



Do Cows Provide More than Milk? –Antibiotic Resistance in Oregon Coastal Environments

Coastal Environments

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Background

Antibiotics are used to fight infections, but they can also become a health risk when over- or misused. Resistance to antibiotics, termed antibiotic resistance, means that bacteria have acquired defense mechanisms, resulting in the antibiotics' no longer being effective. Antibiotic resistance is encoded on genes, and bacteria can acquire it accidentally by spontaneous gene mutation or by passing these resistance genes amongst themselves (horizontal gene transfer).

Bacteria can move antibiotic resistance genes through the food chain (Subbiah et al., 2020). Additionally, low-level exposure to antibiotic residues could happen in water run-off, where human and animal waste collects. This low-level exposure can lead to the selection of a resistant phenotype (Jorgensen & Halling-Sorensen, 2000).

The spread of antibiotic-resistant genes can result in minor bacterial infections becoming untreatable, and pathogens can become resistant to multiple antibiotics rendering last-resort antibiotics ineffective.

Rationale: Manure run-off from agricultural lands and sewage from human settlements could result in the emergence of antibiotic-resistant bacteria in Oregon's estuaries where they could collect in farmed and recreationally harvested shellfish, water, and sediment samples.

Hypothesis: Antibiotic resistance genes can be found in bacteria of Oregon estuarine environments.

Objectives: Determine whether antibiotic resistant bacteria are associated with a) shellfish, b) water, and c) sediment samples collected in Oregon's estuaries.

Results

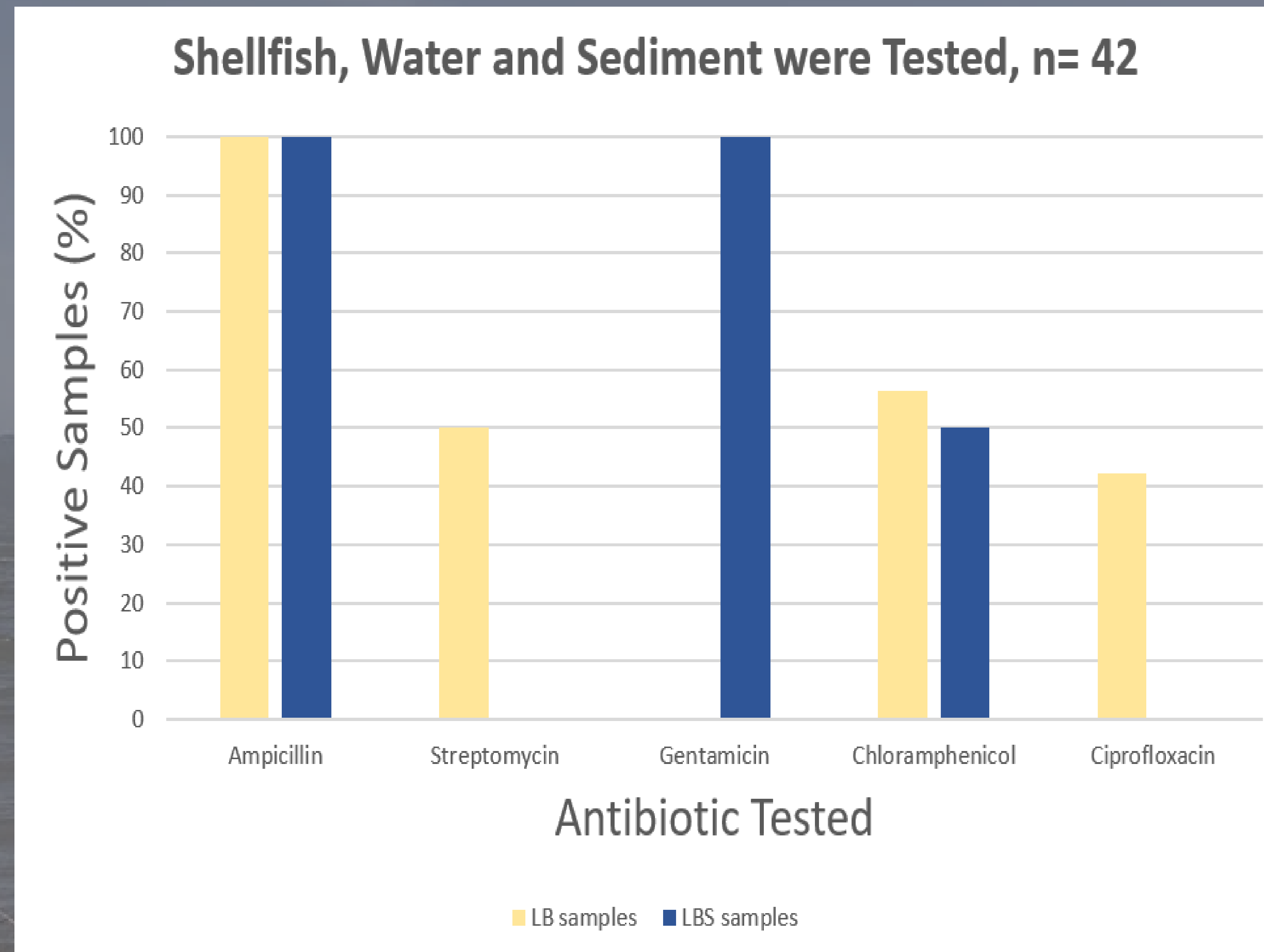


Fig. 1. Preliminary data. Each of the 42 samples (water, sediment, oyster, clam) contained at least one bacterial strain that was resistant to one of the tested antibiotics.

Conclusions

- Antibiotic resistance can be detected in environmental bacteria
- Our results suggest that there are antibiotic resistance genes in bacteria thriving in Oregon coastal environments
- Presence of ciprofloxacin-resistant bacteria suggests human waste sources (this antibiotic is not used in livestock)
- More research must be done, and the experiment should be repeated
- Mindful handling and processing of raw seafood is always prudent
- Antibiotic resistance genes may impact human health



Fig. 2. Pacific oysters, *Crassostrea gigas*. Photo: B. Whitten.

Methodology

Water, sediment, oysters, and clams were sampled/purchased from ten locations between Tillamook Bay, OR to Waldport, OR. The bacteria cultured from these samples were then grown and isolated on LB and LBS agar. LB would grow terrestrial bacterial isolates, while LBS contains seawater and should grow bacteria that can withstand higher salinities. Isolates were subsequently stamped onto agar plates containing antibiotics. If colonies grew on these plates, the sample was considered antibiotic-resistant and counted as a positive result.

Future Direction

- Redo experiment with refined protocols
- Confirm the presence of the antibiotic resistance conferring genes using PCR
- Establish the prevalence of antibiotic resistance in the 4,000+ strains collected rather than the prevalence in the 42 samples
- Test antibiotic resistance against tetracycline (positive results could suggest livestock origin)

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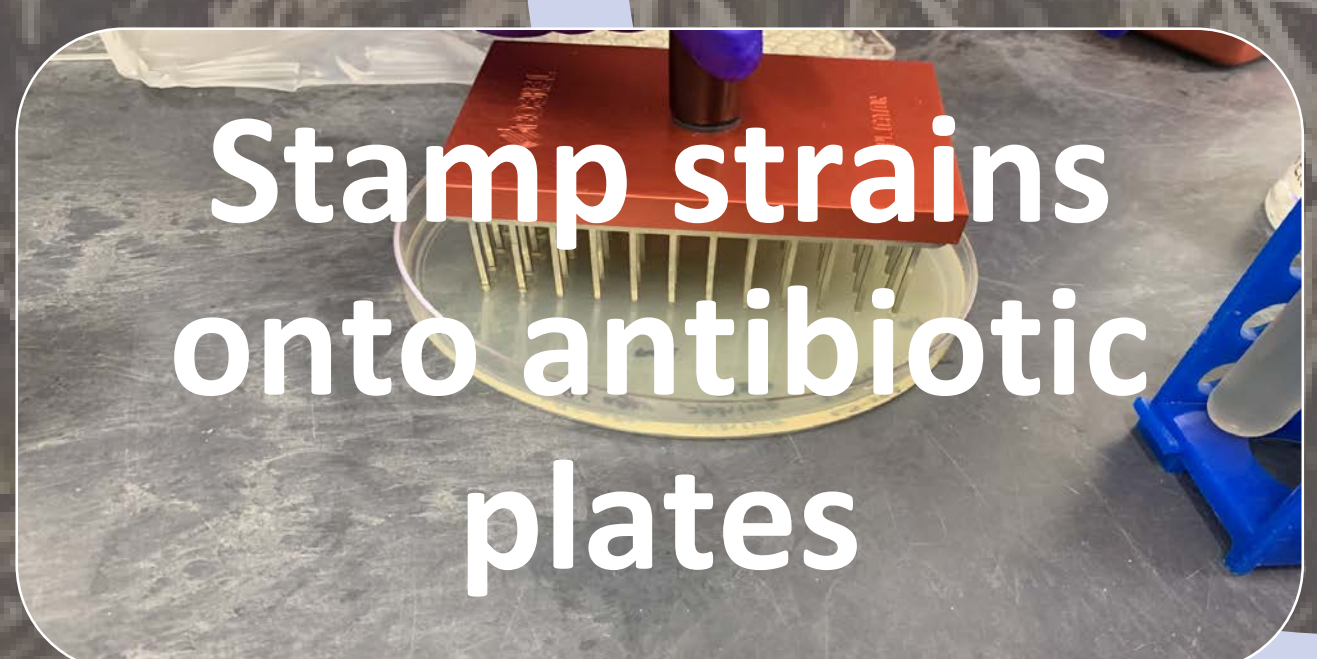
Liu J, Zhu Y, Jay-Russell M, Lemay DG, Mills DA (2020): Reservoirs of antimicrobial resistance genes in retail raw milk. *Microbiome*, 8, 99. Doi: 10.1186/s40168-020-00861-6. (source of the idea and the control strain for this project)



Collect samples



Plate samples & isolate strains



Stamp strains onto antibiotic plates



Fig. 3. Authors: Top row: B. Whitten, J. Jacob, S. Lunda; Bottom row: C. Schubiger (photo).