ABSTRACT

One of the rising interests in bacterial RNA research is due to the enormous discovery of non-protein-coding RNAs (ncRNAs) with diverse regulatory roles in fundamental cellular processes and adaptive responses to environmental changes/ stress conditions, thus affecting virulence. This work reports experimental analyses of ncRNA expression patterns in P. mirabilis, pathogenic bacteria responsible for urinary tract infections in individuals undergoing urinary catheterization. Despite the blooming data on bacterial ncRNAs, no studies have been reported on P. mirabilis ncRNAs. Thus, comparative analysis of known Gram-negative bacterial ncRNAs was performed against P. mirabilis genome. Totally, 53 from 166 of the Gram-negative bacterial ncRNAs have conserved in P. mirabilis with remarkable similarity at homologous regions. Out of these 53 predicted ncRNA candidates, 6 candidates were randomly selected. The selected candidates have highest homology (75-100%), highly conserved and having similarity percentage above 70%. These ncRNAs candidates have also been extensively studied in other organism. Their expression was verified by Northern blot analyses. Interestingly, all ncRNAs showed expression and with stage-specific regulation during P. mirabilis growth. Selected 6 candidates have shown either constitutive or differential expression during lag, log, and stationary phases of P. mirabilis indicating that the genes are active. PmR-11, PmR-34, PmR-42 and PmR-49 have shown common expression pattern by extensively expressing during lag and log phase while PmR-38 and PmR-35 were highly expressing during stationary phase. Anyhow, all the other 47 ncRNA candidates also need to be validated for their expression to confirm the presence of ncRNA genes. Both sequences and adjacent genes of these ncRNAs were conserved among

Gram-negative bacteria. Collectively, this study reveals the possible role of 6 ncRNAs in P. mirabilis during different growth phases. PmR-11 might bind to Tus mRNA in an antisense manner to regulate its expression, which may lead to the removal of its ter site in turn lead to the homologous recombination of bacterial genomes. PmR-34 might be differentially expressed according to the need of amino acids during the growth phases of P. mirabilis. PmR-35 could be playing a fundamental role in housekeeping, stationary phase stress adaptation and post transcriptional activities as the ncRNA expressed predominantly in stationary phase. PmR-38 might be influencing the activity of certain genes to regulate the bacterial replication processes as a part quorum sensing mechanism of *P. mirabilis*. PmR-42 might be involved in regulation of P. mirabilis replication processes as the ortholog, tmRNA, functions to discharge a stalled ribosome from the end of a incomplete mRNA. PmR-49 might regulate certain genes that includes in the repair of macromolecular damage that aggregated throughout stationary phase. This study also reveals the possible target mRNAs for these 6 ncRNAs which was predicted using RNApredator web server. The secondary structure for the 6 ncRNA candidates was predicted using RNAfold web server. The secondary structure prediction has shown that, there is clear signal of rho-independent terminator at 3'end for the Pmr-11 and PmR-34 ncRNA candidates whereas PmR-35, PmR-38, PmR-42, PmR-49 does not have rho independent termination. However we could not locate the possible promoter for these candidates. The potential in-vivo interactions between the ncRNAs and their target mRNAs could be determined, towards elucidation of the actual function of these ncRNAs.