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**2nd International Conference of Advanced Veterinary
Science and Technologies for Sustainable
Development 17-18 Sept 2022**

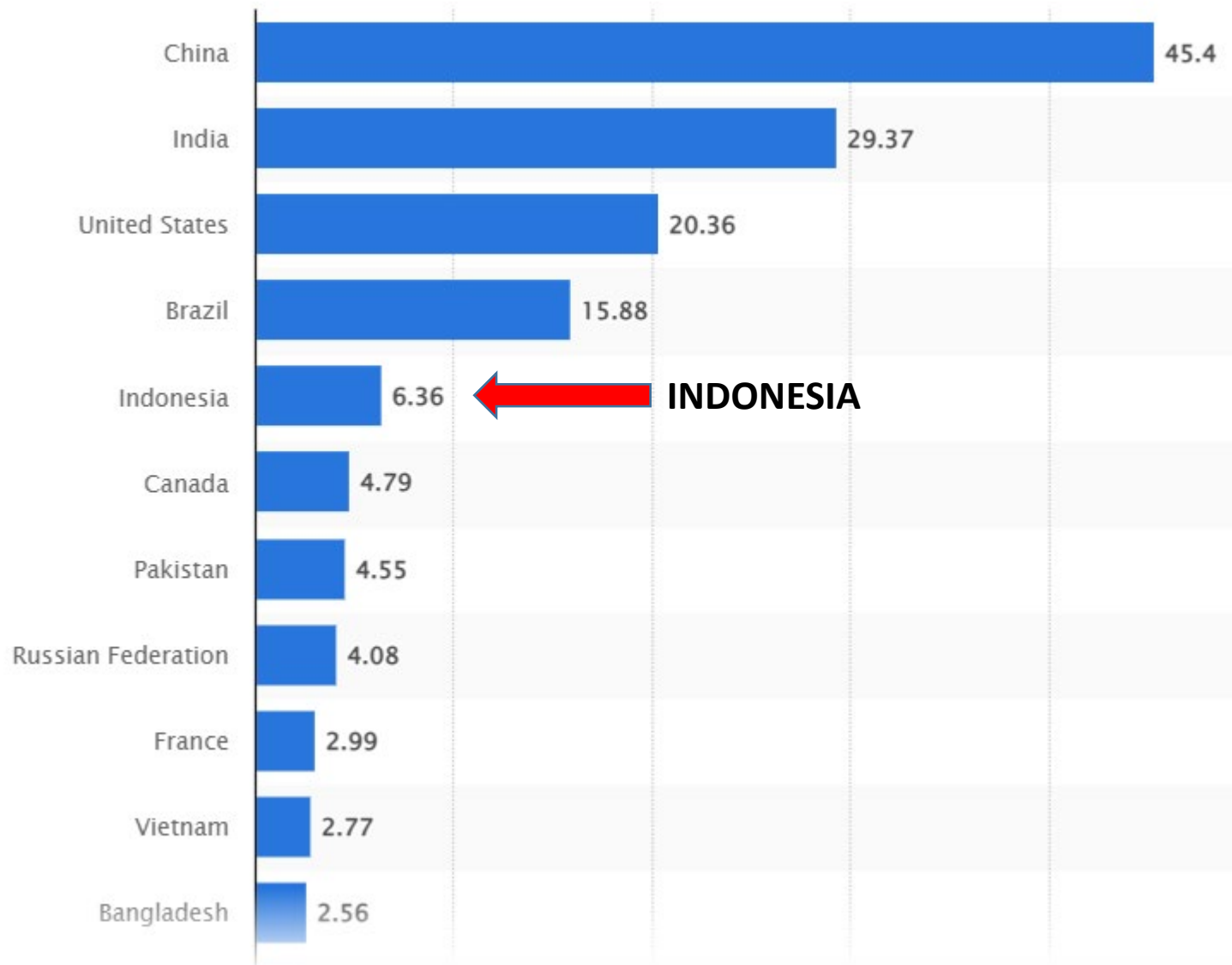
“The Impact of Animal Manure and Pesticide Usage on Vegetable and Soil Microbiome”

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INTRODUCTION

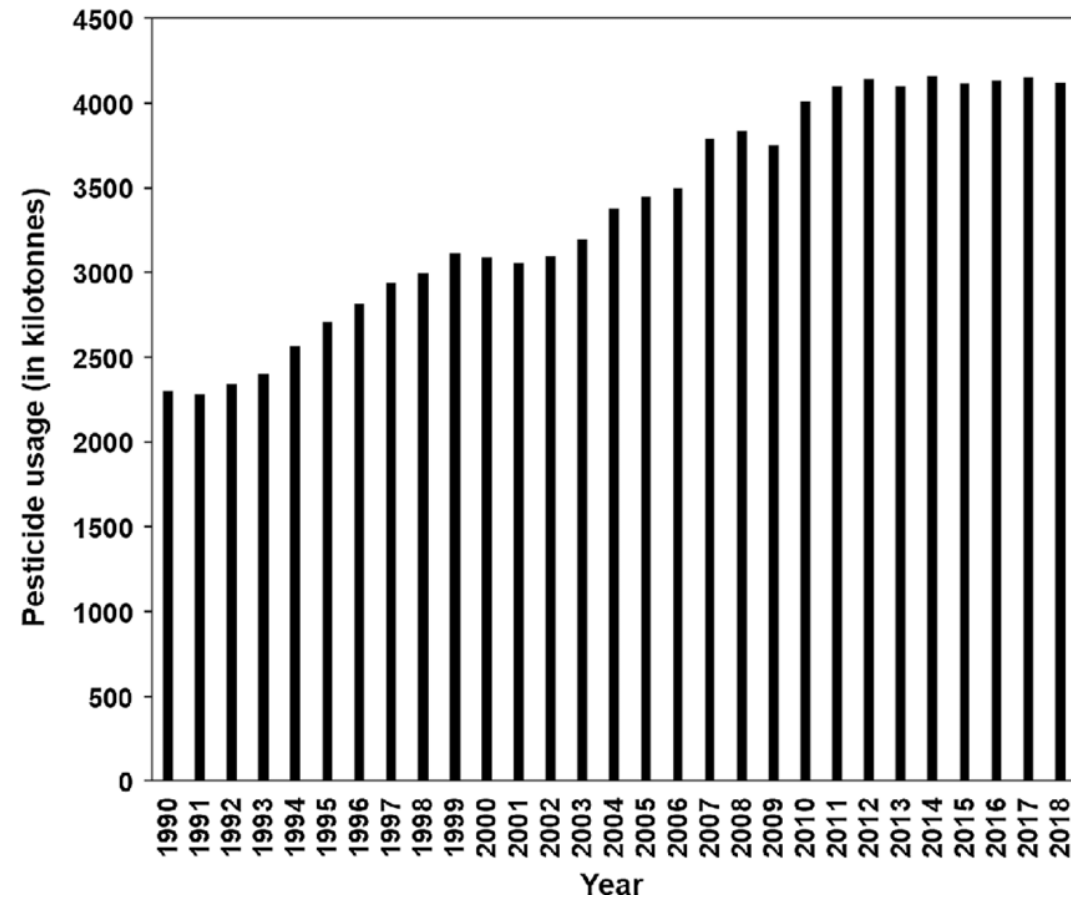
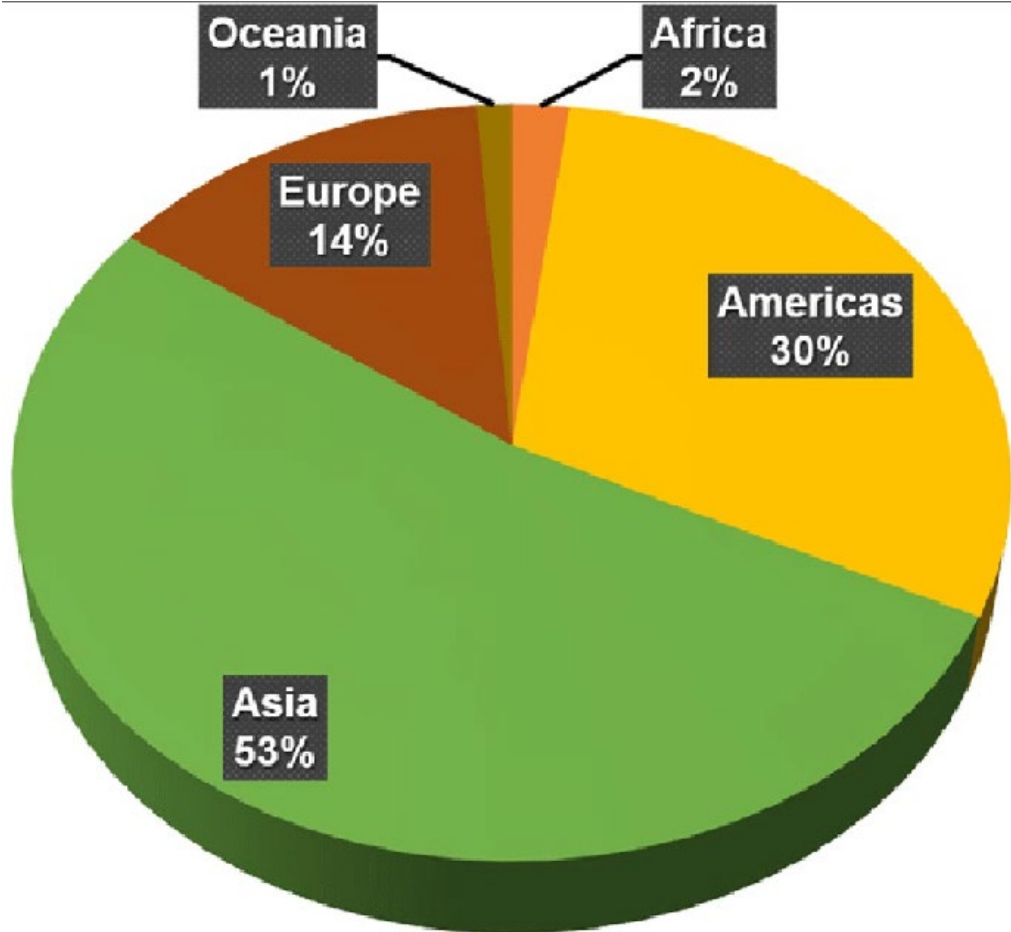
- Agriculture Sector is very important to gain Food Security and Food Safety
- Food production improvement through intensive cultivation
- Fertilizers, pesticides, resistant plants etc. are applied to the agricultural ecosystem
- Is the intensive agriculture for food production improvement always with the consideration for food and the environment safety as well?
- One Health may involve in the implication of this matter



Consumption of fertilizers worldwide in 2019, by country (in milion ton)

<https://www.statista.com/statistics/1287852/global-consumption-fertilizer-by-country/>

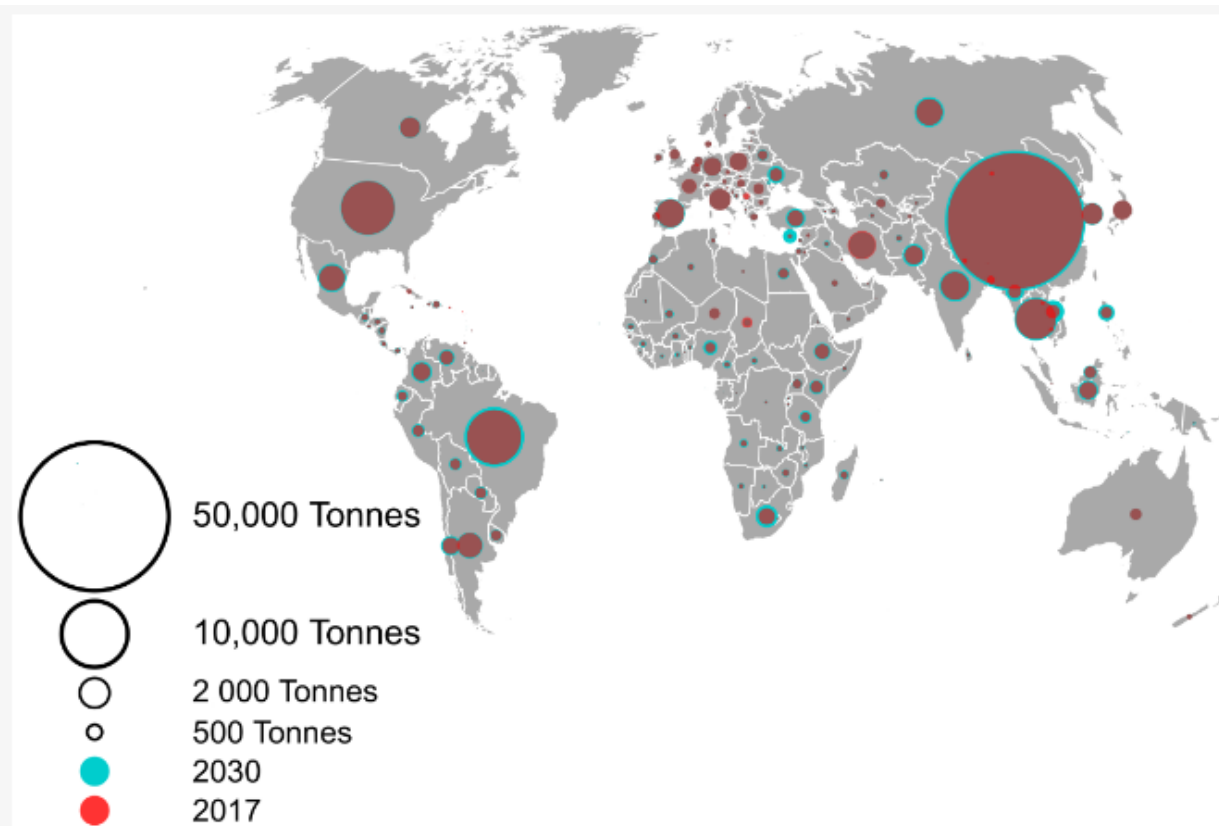
Global distribution of pesticide usage on average for 1990-2018, based on data from FAO (2019)



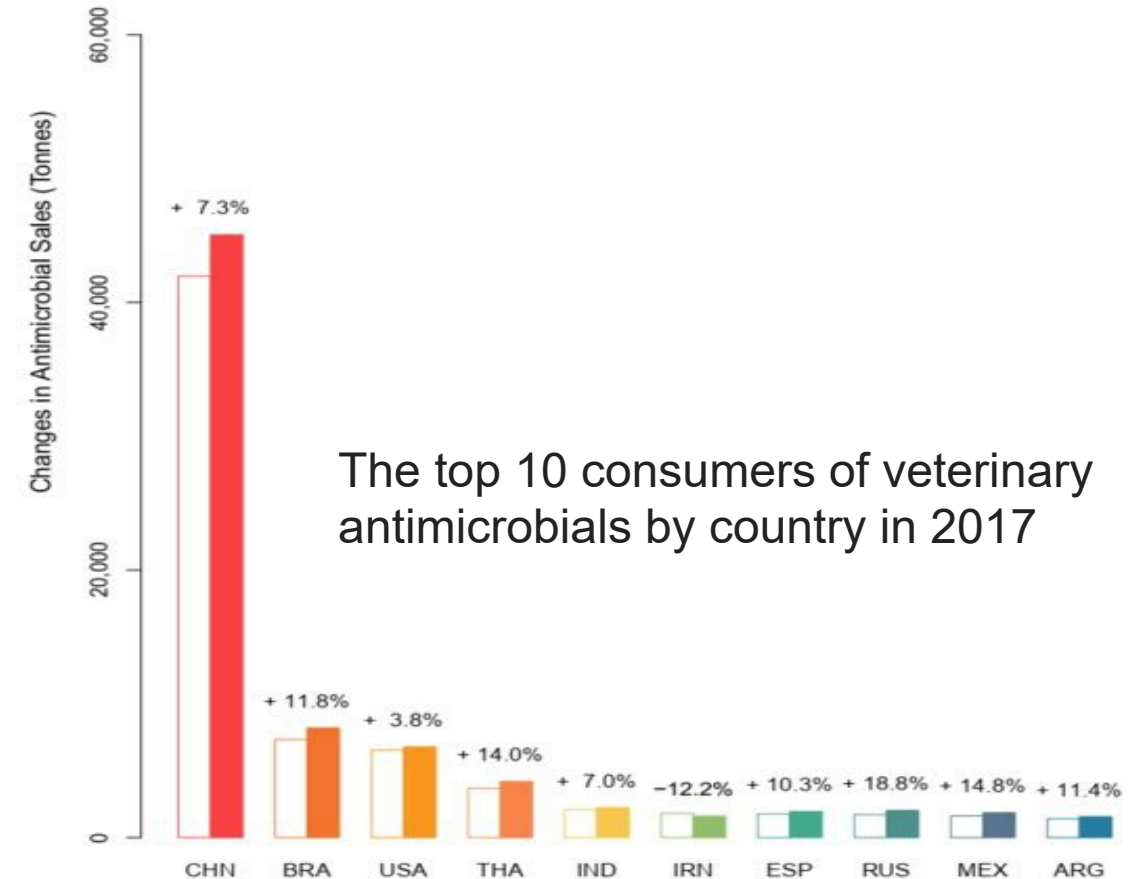
uploaded by Ashish Kapoor

Ponnuchamy et al 2021

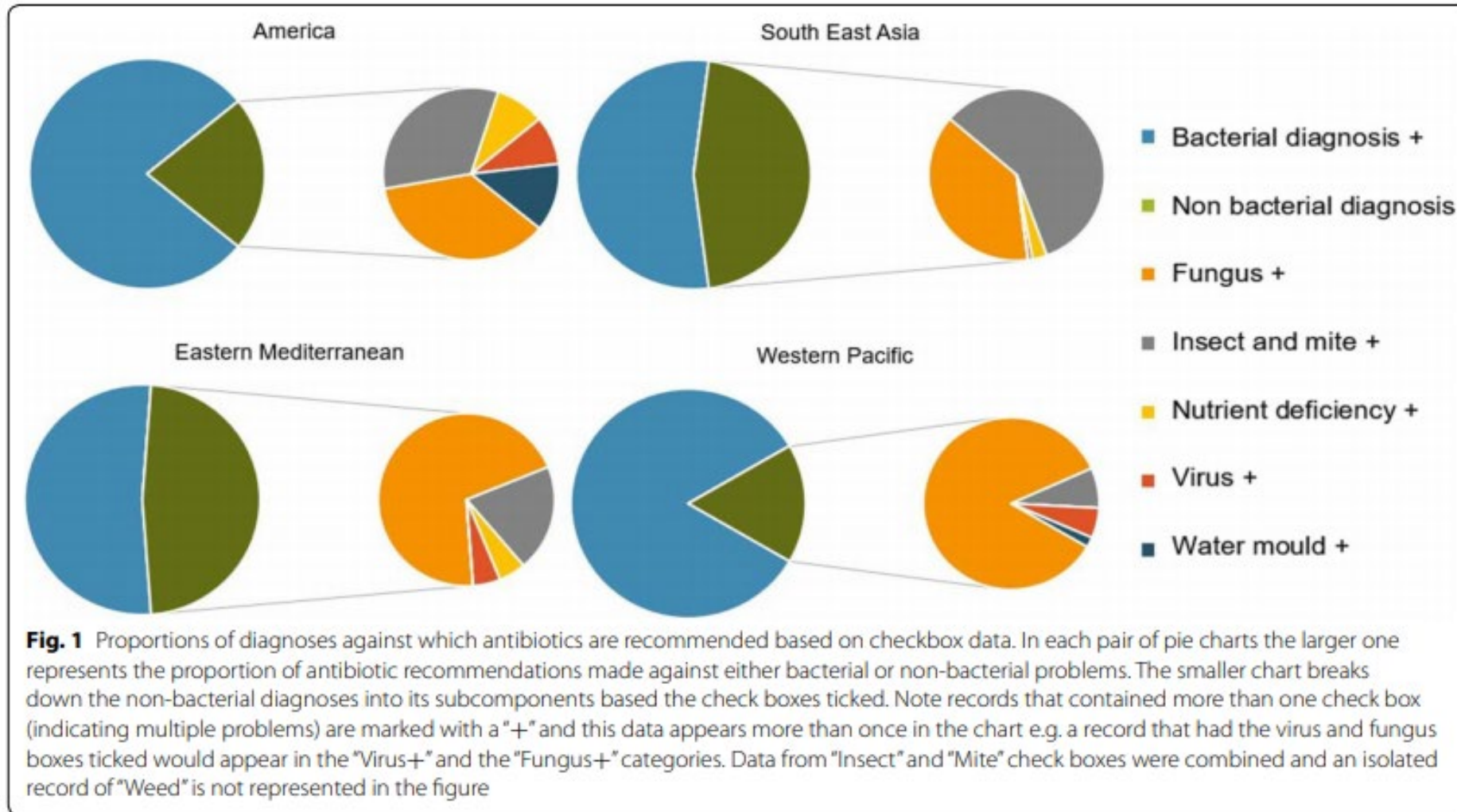
Global Trends in Antimicrobial Use in Food Animals from 2017 to 2030



Antimicrobial consumption per country in 2017 and 2030. The size of the circles corresponds to the amounts of antimicrobials used. Dark red circles correspond to the amounts used in 2017, and the outer blue ring corresponds to the projected increase in consumption in 2030



Synthetic Antibiotic/Pesticide used in Food Production



Taylor and Reeder 2020



**Application of Home
Made Liquid Organic
Fertilizer (HMLOF) /
Pesticides by Small
Holder farmers :
20 ml HMLOF + 5 l
water or 1 l + 100 l water**



Pesticide (insecticides, fungicides, bactericides etc.) applications in intensive agriculture



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- C-organik % : 29,7
- Nitrogen % : 1,65
- Fosfor : 0,5
- Kalium % : 2,3
- pH : 6,6-6,8
- C/N rasio : 18
- Kadar Air % : 11-25



SIMPLE COMPOSTING OF ANIMAL MANURE BEFORE APPLIED AS FERTILIZERAS



Home Made (Small Industry) Liquid Organic Fertilizer (HMLOF)

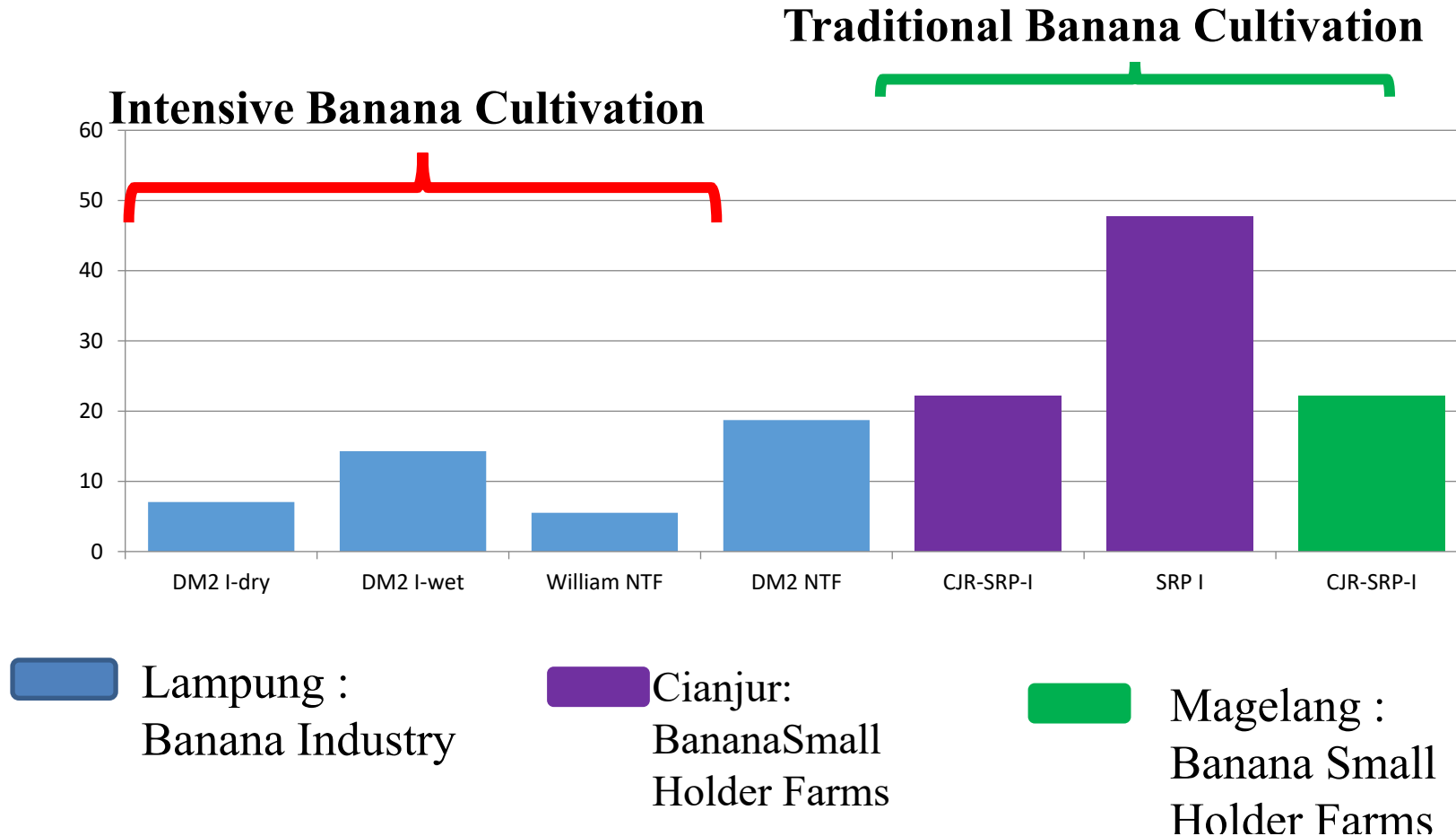


Dung or Mannure

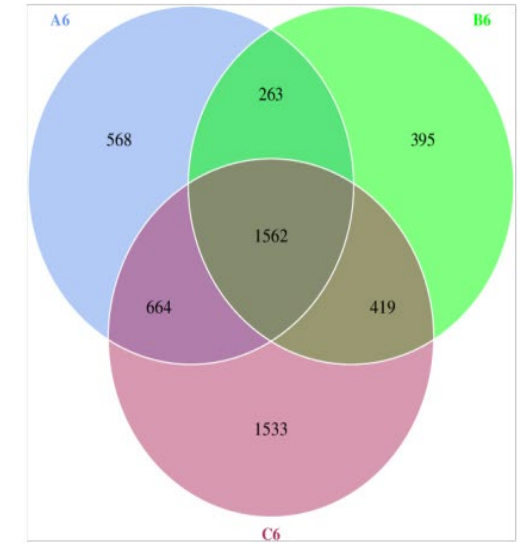
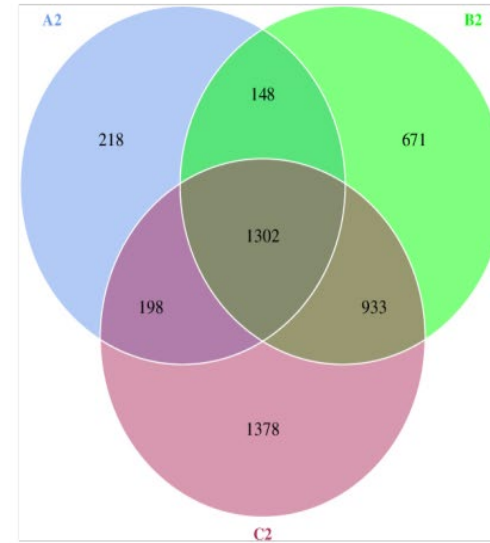
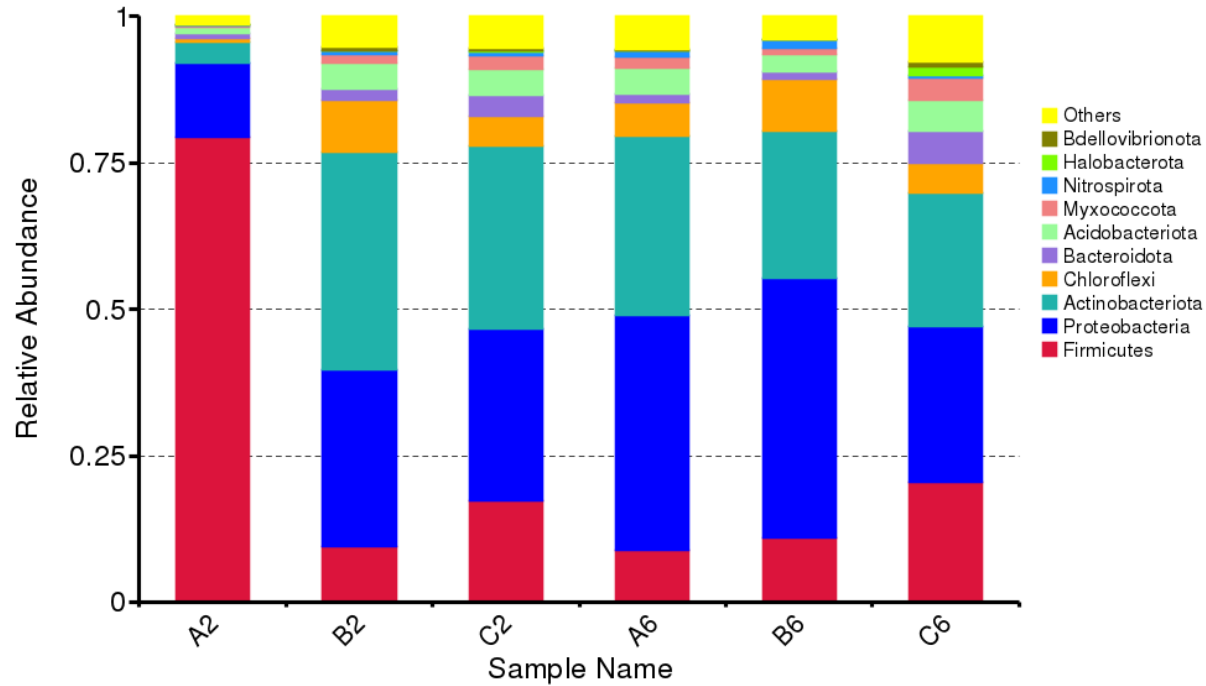




Culturable Dependent Bacterial Population Density in Intensive and Traditional Banana Cultivation



Lower Population Densities of bacteria were found in the Intensive Cultivation lands compared to that in Traditional Cultivation Lands



Metagenomic Analysis of Bacterial diversity and population density of shallot cultivated land : Different Diversity and Population Densities with Different Application of Manure and Pesticides

A and C with the application of manure fertilizers, B without application of manure fertilizer

A2, B2, C2 : 3 x pesticide applications

A6, B6, C6 : 15 x pesticide applications



ANTI MICROBIAL AGENT (AMA)

- Antimicrobials are substances (pesticides, antibiotics, pollutants) that inhibit or kill micro-organisms including antibacterials, antifungals, antivirals, antiparasites, etc.
- Antimicrobials can be natural, semi-synthetic or synthetic.

ANTI MICROBIAL RESISTANCE (AMR)

AMRs are the microbes which are resistant against AMA

ANTIMICROBIAL RESISTANT GENE (ARG)

Microbial genes responsible for the resistance against AMA

Antimicrobial resistance



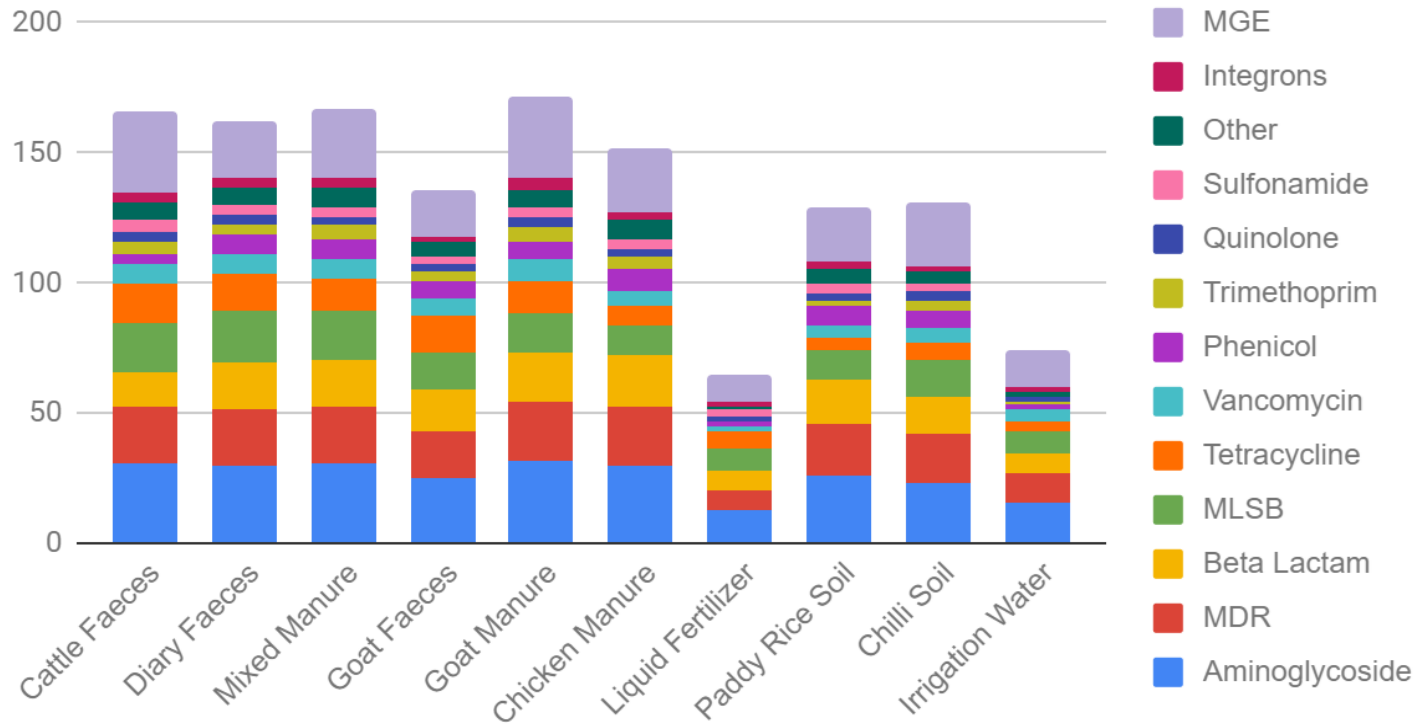
- Antimicrobial resistance is the ability of a micro-organism to withstand the inhibiting or killing action of an antimicrobial.
- In some cases, micro-organisms may be inherently resistant to particular antimicrobials based on specific, defining characteristics of the micro-organism itself.
- This inherent resistance is not caused by agricultural use of medicines.
- **Micro-organisms may acquire resistance via mutations in their DNA, or by exchanging genetic material (genes) with resistant micro-organisms, allowing them to survive exposure to an antimicrobial to which they are normally susceptible**

Detection of Antimicrobial Resistance from Faeces to Manure Fertilized Land At CATI - UGM



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Numbers of Detected Genes in The Center for Agrotechnology Innovation, Universitas Gadjah Mada, Indonesia



Total ARGs identified in all samples :

- Aminoglycoside → 247 genes
- Multidrug resistance → 182 genes
- Beta lactams → 151 genes

Mixed manure and goat manure was processed through drying
Liquid fertilizer was processed by biological fermentation

- This preliminary study shows that the liquid fertilizer can be a good choice for agriculture.
- Our project are still progressing, and we plan to expand the sample to Java Island, Indonesia.

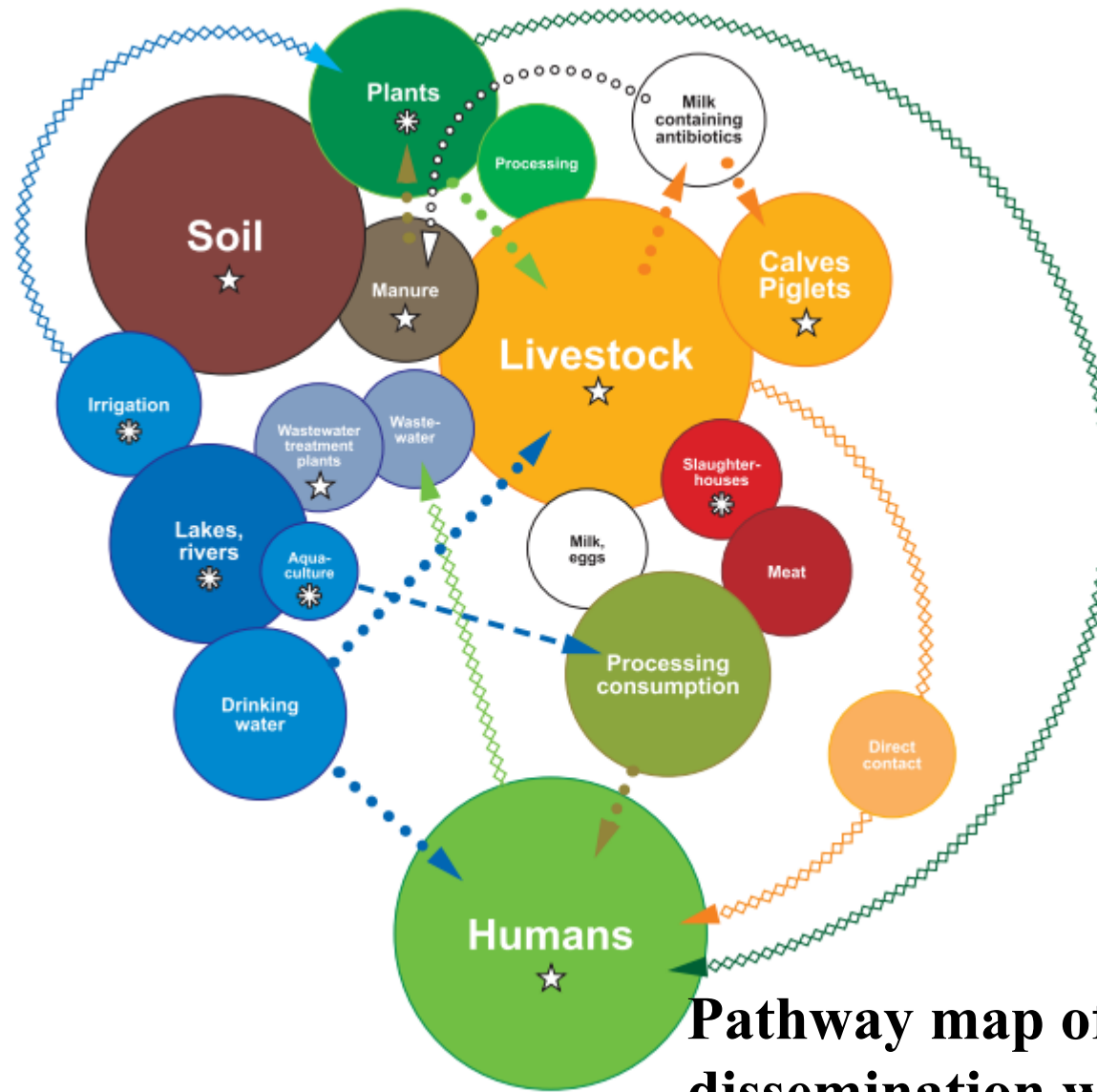
Superbugs



- If bacteria acquire resistance to multiple antimicrobial agents they are called multi-drug resistant.
- The media often refer to bacteria with resistance to a large number of antibiotics as "**superbugs.**"
- When a person or animal is infected by multi-drug resistant bacteria, there are fewer treatment options and recovery may take longer.
- In the worst case, there may be no effective treatments available



- The large volumes of antimicrobials used in agricultural systems can promote resistance.
- Antimicrobials used in agricultural production systems help to maintain animal welfare and plant production, and control animal and plant diseases.
- The application of antimicrobials need to be used **cautiously** in agriculture, human medicine and public health.



Movement of AMA or AMR is indicated by overlapping circles and arrows, respectively; different colors define different groups of reservoirs. Stars indicate the hot spots of ARG and ARB with high bacterial densities, nutrient availability, and selective pressure in the digestive tract of livestock and humans, in manure storage facilities, wastewater treatment plants, and in the rhizosphere. Asterisks indicate possible hot spots of ARG and ARB in water, sediments, and biofilms in aquaculture, rivers, lakes, and irrigation systems, as well as in slaughterhouse facilities and on plant surfaces

Pathway map of AMA (anti microbial agent) and AMR dissemination within agriculture, the environment, and the food processing industry.



Knowledge gaps

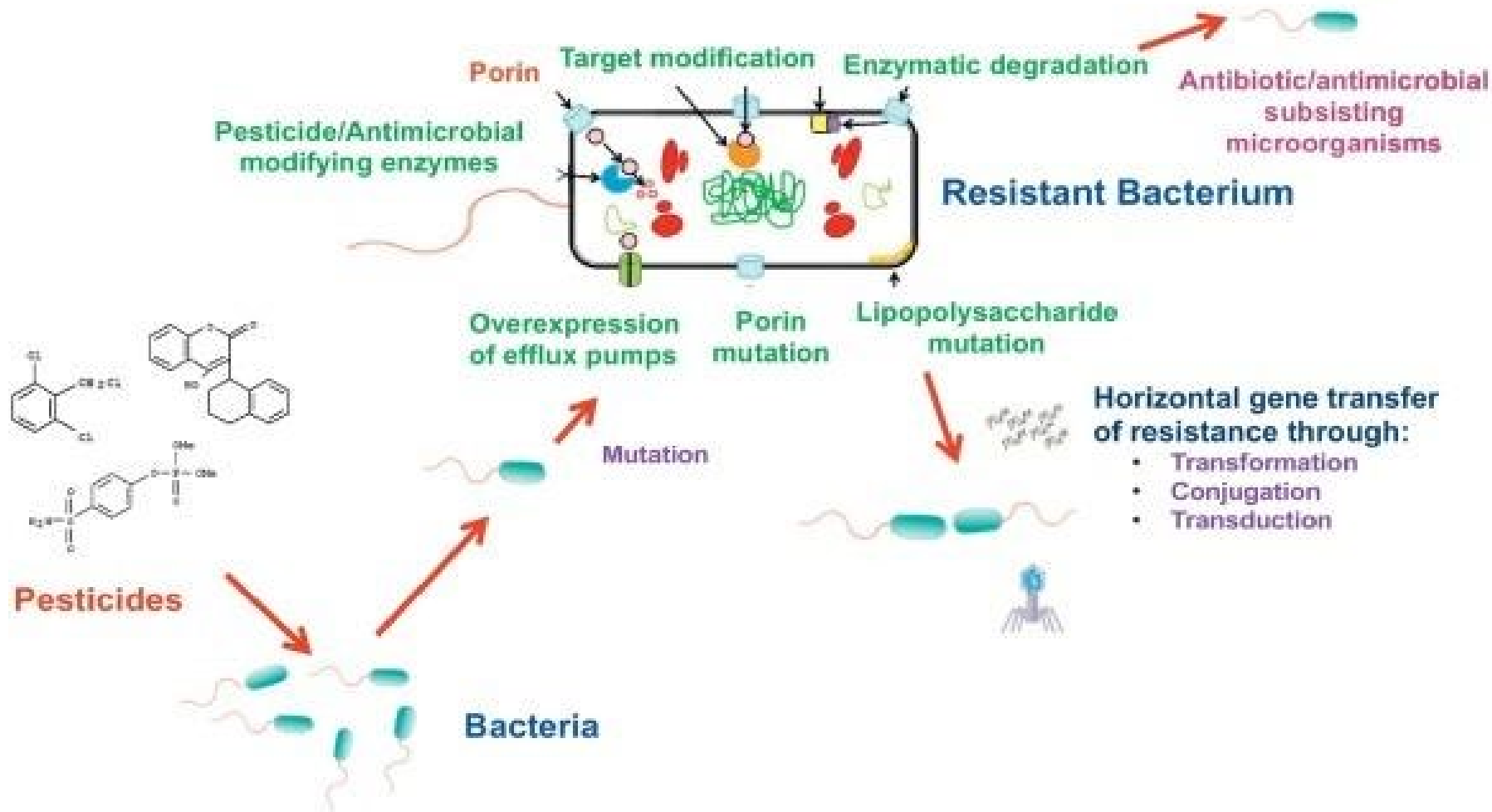
Surveillance data on the use of AMA for animal and plant health
Surveillance data on specific ARG in zoonotic agents and commensal bacteria in livestock, not only their level of AMR
Surveillance data on the amount of AMA, ARB, and their ARG in manure
Effects of different manure treatments on abundance of AMA, ARG, and ARB, as well as on frequency of HGT
Qualitative and quantitative data on sorption and fixation of AMA in soils
Quantitative data on antibiotic potency of AMA that are sorbed and fixed in soils and sediments
Persistence and eventual selection for AMR in AMA-containing soils
Chance of human-associated as well as animal-associated bacteria and pathogens to acquire AMR by MGE from commensals and environmental microbes
Quantitative data on the inoculum needed to add an ARB to the microbiomes of humans and animals
Pathways from agricultural and urban sources of AMA and ARG into sewage and the aqueous environment
Efficiency of wastewater treatment regarding elimination of ARG as well as AMA
AMA and ARB uptake in plants and influence of soil types on uptake
Direct relationship between irrigation water or manure containing ARB and ARG and the abundance and spread of corresponding bacteria or ARG on crops
Chance of human-associated as well as animal-associated bacteria and pathogens to acquire AMR by MGE from bacteria in the phyllosphere
Effects of drying or silaging roughage on the prevalence of ARG and ARB in the phyllosphere
Potential influence of direct use of AMA in plant production on contamination, selection, and spread of environmental, animal, and human resistomes
Evaluation of risk factors for selection and spread of AMR in animals and environmental reservoirs
Evaluation of transmission routes of AMR as well as human exposure data related to agricultural products
Role of biofilms in spread of ARG and ARB in the environment and food processing plants
Kinetics of spontaneous removal of ARG and ARG from soil, water, and food environments

Knowledge gaps regarding AMR in plant and animal agriculture and roles of these sites as sources and sinks of AMR

Thanner et al 2020

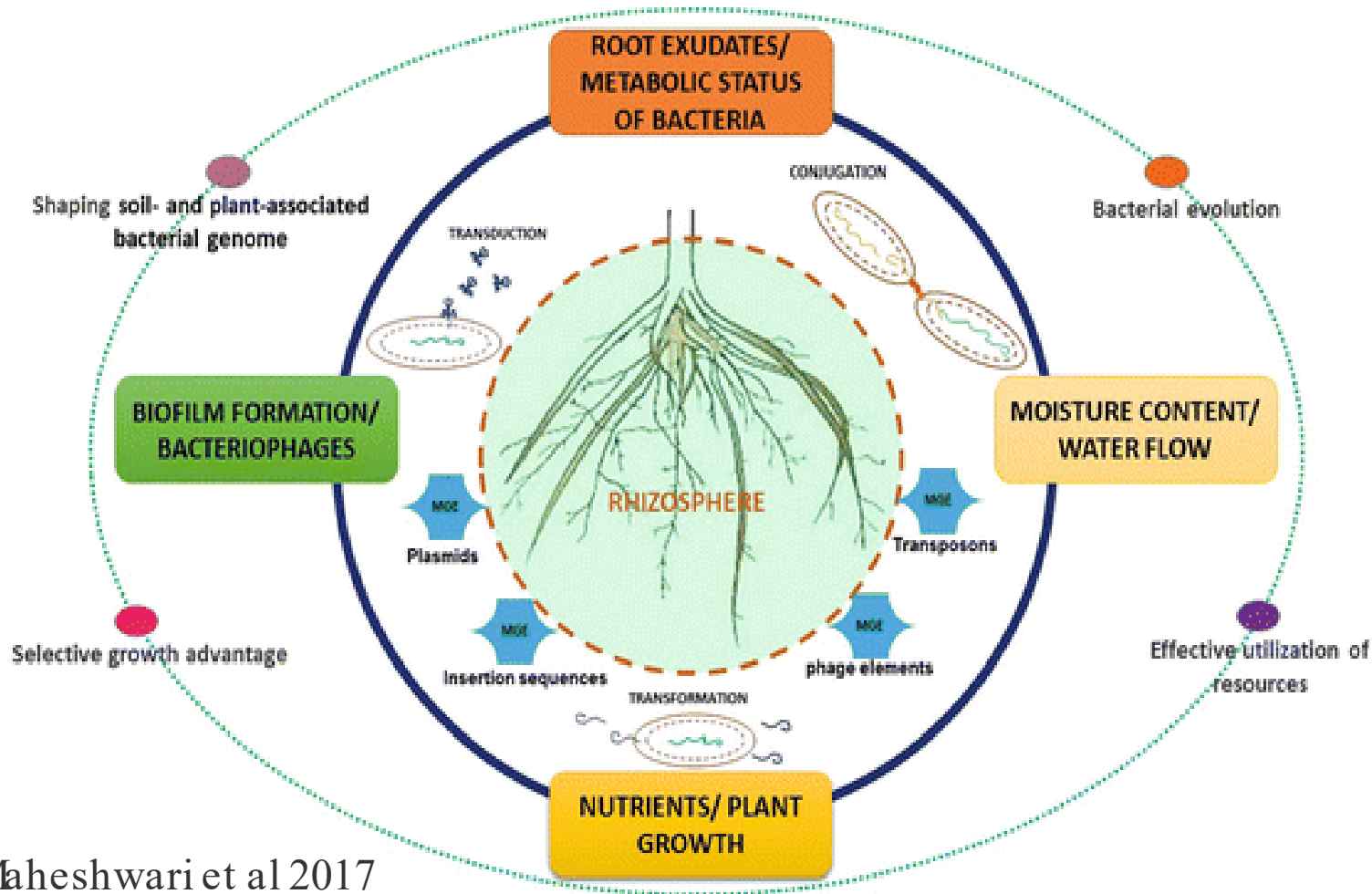
Possible Resistance Mechanisms

Pesticide degraders



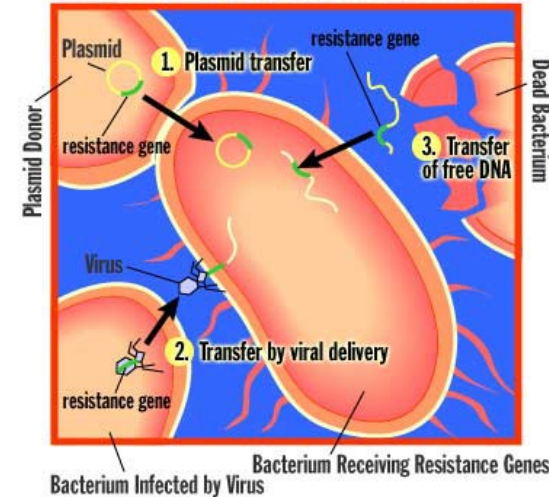
Contribution of pesticide application on AMR development

Horizontal Gene Transfer in the Soil and Rhizosphere



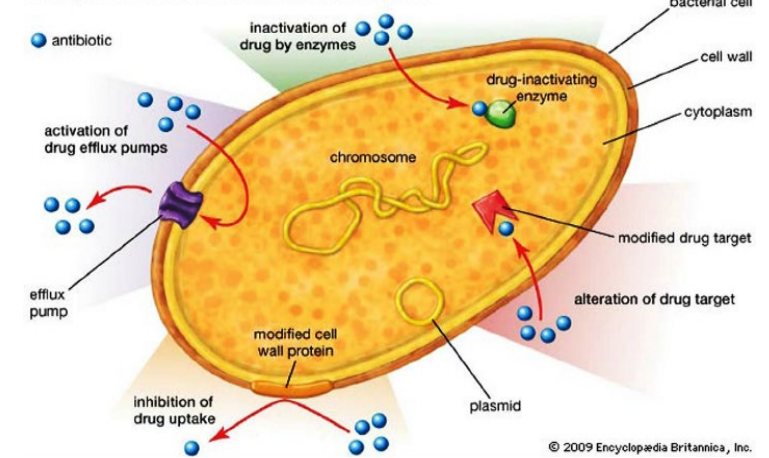
Maheshwari et al 2017

Transferring Resistance Genes



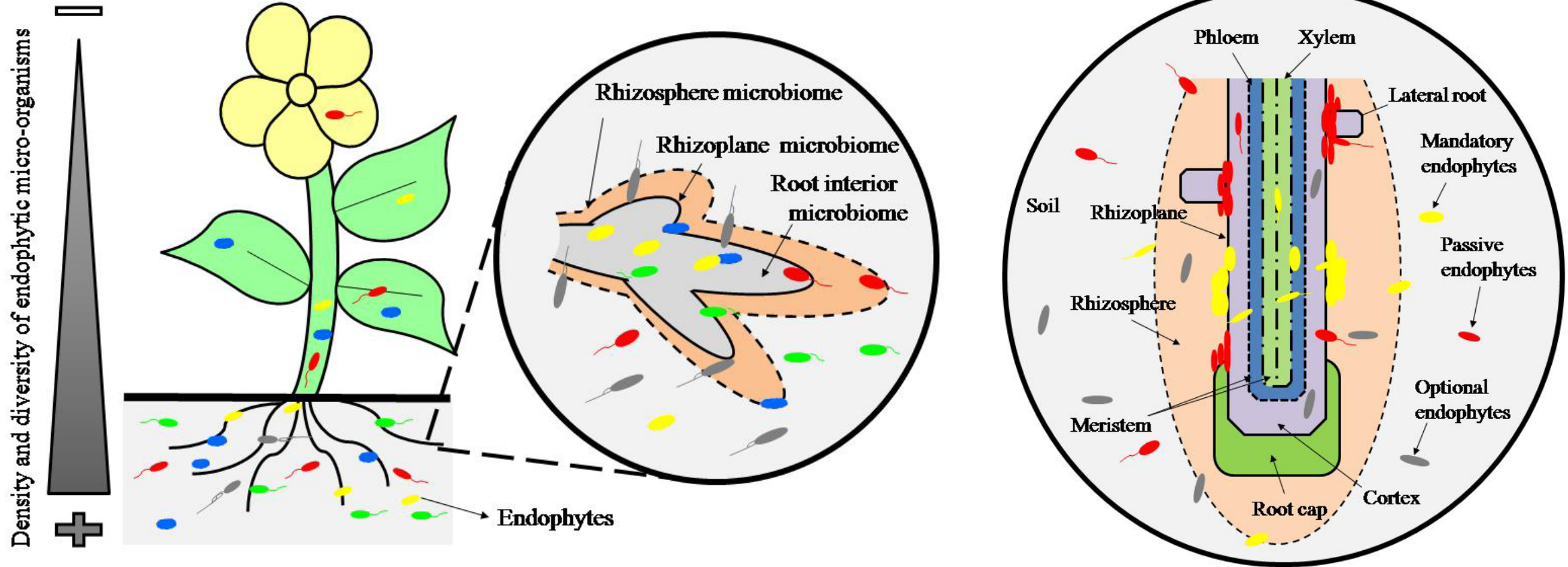
David White et al 2001

Examples of mechanisms of antibiotic resistance



© 2009 Encyclopædia Britannica, Inc.

Bbosa et al 2014

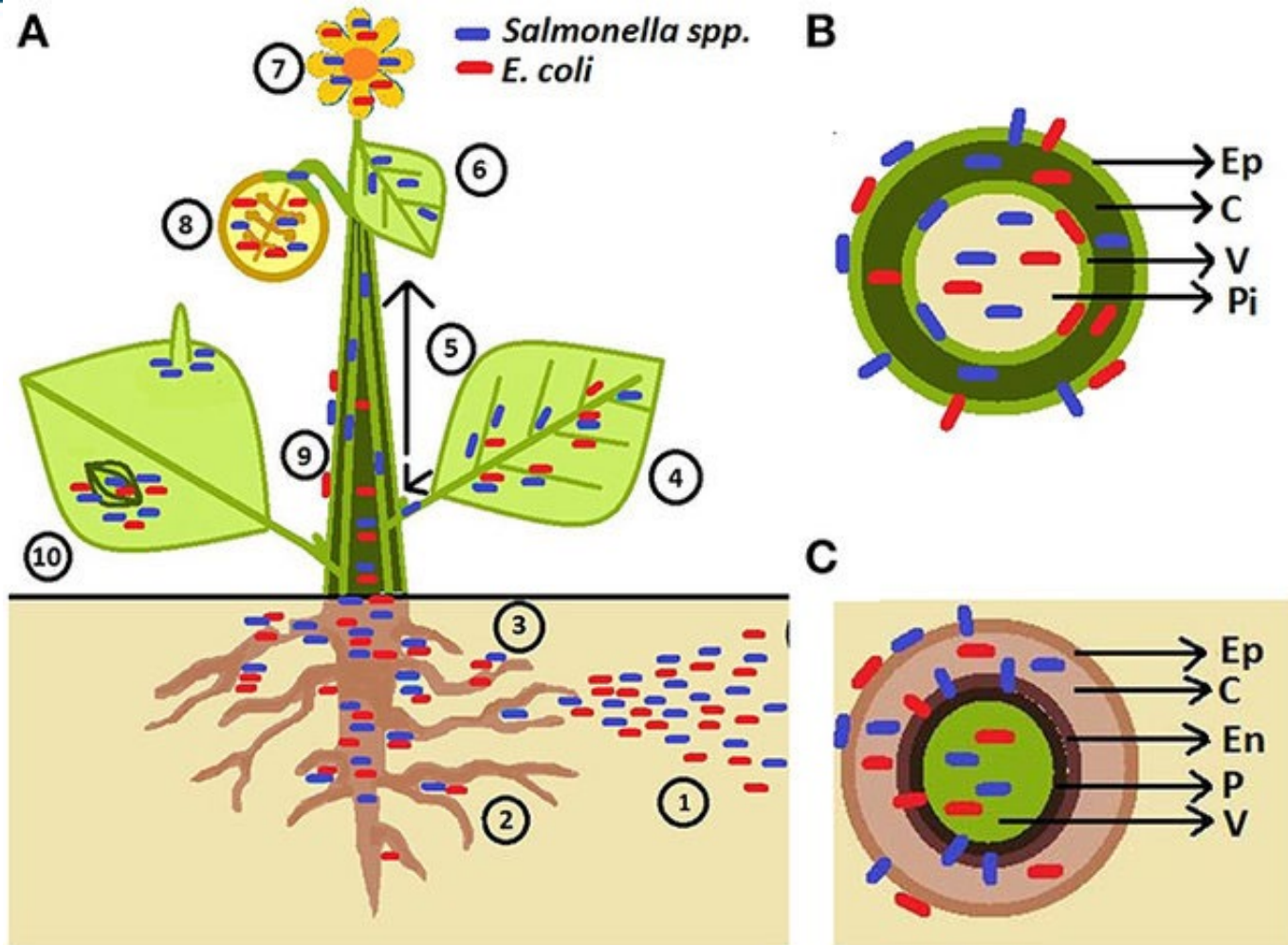


Some Rhizospheric Microbes in the soil may live as Endophytes in the plant tissues

dos Santos et al 2018



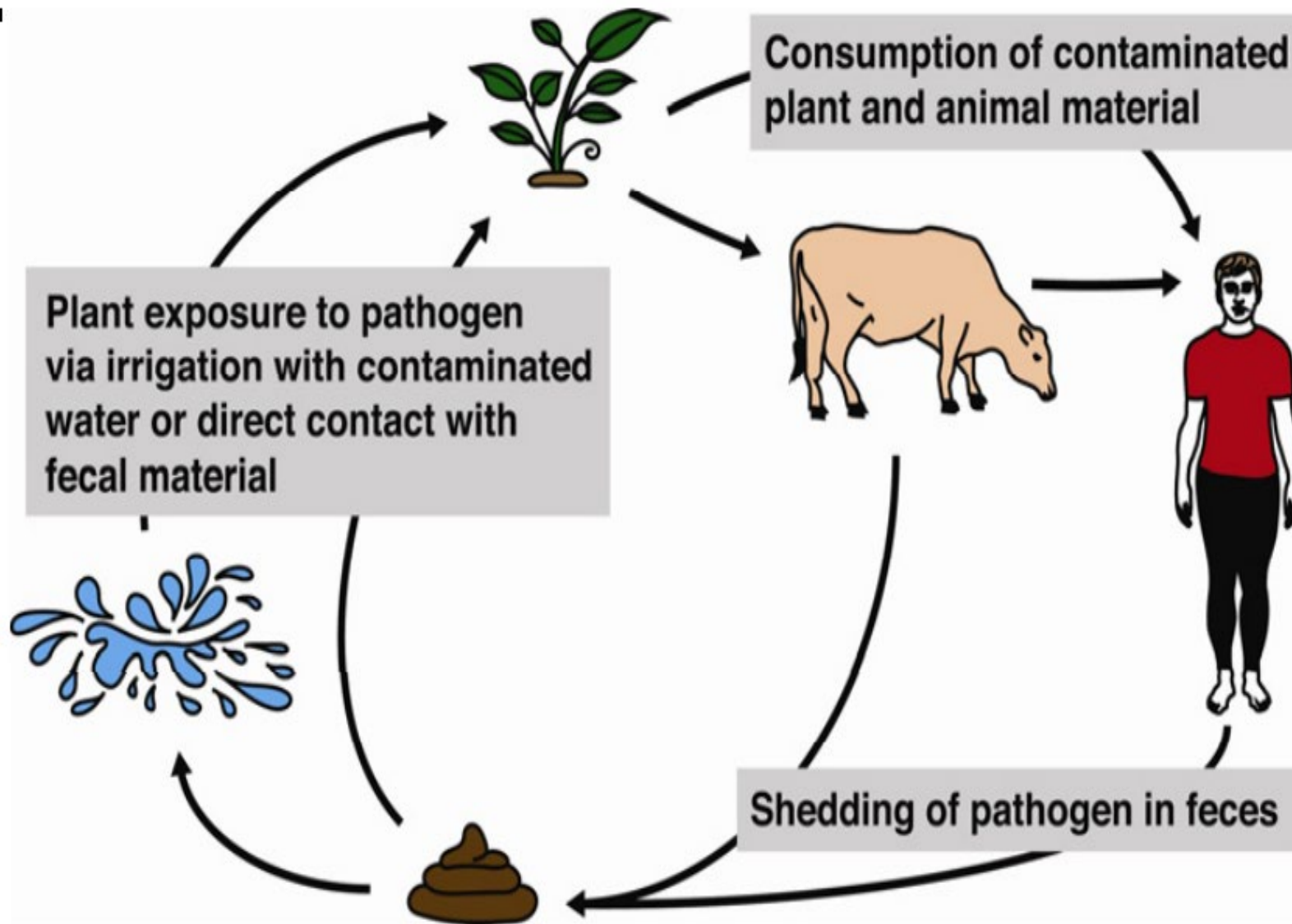
Schematic representation of human pathogen (HP) association with plants



- (A) Pathogens are introduced to soil through contaminated irrigation water, fertilizers, manure, and pesticides
- (B) Stem cross-section showing bacteria located in different tissues (Ep, epidermis; C, cortex; V, vascular tissue; Pi, pith)

Melotto et al 2014

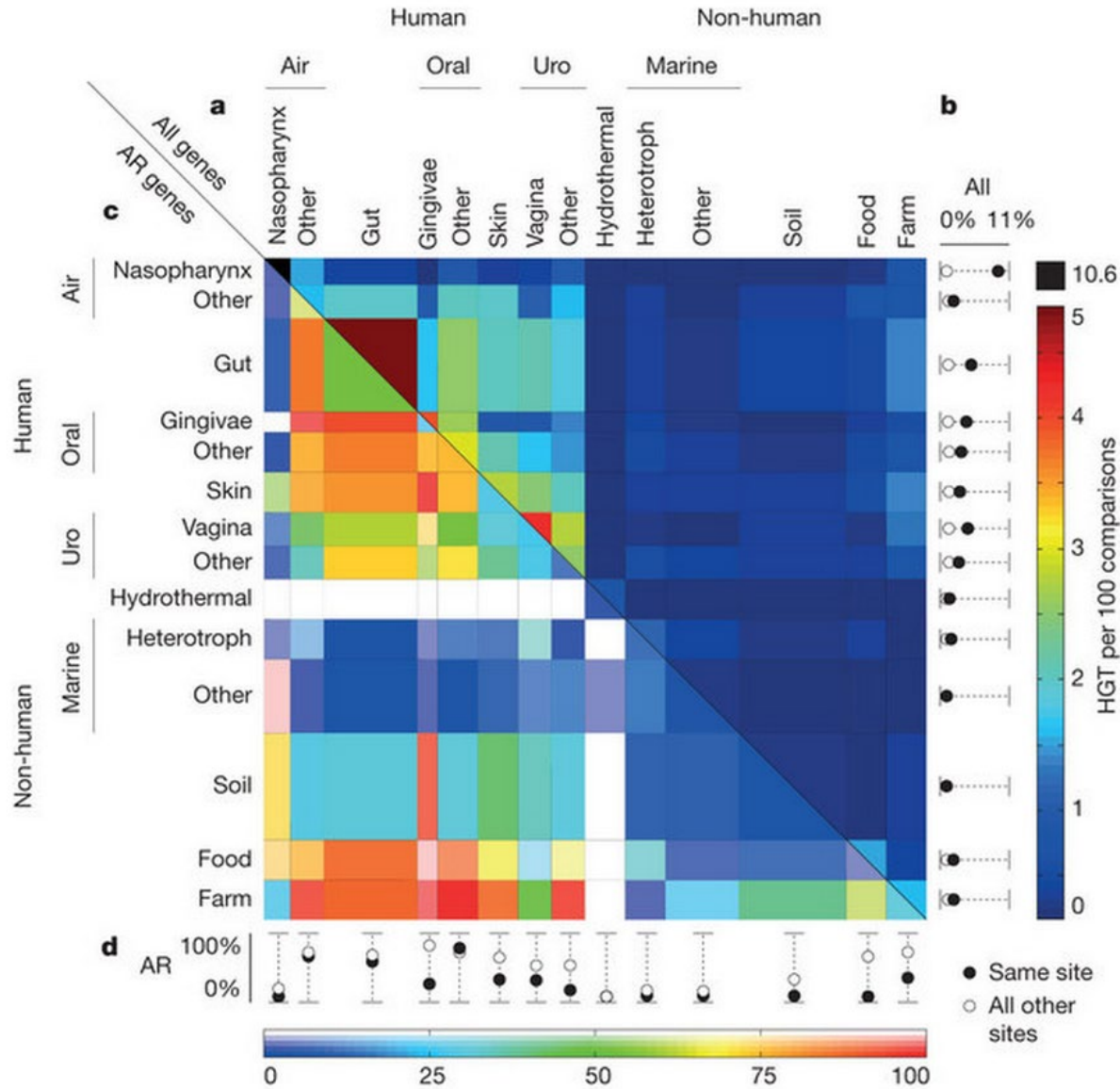
(C) Root cross-section showing bacteria on the root surface, internalizing between the epidermal cells, and colonizing root outer and inner cortex, endodermis (En), pericycle (P) and vascular system



An emerging concept of the ecological cycle of human enteric pathogens on humans, animals, and plants

Fletcher et al 2013

Diagram by Angela Records



Worldwide bacteria network may readily swap beneficial genes

Frequency of horizontal gene transfer between different environments for all functional groups (a,b) and antibiotic resistance genes only (c,d) (credit: Chris S. Smillie et al./Nature)

Conclusion

- The application of Animal Manure and Pesticide may change the diversity and population density of soil microbiome
- The use of Antimicrobial Agent in Agriculture for Food Production may immerge on Antimicrobial Resistance Microbes leading to the spread of AMR into plant, animal and human pathogens
- One Health should be the answer for better management of the AMR spread in the agricultural ecosystem and in the environment



Thank you for attention