

Title of the publication

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Abbreviations: SAM, self-assembled monolayer; OTS, octadecyltrichlorosilane

Introduction

Studying the essentiality of genes helps with identifying the fundamental processes necessary for cell viability [1]. So far, scientists have studied the essential genes in organisms from different domains of life [2]. The results have led to new insights for developing new antibiotics that target essential genes of pathogenic bacteria [3, 4] and synthesising new genomes [5, 6]. Researchers have used different methods for studying the essentiality of genes in prokaryotes. Baba et al. [7] have made a library of single gene deletions using phage lambda Red recombination system to screen essential genes while another group have used antisense RNA knockdowns for this purpose [8]. Another method that is widely used due to its simplicity and accuracy is transposon mutagenesis along with high-throughput sequencing [9, 10, 11, 12, 13, 14, 15]. In this method, pools of single insertion mutants are constructed using transposon mutagenesis and the effect of each mutation on the survival of mutants is evaluated by sequencing the survivors [16]. This can lead to the identification of essential genes.

Although the essentiality of genes has been studied in a variety of organisms, there is still room to study the evolutionary conservation of essentiality. Barquist et al. [17] have used transposon-directed insertion-site sequencing to study the differentiation of the essentiality of genes in *Salmonella* serovars Typhi and Typhimurium which has led to divergence in their pathogenicity and host ranges. We extend this research by studying 12 bacterial strains. These include *Salmonella* enterica subsp. enterica serovar Typhi str. Ty2, *Salmonella* enterica subsp. enterica serovar Enteritidis str. P125109, *Salmonella* enterica subsp. enterica serovar Typhimurium str. SL1344, *Salmonella* enterica subsp. enterica serovar Typhimurium str. A130, *Salmonella* enterica subsp. enterica serovar Typhimurium str. D23580, *Escherichia* coli UPEC ST131, *Escherichia* coli ETEC CS17, *Escherichia* coli

ETEC H10407, *Citrobacter* rodentium ICC168, *Klebsiella* pneumoniae RH201207, *Klebsiella* pneumoniae subsp. pneumoniae Ecl8, and *Enterobacter* cloacae subsp. cloacae NCTC 9394. All these strains are selected from Enterobacteriaceae family.

Enterobacteriaceae is a family that includes bacteria with different host ranges and pathogenicity found in soil, water, plants, animals and humans [18]. In humans, various strains from this family can cause diarrhoea, septicaemia, urinary tract infection, meningitis, respiratory disease, and wound and burn infection [18]. Besides, they can infect poultry and livestock and cause financial losses for farmers [18]. Here, we perform a transposon-directed insertion-site sequencing experiment to study the conservation of essentiality of genes in strains from 5 different species in this family.

{A summary of what we have done}

Results

{An explanation of the experiment}

Clustering orthologous and paralogous genes. To study the essentiality of genes in 12 strains depicted in Fig , We needed to cluster sets of orthologous genes in these strain. Plenty of methods are proposed for this purpose. Altenhoff et al. have compared 15 of these methods [19] and shown that Hieranoid [20] is among three methods that keep a balance between precision and recall. We have used Hieranoid to cluster the sets of orthologous genes. In addition, we intended to study the essentiality of genes in paralogous genes. For this, we have developed a program that clusters all homologous genes using Jackhmmer from HMMER3 package [21].

Discussion

Materials and Methods

ACKNOWLEDGMENTS.

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Reserved for Publication Footnotes

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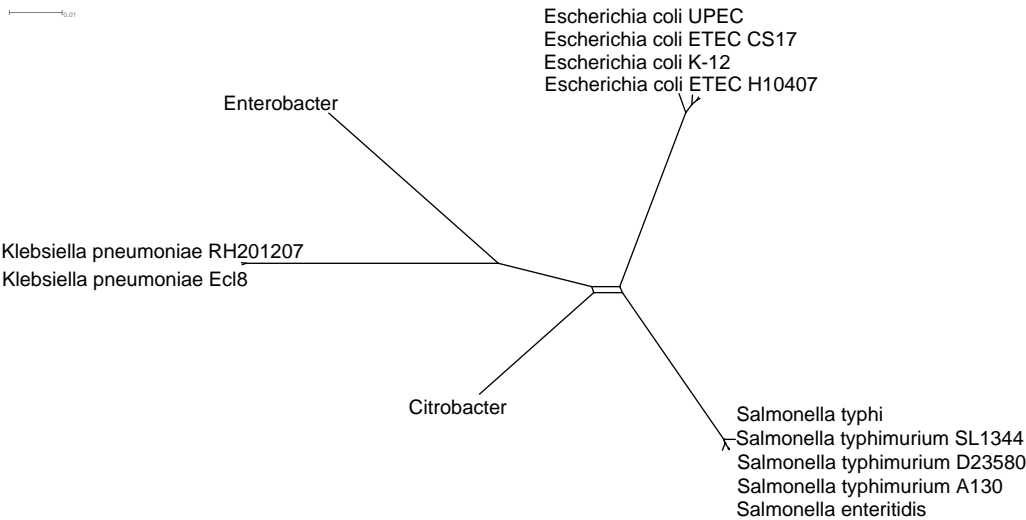


Fig. 1. Species tree. How is it made?