JVARM and NVAL's activities for capacity building on AMR surveillance

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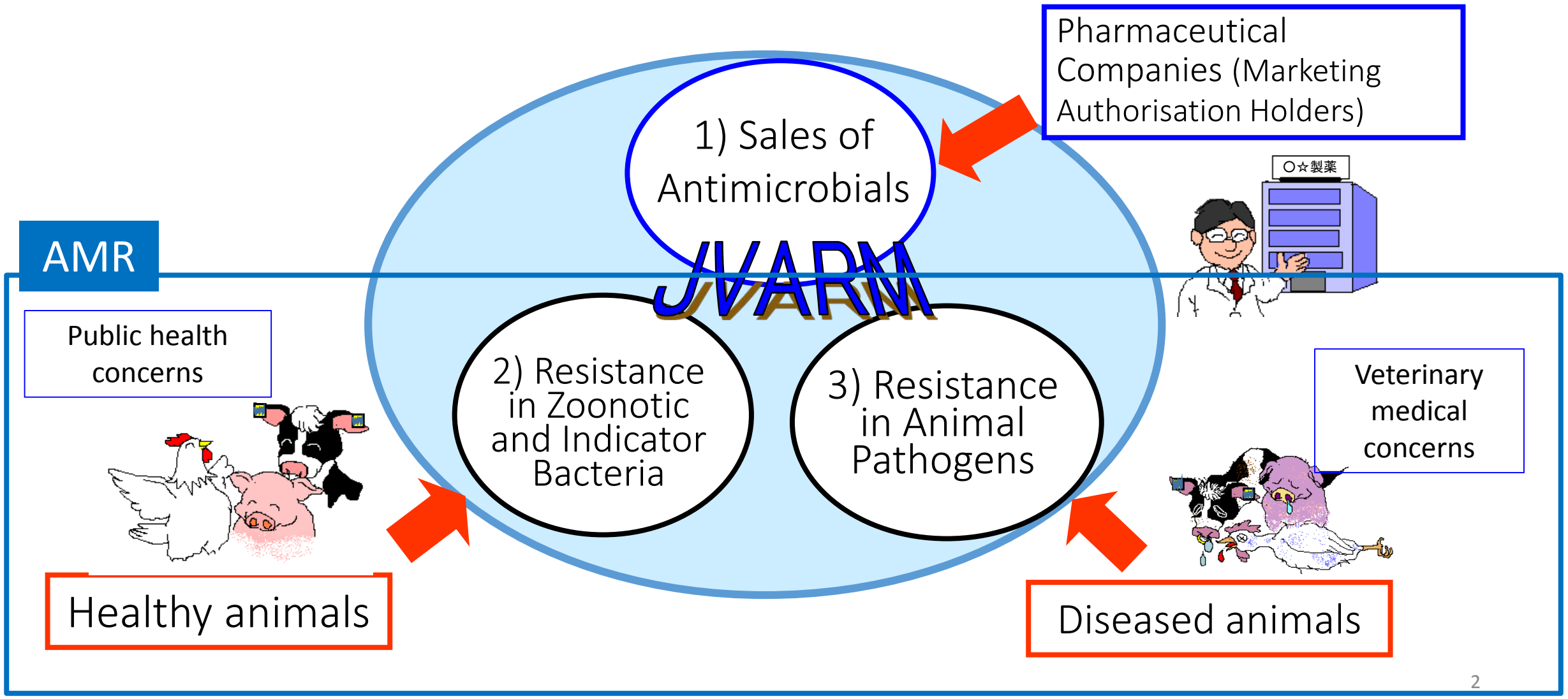
Ministry of Agriculture, Forestry and Fisheries, Japan

NVAL-OIE Joint Seminar on AMR
-Regional reference laboratory activities on AMR surveillance- Oct. 15th 2018

JVARM

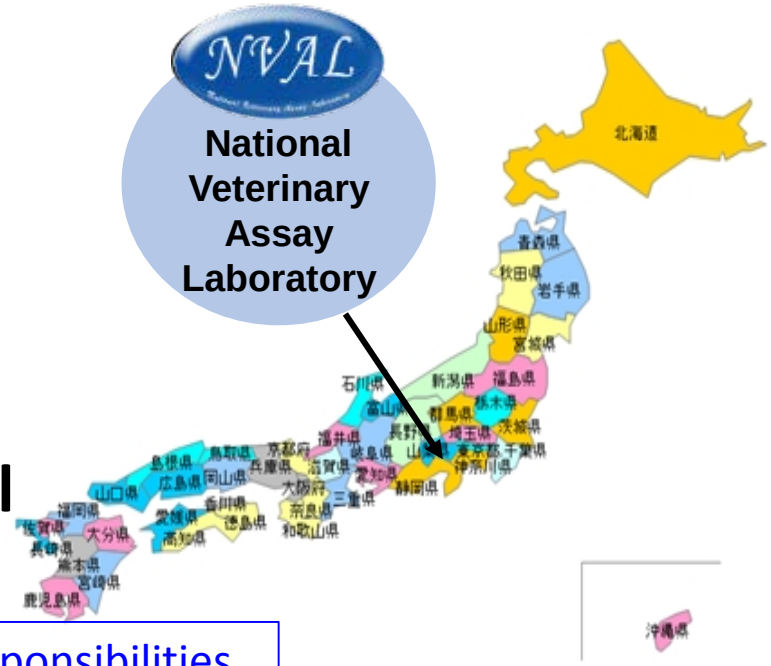
Japanese Veterinary Antimicrobial Resistance Monitoring System



Outline of Livestock and Livestock Hygiene Service Centers in Japan

- **Population (×1,000 people) : 126,933**
- **Number of Livestock (× 1,000 head)**

	2006	2016	Increase/decrease (2016/2006; %)
Dairy cattle	1,636	1,345	-18
Beef cattle	2,775	2,479	-11
Pigs	9,620	9,313	-3
Layers	180,697	173,349	-4
Broiler	103,687	134,395	30



- **Japan has 47 prefectures and each prefecture has several Livestock Hygiene Service Centers (LHSC)**

Role of the LHSC

*Diagnosis of the animal diseases are their basic responsibilities

- Annual inspections under the 'Act on Domestic Animal Infectious Diseases Control'
- Check, guide, advise for farms
- Prevent livestock from infectious diseases etc.

Total No. of LHSC ; 170
Total No. of Vets. ; 2,084

History (1)

JVARM 20 years anniversary

Trial stage; 1999 (Healthy animal's monitoring)
- Settle up the methods (bacterial isolation and MIC).
- Isolate the bacteria from fecal samples, measure the MIC in NVAL

Target bacteria
* *E. coli* (incl. STEC)
* *Enterococcus*
* *Salmonella*
* *Campylobacter*

Samples; fecal samples from Cattle, pig & chicken
*Prefecture staff collected fecal samples from farm → send NVAL (within 3 days on 4℃)
*Collected 515 samples (178 cattle, 179 pigs & 158 broilers) from all 47 prefectures

Isolation/identification; colony morphology, biochemical properties, serotyping
(2 isolates / sample → exp. *E. coli* ;1,018 isolates)

MIC; Agar dilution methods

History (2)

JVARM 20 years anniversary

1st - 2nd stage; 2000-2007

- Establish the monitoring system (Involvement of Local Government)
- Training course for local government staff (bacterial identification & MIC measurement)

3rd - 4th stages; 2008-2011

- Review the monitoring system (ex. time span, isolation of *Salmonella*)
- MIC method was changed in 2010 (Agar dilution→ Broth micro dilution, OTC→TC, ERFX→CPFX, add CTX etc.)

5th - 6th stages; 2012-2015

- Monitoring in Slaughterhouses started in 2012 and completely shifted in 2015

7th stage; 2016-2020 - Follow the AMR National Action Plan -

- Enhanced monitoring in animal pathogens (Active monitoring was started from 2016 for *Salmonella* and *S. aureus* to keep a good relationship with Local Government.)
- Expand the AMR monitoring for farm-raised aquatic animals and companion animals.
- Enhance "One health" approach (collaboration with human medical side) etc.

JVARM publications in the early stage (incl. 1999 isolates)

1. Kijima-Tanaka, M., Ishihara, K., Morioka, A., Kojima, A., Ohzono, T., Ogikubo, K., Takahashi, T., Tamura, Y. 2003. A national surveillance of antimicrobial resistance in *Escherichia coli* isolated from food-producing animals in Japan. J Antimicrob Chemother. 51: 447-451.

2. Esaki, H., Morioka, A., Kojima, A., Ishihara, K., Asai, T., Tamura, Y., Izumiya, H., Terajima, J., Watanabe, H., Takahashi, T. 2004. Epidemiological characterization of *Salmonella* Typhimurium DT104 prevalent among food-producing animals in the Japanese veterinary antimicrobial resistance monitoring program (1999-2001). Microbiol Immunol. 48: 553-556.

3. Ishihara, K., Kira, T., Ogikubo, K., Morioka, A., Kojima, A., Kijima-Tanaka, M., Takahashi, T., Tamura, Y. 2004. Antimicrobial susceptibilities of *Campylobacter* isolated from food-producing animals on farms (1999-2001): results from the Japanese Veterinary Antimicrobial Resistance Monitoring Program. Int J Antimicrob Agents. 24: 261-267.

4. Kojima, A., Ishii, Y., Ishihara, K., Esaki, H., Asai, T., Oda, C., Tamura, Y., Takahashi, T., Yamaguchi, K. 2005. Extended-spectrum-beta-lactamase-producing *Escherichia coli* strains isolated from farm animals from 1999 to 2002: report from the Japanese Veterinary Antimicrobial Resistance Monitoring Program. Antimicrob Agents Chemother. 49: 3533-3537.

5. Kijima-Tanaka, M., Ishihara, K., Kojima, A., Morioka, A., Nagata, R., Kawanishi, M., Nakazawa, M., Tamura, Y., Takahashi, T. 2005. A national surveillance of Shiga toxin-producing *Escherichia coli* in food-producing animals in Japan. J Vet Med B Infect Dis Vet Public Health. 52: 230-237.

Monitoring System in **Farm** (2000-)

Target bacteria;

- Indicator (*E. coli, Enterococcus spp.*), Zoonotic bacteria (*Salmonella spp., Campylobacter spp.*) (-2015)
- Animal pathogen (Clinical isolate; *Salmonella, Stapylococcus, Actinobacillus pleuropneumoniae* etc.)

- MAFF (Ministry of Agriculture, Forestry and Fisheries of Japan)
  - Design risk managements and provide the data for risk assessments to FSC

Report (Every year)

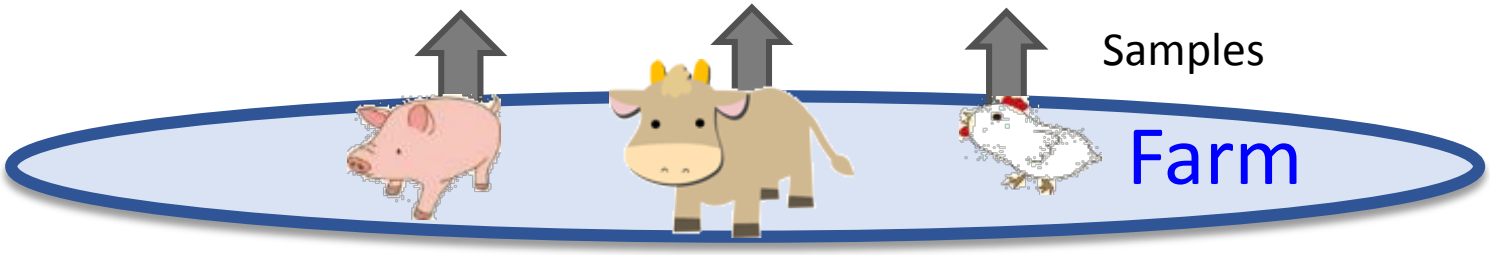
- NVAL（National Veterinary Assay Laboratory）
- FAMIC（Food and Agricultural Materials Inspection Center）(-2015)
  - Sum up, analyze, and evaluate data
  - Research into molecular epidemiology, resistance mechanism

Isolated bacteria, Data

Advice, Training seminar

- Livestock Hygiene Service Center （170 centers）
  - Collect feces in farm, isolate and identify bacteria, and measure MIC

Samples



Monitoring System in **Slaughterhouses** (2012-)

MAFF added the monitoring in slaughterhouses since 2012.
Target bacteria;
- Indicator (*E. coli, Enterococcus spp.*), Zoonotic bacteria (*Salmonella spp., Campylobacter spp.*)

- MAFF (Ministry of Agriculture, Forestry and Fisheries of Japan)
  - Design risk managements and provide the data for risk assessments to FSC

Data ⇄ Report

L（National Veterinary Assay Laboratory）
C（Food and Agricultural Materials Inspection Center）
lyze, and evaluate data
earch into molecular epidemiology, resistance mechanism

Isolated bacteria, Data ⇄ Quality control

organization
houses, isolate and identify bacteria, and measure MIC

amples ⇄ Samples

Slaughterhouses

- Slaughterhouse monitoring -

<Merit>
* More close to the meat
* Sampling is easier
* Could get ‘intestinal contents’

<Demerit>
* No feed back to the farm
* Seldom background data (AMU etc)
* Sample mix-up (insufficient connection between sample and farm) could be occurred.

History (2)

JVARM 20 years anniversary

1st - 2nd stage; 2000-2007
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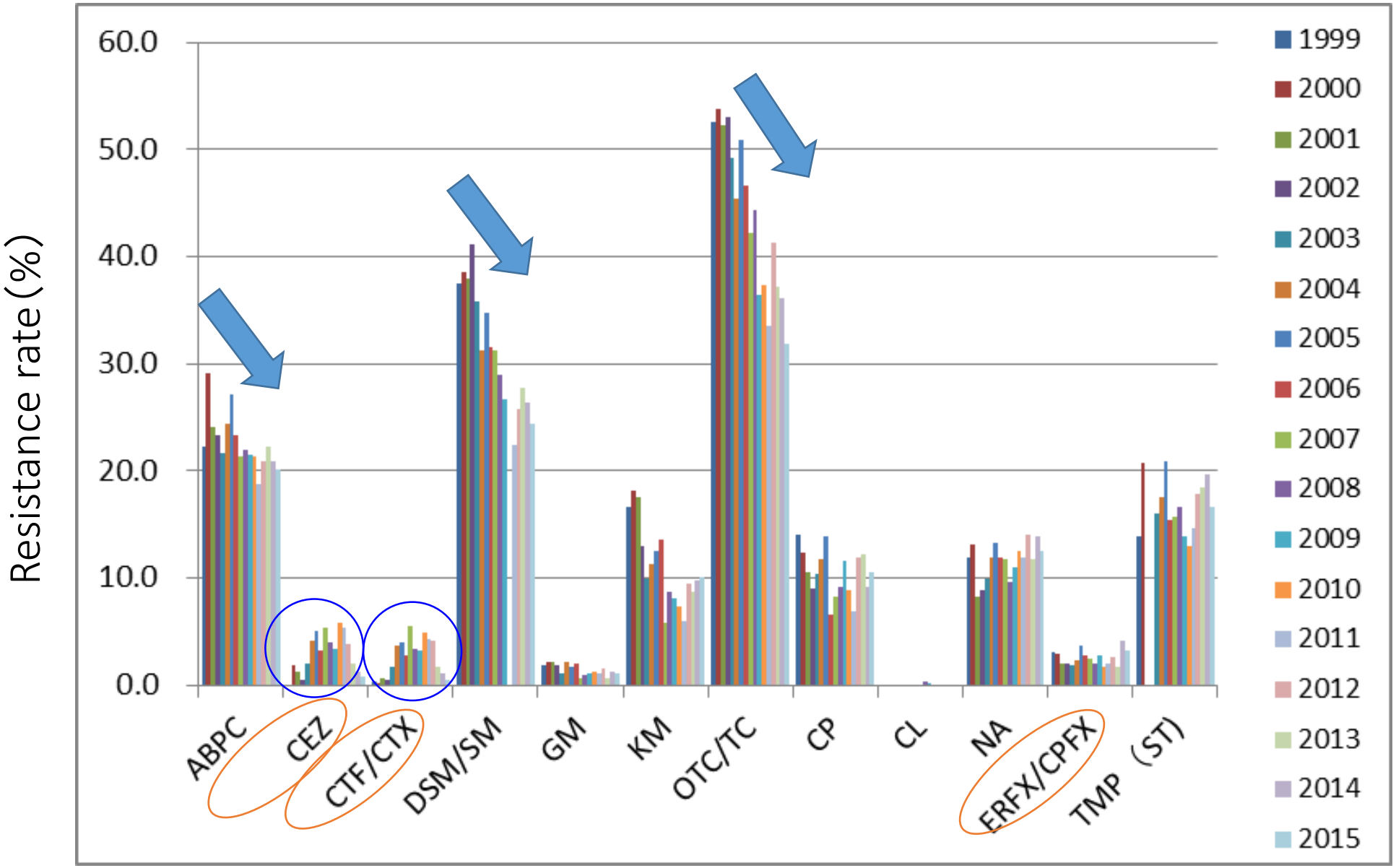
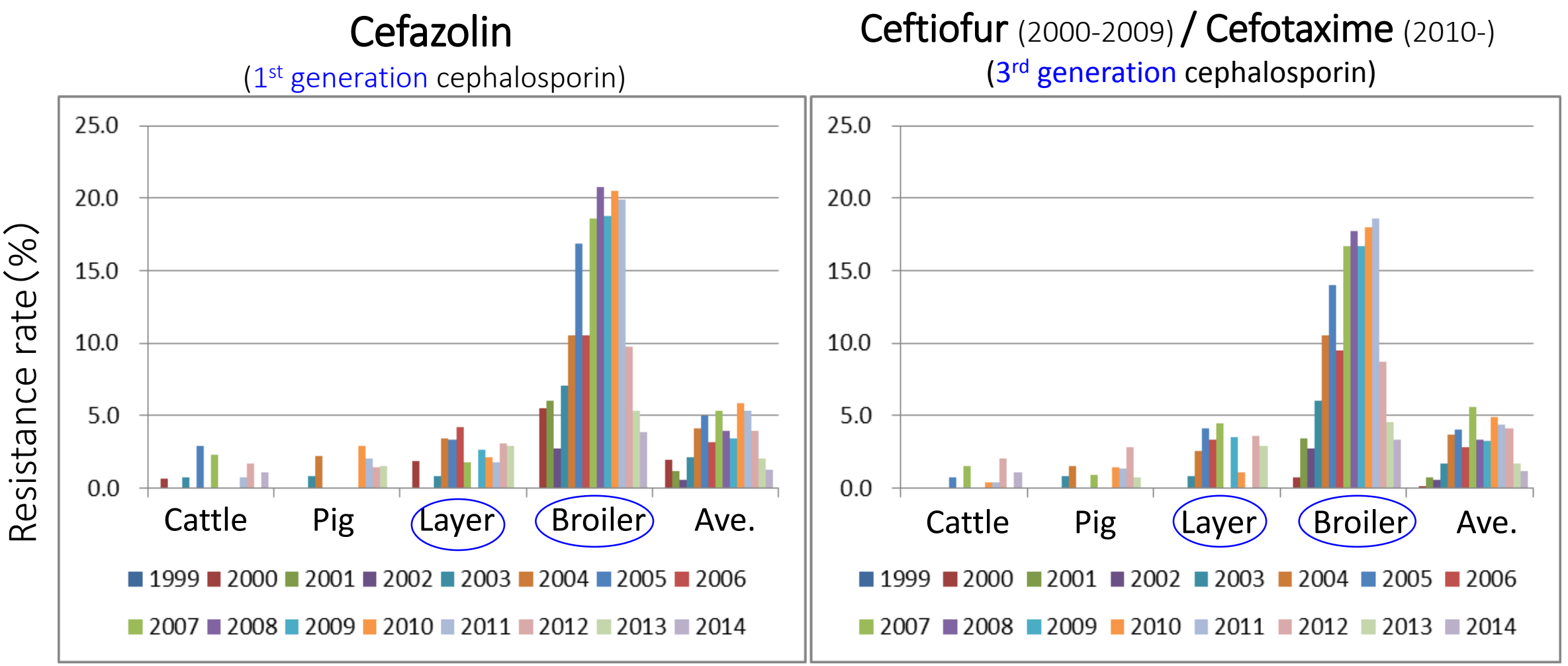
- Enhance "One health" approach (collaboration with human medical side) etc.

Resistance rate in *E. coli* isolated from healthy animals

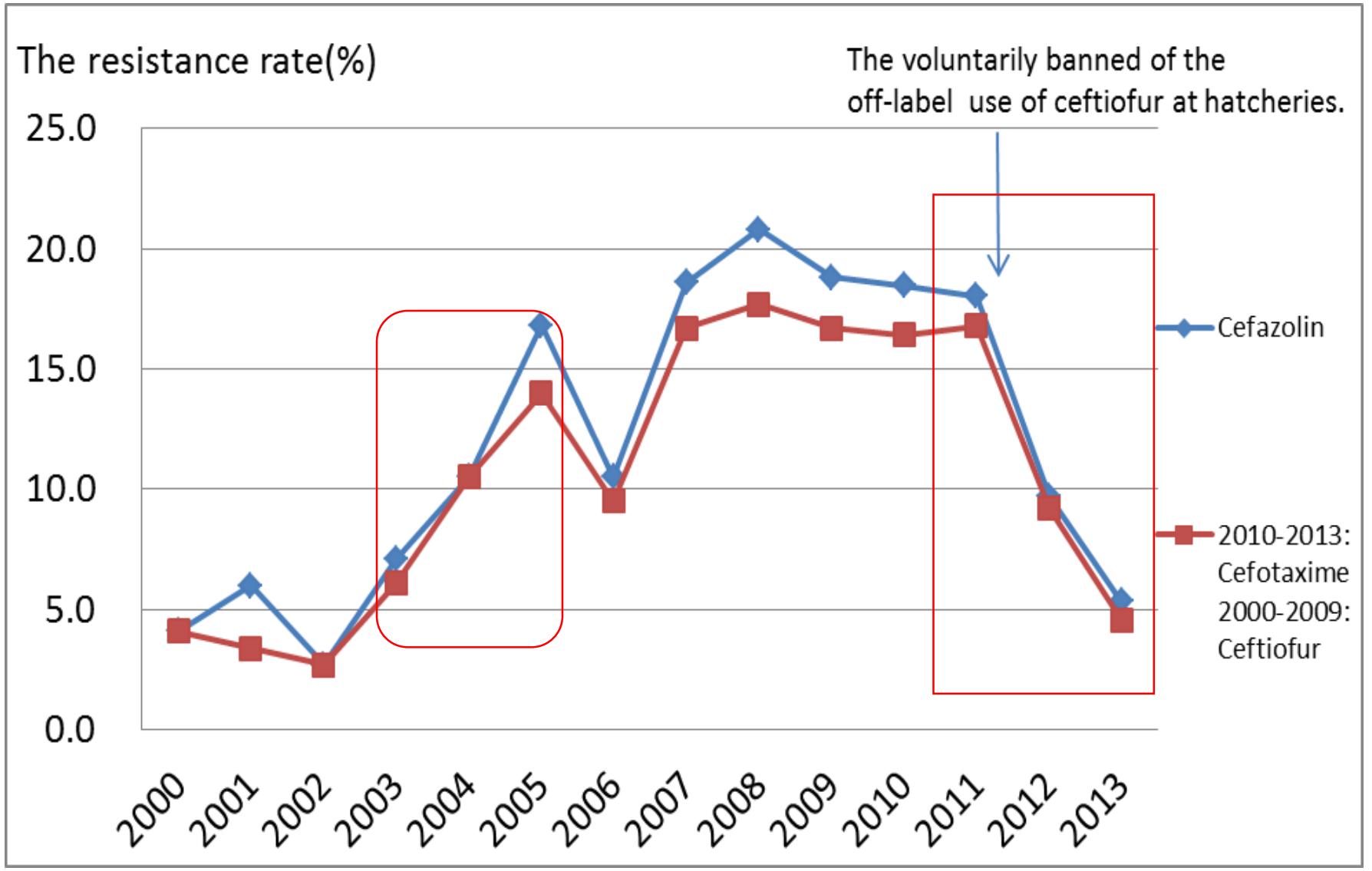


Resistance rate of cephalosporins in *E. coli* isolates from healthy animals



In Japan, cephalosporins have **NOT** been approved for poultry, however, resistance rate was obviously increasing in Layer and Broiler.

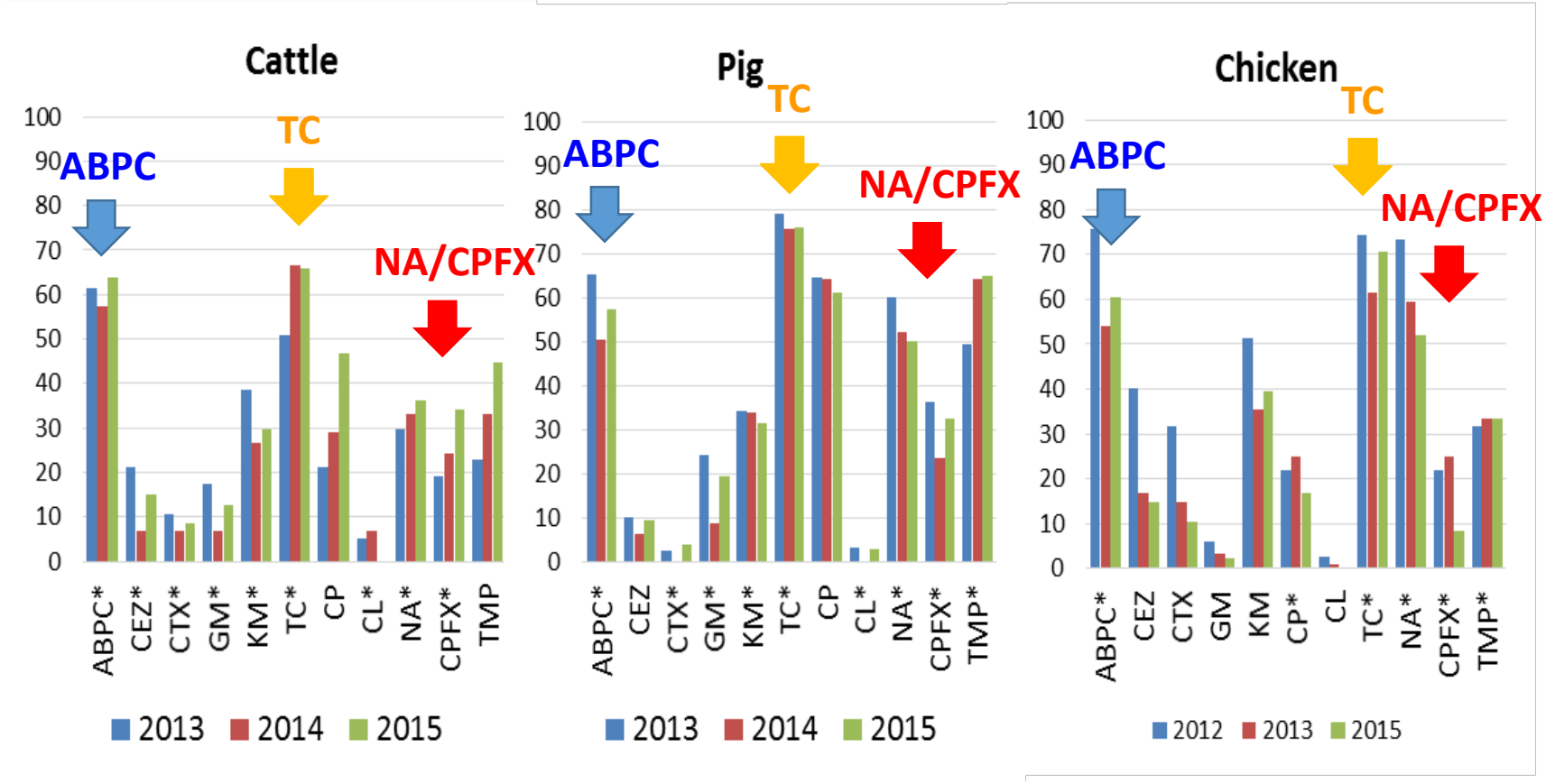
The resistance rate of cephalosporin in *E. coli* isolates from healthy broilers



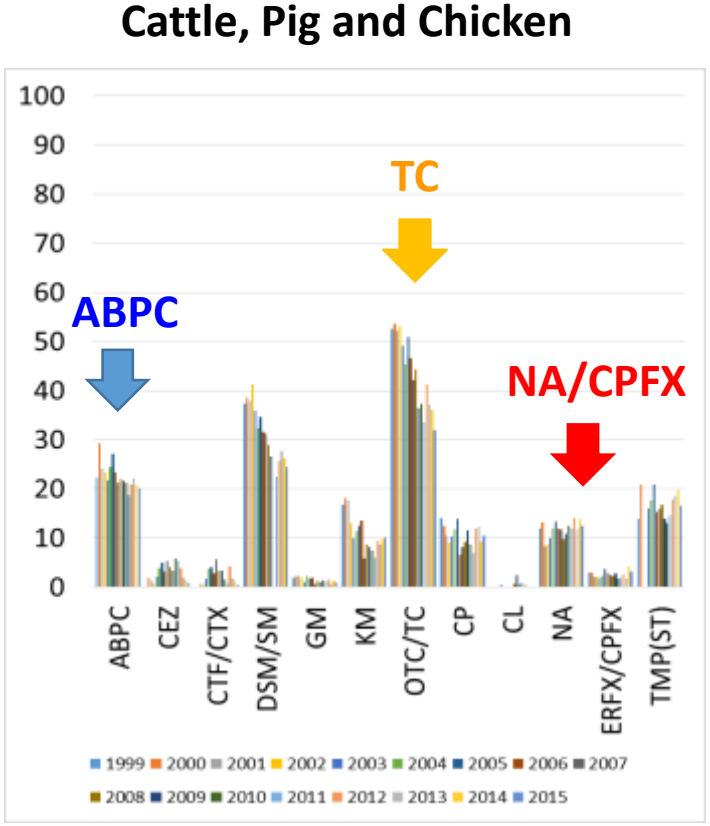
Hiki M et al. Foodborne Pathog Dis. 2015 Jul;12(7):639-43.

Resistance rates in *E. coli* isolated from Diseased and Healthy animals

【Diseased animals】



【Healthy animals】



Significance of Nationwide Monitoring in animal bacterial pathogens

- To detect emerging resistance that may pose a concern for animal and human health;
- To detect changes in susceptibility patterns;
- To provide information for *risk analysis*;
- To provide data for *veterinarians* to inform their treatment decisions;
- To provide information for epidemiological studies and trend analysis.

OIE Terrestrial Animal Health Code Chapter 6.8; Article 6.8.5.

Prefectural Livestock Hygiene Service Center

- To take the interest in the antimicrobial resistance in animals
- To improve the lab technique concerning AMR

We could get the information about the situation of the farm, and could keep the connection with the prefecture.

Current Efforts –AMR Japanese National Action Plan–

6 Key Frameworks

- Awareness through education
- Surveillance & Monitoring
- Infection Prevention & Control
- Optimal use of antimicrobials
- Research & Development
- International Cooperation

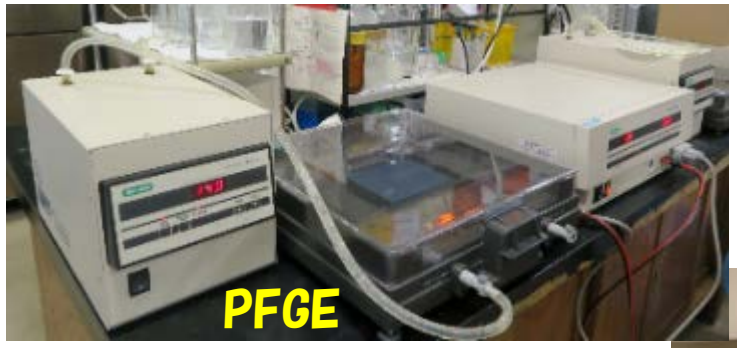
Our Strategies

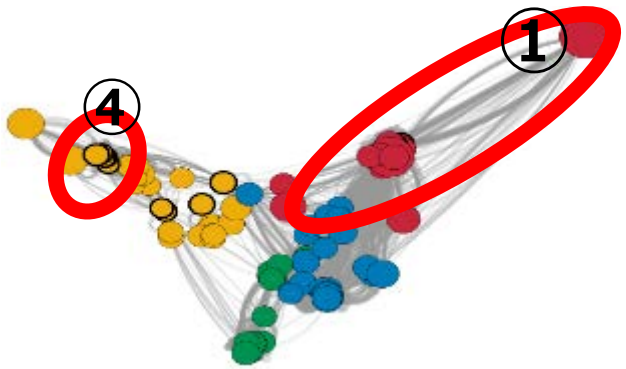
1. Introduce **molecular analysis** approach to AMR surveillance
2. Establish a monitoring system for **companion animals**
3. Strengthening the **One Health Approach** (e.g., between human and animal monitoring)
4. Expand the scope of monitoring in aquaculture
5. Further promoting the risk assessment based policies (e.g., prudent use)
6. **Working with OIE and other international organizations to contribute to the Asian region**

1. Introduce molecular analysis approach to AMR surveillance

Characterization of resistant bacteria with next generation sequencing

- under Japan Agency for Medical Research and Development (AMED) Grant -





Gene network analysis

QIAcube
(Purification of DNA)

2. Establish a monitoring system for companion animals
- Recommendation by Expert Working Group -

1. AMR Monitoring with Clinical Isolates

Every year

1) Sampling Scheme; * From Dogs and Cats.
   * Collect isolates through Private Animal Clinical Laboratories.
   * Sampling should be done nation-widely, and sampling number should be assigned depending on the number of animal hospitals in that regions, generally.
   * One isolate/ animal hospital/ bacterial species and animals.

AMR Monitoring in 2017

2) Target bacteria, origins and isolates numbers are as follows.

	Priority	Target bacteria	Origin of specimen	Isolates No/Year	
				Dog	Cat
Gram Negative (G−)	1	*Escherichia coli*	Urinary/Reproductive organ	100	100
	2	*Klebsiella*	Urinary/Reproductive organ	100	100
	3	*Enterobacter*	Urinary	100	100
	4	*Pseudomonas aeruginosa*	Urinary /Ear	100	100
	4	*Proteus mirabilis*	Urinary /Ear	100	100
	4	*Acinetobacter*	Urinary /Skin	50	50
Gram Posititive (G+)	1	Coaglase−positive *Staphylococcus* (*S. aureus, S. pseudintermedius* etc)	Urinary /Skin	100	100
	2	*Enterococcus* (*E. faecalis, E. faecium* etc)	Urinary /Ear	100	100

←200 each

←200 each

2. AMR Monitoring with isolates from Healthy Animals

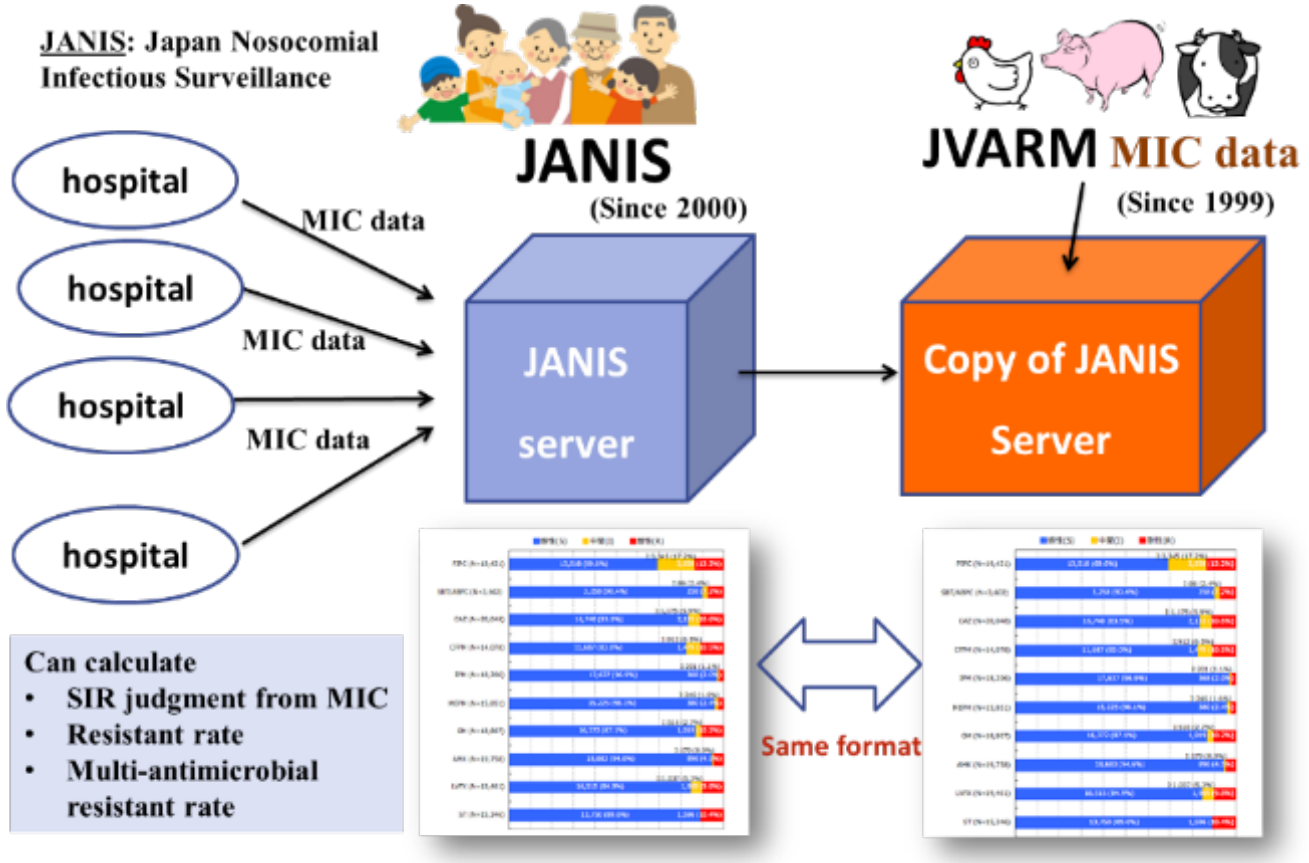
Once in several years

3. Strengthening the **One Health Approach**
(e.g., between human and animal monitoring)

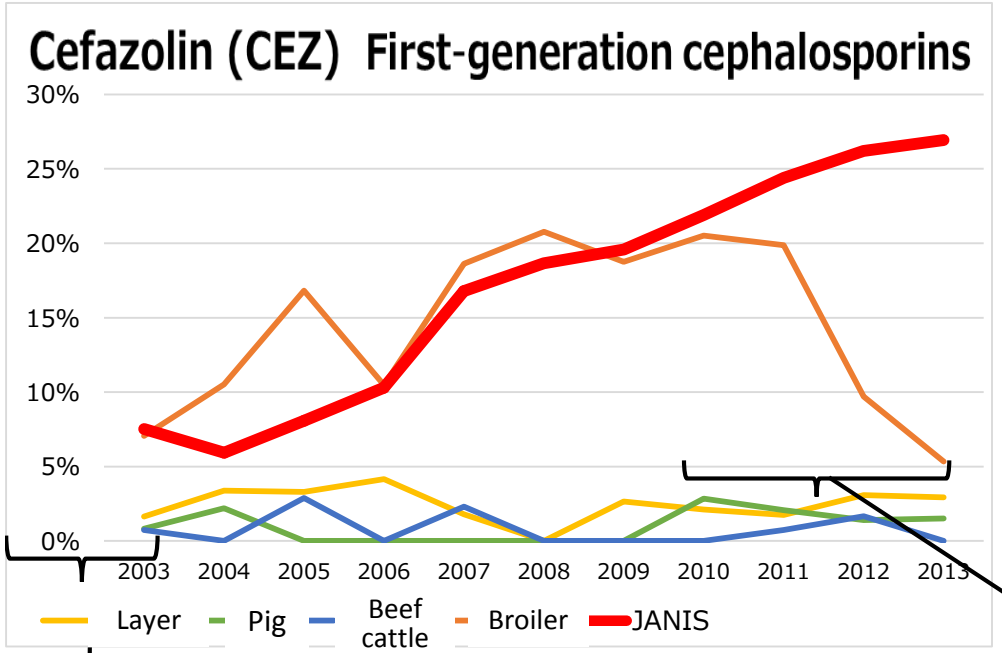
Nippon AMR One Health Report (NAOR) 2017

October 18, 2017
The AMR One Health Surveillance Committee

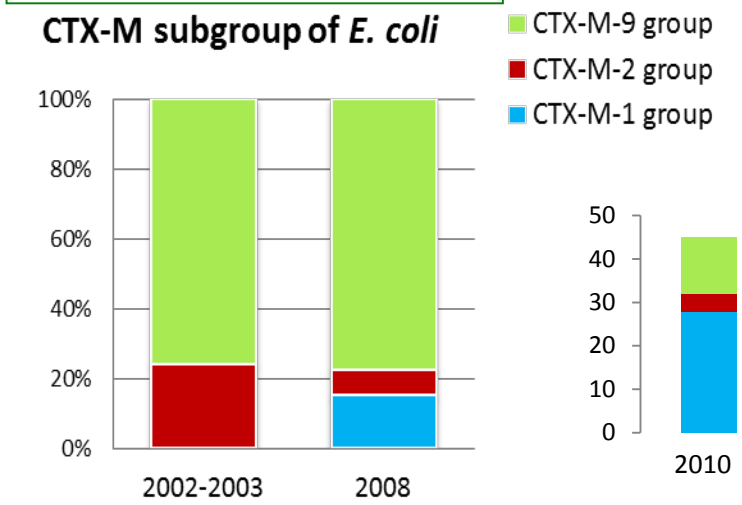
http://www.mhlw.go.jp/file/06-Seisakujouhou-10900000-Kenkoukyoku/0000204347.pdf



Comparison of resistance rate and β-Lactamase genes between human- and animal-derived *Escherichia coli*



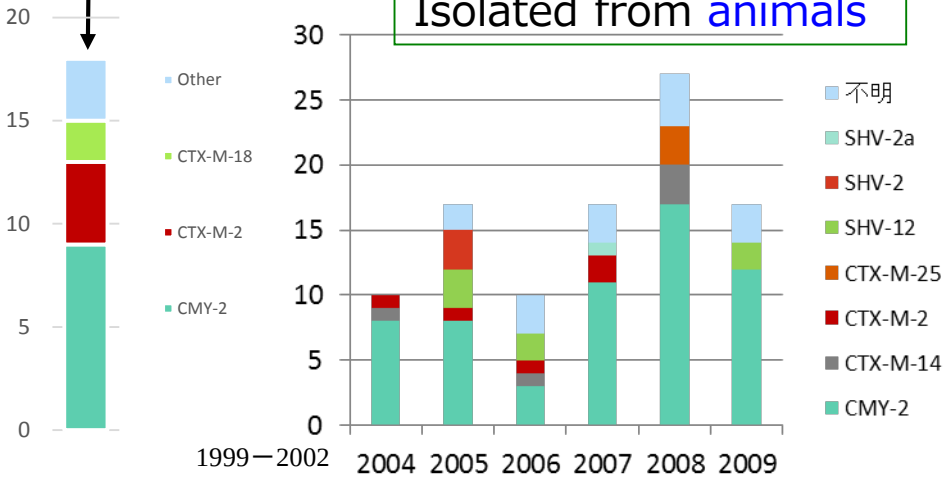
Isolated from human



Suzuki et al. J Antimicrob Chemother. 2009 , 63: 72-9

Whole-Genome Analysis

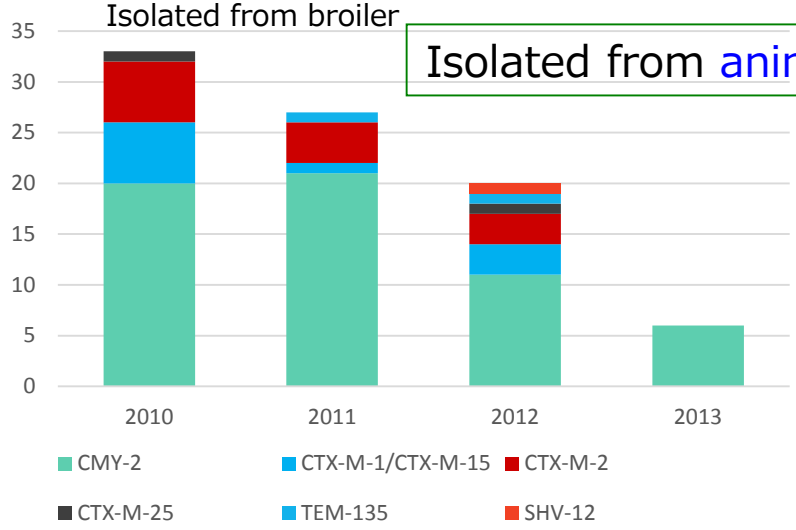
Isolated from animals



Kojima et al. Antimicrob. Agents Chemother. 2005, 49: 3533–3537

Isolated from broiler

Isolated from animals



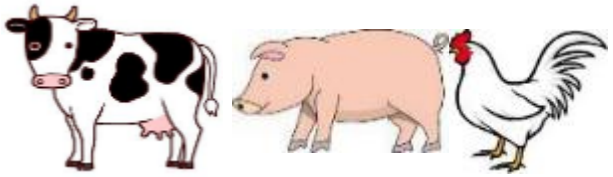
Hiki et al. Foodborne Pathog. Dis. 2015, 12: 639-43

NVAL

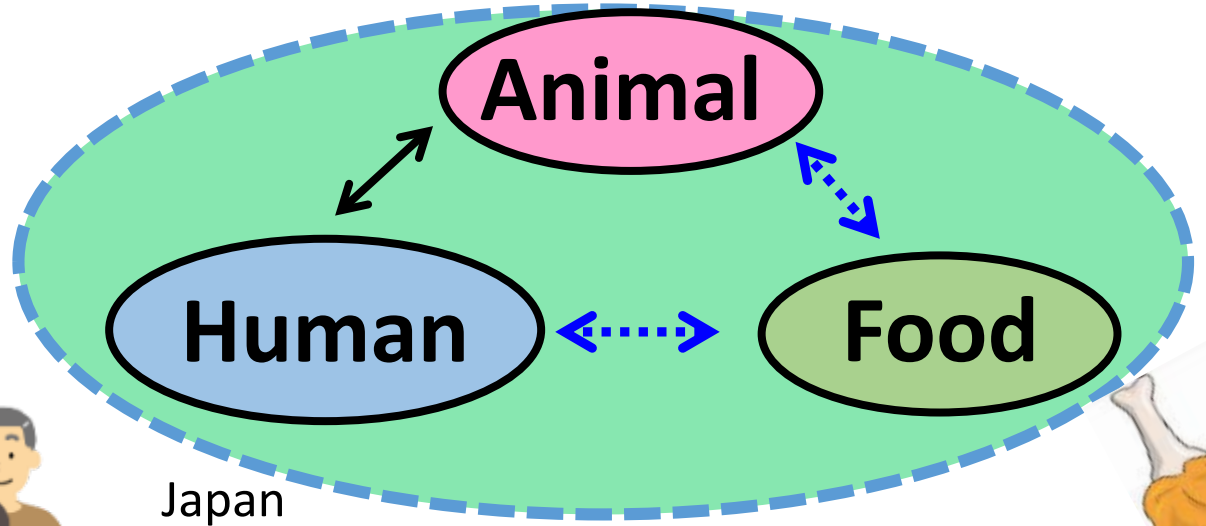
A collaborative research project on One Health Monitoring
- under Health and Labor Sciences Research Grant -

Under the control of Ministry of Agriculture

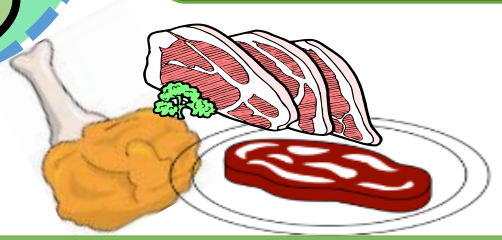
National Veterinary Assay Laboratory



Japanese Veterinary Antimicrobial Resistance Monitoring (JVARM)



Join the project 2017～



National Institute of Infectious Diseases

Japan Nosocomial Infections Surveillance JANIS

Prefectural Institutes of Public Health

Under the control of Ministry of Health

AMR Training Course for local government staff

Target; Local governmental staff

1) Livestock Hygiene Service center (Every year; 3 days training ×2 times; total 44 trainee in 2018)

2) Fisheries experimental station (2017, Nov & Dec ; 2days training ×2 times; total 32 trainee)

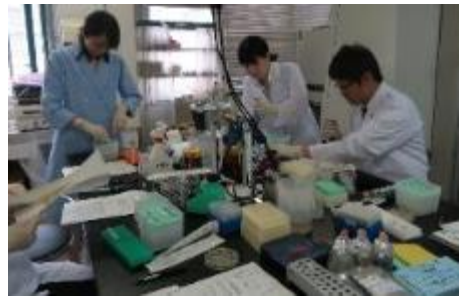
Contents;

* **Technique transfer** ; Isolation/Identification of bacteria, MIC (disk etc), (Option; PFGE, ESBL/MRSA etc)
  Standardize material and methods, Improve technique level, Eye alignment,
* Presentations ; Visiting lecture, MAFF head office staff, NVAL staff,
* **Information sharing** ; Case study by local government staff, Workshop

--Training course for Livestock Hygiene Service center (5/30-6/1 & 6/13-6/15, 2018) --

< Workshop/Lecture > < Isolation/Identification, MIC > < ESBL/MRSA > < PFGE >






OIE Regional Short-Term Training on AMR in Tokyo 2017 (Oct. 16th-24th)

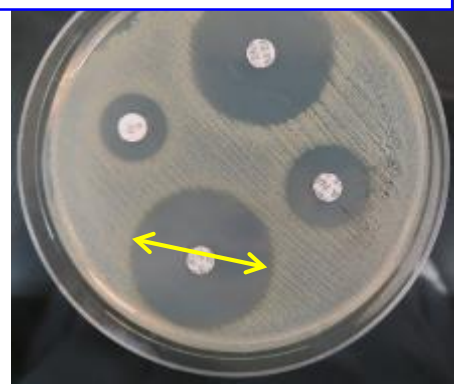
*Participants (12 countries) ; Governmental agency staff on vet AMR in Asia
Bhutan, Cambodia, Chinese Taipei, Hong Kong, Mongolia, Myanmar, Philippines, Republic of Korea
Singapore, Sri Lanka, Thailand & Vietnam

*Laboratory Training Course;
- Pre Advanced course & Advanced course -
<Antimicrobial Sensitivity Tests> <identification of Resistant Bacteria>

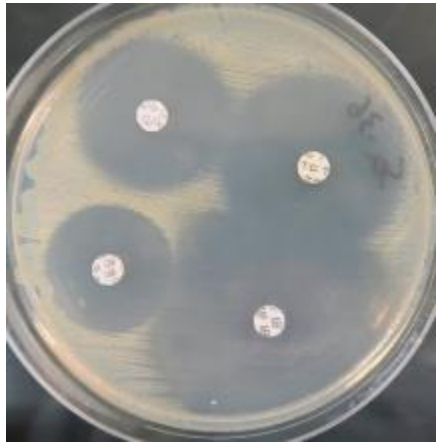
*AMR Joint Seminar; from food safety perspective (in Tokyo Univ.)
<Morning Seminar> Japanese Trials in the National Monitoring and Risk Management
<Afternoon Seminar> From Food Safety Perspective
<Group Work> How to Establish and Strengthen the AMR National Monitoring effectively

*Presentation; Topics for resistance

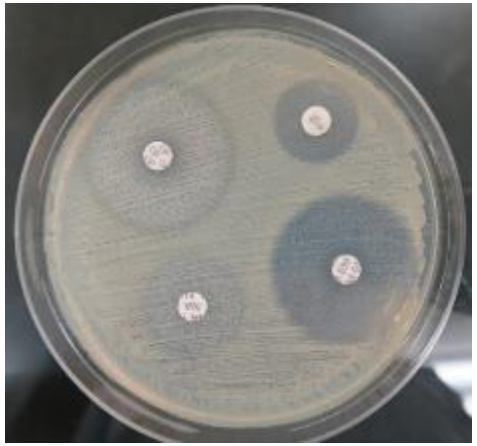
Disk diffusion method



OK→ measure diameter



Overlap/ill-shaped inhibition zone
→ Re-layout the disk and retest

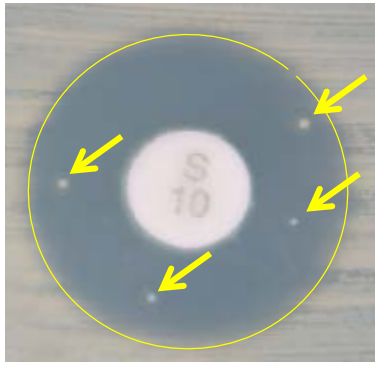


Contamination
→ Cloning, re-identification and retest

＜Eye alignment for inhibition zone＞

Complete inhibition zone

Inner circle shaped by ≧70% of colony



How to judge inner colonies

＜Adjust bacterial concentration＞

＊Adjust Turbidity
＊Incubation Time/℃, Thickness of agar would affect the inhibition zone.

＊Refer to quality control ranges of Reference Strain (exp. ATCC) every test.

Human sector

JANIS

MAFF

Other Asian member states

JVARM network

NVAL

- AMR central lab
- OIE collaborating center

JVARM

International contribution

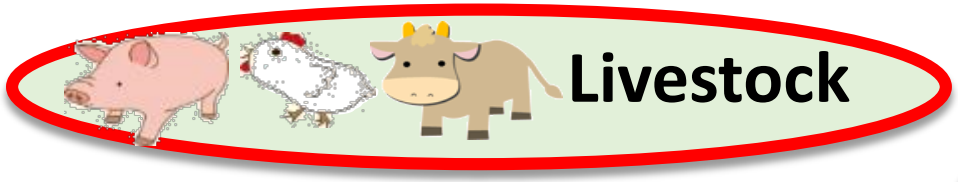
Livestock hygiene service centers

Private test service

Animal hospitals

Academia

Aquatic test facilities



Livestock

Aquatic

Pet

NVAL

Thank you for your attention !

International Cooperation on Harmonisation of Technical Requirements for Registration of Veterinary Medicinal Products