The External Quality Assurance System of the WHO Global Foodborne Infections Network, 2014

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CDC
Centers for Disease Control and Prevention

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Institut Pasteur

DTU Food
National Food Institute

THE EXTERNAL QUALITY ASSURANCE SYSTEM OF THE WHO GLOBAL FOODBORNE INFECTIONS NETWORK YEAR 2014

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List of Abbreviations

AMP, Ampicillin
AST, Antimicrobial Susceptibility Testing
ATCC, American Type Culture Collection
CAZ, Ceftazidime
CDC, Centers for Disease Control and Prevention
CHL, Chloramphenicol
CIP, Ciprofloxacin
CRO, Ceftriaxone
CTX, Cefotaxime
DTU Food, Technical University of Denmark - National Food Institute
EQAS, External Quality Assurance System
ERY, Erythromycin
ESBL, Extended Spectrum Beta-Lactamase
EU, European Union
GEN, Gentamicin
IP, Institute Pasteur
MIC, Minimum Inhibitory Concentration
NAL, Nalidixic Acid
NSSC, National *Salmonella* and *Shigella* Center, Thailand
PHAC, Public Health Agency of Canada
QC, Quality Control
SMX, Sulfamethoxazole
STR, Streptomycin
SXT, Trimethoprim + Sulphonamides
TET, Tetracycline
TMP, Trimethoprim
WHO, World Health Organization
WHO GFN, WHO Global Foodborne Infections Network

1. Introduction

Since 2000, 13 External Quality Assurance System (EQAS) reports have been issued with this report being the 14th. The WHO Global Foodborne Infections Network (WHO GFN) focuses on enhancing World Health Organization (WHO) Member States' capacity to detect and respond to foodborne disease outbreaks by conducting laboratory-based surveillance of *Salmonella* and other foodborne pathogens. Since its inception, the scope of WHO GFN has expanded to include additional foodborne pathogens like *Shigella* and *Campylobacter*. *Salmonella*, *Campylobacter* and *Shigella* are among the most important foodborne pathogens worldwide and account for millions of cases of diarrheal disease and thousands of deaths per year, impacting both developing and industrialized countries. Furthermore, the increased number of *Salmonella* and *Shigella* isolates which are resistant to antimicrobials is of major concern since these isolates are associated with infections characterized by increased morbidity and mortality.

The EQAS is organized annually by the Technical University of Denmark, National Food Institute (DTU Food), Kgs. Lyngby, Denmark in collaboration with Centers for Disease Control and Prevention (CDC) in Atlanta, USA; World Health Organization (WHO) in Geneva, Switzerland; Public Health Agency of Canada (PHAC) in Canada; National *Salmonella* and *Shigella* Center (NSSC), National Institute of Health, Department of Medical Science in Thailand and Institute Pasteur (IP) in Paris, France.

Individual laboratory data are confidential and only known by the participating laboratory, the EQAS Organizer (DTU Food) and possibly the respective WHO GFN regional centre. All summary conclusions are made public. The goal set by WHO GFN aims towards having all national reference laboratories perform *Salmonella* serotyping with a maximum of one deviation out of eight strains tested (error rate of 13%) and performing antimicrobial susceptibility testing (AST) of *Salmonella* and *Shigella* with a maximum error rate of 10% (either <5% very major / major errors and <5% minor errors, or <10% minor errors). Minor deviations are defined as classification of an intermediate strain as susceptible, resistant or vice versa (*i.e.* I $\leftrightarrow$ S or I $\leftrightarrow$R). Major deviation is the classification of a susceptible strain as resistant (*i.e.* S $\rightarrow$ R). Very major deviation is the classification of a resistant strain as susceptible (*i.e.* R $\rightarrow$ S). In this report, the deviations of AST results are divided into two categories, *i.e.* critical deviations which include major and very major deviations, and total deviations which include also the minor deviations. No quality threshold has been determined in relation to identification of *Campylobacter* ssp., serotyping of *Shigella*, or identification of the unknown foodborne pathogen.

In EQAS 2014, the regions were redefined for all countries worldwide. This lead to some reorganization of countries into new regions compared to previous years, why interpreting region based results from 2014 and forward with results before 2014 should be done with care. The countries belonging to each region is listed in Appendix 1.

Appendices 2-5 present additional background information in relation to the WHO EQAS 2014.

2. Summary

The summary report is divided into five sections; the *Salmonella* components, the *Shigella* components, reporting of ESBL *Salmonella* and *Shigella*, the *Campylobacter* components, and identification of the unknown strain. All results reported in the summary can be found in Appendix 1.

***Salmonella* EQAS components**

The acceptance threshold for the EQAS *Salmonella* serotyping component was met by 68% (n = 102) of the 149 participating laboratories. In addition, 57% (n = 85) of the laboratories tested all eight strains with a total at 92% (n = 969) of all tests being correct, representing an increase compared to 2013. The ability to correctly serotype the internal control strain increased to the highest level at 98%, previously only observed in 2011.

On a region-based categorization of participating laboratories, the Caribbean, Central Asia & Middle East, Africa and Latin America all correctly serotyped between 60% and 90% of the test strains, where as China, Russia, Southeast Asia and Europe correctly serotyped between 90 and 99% of the test strains. North America and Oceania both serotyped all eight strains 100% correctly.

The main problem regarding the *Salmonella* serotyping appeared to be with WHO S-14.1 (Orion/Orion var. 15; 3,15:y:1,5), WHO S-14.4 (Napoli; 9,12:l,z13:e,n,x), and WHO S-14.5 (Ohio; 6,7:b:l,w), with 10%, 17% and 14% deviation, respectively.

Concerning the *Salmonella* AST component for the EQAS 2014, the performance recorded was maintained on a similar level as in the EQAS 2013, with low deviations of 3% minor, 1% major, and 1% very major deviations. Deviations categorized by the tested antimicrobials revealed that CIP caused the difficulties of the observed deviations (19%) most likely due to the often observed double zone when performing disk diffusion.

For the 155 laboratories performing the *Salmonella* AST component, only 74% (115 laboratories) reported data for AST of the control strain *E. coli* ATCC 25922. This is an alerting decrease, and it is of extreme importance to once again emphasize that this component represents the true indicator for the laboratory as to the performance of AST.

***Shigella* EQAS components**

The *Shigella* components included in the EQAS consist of serogrouping (i.e. the identification of the species), serotyping (i.e. the further typing of the species), and AST.

For the *Shigella* serogrouping component in EQAS 2014, the deviations observed ranged from 2.4% to 5.6%, for the four *Shigella* strains. This is an increase compared to the very low level of deviations observed in EQAS 2013, with a maximum of 0.9%.

The serotyping component was performed by a total of 83 laboratories for the two strains WHO 2014 SH-14.3 (*S. flexneri*; 2/2b) and WHO 2014 SH-14.4 (*S. boydii*; 2), with deviating results observed at 16.3% and 7.2%, respectively. The serotyping component was not required for the *S. sonnei* serogroup (WHO 2014 SH-14.1 and WHO 2014 SH-14.2). According to the geographical distribution of the participating laboratories, this year Caribbean was again represented. However,

the one laboratory representing the region did not manage to correctly serotype the one strain that they tested. The remaining results, on a region-based categorization, ranged from 58.3% (Africa) to 100% correctly serotyped strains.

For the results of the *Shigella* AST component, the number of participating laboratories increased to the levels before the EQAS 2013, with 116 participating laboratories in EQAS 2014. The results obtained were in 92% of the cases in agreement with the expected results and consistent with previous years. Minor, major and very major deviations were observed in 4%, 1%, and 3% of the reported results, respectively. Categorizing the tested antimicrobials according to the deviations revealed that CIP (34.2%) and CAZ (14.1%) caused difficulties in the AST component.

A region-based categorization of the results revealed correct test results between 76.5% (Caribbean) and 98.4% (Russia), with a very high level of critical deviations observed in Caribbean (18.4%), with the remaining regions all below 10%.

ESBL EQAS component

A part of the EQAS is to detect and confirm ESBL production in the *Salmonella* and *Shigella* strains. If participating in this component of the EQAS, all strains showing reduced susceptibility to cefotaxime (CTX), ceftazidime (CAZ) and/or ceftriaxone (CRO) should be tested for ESBL production.

For the EQAS 2014, one *Salmonella* ESBL-producer (WHO 2014 S-14.7) and two *Shigella* ESBL-producers (WHO 2014 SH-14.1 and WHO 2014 SH-14.2) were included. For the *Salmonella* strain, the gene accounting for the phenotype was CTX-M-3, and the two confirmatory tests (CAZ/Cl:CAZ and CTX/Cl:CTX) showed 11% and 13% of deviations in reporting correct results, respectively. For the *Shigella* strain WHO 2014 SH-14.1 (CTX-M-15), deviations of the confirmatory test results were observed to 9% and 8%, respectively. For the WHO 2014 SH-14.2 (CTX-M-15), the deviating results were observed to 12% and 8%, respectively.

***Campylobacter* EQAS components**

Interpretation of the results for *Campylobacter* in EQAS 2014 only included WHO 2014 C-14.2 due to the fact that the produced lyophilized WHO 2014 C-14.1 proved to be contaminated and therefore could not be distributed.

A total of 101 laboratories participated in the identification of the *C. coli* WHO 2014 C-14.2 strain with a result of 85% correct species identification. On a region-based characterization, the accuracy in *Campylobacter* identification ranged from 57% (Central Asia & Middle East) to 100% (Caribbean, North America, Oceania, and Russian regions).

Concerning the *Campylobacter* AST component in the EQAS 2014, 50 laboratories participated. The overall performance of the AST showed 1.6% major deviations, and 7.2% very major deviations, giving a total of 8.8% critical deviations, the highest level observed in the history of this WHO EQAS.

From the categorization of the antimicrobials, the results showed no problems when testing ERY and GEN. However, NAL, STR, and TET all showed deviations above 10% (11.9%, 16.7%, and

11.1%, respectively). On a region-based characterization, the performance in Africa is noteworthy, with a deviation level of 48.5% critical deviations, whereas Central Asia & Middle East, China, Caribbean, North America, and Latin America all perfectly performed the test without deviations. Europe and Southeast Asia reported deviations at 2.6% and 12.5%, respectively. In EQAS 2014 no laboratories in the Oceania or Russian regions participated in the *Campylobacter* AST component.

For the QC strain *Campylobacter jejuni* ATCC 33560 only 32 laboratories reported AST. Again, we have to emphasize the importance of including this component as it represents the true indicator for the laboratory's performance of AST. In EQAS 2014, the antimicrobials causing most problems were GEN and ERY, however the percentage of laboratories reporting correct AST results for these two compounds increased to 90% and 84% (compared to 82% and 83% in EQAS 2013), respectively.

Identification of unknown culture EQAS component

For this part of the EQAS, an unknown culture is provided for identification. In EQAS 2014, the unknown strain was the Gram negative *Yersinia pseudotuberculosis*.

A total of 122 laboratories participated in this component, with 74% identifying the strain correctly.

3. List of Appendices

Appendix 1: Figures and Tables

Appendix 2: Prenotification

Appendix 3: Expected results

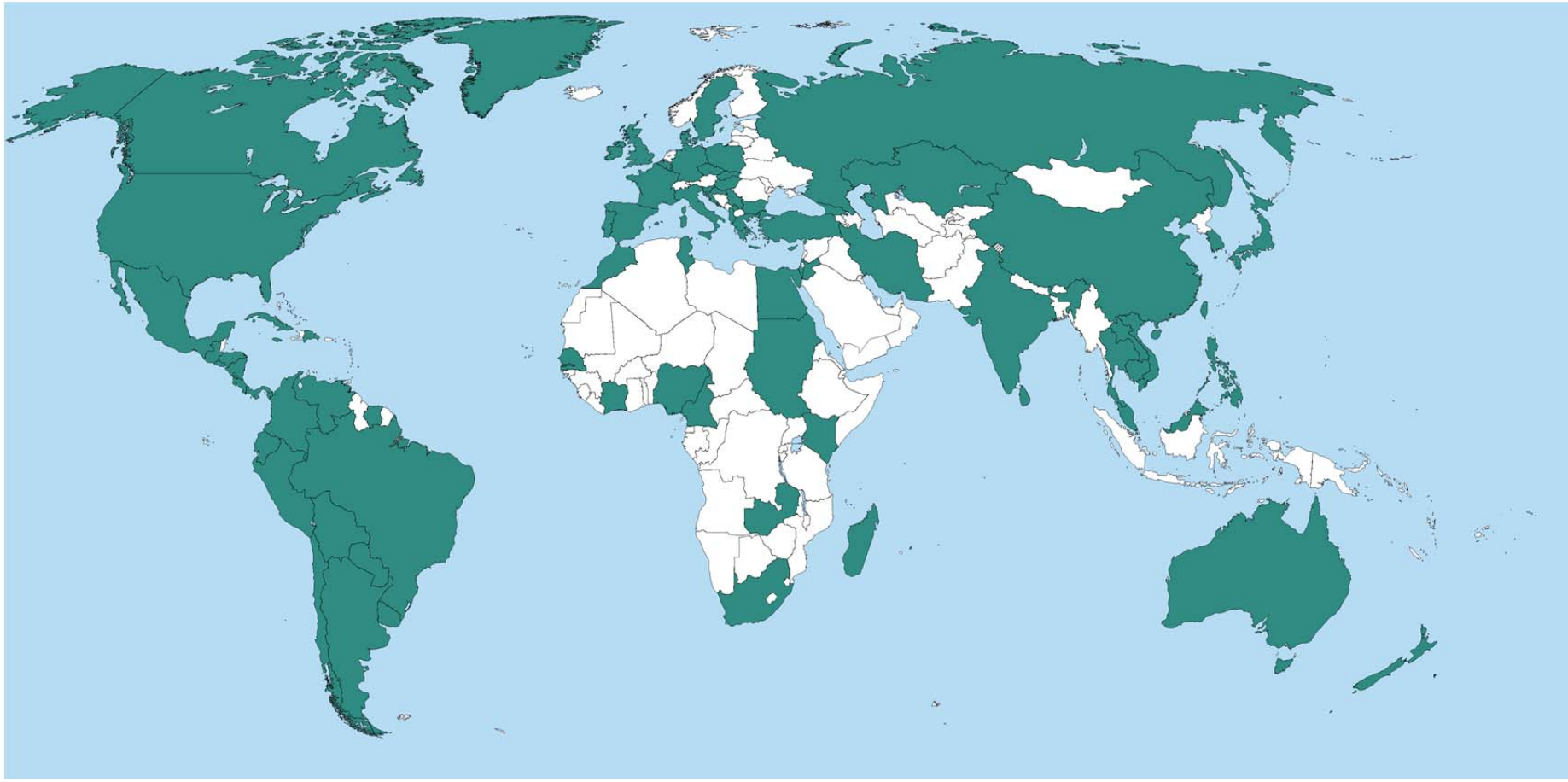
Appendix 4: WHO EQAS 2014 Protocol

Appendix 5a: Subculture and Maintenance of Quality Control Strains

Appendix 5b: Instructions for Opening and Reviving Lyophilized Cultures

Figure and Tables

Figure 1. Countries participating* in the WHO EQAS 2014



*marked in green

List of Countries in the 10 Regions

Africa

Algeria
Angola
Benin
Botswana
Burkina Faso
Burundi
Cameroon
Cameroun
Cape Verde
Central African Republic
Chad
Comoros
Congo (Brazzaville)
Congo, Democratic Republic of the
Cote d'Ivoire (Ivory Coast)
Djibouti
Egypt
Equatorial Guinea
Eritrea
Ethiopia
Gabon
Gambia
Ghana
Guinea
Guinea-Bissau
Kenya
Lesotho
Liberia
Libyan Arab Jamahiriya
Madagascar
Malawi
Mali
Mauritania
Mauritius
Mayotte
Morroco
Mozambique
Namibia
Niger
Nigeria
Reunion
Rwanda
Saint Helena
Sao Tome and Principe
Senegal
Seychelles
Sierra Leone
Somalia
South Africa
South Sudan
Sudan
Swaziland
Tanzania, United Republic of
Togo
Tunisia
Uganda
Western Sahara
Zambia
Zimbabwe

Caribbean

Anguilla
Antigua and Barbuda
Aruba
Bahamas
Barbados
Bonaire, Saint Eustatius and Saba
British Virgin Islands
Cayman Islands
Cuba
Curaçao
Dominica
Dominican Republic
Grenada
Guadeloupe
Haiti
Jamaica
Martinique
Monserrat
Puerto Rico
Saint Lucia
Saint Martin
Saint Vincent and the Grenadines
Saint-Barthélemy
Sint Maarten
St. Kitts and Nevis
Trinidad and Tobago
Turks and Caicos Islands
Virgin Islands (US)

Central Asia & Middle East

Afganistan
Armenia
Azerbaijan
Bahrain
Bangladesh
Bhutan
Georgia
Hong Kong
India
Indonesia
Iran, Islamic rep. Of
Iraq
Israel
Jordan
Kazakhstan
Kuwait
Kyrgyzstan
Lebanon
Macao
Maldives
Mongolia
Myanmar (ex-Burma)
Nepal
Oman
Pakistan
Palestine
Qatar
Saudi Arabia
Syria
Tajikistan
Timor Leste (West)
Turkmenistan
United Arab Emirates
Uzbekistan
Yemen

China

China

Europe

Albania
Andorra
Austria
Guerney and Alderney
Hungary
Iceland
Norway
Poland
Portugal

Belarus
Belgium
Bosnia
Bulgaria
Croatia
Cyprus
Czech Republic
Denmark
Estonia
European Union
Faroe Islands
Finland
France
Germany
Gibraltar
Greece
Ireland
Italy
Jersey
Kosova
Kosovo
Latvia
Liechtenstein
Lithuania
Luxembourg
Macedonia
Malta
Man, Island of
Moldova
Monaco
Montenegro
Netherlands
Romania
San Marino
Serbia
Slovak Republic
Slovakia
Slovenia
Spain
Svalbard and Jan Mayen Islands
Sweden
Switzerland
Turkey
Ukraine
United Kingdom
Vatican City State (Holy See)

Latin America

Argentina
Bolivia
Brazil
Chile
Colombia
Costa Rica
Ecuador
El Salvador
Falkland Islands (Malvinas)
French Guiana
Guatemala
Guyana
Honduras
Mexico
Nicaragua
Panama
Paraguay
Peru
Suriname
Uruguay
Venezuela

North America

Bermuda
Canada
Greenland
Saint Pierre and Miquelon
United States of America

Oceania

Australia
Kiribati
New Zealand
Solomon, Islands
Fiji
Marshall Islands
Papua New Guinea
Tonga
French Polynesia
Micronesia
Samoa
Tuvalu
Guam
New Caledonia
Samoa, American
Vanuatu

Russia

Russia

Southeast Asia

Brunei Darussalam
Cambodia
Japan
Korea, North
Korea, Rep of
Lao PDR
Malaysia
Philippines
Singapore
Sri Lanka
Taiwan
Thailand
Viet Nam

Table 1. EQAS participating laboratories' performance of *Salmonella* serotyping

EQAS iteration	Labs serotyping all provided strains		Correct test results	
	No.	%	No.	%
2000	34	92	165	76
2001	79	82	513	72
2002	80	81	668	91
2003	69	54	692	80
2004	78	61	701	81
2006	105	81	808	85
2007	109	78	920	88
2008	100	66	888	83
2009	119	83	974	86
2010	129	87	998	89
2011	109	89	878	92
2012	122	81	936	83
2013	74	59	812	89
2014	85	57	969	92
Average	97	75	780	85

Table 2. Ability of EQAS participating laboratories to serotype the test *Salmonella* strains

Number of strains correctly serotyped	Participating laboratories													
	EQAS 2000		EQAS 2001		EQAS 2002		EQAS 2003		EQAS 2004		EQAS 2006		EQAS 2007	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
8	9	24	34	35	52	53	66	47	41	32	42	32	66	47
7	9	24	13	14	19	19	29	21	14	11	35	27	29	21
6	4	11	9	9	12	12	13	9	16	13	19	15	13	9
5	3	8	9	9	4	4	11	8	16	13	12	9	11	8
4	3	8	4	4	1	1	7	5	11	9	7	5	7	5
3	4	11	8	8	4	4	6	4	10	8	5	4	6	4
2	2	5	3	3	5	5	2	1	10	8	3	2	2	1
1	2	5	5	5	1	1	6	4	5	4	4	3	6	4
0	1	3	11	11	1	1	0	0	4	3	3	2	0	0
In total	37	100	96	100	99	100	127	100	127	100	130	100	140	100

Number of strains correctly serotyped	Participating laboratories															
	EQAS 2008		EQAS 2009		EQAS 2010		EQAS 2011		EQAS 2012		EQAS 2013		EQAS 2014		AVERAGE EQAS 2000 - 2014	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
8	50	33	76	50	91	61	82	67	68	47	52	41	70	47	57	44
7	36	24	29	19	16	11	17	14	29	20	29	23	32	21	24	19
6	11	7	7	5	12	8	10	8	14	10	15	12	17	11	12	10
5	14	9	13	8	9	6	2	2	9	6	8	6	6	4	9	7
4	12	8	5	3	6	5	4	3	5	3	7	6	5	3	6	5
3	9	6	7	5	2	1	4	3	6	4	7	6	7	5	6	5
2	8	6	5	3	2	1	1	1	10	7	6	5	4	3	5	4
1	9	6	6	4	7	5	3	2	2	1	2	2	4	3	4	4
0	2	1	5	3	3	2	0	0	1	1	0	0	4	3	3	2
In total	151	100	153	100	148	100	123	100	144	100	126	100	149	100	125	100

Table 3. Region-based categorization of EQAS participants' performance of *Salmonella* serotyping

Region	EQAS iteration	No. of labs	No. of strains serotyped	% strains correctly serotyped	Countries participating in EQAS 2014
Africa	2001	6	37	73.0	Cameroun, Egypt, Gambia, Kenya (2), Madagascar, Mauritius, Morroco, Senegal, South Africa, Tunisia
	2002	9	62	87.1	
	2003	11	70	71.4	
	2004	9	51	62.7	
	2006	16	95	71.6	
	2007	11	73	80.8	
	2008	10	71	49.3	
	2009	15	94	75.5	
	2010	13	83	67.5	
	2011	10	57	79.2	
	2012	10	65	60.0	
	2013	8	51	74.5	
	2014	11	63	76.2	
Central Asia & Middle East	2001	10	60	50.0	Bahrain, Georgia, India (2), Israel, Jordan, Kazakhstan
	2002	5	30	83.3	
	2003	5	35	54.3	
	2004	5	33	54.5	
	2006	5	35	74.3	
	2007	5	40	55.0	
	2008	5	34	61.8	
	2009	5	32	46.9	
	2010	5	22	75.9	
	2011	3	23	95.8	
	2012	4	30	56.7	
	2013	5	38	52.6	
	2014	7	37	75.7	
Caribbean	2001	0	0	0	Barbados, Cuba, Jamaica
	2002	0	0	0	
	2003	3	18	61.1	
	2004	2	8	87.5	
	2006	3	14	78.6	
	2007	2	9	77.8	
	2008	3	14	78.6	
	2009	3	12	83.3	
	2010	2	13	92.9	
	2011	1	7	87.5	
	2012	2	16	62.5	
	2013	1	5	100.0	
	2014	3	15	60.0	
Europe	2001	43	323	80.5	Albania, Belgium, Bulgaria, Croatia (2), Cyprus, Czech Republic (2), Denmark (2), France, Germany (2), Greece (3), Hungary, Ireland, Italy (18), Kosova, Luxembourg (2), Malta, Poland (3), Portugal, Serbia (2), Slovak Republic, Slovenia, Spain, Sweden, Turkey, United Kingdom
	2002	50	384	90.0	
	2003	60	401	84.8	
	2004	57	392	84.7	
	2006	52	403	86.4	
	2007	54	415	89.4	
	2008	50	379	82.3	
	2009	47	362	93.1	
	2010	45	332	94.1	
	2011	42	314	94.6	
	2012	47	368	92.9	
	2013	42	309	94.5	
	2014	52	391	96.2	

Table 3 (continued). Region-based categorization of EQAS participants' performance of *Salmonella* serotyping

Region	EQAS iteration	No. of labs	No. of strains serotyped	% strains correctly serotyped	Countries participating in EQAS 2014
North America	2001	4	32	87.5	Canada (10), USA (3)
	2002	2	16	100.0	
	2003	6	41	95.1	
	2004	8	55	81.8	
	2006	10	80	96.3	
	2007	12	94	97.9	
	2008	11	84	95.2	
	2009	12	90	92.2	
	2010	13	103	100.0	
	2011	11	81	97.6	
	2012	14	101	93.1	
	2013	13	92	97.8	
	2014	13	84	100.0	
Oceania	2001	4	30	100.0	Australia (3), New Zealand
	2002	6	43	93.0	
	2003	6	46	93.5	
	2004	5	38	97.4	
	2006	5	37	94.6	
	2007	4	32	100.0	
	2008	4	30	93.3	
	2009	4	32	96.9	
	2010	4	32	100.0	
	2011	4	32	100.0	
	2012	4	32	100.0	
	2013	4	31	100.0	
	2014	4	32	100.0	
Russia	2001	1	8	12.5	Russia (4)
	2002	1	8	62.5	
	2003	1	7	14.3	
	2004	4	26	69.2	
	2006	5	40	80.0	
	2007	8	51	80.4	
	2008	6	40	90.0	
	2009	7	49	91.8	
	2010	8	54	87.1	
	2011	7	48	87.3	
	2012	6	48	87.5	
	2013	2	16	75.0	
	2014	4	30	93.3	
Latin America	2001	11	78	57.7	Argentina, Bolivia, Brazil (2), Chile (2), Colombia (3), Costa Rica (2), Honduras, Mexico (3), Nicaragua, Panama (2), Paraguay, Peru, Uruguay (2), Venezuela (2)
	2002	11	82	87.8	
	2003	13	83	75.9	
	2004	15	88	79.5	
	2006	13	84	84.5	
	2007	15	107	88.8	
	2008	17	120	71.7	
	2009	21	150	77.3	
	2010	22	132	80.0	
	2011	23	144	83.7	
	2012	25	182	73.1	
	2013	22	154	83.1	
	2014	24	166	84.9	

Table 3 (continued). Region-based categorization of EQAS participants' performance of *Salmonella* serotyping

Region	EQAS iteration	No. of labs	No. of strains serotyped	% strains correctly serotyped	Countries participating in EQAS 2014
Southeast Asia	2001	15	113	54.0	Brunei Darussalam, Cambodia, Japan (2), LAO PDR, Malaysia (4), Philippines, Rep of Korea (2), Singapore, Sri Lanka, Taiwan, Thailand (6), Viet Nam
	2002	12	90	92.2	
	2003	15	100	81.0	
	2004	17	130	81.5	
	2006	15	117	84.6	
	2007	19	140	91.4	
	2008	18	125	81.6	
	2009	23	180	81.1	
	2010	24	172	90.5	
	2011	23	180	98.4	
	2012	28	207	77.8	
	2013	22	163	89.6	
	2014	22	166	94.6	
China	2001	4	32	96.9	China (9)
	2002	3	24	100.0	
	2003	8	60	75.0	
	2004	7	46	78.3	
	2006	6	48	85.4	
	2007	10	80	91.3	
	2008	15	108	94.4	
	2009	16	126	95.2	
	2010	10	74	92.5	
	2012	10	78	80.8	
	2013	7	54	92.6	
	2014	9	71	93.0	

Table 4. *Salmonella* serogroups (SG), serotypes (ST) and deviations (D), WHO EQAS 2014

Strain ID	Correct serotype		No. of labs reporting SG	% D_{SG}	No. of labs reporting ST	% D_{ST}	Deviating results (*)
WHO S-14.1	Orion / Orion var. 15	I 3,15:y:1,5	153	4.6	137	10.2	Amager, Amounderness, Elisabethville, Florian, Fufu, Gatineau(3), Gbadago, II 3,10:z:1,5, Lexington var. 15+, Meleagridis, Paratyphi A, Stockholm
WHO S-14.2	Hadar / Istanbul	I 8:z10:e,n,x	158	3.8	137	6.6	Chomedy, Haifa, Mapo, Molade, Newport, Paratyphi C, Santiago, Virginia, Zerifin
WHO S-14.3	IIIa 48:g,z51:-	IIIa 48:g,z51:-	106	10.4	90	7.8	Choleraesuis, Enteritidis, Fitzroy 48:e,h:1,5, II 48:g,m,t:-, IIIb 48:i:z, IV 48:g,z51:-, IV 6,7:z36:-
WHO S-14.4	Napoli	I 9,12:l,z13:e,n,x	155	3.9	133	16.5	Bournemouth, Claibornei, Dublin, Enteritidis, Fallowfield, II .1.,9,12:b:e,n,x, II .1.,9,12:l,w:e,n,x, Itami, Javiana(2), Kapemba, Mahina, Miyazaki, Nordrhein, Zaiman(8)
WHO S-14.5	Ohio	I 6,7:b:l,w	157	1.9	133	14.3	Bonariensis, Colorado(2), Coromandel, Edinburg, Gabon(2), II 6,7:l,z28:1,5:[z42], Infantis(4), Isangi, Jerusalem, Langeveld(4), Montevideo
WHO S-14.6	Enteritidis	I 9,12:g,m:-	156	0.6	145	2.1	Antarctica, Macclesfield, Panama
WHO S-14.7	Typhimurium	I 4,12:i:1,2	160	1.3	141	4.3	Agama, Farsta, Gloucester(2), Lagos, Tripoli
WHO S-14.8	Kentucky	I 8:i:z6	157	4.5	139	4.3	Agama, Hadar, Newport, Paratyphi A, Stuttgart, Warnow

*number of participants reporting the specified deviating result

Table 5. EQAS participating laboratories' performance of internal quality control strain (WHO S-14.6, *Salmonella* Enteritidis) serotyping

EQAS iteration	Labs serotyping *S.* Enteritidis correctly	
	No.	%
2000	34	92
2001	64	84
2004	113	95
2006	116	94
2007	135	96
2008	139	96
2009	141	93
2010	138	97
2011	128	98
2012	139	96
2013	130	96
2014	145	98
Average	119	95

Table 6. EQAS participating laboratories' performance of antimicrobial susceptibility testing of *Salmonella* strains

EQAS iteration	**No. of EQAS participating laboratories**	**% correct test results**	**% minor deviations** (S ↔ I or I ↔ R)^	**% major deviations** (S → R)^	**% very major deviations** (R→ S)^	**% critical deviations** (R→ S & S → R)^	**% total deviations** (S → R & R → S & S ↔ I or I ↔ R)^
2000	44	92	4	4	0	4	8
2001	108	91	6	2	1	3	9
2002	119	92	6	2	1	3	9
2003*	147	93	4	3	0	3	7
2004	152	93	4	2	1	3	7
2006	143	88	8	3	1	4	12
2007	143	93	4	2	1	3	7
2008	168	91	4	2	3	5	9
2009	153	94	3	2	1	3	6
2010	152	92	4	3	2	5	8
2011	127	91	4	2	3	5	9
2012	159	94	3	2	1	3	6
2013	145	95	3	2	0	2	5
2014	155	95	3	1	1	2	5
Average*	137	93	4	2	1	3	8

***Data do not include one strain which may have lost resistance due to transport or storage stress

^S, susceptible; I, intermediate; R, resistant

Table 7. Antimicrobial susceptibility test results (number of R/I/S) for the EQAS 2014 *Salmonella* strains*

Strain	Antimicrobial^											
	AMP	**CTX**	**CAZ**	**CRO**	**CHL**	**CIP**	**GEN**	**NAL**	**SMX**	**TET**	**SXT**	**TMP**
WHO S-14.1	5/1/**141**	2/1/**125**	3/2/**121**	0/1/**101**	1/0/**133**	1/12/**132**	2/2/**139**	1/1/**132**	4/1/**64**	3/2/**133**	2/1/**127**	0/0/**69**
WHO S-14.2	4/0/**144**	3/1/**123**	1/1/**124**	1/2/**99**	2/1/**131**	21/**80**/44	3/2/**139**	**133**/0/3	8/0/**63**	**133**/3/4	3/0/**128**	0/0/**67**
WHO S-14.3	4/0/**143**	0/0/**126**	0/0/**126**	0/0/**101**	0/0/**133**	0/4/**140**	5/0/**138**	2/0/**133**	1/0/**70**	3/1/**136**	0/1/**128**	0/0/**67**
WHO S-14.4	**144**/0/3	2/1/**124**	3/2/**119**	0/0/**103**	0/0/**135**	3/9/**134**	2/2/**140**	2/2/**131**	**69**/0/0	**135**/1/4	**122**/0/8	**64**/0/2
WHO S-14.5	2/2/**142**	0/0/**126**	3/0/**122**	0/0/**102**	0/2/**132**	0/10/**135**	3/1/**140**	3/1/**130**	5/2/**62**	3/6/**129**	2/0/**128**	0/0/**66**
WHO S-14.6	10/10/**127**	5/5/**117**	8/0/**117**	1/0/**101**	0/7/**128**	0/13/**131**	**134**/4/6	1/3/**132**	**60**/0/9	7/11/**120**	2/0/**127**	0/0/**66**
WHO S-14.7	**145**/0/2	**119**/1/6	**114**/6/5	**100**/0/5	**129**/3/1	**46**/69/30	**136**/3/3	**132**/0/1	**73**/0/0	**135**/0/1	**128**/0/1	**68**/0/2
WHO S-14.8	**144**/0/3	5/1/**121**	7/2/**117**	4/1/**95**	3/0/**131**	**137**/4/6	**120**/11/12	**133**/0/2	**67**/0/3	**133**/1/3	5/1/**125**	5/0/**65**

^For antimicrobial abbreviations: see List of Abbreviations page 1

*In bold: expected interpretation. Grey cell: <90% of laboratories did correct interpretation. R, resistant/I, intermediate/ S, susceptible.

Table 8. EQAS participants' performance of *Salmonella* strains antimicrobial susceptibility testing categorized by antimicrobial

EQAS iteration	No. of labs	Performance	Antimicrobial[∞]																
			AMC	AMP	CAZ	CHL	CIP	POD	CRO	CTX	GEN	KAN	NAL	SMX	STR	SXT	TET	TMP	XNL	OVERALL
2000	44	No. of tests	-	343	-	343	334	-			343	312	328	248	312	-	335	295	-	3,193
		% critical deviations*	-	6	-	4	1	-			4	4	1	3	4	-	6	1	-	3
		% total deviations^	-	8	-	7	6	-			5	16	4	5	12	-	13	1	-	8
2001	108	No. of tests	-	822	-	814	813	-			821	623	726	431	679	757	804	416	-	7,706
		% critical deviations*	-	4	-	2	1	-			2	2	2	6	7	2	7	1	-	3
		% total deviations^	-	7	-	3	4	-			4	7	8	9	27	5	18	2	-	9
2002	119	No. of tests	-	918	-	903	911	-			905	680	885	495	718	724	861	499	-	8,499
		% critical deviations*	-	2	-	2	0	-			2	2	2	4	4	7	3	3	-	3
		% total deviations^	-	3	-	3	2	-			16	10	4	4	34	10	7	3	-	9
2003•	147	No. of tests	-	1,019	-	996	995	-			993	738	947	615	768	929	995	582	-	9,577
		% critical deviations*	-	2	-	1	0	-			2	2	1	4	9	2	4	1	-	3
		% total deviations^	-	4	-	2	1	-			2	6	4	5	39	2	11	1	-	7
2004	152	No. of tests	973	1,178	-	1,159	1,162	-	-	995	1,201	-	1,130	734	947	1051	1,122	729	-	12,381
		% critical deviations*	6	3	-	2	0	-	-	0	2	-	1	5	1	3	5	2	-	3
		% total deviations^	12	5	-	2	1	-	-	14	3	-	4	8	21	4	11	2	-	7
2006	143	No. of tests	950	1,092	769	1,060	1,110	305	-	956	1,078	-	1,035	649	896	996	1,054	607	225	12,782
		% critical deviations*	9	2	7	3	2	1	-	7	3	-	2	6	5	3	9	1	2	4
		% total deviations^	22	3	11	15	6	26	-	15	7	-	6	7	22	5	20	2	9	12
2007	143	No. of tests	908	1,114	830	1,105	1,101	389	-	914	1,111	-	1,092	678	875	971	1,047	583	258	12,976
		% critical deviations*	6	5	1	0	1	4	-	1	3	-	2	5	4	3	4	1	0	3
		% total deviations^	17	7	1	6	1	16	-	2	4	-	3	6	26	3	11	2	6	7
2008	168	No. of tests	-	1,331	961	1,226	1,307	-	791	1,104	1,265	-	1,168	718	867	1,155	1,249	696	-	13,858
		% critical deviations*	-	3	3	1	19	-	3	3	4	-	2	4	7	3	6	2	-	5
		% total deviations^	-	8	6	11	21	-	6	6	6	-	4	5	25	4	13	2	-	9
2009	153	No. of tests	-	1,206	921	1,108	1,190	-	775	1,009	1,143	-	1,095	624	864	1,042	1,114	616	-	12,707
		% critical deviations*	-	3	1	1	8	-	0	1	2	-	1	7	9	3	4	1	-	3
		% total deviations^	-	6	1	2	10	-	1	2	3	-	3	9	30	4	10	1	-	6
2010	152	No. of tests	-	1,173	937	1,118	1,194	-	787	1,026	1,133	-	1,096	566	800	1,012	1,134	604	-	12,580
		% critical deviations*	-	4	2	1	3	-	4	4	5	-	1	14	19	4	5	1	-	5
		% total deviations^	-	5	3	2	3	-	8	8	6	-	2	17	55	4	9	1	-	9

Table 8 (continued). EQAS participants' performance of *Salmonella* strains antimicrobial susceptibility testing categorized by antimicrobial.

EQAS iteration	No. of labs	Performance	Antimicrobial[∞]																
			AMC	AMP	CAZ	CHL	CIP	POD	CRO	CTX	GEN	KAN	NAL	SMX	STR	SXT	TET	TMP	XNL	OVERALL
2011	127	No. of tests	-	1099	829	988	1070	-	744	909	999	-	993	542	682	988	1017	493	-	11,353
		% critical deviations*	-	5	3	2	20	-	3	4	4	-	7	4	3	3	4	1	-	5
		% total deviations^	-	6	4	2	21	-	3	6	5	-	15	5	42	3	10	2	-	9
2012	159	No. of tests	-	1228	993	1159	1245	-	834	1058	1161	-	1136	584	814	1054	1163	613	-	13,042
		% critical deviations*	-	3	2	1	11	-	2	4	3	-	2	5	2	1	2	1	-	3
		% total deviations^	-	5	2	2	12	-	3	5	4	-	4	7	35	2	5	1	-	7
2013	145	No. of tests	-	1121	898	1027	1134	-	763	1011	1086	-	1027	491	-	946	1060	545	-	11,109
		% critical deviations*	-	2	3	0	2	-	1	3	3	-	2	4	-	2	3	2	-	2
		% total deviations^	-	3	3	1	18	-	2	6	6	-	6	5	-	2	5	2	-	5
2014	155	No. of tests	-	1176	1003	1072	1161	-	817	1014	1147	-	1078	561	-	1039	1107	541	-	11716
		% critical deviations*	-	3	3	1	3	-	1	2	3	-	1	5	-	2	3	2	-	2
		% total deviations^	-	4	4	2	19	-	2	3	5	-	2	6	-	3	5	2	-	5
Average•	137	No. of tests	944	1059	905	937	904	347	787	1000	1028	588	909	567	769	974	1004	559	242	10154
		% critical deviations*	7	3	2	1	5	3	2	2	3	3	2	5	6	3	4	1	1	3
		% total deviations^	17	5	4	3	10	21	4	7	5	10	5	7	31	4	10	2	8	8

[∞]For antimicrobial abbreviations: see List of Abbreviations page 1

*R→ S & S → R (R, resistant; S, susceptible)

^S→R & R→S & S↔I or I↔R (I, intermediate)

• Data do not include one strain which may have lost resistance due to transport or storage stress

-, not determined

Table 9. Region-based categorization of EQAS participants' performance of *Salmonella* AST

Region	EQAS iteration	No. of labs	% correct test result	% minor deviations (S ↔ I or I ↔ R)^	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (S → R & R → S)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Africa	2001	7	80.1	9.6	7.7	2.5	10.2	19.8	Cameroun, Egypt, Gambia (2), Ivory Coast, Kenya (4), Madagascar, Mauritius, Morroco, Nigeria (3), Senegal, South Africa, Sudan, Tunisia, Zambia
	2002	10	94.3	4.1	1.0	0.6	1.6	5.7	
	2003	13	86.9	6.6	2.8	3.7	6.5	13.1	
	2004	11	85.7	7.2	5.2	1.9	7.1	14.3	
	2006	20	85.8	7.5	4.1	2.7	6.8	14.3	
	2007	16	90.7	4.4	4.0	0.9	4.9	9.3	
	2008	19	83.8	6.5	5.5	4.2	9.7	16.2	
	2009	22	90.1	4.5	3.6	1.8	5.4	9.9	
	2010	22	84.7	6.0	6.5	2.8	9.3	15.3	
	2011	17	87.0	5.0	4.7	3.3	8.0	13.0	
	2012	18	89.4	5.3	3.5	1.9	5.4	10.6	
	2013	16	92.0	3.2	4.0	0.9	4.9	8.0	
	2014	20	92.5	3.8	2.0	1.7	3.7	7.5	
Central Asia & Middle East	2001	10	87.7	6.3	5.2	0.8	6.0	12.3	Bahrain, Georgia, India (9), Islamic rep. of Iran (3), Israel, Jordan, Kazakhstan
	2002	6	83.4	9.8	6.6	0.2	6.8	16.6	
	2003	8	89.9	4.5	4.0	1.6	5.6	10.1	
	2004	10	87.5	6.7	5.5	0.3	5.8	12.5	
	2006	7	79.2	10.5	9.8	0.5	10.3	20.8	
	2007	8	87.8	5.0	6.2	1.1	7.3	12.2	
	2008	12	86.1	6.5	4.0	3.4	7.4	13.9	
	2009	6	93.7	4.3	0.9	1.1	2.0	6.3	
	2010	7	95.8	2.6	0.2	1.4	1.6	4.2	
	2011	4	91.8	4.1	1.8	2.3	4.1	8.2	
	2012	8	92.8	4.4	1.6	0.7	2.3	6.6	
	2013	8	93.6	5.2	1.0	0.1	1.2	6.4	
	2014	17	91.0	4.2	2.9	2.0	4.9	9.0	
Caribbean	2001	2	83.5	9.5	7.0	0.0	7.0	16.5	Barbados, Cuba, Dominican Republic, Jamaica
	2002	1	95.8	4.2	0.0	0.0	0.0	4.2	
	2003	8	91.7	6.4	1.5	0.5	2.0	8.4	
	2004	8	94.1	3.1	1.9	0.9	2.8	5.9	
	2006	5	92.1	5.4	1.6	1.0	2.6	8.0	
	2007	4	95.0	3.1	0.9	0.9	1.8	5.0	
	2008	5	90.7	5.5	0.9	2.9	3.8	9.3	
	2009	4	93.2	1.8	3.2	1.8	5.0	6.8	
	2010	4	90.9	5.4	2.7	0.7	3.4	8.8	
	2011	2	96.5	1.4	0.0	2.1	2.1	3.5	
	2012	4	91.1	1.5	6.7	0.7	7.4	8.9	
	2013	3	90.2	2.6	7.3	0.0	7.3	9.8	
	2014	4	78.3	4.7	9.4	7.6	17.0	21.7	

Table 9 (continued). Region-based categorization of EQAS participants' performance of *Salmonella* antimicrobial susceptibility testing

Region	EQAS iteration	No. of labs	% correct test result	% minor deviations (S ↔ I or I ↔ R)^	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (S → R & R → S)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Europe	2001	47	91.3	5.7	2.7	0.3	3.0	8.7	Albania, Belgium, Bulgaria, Croatia (2), Cyprus, Denmark (2), France, Germany, Greece (3), Hungary, Ireland, Italy (10), Kosova, Luxembourg (2), Malta, Poland (3), Portugal, Serbia (2), Slovak Republic, Slovenia, Spain, Turkey, United Kingdom
	2002	57	92.7	5.2	1.2	0.9	2.1	7.3	
	2003	64	92.9	3.8	1.0	2.3	3.3	7.1	
	2004	58	93.5	4.3	1.4	0.8	2.2	6.5	
	2006	54	88.7	7.0	3.8	0.6	4.4	11.3	
	2007	49	94.2	3.7	1.6	0.4	2.0	5.7	
	2008	51	91.2	4.4	2.5	1.9	4.4	8.8	
	2009	40	95.1	2.6	1.3	0.9	2.2	4.8	
	2010	39	92.4	4.1	1.2	2.3	3.5	7.6	
	2011	36	92.5	4.5	1.7	1.3	3.0	7.5	
	2012	40	95.5	2.8	1.2	0.4	1.7	4.5	
	2013	37	95.7	2.5	1.4	0.3	1.7	4.2	
	2014	40	96.6	2.1	0.8	0.5	1.3	3.4	
North America	2001	4	95.8	3.8	0.0	0.4	0.4	4.2	Canada (4), USA (4)
	2002	3	90.5	6.9	0.6	2.0	2.6	9.5	
	2003	7	93.4	5.2	0.0	1.4	1.4	6.6	
	2004	9	94.2	4.2	1.8	0.0	1.8	6.0	
	2006	8	94.8	2.9	1.0	1.3	2.3	5.2	
	2007	10	95.4	2.9	0.8	0.8	1.6	4.6	
	2008	14	96.4	0.6	0.4	2.6	3.0	3.6	
	2009	10	98.7	0.0	0.4	0.9	1.3	1.3	
	2010	11	94.8	2.6	0.2	2.4	2.6	5.2	
	2011	9	92.1	2.6	1.5	3.8	5.3	7.9	
	2012	10	96.0	2.1	1.0	0.9	1.9	4.0	
	2013	7	98.4	1.3	0.0	0.2	0.2	1.6	
	2014	8	96.9	2.2	0.4	0.6	0.9	3.1	
Oceania	2001	6	91.8	4.7	2.7	0.9	3.6	8.2	Australia (3), New Zealand, Tuvalu
	2002	7	91.7	6.2	0.0	2.0	2.0	8.3	
	2003	9	94.3	2.5	1.2	2.0	3.2	5.7	
	2004	11	97.1	2.5	0.3	0.1	0.4	2.9	
	2006	7	93.4	4.6	0.9	1.1	2.0	6.6	
	2007	1	98.9	1.1	0.0	0.0	0.0	1.1	
	2008	4	93.9	3.8	0.0	2.3	2.3	6.1	
	2009	4	95.9	3.2	0.3	0.6	0.9	4.1	
	2010	4	92.5	4.6	0.6	2.3	2.9	7.5	
	2011	4	93.8	5.6	0.6	0.0	0.6	6.2	
	2012	4	95.5	3.1	0.6	0.9	1.4	4.5	
	2013	4	96.8	2.9	0.0	0.3	0.3	3.2	
	2014	5	97.4	2.0	0.0	0.6	0.6	2.6	

Table 9 (continued). Region-based categorization of EQAS participants' performance of *Salmonella* antimicrobial susceptibility testing.

Region	EQAS iteration	No. of labs	% correct test result	% minor deviations (S ↔ I or I ↔ R)^	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (S → R & R → S)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Russia	2001	1	81.9	15.3	2.8	0.0	2.8	18.1	Russia (4)
	2002	1	84.5	9.9	5.6	0.0	5.6	15.5	
	2003	1	100.0	0.0	0.0	0.0	0.0	0.0	
	2004	4	91.2	6.6	1.5	0.7	2.2	8.8	
	2006	5	87.4	8.2	2.7	1.7	4.4	12.6	
	2007	8	88.9	5.8	4.8	0.4	5.2	11.0	
	2008	6	92.2	4.7	1.4	1.7	3.1	7.8	
	2009	6	93.8	2.1	3.3	0.8	4.1	6.2	
	2010	8	94.3	3.3	1.3	1.1	2.4	5.7	
	2011	7	90.0	4.8	3.2	2.0	5.2	10.0	
	2012	6	97.4	2.0	0.0	0.6	0.6	2.6	
	2013	2	98.2	1.8	0.0	0.0	0.0	1.8	
	2014	4	98.2	0.3	0.9	0.6	1.5	1.8	
Latin America	2001	11	90.8	6.9	1.4	1.0	2.4	9.2	Argentina, Bolivia, Brazil (2), Chile (2), Colombia (2), Costa Rica, Ecuador (2), El Salvador, Guatemala, Honduras, Mexico (2), Nicaragua, Panama, Paraguay, Peru, Suriname, Uruguay (2), Venezuela
	2002	13	93.7	4.6	0.7	1.0	1.7	6.3	
	2003	12	90.8	4.2	2.0	3.0	5.0	9.2	
	2004	17	94.4	4.7	0.8	0.1	0.9	5.6	
	2006	16	88.7	6.3	4.5	0.6	5.1	11.3	
	2007	17	94.9	1.8	1.9	1.4	3.3	5.0	
	2008	20	93.0	3.4	1.5	2.1	3.6	7.0	
	2009	20	95.6	2.1	1.1	1.2	2.3	4.4	
	2010	23	90.8	2.1	5.6	1.4	7.1	9.2	
	2011	22	90.8	2.8	3.1	3.3	6.4	9.2	
	2012	25	94.4	1.6	3.0	1.0	4.0	5.6	
	2013	25	95.5	2.6	1.2	0.3	1.5	4.2	
	2014	24	96.5	1.9	1.1	0.6	1.7	3.5	
China	2001	4	98.9	0.8	0.0	0.3	0.3	1.1	Brunei Darussalam, Cambodia, Japan (2), LAO PDR, Malaysia (4), Philippines, Rep of Korea (2), Sri Lanka (2), Taiwan, Thailand (8), Viet Nam
	2002	3	96.0	4.0	0.0	0.0	0.0	4.0	
	2003	8	90.1	3.6	2.8	3.6	6.4	10.0	
	2004	8	96.0	3.2	0.7	0.1	0.8	4.0	
	2006	6	89.6	7.0	2.9	0.5	3.4	10.4	
	2007	10	98.3	1.1	0.3	0.2	0.5	1.6	
	2008	18	92.8	3.7	0.8	2.7	3.5	7.2	
	2009	14	94.8	2.2	2.1	0.8	2.9	5.1	
	2010	9	92.1	4.5	1.6	1.8	3.4	7.9	
	2012	9	95.3	3.0	0.5	1.2	1.6	4.7	
	2013	8	96.9	2.0	0.5	0.5	1.0	3.1	
	2014	24	95.5	2.9	1.0	0.6	1.6	4.5	

^S. susceptible; I. intermediate; R. resistant

Table 9 (continued). Region-based categorization of EQAS participants' performance of *Salmonella* antimicrobial susceptibility testing.

Region	EQAS iteration	No. of labs	% correct test result	% minor deviations (S ↔ I or I ↔ R)^	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (S → R & R → S)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Southeast Asia	2001	16	88.1	7.7	2.3	1.9	4.2	11.9	China (8)
	2002	18	89.0	8.1	1.4	1.6	3.0	11.0	
	2003	17	87.4	5.2	4.7	2.7	7.4	12.6	
	2004	16	92.8	4.4	2.3	0.5	2.8	7.2	
	2006	15	90.0	8.1	1.2	0.8	2.0	10.0	
	2007	20	93.9	4.0	1.4	0.7	2.1	6.1	
	2008	19	90.5	4.7	2.2	2.6	4.8	9.5	
	2009	27	91.8	4.1	3.0	1.2	4.2	8.3	
	2010	25	92.8	3.8	1.5	1.9	3.4	7.2	
	2011	26	90.5	3.5	2.4	3.5	5.9	9.5	
	2012	35	91.7	3.9	3.5	0.9	4.4	8.3	
	2013	35	93.4	3.2	2.5	0.7	3.2	6.4	
	2014	8	97.0	1.2	0.1	1.6	1.8	3.0	

^S. susceptible; I. intermediate; R. resistant

Table 10. EQAS participants' performance of antimicrobial susceptibility testing of quality control strain *Escherichia coli* ATCC 25922

		Method	Perfor-mance[5.6]	AMC	AMP	CAZ	CHL	CIP	POD	CRO	CTX	ENR[2]	FFN[2]	FIS (SMX)[3]	GEN	NAL	STR	SXT	TET	TMP	XNL[2]
Accepted interval[1]		**MIC (µg/ml)**		2-8	2-8	0.06-0.5	2-8	0.004-0.016	0.25-1	0.03-0.12	0.03-0.12	0.008-0.03	2-8	8-32	0.25-1	1-4	4-16[4]	≤0.5/9.5	0.5-2	0.5-2	0.25-1
		Disks (mm)		18-24	16-22	25-32	21-27	30-40	23-28	29-35	29-35	32-40	22-28	15-23	19-26	22-28	12-20	23-29	18-25	21-28	26-31
EQAS iteration (total no. of participants)	**2000** (44)	MIC & Disk	No.[5]	-	37	-	38	35	-	-	-	-	-	19	39	37	36	-	42	31	-
			%[6]	-	27	-	37	20	-	-	-	-	-	53	23	35	22	-	42	30	-
	2001 (107)	MIC & Disk	No.[5]	-	97	-	97	97	-	-	-	-	-	53	99	74	81	90	96	50	-
			%[6]	-	19	-	20	14	-	-	-	-	-	34	12	14	12	14	22	22	-
	2002 (114)	MIC & Disk	No.[5]	-	109	-	107	108	-	-	-	-	-	57	108	102	82	102	102	66	-
			%[6]	-	16	-	15	14	-	-	-	-	-	26	12	14	11	12	13	11	-
	2003 (144)	MIC & Disk	No.[5]	-	140	-	137	138	-	-	-	-	-	82	138	132	105	129	137	79	-
			%[6]	-	14	-	22	9	-	-	-	-	-	17	9	16	9	14	19	14	-
	2004 (140)	MIC & Disk	No.[5]	117	132	-	128	132	-	-	111	-	-	84	134	126	110	120	129	87	-
			%[6]	13	10	-	13	8	-	-	18	-	-	16	10	9	6	11	13	9	-
	2006 (137)	MIC & Disk	No.[5]	116	133	96	126	127	39	-	115	19	-	74	131	122	106	122	125	74	32
			%[6]	9	14	15	18	8	12	-	21	63	-	29	14	20	11	19	12	17	22
	2007 (126)	MIC & Disk	No.[5]	102	124	92	123	121	47	-	104	-	13	64	124	120	97	107	117	67	35
			%[6]	8	11	9	14	12	9	-	16	-	0	22	6	7	6	13	7	10	11
	2008 (147)	MIC & Disk	No.[5]	-	147	111	135	144	-	-	124	-	-	71	145	136	101	129	139	79	-
			%[6]	-	12	9	10	8	-	-	14	-	-	14	8	8	12	13	7	13	-
		MIC	No.[5]	-	33	23	24	33	-	-	23	-	-	18	31	23	19	22	28	16	-
			%[6]	-	0	5	0	6	-	-	9	-	-	11	0	0	11	9	0	13	-
		Disk	No.[5]	-	114	89	112	111	-	-	101	-	-	53	114	113	82	107	111	63	-
			%[6]	-	16	10	12	8	-	-	15	-	-	15	11	10	12	14	9	13	-
	2009 (129)	MIC & Disk	No.[5]	-	128	100	121	124	-	88	107	-	-	63	123	117	98	113	122	70	-
			%[6]	-	16	13	15	7	-	16	10	-	-	11	18	13	10	14	14	11	-
		MIC (27)	No.[5]	-	27	19	24	26	-	20	20	-	-	14	25	24	19	21	27	25	-
			%[6]	-	11	11	8	8	-	15	15	-	-	21	12	8	5	19	11	13	-
		Disk (102)	No.[5]	-	101	81	97	98	-	68	87	-	-	49	98	93	79	92	95	55	-
			%[6]	-	16	14	16	6	-	16	9	-	-	10	18	14	11	12	15	11	-
	2010 (116)	MIC & Disk	No.[5]	-	114	97	108	115	-	79	100	-	-	51	112	104	84	101	110	63	-
			%[6]	-	11	9	9	6	-	10	14	-	-	11	11	5	5	12	5	15	-
		MIC (25)	No.[5]	-	25	15	21	25	-	15	17	-	-	12	24	19	17	17	24	11	-
			%[6]	-	12	20	10	8	-	7	18	-	-	8	13	16	18	18	17	36	-
		Disk (91)	No.[5]	-	89	82	87	90	-	64	83	-	-	39	88	85	67	84	86	52	-
			%[6]	-	9	6	8	4	-	9	11	-	-	10	9	2	1	10	1	8	-

Table 10 (continued). EQAS participants' performance of antimicrobial susceptibility testing of quality control strain *Escherichia coli* ATCC 25922

		Method	Perfor-mance[5.6]	AMC	AMP	CAZ	CHL	CIP	POD	CRO	CTX	ENR[2]	FFN[2]	FIS (SMX)[3]	GEN	NAL	STR	SXT	TET	TMP	XNL[2]
Accepted interval[1]		**MIC (µg/ml)**		2-8	2-8	0.06-0.5	2-8	0.004-0.016	0.25-1	0.03-0.12	0.03-0.12	0.008-0.03	2-8	8-32	0.25-1	1-4	4-16[4]	≤0.5/9.5	0.5-2	0.5-2	0.25-1
		Disks (mm)		18-24	16-22	25-32	21-27	30-40	23-28	29-35	29-35	32-40	22-28	15-23	19-26	22-28	12-20	23-29	18-25	21-28	26-31
EQAS iteration (total no. of participants)	**2011** (112)	MIC & Disk	No.[5]	-	111	89	102	109	-	76	96	-	-	50	103	103	72	99	107	51	-
			%[6]	-	17	4	11	7	-	7	9	-	-	8	11	8	4	16	7	14	-
		MIC (23)	No.[5]	-	23	15	18	22	-	16	15	-	-	13	22	19	17	16	21	11	-
			%[6]	-	4	7	0	9	-	6	0	-	-	8	9	0	6	6	5	0	-
		Disk (89)	No.[5]	-	88	74	84	87	-	60	81	-	-	37	81	84	55	83	86	40	-
			%[6]	-	20	4	13	7	-	7	11	-	-	8	11	10	4	18	8	18	-
	2012 (135)	MIC & Disk	No.[5]	-	134	111	121	131	-	90	115	-	-	53	127	121	89	112	129	66	-
			%[6]	-	13	12	7	6	-	11	10	-	-	11	9	9	8	13	10	21	-
		MIC (37)	No.[5]	-	37	26	31	35	-	23	28	-	-	19	35	31	26	23	35	22	-
			%[6]	-	3	4	0	3	-	0	4	-	-	5	3	3	8	0	0	9	-
		Disk (98)	No.[5]	-	97	85	90	96	-	67	87	-	-	34	92	90	63	89	94	44	-
			%[6]	-	16	14	9	7	-	15	11	-	-	15	11	11	8	16	14	27	-
	2013 (122)	MIC & Disk	No.[5]	-	117	100	112	119	-	82	107	-	-	44	113	113	-	101	114	59	-
			%[6]	-	12	7	5	7	-	4	8	-	-	10	6	11	-	8	8	11	-
		MIC (33)	No.[5]	-	31	25	28	32	-	19	27	-	-	17	32	28	-	22	32	22	-
			%[6]	-	6	4	4	13	-	5	11	-	-	18	9	11	-	5	6	5	-
		Disk (89)	No.[5]	-	86	75	84	87	-	63	80	-	-	27	81	85	-	79	82	37	-
			%[6]	-	13	8	6	5	-	5	6	-	-	7	4	9	-	10	7	8	-
	2014 (115)	MIC & Disk	No.[5]	-	111	99	101	108	-	75	97	-	-	49	111	103	-	102	104	50	-
			%[6]	-	5	7	7	6	-	7	14	-	-	14	8	8	-	8	7	2	-
		MIC (28)	No.[5]	-	27	21	24	27	-	16	22	-	-	16	28	24	-	21	25	12	-
			%[6]	-	4	5	4	15	-	6	14	-	-	0	14	8	-	14	0	0	-
		Disk (87)	No.[5]	-	84	78	77	81	-	59	75	-	-	33	83	79	-	81	79	38	-
			%[6]	-	6	8	8	4	-	7	15	-	-	21	6	8	-	6	9	3	-

[0]For antimicrobial abbreviations: see List of Abbreviations page 1

[1]CLSI standard. Performance Standards for Antimicrobial Disk and Dilution Susceptibility testing. 22nd Informational supplement. CLSI document M100-S22. 2012 Wayne. PA. USA

[2]CLSI standard. Performance Standards for Antimicrobial Disk and Dilution Susceptibility Tests for bacteria Isolated from Animals. M31-A3. 3rd Edition [Approved Standard]. 2008. Wayne. PA. USA

[3]FIS (sulfisoxazole) covers the group of SMX (sulfonamides)

[4]Quality control range developed by the manufacturer of Sensititre®

[5]No.. number of laboratories performing the analysis

[6]%. percentage of laboratories reporting erroneous results

-. not determined

Table 11. *Shigella* serotypes (ST) and deviations (D). WHO EQAS 2014

Strain	Correct serotype		No. of labs reporting correct identification	D (%)	Deviating results	No. of labs reporting correct ST	D (%)	Deviating results (*)
WHO 2014 SH-14.1	*S. sonnei*	N/A	120	3,2	4	NA	NA	NA
WHO 2014 SH-14.2	*S. sonnei*	N/A	116	4,1	5	NA	NA	NA
WHO 2014 SH-14.3	*S. flexneri*	2 / 2b	122	2,4	3	67	16,3	2a(9), var. X(4)
WHO 2014 SH-14.4	*S. boydii*	2	117	5,6	7	64	7,2	1(2), 3, 5, 11

*number of participants reporting deviating result

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Table 12. Region-based categorization of laboratories performing *Shigella* serotyping in 2014

Region	Year	No. of laboratories	No. of strains serotyped	Strains serotyped correctly (%)	Countries participating in the 2014 iteration
Africa	2009	8	18	72.2	Egypt, Ivory Coast, Kenya, Mauritius, South Africa, Tunisia,
	2010	7	16	62.5	
	2011	4	10	100.0	
	2012	5	18	90.0	
	2013	5	8	62.5	
	2014	6	12	58.3	
Central Asia & Middle East	2009	3	5	100.0	Bahrain, India (2), Iran (Islamic rep. of), Israel, Jordan
	2010	3	6	83.3	
	2011	2	6	100.0	
	2012	3	9	81.8	
	2013	4	8	100.0	
	2014	8	20	85.0	
China	2009	13	35	100.0	China (9)
	2010	9	23	91.3	
	2011	-	-	-	
	2012	8	29	90.6	
	2013	6	11	100.0	
	2014	9	33	93.9	
Caribbean	2009	-	-	-	Barbados, Jamaica
	2010	-	-	-	
	2011	-	-	-	
	2012	1	1	33.3	
	2013	-	-	-	
	2014	2	3	66.7	
Europe	2009	15	40	92.5	Albania, Belgium, Bulgaria, Czech Republic, Denmark, Germany (2), Greece, Ireland, Italy (2), Luxembourg, Malta, Poland, Portugal, Serbia (2), Slovenia (2), Spain, Sweden, Turkey, United Kingdom
	2010	15	35	85.7	
	2011	16	42	92.9	
	2012	19	63	86.3	
	2013	18	31	96.8	
	2014	23	58	84.5	
North America	2009	7	18	100.0	Canada (7), USA
	2010	7	20	100.0	
	2011	6	16	100.0	
	2012	8	25	80.6	
	2013	8	14	100.0	
	2014	8	24	95.8	

Table 12 (continued). Region-based categorization of laboratories performing *Shigella* serotyping in 2014

Region	Year	No. of laboratories	No. of strains serotyped	Strains serotyped correctly (%)	Countries participating in the 2014 iteration
Oceanic	2009	3	8	100.0	Australia (3), New Zealand
	2010	3	8	100.0	
	2011	3	8	100.0	
	2012	3	12	100.0	
	2013	4	10	100.0	
	2014	4	13	100.0	
Russia	2009	6	18	83.3	Russia (3)
	2010	7	20	75.0	
	2011	6	18	88.9	
	2012	5	16	80.0	
	2013	2	4	100.0	
	2014	3	10	100.0	
Latin America	2009	16	40	97.5	Argentina, Brazil (2), Chile (2), Colombia, Costa Rica, Ecuador, El Salvador, Honduras, Mexico (2), Nicaragua, Panama, Paraguay, Peru, Uruguay, Venezuela
	2010	13	33	78.8	
	2011	15	37	94.6	
	2012	19	58	80.6	
	2013	16	30	93.3	
	2014	18	54	87.0	
Southeast Asia	2009	11	30	90.0	Japan (2), LAO PDR, Malaysia, Philippines, Rep of Korea, Sri Lanka, Taiwan, Thailand (4)
	2010	14	32	87.5	
	2011	13	33	84.8	
	2012	14	47	90.4	
	2013	9	17	100.0	
	2014	12	30	83.3	

Table 13. EQAS participating laboratories' performance of *Shigella* strains antimicrobial susceptibility testing

EQAS iteration	No. of participating laboratories	% correct test results	% minor deviations (S ↔ I or I ↔ R)^	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (S → R & R → S)^	% total deviations (S → R & R → S & S ↔ I or I ↔ R)^
2008	15	95	2	2	1	3	5
2009	111	96	2	1	1	2	4
2010	114	91	2	1	6	7	9
2011	107	92	2	1	4	5	7
2012	120	91	3	1	5	6	9
2013	99	91	6	2	2	4	10
2014	116	92	4	1	3	4	8

^S. susceptible; I. intermediate; R. resistant

Table 14. Antimicrobial susceptibility test results (number of R/I/S) for the EQAS 2014 *Shigella* strains*

Strain	Antimicrobial[∞]											
	AMP	**CTX**	**CAZ**	**CRO**	**CHL**	**CIP**	**GEN**	**NAL**	**SMX**	**TET**	**SXT**	**TMP**
WHO SH-14.1	**109**/0/1	**98**/0/0	**80**/6/12	**82**/1/1	0/0/**96**	9/**56**/46	6/1/**100**	**89**/10/1	**47**/0/0	**95**/3/2	**103**/0/2	**45**/0/2
WHO SH-14.2	**106**/1/3	**94**/0/2	**68**/10/20	**77**/3/1	1/0/**97**	0/6/**106**	**95**/4/6	4/0/**99**	**46**/0/1	**96**/2/1	**100**/0/3	**47**/0/1
WHO SH-14.3	**106**/1/3	0/1/**96**	3/0/**93**	3/0/**80**	**83**/7/6	**23**/52/35	3/0/**104**	**98**/0/2	**44**/0/3	**92**/2/5	**99**/0/4	**44**/1/4
WHO SH-14.4	**107**/0/4	3/0/**95**	3/1/**94**	1/0/**82**	1/0/**95**	0/3/**105**	2/1/**102**	3/1/**98**	**42**/0/5	**89**/5/6	7/1/**94**	3/1/**41**

[∞]For antimicrobial abbreviations: see List of Abbreviations page 1

*In bold: expected interpretation. Grey cell: <90% of laboratories did correct interpretation. R. resistant; I. intermediate; S. susceptible.

Table 15. EQAS laboratories' performance of *Shigella* strains antimicrobial susceptibility testing categorized by antimicrobial

EQAS iteration	No. of labs	Lab performance	Antimicrobial													
			AMP	CAZ	CHL	CIP	CTX	GEN	NAL	SMX	STR	SXT	TET	TMP	CRO	OVERALL
2008	15	No. of tests	52	44	51	48	48	50	52	7	27	52	52	4	42	529
		% critical deviations*	1	2	1	-	2	1	-	-	4	2	4	-	2	1.5
		% total deviations^	1	2	1	-	2	1	-	-	9	2	8	-	2	2.2
2009	111	No. of tests	423	358	388	426	372	396	388	211	293	388	386	218	301	4.548
		% critical deviations*	2.4	0.3	2.1	0.2	1.1	2.5	0.5	3.8	5.8	2.3	2.8	1.8	0.3	1.9
		% total deviations^	3.8	0.3	4.6	0.9	1.1	3.5	1.5	3.8	18.1	3.6	7.5	1.8	0.6	3.8
2010	114	No. of tests	424	344	402	434	377	403	382	194	275	363	410	218	291	4.517
		% critical deviations*	1.7	0.6	3.5	40.8	2.4	3.5	2.1	4.6	8.0	8.3	4.4	3.7	0.0	6.4
		% total deviations^	1.9	1.2	9.2	77.9	3.0	5.5	3.0	6.0	14.6	13.8	5.9	3.8	0.0	11.2
2011	107	No. of tests	403	322	353	396	343	359	369	179	246	371	376	178	289	4.184
		% critical deviations*	5.5	5.2	2.2	38.9	2.7	3.3	4.0	1.7	3.6	3.2	2.7	2.2	2.0	5.5
		% total deviations^	7.7	12.0	4.2	40.7	2.7	4.4	11.0	1.7	10.5	3.2	3.5	2.2	2.0	7.7
2012	120	No. of tests	462	376	427	464	400	430	442	196	291	396	426	215	337	4.862
		% critical deviations*	2.6	0.8	5.6	35.3	2.0	4.9	1.6	1.5	9.3	6.3	5.4	1.9	0.9	6.0
		% total deviations^	3.9	0.8	11.5	38.6	3.8	6.3	3.2	2.0	27.1	8.1	7.5	4.2	2.1	9.2
2013	99	No. of tests	-	351	379	420	384	392	393	164	-	346	392	193	309	3723
		% critical deviations*	-	1.1	2.1	8.3	3.4	2.3	3.3	1.8	-	5.8	2.8	3.1	1.0	3.4
		% total deviations^	-	0.3	0.6	2.0	0.9	0.6	0.8	1.1	-	1.7	0.7	1.6	0.3	9.5
2014	116	No. of tests	441	390	386	441	389	424	405	188	-	413	398	189	331	4395
		% critical deviations*	2.5	9.7	2.1	7.9	1.3	4.0	2.5	4.8	-	3.9	3.5	5.3	2.1	4.1
		% total deviations^	2.9	14.1	3.9	34.2	1.5	5.4	5.2	4.8	-	4.1	6.5	6.3	3.9	8.1

∞For antimicrobial abbreviations: see List of Abbreviations page 1
*R→ S & S → R (R. resistant; S. susceptible)
^S→R & R→S & S↔I or I↔R (I. intermediate)
-. not determined

Table 16. Region-based categorization of EQAS participating laboratories' performance of antimicrobial susceptibility tests for *Shigella* strains

Region	Year	No. of labs	% correct test result	% minor deviations (S↔I or I↔R)^	% major deviations (S→R)^	% very major deviations (R→ S)^	% critical deviations (R→ S & S → R)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Africa	2009	17	93.3	2.4	3.5	0.8	4.3	6.8	Cameroun, Egypt, Gambia (2), Ivory Coast, Kenya (4), Madagascar, Mauritius, Nigeria (2), Senegal, South Africa, Sudan, Tunisia, Zambia
	2010	16	84.8	2.5	2.7	10.0	12.7	15.2	
	2011	16	86.0	1.8	3.6	8.3	11.9	13.7	
	2012	17	82.6	4.2	2.5	10.7	13.2	17.4	
	2013	14	87,6	7,2	2,5	2,7	5,2	12,4	
	2014	18	85.3	6.1	2.3	6.4	8.7	14.7	
Central Asia & Middle East	2009	5	94.8	0.9	3.0	1.3	4.4	5.2	Bahrain, Georgia, India (9), Islamic rep. of Iran (3), Israel, Jordan
	2010	6	90.6	1.2	1.6	6.7	8.3	9.4	
	2011	4	92.9	1.6	0.5	4.9	5.4	7.1	
	2012	6	92.3	4.0	2.0	1.3	3.4	7.4	
	2013	6	86,9	8,5	3,9	0,8	4,6	13,1	
	2014	16	85.6	6.7	1.7	6.0	7.7	14.4	
Caribbean	2009	4	95.6	1.5	0.7	2.2	2.9	4.4	Barbados, Dominican Republic, Jamaica
	2010	4	88.5	1.5	3.8	6.2	10.0	11.5	
	2011	1	97.7	2.3	0.0	0.0	2.3	2.3	
	2012	3	84.6	1.9	7.7	5.8	13.5	15.4	
	2013	2	87,5	9,4	0,0	3,1	3,1	12,5	
	2014	3	76.5	5.1	7.1	11.2	18.4	23.5	
Europe	2009	22	98.1	1.1	0.7	0.1	0.8	1.9	Albania, Belgium, Bulgaria, Croatia, Cyprus, Denmark, Germany, Greece (2), Ireland, Italy (6), Luxembourg, Malta, Poland (2), Serbia (2), Slovenia, Spain, Turkey, United Kingdom
	2010	27	93.6	1.5	0.9	3.9	4.8	6.4	
	2011	24	94.8	2.2	0.5	2.5	3.0	5.1	
	2012	24	96.6	1.7	0.4	1.4	1.7	3.4	
	2013	23	93,6	4,8	1,2	0,3	1,5	6,4	
	2014	26	96.0	3.2	0.1	0.7	0.8	4.0	

Table 16 (continued) Region-based categorization of EQAS participating laboratories' performance of antimicrobial susceptibility tests for *Shigella* strains

Region	Year	No. of labs	% correct test result	% minor deviations (S↔I or I↔R)^	% major deviations (S→R)^	% very major deviations (R→ S)^	% critical deviations (R→ S & S → R)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
North America	2009	6	100.0	0.0	0.0	0.0	0.0	0.0	Canada (2), USA
	2010	7	95.0	0.0	0.0	5.0	5.0	5.0	
	2011	4	90.1	0.7	3.3	5.9	9.2	9.9	
	2012	6	89.5	0.0	2.1	8.4	10.5	10.5	
	2013	4	95,2	3,2	0,0	1,6	1,6	4,8	
	2014	3	95.4	2.8	0.0	1.9	1.9	4.6	
Oceanic	2009	-	-	-	-	-	-	-	Australia, Tuvalu
	2010	1	90.0	10.0	0.0	0.0	0.0	10.0	
	2011	1	92.5	5.0	0.0	2.5	2.5	7.5	
	2012	1	90.0	7.5	0.0	2.5	2.5	10.0	
	2013	1	95,5	4,5	0,0	0,0	0,0	4,5	
	2014	2	96.2	3.8	0.0	0.0	0.0	3.8	
Russia	2009	6	95.5	1.6	1.6	1.3	2.9	4.6	Russia (3)
	2010	7	92.1	2.9	1.5	3.5	5.0	7.9	
	2011	6	94.4	3.6	0.0	2.0	2.0	5.6	
	2012	5	96.8	1.4	0.5	1.4	1.8	3.2	
	2013	2	95,2	4,8	0,0	0,0	0,0	4,8	
	2014	3	98.4	0.8	0.0	0.8	0.8	1.6	
Latin America	2009	20	98.3	1.1	0.4	0.3	0.7	1.7	Argentina, Bolivia, Brazil (2), Chile (2), Colombia, Costa Rica, Ecuador (2), El Salvador, Guatemala, Honduras, Mexico (2), Nicaragua, Panama, Paraguay, Peru, Suriname, Uruguay (2), Venezuela
	2010	22	92.1	1.3	2.1	4.5	6.6	7.9	
	2011	20	94.0	1.5	1.3	3.2	4.5	6.0	
	2012	24	91.7	1.3	0.6	6.5	7.1	8.3	
	2013	23	94.1	3.9	1.2	0.8	2.0	5.9	
	2014	23	94.4	3.3	0.5	1.9	2.3	5.6	

^S. susceptible; I. intermediate; R. resistant.

Table 16 (continued) Region-based categorization of EQAS participating laboratories' performance of antimicrobial susceptibility tests for *Shigella* strains

Region	Year	No. of labs	% correct test result	% minor deviations (S↔I or I↔R)^	% major deviations (S→R)^	% very major deviations (R→ S)^	% critical deviations (R→ S & S → R)^	% total deviations (S→R & R→S & S↔I or I↔R)^	Countries participating in the 2014 iteration
Southeast Asia	2009	18	94.1	3.9	0.3	1.7	2.0	5.9	Cambodia, Japan (2), LAO PDR, Malaysia, Philippines, Rep of Korea, Sri Lanka, Taiwan, Thailand (3), Viet Nam
	2010	16	90.5	2.4	0.7	6.4	7.1	9.5	
	2011	19	90.0	2.1	0.8	6.1	6.9	9.0	
	2012	27	87.1	5.1	1.9	5.6	7.6	12.7	
	2013	19	86,2	7,5	2,9	3,1	6,0	13,5	
	2014	13	92.5	4.0	1.1	2.4	3.5	7.5	
China	2009	12	96.3	2.2	1.0	0.5	1.5	3.7	China (8)
	2010	8	92.7	1.2	0.6	5.5	6.1	7.3	
	2011	-	-	-	-	-	-	-	
	2012	7	90.3	2.9	0.0	6.8	6.8	9.7	
	2013	5	92,7	3,4	0,4	3,4	3,9	7,3	
	2014	8	94.6	2.2	0.3	3.0	3.2	5.4	

^S. susceptible; I. intermediate; R. resistant.

Table 17. Proportion of laboratories that obtained the expected result. Number (n/N) and percentages of laboratories which correctly detected and confirmed the ESBL and non ESBL producing *Salmonella* and *Shigella* strains.

Isolate no.	Expected interpretation	Confirmatory tests	
		CAZ/Cl:CAZ	CTX/Cl:CTX
WHO 2014 S-14.1	non ESBL	23/24 (96%)	28/29 (97%)
WHO 2014 S-14.2	non ESBL	23/24 (96%)	28/29 (97%)
WHO 2014 S-14.3	non ESBL	24/24 (100%)	28/29 (97%)
WHO 2014 S-14.4	non ESBL	24/25 (96%)	30/30 (100%)
WHO 2014 S-14.5	non ESBL	24/24 (100%)	28/29 (97%)
WHO 2014 S-14.6	non ESBL	23/24 (96%)	29/30 (97%)
WHO 2014 S-14.7	ESBL-producer	62/70 (89%)	68/78 (87%)
WHO 2014 S-14.8	non ESBL	23/25 (92%)	29/30 (97%)
WHO 2014 SH-14.1	ESBL-producer	50/55 (91%)	59/64 (92%)
WHO 2014 SH-14.2	ESBL-producer	45/51 (88%)	56/61 (92%)
WHO 2014 SH-14.3	non ESBL	21/21 (100%)	25/26 (96%)
WHO 2014 SH-14.4	non ESBL	20/22 (91%)	25/27 (93%)

Table 18. EQAS participating laboratories' performance of *Campylobacter* strains identification

EQAS iteration	No. of labs	Correct species	Strain no.	No. of results submitted	% correct identification	Deviating results (*)
2003	97	*C. jejuni*	# 1	93	88%	*C. coli* (9) *C. lari* (3)
	97	*C. coli*	# 2	93	84%	*C. jejuni* (7) *C. lari* (4) *C. upsaliensis* (4)
2004	109	*C. lari*	# 1	97	79%	*C. coli* (11) *C. jejuni* (8)
	109	*C. jejuni*	# 2	109	87%	*C. coli* (8) *C. lari* (4) *C. upsaliensis* (2)
2006	99	*C. jejuni*	# 1	87	90%	*C. lari* (3) *C. coli* (3) *C. upsaliensis* (3)
	99	*C. coli*	# 2	95	65%	*C. lari* (19) *C. jejuni* (11) *C. upsaliensis* (2)
2007	142	*C. lari*	# 1	98	74%	*C. jejuni* (10) *C. coli* (9) *C. upsaliensis* (7)
	142	*C. coli*	# 2	102	76%	*C. lari* (3) *C. jejuni* (20) *C. upsaliensis* (2)
2008	154	*C. lari*	# 1	109	62%	*C. coli* (14) *C. jejuni* (18) *C. upsaliensis* (7)
	154	*C. lari*	# 2	109	62%	*C. coli* (10) *C. jejuni* (19) *C. upsaliensis* (13)
2009	131	*C. coli*	# 1	87	77%	*C. upsaliensis* (10) *C. jejuni* (9) *C. lari* (1)
	131	*C. jejuni*	# 2	87	95%	*C. upsaliensis* (3) *C. lari* (1)
2010	130	*C. jejuni*	# 1	88	92%	*C. coli* (4) *C. lari* (3) *C. upsaliensis* (1)
	130	*C. coli*	# 2	84	85%	*C. jejuni* (11) *C. lari* (2) *C. upsaliensis* (2)
2011	132	*C. coli*	# 1	81	59%	*C. jejuni* (19) *C. lari* (13) *C. upsaliensis* (1)
	132	*C. coli*	# 2	79	70%	*C. jejuni* (17) *C. lari* (5) *C. upsaliensis* (2)
2012	135	*C. jejuni*	# 1	112	96%	*C. coli* (4)
	135	*C. jejuni*	# 2	103	85%	*C. coli* (10) *C. lari* (5) *C. upsaliensis* (1)
2013	123	*C. coli*	# 1	95	82%	*C. jejuni (13)* *C. lari* (3) *C. upsaliensis* (1)
	123	*C. coli*	# 2	92	84%	*C. jejuni* (9) *C. lari* (4) *C. upsaliensis* (2)
2014	101	*C. coli*	#2	101	85 %	*C. jejuni* (8) *C. lari* (6) *C. upsaliensis* (1)

*number of participants reporting the specified deviating result

Table 19. Region-based categorization of EQAS 2014 participating laboratories' performance of *Campylobacter* strains identification

Region	Year	No. of labs	No. of strains identified	% strains correctly identified	Countries participating in the 2014 iteration
Africa	2009	9	15	53	Cameroun, Kenya (2), Madagascar, Mauritius, Nigeria, Senegal, South Africa, Tunisia
	2010	7	13	77	
	2011	10	19	32	
	2012	9	17	82	
	2013	9	17	41	
	2014	9	9	67	
Central Asia & Middle East	2009	14	27	85	Bahrain, India (3), Islamic rep. of Iran(2), Israel
	2010	13	26	89	
	2011	2	4	50	
	2012	11	22	96	
	2013	1	8	50	
	2014	7	7	57	
Caribbean	2009	2	4	100	Barbados, Jamaica
	2010	3	6	67	
	2011	1	2	0	
	2012	4	7	57	
	2013	2	4	100	
	2014	2	2	100	
Europe	2009	29	55	89	Bulgaria, Czech Republic, Denmark (2), Germany (2), Greece, Hungary, Italy (8), Luxembourg (2), Malta, Poland (2), Serbia (2), Slovenia, Spain, Turkey
	2010	29	57	97	
	2011	25	48	85	
	2012	29	56	95	
	2013	26	51	88	
	2014	26	26	89	
North America	2009	10	19	90	Canada (6), USA (4)
	2010	11	22	86	
	2011	9	18	78	
	2012	13	26	96	
	2013	10	18	100	
	2014	10	10	100	
Oceania	2009	2	4	100	New Zealand
	2010	2	3	100	
	2011	2	4	100	
	2012	2	4	100	
	2013	2	4	100	
	2014	1	1	100	

Table 19 (continued). Region-based categorization of EQAS 2014 participating laboratories' performance of *Campylobacter* strains identification

Region	Year	No. of labs	No. of strains identified	% strains correctly identified	Countries participating in the 2014 iteration
Russia	2009	2	4	100	Russia (3)
	2010	2	4	100	
	2011	2	4	50	
	2012	5	10	80	
	2013	1	2	100	
	2014	3	3	100	
Latin America	2009	14	26	89	Argentina, Bolivia, Brazil (2), Chile (2), Colombia (3), Costa Rica, Ecuador, El Salvador, Guatemala, Mexico, Panama, Paraguay, Peru, Suriname, Uruguay (2), Venezuela (2)
	2010	19	37	78	
	2011	19	37	49	
	2012	22	40	95	
	2013	20	36	83	
	2014	22	22	86	
Southeast Asia	2009	10	20	90	Brunei Darussalam, Cambodia, Japan (2), LAO PDR, Malaysia, Philippines, Rep of Korea (2), Taiwan, Thailand (3)
	2010	14	27	93	
	2011	12	24	67	
	2012	17	33	85	
	2013	15	28	89	
	2014	13	13	92	
China	2009	12	24	92	China (8)
	2010	10	20	85	
	2011	-	-	-	
	2012	-	-	-	
	2013	5	10	90	
	2014	8	8	75	

Table 20. EQAS participants' performance of *Campylobacter* strains antimicrobial susceptibility testing

EQAS iteration	No. of labs	% correct test results	% major deviations (S → R)^	% very major deviations (R → S)^	% critical deviations (R → S & S → R)^
2009	25	91.4	4.5	4.1	8.6
2010	37	91.3	4.2	4.5	8.7
2011	38	93.8	2.8	3.4	6.2
2012	47	93.6	5.0	1.5	6.4
2013	47	92.4	5.0	2.6	7.6
2014	50	91.2	1.6	7.2	8.8

^S. susceptible; R. resistant

Table 21. Antimicrobial susceptibility test results (number of R/S) for the EQAS 2014 *Campylobacter* strains*

Strain	Antimicrobial^					
	CIP	ERY	GEN	NAL	STR	TET
WHO 2014 C-14.2	**45**/0/4	1/0/**45**	3/0/**42**	**37**/0/5	**20**/0/4	**40**/0/5

^For antimicrobial abbreviations. see List of Abbreviations page 1

*In bold: expected interpretation. Grey cell: <90% of laboratories did correct interpretation. R. resistant; S. susceptible

Table 22. EQAS participants' performance of *Campylobacter* antimicrobial susceptibility testing categorized by antimicrobial

EQAS iteration	No. of labs	Lab performance	Antimicrobial						
			CHL	CIP	ERY	GEN	NAL	STR	TET
2009	25	No. of tests	37	46	46	43	41	34	45
		% critical deviations*	8.1	6.5	10.9	2.3	9.8	11.8	11.1
2010	37	No. of tests	44	70	71	59	53	39	68
		% critical deviations*	4.5	7.1	11.3	10.2	7.5	10.3	8.8
2011	38	No. of tests	41	67	62	65	62	30	60
		% critical deviations*	0.0	6.0	6.5	3.1	8.1	13.3	8.3
2012	47	No. of tests	70	84	81	81	39	53	74
		% critical deviations*	4.3	6.0	6.2	7.4	5.1	11.3	5.4
2013	47	No. of tests	71	90	87	82	79	51	86
		% critical deviations*	5.6	6.7	8.0	0.0	8.9	23.5	8.1
2014	50	No. of tests	-	49	46	45	42	24	45
		% critical deviations*	-	8.2	2.2	6.7	11.9	16.7	11.1

^For antimicrobial abbreviations. see List of Abbreviations page 1

*R→ S & S → R (R. resistant; S. susceptible

Table 23. Region-based categorization of EQAS 2014 participants' performance of antimicrobial susceptibility testing of *Campylobacter* strains

Region	Year	No. of labs	% correct test result	% major deviations (S → R)^	% very major deviations (S → R)^	% critical deviations (R→S & S→R)^	Countries participating in the 2014 iteration
Africa	2009	2	75.0	10.7	14.3	25.0	Cameroun, Kenya (2), Madagascar, Nigeria, Senegal, Tunisia
	2010	2	95.2	0.0	4.8	4.8	
	2011	7	85.0	3.3	11.7	15.0	
	2012	4	94.3	0.0	5.7	5.7	
	2013	5	90.9	5.5	3.6	9.1	
	2014	7	51.5	39.4	9.1	48.5	
Central Asia & Middle East	2009	0	-	-	-	-	India, Islamic rep. of Iran, Israel
	2010	0	-	-	-	-	
	2011	1	75.0	0.0	25.0	25.0	
	2012	2	93.8	6.3	0.0	6.3	
	2013	3	93.3	3.3	3.3	6.7	
	2014	3	100.0	0.0	0.0	0.0	
China	2009	2	95.2	4.8	0.0	4.8	China (6)
	2010	1	100.0	0.0	0.0	0.0	
	2011	0	-	-	-	-	
	2012	2	88.5	7.7	3.8	11.5	
	2013	3	95.2	2.4	2.4	4.8	
	2014	6	100.0	0.0	0.0	0.0	
Caribbean	2009	0	-	-	-	-	Cuba, Jamaica
	2010	0	-	-	-	-	
	2011	0	-	-	-	-	
	2012	1	75.0	25.0	0.0	25.0	
	2013	1	100.0	0.0	0.0	0.0	
	2014	2	100.0	0.0	0.0	0.0	
Europe	2009	10	94.8	3.0	2.2	5.2	Denmark, Germany, Greece, Hungary, Italy (3), Luxembourg (2), Malta, Poland (2), Serbia, Slovenia, Spain, Turkey
	2010	13	100.0	0.0	0.0	0.0	
	2011	11	100.0	0.0	0.0	0.0	
	2012	16	97.3	1.6	1.1	2.7	
	2013	16	94.9	3.5	1.5	5.1	
	2014	16	97.4	1.3	1.3	2.6	
North America	2009	2	100.0	0.0	0.0	0.0	Canada, USA (3)
	2010	5	93.8	6.3	0.0	6.3	
	2011	5	100.0	0.0	0.0	0.0	
	2012	5	100.0	0.0	0.0	0.0	
	2013	3	100.0	0.0	0.0	0.0	
	2014	4	100.0	0.0	0.0	0.0	

^S. susceptible; R. resistant

Table 23 (continued). Region-based categorization of EQAS 2014 participants' performance of antimicrobial susceptibility testing of *Campylobacter* strains

Region	Year	No. of labs	% correct test result	% major deviations (S → R)^	% very major deviations (S → R)^	% critical deviations (R→S & S→R)^	Countries participating in the 2014 iteration
Oceania	2009	0	-	-	-	-	- none -
	2010	0	-	-	-	-	
	2011	1	100.0	0.0	0.0	0.0	
	2012	0	-	-	-	-	
	2013	0	-	-	-	-	
	2014	0	-	-	-	-	
Russia	2009	0	-	-	-	-	- none -
	2010	1	78.6	7.1	14.3	21.4	
	2011	1	100.0	0.0	0.0	0.0	
	2012	0	-	-	-	-	
	2013	0	-	-	-	-	
	2014	0	-	-	-	-	
Latin America	2009	5	93.2	6.8	0.0	6.8	Argentina, Brazil, Chile (2), Costa Rica, Paraguay
	2010	8	89.6	6.0	4.5	10.4	
	2011	7	96.8	0.0	3.2	3.2	
	2012	7	95.2	3.2	1.6	4.8	
	2013	7	92.4	4.5	3.0	7.6	
	2014	6	100.0	0.0	0.0	0.0	
Southeast Asia	2009	4	84.4	4.4	11.1	15.6	Philippines, Rep of Korea (2), Thailand (3)
	2010	7	77.2	9.8	13.0	22.9	
	2011	5	85.1	9.0	6.0	14.0	
	2012	10	85.8	13.3	0.9	14.2	
	2013	9	84.8	10.7	4.5	15.2	
	2014	6	87.5	12.5	0.0	12.5	

^S. susceptible; R. resistant

Table 24. EQAS participants' performance of antimicrobial susceptibility testing of *Campylobacter jejuni* ATCC 33560

	Method used	Incubation conditions	Labs' performance[1, 2]	Antimicrobial[3]					
				CHL	CIP	ERY	GEN	NAL	TET
EQAS 2010 (N=20)	Microdilution	42°C / 24h	**No.[1]**	3	6	6	6	4	6
			%[2]	67	83	100	83	75	83
	Microdilution	36-37°C / 48h	**No.[1]**	5	8	8	8	7	8
			%[2]	80	88	88	75	86	88
	Agardilution	42°C / 24h	**No.[1]**	-	6	6	6	-	-
			%[2]	-	100	83	83	-	-
	Agardilution	36-37°C / 48h	**No.[1]**	-	0	0	0	-	-
			%[2]	-	0	0	0	-	-
	Overall	Overall	**No.[1]**	8	20	20	20	11	14
			%[2]	75	90	90	80	82	86
EQAS 2011 (N=26)	Microdilution	42°C / 24h	**No.[1]**	4	9	9	8	7	9
			%[2]	100	67	100	88	100	67
	Microdilution	36-37°C / 48h	**No.[1]**	6	8	6	8	7	7
			%[2]	83	88	100	75	86	86
	Agardilution	42°C / 24h	**No.[1]**	-	8	8	8	-	-
			%[2]	-	88	63	100	-	-
	Agardilution	36-37°C / 48h	**No.[1]**	-	1	1	1	-	-
			%[2]	-	0	0	100	-	-
	Overall	Overall	**No.[1]**	10	26	24	25	14	16
			%[2]	90	77	83	88	93	75
EQAS 2012 (N=34)	Microdilution	42°C / 24h	**No.[1]**	9	12	12	12	10	12
			%[2]	67	75	83	83	80	75
	Microdilution	36-37°C / 48h	**No.[1]**	7	9	8	8	8	8
			%[2]	100	89	100	63	88	88
	Agardilution	42°C / 24h	**No.[1]**	-	9	7	9	-	-
			%[2]	-	89	86	89	-	-
	Agardilution	36-37°C / 48h	**No.[1]**	-	4	4	4	-	-
			%[2]	-	50	100	100	-	-
	Overall	Overall	**No.[1]**	34	80	75	78	43	50
			%[2]	82	81	88	83	86	80

[1]No.. number of labs performing the analysis, [2]%. percentage of labs reporting correct results, [3]For antimicrobial abbreviations: see List of Abbreviations page 1, -. not determined

Table 24 (continued). EQAS participants' performance of antimicrobial susceptibility testing of *Campylobacter jejuni* ATCC 33560

	Method used	Incubation conditions	Labs' performance[1, 2]	Antimicrobial[3]					
				CHL	CIP	ERY	GEN	NAL	TET
EQAS 2013 (N=47)	Microdilution	42°C / 24h	No.[1]	6	8	8	8	7	8
			%[2]	83	88	100	88	86	100
	Microdilution	36-37°C / 48h	No.[1]	8	12	12	11	11	12
			%[2]	88	92	83	73	91	75
	Agardilution	42°C / 24h	No.[1]	-	9	9	8	-	-
			%[2]	-	89	67	75	-	-
	Agardilution	36-37°C / 48h	No.[1]	-	7	7	6	-	-
			%[2]	-	86	86	100	-	-
	Overall	Overall	No.[1]	14	36	36	33	18	20
			%[2]	86	89	83	82	89	85
EQAS 2014 (N=32)	Microdilution	42°C / 24h	No.[1]	-	10	10	10	10	10
			%[2]	-	90	100	80	100	90
	Microdilution	36-37°C / 48h	No.[1]	-	10	10	9	8	10
			%[2]	-	100	80	89	100	100
	Agardilution	42°C / 24h	No.[1]	-	7	7	7	-	-
			%[2]	-	100	71	100	-	-
	Agardilution	36-37°C / 48h	No.[1]	-	5	5	5	-	-
			%[2]	-	80	80	100	-	-
	Overall	Overall	No.[1]	-	32	32	31	18	20
			%[2]	-	94	84	90	100	95

[1]No.. number of labs performing the analysis, [2]%. percentage of labs reporting correct results, [3]For antimicrobial abbreviations: see List of Abbreviations page 1, -. not determined

Table 25. EQAS participating laboratories' performance of unknown strain identification

EQAS iteration	Strain ID	No. of participating labs	Percentage (%) of labs performing correct identification
2003	*E. coli* O157	115	99
2004	*Shigella flexneri*	121	94 (*Shigella)* 74 (*S. flexneri)*
2006	*Yersinia enterocolitica* O3	134	93 (*Yersinia)* 89 (*Y. enterocolitica)* 66 (*Y. enterocolitica* O3)
2007	*Vibrio parahaemolyticus*	86	83
2008	*Enterobacter sakasakii*	128	92
2009	*Vibrio mimicus*	56	48
2010	*Citrobacter spp.*	115	90
2011	*Aeromonas hydrophila*	106	83
2012	*Salmonella* Paratyphi B var. Java	134	23% (*Salmonella* spp) 7% (*Salmonella* O:B) 24% (*Salmonella* Paratyphi B var. java. In total 54% Deviations: *Citrobacter freundii* (1), *Edwardsiella* sp (1), *Escherichia fergusonii* (1), *Proteus mirabilis* (1), *Salmonella* serovar X* (24), *Salmonella* serovar Paratyphi B (34) * incorrect serovar
2013	*E. coli* O157:H16 non-VTEC	129	82% including: *Escherichia coli* non-VTEC *Escherichia coli* O157 non-VTEC *Escherichia coli* O157:H16 non-VTEC *E. coli* non-VTEC *E. coli* O157 non-VTEC *E. coli* O157:H16 non-VTEC Deviations: *Escherichia coli* O157 H7 (9), *Escherichia hermannii* (2), *Shigella sonnei* (2), *E.coli* EHEC, *Escherichia coli* O114: nonmotile, *Escherichia coli* O157:H12, *Escherichia coli* O157:H16, Stx1+, *Escherichia coli* O157:H45, *Escherichia coli* O157:H7/ Verotoxin negative, *Escherichia fergusonii*, *Esherichia coli* STEC, *Vibrio mimicus*, *Citrobacter amalonaticus*
2014	*Yersinia pseudotuberculosis*	122	74% Correct, including: *Yersinia pseudotuberculosis* *Yersinia pseudotuberculosis* API 20 E [1014100] *Yersinia pseudotuberculosis* I *yersinia pseudotuberculosis* O:1b *Yersinia pseudotuberculosis* O1 *YERSINIA SPECIES* Deviations: *Acinetobacter baumannii, bacteria Sphingomonas paucimoilis, Burkolderia sp., Citrobacter freundi, corynebacterium species*, Gram negative *sphingomonas paucimobilis, HELICOBACTER, Pasteurella maisi, Pasteurella sp., Pseudomonas luteola, Rhizobium radiobacter* (5), *Salmonella typhi, Shigella flexineri, Sphingomonas paucimobilis* (4), unknown, *Vibrio metschnikovii, Yersinia enterocolitica* (4), *Yersinia similis, Yestina pestis*

M00-06-001/01.12.2011

Kgs. Lyngby, Denmark, April 2014

SIGN-UP FOR EQAS 2014

Greetings to the WHO Global Foodborne Infections Network (WHO GFN) Members:

WHO GFN strives to increase the quality of laboratory-based surveillance of *Salmonella* and other foodborne pathogens by encouraging national and regional reference laboratories that attended WHO GFN training courses to participate in the External Quality Assurance System (EQAS). The 2013 EQAS cycle is completed, and we are pleased to announce the launch of the 2014 EQAS cycle.

WHY PARTICIPATE IN EQAS?

EQAS provides the opportunity for proficiency testing which is considered an important tool for the production of reliable laboratory results of consistently good quality.

WHAT IS OFFERED IN EQAS?

This year, WHO EQAS offers the following components:

- Serogrouping, serotyping and antimicrobial susceptibility testing of eight *Salmonella* isolates;
- Serotyping and antimicrobial susceptibility testing of four *Shigella* isolates;
- Species identification and antimicrobial susceptibility testing of two *Campylobacter* isolates;
- Identification of one unknown bacterial isolate.

WHO SHOULD PARTICIPATE IN EQAS 2014?

All national and regional reference laboratories which perform analysis on *Salmonella*, *Shigella* and/or *Campylobacter* and are interested in participating in an external quality assurance program are invited to participate.
We expect that all national and regional reference laboratories that attended WHO GFN Training Courses will participate in EQAS.
The WHO GFN Regional Centers in cooperation with the EQAS Coordinator will evaluate the list of laboratories that sign up for EQAS 2014. Laboratories which signed up and received bacterial isolates in year 2013 but did not submit any result should provide a consistent explanation for this if they want to participate in 2014.

COST FOR PARTICIPATING IN EQAS

There is no participation fee in EQAS 2014. Laboratories should, however, cover the expenses for parcel shipment if they can afford it. If FedEx has ‘Dangerous Goods-service’ in your country or if you have a DHL-account no, please provide your FedEx or DHL import account number (for import of UN3373 Biological Substance Category B) in the sign-up form or, alternatively, to the EQAS Coordinator (please find contact information below). We need this information at this stage to save time and resources. Participating laboratories are responsible for paying any expenses related to taxes or custom fees applied by their country.

HOW TO SIGN- UP FOR EQAS 2014

This link will open a sign-up webpage: http://eqas.food.dtu.dk/who/signup

In this webpage, you will be asked to provide the following information:

- Name of institute, department, laboratory, and contact person
- Complete mailing address for shipment of bacterial isolates (no post-office box number)
- Telephone and fax number, e-mail address
- FedEx or DHL import account number (if available)
- Approximate number of *Salmonella* isolates annually serogrouped/serotyped
- Approximate number of *Salmonella* isolates annually tested for antimicrobial susceptibility
- Availability of ATCC reference strains
- Components of EQAS 2014 you plan to participate in
- Level of reference function in your country

If you experience any problem in the sign-up webpage, please try again a few days later. If problems persist after several attempts, please contact the EQAS Coordinator Susanne Karlsmose: E-mail suska@food.dtu.dk; fax +45 3588 6341.

TIMELINE FOR SHIPMENT OF ISOLATES AND AVAILABILITY OF PROTOCOLS

Due to increased number of participants in WHO EQAS, a number of different institutions will ship the bacterial isolates, and you will receive information concerning the institution shipping your parcel. The bacterial isolates will be shipped between August and September 2014.

In order to minimize delays, **please send a valid import permit to the EQAS coordinator**. Please apply for a permit to receive the following (according to your level of participation): "UN3373, Biological Substance Category B": eight *Salmonella* strains, four *Shigella* strains, two *Campylobacter*, one *Campylobacter* reference strain (for new participants performing antimicrobial susceptibility testing on *Campylobacter*), one *Escherichia coli* reference strain (for new participants performing antimicrobial susceptibility testing on *Salmonella* and/or *Shigella*) and an unknown isolate (enteric bacteria) between August and September 2014.

Protocols and all relevant information will be available for download from the website http://www.antimicrobialresistance.dk/233-169-215-eqas.htm.

DEADLINE FOR SUBMITTING RESULTS TO THE NATIONAL FOOD INSTITUTE

Results must be submitted to the National Food Institute (DTU Food) by **31st December 2014** through the password-protected website. An evaluation report will be generated upon submission of results. Full anonymity is ensured, and only DTU Food and the WHO GFN Regional Centre in your region will have access to your results.

Deadline for sign-up for EQAS 2014 is 30th May 2014

			Ampicillin		Cefotaxime		Ceftazidime		Ceftriaxone		Chloramphenicol		Ciprofloxacin		Gentamicin		Nalidixic acid		Sulfonamides		Tetracycline		Trimethoprim		Trim/Sulfa	
			AMP		**CTX**		**CAZ**		**CRO**		**CHL**		**CIP**		**GEN**		**NAL**		**SMX**		**TET**		**TMP**		**SXT**	
WHO 2014 S-14.1	*Salmonella* Orion / Orion var. 15	I 3,15:y:1,5	<= 1	SUSC	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	<= 0.015	SUSC	<= 0.5	SUSC	<= 4	SUSC	= 32	SUSC	<= 2	SUSC	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 S-14.2	*Salmonella* Hadar / Istanbul	I 8:z10:e,n,x	<= 1	SUSC	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	= 0.5	INTER	<= 0.5	SUSC	> 128	RESIST	= 32	SUSC	= 64	RESIST	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 S-14.3	*Salmonella* IIIa 48:g,z51:-	IIIa 48:g,z51:-	<= 1	SUSC	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	= 0.03	SUSC	<= 0.5	SUSC	= 8	SUSC	= 16	SUSC	<= 2	SUSC	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 S-14.4	*Salmonella* Napoli	I 9,12:l,z13:e,n,x	> 64	RESIST	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	= 0.03	SUSC	<= 0.5	SUSC	<= 4	SUSC	> 1024	RESIST	> 64	RESIST	> 32	RESIST	> 4	RESIST
WHO 2014 S-14.5	*Salmonella* Ohio	I 6,7:b:l,w	<= 1	SUSC	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	= 0.03	SUSC	<= 0.5	SUSC	<= 4	SUSC	= 64	SUSC	<= 2	SUSC	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 S-14.6	*Salmonella* Enteritidis	I 9,12:g,m:-	= 2	SUSC	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	= 0.03	SUSC	> 32	RESIST	<= 4	SUSC	> 1024	RESIST	<= 2	SUSC	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 S-14.7	*Salmonella* Typhimurium	I 4,12:i:1,2	> 64	RESIST	> 4	RESIST	> 8	RESIST	> 64	RESIST	> 128	RESIST	= 1	RESIST	> 32	RESIST	> 128	RESIST	> 1024	RESIST	> 64	RESIST	> 32	RESIST	> 4	RESIST
WHO 2014 S-14.8	*Salmonella* Kentucky	I 8:i:z6	> 64	RESIST	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	> 8	RESIST	= 16	RESIST	> 128	RESIST	> 1024	RESIST	= 64	RESIST	<= 0.25	SUSC	<= 0.12	SUSC
WHO 2014 SH-14.1	sonnei		> 64	RESIST	> 4	RESIST	= 4	RESIST	= 64	RESIST	<= 8	SUSC	= 0.12	INTER	= 1	SUSC	= 64	RESIST	> 1024	RESIST	= 64	RESIST	> 32	RESIST	> 4	RESIST
WHO 2014 SH-14.2	sonnei		> 64	RESIST	> 4	RESIST	= 2	RESIST	= 64	RESIST	<= 8	SUSC	<= 0.015	SUSC	> 32	RESIST	<= 4	SUSC	> 1024	RESIST	= 64	RESIST	> 32	RESIST	> 4	RESIST
WHO 2014 SH-14.3	flexneri 2b		> 64	RESIST	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	= 32	RESIST	= 1	RESIST	= 1	SUSC	> 128	RESIST	> 1024	RESIST	= 64	RESIST	> 32	RESIST	> 4	RESIST
WHO 2014 SH-14.4	boydii 2		> 64	RESIST	<= 0.25	SUSC	<= 0.5	SUSC	<= 0.25	SUSC	<= 8	SUSC	<= 0.015	SUSC	<= 0.5	SUSC	<= 4	SUSC	> 1024	RESIST	= 32	RESIST	<= 0.25	SUSC	= 0.25	SUSC

			Ciprofloxacin		Erythromycin		Gentamicin		Nalidixic acid		Streptomycin		Tetracycline	
			CIP		**ERY**		**GEN**		**NAL**		**STR**		**TET**	
WHO 2014 C-14.1	*No strain, this year, with this code (due to problems with the lyophilization)*													
WHO 2014 C-14.2	*C. coli*		= 8	RESIST	<= 1	SUSC	<= 0.12	SUSC	= 64	RESIST	> 16	RESIST	> 64	RESIST

WHO B-14.1	*Yersinia pseudotuberculosis*

WHO Collaborating Centre
External Quality Assurance System (EQAS) 2014

DTU Food
National Food Institute

WHO Collaborating Centre
for Antimicrobial Resistance in
Foodborne Pathogens
www.antimicrobialresistance.dk

PROTOCOL for

- serotyping and antimicrobial susceptibility testing of *Salmonella*
- serotyping and antimicrobial susceptibility testing of *Shigella*
- identification and antimicrobial susceptibility testing of *Campylobacter*
- identification of an unknown enteric pathogen

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1 INTRODUCTION

In 2000, the Global Foodborne Infections Network (formerly known as WHO Global Salm-Surv) launched an External Quality Assurance System (EQAS). The EQAS is organized by the National Food Institute, Technical University of Denmark (DTU Food), in collaboration with partners and Regional Sites in WHO GFN.

Various aspects of the proficiency test scheme may from time to time be subcontracted. When subcontracting occurs, it is placed with a competent subcontractor and the National Food Institute is responsible for the subcontractor's work.

The WHO EQAS 2014 includes

- serotyping and antimicrobial susceptibility testing of eight *Salmonella* strains,
- serotyping and antimicrobial susceptibility testing of four *Shigella* strains,

Technical University of Denmark DTU

- antimicrobial susceptibility testing of the *Escherichia coli* ATCC 25922 (CCM 3954) reference strain for quality control,
- identification and antimicrobial susceptibility testing of one thermophilic *Campylobacter* isolate (note, this year, only one *Campylobacter* strain is included, due to unfortunate issues with the lyophilisation of the strains),
- antimicrobial susceptibility testing of *Campylobacter jejuni* ATCC 33560 (CCM 6214) reference strain for quality control,
- identification of one 'unknown' bacterial isolate.

All participants will receive the strains according to the information they reported in the sign-up form.

The above-mentioned reference strains are included in the parcel only for new participants of the EQAS who did not receive them previously. The reference strains are original CERTIFIED cultures provided free of charge, and should be used for future internal quality control for antimicrobial susceptibility testing in your laboratory. The reference strains will not be included in the years to come. Therefore, please take proper care of these strains. Handle and maintain them as suggested in the manual 'Subculture and Maintenance of QC Strains' available on the WHO Collaborating Centre website (see www.antimicrobialresistance.dk).

2 OBJECTIVES

The main objective of this EQAS is to support laboratories to assess and if necessary improve the quality of serotyping and antimicrobial susceptibility testing of enteric human pathogens, especially *Salmonella*. A further objective is to assess and improve the comparability of surveillance data on *Salmonella* serotypes and antimicrobial susceptibility reported by different laboratories. Therefore, the laboratory work for this EQAS should be done by using the methods routinely used in your laboratory.

3 OUTLINE OF THE EQAS 2014

3.1 Shipping, receipt and storage of strains

In September 2014 around 200 laboratories located worldwide will receive a parcel containing eight *Salmonella* strains, four *Shigella* strains, one *Campylobacter* strain and one 'unknown' bacterial isolate (according to information reported in the sign-up form). An *E. coli* ATCC 25922 reference strain and a *C. jejuni* ATCC 33560 reference strain will be included for participants who signed up to perform antimicrobial susceptibility testing (AST) and did not receive them previously. All provided strains belong to UN3373, Biological substance category B. ESBL-producing strains could be included in the selected material.

Please confirm receipt of the parcel through the confirmation form enclosed in the shipment

The *Salmonella* and *Shigella* strains, and the 'unknown' bacterial isolate are shipped as agar stab cultures whereas the reference strains and the *Campylobacter* strain are shipped lyophilised. On arrival, the agar stab cultures must be subcultured and prepared for storage in your strain collection (e.g. in a -80°C freezer). This set of cultures should serve as reference if discrepancies are detected during the testing (e.g. they can be used to detect errors such as mis-labelling or contamination). Lyophilised strains must be reconstituted, and you can find below a suggested procedure.

3.2 Serotyping of *Salmonella*

The eight *Salmonella* strains should be serotyped by using the method routinely used in the laboratory. If you do not have all the necessary antisera please go as far as you can in the identification and report the serogroup, since also serogroup results will be evaluated. Serogroups should be reported using terms according to Kauffmann-White-Le Minor (Grimont and Weill, 2007. 9th ed. Antigenic formulae of the *Salmonella* serovars. WHO Collaborating Centre for Reference and Research on *Salmonella*).

Please fill in information concerning the brand of antisera used for typing in the fields available in the database for entering results. In addition, we kindly ask you to report which antisera you think are required to complete the serotyping, if relevant.

3.3 Antimicrobial susceptibility testing of *Salmonella, Shigella* and *Escherichia coli* ATCC 25922

The *Salmonella* and *Shigella* strains as well as the *E. coli* ATCC 25922 reference strain should be tested for susceptibility towards as many as possible of the antimicrobials mentioned in the test form. Please use the methods routinely used in your laboratory.

For reconstitution of the *E. coli* reference strain, please see the document 'Instructions for opening and reviving lyophilised cultures' on the WHO Collaborating Centre website (see www.antimicrobialresistance.dk).

Testing of gentamicin susceptibility may be valuable for monitoring purposes. Therefore we kindly ask you to disregard, for the purpose of this proficiency trial, that the Clinical and Laboratory Standards Institute (CLSI) guidelines state that *Salmonella* and *Shigella* should not be reported as susceptible to aminoglycosides.

The breakpoints used in this EQAS for interpreting MIC results are in accordance with CLSI values (Table 1). Consequently, interpretation of MIC results will lead to categorization of strains into three categories: resistant (R), intermediate (I) and susceptible (S). In the evaluation report you

receive upon result submission, you can find that obtained interpretations in accordance with the expected interpretation will be defined as 'correct', whereas deviations from the expected interpretation will be defined as 'minor' (I ↔ S or I ↔ R), 'major' (S interpreted as R) or 'very major' (R interpreted as S).

Please report the breakpoints that you routinely use in your laboratory for interpretation of antimicrobial susceptibility test results in the fields available in the database (or in the test forms).

Table 1. Interpretive breakpoint for *Salmonella* and *Shigell*a antimicrobial susceptibility testing

Antimicrobials	Reference value, MIC (µg/mL)			Reference value, Disk diffusion (mm)		
	Sensitive	Intermediate	Resistant	Resistant	Intermediate	Sensitive
Ampicillin, AMP	≤8	16	≥32	≤13	14-16	≥17
Cefotaxime, CTX*	≤1	-	>1	≤27	-	>27
Ceftazidime, CAZ*	≤1	-	>1	≤22	-	>22
Ceftriaxone, CRO*	≤1	-	>1	≤25	-	>25
Chloramphenicol, CHL	≤8	16	≥32	≤12	13-17	≥18
Ciprofloxacin, CIP	≤0.06**	0.12-0.5**	≥1**	≤20mm (5µg)** or <23mm (1µg)***	21-30mm (5µg)** or - (1µg)***	≥31mm (5µg)** or ≥23mm (1µg)***
Gentamicin, GEN	≤4	8	≥16	≤12	13-14	≥15
Nalidixic acid, NAL	≤16	-	≥32	≤13	14-18	≥19
Sulfonamides, SMX	≤256	-	≥512	≤12	13-16	≥17
Tetracycline, TET	≤4	8	≥16	≤11	12-14	≥15
Trimethoprim, TMP	≤8	-	≥16	≤10	11-15	≥16
Trimethoprim + sulfamethoxazole, TMP+SMX, SXT	≤2/38	-	≥4/76	≤10	11-15	≥16

Reference values used in this EQAS are according to CLSI (M100-S24), with the following exceptions:
* Reference values are according to CLSI M100-S24 Table 3A. These interpretative criteria are also applied for *Salmonella* and *Shigella* test strains for interpretation of AST results in this EQAS
** These breakpoints should also be applied for *Shigella* test strains for interpretation of AST results in this EQAS
*** The publication by Cavaco LM and Aarestrup FM (J. Clin. Microbiol. 2009. Sep;47(9):2751-8) provides the background for these interpretative criteria in the WHO GFN EQAS. These interpretative criteria are also applied for *Shigella* test strains for interpretation of AST results in this EQAS.

Concerning ciprofloxacin susceptibility tests, please note that for results obtained in this proficiency test, the breakpoints for *Salmonella* are applied for *Shigella* also. These breakpoints for ciprofloxacin take into consideration mechanisms of resistance due to plasmid-mediated quinolone resistance genes (e.g. *qnr*-genes) and one-point-mutation in the gyrase gene.

Important notes*: beta-lactam resistance*
The following tests for detection of Extended-Spectrum Beta-Lactamase (ESBL) production are optional.

All strains showing reduced susceptibility to cefotaxime (CTX), ceftazidime (CAZ) and/or ceftriaxone (CRO) could be tested for ESBL production by confirmatory test. Confirmatory test for ESBL production requires use of both cefotaxime (CTX) and ceftazidime (CAZ) alone, and in combination with a β-lactamase inhibitor (clavulanic acid). Synergy is defined either as i) a ≥ 3 twofold concentration decrease in an MIC for either antimicrobial agent tested in combination with clavulanic acid vs. its MIC when tested alone (E-test 3 dilution steps difference; MIC CTX : CTX/CL or CAZ : CAZ/CL ratio ≥ 8) or ii) a ≥ 5 mm increase in a zone diameter for either antimicrobial agent tested in combination with clavulanic acid vs. its zone when tested alone (CLSI M100 Table 2A; Enterobacteriaceae). The presence of synergy indicates ESBL production.

Of note, MIC values and relative interpretation of cefotaxime (CTX), ceftazidime (CAZ) and/or ceftriaxone (CRO) used for detection of beta-lactamase-producing strains in this EQAS should be reported as found.

3.4 Handling the *Campylobacter* strains

Lyophilised cultures are supplied in vacuum-sealed ampoules. Care should be taken in opening the ampoule, and all instructions given below should be followed closely to ensure the safety of the person who opens the ampoule and to prevent contamination of the culture.

a. Check the number of the culture on the label inside the ampoule
b. Make a file cut on the ampoule near the middle of the plug
c. Disinfect the ampoule with alcohol-dampened gauze or alcohol-dampened cotton wool from just below the plug to the pointed end
d. Apply a red-hot glass rod to the file cut to crack the glass and allow air to enter slowly into the ampoule
e. Remove the pointed end of the ampoule into disinfectant
f. Add about 0.3 ml appropriate broth to the dried suspension using a sterile Pasteur pipette and mix carefully to avoid creating aerosols. Transfer the contents to one or more suitable solid and /or liquid media

g. Transfer the rest of the content of the ampoule to a test tube containing 5-6 ml of a suitable liquid media.
h. Incubate the agar plate and liquid media at a temperature of 42°C at microaerobic conditions for 24-48 hours.
i. Autoclave or disinfect effectively the used Pasteur pipette, the plug and all the remains of the original ampoule before discarding
j. Inoculate a second agar plate from the liquid media with a 10µl loop or a cotton swab if the initial plate had inadequate growth.
k. Select a pure culture with vigorous growth from the agar plate for further work.

Please note that:

- Cultures may need at least one subculture before they can be optimally used
- Unopened ampoules should be kept in a dark and cool place!

For reconstitution of *C. jejuni* ATCC33560 reference strain, please see the document 'Instructions for opening and reviving lyophilised cultures' on the WHO Collaborating Centre website (see www.antimicrobialresistance.dk).

3.5 Identification of *Campylobacter*

The thermophilic *Campylobacter* isolate should be identified to species level.

3.6 Antimicrobial susceptibility testing of *Campylobacter*

The *Campylobacter* test strain and the *C. jejuni* reference strain ATCC33560 should be tested for susceptibility to as many antimicrobials as possible among the ones mentioned in the test form. It should be noted that only MIC methods (i.e. broth or agar dilution methods) are recommendable for AST of *Campylobacter*. Neither the use of disk diffusion nor E-test is recommendable for AST of *Campylobacter*.

In this EQAS, the breakpoints used for interpretation of MIC results for *Campylobacter* are epidemiological cut-off values according to EUCAST (European Committee on Antimicrobial Susceptibility Testing; www.eucast.org; Table 2). Consequently, only two categories of characterisation (resistant, R or susceptible, S) are allowed. In the evaluation report that you receive upon result submission, you can find that obtained interpretations in agreement with the expected interpretation, will be categorised as 'correct', whereas deviations from the expected interpretation will be categorizes as 'incorrect'.

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Please report the breakpoints that you routinely use in your laboratory for interpretation of antimicrobial susceptibility test results, in the fields available in the database (or in the test form).

Note that the interpretation of antimicrobial susceptibility test results for *Campylobacter* requires knowledge of the *Campylobacter* species. If you did not sign-up for *Campylobacter* identification, but perform AST on *Campylobacter*, you are welcome to contact the EQAS Coordinator to obtain information regarding the identity of the *Campylobacter* test strain.

Table 2. Interpretive criteria for *Campylobacter* antimicrobial susceptibility testing

Antimicrobials for *Campylobacter*	MIC (µg/mL) **R is >** *C. jejuni*	MIC (µg/mL) **R is >** *C. coli*
Ciprofloxacin, CIP	0.5	0.5
Erythromycin, ERY	4	8
Gentamicin, GEN	2	2
Nalidixic acid, NAL	16	16
Streptomycin, STR	4	4
Tetracycline, TET	1	2

Reference values for interpretation of *Campylobacter* AST results according to EUCAST

The sub-cultured *Campylobacter* strains should be used for MIC-testing after incubation at 36-37ºC for 48 hours or at 42ºC for 24 hours. Likely, two subcultures are needed prior to MIC-testing to ensure optimal growth.

3.7 Identification of the unknown environmental bacterium

The 'unknown' isolate should be identified to species level and further typed if relevant.

4 REPORTING OF RESULTS AND EVALUATION

We recommend that you write your results in the enclosed test forms and that you read carefully the description in paragraph 5 before entering your results in the web database. For entering your results via the web, you will be guided through all steps on the screen and you will immediately be able to view and print a report evaluating your results. Results in agreement with the expected interpretation are categorised as 'correct', while results deviating from the expected interpretation are categorised as 'incorrect'.

Results must be submitted no later than 31 December 2014.

If you do not have access to the Internet, or if you experience difficulties in entering your results, please return the completed test forms by e-mail, fax or mail to the National Food Institute, Denmark.

All results will be summarized in a report which will be publicly available. Individual results will be anonymous and will only be forwarded to the official GFN Regional Centre in your region.

We are looking forward to receiving your results.

If you have any questions or concerns, please do not hesitate to contact the EQAS Coordinator:

Susanne Karlsmose

National Food Institute, Technical University of Denmark

Kemitorvet, Building 204 ground floor, DK-2800 Lyngby - DENMARK

Tel: +45 3588 6601, Fax: +45 3588 6341

E-mail: suska@food.dtu.dk

It is possible to communicate with the EQAS organisers in other languages than English. However, this is not a direct contact with the EQAS organisers since translation of the message is required. The following languages may be used: Chinese, French, Portuguese, Russian and Spanish.

5 HOW TO ENTER RESULTS IN THE INTERACTIVE DATABASE

Please carefully read these instructions before entering the web page. Remember that you need by your side the completed test forms and the breakpoint values you used.

In general, you can browse back and forth in the pages of the database. Always remember to save your input before leaving a page.

1) Enter the WHO Collaborating Centre website (from http://www.antimicrobialresistance.dk), then
    a. Click on 'EQAS'
    b. Click on the link for the interactive database (http://eqas.food.dtu.dk/who)
    c. Write your username and password in lower-case letters and click on 'Login'.
    You can find your username and password in the letter following your strains.
    Your username and password will remain unchanged in future trials. Do not hesitate to contact us if you experience problems with the login.

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2) Click on 'Materials and methods'
   a. Fill in the fields relative to brand of antisera (very important because we would like to compare results obtained with different brands of antisera)
   b. Fill in the fields relative to the method used for antimicrobial susceptibility testing
   c. Enter the brand of materials, e.g. Oxoid
   d. Fill in the field asking whether your institute serves as a national reference laboratory
   e. In the comment field, report which antisera you think is required to complete your serotyping, if relevant
   f. Click on 'Save and go to next page' – ALWAYS remember to save each page before leaving it!

3) In the data entry page 'Routinely used breakpoints'
   a. Fill in the fields relative to the breakpoints used routinely in your laboratory to determine the antimicrobial susceptibility category. Remember to use the operator keys in order to show – equal to (=), less than (<), less or equal to(≤), greater than (>) or greater than or equal to (≥).

4) In the data entry pages '*Salmonella* strains 1-8',
   a. SELECT the serogroup (O-group) from the drop-down list, DO NOT WRITE – Wait a few seconds – the page will automatically reload, so that the drop-down list in the field "Serotype" only contains serotypes belonging to the chosen serogroup.
   b. SELECT the serotype from the drop-down list – DO NOT WRITE – wait a few seconds and you can enter the antigenic formula (e.g. 1,4,5,12:i:1,2)
   c. Enter the zone diameters in mm or MIC values in µg/ml. Remember to use the operator keys to show e.g. equal to (=), etc.
   d. Enter the interpretation as R (resistant), I (intermediate) or S (susceptible)
   e. If you performed confirmatory tests for ESBL production, select the appropriate result.
   f. If relevant, fill in the field related to comments (e.g. which antisera you miss for complete serotyping)
   g. Click on 'Save and go to next page'

   If you did not perform these tests, please leave the fields empty

5) In the data entry page '*E. coli* reference strain':
   a. Enter the zone diameters in mm or MIC values in µg/ml. Remember to use the operator keys to show e.g. equal to (=), etc.
   b. Click on 'Save and go to next page'

6) In the page 'Identification of *Campylobacter* and unknown sample':
   a. Choose the correct *Campylobacter* species from the pick list

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b. Fill in the field concerning species and type of the unknown bacterial isolate, and report the method used for identification
c. Click on 'Save and go to next page'

If you did not perform these tests, please leave the fields empty

7) The next page is a menu that allows you to review the input pages and approve your input *and finally see and print the evaluated results*
   a. Browse through the input pages and make corrections if necessary. Remember to click on 'save and go to next page' if you make any corrections.
   b. Approve your input. Be sure that you have filled in all the results before approval, as YOU CAN ONLY APPROVE ONCE! The approval blocks your data entry into the interactive database, but allows you to see the evaluated results.
   c. As soon as you have approved your input, an evaluation report will appear.

8) After browsing all pages in the report, you will find a new menu. You can choose 'EQAS 20xx start page', 'Review evaluated results' (a printer friendly version of the evaluation report is also available) or 'Go to WHO GFN homepage'.

End of entering your data – thank you very much!

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SUBCULTURE AND MAINTENANCE OF QUALITY CONTROL STRAINS

1.1 Purpose

Improper storage and repeated subculturing of bacteria can produce alterations in antimicrobial susceptibility test results. The Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) has published a guideline for Quality Control (QC) stock culture maintenance to ensure consistent antimicrobial susceptibility test results.

1.2 References

M100-S21, January 2011 (Performance Standards for Antimicrobial Susceptibility Testing)

M7-A8, January 2009 (Methods for Dilution Antimicrobial Susceptibility Test for Bacteria That Grow Aerobically; Approved Standard)

1.3 Definition of Terms

Reference Culture: A reference culture is a microorganism preparation that is acquired from a culture type collection.

Reference Stock Culture: A reference stock culture is a microorganism preparation that is derived from a reference culture. Guidelines and standards outline how reference stock cultures must be processed and stored.

Working Stock Cultures: A working stock culture is growth derived from a reference stock culture. Guidelines and standards outline how working stock cultures must be processed and how often they can be subcultured.

Subcultures (Passages): A subculture is simply the transfer of established microorganism growth on media to fresh media. The subsequent growth on the fresh media constitutes a subculture or passage. Growing a reference culture or reference stock culture from its preserved status (frozen or lyophilized) is not a subculture. The preserved microorganism is not in a stage of established growth until it is thawed or hydrated and grown for the first time

1.4 Important Considerations

- Do not use disc diffusion strains for MIC determination.
- Obtain QC strains from a reliable source such as ATCC
- CLSI requires that QC be performed either on the same day or weekly (only after 30 day QC validation)
- Any changes in materials or procedure must be validated with QC before implemented
- For example: Agar and broth methods may give different QC ranges for drugs such as glycopeptides, aminoglycosides and macrolides

- Periodically perform colony counts to check the inoculum preparation procedure
- Ideally, test values should be in the middle of the acceptable range
- Graphing QC data points over time can help identify changes in data helpful for troubleshooting problems

1.5 Storage of Reference Strains

Preparation of stock cultures

- Use a suitable stabilizer such as 50% fetal calf serum in broth, 10-15% glycerol in tryptic soy broth, defibrinated sheep blood or skim milk to prepare multiple aliquots.
- Store at -20°C, -70°C or liquid nitrogen. (Alternatively, freeze dry.)
- Before using rejuvenated strains for QC, subculture to check for purity and viability.

Working cultures

- Set up on agar slants with appropriate medium, store at 4-8°C and subculture weekly.
- Replace the working strain with a stock culture at least monthly.
- If a change in the organisms inherent susceptibility occurs, obtain a fresh stock culture or a new strain from a reference culture collection e.g. ATCC.

1.6 Frequency of Testing

Weekly vs. daily testing

Weekly testing is possible if the lab can demonstrate satisfactory performance with daily testing as follows:

- Documentation showing reference strain results from 30 consecutive test days were within the acceptable range.
- For each antimicrobial/organism combination, no more than 3 out of 30 MIC values may be outside the acceptable range.

When the above are fulfilled, each quality control strain may be tested once a week and whenever any reagent component is changed.

Corrective Actions

If an MIC is outside the range in weekly testing, corrective action is required as follows:

- Repeat the test if there is an obvious error e.g. wrong strain or incubation conditions used
- If there is no obvious error, return to daily control testing

The problem is considered resolved only after the reference strain is tested for 5 consecutive days and each drug/organism result is within specification on each day.

If the problem cannot be resolved, continue daily testing until the errors are identified.

Repeat the 30 days validation before resuming weekly testing.

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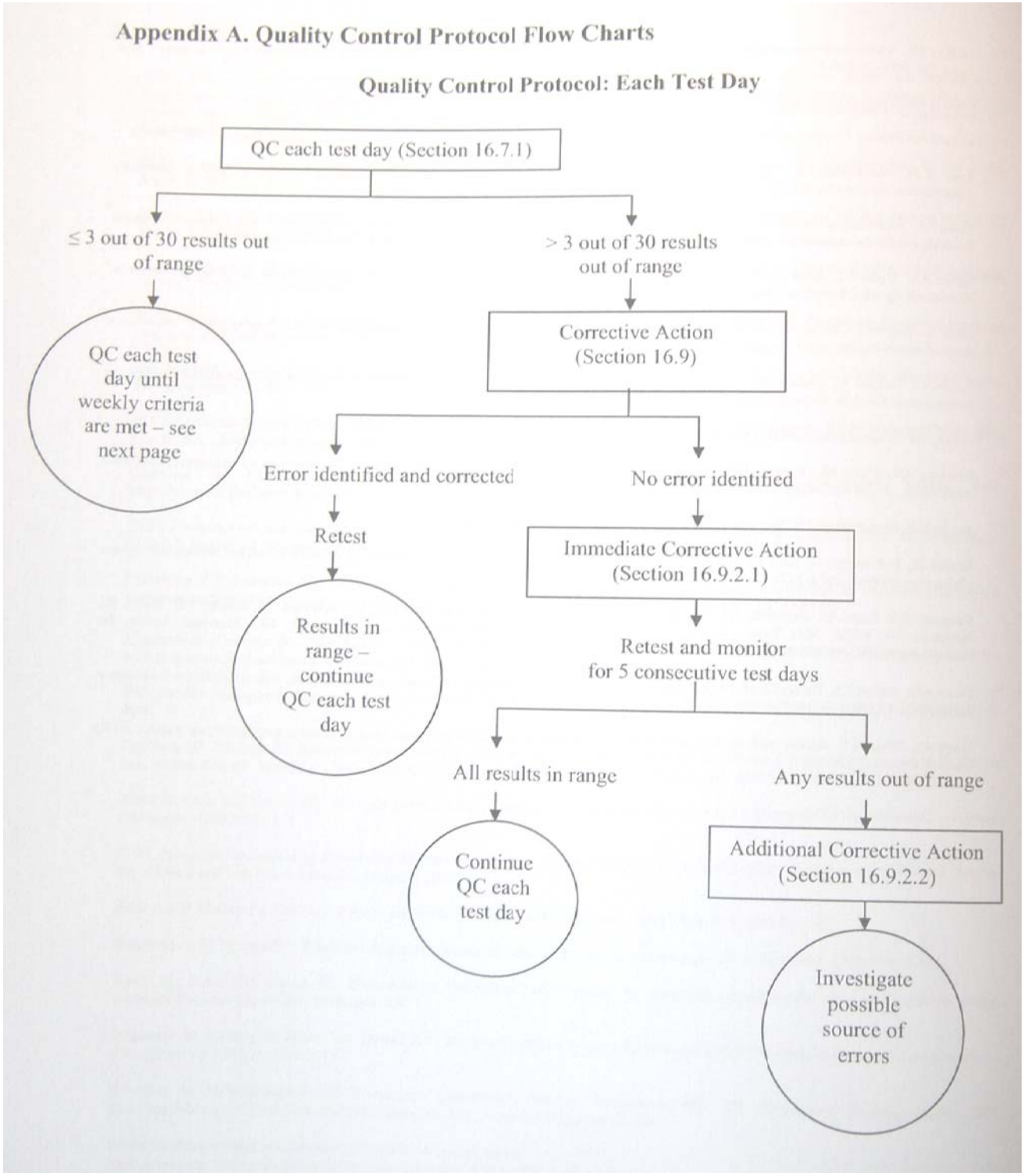
DAILY MIC QC CHART



Reference: CLSI M7-A8, page 44

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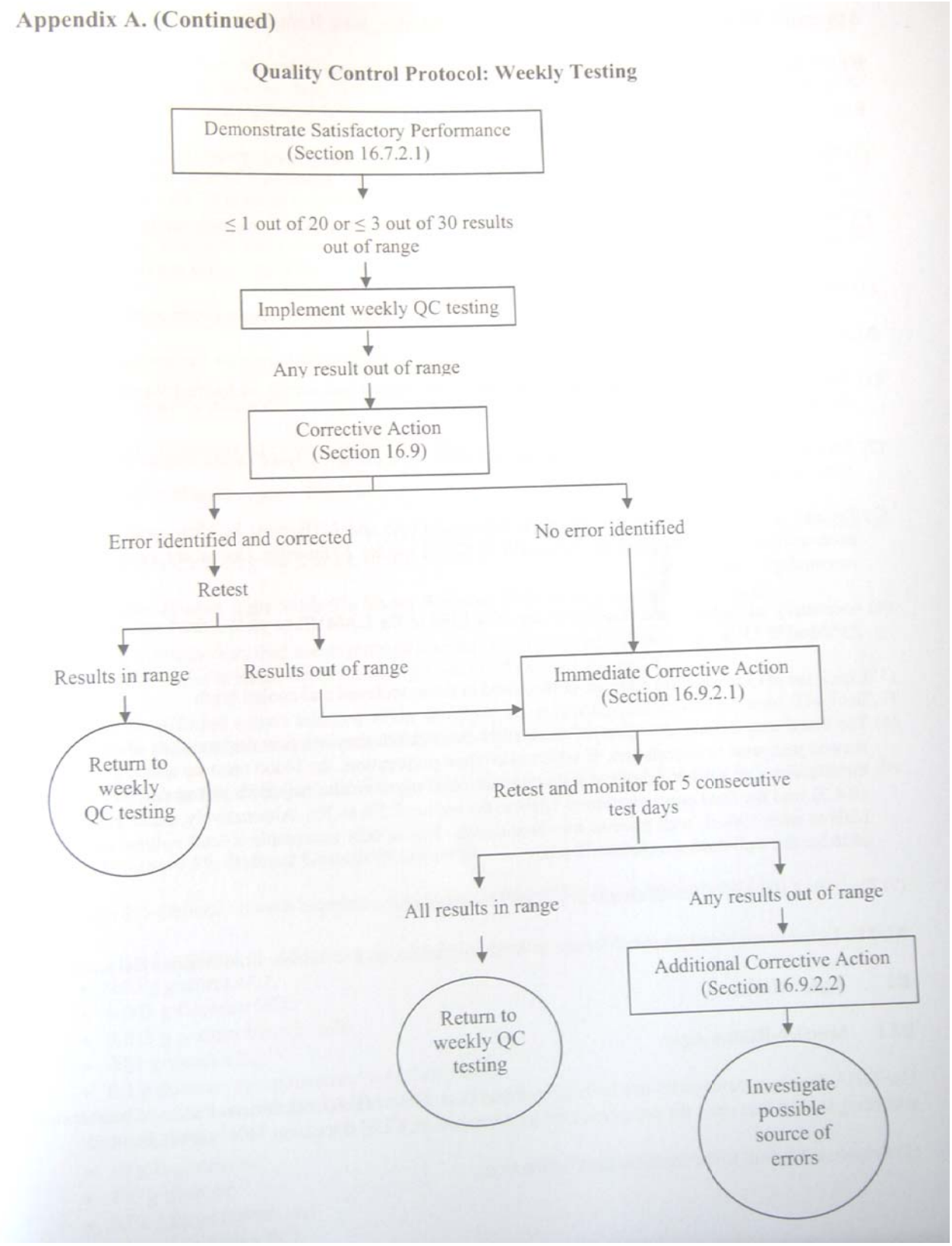
WEEKLY MIC QC CHART

Appendix A. (Continued)



Reference: CLSI M7-A8, page 45

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INSTRUCTIONS FOR OPENING AND REVIVING LYOPHILISED CULTURES

Manual from Czech Collection of Microorganisms (CCM)
Masaryk University
Tvrdého 14
602 00 BRNO
Czech Republic

Lyophilised cultures are supplied in vacuum-sealed ampoules. Care should be taken in opening the ampoule. All instructions given below should be followed closely to ensure the safety of the person who opens the ampoule and to prevent contamination of the culture.

a. Check the number of the culture on the label inside the ampoule
b. Make a file cut on the ampoule near the middle of the plug
c. Disinfect the ampoule with alcohol-dampened gauze or alcohol-dampened cotton wool from just below the plug to the pointed end
d. Apply a red-hot glass rod to the file cut to crack the glass and allow air to enter slowly into the ampoule
e. Remove the pointed end of the ampoule into disinfectant
f. Add about 0.3 ml appropriate broth to the dried suspension using a sterile Pasteur pipette and mix carefully to avoid creating aerosols. Transfer the contents to one or more suitable solid and /or liquid media
g. Incubate the inoculated medium at appropriate conditions for several days
h. Autoclave or disinfect effectively the used Pasteur pipette, the plug and all the remains of the original ampoule before discarding

Please note that:

- Cultures should be grown on media and under conditions as recommended in the CCM catalogue
- Cultures may need at least one subculturing before they can be optimally used in experiments
- Unopened ampoules should be kept in a dark and cool place!

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