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IN VITRO PROFILING OF POTENTIAL FISH PROBIOTICS, *Enterococcus hirae* spp., ISOLATED FROM JADE PERCH *Scortum barcoo* AND SAFETY PROPERTIES ASSESSED USING WHOLE GENOME SEQUENCING

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Exploiting the use of probiotics in aquaculture is one key strategy to promote fish growth while mitigating fish-associated diseases. Among the different types of probiotics, host-derived probiotics exhibit promising efficacy compared to commercial or terrestrial-derived ones as they are adapted to the host environment. This study aimed to identify and characterize potential probiotics from jade perch (*Scortum barcoo*), one of the newly farmed fish species in Singapore, to enhance its disease resistance.

Gut isolates from jade perch were screened for their ability to inhibit the growth of pathogenic *Aeromonas sobria*, *Streptococcus iniae*, and *Streptococcus agalactiae*. Several isolates that demonstrated antagonistic effects against *A. sobria* and *S. iniae* were later identified to be *Enterococcus hirae*. The pathogen inhibition efficacy of this isolate was also examined in both jade perch and tilapia gut suspensions, mimicking more complex host microbial environments. Significant clear zones were found when co-incubated with jade perch gut suspensions, whereas only minimal inhibition zones were produced in tilapia gut suspensions, suggesting the advantage of using host-derived probiotics.

The safety of the final probiotic candidate was evaluated using whole-genome sequencing. This strain did not harbor any known toxins and mobile genetic elements. Conjugation experiments confirmed that its inherent antibiotic resistance gene *aac* (6')-lid was not transferable. Two classes of bacteriocins were found in the probiotic genome, which partially explained its antimicrobial activities against both Gram-positive and Gram-negative pathogens. The use of *E. hirae* as a probiotic for Jade Perch and other freshwater fish is promising as it is acclimatized to aquatic environments, safe and effective.