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Food and Drug Safety Evaluation

New approach for AMR monitoring and surveillance using NGS

27 May 2024, Berlin

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Ministry of Food and Drug Safety



Agenda

1. Antimicrobial resistance (AMR)

2. Next Generation Sequencing (NGS)

Antimicrobial resistance (AMR)



One Health AMR

AMR Info

R&D

Test Method

"Healthy and Happy" with the right antibiotics





Human Consumption



Human Resistance Rate



Livestock Sell

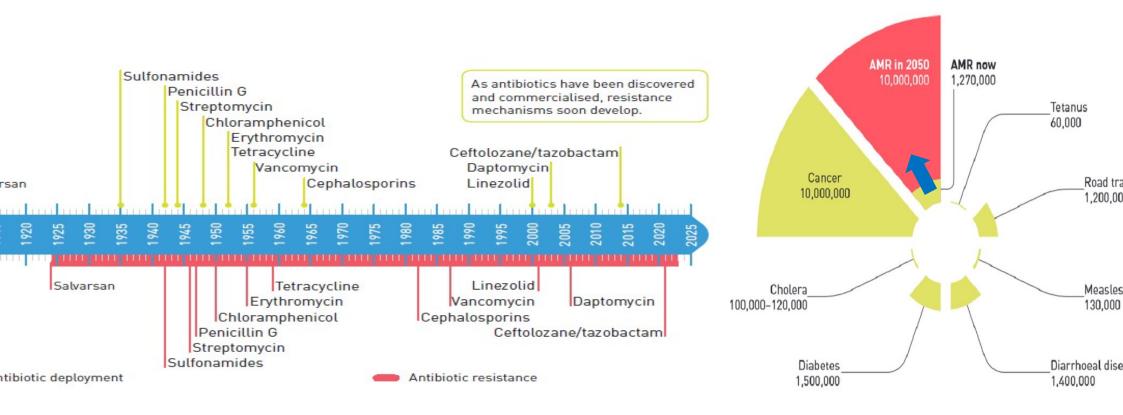


Livestock Resistance Rate



Resistance Rate Comparisons Between Countries

AMR is a global public health concern



Antibiotics and their resistance

Predicted mortality

www.ui

Global AMR response



 Endorsed by the World Health Assembly – member states encouraged to develop NAPs. AMR Multi-Partner Trust Fund

 speared to take a One Health
 approach to disbursing funds to
 support countries in their NAPs.

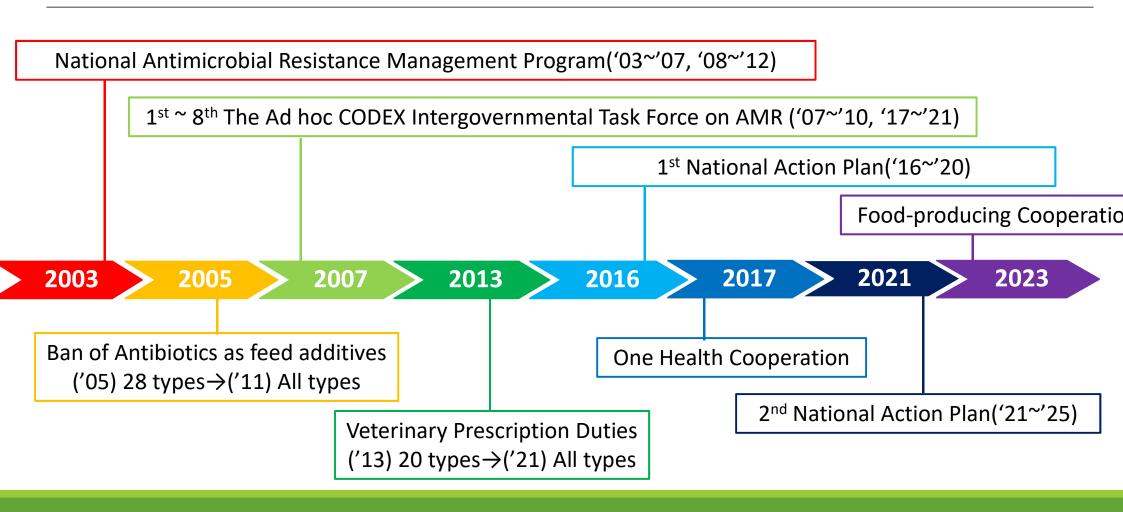


- FAO, WOAH (founded as OIE) and WHO formed the Tripartite.
- Joint leadership role on AMR governance.

- UN Political Declaration on AMR.
- Inflection point to strengthen the response to AMR beyond the health sector.

 Tripartite becomes the Quadripartite as it welcomes UNEP to join the trio.

Korea AMR response



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Next Generation Sequencing (NGS)







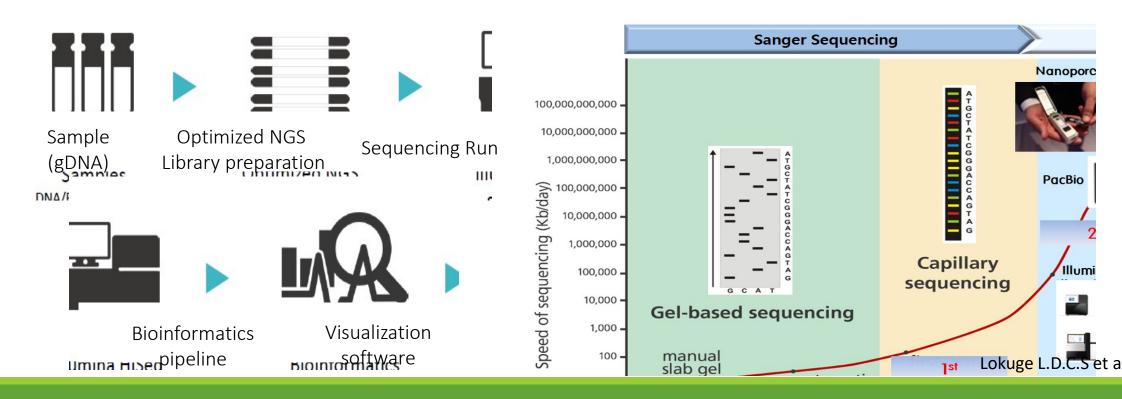




NGS is ···

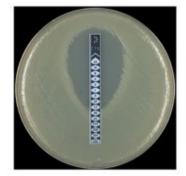
Massively parallel sequencing

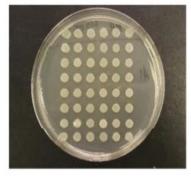
→ Able to provide a "complete" set of data on the genetic material in sample

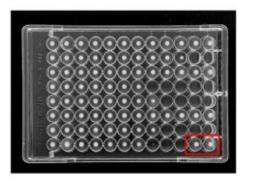


NGS, Antimicrobial susceptibility Test (AST









	1	2	3	4	5	6	7	8	9	10	11	12	ANTIMICROBICS	
A	AUG2	AUG2	AUG2	AUG2	AUG2	FEP	FEP	FEP	FEP	FEP	FEP	FEP	CIP	Ciprofloxacin
	2/1	4/2	8/4	16/8	32/16	0.25	0.5	1	2	4	8	16	AUG2	Amoxicillin / clavulanic acid 2:1 ratio
В	FOX	FOX	FOX	FOX	FOX	FOX	SXT	SXT	SXT	SXT	SXT	SXT	FOX	Cefoxitin
	1	2	4	8	16	32	0.12/2.38	0.25/4.75	0.5/9.5	1/19	2/38	4/76	CHL	Chloramphenicol
С	CHL	CHL	CHL	CHL	CHL	CHL	AMP	AMP	AMP	AMP	AMP	AMP	STR	Streptomycin
	2	4	8	16	32	64	2	4	8	16	32	64	GEN	Gentamicin
D	CIP	CIP	CIP	CIP	CIP	CIP	CIP	CIP	STR	STR	STR	STR	TET	Tetracycline
	0.12	0.25	0.5	1	2	4	8	16	16	32	64	128	NAL	Nalidixic Acid
E	GEN	GEN	GEN	GEN	GEN	GEN	GEN	CAZ	CAZ	CAZ	CAZ	CAZ	CAZ	Ceftazidime
	1	2	4	8	16	32	64	1	2	4	8	16	SXT	Trimethoprim / sulfamethoxazole
F	TET	TET	TET	TET	TET	TET	TET	FIS	FIS	FIS	FIS	FIS	FEP	Cefepime
	2	4	8	16	32	64	128	16	32	64	128	256	CTX	Cefotaxime
G	NAL	NAL	NAL	NAL	NAL	NAL	NAL	СТХ	СТХ	СТХ	СТХ	СТХ	MEM	Meropenem
	2	4	8	16	32	64	128	0.5	1	2	4	8	AMP	Ampicilin
н	MERO	MERO	MERO	MERO	MERO	COL	COL	COL	COL	POS	POS	POS	COL	Colistin
	0.25	0.5	1	2	4	2	4	8	16	CON	CON	CON	FIS	Sulfisoxazole
											4		POS	Positive Control

As the M&S tool, NGS can

1. Predict AMR with mechanistic information

Not only phenotypically determination (R/S), reveal molecular mechanisms

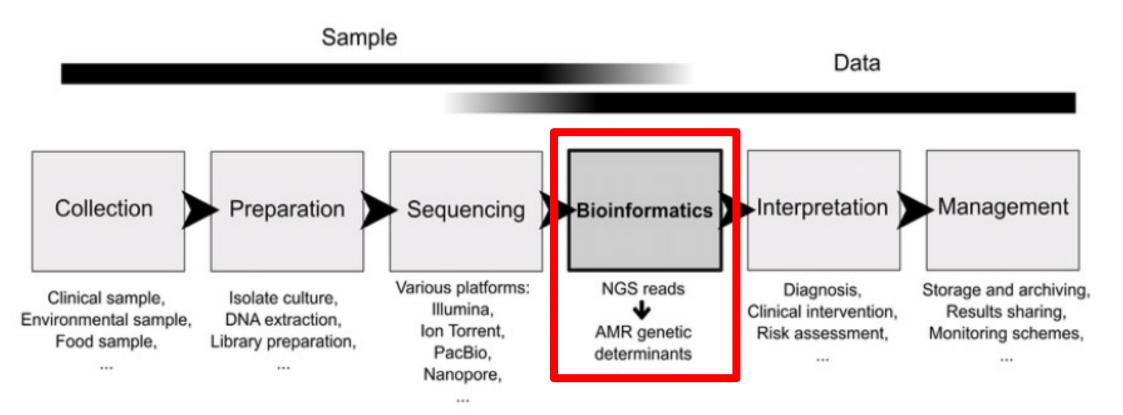
2. Link the different fields: environment, animals and foods

Not culture-dependent method

3. Allow data storage and accumulation

Possible to save for future analyses

Bioinformatics, a key of NGS



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NGS application rules

CATION

n simple rules for the sharing of bacterial notype—Phenotype data on antimicrobial sistance

rovide data in well-defined format

rovide relevant contextual sample metadata lake all samples identifiable rovide raw quantitative data for phenotypic AST results clude phenotyping method hare tabular data files in machine-readable format lake raw genomic data available lake genotypic resistance calls in a reproducible manner

eport novel resistance determinants in a systematic way

Share the data to the fullest extent possible

Chindelevitch, L. et al., 2023

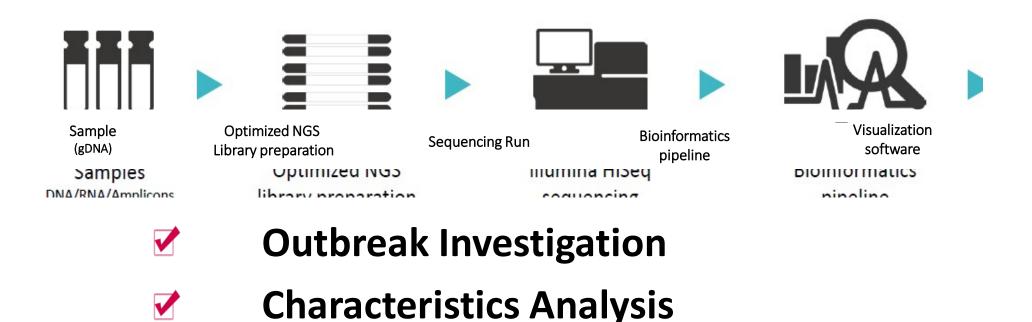
Box 1. Recommended format for reporting genotype and AMR phenotype data

Based on a review of the available formats, we recommend the use of a single file in a tabular format, with 1 row per isolate, and the following information made available for each one:

- Internal ID (this can be helpful as the key for merging genotype and phenotype tables)
- Accession number for the raw genotypic data in databases (NCBI, ENA, and DDBJ [12])
- · Additional accession numbers specific to the isolate, such as the assembled contigs
- Collection date, in a "long format" (e.g., 12 October 2022) to avoid potential confusion
- · Collection location, ideally in an unambiguous format such as longitude and latitude
- · Source of isolation (animal, clinical, environmental, etc.)
- For clinical isolates, the fluid or tissue the isolate is from (blood, sputum, stool, urine, etc.)
- Isolate genus and species
- Experimental approach used to measure phenotypic susceptibility (agar dilution, Etest, Vitek2, etc. [13])
- For each drug or combination tested for susceptibility, ideally the 3 columns specified in Rule 4, otherwise 1 column with the resistance status (susceptible (S), intermediate susceptibility (I), resistant (R))

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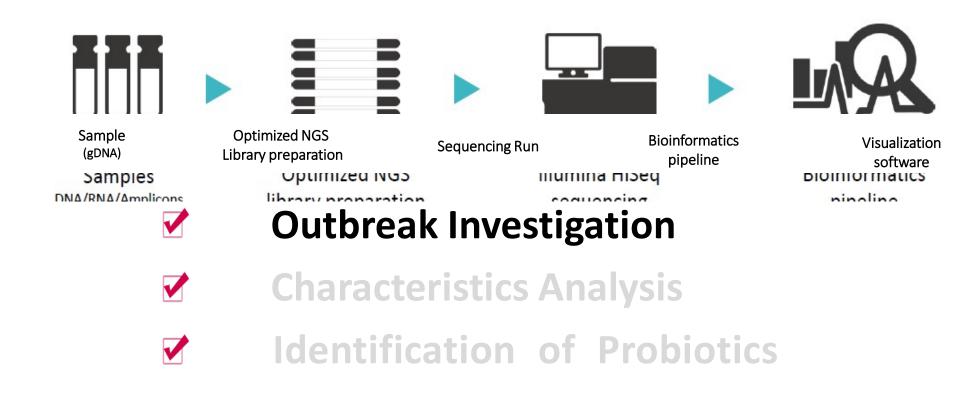
NIFDS NGS application



Identification of Probiotics

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NIFDS NGS application



NGS targeted gene panel

NGS based multigene panel for high throughput detection of food-borne pathogens

15 Target DNAs

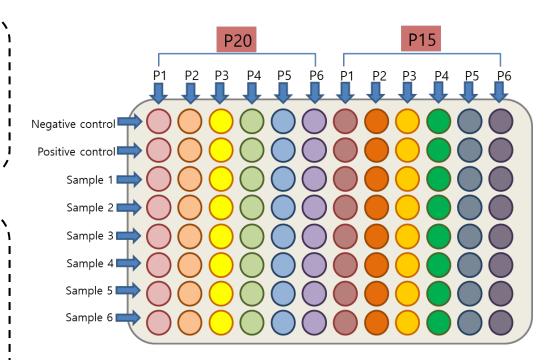
- Campylobacter jejuni (**mapA**)
- Campylobacer coli (ceuE)
- Clostridium perfringens (cpe)
- Vibrio choleae (ctx)
- Vibrio vulnificus (glnA)
- Vibrio parahaemolyticus (tdh)
- Salmonella spp. (his)

- Listeria monocytogens (iap)
- Bacillus cereus (bceT)
- Yersinia enterocolitica (ail)
- Staphylococcus aureus (coa)
- EHEC (VT1, VT2)
- ETEC (STh, STp)

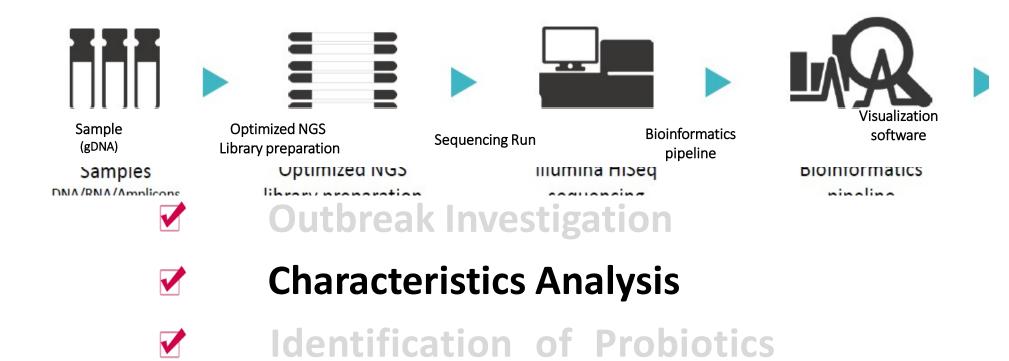
20 Target DNAs

- Campylobacter jejuni (**hipO**)
- Campylobacter coli (glyA)
- Clostridium perfringens (cpa, cpe)
- Vibrio cholera (**hly**)
- Vibrio vulnificus (vvh)
- Vibrio parahaemolyticus (toxR)
- Salmonella spp. (invA)
- Listeria monocytogens (prfA)

- Bacillus cereus (groEL)
- Yersinia enterocolitica (**inv**)
- Staphylococcus aureus (femA)
- EHEC (VT1, VT2)
- ETEC (STh, STp)
- EAEC (aggR)
- EPEC (eaeA, bfpA)
- EIEC (ipaH)



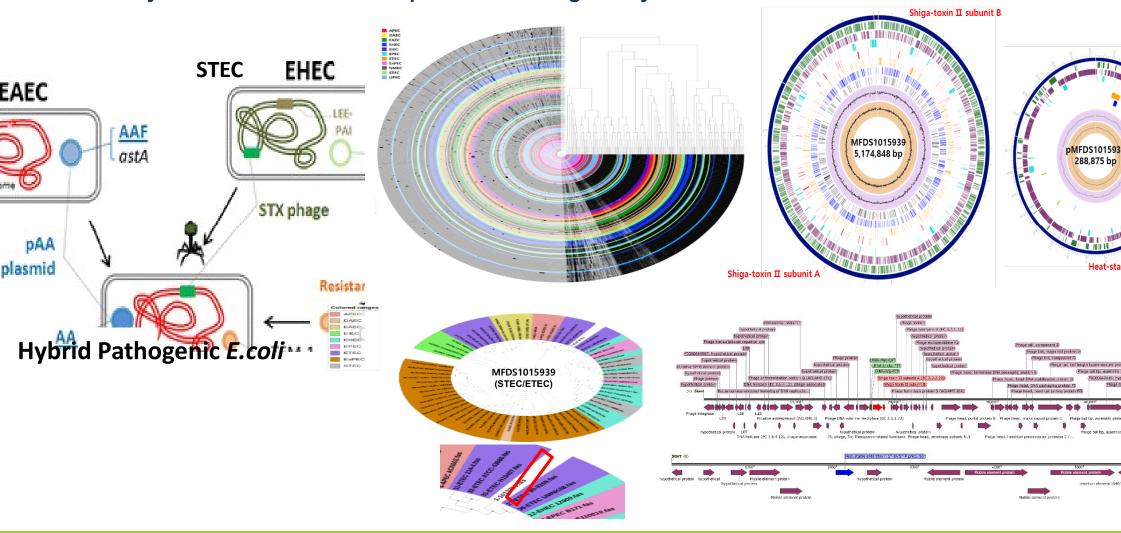
NIFDS NGS application



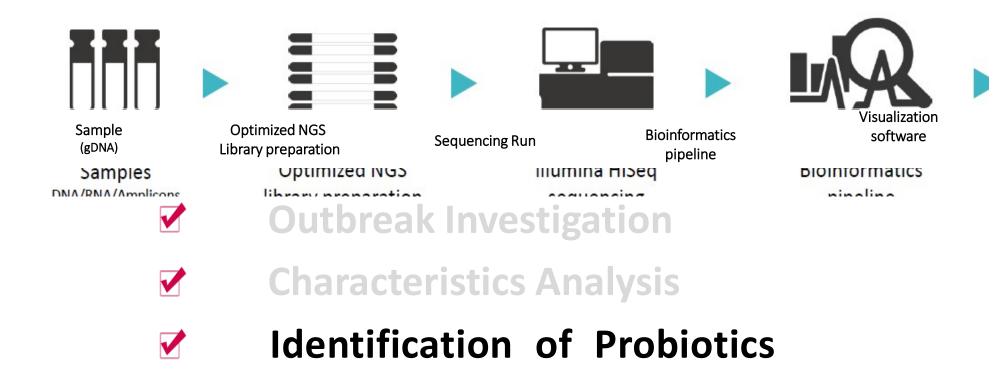
omparative Genomics and Characterization of Hybrid pathogenic *E.coli*

irulence and transcriptome profile of Hybrid pathogenic *E.coli*

orrelation analysis between virulence expression and regulatory factors in *E. coli*



NIFDS NGS application





Lactobacillus plantarum, Lactobacillus rhamnosus, Lactobacillus casei, Lactobacillus paracasei, Lactobacillus fermentum, Bifidobacterium longum, Bifidobacterium breve Bifidobacterium bifidum, Bifidobacterium lactis, Streptococcus thermophilus

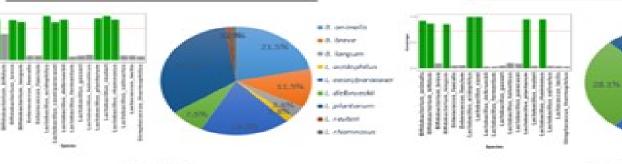
e: Adults take 1 capsule daily. May be taken with or without food, be opened. Contents can be taken directly with water Not intended for children.

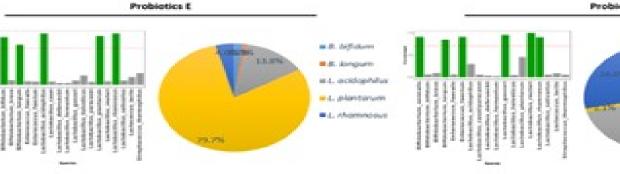
pplement Facts ze 1 Capsule er Container 30 Probletic Blend 218 mg Illus acidophilus illus acidophilus illus asseri illus gasseri illus plantarum illus plantarum illus piantarum illus piantarum illus piantarum illus paracasei illus acidophilus bulgancus illus alivarius illus salivarius illus fermentum acto Cultures (35 Billion CFU) cterium lactis cterium bifidum cterium breve cterium infantis cterium longum iifido Cultures (15 Billion CFU) biotic Cultures 50 Billion CFU¹ ebiotic Fiber Blend 407 mg Potato [Resistant Starch] (tuber), Organic Acacia Fibe ਸ਼ੀ)

late under recommended storage conditions. dry place.

Lactobacillus acidophilus, Lactobacillus casei, Lactobacillus gasseri, Lactobacillus plantarum, Lactobacillus paracasei, Lactobacillus rhamnosus, Lactobacillus fermentum, Lactobacillus bulgaricus, Lactobacillus salivarius, Bifidobacterium longum, Bifidobacterium breve, Bifidobacterium bifidum, Bifidobacterium lactis, Bifidobacterium infantis

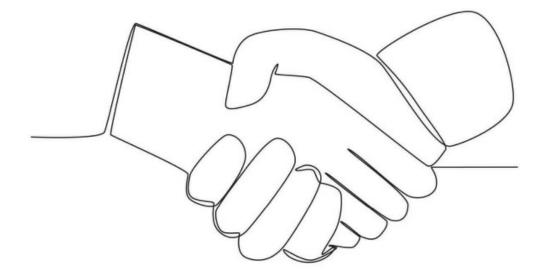
Probletics A. Probleti M. A. Andrewsky M. St. Derman THE RESIDENCE Will Street, Square, ALCOHOLD STATE No. of Contract The property of the But offermouse. B.C. Planning below Probiotics C





Problemies G.

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THANK YOU!