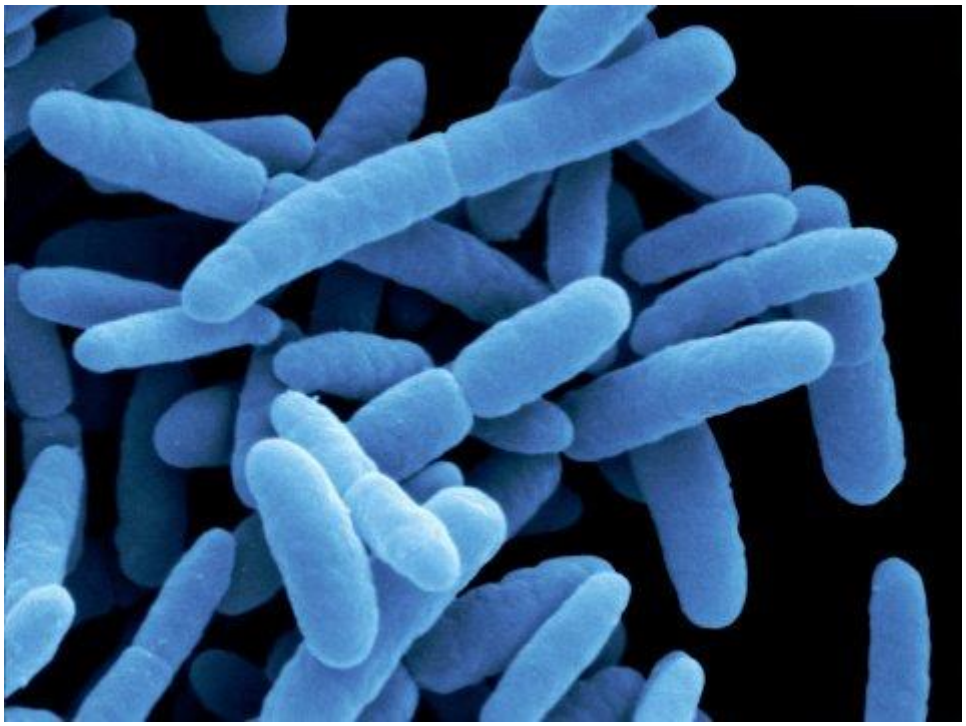
Creación de una red de regulación bacteriana de respuesta a antibióticos



Creation of a bacterial regulatory network of response to antibiotics

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Español

Este documento tiene como función explicar las redes bioinformáticas que se adjuntan en distintos archivos. Los mismos representan redes bioinformáticas de interacciones proteína-proteína en *Escherichia coli*, que reflejan las diferentes rutas bioquímicas que se ven afectadas por niveles sub-MIC de diferentes antibióticos.

En microbiología, se define a la concentración inhibitoria mínima o MIC (*Minimum inhibitory concentration)* como la concentración más baja de antibiótico que, en condiciones *in vitro* establecidas, inhibe el crecimiento poblacional visible de una bacteria objetivo. La naturaleza de las condiciones sub-MIC provocan la adaptación de las bacterias a medios seleccionados mediante mutaciones de bajo coste energético, además de regular la expresión génica y favorece las tasas de transferencia horizontal de genes [1].

Las proteínas y sus interacciones funcionales forman la columna vertebral de la maquinaria celular. Su red de conectividad debe considerarse para la plena comprensión de los fenómenos biológicos [2]. Esta red de conectividad está formada por interacciones proteína-proteína, las cuales se entienden comúnmente como contactos físicos con acoplamiento molecular entre proteínas que ocurren en una célula o en un organismo *in vivo*. Cualquier proteína en el ribosoma o en el aparato transcripcional basal comparte un contacto funcional con las otras proteínas en el complejo, pero ciertamente, no todas las proteínas en el complejo particular interactúan [3].

En este trabajo se han empleado dos programas bioinformáticos, por un lado, CellDesigner (v. 4.4.2) y, por el otro, Cytoscape (v. 3.7.2) cada uno con aplicaciones y características distintas. Para la obtención de la información bibliográfica de las interacciones proteína-proteína se ha utilizado la base de datos STRING (https://string-db.org/).

CellDesigner es una herramienta de modelado de redes de regulación genética y bioquímica. Este programa ayuda a los usuarios a crear fácilmente dichas redes, utilizando una representación gráfica integral y sólidamente definida (SBGN, *Systems Biology Graphical Notation*). CellDesigner es compatible con SBML (*Systems Biology Markup Language*) y tiene un software habilitado para Systems Biology Workbench

(SBW) para que pueda importar/exportar documentos descritos por SBML e integrarse con otros paquetes de software de simulación/análisis habilitados para SBW. La ventaja que posee CellDesigner es un editor de diagramas de procesos con tecnología estandarizada (SBML en este caso) para cada plataforma informática, de modo que pueda beneficiar a la mayor cantidad de usuarios posible [4].

Por su parte, Cytoscape, es un software de código abierto útil para integrar redes de interacción biomolecular con datos de expresión de alto rendimiento y otros estados moleculares en un marco conceptual unificado. Proporciona un entorno de modelado de propósito general para integrar redes y estados de interacción biomolecular [5]. Cytoscape viene provisto de una herramienta de análisis de redes, lo que permite obtener información detallada del funcionamiento, cohesión y estado de una red bioinformática.

STRING es una base de datos de interacciones proteína-proteína conocidas y previstas. Las interacciones incluyen asociaciones directas (físicas) e indirectas (funcionales); provienen de la predicción computacional, de la transferencia de conocimiento entre organismos y de interacciones agregadas de otras bases de datos (primarias) [2].

Debido a que STRING proporciona dos clases de interacciones proteína-proteína, relaciones conocidas y previstas, se han realizado dos redes distintas, por lo que una red contiene exclusivamente interacciones conocidas y otra red contiene relaciones, tanto conocidas como previstas.

La lista de antibióticos, sus concentraciones y genes es la siguiente:

Antibiótico	Concentración	Gen	Referencia
Ciprofloxacino	0.015-256 µg/ml	gyrA	[6]
		parC	
Norfloxacino	0.2-9.7 µg/ml	gyrB	[7]
		parE	
Omeprazol	100 µg/ml	acrA	[8]
		acrB	
		tolC	
		acrR	
Amikacina	0,016 mg/ml	aac(6')-Ib	[9]
		aac(3)-IIa	
		aph(3')-Ia	
		ant(2.)-Ia	
Ácido gálico	0.25 mg/ml	pgaA	[10]

		pgaB	
		pgaC	
		pgaD	
Ciprofloxacino	0.39-25 µg/ml	parC	[11]
Levofloxacino	0.78-25 µg/ml		
Ácido Nalidíxico	50- >200 µg/ml		
Esparfloxacino	0.39-12.5 µg/ml		
Telurito de potasio	0,001-0,8 mg/ml	terD	[12]
Ácido peptidonucleico	0.25-128 µg/ml	mcr-1	[13]
Azitromicina	0.256 -1.024 mg/ml	mph(A)	[14]
Ciprofloxacino	0.007 -128 µg/ml	gyrA	[15]
Ácido Nalidíxico	2-2000 µg/ml	gyrB	
Tetraciclina	0.0015-0.01 mg/ml	acrA	[16]
		acrB	
		tolC	
		soxS	
		rob	
		ompA	
		ompX	
		ompF	
		ompC	
		micF	
		marA	
Ciprofloxacino	2–64 µg/ml	qnrA	[17]
		qnrB	
Ofloxacino	8–64 µg/ml	aac(6')-Ib	
		qepA	
Ácido Nalidíxico	8–64 µg/ml	oqxA	
		oqxB	
Doxiciclina	0.375-192 µg/ml	RS17215	[18]
		RS17640	
		RS08025	
Ceftazidima	0.0625-256 µg /mll	ibpA	[19]
		ibpB	
		tnaA	
Quercetina	1-256 µg/ml	tetA	[20]
		tetB	
		tetM	
		tetS	
Piperacilina/tazobactam	0,032-0,256 mg/ml	TEM-1	[21]
		TEM-30	
		TEM-35	
		TEM-40	
		TEM-135	
Fosfomicina	50 µg/ml	uhpT	[22]
		uhpA	
Norfloxacino	0.094-256 µg/ml	gyrA	[23]
Ciprofloxacino	0.012-32 µg/ml	parC	
		parE	

Ácido Nalidíxico	1-256 µg/ml	marR	
		acrR	
Fluoxetina	0.0005-1 mg/ml	marR	[24]
		rob	
		sdiA	
		cytR	
		crp	
		acrB	
		acrD	
		ompF	
		ompW	
		yadG	
		yadH	
		acrA	
		acrB	
		tolC	
		tsx	
		mdlA	
		mdlB	
		mdfA	
		emrB	
		yciK	
		ampC	
		ampH	
		mrcA	
		mrdA	
		dacA	
		insB	
		insF	
		ymfD	
		yfjH	
		udp	
		cdd	
		tsx	
		mdtF	
		oppA	
		sodA	
		gor	
		ahpF	
		trxB	
		soxR	
		perR	

English

The purpose of this document is to explain the bioinformatics networks that are attached in different files. They represent bioinformatic networks of protein-protein interactions in *Escherichia coli*, which reflect the different biochemical pathways that are affected by sub-MIC levels of different antibiotics.

In microbiology, the Minimum Inhibitory Concentration (MIC) is defined as the lowest concentration of antibiotic that, under established *in vitro* conditions, inhibits the visible population growth of a target bacterium. The nature of sub-MIC conditions provoke the adaptation of bacteria to selected media through low energy cost mutations, in addition to regulating gene expression and favoring horizontal gene transfer rates [1].

Proteins and their functional interactions form the backbone of cellular machinery. Its network connectivity must be considered for the full understanding of biological phenomena [2]. This network of connectivity is formed by protein-protein interactions, which are commonly understood as physical contacts with molecular coupling between proteins that occur in a cell or in an organism in vivo. Any protein on the ribosome or basal transcriptional apparatus shares functional contact with the other proteins in the complex, but certainly not all proteins in the particular complex interact [3].

In this work, two bioinformatics programs have been used, on the one hand, CellDesigner (v. 4.4.2) and, on the other, Cytoscape (v. 3.7.2), each with different applications and characteristics. The STRING database (https://string-db.org/) has been used to obtain bibliographic information on protein-protein interactions.

CellDesigner is a biochemical and genetic regulatory network modeling tool. This program helps users to easily create such networks, using a robust and comprehensive graphical representation (SBGN, Systems Biology Graphical Notation). CellDesigner supports SBML (Systems Biology Markup Language) and has Systems Biology Workbench (SBW) enabled software so that it can import/export SBML-described documents and integrate with other SBW-enabled simulation/analysis software packages. The advantage of CellDesigner is a process diagram editor with standardized technology (SBML in this case) for each computing platform, so that it can benefit as many users as possible [4].

For its part, Cytoscape is open source software useful for integrating biomolecular interaction networks with high-throughput expression data and other molecular states in a unified conceptual framework. It provides a general-purpose modeling environment for integrating biomolecular interaction states and networks [5]. Cytoscape comes equipped with a network analysis tool, which allows obtaining detailed information on the functioning, cohesion and status of a bioinformatics network.

STRING is a database of known and predicted protein-protein interactions. Interactions include direct (physical) and indirect (functional) associations; they come from computational prediction, knowledge transfer between organisms, and aggregate interactions from other (primary) databases [2].

Because STRING provides two classes of protein-protein interactions, known and predicted relationships, two different networks have been realized, so one network contains exclusively known interactions and another network contains relationships, both known and predicted

The list of antibiotics, their concentrations and genes is as follows:

Antibiotic	Concentration	Gene	Reference
Ciprofloxacin	0.015-256 µg/ml	gyrA	[6]
		parC	
Norfloxacin	0.2-9.7 µg/ml	gyrB	[7]
		parE	
Omeprazole	100 µg/ml	acrA	[8]
		acrB	
		tolC	
		acrR	
Amikacin	0,016 mg/ml	aac(6')-Ib	[9]
		aac(3)-IIa	
		aph(3')-Ia	
		ant(2.)-Ia	
Gallic acid	0.25 mg/ml	pgaA	[10]
		pgaB	
		pgaC	
		pgaD	
Ciprofloxacin	0.39-25 µg/ml	parC	[11]
Levofloxacin	0.78-25 µg/ml		
Nalidixic acid	50- >200 µg/ml		
Sparfloxacin	0.39-12.5 µg/ml		
Potassium tellurite	0,001-0,8 mg/ml	terD	[12]

Peptidonucleic acid	0.25-128 µg/ml	mcr-1	[13]
Azithromycin	0.256 -1.024 mg/ml	mph(A)	[14]
Ciprofloxacin	0.007 -128 µg/ml	gyrA	[15]
Nalidixic acid	2-2000 µg/ml	gyrB	
Tetracycline	0.0015-0.01 mg/ml	acrA	[16]
		acrB	
		tolC	
		soxS	
		rob	
		ompA	
		ompX	
		ompF	
		ompC	
		micF	
		marA	
Ciprofloxacin	2–64 µg/ml	qnrA	[17]
		qnrB	
Ofloxacin	8–64 µg/ml	aac(6')-Ib	
		qepA	
Nalidixic acid	8–64 µg/ml	oqxA	
		oqxB	
Doxycycline	0.375-192 µg/ml	RS17215	[18]
		RS17640	
		RS08025	
Ceftazidime	0.0625-256 µg /mll	ibpA	[19]
		ibpB	
		tnaA	
Quercetin	1-256 µg/ml	tetA	[20]
		tetB	
		tetM	
		tetS	
Piperacillin/tazobactam	0,032-0,256 mg/ml	TEM-1	[21]
		TEM-30	
		TEM-35	
		TEM-40	
		TEM-135	
Fosfomycin	50 µg/ml	uhpT	[22]
		uhpA	
Norfloxacin	0.094-256 µg/ml	gyrA	[23]
Ciprofloxacin	0.012-32 µg/ml	parC	
		parE	
Nalidixic acid	1-256 µg/ml	marR	
		acrR	
Fluoxetine	0.0005-1 mg/ml	marR	[24]
		rob	
		sdiA	
		cytR	
		crp	
		acrB	

		acrD	
		ompF	
		ompW	
		yadG	
		yadH	
		acrA	
		acrB	
		tolC	
		tsx	
		mdlA	
		mdlB	
		mdfA	
		emrB	
		yciK	
		ampC	
		ampH	
		mrcA	
		mrdA	
		dacA	
		insB	
		insF	
		ymfD	
		yfjH	
		udp	
		cdd	
		tsx	
		mdtF	
		oppA	
		sodA	
		gor	
		ahpF	
		trxB	
		soxR	
		perR	

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