



**Additional file 3. Molecular analysis of the *Ruminiclostridium cellulolyticum* mutant strains.**

**A.** Southern blot analysis of the strains. Genomic DNA or pMTLcbpA and pMTLcdpB were digested by PstI or EcoRI. After migration and transfert, the membrane was probed with a labeled probe targeting the erythromycine resistance cassette. The size of the detected fragments is consistent with theoretical sizes : MLTcbpA, 2.8 kb; MTLcdpA, 6.6 kb ; MTLcdpB, 4.2 kb ; MTLcdpC, 7.8 kb. **B.** PCR analysis of genomic DNA using primers hybridizing upstream and downstream the insertion site in the respectives target genes. Insertion of the intron increases the size by 1,78 kb in the mutant strains compared to the WT genomic DNA.