6 Conclusions

- Bioinformatics analysis of the small RNA RsaF indicated the hyaluronate lyase (HysA) and serine protease like protein (SplD) as potential targets.
- RsaF functions as a positive regulator of hysA expression, since overexpression of *rsaF* increased *hysA* mRNA levels and enzymatic activity by 2-4 fold, which was confirmed by a corresponding decrease (0.1 < fold) in hysA expression in the disruption strain. The enhanced biofilm formation with tightly packed cells in presence of exogenous HA in the *rsaF* disruption and the reduced biofilm formation RsaF overexpression strain reiterates the role of RsaF as a positive regulator of HysA expression
- A functional RsaF was essential for the expression of SplD, as disruption of the *rsaF* decreased the *splD* mRNA levels to 0.005 fold and the total protease activity significantly. RsaF seemed to influence translation rate, since overexpression did not significantly change in *splD* mRNA levels but resulted in an increase in protease activity, as observed in zymography.
- The influence of RsaF on *hysA and splD* expression was mediated by RNA-RNA interaction between RsaF and target mRNAs
- Overexpression of RsaF demonstrated an increased stability of *hysA* and *splD* mRNAs.
- Overall, The findings demonstrate a positive regulatory role for small RNA RsaF in the expression of the virulence factors HysA and SplD.