

MICROBIAL RESISTANCE – actual situation in the Slovak Republic

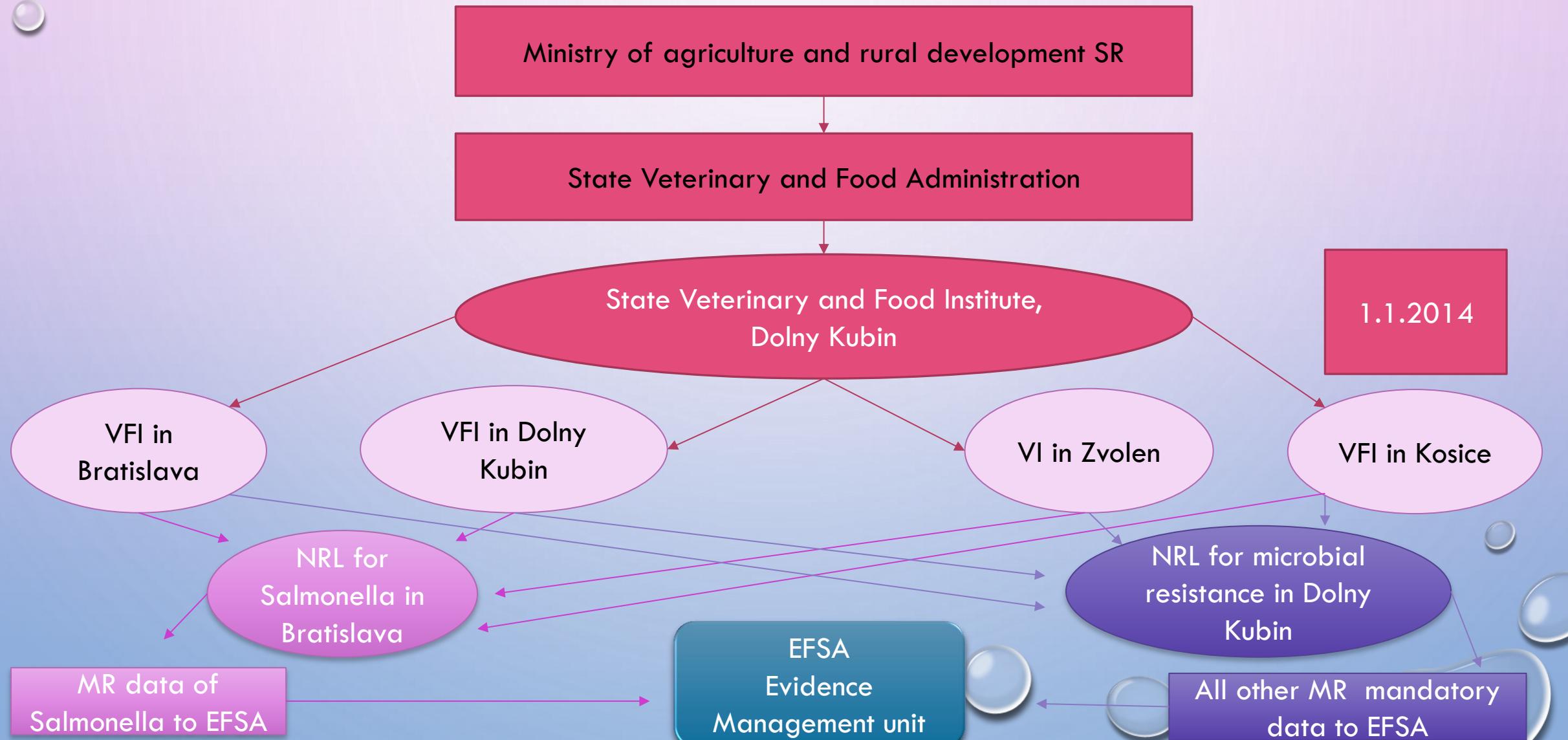
National reference laboratory for microbial resistance

Veterinary and Food Institute in Dolny Kubin

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MICROBIAL RESISTANCE - actual situation in the SR

- The organizational structure – MR data collection - data transmission to EFSA



MICROBIAL RESISTANCE - actual situation in the SR

- Microbial resistance in Salmonella isolates:
- Active monitoring: 4 monitoring programs ongoing in SR:
 - National monitoring program of *Salmonella* infection in breeding flocks of *Gallus Gallus*
 - National monitoring program of *Salmonella* infection in laying hens of *Gallus Gallus*
 - National monitoring program of *Salmonella* infection in broilers of *Gallus Gallus*
 - National monitoring program of *Salmonella* infection in turkey
- Passive monitoring: clinical samples from animals (faeces, rectal/cloacal swabs, carcasses, food and feed (data not included)



MICROBIAL RESISTANCE - actual situation in the SR

- the highest level of microbial resistance in *Salmonella* isolates – poultry flocks and poultry meat – the focus of this presentation
- In 2014: 100 positive samples for *Salmonella* spp.
 - ✓ in the flocks - 23 isolates from broilers before slaughter
 - 5 isolates from one-day old broilers
 - 1 isolate from fattening turkey
 - ✓ at the slaughterhouse - 58 isolates from broilers carcass
 - 1 isolate from turkey carcass
 - ✓ food from retail - 11 isolates from poultry meat
 - 1 isolate from turkey meat

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Salmonella* spp. from poultry and poultry meat – qualitative data

Evaluation according to serovar:

S. Enteritidis: 25 positive – 12x fully sensitive

Cp – R: 6x; ACp – R: 1x; Nx Cp – R: 4x;
Nx Cp Ct – R: 2x

S. Infantis: 59 positive – 0x sensitive

SuTetNxCp – R: 46 x; ASuTetNxCp – R: 11x;
SuTetNxCpTg – R: 2x

S. Newport: 2 positive – 0x sensitive

NxCp – R: 1x; SuTetNxCp – R: 1x.

S. Kentucky: 1 positive – 0x sensitive

TetNxCpCol – R: 1x

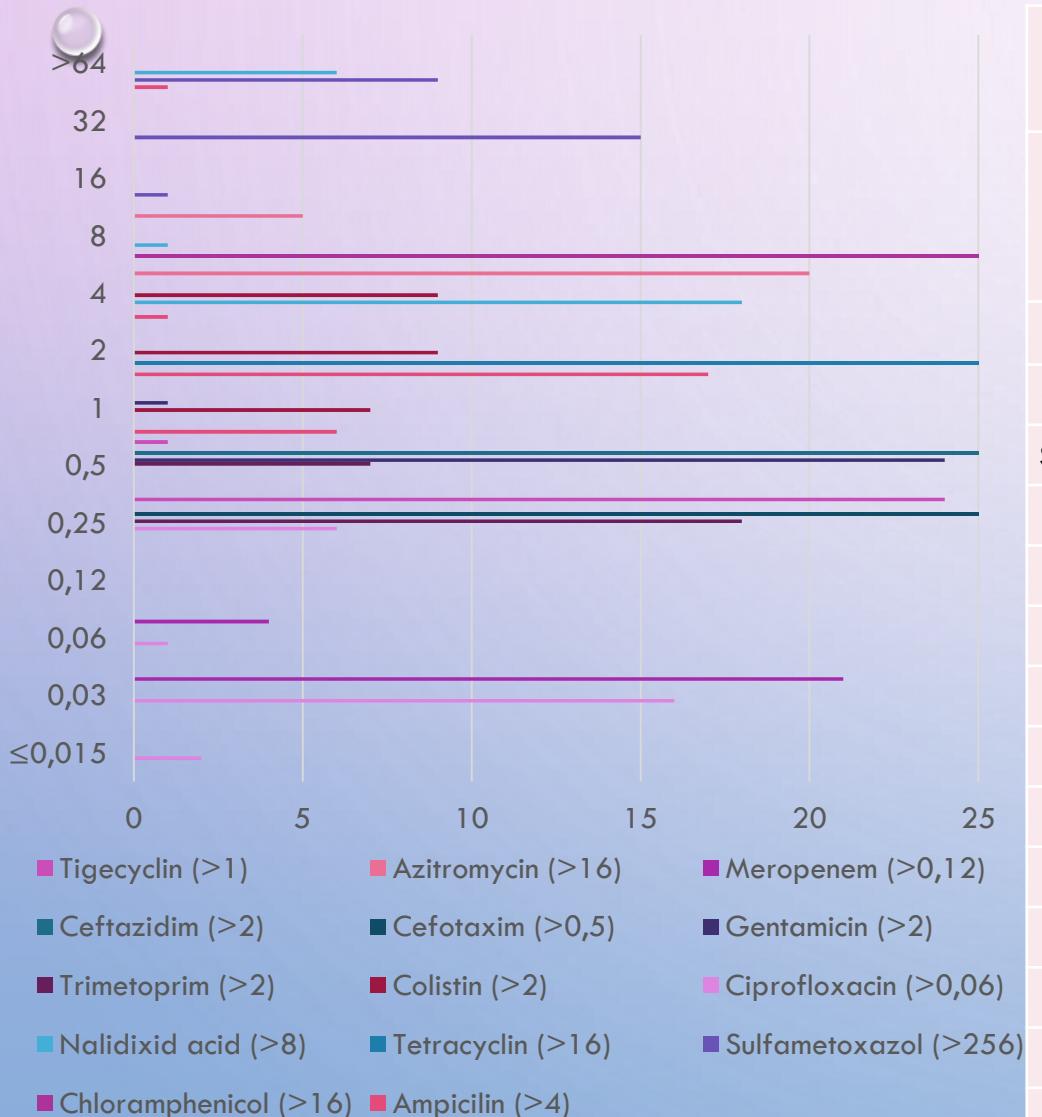
Other serovar (*S. Typhimurium*, *S. Indiana*, *S. Mbandaka*, *S. Lille*) – fully sensitive

61 MDR isolates, 0 5-DR isolates

<i>Salmonella</i> spp.		Broilers, turkeys, poultry meat									
Serotype		<i>Enteritidis</i>		<i>Infantis</i>		<i>Newport</i>		Kentucky		Other serotype	
No. of tested isolates	25		59			2		1		13	
Antimicrobials	n	R%	n	R%	n	R%	n	R%	n	R%	
Ampicilin (A)	1	4	11	18,6	0	0	0	0	0	0	
Chloramphenicol (Chl)	0	0	0	0	0	0	0	0	0	0	
Sulfametoxazol (Su)	0	0	59	100	1	50	0	0	0	0	
Tetracyclin (Tet)	0	0	59	100	1	50	1	100	0	0	
Nalidixid acid (Nx)	6	24	59	100	2	100	1	100	0	0	
Ciprofloxacin (Cp)	6	24	59	100	2	100	1	100	0	0	
Colistin (Col)	9	36	0	0	0	0	1	100	0	0	
Trimetoprim (W)	0	0	0	0	0	0	0	0	0	0	
Gentamicin (Gen)	0	0	0	0	0	0	0	0	0	0	
Cefotaxim (Ctx)	0	0	0	0	0	0	0	0	0	0	
Ceftazidim (Caz)	0	0	0	0	0	0	0	0	0	0	
Meropenem (Mero)	0	0	0	0	0	0	0	0	0	0	
Azitromycin (Azi)	0	0	0	0	0	0	0	0	0	0	
Tigecyclin (Tg)	0	0	2	3,4	0	0	0	0	0	0	

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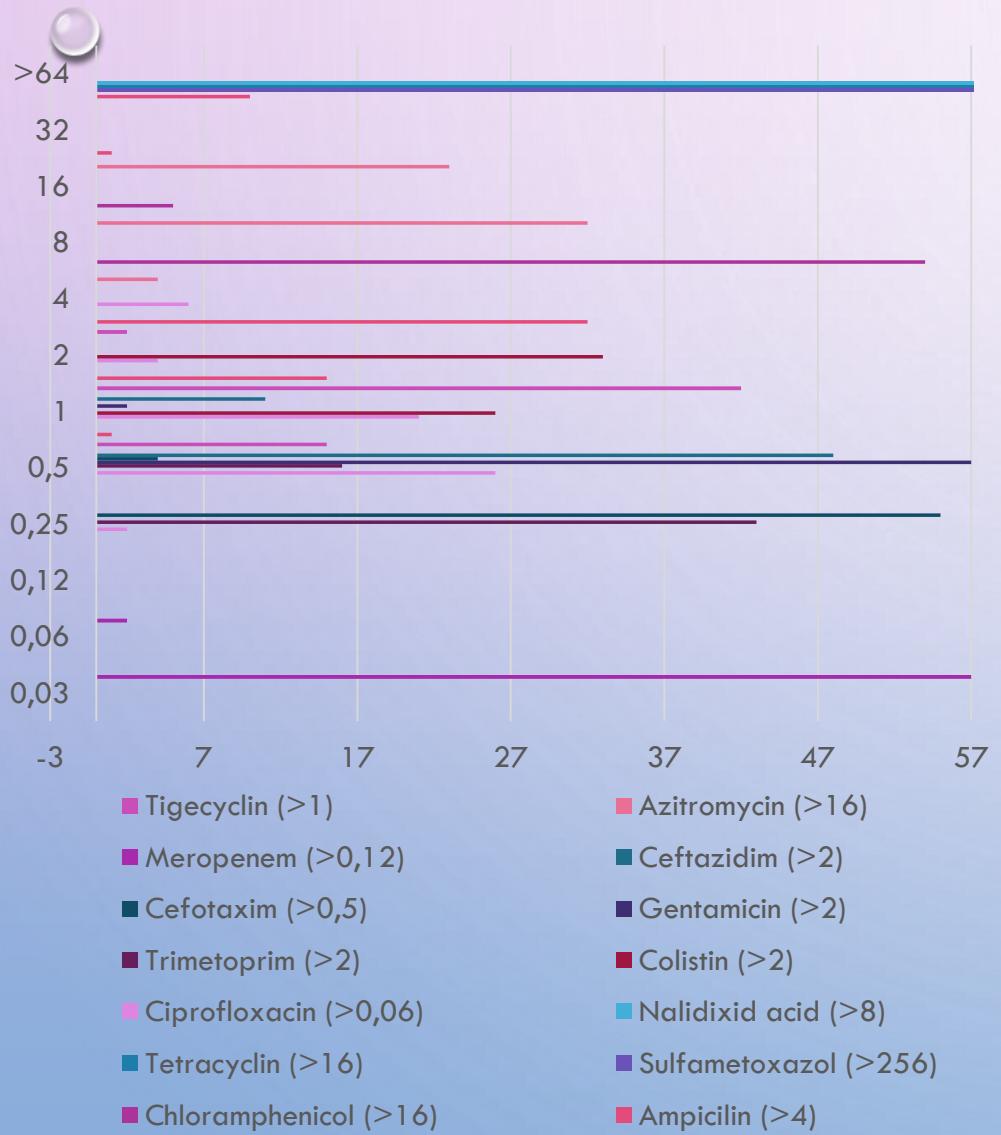
Profile of *Salmonella Enteritidis* from poultry and poultry meat – quantitative data (25 isolates)



S. Enteritidis	Poultry and poultry meat												
	Minimal inhibition concentration of antimicrobial (µg/ml)												
Antimicrobials	≤0,015	0,03	0,06	0,12	0,25	0,5	1	2	4	8	16	32	>256
Ampicillin (>8)							6	17	1				1
Chloramphenicol (>16)												25	
Sulfametoxyzol (>256)											1	24	
Tetracyclin (>8)									25				
Nalidixid acid (>8)										18	1		6
Ciprofloxacin (>0,06)	2	16	1				6						
Colistin (>2)								7	9	9			
Trimetoprim (>2)							18	7					
Gentamicin (>2)							24	1					
Cefotaxim (>0,5)							25						
Ceftazidim (>2)							25						
Meropenem (>0,12)	21	4											
Aztreonam (NA)										20	5		
Tigecycline (>1)							24	1					

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Salmonella Infantis* from poultry and poultry meat – quantitative data (59 isolates)



S. Infantis	Poultry and poultry meat												
	Minimal inhibition concentration of antimicrobial ($\mu\text{g}/\text{ml}$)												
Antimicrobials	$\leq 0,015$	0,03	0,06	0,12	0,25	0,5	1	2	4	8	16	32	>64
Ampicilin (>4)							1	15	32		1	10	
Chloramphenicol (>16)										54	5		
Sulfametoxazol (>256)												59	
Tetracyclin (>16)												59	
Nalidixid acid (>8)												59	
Ciprofloxacin (>0,06)		2	26	21	4	6							
Colistin (>2)							26	33					
Trimetoprim (>2)		43	16										
Gentamicin (>2)							57	2					
Cefotaxim (>0,5)							55	4					
Ceftazidim (>2)							48	11					
Meropenem (>0,12)	57	2											
Azitromycin (>16)									4	32	23		
Tigecyclin (>1)							15	42	2				

MICROBIAL RESISTANCE - actual situation in the SR

Conclusion – prevalence of resistance among *Salmonella* isolates in 2014

- CHL, W, GEN - zero resistant
- CAZ, CTX - zero resistance to Cephalosporine 3rd generation — **no ESBL/AmpC *Salmonella* producer findings**
- MERO – zero resistance – **no carbapenemase *Salmonella* producer findings**
- TET, SUL, AMP – moderate – extreme high resistance
- CIP, NAX – extreme high resistance (100% R *S. Infantis*, *S. Infantis* is responsible for most chinolons and fluorochinolons resistance in *Salmonella*)
- COL – low – moderate level of resistance. *S. Enteritidis* alone is responsible for most colistin resistance in *Salmonella* from broilers and layers
- New tested antimicrobials:
- TGC: tetracycline derivate antibiotic – developed in response to the growing rate of MR in bacteria e.g. STA, *Acinetobacter*, *E. coli* - for treating complicated infection. Low resistance – *S. Infantis* the main serovar R to TGC, serovar association
- AZI: macrolide to use in the treatment of invasive *Salmonella* and *Shigella* infection, no one *Salmonella* R isolate
- MDR: 61 isolates showed MDR, no penta resistant pattern observe

CIP, NAX, SSS, TET and follow by the same pattern with AMP, the most observe patterns

MICROBIAL RESISTANCE - actual situation in the SR

- Microbial resistance in *Campylobacter* isolates:
- Active monitoring: According to Desicion 2013/652/EU - Harmonised monitoring and reporting of AMR in zoonotic (including *Campylobacter*) and commensal bacteria
- Internal study for setting prevalence and MR of *Campylobacter jejuni* and *C. coli* from pigs at the slaughter
- Passive monitoring: clinical samples from animals (faeces, rectal/cloacal swabs, carcasses) - data not included
- the highest level of microbial resistance in *Campylobacter* isolates – caecal samples gathered at slaughter from broilers and pigs – the focus of this presentation
- 74 positive samples for *Campylobacter* spp.
 - ✓ 11 *C. jejuni* isolates from broilers; 3 *C. jejuni* isolates from pigs
 - ✓ 36 *C. coli* isolates from broilers; 17 *C. coli* isolates from pigs
 - ✓ 7 isolates other species than *C. jejuni* and *C. coli* (missing interpretation criterium) from broilers

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Campylobacter* spp. in broilers and pigs caecum - qualitative data

Evaluation according to animal and bacterial species:

Broilers vs. Pig – *C. jejuni*:

- broilers: 11 positive – 3x fully sensitive

CpNx – R: 3x; CpNxTet – R: 2x; CpNxEry – R: 2x;

CpNxStrTet – R: 1x

- pigs: 3 positive – zero fully sensitive

CpNxTetStr – R: 2x; CpNxEryTet – R: 1x

Broilers vs. Pig – *C. coli*:

- broilers: 36 positive – 3x fully sensitive

CpNx – R: 16x; CpNxTet – R: 10x; CpNxStrTet – R: 5x

- pigs: 17 positive – zero fully sensitive

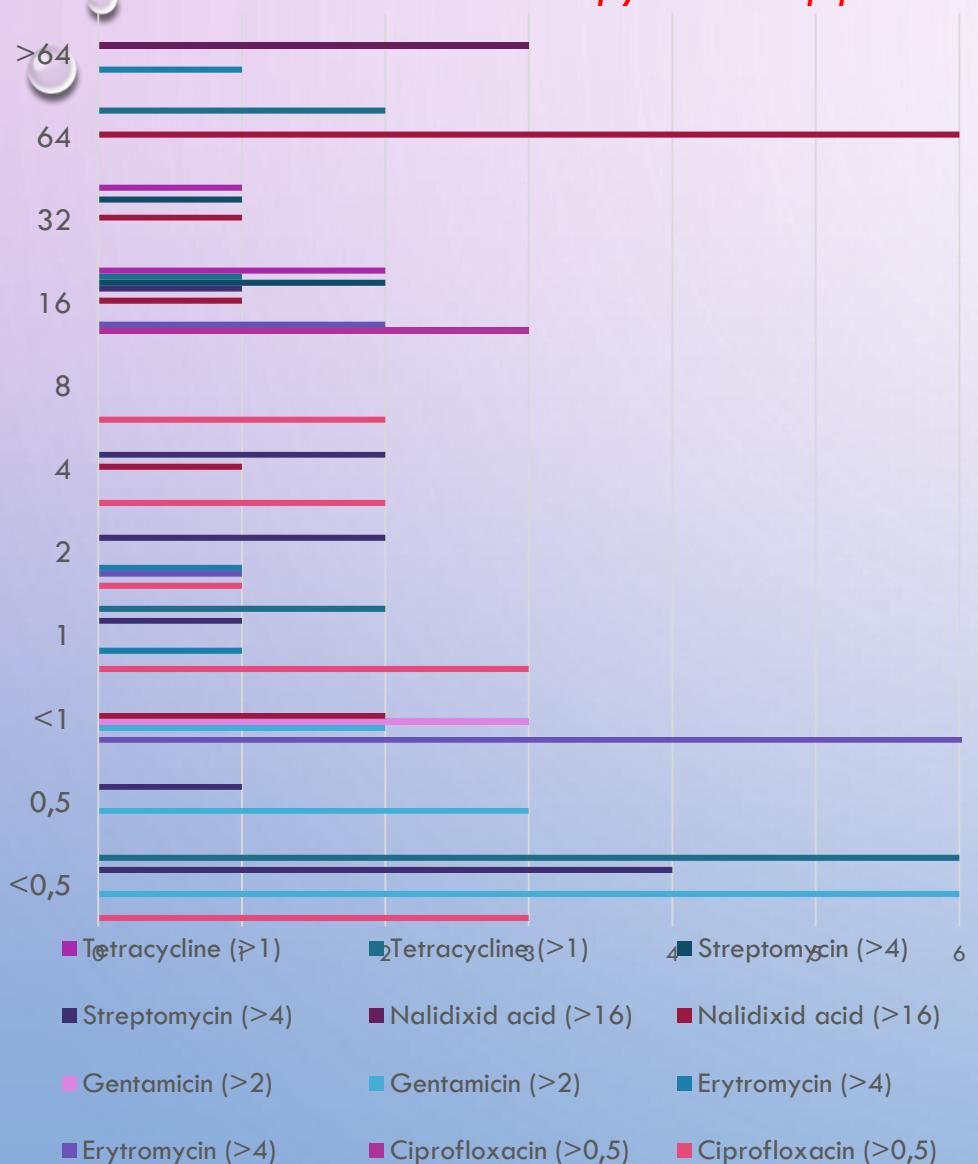
CpNxTetStr – R: 9x; CpNxTetStrEry – R: 3x

7 MDR vs. 16 MDR isolates, 0 5-DR isolates

<i>Campylobacter</i> spp.		Broilers caecum			
Species	<i>C. Jejuni</i>			<i>C. Coli</i>	
No.of tested isolates	n	R%		n	R%
Ciprofloxacin (>1)	7	64	Ciprofloxacin (>1)	33	92
Erytromycin (>8)	2	18	Erytromycin (>16)	1	3
Gentamicin (>4)	0	0	Gentamicin (>4)	0	0
Nalidixid acid (>32)	8	73	Nalidixid acid (>32)	33	92
Streptomycin (>8)	1	9	Streptomycin (>8)	7	19
Tetracycline (>2)	3	27	Tetracycline (>4)	16	44
<i>Campylobacter</i> spp.		Pig caecum			
Species	<i>C. Jejuni</i>			<i>C. Coli</i>	
No.of tested isolates	n	R%		n	R%
Ciprofloxacin (>1)	3	100	Ciprofloxacin (>1)	14	82
Erytromycin (>8)	1	33	Erytromycin (>16)	4	24
Gentamicin (>4)	0	0	Gentamicin (>4)	0	0
Nalidixid acid (>32)	3	100	Nalidixid acid (>32)	16	94
Streptomycin (>8)	3	100	Streptomycin (>8)	16	94
Tetracycline (>2)	3	100	Tetracycline (>4)	14	82

MICROBIAL RESISTANCE - actual situation in the SR

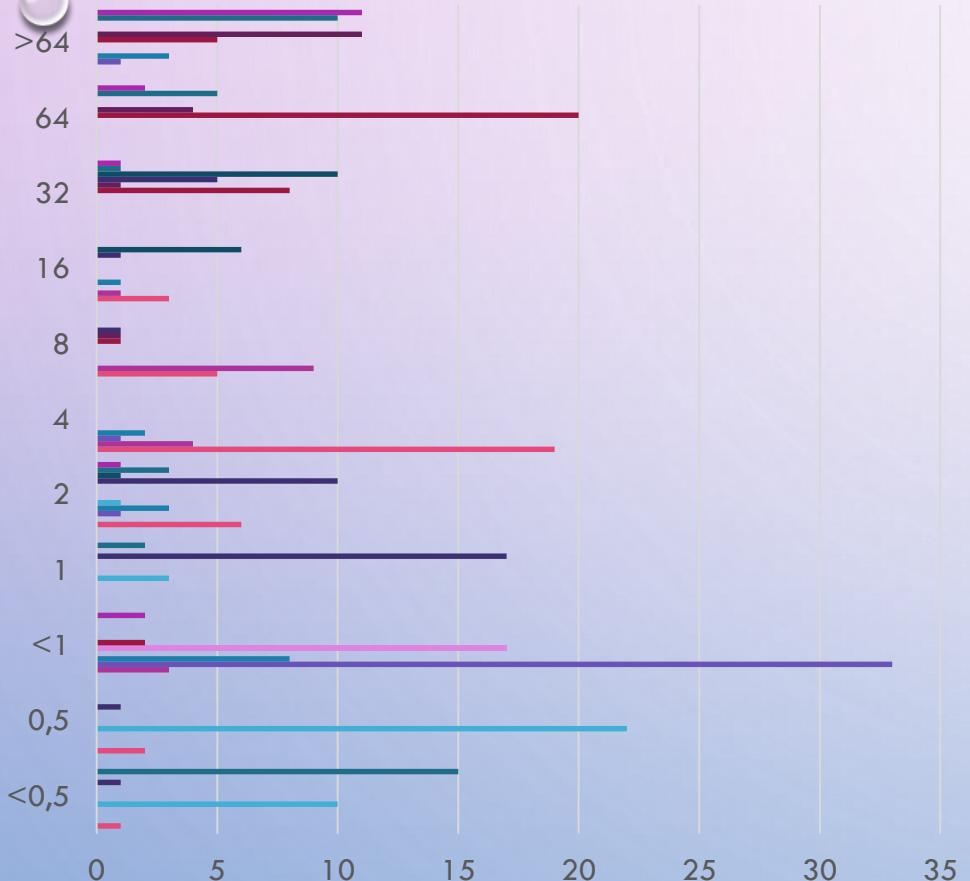
Profile of *Campylobacter jejuni* in broilers and pigs caecum - quantitative data (11/3 isolates)



C. jejuni	Broilers and pigs caecum									
	Minimal inhibition concentration of antimicrobial ($\mu\text{g/ml}$)									
Antimicrobials	<0,5	0,5	1	2	4	8	16	32	64	>64
Ciprofloxacin ($>0,5$)	broilers	3			3	1	2	2		3
	pigs								3	
Erytromycin (>4)	broilers		8		1			2		
	pigs			1	1					1
Gentamicin (>2)	broilers	6	3	2						
	pigs			3						
Nalidixic acid (>16)	broilers			2		1		1	1	6
	pigs									3
Streptomycin (>4)	broilers	4	1		1	2	2		1	
	pigs								2	1
Tetracycline (>1)	broilers	6			2			1		2
	pigs							2	1	

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Campylobacter coli* in broilers and pigs caecum - quantitative data (36/17 isolates)



C. coli		Broilers and pigs caecum										
		Minimal inhibition concentration of antimicrobial ($\mu\text{g}/\text{ml}$)										
Antimicrobials		<0,5	0,5	1	—	2	4	8	16	32	64	>64
Ciprofloxacin (>0,5)	broilers	1	2			6	19	5	3			
	pigs			3		4	9	1				
Erytromycin (>4)	broilers			33		1	1					1
	pigs			8		3	2					3
Gentamicin (>2)	broilers	10	22		3	1						
	pigs			17								
Nalidixic acid (>16)	broilers			2				1		8	20	5
	pigs							1		1	4	11
Streptomycin (>4)	broilers	1	1		17	10		1	1	5		
	pigs					1			6	10		
Tetracycline (>1)	broilers	15		2	3						1	5
	pigs			2		1					1	2

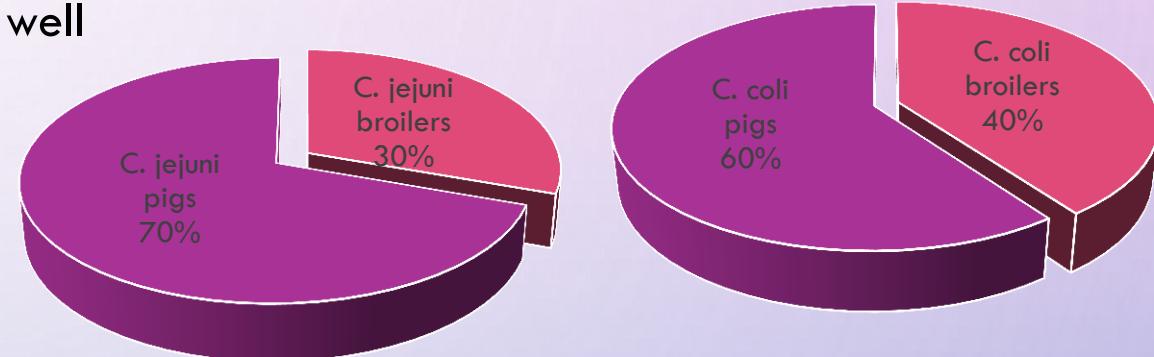
MICROBIAL RESISTANCE - actual situation in the SR

Conclusion – prevalence of resistance among *Campylobacter* isolates

- Bacterial species and animal species differences in MR as well
- MDR: 7 vs. 16 isolates showed MDR, no 5DR

broilers: CIP, NAX, follow by the same pattern with
TET, the most observe patterns

pigs: CIP,NAX,TET, STR the most observe patterns



- GEN – zero resistance in both species (no selective pressure, no oral administration)
- ERY – low resistance in both species (in pig little bit higher R level – selective atb pressure)
- STR – moderate – very high level of resistance – species differences (only 3 *C.coli* from pigs) – no bifunctional enzyme production
- CIP, NAX, TET high - extrem high resistance – probably onepoint chrom.mutation

MICROBIAL RESISTANCE - actual situation in the SR

- Microbial resistance in *Escherichia coli* isolates:
- Active monitoring: According to Desicion 2013/652/EU - Harmonised monitoring and reporting of AMR in zoonotic and commensal bacteria (including *Escherichia coli*)
- Passive monitoring: clinical samples from animals (faeces, swabs, rectal/cloacal swabs, carcasses) - data not included
- the highest level of microbial resistance in *Escherichia coli* isolates – caecal samples gathered at slaughter from broilers – the focus of this presentation
- 86 positive samples for commensal *Escherichia coli*

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Escherichia coli* from broilers caecum – qualitative data

Evaluation:

E.coli: 86 positive – 1x fully sensitive

Lot of combinations of R patterns

NxCp – R: 79x; SuTCpTeNxChA – R: 8x;
STCpTeNxA – R: 7x

Fot,Caz – R: 10x

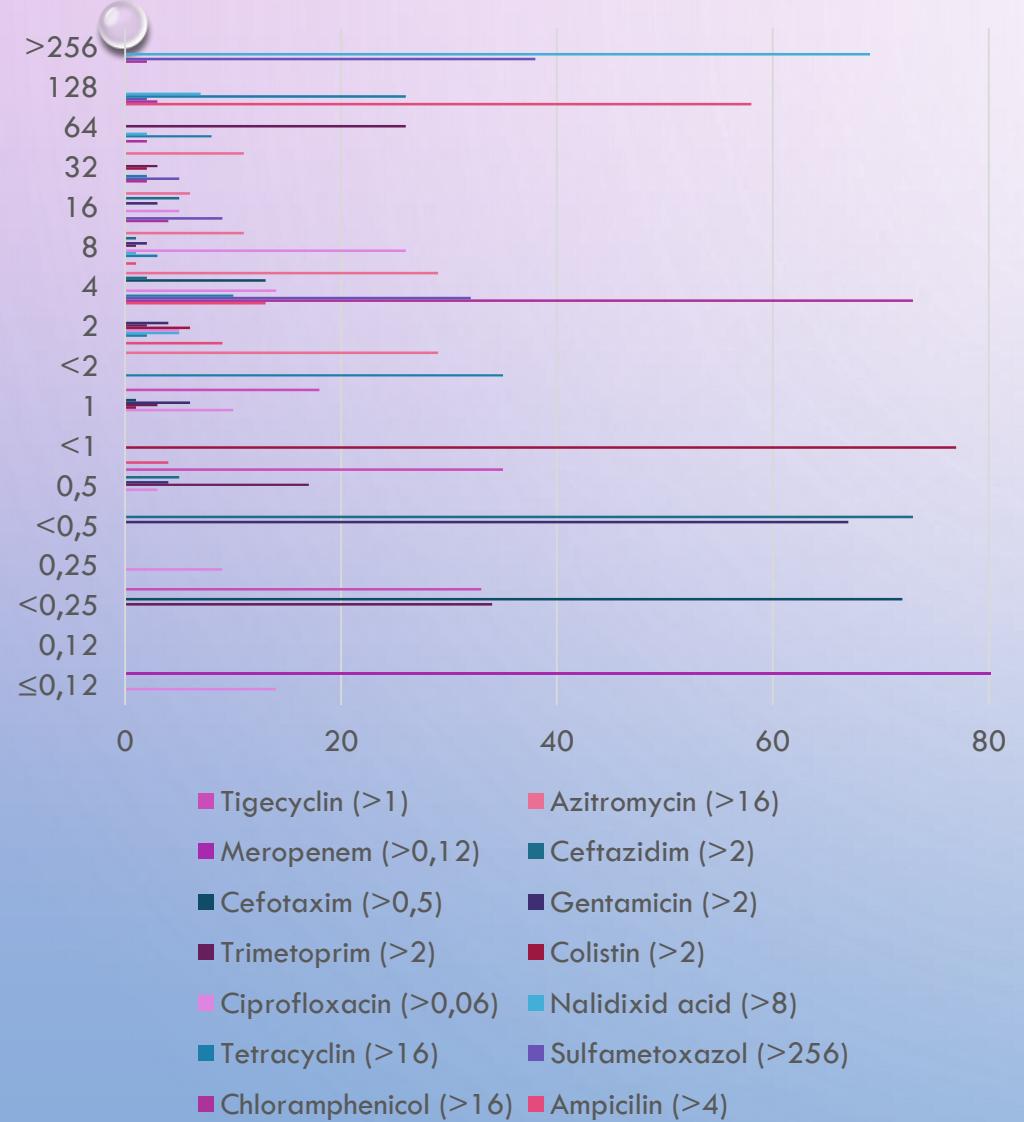
NxCp – R: 79x

53 MDR isolates, 20 - 5-DR isolates

<i>Escherichia coli</i>	Broilers caecum	
No. of tested isolates	86	
Antimicrobials	n	R%
Ampicilin (>8)	58	67
Chloramphenicol (>16)	7	8
Sulfametoxazol (>64)	38	44
Tetracyclin (>8)	36	42
Nalidixid acid (>16)	78	91
Ciprofloxacin (>0,06)	72	84
Colistin (>2)	2	2
Trimetoprim (>2)	30	35
Gentamicin (>2)	5	6
Cefotaxim (>0,25)	14	16
Ceftazidim (>0,5)	8	9
Meropenem (>0,12)	0	0
Azitromycin (>16)	11	13
Tigecyclin (>1)	0	0

MICROBIAL RESISTANCE - actual situation in the SR

Profile of *Escherichia coli* in broilers caecum - quantitative data (86 isolates)



MICROBIAL RESISTANCE - actual situation in the SR

Conclusion – prevalence of resistance among *Escherichia coli* isolates in 2014

- MERO, GEN, CHL – zero – low level R - – **no carbapenemase *Escherichia coli* producer findings**
- COL – 2% - *E. coli* marker – EURL-AMR
- SSS, W – moderate resistance
- TET – high R – 67% - vs. TGC: tetracycline derivate antibiotic (new atb) – 0% - growth promoter before banning
- AMP – high resistance – (detto *Salmonella*) – depends on the content of bacterial wall
- CAZ, CTX – low R to Cephalosporine 3rd generation — **ESBL/AmpC *Escherichia coli* producer findings (without specification)**
- CIP, NAX – extreme high resistance: phenotypic ID of mechanism: susp. 1 point chrom.mutation – multi chrom.m. (qnr)
- MDR: 53 isolates showed MDR, 20 isolates - penta resistant patern observe
CIP, NAX - the most observe patterns

MICROBIAL RESISTANCE - actual situation in the SR

- Prevalence of ESBL/AmpC and carbapenemase resistance among *Escherichia coli* isolates in 2015

- Active monitoring: According to Desicion 2013/652/EU - Harmonised monitoring and reporting of AMR in zoonotic and commensal bacteria, including specific ESBL- carbapenemase monitoring

- ✓ caecal samples gathered at slaughter from pigs
- ✓ bovine meat from retail
- ✓ pork meat from retail

MICROBIAL RESISTANCE - actual situation in the SR

Prevalence of ESBL/AmpC and carbapenemase resistance among *Escherichia coli* isolates in 2015

- Preliminary results to 1.10.2015

✓ caecal samples gathered at slaughter from pigs: 119 / 43 positive – 36%

17x AmpC phenotype

7x ESBL phenotype

19x AmpC+ESBL phenotype

no carbapenemase findings

✓ bovine meat from retail: 125 / 5 positive – 4%

5x AmpC phenotype

no carbapenemase findings

✓ pork meat from retail: 132 / 4 positive – 3%

1x AmpC phenotype

2x ESBL phenotype

1x AmpC+ESBL phenotype

no carbapenemase findings

THANKS FOR YOUR ATTENTION