ABSTRACT

Dengue is a vector-borne disease that lack vaccine or effective treatment, resulting in vector control being the primary disease control strategy. *Wolbachia*, an intracellular bacterium that can spread through vector population via cytoplasmic incompatibility (CI), has been shown to inhibit the transmission of a number of the deadly human pathogens such as dengue, Chikungunya viruses and filarial nematode in *mosquitoes*. In order to utilize *Wolbachia* to make mosquitoes resistant to the pathogens, a more efficient vector population replacement strategy required to block the transmission of pathogens to the humans completely and rapidly. The main objective of the current study was to evaluate the potential of *w*AlbB *Wolbachia* induced *Ae. aegypti* population to invade wild population with high frequency of infection through various release ratios. For this purpose three experimental (1,2,3) and two control (+ve, -ve) groups with constant number of wild males and females (1:1) were designed under semi-field conditions. In group 1-4 *Wolbachia* infected females (Wcl♀) were remained constant (20) while *Wolbachia* infected males (Wcl♂) varied as 20, 40, 80 & 0 with 1:1, 1:2, 1:4 & 1:0 ratios while negative control (5) did not have any *Wolbachia* infected male and female. After one week of 1st release, *Wolbachia* infected population was released again in 2nd release with fixed ratio (05 Wcl♀ constant X Wcl♂ 10, 20, 40) to boost the replacement strategy. Presence of *Wolbachia* was confirmed by PCR in each successive generations using *wsp Wolbachia* specific primers. The results indicated that the release of additional infected males (group 3) into the population can accelerate the population replacement by increasing the frequency of incompatible mating and results in decline of uninfected population. *Wolbachia* invasion was evaluated on three parameters; oviposition count, eggs hatch rate and *Wolbachia* induced population replacement frequency in successive generations. There was no significant difference ($p > 0.05$) in egg laying capacity of females of various experimental groups compared with control at 1-4 generations. However, significantly lower ($p \leq 0.05$) hatch rate (48%) was observed in release ratio 1:4 of *Wolbachia* infected females X males (group 3) at

generation 1. In addition highest average population replacement was also observed in group 3 and group 2 (100%± 0.00) at generation 3 (F3) and generation 4 (F4) respectively. There is a direct correlation between an increased ratios of infected males released with the increase rate of population replacement. In conclusion 100% *Wolbachia* infected population replacement occurred in group 3 with maximum *Wolbachia* infected males as rapid as within 3-4 months period. There is a dire need for further research on mass rearing of *Wolbachia* infected males to use in the field trials in specific localities for dengue vector population replacement and suppression in Pakistan to control the disease.