

Anhang 1: Ergebnisse der Ringversuchsserie „Bakterien- und Pilzgenom-Nachweis (PCR/NAT)“ November 2013

Anhang 1 zu:

Reischl U, Schneider W, Holzmann T, Ehrenschwender M, Maaß M, Straube E, Frangoulidis D, Grass G, Splettstösser W, Fingerle V, Sing A, Jacobs E, Reiter-Owona I. Bakterien- und Pilzgenom-Nachweis PCR/NAT: Auswertung des Ringversuchs November 2013 von INSTAND e.V. zur externen Qualitätskontrolle molekularbiologischer Nachweisverfahren in der bakteriologischen Diagnostik. GMS Z Forder Qualitätssich Med Lab. 2014;5:Doc01. DOI: 10.3205/lab000011, URN: urn:nbn:de:0183-lab0000116

Attachment 1: Results of the proficiency testing scheme “Bacterial and Fungal Genome Detection (PCR/NAT)” November 2013

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PCR-/NAT *C. trachomatis* & GO (RV 530) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325301	Ø/++	63	<i>Neisseria gonorrhoeae</i> (~ 1x10 ⁴ CFU/mL)
1325302	Ø/+	63	<i>Neisseria gonorrhoeae</i> (~ 1x10 ³ CFU/mL)
1325303	++/Ø	61	<i>Chlamydia trachomatis</i> (~ 5x10 ³ IFU/mL)
1325304	+/+	62	<i>Chlamydia trachomatis</i> (~ 1x10 ³ IFU/mL) <i>Neisseria gonorrhoeae</i> (~ 1x10 ³ CFU/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=148</i>	<i>Probennummer (Sample no.)</i>				<i>Inhibition</i>				
	1325301	1325302	1325303	1325304	1325301	1325302	1325303	1325304	
<i>Befund Result</i>									
Positiv CT	0	0	145	16	n.d.	0	0	0	0
Positiv CT & GO	1	0	0	129	nein/no	148	148	148	148
Positiv GO	143	134	0	3	ja/yes	0	0	0	0
Negativ	4	11	3	0					
Fraglich/ <i>questionable</i>	0	3	0	0					

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
GenProbe CT/NG [20] (n=10)	24	24/38 [§]	63	0	0/0	0
LightMix CT/NG [21] (n=8)	30	30/32	94	0	0/0	0
Roche COBAS [22] (n=32)	122	122/128	95	0	0/0	0
Cepheid Xpert CT/NG [23] (n=1)	4	4/4	100	0	0/0	0
BD ProbeTec [24] (n=19)	74	74/75 [§]	99	0	0/0	0
Artus CT [25] (n=3)	11	11/12	92	0	0/0	0
Abbott RealTime CT/NG [26] (n=30)	119	119/120	99	0	0/0	0
Other commercial tests [27] (n=38)	145	145/152	95	0	0/0	0
<i>In house</i> PCR assay [28] (n=13)	50	50/52	96	0	0/0	0
Andere/k.A./other [29] (n=6)	20	20/24	83	0	0/0	0

[§] Due to reporting questionable results, the number of true results (denominator in the „relative“ column) has been reduced.

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.

Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants

Tabelle 4: Häufigkeit der Mitteilung verschiedener Befunde. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für Chlamydia trachomatis dargestellt.

Absolute numbers of reported individual results. Note: only the C. trachomatis-specific results are depicted in this table

n=147	Probennummer (Sample no.)			
Befund <i>Result</i>	1325301	1325302	1325303	1325304
Positiv	1	0	145	145
Negativ	146	147	2	2
Fraglich <i>Questionable</i>	0	0	0	0

	Inhibition			
	1325301	1325302	1325303	1325304
n.d.	0	0	0	0
nein <i>no</i>	147	147	147	147
ja <i>yes</i>	0	0	0	0

Tabelle 5: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für ***Chlamydia trachomatis*** dargestellt. *Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods. Note: only the C. trachomatis-specific results are depicted in this table.*

NAT-Methode (nur CT) [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
GenProbe CT/NG [20] (n=10)	20	20/20	100	20	20/20	100
LightMix CT/NG [21] (n=8)	14	14/16	88	16	16/16	100
Roche COBAS [22] (n=32)	64	64/64	100	64	64/64	100
Cepheid Xpert CT/NG [23] (n=1)	2	2/2	100	2	2/2	100
BD ProbeTec [24] (n=19)	38	38/38	100	38	38/38	100
Artus CT [25] (n=3)	6	6/6	100	6	6/6	100
Abbott RealTime CT/NG [26] (n=30)	60	60/60	100	60	60/60	100
Other commercial tests [27] (n=37)	73	73/74	99	74	74/74	100
In house PCR assay [28] (n=13)	25	25/26	96	25	25/26	96
Andere/k.A./other [29] (n=6)	12	12/12	100	12	12/12	100

Tabelle 6: Häufigkeit der Mitteilung verschiedener Befunde. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für ***Neisseria gonorrhoeae*** dargestellt. *Absolute numbers of reported individual results. Note: only the N. gonorrhoeae-specific results are depicted in this table.*

n=144	Probennummer (Sample no.)			
Befund <i>Result</i>	1325301	1325302	1325303	1325304
Positiv	144	134	0	132
Negativ	0	7	144	12
Fraglich <i>Questionable</i>	0	3	0	0

	Inhibition			
	1325301	1325302	1325303	1325304
n.d.	0	0	0	0
nein <i>no</i>	144	144	144	144
ja <i>yes</i>	0	0	0	0

Tabelle 7: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für *Neisseria gonorrhoeae* dargestellt.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods. Note: only the N. gonorrhoeae-specific results are depicted in this table.

NAT-Methode (nur GO) [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
GenProbe CT/NG [20] (n=10)	14	14/28 [§]	50	10	10/10	100
LightMix CT/NG [21] (n=8)	24	24/24	100	8	8/8	100
Roche COBAS [22] (n=30)	90	90/90	100	30	30/30	100
Cepheid Xpert CT/NG [23] (n=1)	3	3/3	100	1	1/1	100
BD ProbeTec [24] (n=19)	55	55/56 [§]	98	19	19/19	100
Artus CT [25] (n=3)	8	8/9	89	3	3/3	100
Abbott RealTime CT/NG [26] (n=30)	89	89/90	99	30	30/30	100
Other commercial tests [27] (n=37)	110	110/111	99	37	37/37	100
<i>In house</i> PCR assay [28] (n=13)	38	38/39	97	13	13/13	100
Andere/k.A./other [29] (n=5)	14	14/15	93	5	5/5	100

[§] Due to reporting questionable results, the number of true results (denominator in the „relative“ column) has been reduced.

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.

Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants

Comments:

¹⁾ Four of the 148 participants performed only *C. trachomatis* detection, one of the 148 participants performed only *N. gonorrhoeae*-specific assays, whereas the other 143 laboratories detected both species.

PCR-/NAT *Chlamydia trachomatis* (RV 531) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.

Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325311	+	61	<i>Chlamydia trachomatis</i> (~ 1x10 ³ IFU/mL)
1325312	∅	62	<i>Escherichia coli</i> K12
1325313	+	61	<i>Chlamydia trachomatis</i> (~ 1x10 ³ IFU/mL)
1325314	++	61	<i>Chlamydia trachomatis</i> (~ 5x10 ³ IFU/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.

Absolute numbers of reported individual results.

<i>n=102</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325311</i>	<i>1325312</i>	<i>1325313</i>	<i>1325314</i>		<i>1325311</i>	<i>1325312</i>	<i>1325313</i>	<i>1325314</i>
Befund <i>Result</i>									
Positiv	100	3	95	110	n.d.	0	0	0	0
Negativ	2	98	6	2	nein <i>no</i>	102	102	102	102
Fraglich <i>Questionable</i>	0	1	1	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
Hain GenoQuick CT [20] (n=17)	48	48/51	94	14	14/17	82
TIB Molbiol LightMix CT [21] (n=1)	2	2/3	67	1	1/1	100
Roche COBAS CT [22] (n=26)	76	76/78	97	26	26/26	100
Cepheid Xpert CT/NG [23] (n=5)	15	15/15	100	5	5/5	100
BD ProbeTec [24] (n=13)	38	38/39	97	13	13/13	100
Artus CT [25] (n=11)	33	33/33	100	11	11/11	100
Abbott CT/NG [26] (n=4)	10	10/12	83	4	4/4	100
Other commercial tests [27] (n=14)	42	42/42	100	14	14/14	100
<i>In house</i> PCR assay [28] (n=10)	28	28/29 [§]	97	9	9/9 [§]	100
Andere/k.A. <i>l other</i> [29] (n=1)	3	3/3	100	1	1/1	100

[§] Due to reporting questionable results, the number of true results (denominator in the „relative“ column) has been reduced.

PCR-/NAT *Bordetella pertussis* (RV 532) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.

Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325321	∅	62	<i>Escherichia coli</i> K12
1325322	++	61	<i>Bordetella pertussis</i> (~ 1x10 ⁴ CFU/mL)
1325323	∅	62	<i>Escherichia coli</i> K12
1325324	∅	62	<i>Bordetella parapertussis</i> (~ 1x10 ⁴ CFU/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.

Absolute numbers of reported individual results.

<i>n=116</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325321</i>	<i>1325322</i>	<i>1325323</i>	<i>1325324</i>		<i>1325321</i>	<i>1325322</i>	<i>1325323</i>	<i>1325324</i>
<i>Befund Result</i>									
Positiv	0	115	0	3	n.d.	1	1	1	1
Negativ	116	1	116	113	nein no	115	115	115	115
Fraglich Questionable	0	0	0	0	ja yes	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
TIB Molbiol LightMix BP [20] (n=12)	12	12/12	100	36	36/36	100
Diagenode <i>B.pertussis</i> [21] (n=8)	8	8/8	100	24	24/24	100
GenoQuick Bordetella [22] (n=7)	7	7/7	100	21	21/21	100
Other commercial tests [27] (n=37)	37	37/37	100	109	109/111	98
<i>In house</i> PCR assay [28] (n=52)	51	51/52	98	155	155/156	99
Anderer/k.A./other [29] (n=1)	1	1/1	100	3	3/3	100

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.
Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants

PCR-/NAT *Helicobacter pylori* (RV 533) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.

Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325331	∅	62	<i>Escherichia coli</i> K12
1325332	+++	61/72	<i>Helicobacter pylori</i> (~ 1x10 ⁶ CFU/mL) Clarithromycin susceptible (wildtype 23S rDNA sequence)
1325333	++	61/72	<i>Helicobacter pylori</i> (~ 1x10 ⁵ CFU/mL) Clarithromycin susceptible (wildtype 23S rDNA sequence)
1325334	+	61/72	<i>Helicobacter pylori</i> (~ 1x10 ⁴ CFU/mL) Clarithromycin susceptible (wildtype 23S rDNA sequence)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.

Absolute numbers of reported individual results.

<i>n=35</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	1325331	1325332	1325333	1325334		1325331	1325332	1325333	1325334
Befund <i>Result</i>									
Positiv	0	35 ¹⁾	35 ¹⁾	35 ¹⁾	n.d.	0	0	0	0
Negativ	35	0	0	0	nein <i>no</i>	35	35	35	35
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Comments:

¹⁾ Twenty nine of the 35 participants reported results for molecular Clarithromycin-susceptibility testing. With the exception of 1 laboratory, all reported results were correct.

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
Hain GenoType Helico [25] (n=12)	36	36/36	100	12	12/12	100
Ingenetix ClariRes [26] (n=3)	9	9/9	100	3	3/3	100
Commercial assay [27] (n=5)	15	15/15	100	5	5/5	100
<i>In house</i> PCR assay [28] (n=16)	48	48/48	100	16	16/16	100
Andere/k.A./other [29] (n=1)	3	3/3	100	1	1/1	100

PCR-/NAT EHEC/STEC (RV 534) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325341	+++	61/71, 77, 78	EHEC (~1x10 ⁵ CFU/mL) (<i>stx-1</i> , <i>eae</i> , <i>hlyA</i> and O157 positive)
1325342	++	61/71, 72, 77, 78	EHEC (~1x10 ⁴ CFU/mL) (<i>stx-1</i> , <i>stx-2</i> , <i>eae</i> , <i>hlyA</i> and O157 positive)
1325343	+++	61/72, 76, 77	EHEC (~1x10 ⁵ CFU/mL) (<i>stx-2f</i> and <i>eae</i> positive)
1325344	Ø	62	<i>Escherichia coli</i> K12 (negative for <i>eae</i> and <i>hlyA</i>)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=104</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	1325341	1325342	1325343	1325344		1325341	1325342	1325343	1325344
Befund <i>Result</i>									
Positiv	101 ¹⁾	98 ¹⁾	25 ¹⁾	2	n.d.	1	1	1	1
Negativ	3	6	79 ²⁾	102	nein <i>no</i>	103	103	103	103
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Comments:

¹⁾ Partial or complete shiga toxin subtyping, *eae*-, and *hlyA*-detection was performed by 95 laboratories. With the exception of 5 laboratories, all reported results were correct.

²⁾ As the clinical relevance of shiga toxin 2f positive EHEC is not proven yet, negative PCR results were not rated as "false negative".

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
Hain GenoType EHEC [20] (n=24)	47	47/72	65	24	24/24	100
Hyplex EHEC [21] (n=5)	10	10/15	67	5	5/5	100
r-Biopharm RIDAGENE [22] (n=14)	36	36/42	86	13	13/14	93
Other commercial tests [27] (n=9)	21	21/27	78	9	9/9	100
<i>In house</i> PCR assay [28] (n=51)	110	110/153	72	50	50/51	98
Anderer/ k.A./other [29] (n=3)	4	4/9	44	3	3/3	100

PCR-/NAT *Borrelia burgdorferi* (RV 535) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325351	(++)	61	<i>Borrelia bissetii</i> (~1x10 ⁴ organisms/mL)
1325352	∅	62	<i>Escherichia coli</i> K12 (negative for <i>eae</i> and <i>hlyA</i>)
1325353	++	61	<i>Borrelia spielmanii</i> (~1x10 ⁴ organisms/mL)
1325354	++	61	<i>Borrelia afzelii</i> (~1x10 ⁴ organisms/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=96</i>	<i>Probennummer (Sample no.)</i>				<i>Inhibition</i>				
	<i>1325351</i>	<i>1325352</i>	<i>1325353</i>	<i>1325354</i>	<i>1325351</i>	<i>1325352</i>	<i>1325353</i>	<i>1325354</i>	
<i>Befund Result</i>									
Positiv	87	1	92	91	n.d.	1	1	1	1
Negativ	7 ¹⁾	95	4	5	nein no	95	95	95	95
Fraglich Questionable	2 ¹⁾	0	0	0	ja yes	0	0	0	0

Comments:

¹⁾ As sample #1325351 contained a very rare *Borrelia* isolate (*B. bissetii*), negative PCR results were not rated as "false negative".

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
artus <i>Borrelia</i> LC Kit [20] (n=21)	60	60/63	95	20	20/21	95
Demeditec GenFlow [21] (n=5)	15	15/15	100	5	5/5	100
LightMix <i>Borrelia</i> [22] (n=4)	12	12/12	100	4	4/4	100
Other/commercial tests [27] (n=13)	36	36/38 [§]	95	13	13/13	100
<i>In house</i> PCR assay [28] (n=53)	147	147/158 [§]	93	53	53/53	100

[§] Due to reporting questionable results, the number of true results (denominator in the "relative" column) has been reduced.

PCR-/NAT *Legionella pneumophila* (RV 536) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325361	+++	61	<i>Legionella pneumophila</i> SG3 (~ 1x10 ⁶ CFU/mL)
1325362	+	61	<i>Legionella pneumophila</i> SG3 (~ 1x10 ⁴ CFU/mL)
1325363	∅	62	<i>Chlamydia pneumoniae</i> (~ 1x10 ⁶ IFU/mL)
1325364	∅	62	<i>Escherichia coli</i> K12

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=84</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325361</i>	<i>1325362</i>	<i>1325363</i>	<i>1325364</i>		<i>1325361</i>	<i>1325362</i>	<i>1325363</i>	<i>1325364</i>
Befund <i>Result</i>									
Positiv	84	78	0	1	n.d.	0	0	0	0
Negativ	0	6	84	83	nein <i>no</i>	84	84	84	84
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwenden verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
LightMix <i>Legionella</i> [25] (n=7)	14	14/14	100	14	14/14	100
GeneProof <i>L.pneumophila</i> [26] (n=5)	10	10/10	100	10	10/10	100
Other commercial tests [27] (n=27)	51	51/54	94	53	53/54	98
<i>In house</i> PCR assay [28] (n=45)	88	88/90	97	90	90/90	100
Andere/k.A./other [29] (n=2)	3	3/4	75	4	4/4	100

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.
Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants.

PCR-/NAT *Salmonella enterica* (RV 537) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325371	+++	61	<i>S. enterica</i> ser. Typhi (~ 1x10 ⁶ CFU/mL)
1325372	∅	62	<i>Escherichia coli</i> K12
1325373	+	61	<i>S. enterica</i> ser. Typhi (~ 1x10 ⁴ CFU/mL)
1325374	++	61	<i>S. enterica</i> ser. Typhi (~ 1x10 ⁵ CFU/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=11</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325371</i>	<i>1325372</i>	<i>1325373</i>	<i>1325374</i>		<i>1325371</i>	<i>1325372</i>	<i>1325373</i>	<i>1325374</i>
<i>Befund Result</i>									
Positiv	11	0	10	10	n.d.	0	0	0	0
Negativ	0	11	1	1	nein <i>no</i>	11	11	11	11
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number *)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
Foodproof <i>Salmonella</i> kit [21] (n=1)	3	3/3	100	1	1/1	100
Other commercial tests [27] (n=2)	6	6/6	100	2	2/2	100
<i>In house</i> PCR assay [28] (n=8)	22	22/24	92	8	8/8	100
Andere/k.A./other [29] (n=1)	3	3/3	100	1	1/1	100

PCR-/NAT *Listeria spp.*
(RV 538) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325381	++	61 /71	<i>Listeria monocytogenes</i> (~ 1x10 ⁵ CFU/mL)
1325382	+	61 /71	<i>Listeria monocytogenes</i> (~ 1x10 ⁴ CFU/mL)
1325383	(+)	61 /71	<i>Listeria monocytogenes</i> (~ 1x10 ³ CFU/mL)
1325384	∅	62	<i>Escherichia coli</i> K12

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=25</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	1325381	1325382	1325383	1325384		1325381	1325382	1325383	1325384
<i>Befund Result</i>									
Positiv	23	23	23	1	n.d.	0	0	0	0
Negativ	2	2	2 ¹⁾	24	nein no	25	25	25	25
Fraglich <i>Questionable</i>	0	0	0	0	ja yes	0	0	0	0

Comments:

¹⁾ As sample #1325383 contained a low number of *Listeria monocytogenes* target organisms, negative PCR results were not rated "false negative", but the participants may consider to improve the analytical sensitivity of corresponding PCR assays.

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number *)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
TIB Molbiol LightMix LM [21] (n=2)	6	6/6	100	2	2/2	100
Ingenetix BactoReal LM [22] (n=2)	6	6/6	100	2	2/2	100
Other commercial tests [27] (n=3)	6	6/9	67	3	3/3	100
<i>In house</i> PCR assay [28] (n=18)	51	51/54	94	17	17/18	94

PCR-/NAT MRSA/cMRSA

(RV 539) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325391	∅	62/72,76	MSSA + CoNS (<i>S. aureus</i> , CoNS, oxa ^R , PVL-neg) (~ 1x10 ⁴ CFU/mL)
1325392	++	61/72	MRSA (<i>S. aureus</i> , oxa ^R , PVL-neg) (~ 1x10 ³ CFU/mL)
1325393	+++	61/71,72	cMRSA (<i>S. aureus</i> , oxa ^R , PVL-pos, spa:t 310) (~ 1x10 ⁴ CFU/mL)
1325394	∅	62	<i>Escherichia coli</i> K12

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=241</i>	<i>Probennummer (Sample no.)</i>				<i>Inhibition</i>				
<i>Befund Result</i>	<i>1325391</i>	<i>1325392</i>	<i>1325393</i>	<i>1325394</i>	<i>1325391</i>	<i>1325392</i>	<i>1325393</i>	<i>1325394</i>	<i>1325394</i>
Positiv	16	237	241 ¹⁾	1	n.d.	2	2	2	2
Negativ	214	4	0	240	nein no	239	239	239	239
Fraglich Questionable	11 ²⁾	0	0	0	ja yes	0	0	0	0

Comments:

- ¹⁾ A dedicated cMRSA identification (*lukFS* gene detection) was performed by 57 laboratories, but obviously only positive results (if observed) were reported. All reported results for cMRSA were correct.
- ²⁾ Bei Teilnehmern, die die Verwendung eines Testsystems aufgeführt haben, das auf einer getrennten Erfassung von *S. aureus*-spezifischen Markern und dem *mecA* Gen beruht, wurden die als „fraglich“ klassifizierten Ergebnisse bei der Erteilung der Zertifikate als „richtig“ bewertet.
For participants who indicated the use of assay concepts for the independent detection of the mecA gene and a S. aureus species marker gene, "questionable" results were accepted in the course of issuing the official QC certificates.

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
BD GeneOhm MRSA [20] (n=20)	40	40/40	100	40	40/40	100
GT MRSA Direct/GQ MRSA [21] (n=30)	60	60/60	100	58	58/60	97
Hyplex <i>StaphyloResist</i> [22] (n=4)	8	8/8	100	4	4/5 [§]	80
LightCycler Kits [23] (n=1)	2	2/2	100	1	1/1 [§]	100
Cepheid Xpert/GeneXpert [24] (n=82)	164	164/164	100	161	161/161 [§]	100
LC MRSA Advanced (Roche) [25] (n=31)	62	62/62	100	61	61/61 [§]	100
TIB Molbiol LightMix MRSA [26] (n=1)	2	2/2	100	2	2/2	100
Commercial assay kit [27] (n=35)	69	69/70	99	66	66/69 [§]	96
<i>In house</i> PCR assay [28] (n=36)	70	70/72	97	58	58/68 [§]	85
Andere/k.A./other [29] (n=16)	31	31/32	97	31	31/32	97

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.

Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants.

§ Due to reporting questionable results, the number of true results (denominator in the „relative“ column) has been reduced.

PCR-/NAT *Chlamydia pneumoniae* (RV 540) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325401	+	61	<i>Chlamydia pneumoniae</i> (~ 1x10 ⁴ IFU/mL)
1325402	∅	62	<i>Mycoplasma pneumoniae</i> (~ 10 ⁵ genome copies/mL)
1325403	∅	62	<i>Escherichia coli</i> K12
1325404	++	61	<i>Chlamydia pneumoniae</i> (~ 1x10 ⁵ IFU/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=94</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325401</i>	<i>1325402</i>	<i>1325403</i>	<i>1325404</i>		<i>1325401</i>	<i>1325402</i>	<i>1325403</i>	<i>1325404</i>
Befund <i>Result</i>									
Positiv	94	1	1	93	n.d.	0	0	0	0
Negativ	0	93	93	1	nein <i>no</i>	94	94	94	94
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.
Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
TIB Molbiol LightMix CP [21] (n=12)	23	23/24	96	23	23/24	96
Diagenode MP/CP [22] (n=6)	12	12/12	100	11	11/12	92
AmpliGnost CP PCR Kit [23] (n=2)	4	4/4	100	4	4/4	100
Other commercial tests [27] (n=32)	64	64/64	100	64	64/64	100
<i>In house</i> PCR assay [28] (n=43)	86	86/86	100	86	86/86	100

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.
Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants.

PCR-/NAT *Mycoplasma pneumoniae* (RV 541) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325411	∅	62	<i>Mycoplasma genitalium</i> (~ 10 ⁴ genome copies/mL)
1325412	++	61	<i>Mycoplasma pneumoniae</i> (~ 10 ⁵ genome copies/mL)
1325413	+	61	<i>Mycoplasma pneumoniae</i> (~ 10 ⁴ genome copies/mL)
1325414	∅	62	<i>Mycoplasma genitalium</i> (~ 10 ⁵ genome copies/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=104</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325411</i>	<i>1325412</i>	<i>1325413</i>	<i>1325414</i>		<i>1325411</i>	<i>1325412</i>	<i>1325413</i>	<i>1325414</i>
Befund <i>Result</i>									
Positiv	4	103	94	4	n.d.	0	0	0	0
Negativ	100	1	10	100	nein <i>no</i>	104	104	104	104
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
LightMix <i>M.pneumoniae</i> [20] (n=10)	19	19/20	95	20	20/20	100
Minerva Venor Mp [22] (n=1)	2	2/2	100	2	2/2	100
AmpliGnost MP PCR Kit [23] (n=1)	2	2/2	100	2	2/2	100
Diagenode MP/CP [24] (n=6)	12	12/12	100	12	12/12	100
Commercial assay/kit [27] (n=34)	64	64/68	94	66	66/68	97
<i>In house</i> PCR assay [28] (n=52)	99	99/104	95	98	98/104	94
Andere/k.A./other [29] (n=1)	1	1/2	50	2	2/2	100

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.

Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants.

PCR-/NAT *C. burnetii* & *B. anthracis* (RV 542) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325421	Ø/Ø	64	<i>Escherichia coli</i> K12
1325422	+ / ++	62	<i>Coxiella burnetii</i> (~ 1x10 ³ genome copies/mL) <i>Bacillus anthracis</i> (~ 5x10 ⁴ genome copies/mL)
1325423	Ø / +	63	<i>Bacillus anthracis</i> (~ 1x10 ⁴ genome copies/mL)
1325424	++ / Ø	61	<i>Coxiella burnetii</i> (~ 1x10 ⁴ genome copies/mL)

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für *Coxiella burnetii* dargestellt.

Absolute numbers of reported individual results. Note: only the C. burnetii-specific results are depicted in this table

<i>n=20</i>	<i>Probennummer (Sample no.)</i>				<i>Inhibition</i>				
	1325421	1325422	1325423	1325424	1325421	1325422	1325423	1325424	
<i>Befund Result</i>									
Positiv	0	20	0	20	n.d.	0	0	0	0
Negativ	20	0	20	0	nein no	20	20	20	20
Fraglich <i>Questionable</i>	0	0	0	0	ja yes	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für *Coxiella burnetii* dargestellt.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

Note: only the C. burnetii-specific results are depicted in this table.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
LightMix <i>C. burnetii</i> [20] (n=2)	4	4/4	100	4	4/4	100
<i>In house</i> PCR assay [28] (n=17)	34	34/34	100	34	34/34	100
Andere/k.A./other [29] (n=1)	2	2/2	100	2	2/2	100

Tabelle 4: Häufigkeit der Mitteilung verschiedener Befunde. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für ***Bacillus anthracis*** dargestellt.

Absolute numbers of reported individual results. Note: only the ***B. anthracis***-specific results are depicted in this table.

n=14	Probennummer (Sample no.)					Inhibition			
	1325421	1325422	1325423	1325424		1325421	1325422	1325423	1325424
Befund Result									
Positiv	0	14	14	0	n.d.	0	0	0	0
Negativ	14	0	0	14	nein no	14	14	14	14
Fraglich Questionable	0	0	0	0	ja yes	0	0	0	0

Tabelle 5: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden. Anmerkung: in dieser Tabelle sind nur die Ergebnisse für ***Bacillus anthracis*** dargestellt.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods. Note: only the ***B. anthracis***-specific results are depicted in this table.

NAT-Methode [Code] (total number*)	NAT richtig positiv True positive results			NAT richtig negativ True negative results		
	Absolut Absolute	Relativ Relative	%	Absolut Absolute	Relativ Relative	%
LightMix <i>B. anthracis</i> [21] (n=3)	6	6/6	100	6	6/6	100
In house PCR assay [28] (n=10)	20	20/20	100	20	20/20	100
Andere/k.A./other [29] (n=1)	2	2/2	100	2	2/2	100

Comments:

¹⁾ Nine of the 23 participants performed only a *Coxiella burnetii* detection, three of the 23 participants performed only *Bacillus anthracis*-specific assays, whereas the other eleven laboratories detected both species.

PCR-/NAT *Francisella tularensis* (RV 543) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325431	+++	61	<i>Francisella tularensis holarctica</i> (~ 1x10 ⁶ CFU/mL)
1325432	∅	62	<i>Escherichia coli</i> K12
1325433	++	61	<i>Francisella tularensis holarctica</i> (~ 1x10 ⁵ CFU/mL)
1325434	++/ ∅*	61	<i>Francisella tularensis novicida</i> (~ 1x10 ⁵ CFU/mL)

* siehe Ergebnisdiskussion/see Discussion section

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=15</i>	<i>Probennummer (Sample no.)</i>				<i>Inhibition</i>				
	<i>1325431</i>	<i>1325432</i>	<i>1325433</i>	<i>1325434</i>	<i>1325431</i>	<i>1325432</i>	<i>1325433</i>	<i>1325434</i>	
<i>Befund Result</i>									
Positiv	15	0	15	14	n.d.	0	0	0	0
Negativ	0	15	0	1*	nein no	15	15	15	15
Fraglich <i>Questionable</i>	0	0	0	0	ja yes	0	0	0	0

* siehe Ergebnisdiskussion/see Discussion section

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number*)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
LightMix <i>F. tularensis</i> [20] (n=4)	12	12/12	100	4	4/4	100
Commercial assay/kit [27] (n=1)	3	3/3	100	1	1/1	100
In house PCR assay [28] (n=12)	35	35/36	97	12	12/12	100

* Durch Mehrfachnennung oder fehlende Angabe kann die absolute Zahl der Ergebnisse (Tab. 2) von der Anzahl der Teilnehmer abweichen.
Due to reporting results of multiple assay systems or missing specifications, the effective numbers are not correlating with the number of participants.

PCR-/NAT *Pneumocystis jirovecii* (RV 560) November 2013

Tabelle 1: Probenzusammensetzung und erwartetes Ergebnis.
Sample composition and expected results.

	<i>Erwartet/expected</i>		<i>Probenzusammensetzung/Sample composition</i>
1325601	++	61	<i>Pneumocystis jirovecii</i> (~ 3x10 ⁵ genome copies/mL)
1325602	∅	62	<i>Escherichia coli</i> K12
1325603	+	61	<i>Pneumocystis jirovecii</i> (~ 9x10 ⁴ genome copies/mL)
1325604	∅	62	<i>Escherichia coli</i> K12

Tabelle 2: Häufigkeit der Mitteilung verschiedener Befunde.
Absolute numbers of reported individual results.

<i>n=59</i>	<i>Probennummer (Sample no.)</i>					<i>Inhibition</i>			
	<i>1325601</i>	<i>1325602</i>	<i>1325603</i>	<i>1325604</i>		<i>1325601</i>	<i>1325602</i>	<i>1325603</i>	<i>1325604</i>
Befund <i>Result</i>									
Positiv	58	0	59	0	n.d.	0	0	0	0
Negativ	1	59	0	59	nein <i>no</i>	59	59	59	59
Fraglich <i>Questionable</i>	0	0	0	0	ja <i>yes</i>	0	0	0	0

Tabelle 3: Häufigkeit richtig positiver und richtig negativer NAT-Befunde bei Anwendern verschiedener Methoden.

Absolute numbers and relative frequency of reported true positive and true negative results among various NAT methods.

NAT-Methode [Code] (total number *)	NAT richtig positiv <i>True positive results</i>			NAT richtig negativ <i>True negative results</i>		
	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%	Absolut <i>Absolute</i>	Relativ <i>Relative</i>	%
Myconostica MycAssay [20] (n=1)	1	1/2	50	2	2/2	100
TIB Molbiol LightMix PJ [21] (n=7)	14	14/14	100	14	14/14	100
AmpliGnost PJ PCR Kit [22] (n=5)	10	10/10	100	10	10/10	100
Sacace PJ RealTM [23] (n=1)	2	2/2	100	2	2/2	100
Commercial assay/kit [27] (n=11)	22	22/22	100	22	22/22	100
<i>In house</i> PCR assay [28] (n=33)	66	66/66	100	66	66/66	100
Andere/k.A./other [29] (n=1)	2	2/2	100	2	2/2	100