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Metagenomic approaches to unravel Antimicrobial peptides from the zebrafish gut

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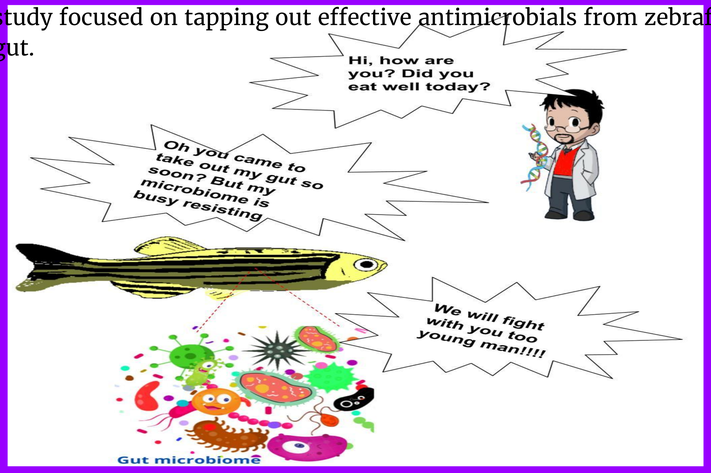
ABSTRACT

The gut microflora plays a significant role in protecting the host from serious health complications. A disruption in the normal gut balance is an established reason in various pathological conditions. The recent advancements in genomics unravels mysteries between the co-existence of the host and gut microbiome. Development of potential antimicrobials relies on tapping them from the natural resources closely related animal models. This study is focused on elucidating functional antimicrobial resistance mechanisms adapted by the gut-biome of zebrafish. Metagenomic strategies applied to the zebrafish gut biome demonstrated the abundance of diverse inhabited flora. Gene ontology analysis through Macrel and Pubseed servers divulged the antimicrobial mechanisms and elucidated 2,356 antimicrobial peptides. Screening of the potential antimicrobial peptides based on their properties enabled in the identification of few promising antimicrobial peptide candidates active against *Klebsiella pneumoniae, Pseudomonas aeruginosa, Staphylococcus aureus, E.Coli* and *Bacillus cereus*. Molecular docking and dynamics methods were employed to persuade a single potential antimicrobial peptide with the sequence "MPPYLHEIQPHTASNCQTELVIKL". Metabolic model construction and the flux balance analysis performed on the metagenome assured the existence of multitude of antimicrobial biosynthetic pathways including puromycin, tetracycline, novobiocin, streptomycin and cephalosporins. The work established the key potency of the zebrafish gut commensals in tackling the antibiotic resistance strategies. The study is novel and expands our knowledge on the zebrafish gut biodiversity with an emphasis on tapping out potential antimicrobial peptides to be developed against most of the resistant bacterial species.

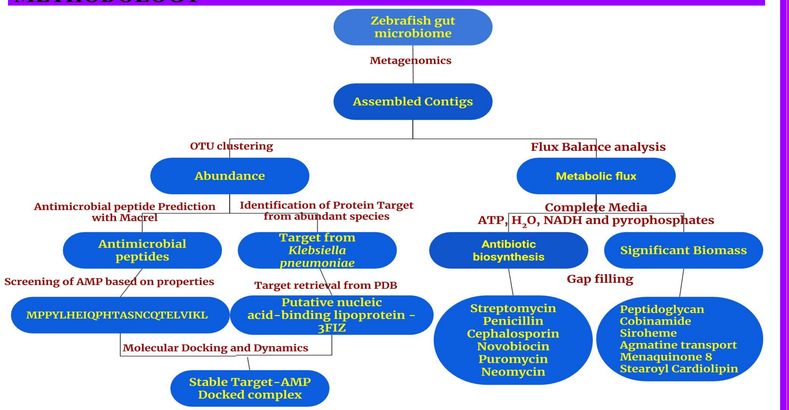
Keywords: antimicrobial peptides, zebrafish, metagenomics, molecular docking, peptide dynamics, stability, flux balance analysis

INTRODUCTION

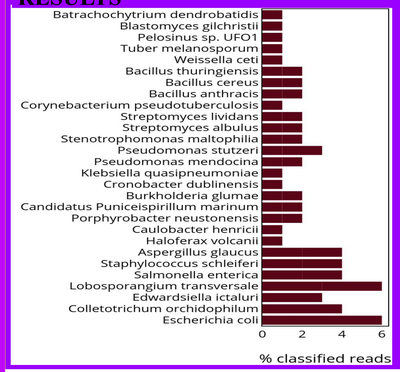
Exhaustive abuse of antimicrobials caused devastating resistance mechanisms and an alternative antidote is the most sought development in the post-antibiotic era. Zebrafish gut that dwell diverse microbes has been extensively studied since 1960 and are significant assets of antimicrobial fulcrum. Unexploited natural resources are excellent manoeuvre for antimicrobials. Hence the study focused on tapping out effective antimicrobials from zebrafish gut.



METHODOLOGY



RESULTS



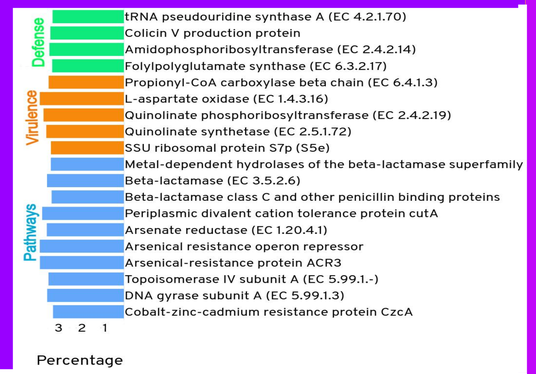


Figure 1: Abundant organisms from the gut **Figure 2: Pathways, Defensins and Virulence mechanisms**

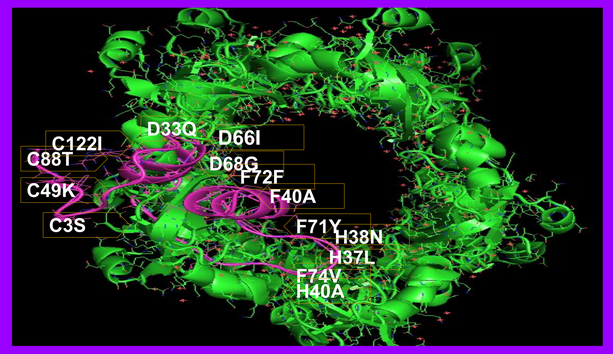


Figure 3: Peptide1 (pink) interacted with 20 residues of target(green) with an energy of -429.34 Kcal/mol.

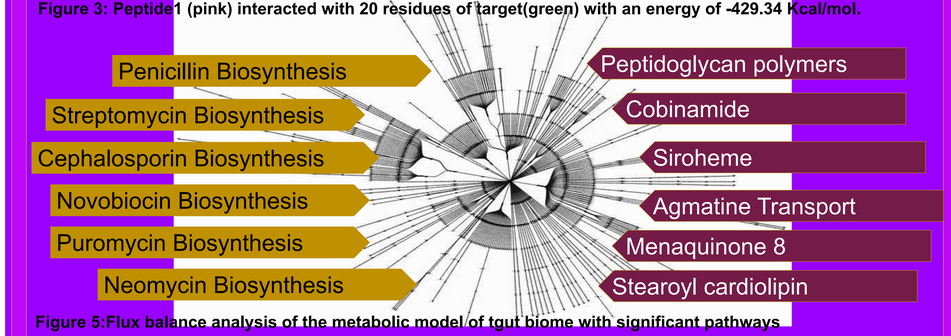


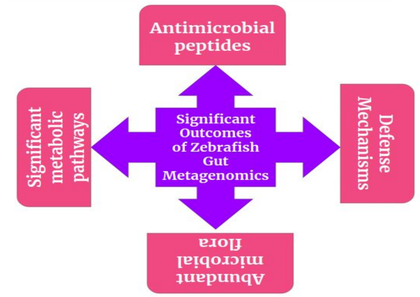
Figure 5:Flux balance analysis of the metabolic model of tgut biome with significant pathways

DISCUSSION

We enumerated the abundant microbes that are part of the zebrafish gut and their defense mechanisms using metagenomics approaches

- E. coli, Lobosporangium transversale, Colletotrichum orchidophylum, Salmonella enterica, staphylococcus schleiferi, and Aspergillus glaucus were abundant.
- Abundant enzymes were t-RNA pseudouridine synthase A, Colicin V production protein, Acetyl-coenzyme A carboxyl transferase, folylpolyglutamate synthase, Quinolinate synthase, small ribosomal unit protein S7p, beta lactamase, copper, zinc, cadmium, and mercury resistant enzymes.
- Peptide 1, "MPPYLHEIQPHTASNCQTELVIKL" showed promising results with a net charge of +2.5 and 53% hydrophobicity.
- Valid 3D structure of peptide 1 as inferred from Ramachandran plot with 92.1% of residues lying in the allowed region.
- Molecular Docking of the peptide with the putative nucleic acid binding protein of Klebsiella pneumonia rendered an excellent binding score of 1124 and energy of -429.34 Kcal/mol.
- The peptide contacted the target and established a strong interaction with glycine, threonine, isoleucine, and glutamine of chains C and F.
- Molecular dynamics confirmed the stability of target AMP complex with an average fluctuation of 1A^0
- From the flux balance analysis, the gut biome of zebrafish was inferred to be an excellent source of various antimicrobials, cardio and neuroprotective agents and act as amazing models for heart and neurology.

CONCLUSION



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