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## Investigation of pork meat in chicken- and beef-based commercial products by ELISA and real-time PCR sold at retail in Kosovo

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The authors are fully responsible for both the content and the formal aspects of the electronic supplementary material. No editorial adjustments were made.

### Electronic supplementary material

Supplementary Figure S1  
Supplementary Tables S1–S2

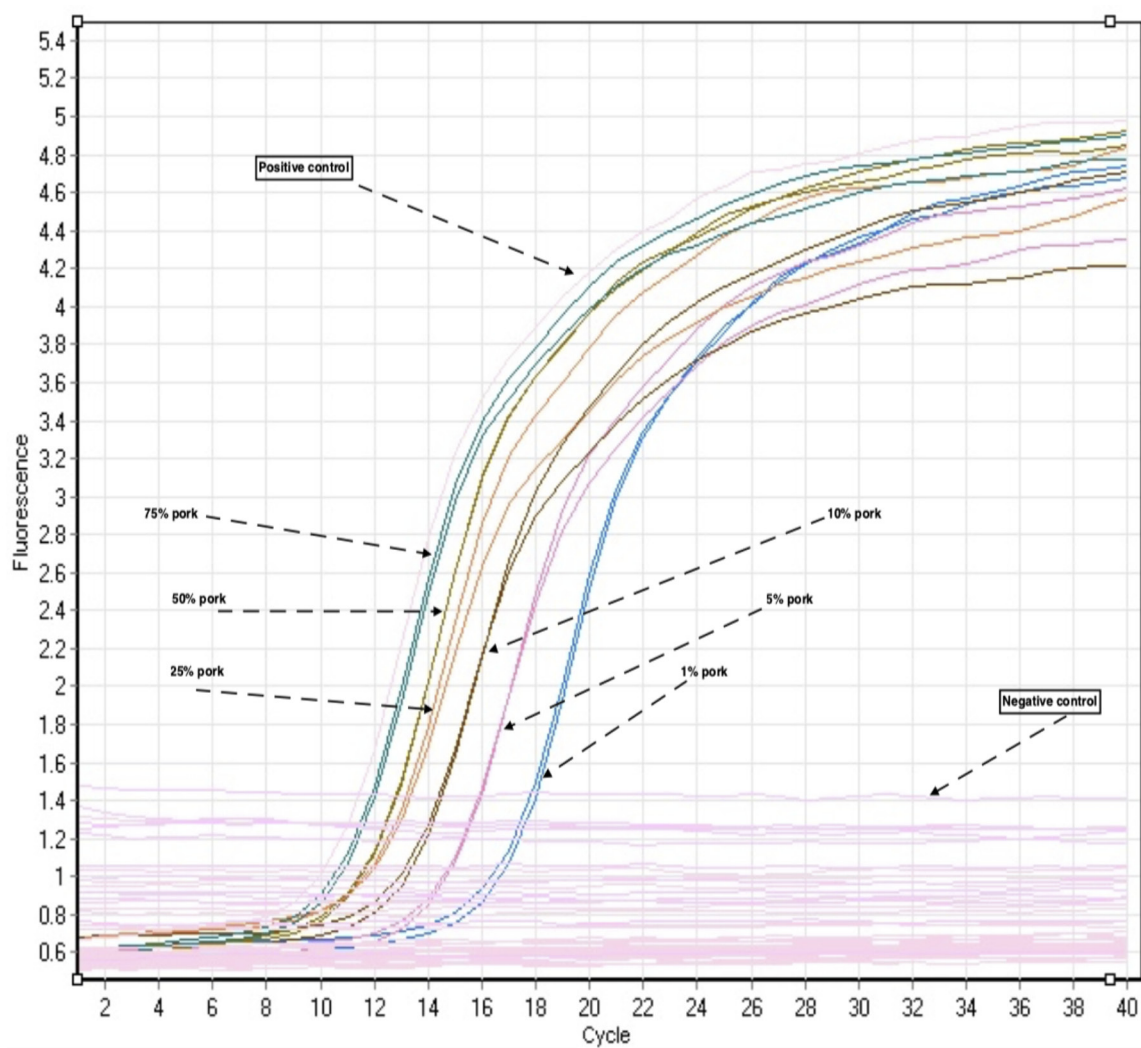


Figure S1A. Standardised food-matrices with known amount of pork (1–75%) in beef-based meat products

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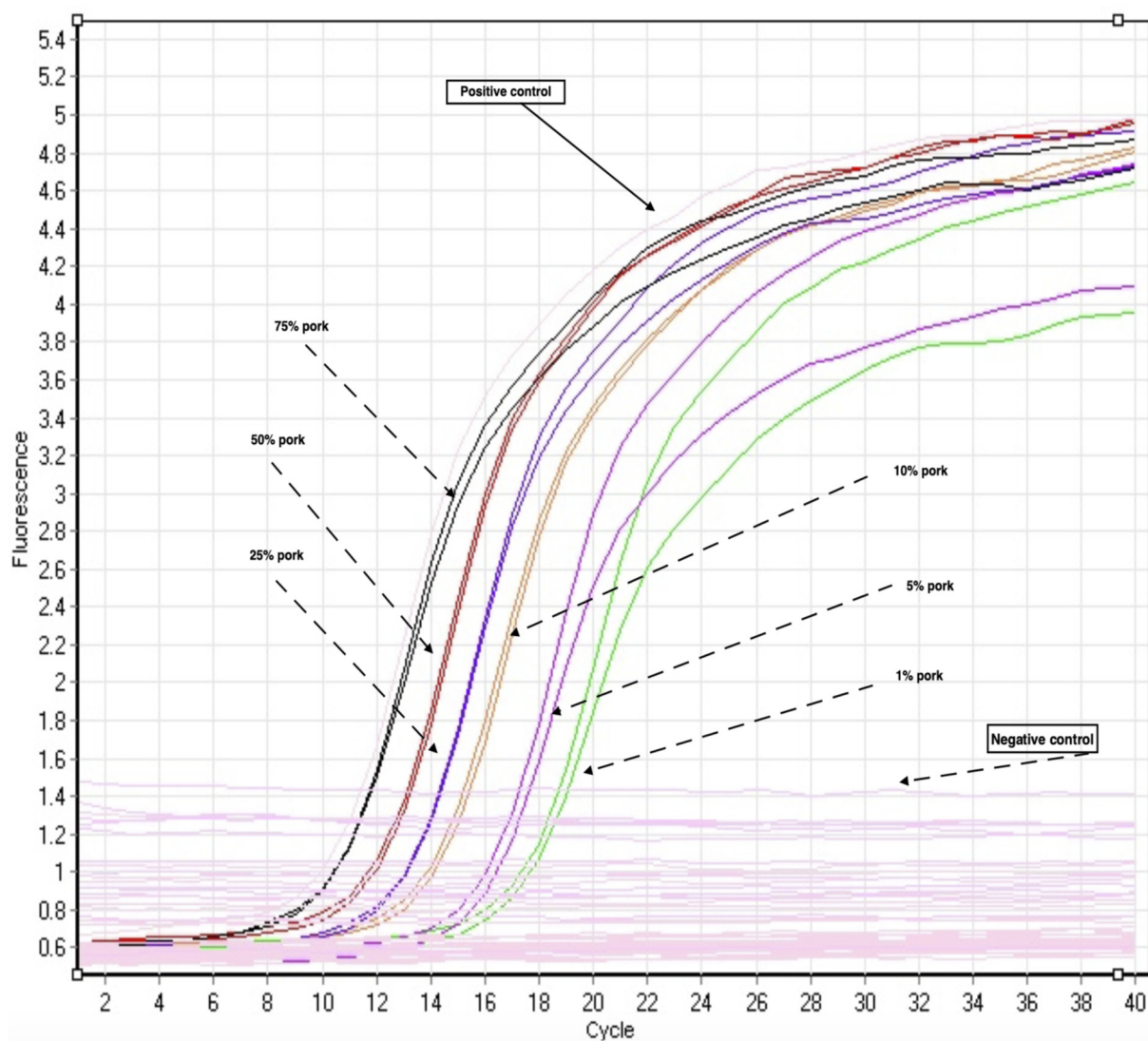


Figure S1B. Standardised food-matrices with known amount of pork (1–75%) in chicken-based meat products

<https://doi.org/10.17221/164/2020-CJFS>Table S1. Results of adulterated pork in chicken and beef commercial processed meat products tested with PCR *mericon* kit, PCR with designed primer and ELISA commercial kit

ID	Product type	Declaration of species content	ELISA commercial kit (% w/w)	Real-time PCR commercial kit	Real-PCR pork primer	Origin (ISO)
1	salami	100% chicken	< 0.02	not tested	not tested	KS
2	salami	100% chicken	< 0.02	not tested	not tested	KS
3	salami	100% chicken, MDM halal	< 0.02	not tested	not tested	KS
4	salami	100% chicken	< 0.02	not tested	not tested	KS
5	salami	68% MDM, meat skin, proteins, water halal	< 0.02	not tested	not tested	SI
6	salami	55% MDM, 15% minced meat, 5% skin, proteins, water	< 0.02	not tested	not tested	BA
7	salami	50% meat, 37% MDM, skin, water	< 0.02	not tested	not tested	NL
8	salami	100% chicken	< 0.02	not tested	not tested	KS
9	salami	100% chicken	< 0.02	not tested	not tested	KS
10	<b>sausage</b>	100% chicken, MDM, skin	0.02–0.1	+	+	<b>MK</b>
11	sausage	100% MDM	< 0.02	not tested	not tested	KS
12	sausage	100% chicken meat	< 0.02	not tested	not tested	KS
13	sausage	100% chicken meat	< 0.02	not tested	not tested	KS
14	sausage	100% MDM	< 0.02	not tested	not tested	AL
15	salami	82% chicken meat, water	< 0.02	not tested	not tested	SI
16	sausage	94 % MDM chicken and trout meat	< 0.02	not tested	not tested	IT
17	sausage	chicken-MDM 54%, trout-MDM 11 %	< 0.02	not tested	not tested	IT
18	sausage with pistachios	MDM 48%, meat 13%	< 0.02	not tested	not tested	NL
19	sausage	chicken meat 50%, trout 20%, skin	< 0.02	not tested	not tested	DE
20	pâté	30% cooked chicken meat, milk proteins	< 0.02	not tested	not tested	BA
21	pâté picante	30% cooked chicken meat, milk proteins	< 0.02	not tested	not tested	SI
22	pâté	100% chicken, halal	< 0.02	not tested	not tested	KS
23	pâté	35% chicken meat, 5% chicken skin, 9% proteins	< 0.02	not tested	not tested	BA
24	pâté	100% chicken MDM. halal	< 0.02	not tested	not tested	KS
25	pâté	37% chicken meat	< 0.02	not tested	not tested	HR
26	pâté	35% MDM	< 0.02	not tested	not tested	MK
27	pâté	35% MDM, 20% chicken connective tissue	< 0.02	not tested	not tested	MK
28	salami, premium	70% MDM	< 0.02	not tested	not tested	SI
29	pâté	45% minced chicken, skin, liver	< 0.02	not tested	not tested	SI
30	pâté picante	35% chicken meat, 20% fat, liver	< 0.02	not tested	not tested	BA
31	ragu sauce	30% chicken meat, 10% fat	< 0.02	not tested	not tested	BA
32	<b>sausage</b>	52% chicken meat, 13 % pork MDM	<b>0.1–0.5</b>	+	+	<b>IT</b>
33	sausage	68% MDM chicken and 22% trout meat	< 0.02	not tested	not tested	IT
34	<b>sausage</b>	55% chicken MDM, 33% trout	<b>0.02–0.1</b>	+	+	<b>IT</b>
35	sausage	100% vegetal	< 0.02	not tested	not tested	IT
36	file	100% chicken	< 0.02	not tested	not tested	IT
37	file	100% chicken Brust	< 0.02	not tested	not tested	KS
38	sausage	100% beef	< 0.02	-	-	KS
39	file	100% beef	< 0.02	not tested	not tested	BA
40	file	60% beef, rest: wheat, egg, milk	< 0.02	not tested	not tested	HR
41	ragu sauce	45% beef meat, rest: wheat, egg, milk	< 0.02	not tested	not tested	HR

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Table S1. To be continued

ID	Product type	Declaration of species content	ELISA commercial kit (% w/w)	Real-time PCR commercial kit	Real-PCR pork primer	Origin (ISO)
42	file	60% beef, 8% proteins	<b>0.02–0.1</b>	+	+	<b>BA</b>
43	pâté	60% beef meat, 10% beef fat	< 0.02	not tested	not tested	BA
44	salami	100% beef	< 0.02	-	not tested	KS
45	salami	100% beef, halal	< 0.02	not tested	not tested	KS
46	salami	100% beef	< 0.02	-	-	KS
47	salami	100% beef, premium meat, fat	< 0.02	not tested	not tested	KS
48	salami	100% beef, halal	< 0.02	not tested	not tested	KS
49	<b>sausage</b>	mixed beef and chicken	<b>0.02–0.1</b>	+	+	<b>KS</b>
50	dried meat	100% beef	< 0.02	not tested	not tested	MK
51	file	100% beef	< 0.02	-	not tested	KS
52	Balkan sausage-file	100% beef	< 0.02	not tested	not tested	KS
53	<b>Balkan sausage-file</b>	100% beef meat and fat	<b>0.02–0.1</b>	+	+	<b>MN</b>
54	<b>Balkan sausage-file</b>	100% beef	<b>0.1–0.5</b>	+	+	<b>SI</b>
55	Balkan sausage (home made)	100% beef	< 0.02	not tested	not tested	KS
56	Balkan sausage (home made)	100% beef	< 0.02	not tested	not tested	KS
57	<b>Balkan sausage (home made)</b>	mixed beef and chicken MDM	0.02–0.1	+	+	<b>KS</b>
58	<b>Balkan sausage (home made)</b>	100% beef	0.02–0.1	+	+	<b>KS</b>
59	<b>Balkan sausage (home made-picante)</b>	40% beef meat, 40% chicken MDM	0.02–0.1	+	+	<b>KS</b>
60	<b>Balkan sausage (home made)</b>	100% beef-trimming	0.02–0.1	+	+	<b>KS</b>
61	Balkan sausage industrial	100% beef	< 0.02	not tested	not tested	KS
62	Balkan sausage industrial	30% beef meat, 20% chicken meat	< 0.02	not tested	not tested	KS

(+) – positive sample; (–) – negative sample; BA – Bosnia and Herzegovina; DE – Germany; HR – Croatia; IT – Italy; KS – Kosovo, MN – Monte Negro, MK – North Macedonia, NL – Netherland; SI – Slovenia; MDM – mechanically deboned meat; ELISA – enzyme-linked immunosorbent assay; PCR – polymerase chain reaction; ID 38, 44, 46, 51 were screened negative on ELISA but were on the upper detection limit, therefore are tested for confirmation of the negative results with *mericon* real-time PCR; and ID 38 and ID 44 are additionally tested for confirmation of negative results with pork specific primers; bold – positive sample



Table S2A. ELISA results for chicken samples

Assay: PiG; Sample-ID-Chicken					Assay: PiG; Sample-ID-Chicken									
	1	2	3	4		4	5	6	7	8	9	10	11	12
A	NC	M2	M6	NC	A	NC	M11	M15	M19	NC	M24	M29	M33	M37
B	NC	M2	M6	NC	B	NC	M11	M15	M19	NC	M24	M29	M33	M37
C	LC	M3	M7	LC	C	LC	M12	M16	M20	LC	M25	M30	M34	NC
D	LC	M3	M7	LC	D	LC	M12	M16	M20	LC	M25	M30	M34	NC
E	HC	M4	M8	HC	E	HC	M13	M17	M21	HC	M26	M31	M35	LC
F	HC	M4	M8	HC	F	HC	M13	M17	M21	HC	M26	M31	M35	LC
G	M1	M5	M9		G	M10	M14	M18	M22	M23	M27	M32	M36	HC
H	M1	M5	M9		H	M10	M14	M18	M22	M23	M27	M32	M36	HC
Results: Sample-OD @ 450nm					Results: Sample-OD @ 450nm									
	1	2	3	4		4	5	6	7	8	9	10	11	12
A	0.093	0.065	0.065	0.271	A	0.136	0.102	0.1	0.097	0.124	0.093	0.099	0.088	0.0351
B	0.089	0.056	0.057	0.129	B	0.133	0.101	0.097	0.094	0.155	0.087	0.085	0.095	0.039
C	1.876	0.060	0.058	1.162	C	2.210	0.091	0.088	0.083	2.070	0.08	0.086	0.394	0.148
D	1.950	0.068	0.078	1.128	D	2.118	0.092	0.09	0.086	2.038	0.086	0.094	0.384	0.154
E	3.438	0.059	0.066	2.906	E	3.500	0.065	0.093	0.061	3.342	0.061	0.061	0.062	2.000
F	3.500	0.066	0.059	3.500	F	3.500	0.103	0.128	0.099	3.500	0.094	0.099	0.055	1.967
G	0.076	0.086	0.063		G	0.191	0.129	0.120	0.115	0.12	0.126	2.731	0.057	3.500
H	0.083	0.078	0.100		H	0.202	0.131	0.133	0.122	0.127	0.128	2.850	0.101	3.158

ELISA – enzyme-linked immunosorbent assay; NC – negative control/not detected; LC – low control/low detection; HC – high control/high detection; yellow field – positive samples

Interpretation – not detected: < 0.02% w/w (NC); low detected: 0.02–0.1% w/w (> NC & < LC); moderate detection: 0.1–0.5% w/w (> LC & < HC); high detected: > 0.5% (> HC)

Table S2B. ELISA results for beef samples

Assay: PiG; Sample-ID-Beef								
	1	2	3	4	5	6	7	8
A	NC	M39	M43	M47	NC	M52	M56	M60
B	NC	M39	M43	M47	NC	M52	M56	M60
C	LC	M40	M44	M48	LC	M53	M57	M61
D	LC	M40	M44	M48	LC	M53	M57	M61
E	HC	M41	M45	M49	HC	M54	M58	M62
F	HC	M41	M45	M49	HC	M54	M58	M62
G	M38	M42	M46	M50	M51	M55	M59	NC
H	M38	M42	M46	M50	M51	M55	M59	NC
Results: Sample-OD @ 450nm								
	1	2	3	4	5	6	7	8
A	0.081	0.057	0.052	0.053	0.05	0.055	0.046	0.699
B	0.062	0.055	0.048	0.046	0.049	0.057	0.051	0.691
C	1.752	0.054	0.056	0.056	1.667	0.14	0.232	0.068
D	1.744	0.06	0.082	0.062	1.591	0.132	0.237	0.063
E	3.139	0.062	0.066	0.088	2.872	1.981	0.117	0.081
F	3.159	0.053	0.053	0.068	2.929	2.005	0.105	0.055
G	0.077	0.12	0.068	0.067	0.073	0.079	0.381	0.059
H	0.083	0.152	0.071	0.071	0.076	0.074	0.399	0.068

ELISA – enzyme-linked immunosorbent assay; NC – negative control/not detected; LC – low control/low detection; HC – high control/high detection; yellow field – positive samples

Interpretation – not detected: < 0.02% w/w (NC); low detected: 0.02–0.1% w/w (> NC & < LC); moderate detection: 0.1–0.5% w/w (> LC & < HC); high detected: > 0.5% (> HC)