Characterization of Rolling- Circle Plasmid Isolated from *Salmonella spp.*

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Small multicopy plasmids are frequently isolated from Enterobacteriaceae. In Salmonella, low molecular weight plasmids are found only in about 10% of Salmonella strains and their biological functions are largely unknown. However, many plasmids in Salmonella control important properties, such as, virulence factors, heavy metals and antibiotics resistance, phages or utilizations of alternative carbon sources. We have isolated a plasmid (pVCM1) from one strain of *Samonella enteretidis* isolated from broilers carcass. The strains were grown in liquid or solid Luria-Bertani broth at 37 °C. Plasmids were purified by QIAprep Miniprep kit (Qiagen GmbH, Germany) according to the manufacturer's instructions, separated on 1.0% agarose electrophoresis and visualized by ethidium bromide staining for analysis. Plasmids were digested with *Eco*RI enzyme and subcloned in the pUC18 plasmid. The plasmidal stability was evaluated, inoculating *E. coli* cells transformated with pVCM1 plasmid in liquid Luria-bertani broth supplemented with ampicillin. The total DNA sequence of plasmid pVCM1 has 1981 pb. Blast search resulted that pVCM1 showed 99% of identity with pB and 92% with pJ, which were isolated from *Salmonella enteretidis.* Only one ORF was founded in pVCM1. The protein encoded by this ORF showed extensive homology to Rep proteins of plasmids that replicates by rolling-circle mechanism. pVCM1 was stable after 240 generations. The *rep* gene was amplified and cloned in the pGEMT-easy vector. The 1981 pb pVCM1 plasmid is homologous to pB and pJ plasmids isolated from *Salmonella* detected in Czech Republic, possess only one ORF, what codified for a replicase protein.

Key-Words: plasmid, rollin-circle replication, *Salmonella*