Genomics Guided Target Identification and Virtual Screening of Natural Compound Library to Propose Inhibitors Against *Shigella* Spp.

ABSTRACT

Thesis submitted for the Degree of

Doctor of Philosophy (Science)

In Biotechnology

By

Sarmishta Mukhopadhyay

Postgraduate and Research Department of Biotechnology

St. Xavier's College (Autonomous), Kolkata

Registration No. Ph.D./21/BMBT/01

Affiliated to the University of Calcutta

2024

Genomics Guided Target Identification and Virtual Screening of Natural Compound Library to Propose Inhibitors Against *Shigella* Spp.

ABSTRACT

Thesis submitted for the Degree of

Doctor of Philosophy (Science)

In Biotechnology

By

Sarmishta Mukhopadhyay

Postgraduate and Research Department of Biotechnology

St. Xavier's College (Autonomous), Kolkata

Registration No. Ph.D./21/BMBT/01

Affiliated to the University of Calcutta

2024

Abstract

Since the inception of civilization, diarrheal illnesses have detrimentally affected human well-being, and even today in the twenty-first century, they remain the second-most prevalent reason of morbidity and death among youngsters. The most commonly recognized etiologic agent for diarrheal mortality in children is the Gram-negative bacterium *Shigella*, which is accountable for 13.2% of diarrheal episodes across the globe. While *Shigella* was first identified in 1898, the manifestations of shigellosis remain evident today, a century after the disease was discovered. Within the Enterobacteriaceae family, *Shigella* is a facultative anaerobe that is gram-negative, non-flagellated, non-spore-forming, and non-lactose-fermenting. The disease is mostly spread via the faecal channel, which involves food, faeces, fingers, flies, and fomites as the main vectors, routes, and factors that trigger disease outbreak. Because *Shigella* only requires a minimal infectious load (about 10–100 organisms), the infection spreads effortlessly. The primary hallmark of shigellosis is acute ulcerative damage to the large intestine; with signs of fever, vomiting, cramping in the abdomen, diarrhea, and tenesmus. Symptoms emerge between 2 and 5 days after exposure with the bacteria. Sulfonamides had originally made fighting shigellosis easier, but to our disappointment, the efficacy of the remedy did not endure long and more than 80% of the isolates acquired sulfonamide resistance. This was followed by a quick emergence of *Shigella* isolates that were resilient to a broad range of medications, including tetracycline, fluoroquinolones, and nalidixic acid, and hence unresponsive to therapy. Currently, shigellosis is treated with third-generation cephalosporins (ceftriaxone) with varying degrees of success. The pressing need for new therapeutic alternatives to control the threat of shigellosis has been made explicit by the present trend in antibiotic resistance, the rise in non-typeable strains, and the uneven global distribution of different *Shigella* serotypes. In this regard, this work undertakes a comprehensive genomic analysis of

Shigella flexneri with the goal to identify promising and broad spectrum novel therapeutic targets in this bacterium and propose potential small molecule inhibitors of herbal origin against them. In the upstream part, the study involved the isolation and identification of a multidrug resistant *Shigella flexneri* strain from urban environmental sample. This was followed by a pangenome analysis to determine the core proteins which were eventually filtered by an efficient subtractive genomics approach to identify a set of *Shigella* specific proteins that do not exist in the corresponding host. After acquiring the likely targets, the next stage was to find inhibitors from natural herbal resources. Since the dawn of time, medicinal plants have been utilized in conventional therapies to heal illnesses in humans and animals. Nevertheless, substantial scientific investigation must be conducted due to the dearth of concrete evidence confirming the curative effect of these drugs against certain infectious diseases. Consequently, the latter part of this work focussed on evaluating the antimicrobial qualities of selected medicinal plants against *Shigella flexneri*. To rapidly and reliably screen natural compounds derived from plants for antibacterial agents, virtual screening, molecular docking, and molecular simulation were executed. 10 unique and widespread pharmacological targets were identified in the investigation, and 3 compounds were deemed as viable therapeutic candidates against the anticipated targets based on their affinity for binding and analysis of the ADMET attributes. The findings of this study corroborate the long-standing assertion of the ethnomedicinal literatures for the perpetual application of these plants for soothing infectious maladies. The prospective targets together with the small molecules proposed through this work, thus opens up the avenue to replace existing medications with more efficient natural therapeutics upon further in-vitro and in-vivo investigations, to effectively manage *Shigella* superbugs.

Keywords: *Shigella*, MDR, comparative genomics, drug targets, natural products.

Sarmishta Mukhopadhyay