molecular approaches for evaluating the risk of pathogenic Yersinia enterocolitica along the pork production chain

Networking : tool for an excellent research: a public veterinary health without borders to face new emergencies : atti 4° Convegno Nazionale sulla Ricerca in Sanità Pubblica Veterinaria : Roma 6 Aprile 2017 / edited by Marina Bagni, Antonio Petrini, Antonio Lavazza. - [Teramo] : Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise, 2017. - (Veterinaria Italiana. Collana di Monografie ; Anno 53, Monografia ; 26) p 74 [Nr. Estr. 7908]

Convegno Nazionale sulla Ricerca in Sanità Pubblica Veterinaria (4° : Roma : 6 Aprile 2017)

Enteropathogenic *Yersinia* is the third most common cause of bacterial enteritis in European countries and infection is most often acquired by raw or undercooked pig meat. Slaughtered pigs are considered the principal animal reservoir for pathogenic strains of *Y. enterocolitica* (Ye). The presence of Ye in the pork production chain was detected by ISO Real-time PCR method. In order to evaluate the pathogenetic variability of Ye, different virulence genes were detected using molecular platforms with simultaneous detection. The platforms have been used to evaluate 153 strains of Ye and 130 samples taken along the pork chain. The isolated strains showed the presence of *ail* and *ystA* genes only in pathogenic bio-serotypes and in a strain 1A, isolated from a human case. The *ystB* locus has been identified in four non-pathogenic bio-serotypes, *fes* and *fepD* genes in most of the non-pathogenic bio-serotypes, and *virF* only in serotype 0:3. The results obtained from the analysis of samples showed the absence of all *ystA* and *virF* genes, and the detection of *ystB*, *fes* and *fepD* in about 45% of the samples. The developed methods allow the rapid identification and characterization of Ye, in order to estimate the microbiological risk along the pork chain.

Di_Paolo A, Cornell S, Ortenzi R, Sebastiani C, Ciullo M, Spaccini G, Biagetti M, Valiani A, Pezzotti G, Cammi° G, Arrigoni° N, Mazzone PM

Survival of *Mycobacterium avium* subsp. *paratuberculosis* during Pecorino cheese production : preliminary data

18th International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD), June 7-10, 2017 : Sorrento, Italy : abstract book / [s.l. : s.n., 2017]. - p 290 (Poster 149). - 6 bib ref [Nr. Estr. 7599]

International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD) (18th : Sorrento : June 7-10, 2017)

Filipello V, Gallina S, Amato E, Losio° MN, Pontello M, Decastelli L, Lomonaco S

Diversity and persistence of *Listeria monocytogenes* within the Gorgonzola PDO production chain and comparison with clinical isolates from the same area

Int J Food Microbiol. - Vol. 245 (2017). - p 73-78. - 50 bib ref [Nr. Estr. 7518]

Listeria monocytogenes causes invasive syndromes with high fatality rates in specific population groups. Cheeses have been commonly implicated in outbreaks worldwide. Gorgonzola is a cheese only produced in Northwestern Italy (it is the third Italian cheese in terms of production and export) and *L. monocytogenes* is frequently isolated from the production chain. The aims of this study were to assess the distribution of *L. monocytogenes* Virulence Types (VTs) in isolates collected in Gorgonzola processing plants and to determine the presence of Epidemic Clones (ECs). Fifty-Six *L. monocytogenes* strains collected between 2004 and 2016 from cheese and environmental samples were subtyped with Multi-Virulence-Locus Sequence Typing (MVLST) and compared to previously typed strains. Most isolates ($n = 50$) belonged to two new VTs (VT113 and VT114). The remaining isolates belonged to previously identified VTs: VT14-ECVIII (milk chocolate outbreak, 1994, USA) and VT80 (ricotta salata outbreak, 2012, USA). VT14, VT80 and VT113 were shared with isolates from apparently sporadic human cases in the same geographical area and temporal period (Piedmont and Lombardy, 2005–2016). The overall *L. monocytogenes* population appears to be homogeneous and may be characteristic of Gorgonzola production. Nevertheless, the detection in cheese and environmental samples of VTs observed in clinical isolates or outbreak related strains (VT80, VT14) contributed to better describe the current scenario and pointed out the need for increased surveillance.

Finazzi° G, Bertasi° B, Bornati° L, Benevenia° R, Losio° MN, Varisco° G

Diagnosis of human botulism in Northern Italy during 2012-2016

Since hospitals usually do not have specific analytical protocols, the differential diagnosis between food botulism and other diseases characterized by neurological symptoms is difficult. Microbiology Dep. of IZSLER provides support to Northern Italy Hospitals in detecting preformed toxin and C. botulinum (Cb) strains in food or human biological samples. Description of the problems Toxin detection is carried out with the Mouse test, that requires equipped structures authorized for the use of laboratory animals. Detection of Cb is performed by a multiplex PCR. In case of foodborne illness, beyond the diagnostic confirmation, it is challenging to identify the implicated food. Botulism symptoms could appear even 4-5 days after ingestion of a contaminated food, therefore it is difficult to have a full medical history of the potentially responsible foods and in most cases leftovers are no longer available for diagnostic confirmation. Results In the last 5 years, we have been contacted for 38 events of suspected human botulism: 9 were confirmed and involved 11 persons. In 5 cases the suspected food was homemade vegetable preserve (not available in 2 cases), in 1 case vacuum preserved smoked salmon, in 1 case chamomile sachets, and in 2 cases no hypothesis were made. Cb Type A, B and E were identified: 3 patients Type A (2 linked to positive homemade canned chickpeas), 5 patients Type B (1 linked to positive homemade cherries preserve), 1 Type E. In 2 cases the strains were not typed. Lessons Food botulism was not confirmed in 31 out of 38 events, still providing diagnostic elements to direct clinical suspicions to other diseases. These data confirm the validity of the diagnostic methods used, but also underline the difficulty of identifying with certainty the food involved in confirmed cases.

Finazzi° G, Losio° MN, Bertasi° B, Panteghini C, Pavoni° E, D'Incau° M, Bonomini A, Pedroni P

Epidemiological study of Salmonella spp. strains isolated in Lombardia from 2012-2016

European Symposium on Food Safety : 29-31 May 2017 Brussels, Belgium / [s.l. : s.n., 2017]. - p 92 (poster p1-22) [Nr. Estr. 7572]

European Symposium on Food Safety : Brussels, Belgium : 29-31 May 2017)

Introduction: In cases of foodborne illness, the biggest challenge is to identify the offending food. Symptoms could appear after a few hours, but, sometimes four to five days after ingestion of a contaminated food. As a consequence, it is difficult to have a complete medical history of the potentially responsible foods; and in most cases, they are not available for diagnostic confirmation because they have been completely consumed or thrown away. Purpose: The objects of this study were to analyze the data obtained from molecular characterization of Salmonella strains, isolated from food and environmental farm samples, rather than from humans; to evaluate the possible correlations; and to speculate on possible reservoirs for Salmonella that can cause human illness. Methods: A total of 1809 Salmonella spp. strains were isolated from food (1169), farm (464), and the Lombardia environment (176), along with 148 human strains made available by Hospitals from the same region were serotyped according to ISO 6579-3:2014 and molecular typed by PFGE as per the CDC (Atlanta) Pulsnet System. The data obtained were elaborated and compared using BioNumerics software (Applied Maths). Results: Among human strains monophasic Salmonella Typhimurium was the serotype most frequently isolated (42%), followed by Salmonella Typhimurium (7.4%), and Salmonella Enteritidis (4.7%). Mono-phasic Salmonella Typhimurium was, also, the serotype most isolated from other sources (25.4% of strains considered); Salmonella Derby and Salmonella Infantis, (16.4% and 11.2%, respectively), were the other prevalent serotypes. Molecular comparison was able to point out some situation of close affinity between strains isolated from human and food. Significance: This work demonstrates a good approach for obtaining a picture of circulating strains of Salmonella in Lombardia. The approach could be used to create a system of epidemiological surveillance and to schedule monitoring plans for production chains that represent the greatest potential as reservoirs for human infections.

Fontana° MC, Bardasi° L, Rubini° S, Taddei° R, Karaman° I, Barbieri S, Formaglio A, Guidi E, Bergamini M

Gestione di un caso di latte crudo bovino destinato al consumo umano contaminato da *Listeria monocytogenes*

50. Congresso Nazionale Società Italiana di Igiene (SITI) : Torino, 22-25 Novembre 2017 : volume degli atti / [s.l. : s.n., 2017]. - p 283 [Nr. Estr. 7788]

Congresso Nazionale Società Italiana di Igiene (SITI) (50. : Torino : 22-25 Novembre 2017)

INTRODUZIONE: La vendita diretta di latte crudo bovino attraverso distributori automatici e autorizzata in Italia dal 2004 e viene monitorata per i pericoli microbiologici in base alla normativa vigente. *Listeria monocytogenes* rientra fra i patogeni ricercati, è ampiamente diffusa nell'ambiente ed è un patogeno opportunista dell'uomo e degli animali in grado di causare manifestazioni cliniche molto gravi quali meningoccefaliti, setticemie, aborti. La presenza di microrganismi patogeni nel latte crudo dipende da vari fattori correlati al tipo di allevamento e di alimentazione, alle pratiche igieniche utilizzate in allevamento e alla gestione dei distributori di latte. La contaminazione può essere di natura ambientale o derivare dall'eliminazione diretta del microrganismo patogeno attraverso il latte. Viene descritto un caso di presenza di *L. monocytogenes* in latte crudo destinato al consumo diretto. **MATERIALI E METODI:** Durante le analisi svolte nell'ambito del controllo ufficiale periodico a carico di questa tipologia di prodotto, tre campioni prelevati presso distributori afferenti ad uno stesso allevamento sono risultati non conformi per presenza di *L. monocytogenes*. Sono state considerate le possibili fonti di infezione in allevamento quali mangime ed animali portatori: sono stati esaminati per *L. monocytogenes* 3 campioni di insilato prodotto in azienda e 78 campioni di latte prelevati dai singoli soggetti in lattazione (campioni costituiti da pool del latte del quart' delta mamnella). La fase di screening per la presenza del patogeno è stata effettuata con metodo PCR Real Time AFNOR BRD 07/10-04/05, la conferma microbiologica e la numerazione rispettivamente con ISO 11290-1:1996 Amd1:2004 e ISO 11290-2:1998 Amd1:2004. **RISULTATI:** I campioni di mangime sono risultati non contaminati mentre fra i campioni di latte dei soggetti in lattazione uno risultato contaminato da *L. monocytogenes* (500 UFC/mL). **CONCLUSIONI:** Il latte crudo è risultato contaminato dall'eliminazione diretta di *L. monocytogenes* da parte di un bovino portatore asintomatico presente in allevamento. Il controllo periodico di questa matrice ha consentito la pronta individuazione di un rischio. L'identificazione della causa con esclusione del soggetto ha riportato la matrice in condizioni di idoneità al consumo umano.

Franzini° G, Micheli A, Daminelli° P, Cosciani_Cunico° E, Dalzini° E, Spagnoli° F, Todeschi° S, Losio° MN

Aspetti igienico-sanitari della carne separata meccanicamente : dati preliminari

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locali e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia, 13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 23-24 (Poster P002-74). - 1 bib ref [Nr. Estr. 7641]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

La carne separata meccanicamente (CSM) è un prodotto diverso dalla carne generalmente intesa e le sue caratteristiche potrebbero renderla più sensibile alla contaminazione microbiologica (EFSA, 2013). Al fine di valutare gli aspetti igienico-sanitari sono stati confrontati 6 lotti di CSM (4 di polio e 2 di suino) e di carne macinata tradizionale (CM) durante la shelf-life con conservazione ad 8°C; sono state condotte misurazioni del valore di pH, numerazioni della flora indigena (carica batterica totale, CBT) e ricerca genotipica dei principali patogeni correlati, *Salmonella* spp. (Ss), *Campylobacter* spp. (Cs) e *Listeria monocytogenes* (Lm). Attraverso challenge test è stato calcolato il tasso di crescita (rate) di Ss a 15°C e il suo tasso di inattivazione (rate) a 55°C (2). La differenza nella CBT tra le due matrici è risultata statisticamente significativa ($P < 0,05$): in CSM è inferiore

(5,65±0,64 vs 6,39±0,73 log UFC/g), ma aumenta più rapidamente (9±0,33 vs 8,07±0,53 log UFC/g). Il pH b risultato pad a 5,80±0,14 in CSM e a6,24±0,18 in CM. A tempo 0 la CSM b risultata Cs-positiva (0,39%, I.C. 0,20-0,61), Lm-positiva (0,17%, I.C. 0,06-0,39), Ss-positiva (0,22%, IC 0,09-0,45), mentre la CM b risultata esclusivamente Cs-positiva (0,22% I.C. 0,09-0,45). In CSM la rate di Ss e risultata maggiore (P<0,05) rispetto a CM a 15 °C (0,04±0,007 vs 0,02±0,003) e a 55°C (-2,84±0,27 vs - 2,3±0,26). I dati ottenuti confermano l'esistenza di una differenza tra le due matrici nei confronti dei microrganismi. CSM è risultata maggiormente contaminata da patogeni alimentari; la concentrazione di Ss aumenta e diminuisce più velocemente in questa matrice, nelle condizioni testate. E quindi necessario un trattamento adeguato dal punto di vista igienico-sanitario durante la produzione, la successiva trasformazione e al momento del consumo prestando attenzione ai possibili pericoli biologici correlati. Lavoro finanziato dal Ministero della salute nell'ambito del PRC 003/2013: applicazione di nuove metodiche per l'identificazione dei pericoli microbiologici, chimici e fisici relativi al consumo di carni avicole e suine separate meccanicamente.

Galuppini° E, Finazzi° G, Filipello° V, Tilola° M, Zani L, Colagiorgi A, Campagna° D, Di_Ciccio PA, Ianieri A, Losio° MN, Luini° M

Caratterizzazione di isolati di Staphylococcus aureus provenienti da prodotti lattiero-caseari a latte crudo tipici delle valli bresciane

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locali e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia, 13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 30 (Poster P015-52). - 2 bib ref [Nr. Estr. 7647]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

La tossinfezione da *S. aureus* è una malattia alimentare a diffusione globale causata da enterotossine stafilococciche (SE) preformate in alimenti contaminati. *S. aureus* è isolato spesso da prodotti lattierocaseari, soprattutto a latte crudo, che potrebbero rappresentare una minaccia per la salute umana (D'Amico et al., 2011). Nelle regioni alpine prodotti a latte crudo sono stagionalmente realizzati in piccoli caseifici d'alpeggio artigianali. La contaminazione dei prodotti, è legata al rilascio nel latte di *S. aureus* da parte di animali con mastite subclinica, oppure all'igiene di operatori carrier, durante le fasi di lavorazione (Rola et al., 2016). I dati sulla caratterizzazione di *S. aureus* isolati da prodotti di alpeggio sono tuttavia scarsi. Lo scopo di questo lavoro è stato di caratterizzare 49 isolati di *S. aureus* da prodotti di alpeggio valutandone i) i genotipi con MLST e spa typing; ii) la meticillino-resistenza con PCR per *mecA* e *mecC*; iii) i profili enterotossigeni con PCR multiplex per sea-b-c-d-e-g-h-i-j-p-r; iv) la produzione di biofilm su polistirene a 37°C; v) la presenza di geni biofilm associati (PCR per 10 geni marker). I geni delle SE sono state trovate nel 62.5% (n=25) degli isolati. Con MLST e spa typing è stato possibile identificare 3 principali cluster di isolati: ST8/t2935/sea-d-j-r rappresentato principalmente da isolati di matrice bovina (n=16, 33%); S171/t524/sed (n=9, 18%) e ST97/sed (n=7, 12%). Sono stati identificati 2 isolati meticillino-resistenti (MRSA) appartenenti a un cluster di 3 isolati tipizzati come ST1/t1277seh. produzione di biofilm è stata osservata nel 38% degli isolati (n=19) di cui 2 forti, 8 moderati e 9 deboli produttori. La presenza di geni biofilm associati sembra non essere predittiva del fenotipo. I ceppi enterotossigeni risultano molto comuni nei prodotti di alpeggio; la maggior parte degli isolati sembrano essere correlati a mastite, ma sono stati identificati anche ceppi isolati prevalentemente da casi umani. I MRSA sembrano avere un ruolo minore.

Gamba° V, Bertoni° L, Facchetti° S, Bozzoni° E, Pellicciotti° S, Berardi° A, Moneta° C, Tognoli° N, Ferretti° E

Residues of coccidiostats in eggs : results of seven-year monitoring in Lombardia and Emilia-Romagna

5th MS-Food Day : Bologna, October 11-13, 2017 / [s.l. : s.n, 2017]. - p 202-205 (Poster P19). - 5 bib ref [Nr. Estr. 7718]

Mass Spectrometry Food Day (5th : Bologna : October 11-13, 2017)

The LC-MS/MS method for the detection of coccidiostats in eggs was introduced in Lombardia and Emilia Romagna at the end of 2009. Here, we give a general overview of the results obtained between 2010 and 2016, and we characterize the population of tested samples in relation to the type of farm in which eggs were produced.

Gamba° V, Bolzoni° G, Monastero° P, Daminelli° P

Persistence of antibiotic residues in milk during the cheese making process : Desfuoylceftiofur case study

5th MS-Food Day : Bologna, October 11-13, 2017 / [s.l. : s.n, 2017]. - p 209-211 (Poster P21). - 3 bib ref [Nr. Estr. 7719]

Mass Spectrometry Food Day (5th : Bologna : October 11-13, 2017)

We have determined the persistence of Desfuoylceftiofur in dairy products after the cheese production process. Upon spiking milk with the antibiotic at a concentration equal to the Maximum Residue Limit (MRL), we show that almost half of the analyte persists in whey, while its concentration increases in curd, exceeding the MRL.

Genchi M, Vismarra A, Mangia C, Faccini° S, Vicari° N, Rigamonti° S, Prati° P, Marino AM, Kramer L, Fabbì° M

Lack of viable parasites in cured 'Parma Ham' (PDO), following experimental Toxoplasma gondii infection of pigs

Food Microbiol. - Vol. 66 (2017). - p 157-164. - 36 bib ref [Nr. Estr. 7547]

Twelve Large White pigs were experimentally infected with 1000 Toxoplasma gondii oocysts/each. Serology was carried out at different time points post infection (p.i.) and animals were slaughtered at four months p.i. One of two thighs was examined for T. gondii infection status by PCR and bioassay in mice. The other thigh was processed for Parma ham production. Four thighs were examined after twelve months of curing, four after fourteen months and four were examined after sixteen months. Cured hams were analyzed by PCR, bioassay and in-vitro cultivation on Vero cells followed by real-time PCR. Pigs seroconverted from day 21 p.i. Bioassays were positive for all fresh thighs, but negative for cured hams. PCR was positive for parasite DNA from most thighs both at slaughter and post curing, but parasite growth was not observed following in vitro cultivation and real-time PCR. Results indicate that the curing process of Parma Ham (PDO), when carried out according to the Parma Ham consortium regulations, can inactivate T. gondii tissue cysts. Results would suggest that food-borne transmission of T. gondii to consumers from Parma ham can be excluded.

Giacometti F, Bonilauri° P, Piva S, Scavia G, Amatiste S, Bianchi DM, Losio° MN, Bilei S, Cascone G, Comin D, Daminelli° P, Decastelli L, Merialdi° G, Mioni R, Peli A, Petruzzelli A, Tonucci F, Liuzzo G, Serraino A

Paediatric HUS cases related to the consumption of raw milk sold by vending machines in

Italy : quantitative risk assessment based on Escherichia coli O157 official controls over 7 years

Zoonoses Public Health. - Vol. 64 (2017). - p 505-516. - 35 bib ref [Nr. Estr. 7907]

A quantitative risk assessment (RA) was developed to estimate haemolytic uremic syndrome (HUS) cases in paediatric population associated with the consumption of raw milk sold in vending machines in Italy. The historical national evolution of raw milk consumption phenomenon since 2008, when consumer interest started to grow, and after 7 years of marketing adjustment, is outlined. Exposure assessment was based on the official Shiga toxin-producing Escherichia coli O157:H7 (STEC) microbiological records of raw milk samples from vending machines monitored by the regional Veterinary Authorities from 2008 to 2014, microbial growth during storage, consumption frequency of raw milk, serving size, consumption preference and age of consumers. The differential risk considered milk handled under regulation conditions (4°C throughout all phases) and the worst time-temperature field handling conditions detected. In case of boiling milk before consumption, we assumed that the risk of HUS is fixed at zero. The model estimates clearly show that the public health significance of HUS cases due to raw milk STEC contamination depends on the current variability surrounding the risk profile of the food and the consumer behaviour has more impact than milk storage scenario. The estimated HUS cases predicted by our model are roughly in line with the effective STEC O157 associated HUS cases notified in Italy only when the proportion of consumers not boiling milk before consumption is assumed to be 1%. Raw milk consumption remains a source of E. coli O157:H7 for humans, but its overall relevance is likely to have subsided and significant caution should be exerted for temporal, geographical and consumers behaviour analysis. Health education programmes and regulatory actions are required to educate people, primarily children, on other STEC sources.

Grisenti MS, Meriardi° G, Ramini° M, Bardasi° L, Frustoli MA, Barbuti S, Daminelli° P, Brutti A

Valutazione dell'utilizzo del trattamento ad alte pressioni come ulteriore strumento per il controllo di Listeria monocytogenes e Salmonella enterica in salami italiani = Evaluation of High Pressure Processing as an additional hurdle to control Listeria monocytogenes and Salmonella enterica in Italian salami

Industria Conserve. Quaderno Tecnico. - Vol. no 9 (2017). - p 22-30. - 26 bib ref [Nr. Estr. 7632]

I salami sono prodotti carni fermentati e stagionati, tradizionalmente considerati come sicuri sulla base delle caratteristiche di basso pH, bassa attività dell'acqua e presenza di conservanti. Questi prodotti, infatti, nel corso della stagionatura sviluppano condizioni tali da garantirne la sicurezza sanitaria; in particolare la caduta di pH determinata dalla fermentazione lattica, la riduzione dei valori di aw legata all'asciugatura che si verifica nel corso della stagionatura, l'aggiunta di sale e di additivi, la presenza di una cospicua flora microbica tipica costituiscono una combinazione sinergica tale da contrastare la crescita di microrganismi patogeni. Tuttavia, diversi casi di salmonellosi di origine alimentare sono stati associati al consumo di salame e Listeria monocytogenes e stata spesso isolata da questi prodotti, a dimostrazione del fatto che l'assenza di un definito step di letalità potrebbe, in alcuni casi, determinare il fallimento nel controllo dei rischi microbiologici. Trattamenti ad alte pressioni (HPP) vengono applicati per migliorare la qualità microbiologica di diversi alimenti. Recentemente in Italia, l'utilizzo delle alte pressioni è stato esteso anche per il trattamento di salumi stagionati e/o fermentati. Al fine di valutare l'efficacia dell'HPP per l'inattivazione di Salmonella e L. monocytogenes in campioni di salame artificialmente contaminati, è stato allestito un esperimento utilizzando due differenti tipologie di salami italiani. In considerazione degli effetti conosciuti sulla baroresistenza dei microrganismi in riferimento all'attività dell'acqua e al contenuto di grasso dell'alimento in cui sono contenuti, l'esperimento è stato incentrato su prodotti con caratteristiche standard: contenuto di grassi variabile tra il 28-32% e aw compresa tra 0,90 e 0,92. I salami stagionati sono stati forniti da sette differenti aziende produttrici e complessivamente sono state utilizzate: 4 produzioni di salame a grana fine e tre a grana media. I campioni sono stati contaminati in laboratorio e una parte è stata sottoposta a trattamento HPP, mentre altri sono stati utilizzati come controlli. Il processo HPP è stato applicato, per entrambi i patogeni, a una pressione di 600 MPa per 5

minuti con temperatura dell'acqua di 5°C, e per Salmonella anche di 12°C e 20°C. I dati riportati in questo studio confermano l'efficacia del trattamento HPP nel ridurre significativamente le concentrazioni dei patogeni studiati nella matrice salame stagionato.

Salami are cured fermented pork meat products. The low pH level reached by effect of the fermentation along with the reducing of water activity during seasoning, the presence of preservatives and the high concentration of Lactobacilli allow these products to achieve a general good hygienic standard. Nevertheless, several cases of food borne salmonellosis have been linked to salami consumption and Listeria monocytogenes is often isolated from salami, proving that the absence of a defined lethal step can in some circumstance determine a failure in the control of microbiological hazards. High Pressure Processing (HPP) has been applied to several foodstuff in order to increase their microbiological quality Recently HPP has been applied also to pork products in Italy In order to evaluate and validate the efficacy of HPP on the reduction of an artificial contamination with Salmonella and L. monocytogenes in Italian salami, an experimental trial was performed on different types of these products. In consideration of the expected impact of water activity and fat on the effectiveness of HPP the experiment was focused on products with standard characteristic: fat content ranging between 28-32% and aw 0.90-0.92. Samples obtained from seven different plants (4 fine grained and 3 medium grained) were contaminated in the laboratory and part of the samples were submitted to HPP while other samples were used as controls. For both pathogens the HPP process was applied, at a pressure of 600 MPa for 5 minutes with water temperature of 5°C and for Salmonella also at 12°C and 20°C. The results of this work can help food processors to set optimum technological parameters of high pressure processing to inactivate pathogenic microorganisms in salami and to evaluate the application of HPP as lethality or post lethality treatment.

Losio° MN, Dalzini° E, Pavoni° E, Merigo° D, Finazzi° G, Daminelli° P

A survey study on safety and microbial quality of “gluten-free” products made in Italian pasta factories

Food Control. - Vol. 73 (2017). - p 316-322. - 60 bib ref [Nr. Estr. 7399]

The rising prevalence of celiac disease leads to an increased demand of “gluten-free” products. A survey study on the gluten content and on the microbiological quality of “gluten-free” flour, and processing flour products, was carried out from 2010 to 2015 in Northern Italy. Overall 12,419 samples were analyzed, and 94.7% contained a gluten concentration less than 5 mg kg⁻¹ (lower limit of detection). Only 0.1% of samples showed a gluten concentration above 80 mg kg⁻¹ (maximum limit of detection). In the remaining 5.2%, the gluten concentration was between 5 and 80 mg kg⁻¹, underlining how a gluten-free diet completely devoid of gluten is unrealistic. The microbiological quality of these products was investigated. Overall, the majority of samples revealed microbial loads of less than 1 I g CFU g⁻¹ (lower limit of detection). High levels of spoilage bacteria were found in egg-containing products. Total mesophilic bacteria were counted in all analyzed food categories with concentrations up to about 6, 8 and 9 I g CFU g⁻¹ in dry pasta, flours and egg products respectively. Listeria monocytogenes was found only in one sample, whereas Salmonella spp. was never found. Buckwheat flour was the most frequently contaminated product by presumptive Bacillus cereus, with a prevalence of 12.5%. Also, a contamination by Coagulase-Positive Staphylococci was found during this investigation, especially in buckwheat dry pasta and flour and in egg dry pasta, with a prevalence of 54.7%. This study aimed to enhance the knowledge about the “gluten-free” products which are still poorly studied, even if their impact on the food market is increasingly considerable.

Losio° MN, Galuppini° E, Pavoni° E, Benevenia° R, Bertasi° B

Monitoraggio preliminare in campioni a rischio di contaminazione da virus dell'Epatite E

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locale e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia,13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 36

(Poster P027-85). - 5 bib ref [Nr. Estr. 7645]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

L'infezione da HEV, in Europa, è considerata un problema emergente di sanità pubblica. Fino a pochi anni fa, la maggior parte dei casi era individuata in viaggiatori provenienti da aree endemiche. Recentemente, invece, si è registrato un incremento del numero di casi sporadici in soggetti senza anamnesi di viaggi all'estero, ascrivibili a ceppi di HEV endemici per i quali la trasmissione più probabile è stata di origine alimentare (ciclo oro-fecale) o zoonotica (considerando il suino come principale serbatoio animale). Studi effettuati in diversi Paesi europei hanno evidenziato un valore medio di sieroprevalenza del 21% nell'uomo e del 50% nel suino (Adlhoch et al., 2016; Pavoni et al., 2015). Queste evidenze, rafforzate dalla stretta somiglianza genomica fra ceppi suini ed umani (Pavoni et al., 2015; Di Bartolo et al., 2012), hanno indotto anche gli organismi europei di riferimento per la sorveglianza e il monitoraggio delle zoonosi e dei rischi ad esse connessi (ECDC, EFSA, IMA) ad affrontare le problematiche del possibile rischio alimentare/zoonotico legato a HEV (EFSA, 2011). Per questo motivo, nel periodo giugno-dicembre 2016, presso l'IZSLER di Brescia sono stati analizzati 372 campioni tra cui mitili, acqua potabile, frutti di bosco, vegetali della IV gamma, preparazioni gastronomiche e fegati di suino prelevati al macello. A seguito di una fase di omogeneizzazione dei campioni e di estrazione dell'RNA virale, è stata eseguita l'amplificazione sul cDNA con una metodica Real Time PCR precedentemente descritta (Martinez-Martinez et al., 2011). Nessuna delle matrici considerate è risultata positiva per la presenza di RNA virale. Questi risultati preliminari, ottenuti mediante un progetto autofinanziato dall'Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna, saranno ulteriormente implementati nell'ambito del progetto CCM2016 Epatite E, un problema emergente in sicurezza alimentare: approccio One Health per la valutazione del rischio.

Losio^o MN, Tilola^o M, Mangeri^o L, Benevenia^o R, Delibato E, Crescenzi E, De_Santis P, Bertasi^o B

Application of digital PCR to perform species identification in food

European Symposium on Food Safety : 29-31 May 2017 Brussels, Belgium / [s.l. : s.n., 2017]. - p 104-105 (poster P2-09) [Nr. Estr. 7575]

European Symposium on Food Safety : Brussels, Belgium : 29-31 May 2017)

Introduction: The setting-up of new methods for species identification in food is a very important topic to avoid frauds. In fact, to verify foodstuffs compliance to labels and to discriminate accidental contamination from voluntary addition, it is necessary to quantify ingredients. To date, European legislation does not indicate how to achieve this goal, but several studies are in progress to develop and standardize new quantitative methods. **Purpose:** The aim of this work was to verify the applicability of Digital PCR for species identification in food; in particular, to quantify chicken after artificial contamination in raw meat. **Methods:** Digital PCR (Quant Studio 3D®) was applied to evaluate three topics: 1) Linearity between the spiking percentage of chicken (100% to 0.1%) and related observed genomic copies. 2) Correspondence between different targets (chicken, turkey, beef, pork) and a housekeeping gene (miostatin) to develop a ratio comparable with known target percentage. 3) Repeatability of 1% chicken in different meat categories. **Results:** Linearity between target percentage in spiked material and related genomic copies value was verified ($R^2=0.9996$). However, due to DNA extracts' moot quantifications, several troubles were observed in attributing the exact copies value to each spiking level. Miostatin was confirmed as the housekeeping gene for chicken, while a discrepancy between target and miostatin copies was observed for the other tested species. Finally, chicken genomic copies in swine and bovine raw meat, were not comparable for each sample. **Significance:** These data suggest that the digital PCR system could be applied in species identification. However, further studies will be necessary to investigate the possibility of different housekeeping genes and to validate repeatability studies in order to verify the extraction efficiency.

Mangeri L, Losio° MN, Rubini° S, Bertasi° B, Pavoni° E, Meletti° F, Galuppini° E

Monitoraggio e determinazione del virus dell'Epatite A e di norovirus in molluschi eduli lamellibranchi del nord Adriatico

Le sinergie tra grande distribuzione organizzata, industria, piccole produzioni locale e controllo ufficiale : tutela del consumatore, difficoltà e prospettive : XXVII Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) : Perugia,13-14-15 Settembre 2017 / [s.l. : s.n., 2017]. - p 39 (Poster P034-102). - 1 bib ref [Nr. Estr. 7648]

Convegno Nazionale Associazione Italiana Veterinari Igienisti (AIVI) (27. : Perugia : 13-14-15 Settembre 2017)

Dati epidemiologici riguardanti le tossinfezioni trasmesse con i prodotti alimentari hanno evidenziato come i virus enterici rappresentino un rischio in aumento per la salute pubblica. Negli ultimi anni, le Autorità Competenti hanno messo in atto piani di monitoraggio per il controllo della salubrità degli alimenti nei confronti delle contaminazioni virali. Il piano di sorveglianza delle zone di produzione di molluschi bivalvi attuato dal servizio sanitario regionale dell'Emilia Romagna ha raccolto, nel periodo compreso tra dicembre 2016 e marzo 2017, 70 campioni di molluschi eduli lamellibranchi. Tra questi, cozze (*Mytilus galloprovincialis*), vongole (*Tapes philippinarum*) e ostriche (*Crassostrea* spp.) prelevati lungo la costa romagnola. I campioni sono stati sottoposti ad analisi qualitativa per la ricerca del virus dell'Epatite A e dei norovirus GI-GII, in conformità alla norma ISO/TS 15216-2:2013, che ha permesso di rilevare, mediante One-Step Real Time PCR, 27 campioni positivi a norovirus GII. Successivamente si è proceduto alla quantificazione del virus nelle matrici contaminate secondo quanto previsto dalla norma ISO/IS 15216-1:2013, la quale ha evidenziato le differenti concentrazioni del virus nei campioni positivi. URNA di questi ultimi è stato infine retrotrascritto ed amplificato mediante una semi-nested PCR avente come target una zona della ORF2. 1 prodotti di PCR sono stati sottoposti a sequenziamento per approfondire le eventuali correlazioni tra i genogruppi (Kojima et al., 2002).

Marcoccia° D, Pellegrini M, Fiocchetti M, Lorenzetti S, Marino M

Food components and contaminants as (anti)androgenic molecules

Genes & Nutrition. - Vol. 12 (2017). - Article no. 6 (16 p). - 171 bib ref (ultimo accesso 03/11/2017 <https://genesandnutrition.biomedcentral.com/articles/10.1186/s12263-017-0555-5>) [Nr. Estr. 7654]

Androgens, the main male sex steroids, are the critical factors responsible for the development of the male phenotype during embryogenesis and for the achievement of sexual maturation and puberty. In adulthood, androgens remain essential for the maintenance of male reproductive function and behavior. Androgens, acting through the androgen receptor (AR), regulate male sexual differentiation during development, sperm production beginning from puberty, and maintenance of prostate homeostasis. Several substances present in the environment, now classified as endocrine disruptors (EDCs), strongly interfere with androgen actions in reproductive and non-reproductive tissues. EDCs are a heterogeneous group of xenobiotics which include synthetic chemicals used as industrial solvents/lubricants, plasticizers, additives, agrochemicals, pharmaceutical agents, and polyphenols of plant origin. These compounds are even present in the food as components (polyphenols) or food/water contaminants (pesticides, plasticizers used as food packaging) rendering the diet as the main route of exposure to EDCs for humans. Although huge amount of literature reports the (anti)estrogenic effects of different EDCs, relatively scarce information is available on the (anti)androgenic effects of EDCs. Here, the effects and mechanism of action of phytochemicals and pesticides and plasticizers as possible modulators of AR activities will be reviewed taking into account that insight derived from principles of endocrinology are required to estimate EDC consequences on endocrine deregulation and disease.

Morandi S, Cremonesi P, Povolo M, Capra E, Silveti T, Castiglioni B, Ribeiro MG, Alves AC, da_Costa GM, Luini^o M, Brasca M

Prototheca blaschkeae subsp. brasiliensis subsp. nov., isolated from cow milk

Int J Syst Evol Microbiol. - Vol. 67 (2017). - p 3865-3871. - 21 bib ref [Nr. Estr. 7686]

A strain of an achlorophyllic alga, named PR24T, was isolated from cow milk samples from the state of Minas Gerais, Brazil. Based on 18S rDNA, 28S rRNA, D1/D2 region of the LSU rDNA and SSU rRNA gene sequence similarities, this strain was found to be a member of the genus *Prototheca* and closely related to *Prototheca blaschkeae* SAG2064T. However, the novel strain could easily be distinguished from recognized *Prototheca* species by internal transcribed spacer, species-specific PCR, single-strand conformation polymorphism-PCR analysis and phenotypic characteristics. The inability to grow in Sabouraud broth at pH 4.0 and the different cellular fatty acid composition clearly distinguished PR24T from the reference strain of *P. blaschkeae*. The combination of genotypic and phenotypic data indicates that strain PR24T represents a subspecies of *P. blaschkeae*, for which the name *Prototheca blaschkeae* subsp. *brasiliensis* subsp. nov. is proposed. The respective type strain is PR24T (=DSM 103592T=IHEM 26958T).

Naas HT, Almajdoubi Z, Garbaj AM, Azwai SM, Gammoudi FT, Abolghait SK, Moawad AA, Barbieri^o I, Eshamah HL, Eldaghayes IM

Molecular identification and antibiogram of Enterococcus spp. isolated on Enterococcus selective differential (ESD) media from meat, meat products and seafood in Libya

J Microbiol Biotechnol Food Science. - Vol. 6 no 6 (2017). - p 1264-1268. - 29 bib ref [Nr. Estr. 7703]

This study was conducted to investigate the presence of *Enterococcus* spp. in meat, meat products and seafood. A hundred and four samples were randomly collected from different geographic localities in Libya. The samples were subjected to microbiological analysis for enumeration and isolation of *Enterococcus* spp. by conventional cultural and molecular identification using PCR and partial sequencing of 16S rDNA techniques. Out of 104 samples, 73 (70.2%) isolates were found to be enterococci based on their cultural characteristics on ESD medium. However, out of 36 samples subjected to molecular identification, only six isolates were confirmed to be *Enterococcus* spp. using PCR and partial sequencing of 16S rDNA technique. All enterococci strains tested for their antibiotic sensitivity profiles showed high percentage of multi-resistance phenotype. These results can be used for further studies on enterococci as an emerging food borne pathogen and its role in human infection in Libya and would suggest that meat, meat products and seafood might play a role in the spreading of enterococci through the food chain with antimicrobial resistance characteristics.

Piva S, Gariano GR, Bonilauri^o P, Giacometti F, Decastelli L, Florio D, Massella E, Serraino A

Occurrence of putative virulence genes on Arcobacter butzleri isolated from three different environmental sites throughout the dairy chain

J Appl Microbiol. - Vol. 122 no (2017). - p 1071-1077. - 41 bib ref (ultimo accesso 05/07/2017 <http://onlinelibrary.wiley.com/wol1/doi/10.1111/jam.13403/abstract>) [Nr. Estr. 7578]

Aims: This comparative study investigated the occurrence of *cadF*, *cj1349*, *ciaB*, *pldA*, *tlyA*, *hecA*, *hecB*, *mviN*, *irgA* and *IroE* genes in 212 *Arcobacter butzleri* isolated from three different environmental sites linked to the dairy chain (farms, industrial and artisanal dairy plants) located in three Italian regions (Lombardy, Emilia-Romagna and Calabria). **Methods and Results:** According to the presence of these genes, different pathotypes (P-types) were determined. The main genes

detected were *ciaB*, *mviN*, *tlyA*, *cj1349*, *pldA* and *cadF*, while the least common genes were *iroE*, *hecA*, *hecB* and *irgA*. *TlyA*, *irgA*, *hecA*, *hecB* and *iroE*, which were significantly more frequent in isolates recovered in industrial dairy plants. Twelve P-types were detected. The occurrence of the most frequently detected P-types (P-types 1, 2, 3 and 5) differed significantly ($P < 0.001$) in relation to both the environmental site and geographical area of isolation. The highest diversity in P-types was observed in industrial dairy plants and in the Calabria region. Conclusions: The results of this study show a correlation between the occurrence of putative virulence genes and virulence genotype variability depending on the environmental site and geographical origin of the isolates. Significance and Impact of the Study: The present study provides insights into the similar distribution of putative virulence genes in a dairy chain and other sources' isolates and also into a geographical distribution of some P-types. We have shown that industrial dairy plants may represent an environmental site favouring a selection of the isolates with a higher pathogenetic pattern.

Ricchi° M, Bertasio° C, Boniotti° MB, Vicari° N, Russo° S, Tilola° M, Bellotti° MA, Bertasi° B

Comparison among the quantification of bacterial pathogens by qPCR, dPCR, and cultural methods

Front Microbiol. - Vol. 8 (2017). - Article 1174 (15 p). - 42 bib ref (ultimo accesso 08/09/2017 <http://journal.frontiersin.org/article/10.3389/fmicb.2017.01174/full>) [Nr. Estr. 7633]

The demand for rapid methods for the quantification of pathogens is increasing. Among these methods, those based on nucleic acids amplification (quantitative PCRs) are the most widespread worldwide. Together with the qPCR, a new approach named digital PCR (dPCR), has rapidly gained importance. The aim of our study was to compare the results obtained using two different dPCR systems and one qPCR in the quantification of three different bacterial pathogens: *Listeria monocytogenes*, *Francisella tularensis*, and *Mycobacterium avium* subsp. *paratuberculosis*. For this purpose, three pre-existing qPCRs were used, while the same primers and probes, as well as PCR conditions, were transferred to two different dPCR systems: the QX200 (Bio-Rad) and the Quant Studio 3D (Applied Biosystems). The limits of detection and limits of quantification for all pathogens, and all PCR approaches applied, were determined using genomic pure DNAs. The quantification of unknown decimal suspensions of the three bacteria obtained by the three different PCR approaches was compared through the Linear Regression and Bland and Altman analyses. Our results suggest that, both dPCRs are able to quantify the same amount of bacteria, while the comparison among dPCRs and qPCRs, showed both over and under-estimation of the bacteria present in the unknown suspensions. Our results showed qPCR over-estimated the amount of *M. avium* subsp. *paratuberculosis* and *F. tularensis* cells. On the contrary, qPCR, compared to QX200 dPCR, under-estimated the amount of *L. monocytogenes* cells. However, the maximum difference among PCR approaches was $<0.5 \text{ Log}_{10}$, while cultural methods underestimated the number of bacteria by one to two Log_{10} for *Francisella tularensis* and *Mycobacterium avium* subsp. *paratuberculosis*. On the other hand, cultural and PCR methods quantified the same amount of bacteria for *L. monocytogenes*, suggesting for this last pathogen, PCR approaches can be considered as a valid alternative to the cultural ones.

Robotti E, Campo F, Riviello M, Bobba° M, Manfredi M, Mazzucco E, Gosetti F, Calabrese G, Sangiorgi° E, Marengo E

Optimization of the extraction of the volatile fraction from honey samples by SPME-GC-MS, experimental design, and multivariate target functions

J Chem. - Vol. 2017). - Article ID 6437857 (14 p) - 56 bib ref (ultimo accesso 31/03/2017 <https://doi.org/10.1155/2017/6437857>) [Nr. Estr. 7527]

Head space (HS) solid phase microextraction (SPME) followed by gas chromatography with mass

spectrometry detection (GC-MS) is the most widespread technique to study the volatile profile of honey samples. In this paper, the experimental SPME conditions were optimized by a multivariate strategy. Both sensitivity and repeatability were optimized by experimental design techniques considering three factors: extraction temperature (from 50 C to 70 C), time of exposition of the fiber (from 20 min to 60 min), and amount of salt added (from 0 to 27.50%). Each experiment was evaluated by Principal Component Analysis (PCA) that allows to take into consideration all the analytes at the same time, preserving the information about their different characteristics. Optimal extraction conditions were identified independently for signal intensity (extraction temperature: 70 C; extraction time: 60 min; salt percentage: 27.50% w/w) and repeatability (extraction temperature: 50 C; extraction time: 60 min; salt percentage: 27.50% w/w) and a final global compromise (extraction temperature: 70 C; extraction time: 60 min; salt percentage: 27.50% w/w) was also reached. Considerations about the choice of the best internal standards were also drawn. The whole optimized procedure was then applied to the analysis of a multiflower honey sample and more than 100 compounds were identified.

Rubini° S, Barbieri S, Luppi° A, D'inciau° M, Melloni° R, Russotto° C, Govoni G, Guidi E, Bergamini M, Meriardi° G

Antimicrobial susceptibility of Salmonella typhimurium strains isolated from shellfish and seawater

10th European Public Health Conference : 1-4 November 2017 Stockolm, Sweden / [s.l. : s.n., 2017]. - 1 p (ultimo accesso 09/02/2018
<https://eph2017-eupha.ipostersessions.com/Default.aspx?s=gallery>) [Nr. Estr. 7784]

European Public Health Conference (10th : Stockolm, Sweden : 1-4 November 2017)

The Shellfish Monitoring plan, mandatory in European Union, in Emilia Romagna has been implemented since 1997. *S. enterica* subsp. *enterica* serovar Typhimurium is listed between the ten most common *Salmonella* serovars isolated from humans in the European countries. It has been isolated mainly from eggs, poultry meat, and porky meat. In the EU, over 100,000 human cases are reported each year In the study area (Figure 1), from 1997 to 2016, 11821 samples have been analyzed. ISO methods were applied. Serotyping was performed by using commercial antisera. Antibiotic-resistance has been carried out with the Kirby-Bauer method. The susceptibility of 655. Typhimurium strains to 17 antibiotics has been tested : amoxicillin + clavulanic acid (AMC), ampicillin (AMP), amikacin (AMK), cefotaxime (CTX), ceftiofur (EFT), cephalothin (CEF), chloramphenicol (C), colistin sulphate (CS), enrofloxacin (ENR), florfenicol (FFC), gentamicin (CN), kanamycin (K), nalidixic acid (NA), streptomycin (S), sulphonamide (SU), trimethoprim—sulfamethoxazole (SXT) and tetracycline (TE). Strains have been classified as susceptible, intermediate and resistant to antibiotics. Intermediate isolates were grouped with resistant isolates. RESULTS From 1997 to 2016 273 *Salmonella* strains were isolated. *S. Typhimurium* was the most frequently isolated serovar (83 strains, 2,31%) both in seawater and in molluscan shellfish. The 655. Typhimurium strains showed resistance to: AMC and S (30.8%), AMP and TE (29.2%), C (23.1%), FFC (21.5%), CEF (16.9%), SU (10.8%), CN and K (1.5%). No resistances were observed to the remaining antibiotics tested. . CONCLUSIONS Mollusks are not the most important source of *Salmonella* infection in humans but the isolation of *Salmonella* strains in these products increased in these last years, for this reason it should not be ignored. The widespread habit of eating raw or undercooked mollusks is a hazardous behavior that increases tremendously the public health concerns.

Rubini° S, Pigozzi S, Contiero L, Bille L, Bordin P, Menotta° S, Boschetti L, Cangini M, Milandri A, Orletti R, Piersanti A, Barile N, Latini M

Lipophilic marine biotoxins in commercial clams harvested along the western Adriatic Sea

11th International Conference on Molluscan Shellfish Safety : 14th -18th May, 2017 : Galway,

Ireland / [s.l. : s.n., 2017]. - 1 p. (ultimo accesso 09/02/2018
<http://programme.exordo.com/icmss2017/delegates/presentation/107/>) [Nr. Estr. 7787]

International Conference on Molluscan Shellfish Safety (11th : Galway, Ireland : 14th -18th May, 2017)

In the western Adriatic Sea there are a number of commercially exploited clam species, but the fishery for manila clams (*Ruditapes philippinarum*) and striped venus (*Chamelea gallina*) is by far the most important. In Italy both species are considered at low risk of contamination by marine biotoxins for the goals of the shellfish monitoring programme organized under Reg. (EC) No 854/2004, since historical data have never shown positivity for any of the regulated compounds. This is reflected in a reduced sampling frequency for both species. However, in May 2012 okadaic acids were detected for the first time in manila clams from the brackish lagoon of Sacca di Goro (Emilia Romagna). The episode was linked with high abundances of *Prorocentrum lima* and showed a maximum of 1154 pg OA eq./kg in summer 2013. The phenomenon became endemic and in 2016 shellfish farmers abandoned the areas where prolonged closures prevented a profitable economic exploitation of the resource. Following this unexpected event, lipophilic marine biotoxin results in clams have been collected to provide new evidences for a review of the current risk assessment. Data produced in the period 2013-2016 were gathered for the western Adriatic Sea. A total of 1535 samples have been analysed using LC-MS/MS. Manila clams accounted for 77% of the analysed shellfish. Only 4% of the samples contained toxin levels higher than the current EU regulatory limit (160 pg/kg of edible part). Most of them belonged to species *Ruditapes philippinarum* (97%) from transitional waters.

Sangiorgi^o E, Berneri^o R, Cotti_Piccinelli^o E, Rovellini P, Piro R, Miano B

Fighting the food frauds by means of lipid analysis : palm oil addition

8th International symposium on recent advances in food analysis : November 7-10, 2017, Prague, Czech Republic : book of abstracts / editors, Jana Pulkrabova ...[et al.]. - [Praha : University of Chemistry and Technology, 2017]. - p 81 [Nr. Estr. 7712]

International symposium on recent advances in food analysis (8th : Prague, Czech Republic : November 7-10, 2017)

Palm oil (PO) is the most widely consumed vegetable oil on the planet, and it is in most part of the packaged products sold in the supermarket. The demand for PO has increased steeply in last time as substitute of animal fat and as an alternative to hydrogenated vegetable oils. It is produced in tropical areas and its rapid expansion threatens some of the planet's most important and sensitive habitats. It has a high percent of saturated fatty acids that are not favorable to the health and recently the problems concerning food safety have evidenced the dangerous content of 3-MCPD in it (efsa.2016.4426). The consciousness of these facts has led the consumers to request food with no PO as ingredient. Starting from December 2014, all food contain palm oil sold in Europe must state it in the ingredient list (EU Reg 1169/2011). Today in Italian market many food items are now advertised as PO free but because of its low price and rheological and stability properties it is possible its use without a label declaration. To avoid and prevent this food fraud it is important to have reliable and easy analytical methods to verify this statement. This presentation investigates different methods to determine the presence of PO in food, choosing between classical analysis with chromatographic separations (GC and HPLC) and the fast Direct Analysis in Real Time approach (DART-MS). Different mixture of sunflower oil and rapeseed oil with PO were used to determine a LOD of PO addition. For GC, HPLC and DART-MS 21 samples were analyzed and for DART-HRMS 63 samples. For the determination of fatty acids and phytosterols, GC-FID was used; for the determination of tocopherols and tocotrienols HPLC with fluorescence detector was used; for TAGs, DAGs and MAGs an HPLC coupled with RI detector was used. Data were elaborated using Unscrambler and Metaboanalyst programs. Fatty acid composition is quite different from other vegetable oils and phytosterols content is much lower in PO; nevertheless, these two analysis were not useful for our target and statistical analysis of this data could scarcely differentiate products with PO from the others. The tocopherols and tocotrienols analysis allowed to distinguish the PO presence in the samples and could act as confirmatory analysis. The TAGs profile is characteristic

for PO so that it was possible recognize presence of this oil in false labelled as PO free food. With DART-ion trap in MS-MS mode (PPP as precursor ion) it was possible to detect PO presence. Using chemometric evaluation on DART-HRMS and the ratio PP0/000 data it was possible to define a threshold TAGs ratio for the presence of PO in the most of food products with more than 1% of PO. It was demonstrated that the use of an DART method allows to detect PO addition in food samples directly on sample with a minimum preparation. Fatty acids, fytosterols and DART-MS could be considered screening methods. Tocopherols and tocotrienols determination is a confirmatory analysis.

Sangiorgi° E, Folegatti L, Berneri° R, Cotti_Piccinelli° E

Fighting the food fraud : detecting vegetable oils in milk products

8th International symposium on recent advances in food analysis : November 7-10, 2017, Prague, Czech Republic : book of abstracts / editors, Jana Pulkrabova ...[et al.]. - [Praha : University of Chemistry and Technology, 2017]. - p 169 [Nr. Estr. 7713]

International symposium on recent advances in food analysis (8th : Prague, Czech Republic : November 7-10, 2017)

The adulteration of food is of a primary importance for all the actors involved in the food chain, from the producers to the consumers. The adulteration frequently involves the replacements of one or more expensive ingredients with cheaper substitutes. The growing interest in the food authenticity requires reliable and suitable verification methods to prevent deliberate or accidental mislabeling. One of the sectors more involved in food frauds is the cheese and milk product area where the milk fat, for economical or rheological reasons, could be partially substituted with cheaper vegetable oils. To detect vegetable oils added to milk fat sterols analysis, dehydroxylated sterols analysis, fatty acids profile and triacylglycerols (TAGs) analysis were used. 22 cheese and milk product samples were analyzed, either from retails or ethnic markets. GC-FID was used to determine sterols, dehydroxylated sterols and fatty acids profile. A DART source coupled with an ion trap detector was used to determine the lipid mass profile of the samples. The reference method ISO 12078:2006 allows to detect the presence of vegetable oils in milk fat through the analysis of phytosterols, mainly 13-sitosterol, campesterol and stigmasterol. Opposite, the milk fat has cholesterol as the main sterol (97-98%) with little amount of 17-cholesterol and 24-metylencholesterol only. Fatty acids analysis has to be accompanied with statistical analysis and could act as screening method. The lipid profile obtained with DART-MS analysis could highlight the long chain fatty acid TAGs of vegetable oils that are not present in milk fat. This analysis could be a screening method too. Among the samples analyzed, cheese and spreadable products, two of them revealed the presence of vegetable oils.

Scavia G, Alfonsi V, Taffon S, Escher M, Bruni R, De_Medici D, Di_Pasquale S, Guizzardi S, Cappelletti B, Iannazzo S, Losio° MN, Pavoni° E, Decastelli L, Ciccaglione AR, Equestre M, Tosti ME, Rizzo C and National Italian Task Force on Hepatitis A

A large prolonged outbreak of hepatitis A associated with consumption of frozen berries, Italy, 2013–14

J Med Microbiol. - Vol. 66 no 3 (2017). - p 342-348. - 35 bib ref [Nr. Estr. 7577]

Purpose. In 2013/2014, Italy experienced one of the largest community-wide prolonged outbreaks of hepatitis A virus (HAV) throughout the country. The article provides a comprehensive description of the outbreak and the investigation carried out by a multidisciplinary National Task Force, in collaboration with regional and local public health authorities. Control strategies of food-borne HAV infection in both the human and food sectors are also described. Methodology. Enhanced human epidemiological and microbiological surveillance together with microbiological monitoring of HAV in food and trace-back investigation were conducted. Results. A total of 1803 HAV cases were

identified from 1 January 2013 to 31 August 2014, in Italy. Sequencing was possible for 368 cases (20.4 %), mostly collected between 1 January 2013 and 28 February 2014, and 246 cases (66.8 %) harboured an HAV outbreak strain. Imported frozen berries contaminated with HAV were identified as the vehicle of the outbreak which also involved many other European countries in 2013 and 2014. Epidemiological evidence obtained through a case–control study was supported by the finding of a 100 % nucleotide similarity of the VP1/2A sequences of HAVs detected in human and food samples. Trace-back investigation revealed an extremely complex supplying network with no possibility for a point source potentially explaining the vast contamination of berries found in Italy. Conclusion. The investigation benefited from an excellent collaboration among different sectors who shared proactively the available information. Our findings highlight the importance of considering frozen berries among the highest risk factors for HAV.

Scavia G, Maugliani A, Chiani P, Michelacci V, Minelli F, Tozzoli R, Caprioli A, Bilei S, Bossù T, Marconi P, De_Santis L, Materassi M, Peruzzi L, Luini° MV, Morabito S

A severe transnational foodborne outbreak of infection by shigatoxin-producing E. coli O26 in a transnational community established in Italy, 2016

18th International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD), June 7-10, 2017 : Sorrento, Italy : abstract book / [s.l. : s.n., 2017]. - p 123, (Poster 052). - 7 bib ref [Nr. Estr. 7587]

International Symposium of the World Association of Veterinary Laboratory Diagnosticians (WAVLD) (18th : Sorrento : June 7-10, 2017)

Serraino A, Bonilauri° P, Giacometti F, Ricchi° M, Cammi° G, Piva S, Zambrini V, Canever A, Arrigoni° N

Investigation into Mycobacterium avium ssp. paratuberculosis in pasteurized milk in Italy

J Dairy Sci. - Vol. 100 no 1 (2017). - p 118-123. - 54 bib ref (ultimo accesso 06/06/2018 [https://www.journalofdairyscience.org/article/S0022-0302\(16\)30747-0/fulltext](https://www.journalofdairyscience.org/article/S0022-0302(16)30747-0/fulltext)) [Nr. Estr. 7499]

This study investigated the presence of viable Mycobacterium avium ssp. paratuberculosis (MAP) in pasteurized milk produced by Italian industrial dairy plants to verify the prediction of a previously performed risk assessment. The study analyzed 160 one-liter bottles of pasteurized milk from 2 dairy plants located in 2 different regions. Traditional cultural protocols were applied to 500 mL of pasteurized milk for each sample. The investigation focused also on the pasteurization parameters and data on the microbiological characteristics of raw milk (total bacterial count) and pasteurized milk (Enterobacteriaceae and Listeria monocytogenes). No sample was positive for MAP, the pasteurization parameters complied with European Union legislation, and the microbiological analysis of raw and pasteurized milk showed good microbiological quality. The results show that a 7-log (or >7) reduction could be a plausible value for commercial pasteurization. The combination of hygiene practices at farm level and commercial pasteurization yield very low or absent levels of MAP contamination in pasteurized milk, suggesting that pasteurized milk is not a significant source of human exposure to MAP in the dairies investigated.

Suffredini° E, Barbieri S, Rubini° S, Losardo° M, Pavoni° E, Bertasi° B, Bolognesi° E, Govoni G, Berardelli C, Boschetti L, Bergamini M, Meriardi° G

Virulence factors in Vibrio parahaemolyticus isolates from bivalve shellfish

10th European Public Health Conference : 1-4 November 2017 Stockholm, Sweden / [s.l. : s.n., 2017]. - 1 p (ultimo accesso <https://eph2017-eupha.ipostersessions.com/Default.aspx?s=gallery>) [Nr. Estr. 7785]

Tamba° M, Bolzoni° L, Casadei° G, Carra° E, Gualanduzzi C, Borrini BM, Bergamini° F, Degl'Innocenti° C, Pongolini° S

Il contributo dei laboratori alle indagini epidemiologiche sui casi di malattia trasmessa da alimenti, un "nuovo" strumento di prevenzione

XLI Congresso dell'Associazione Italiana di Epidemiologia "L'epidemiologia oggi : evidenze, comunicazione e partecipazione" : Mantova 25-27 Ottobre 2017 : abstract / [s.l. : s.n., 2017]. - p 245 [Nr. Estr. 8023]

Congresso dell'Associazione Italiana di Epidemiologia (41. : Mantova : Mantova 25-27 Ottobre 2017)

Introduzione: Le malattie trasmesse da alimenti (MTA) rappresentano un problema di sanità pubblica prioritario in Europa. La loro prevenzione richiede un approccio One Health poiché coinvolge diversi settori (sanità pubblica, ambientale, medicina veterinaria, sicurezza alimentare) non ancora totalmente integrati. Un contributo alla sorveglianza delle MTA può venire dal laboratorio: la tipizzazione genetica degli isolati da pazienti, animali, alimenti e ambiente può aiutare l'indagine epidemiologica, permettendo la conferma del collegamento tra paziente ed alimento o l'individuazione di cluster di pazienti che potrebbero non essere individuati attraverso le classiche procedure di indagine. Obiettivi: Sperimentare in campo l'uso della genotipizzazione di isolati da pazienti, alimenti e animali nell'indagine epidemiologica sui casi di MTA da Salmonella. Metodi: Nel settembre 2016 il Centro Enternet regionale (IZSLER PR) ha rilevato un cluster di 17 isolati di Salmonella enteritidis (SE) con il medesimo profilo PFGE provenienti da pazienti residenti in 2 province emiliane. Dal confronto con le notifiche di malattia infettiva pervenute in regione è stato possibile appurare che 11 casi erano stati classificati come sporadici, mentre 6 erano stati attribuiti ad un focolaio causato dal consumo di una torta farcita con crema a base di uovo crudo. La stessa SE è stata isolata dalle uova rimaste, permettendo l'individuazione dell'azienda di origine. Sui casi sporadici è stato chiesto un supplemento di indagine per raccogliere oltre alla storia alimentare recente, anche l'elenco degli esercizi commerciali abituali di acquisto degli alimenti. Contemporaneamente a stessa SE è stata isolata da galline di un allevamento del modenese, conferite a IZSLER dal veterinario aziendale. La positività del gruppo è stata confermata dai controlli ufficiali. È stato acquisito l'elenco dei clienti ed effettuato il richiamo delle uova. Sono quindi stati tipizzati 27 ceppi di SE isolati nello stesso allevamento dal 2006 al 2016. Tutti i ceppi sono stati poi sottoposti ad una ulteriore tipizzazione tramite MLVA. Risultati: Dei 17 isolati umani del cluster, 13 avevano il medesimo genotipo PFGE/MLVA, mentre 4 presentavano diverso profilo MLVA. Per tutti i 6 casi del focolaio e per 3 dei 7 casi sporadici con stesso profilo genetico è stato possibile evidenziare anche un collegamento epidemiologico con l'allevamento infetto. Il medesimo genotipo PFGE/MLVA dei pazienti è stato riscontrato negli animali, nelle uova e nei prelievi ambientali svolti nell'azienda infetta a partire dal 2009, suggerendo la persistenza del ceppo in azienda e la scarsa efficacia delle disinfezioni attuate. Conclusioni: Dopo la macellazione degli animali infetti, non sono stati individuati altri pazienti con SE col profilo PFGE/MLVA cluster. La genotipizzazione degli isolati da pazienti, animali e alimenti può aiutare ad individuare la fonte di contaminazione, contribuendo così alla prevenzione delle MTA.

Terio V, Bottaro M, Pavoni° E, Losio° MN, Serraino A, Giacometti F, Martella V, Mottola A, Di_Pinto A, Tantillo G

Occurrence of hepatitis A and E and norovirus GI and GII in ready-to-eat vegetables in Italy

Int J Food Microbiol. - Vol. 249 (2017). - p 61-65. - 64 bib ref [Nr. Estr. 7533]

Fresh vegetables and their ready-to-eat (RTE) salads have become increasingly recognized as potential vehicles for foodborne diseases. The EU Reg. 1441/2007 establishes microbiological criteria for bacterial pathogens for products placed on the market during their shelf-life (i.e. Salmonella spp., Listeria monocytogenes) for pre-cut fruits and vegetables (RTE) whilst it does not

address the problem of contamination by enteric viruses. In this study we investigated the contamination by hepatitis A virus (HAV), hepatitis E virus (HEV) and norovirus (NoV) in 911 ready-to-eat vegetable samples taken from products at retail in Apulia and in Lombardia. The vegetable samples were tested using validated real-time PCR (RT-qPCR) assays, ISO standardized virological methods and ISO culturing methods for bacteriological analysis. The total prevalence of HAV and HEV was 1.9% (18/911) and 0.6% (6/911), respectively. None of the samples analysed in this study was positive for NoV, *Salmonella* spp. or *Listeria monocytogenes*. The detection of HAV and HEV in RTE salads highlights a risk to consumers and the need to improve production hygiene. Appropriate implementation of hygiene procedures is required at all the steps of the RTE vegetable production chain and this should include monitoring of emerging viral pathogens.