PROMISE

"Protection of consumers by microbial risk mitigation through combating segregation of expertise"

P 14, VETFAK - Croatia

Team leader:

Prof. Estella Prukner-Radovcic, DVM, PhD, Dipl ECPVS

Faculty of Veterinary Medicine University of Zagreb

prukner@vef.hr

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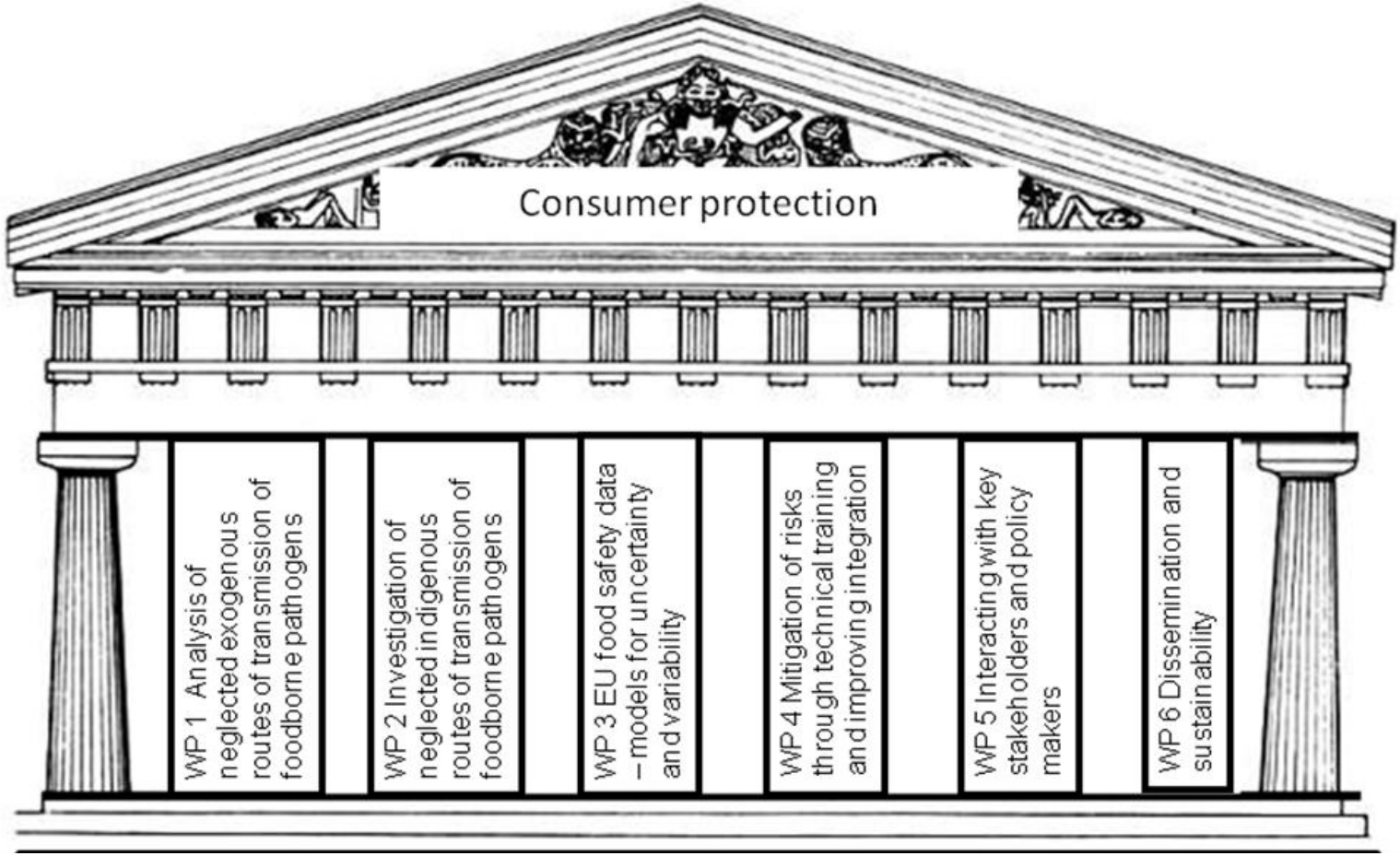

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PARTNERS

•	1 VETERINAERMEDIZINISCHE UNIVERSITAET WIEN VUW	Austria
•	2 BUNDESINSTITUT FUER RISIKOBEWERTUNG BFR	Germany
•	3 INSTITUTE OF FOOD RESEARCH IFR	United Kingdom
•	4 AGRICULTURAL UNIVERSITY OF ATHENS AUA	Greece
•	5 AGRICULTURE AND FOOD DEVELOPMENT AUTHORITY TEAGASC	Ireland
•	6 UNIVERSIDAD DE BURGOS UoB	Spain
•	7 VYZKUMNY USTAV VETERINARNIHO LEKARSTVI VRIB	Czech Republic
•	8 UNIVERZA V LJUBLJANI UL	Slovenia
•	9 MAGYAR TUDOMANYOS AKADEMIA ALLATORVOS-VMRI	Hungary
•	10 VYSKUMNY USTAV POTRAVINARSKY VUP	Slovakia
•	11 UNIVERSITATEA DUNAREA DE JOS DIN GALATI UDJG	Romania
•	12 KALITE SISTEM LABORATUVARLARI AS KSL	Turkey
•	13 DI ANDREAS MOSER RTD SERVICES -RTDS RTDS	Austria
•	**14 UNIVERSITY OF ZAGREB-Faculty of Veterinary Medicine VETFAC**	**Croatia**
•	15 OSTERREICHISCHE AGENTUR FUR GESUNDHEIT UND	
•	ERNAHRUNGSSICHERHEIT GMBH AGES	Austria
•	16 FOOD SAFETY AUTHORITY OF IRELAND FSAI	Ireland
•	17 ENIEOS FOREAS ELEGHOU TROFUNON HFSA	Greece
•	18 GIDA GUVENLIGI DERNEGI TFSA	Turkey
•	19 AUTORITATEA NATIONALA SANITARA VETERINARA SI PENTRU	
•	SIGURANTA ALIMENTELOR NSF	Romania
•	20 URAD VEREJNEHO ZDRAVOTNICTVA SLOVENSKEJ REPUBLIKY-SK	Slovakia

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Croatian team (P 14)

Faculty of Veterinary Medicine University of Zagreb

– *Department of Poultry Diseases with Clinic*

- Team leader:

Prof. Estella Prukner-Radovčić, DVM, PhD, Dipl.ECPVS

Post-doc: Danijela Horvatek, DVM, PhD

PhD student: Maja Lukač, DVM

– *Department of Food Hygiene*

Prof. Lidija Kozačinski, DVM, PhD

PhD student: Sandra Gutić, DVM

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Croatian part in PROMISE

- WP 1 - Analysis of neglected exogenous routes of transmission of foodborne pathogens:
  - Task leader 1.1 – Standardisation of sampling and methodology
  - Study of illegal food imports: border stations - 100 samples – Salmonella
- WP 5 - Interaction with key stakeholders and policy makers
- WP 6 - Dissemination and sustainability

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Laboratories

VETFAK

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WP 1.
Analysis of neglected EXOGENOUS routes of transmission of foodborne pathogens

- **Task 1.1**

STANDARDISATION of SAMPLING and METHODOLOGY of CULTURE AND IDFENTIFICATION OF FOODBORNE PATHOGENS

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COOPERATION

Table 1. List of participants

Participant No.	Organisation name	Abbreviation	Country
1	University for Veterinary Medicine	VUW	Austria
2	Federal Institute for Risk Assessment	BFR	Germany
4	Agricultural University of Athens	AUA	Greece
6	University of Burgos	UoB	Spain
7	Veterinary Research Institute	VRIB	Czech Rep
8	University of Ljubljana	UL	Slovenia
9	Veterinary Medical Research Institute, Hungarian Academy of Sciences (VMRI-MTA)	VMRI	Hungary
11	University Dunarea de Jos Galati	UDJG	Romania
12	Kalite Sistem Lab oratuarlar Grubu	KSL	Turkey
14	Faculty of Veterinary Medicine University of Zagreb	VETFAC	Croatia

Samples

- **Sample** is a **25 g of material x 4** (for 4 pathogens and commensals) of raw or ready-to-eat confiscated food of animal origin, taken by the border stations by veterinary and food control officials, and kept for PROMISE partner laboratories at **4°C for ≥24 hours**.

- **Categories of samples** are:
  - Milk and all kinds of milk products including milk powder and baby-food with milk;
  - Eggs and egg products, including egg powders;
  - Meat and raw meat products (i.e. sausages, hams), and fishes;
  - In case of availability: hunted wild animals, or “bush meat” also to be sampled.

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Sampling plan divided by the PROMISE Consortium

Partner	BFR	VMRI	VRI	VUW	VFUZ	UL	AUA	UDJG	UoB	KSL*
Country/Partner	GE/P2	HU/P9	CZ/P7	AT/P1	**CR/P14**	SL/P8	GR/P4	RO/P11	SP/P3	TU/12
No of border	1	6	NA	1	**2**	2	1	1	1	NA
Type of border	Air-port	Ground border	Ground -fecal sample	Air-port	**Sea-port, Ground border, Airport**	Ground border and Airport	Sea-port, Airport, Ground border	Ground border	Airport	NA
No of samples	**1000**	**200**	**100**	**500**	**100**	**100**	**200**	**100**	**100**	**100**
Salmonella	y	y	n	y	**y**	y	y	y	y	?
Listeria	y	n	n	y	**n**	n	y	n	y	Y
VTEC	y	y	n	y	**n**	y	y	y	y	?
Campylobacter	y	y	n	y	**n**	y	n	y	y	?
E. coli MDR	n	y	n	y	**n**	n	n	n	y	?
Others	-	-	-	Staph	-	Staph	Staph	n		?
Bacterial DNA	+	+	+++	+	+	+	+	+	+	?

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ANALYTICAL METHODS

- All methods that are used for the subsequent investigations are harmonized between participants.
- For the sampling methods, and bacterial culture techniques, the appropriate **ISO methods** are chosen.
- The use of ISO methods will allow a uniform basis for isolation and identification of the respective foodborne pathogens and thus, it will allow comparison of the results.
- Regarding **standardization** of all testing capabilities and to obtain the relevant data about the ISO methods already used or recently adopted in participant's laboratories, instead of on training workshop, a **questionnaire** was send by a Task leader to all involved parties.

- The partners were asked to specify the methods used for the detection of different pathogens and to describe the differences if some other methods are used, then ISO. All involved partner has decided to use ISO methods as originally accepted at the kick-off meeting in Vienna (30-31st January 2012).

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Microorganism	Method to be used	Comments
Salmonella spp.	Microbiology of food and animal feeding stuffs — Horizontal method for the detection of Salmonella spp. (ISO 6579:2002)	
Listeria spp.	Microbiology of food and animal feeding stuffs -- Horizontal method for the detection and enumeration of *Listeria monocytogenes* -- Part 1: Detection method (ISO 11290-1:1996) Microbiology of food and animal feeding stuffs -- Horizontal method for the detection and enumeration of *Listeria monocytogenes* -- Part 2: Enumeration method (ISO 11290-2:1998)	
Verotoxigenic *E. coli* (VTEC)	VTEC O157 Microbiology of food and animal feeding stuffs -- Horizontal method for the detection of Escherichia coli O157 (ISO 16654:2001)	VTEC non-O157 (Optional: until the new ISO for O157+non-O157 will be published, soon*) 1) Keep (-80C with 20% gycerol,) 2 x 2ml of enrichment cultures of O157-tests (ISO16654:2001) Further alternatives: 2) DIN 10118 (ASU §64 LFGB, L00.00-92) or ASU §64 LFGB, L07.18-1 3) method by CRL-VTEC (Rome, Italy) ---------- * Under development ISO WD/TS13136 (detection of O157, O26, O145, O111, O103)
Campylobacter spp.	Microbiology of food and animal feeding stuffs -- Horizontal method for detection and enumeration of Campylobacter spp. – Part 1: Detection method (ISO 10272-1:2006)	PCR analysis using primers by Wang et al. (2002), Linton et al. (2007), Zorman and Smole Možina (2002) for C. jejuni and C. coli and Chaban et al. (2009) for C. lari
Multidrug resistant (MDR) *E. coli*	non-selective TSB culture for all E.coli + indicator agar	
Other bacteria	Microbiology of food and animal feeding stuffs -- Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species) -- Part 1: Technique using Baird-Parker agar medium (ISO 6888-1:1999) Microbiology of food and animal feeding stuffs -- Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species) -- Part 2: Technique using rabbit plasma fibrinogen agar medium (ISO 6888-2:1999) VMRI method for *Pseudomonas aeruginosa*: Z-Broth (Szita et al. 2007) and HiFluoro agar (Sigma)	

Testing methods for antimicrobial resistance (AMR)

The antimicrobials to test for *Salmonella* and *Campylobacter* have already been agreed as follows:

- ***a) Salmonella*** will be isolated from meat and milk products and samples from about:
  200 live or dead illegally imported animals, if available at borders (DIN ISO 6579). Isolates of *S.* Typhimurium will be typed as developed by Partner VRIB. ABR profiles will include resistance to the antibiotics COL, TAZ, SMX, AMP, FFN, TET, GEN, STR, TMP, CHL, KAN, FOT, CIP and NAL.

- ***b) Campylobacter*** will be tested for resistance to the antibiotics CIP, ERY, GEN, NAL, STR, TET and CHL. Testing methods for antimicrobial resistance (AMR), in specific details on *Salmonella* and *Campylobacter* will be finally harmonized with respect to the selection of antimicrobials and the methods to be used (ETest, microdilution, disk diffusion) according to the criteria of the Clinical Laboratory Standards Institute (CLSI), with respect to the partner's preliminary experiences of the first few isolates.

Sampling GENERAL INFORMATIONS- VETFAK

- Period from November 2012 – June 2013
- Total of 100 samples obtained and examined
- Mostly for personal use ???
- Only 8 samples for market (China products)
- Places of confiscation:
  - Sea port Rijeka
  - Four borders crossings between Croatia and Bosnia and Herzegovina (The longest border in Croatia)
  - One border crossing between Croatia and Serbia
  - One border crossing between Croatia and Montenegro

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PLACES OF ORIGIN AND FINAL DESTINATIONS

Bosnia and Herzegovina

- Austria
- Croatia
- Germany
- Slovenia
- Italy

Serbia

- Croatia
- Slovenia

Macedonia

- Croatia
- Slovenia
- Switzerland
- Germany

China

- Hungary

Albania

- EU

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NUMBER OF SAMPLES FROM EACH COUNTRY

- **Places of origin in total:**
  - **BOSNIA (69)**
  - **SERBIA (5)**
  - **MACEDONIA (7)**
  - **ALBANIA (1)**
  - **CHINA (8)**
  - **UNKNOWN (10)**

Personal use

Market

- Final destinations in total:
  - ✓ CROATIA (43)
  - ✓ SLOVENIA (5)
  - ✓ AUSTRIA (22)
  - ✓ GERMANY (16)
  - ✓ ITALY (2)
  - ✓ SWITZERLAND (1)
  - ✓ HUNGARY (8)
  - ✓ UNKNOWN (3)

Personal use

Market

CATEGORY OF THE PRODUCTS

Category of the product	Number in total
Dairy products	**15**
Fresh or fresh frozen meat	**30**
Dry meat	**48**
Eggs	**3**
Fish in the can	**2**
Dehidrated noodles with meat	**2**

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COOPERATION

RESULTS

Type of bacteria isolated	Number of bacteria isolated	Sample	Place of origin	Final destination
Salmonella Enteritidis	**1**	**Fresh beef meat**	**Bosnia and Herzegovina**	**Croatia**
Listeria ivanovi	**1**	**Dry pork sausages**	**Bosnia and Herzegovina**	**Germany**
Listeria grayi	**1**	**Pork product „Svargla“**	**Bosnia and Herzegovina**	**Austria**
MDR *E.coli*	**4**	**Fresh beef meat**	**Bosnia and Herzegovina**	**Croatia, Austria**
	1	**Frozen chicken meat**		
	1	**Fresh chicken meat**		

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Expectations

- While legal imports are well monitored for contamination and alerts are registered through the Rapid Alert System for Food and Feed (RASFF; http://www.efet.gr/docs/rasff/report2008_en.pdf) notification systems, ”gates” into the EU-27 could exist where food supply chains are not controllled.

  These **uncontrolled imports** present the risk that new strains of traditional pathogens will be transferred from third countries into the European Union.

- To improve the **cooperation** between the experts from **human medicine, veterinary medicine and food safety**, to effectively monitor the incidence of diseases in animals, food and people

- Participation in experts groups will strengthen **collaboration between the countries** (members of this project) and further enhance the possibility to be involved in new “FP8” projects.
- **Young scientists** involved in this project will have the opportunity to work in foreign scientific institutions and acquire experience and skills which can be applied in their own laboratory

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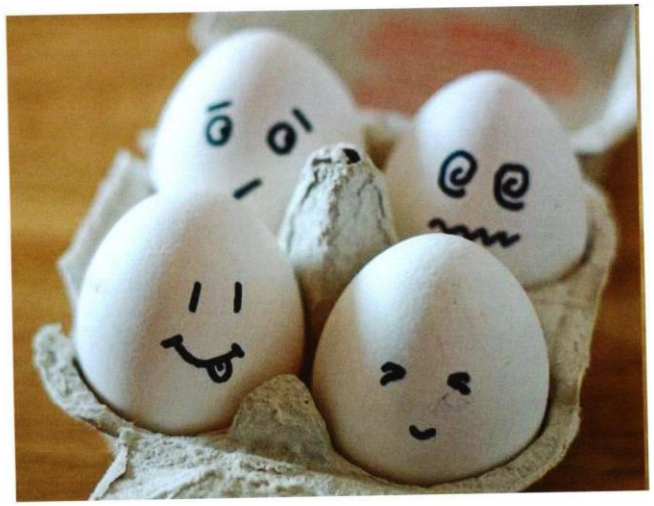
Hvala na pažnji!



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