

# Characterization of Gene Families Encoding Beta-Lactamases of Gram-Negative Rods Isolated from Ready-to-Eat Vegetables in Mexico City.

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## **Resumen.**

Las bacterias resistentes a la betalactamasa, comunes en hospitales de tercer nivel, han surgido como un problema mundial de salud debido al consumo de vegetales listos para comer. Nuestro objetivo fue caracterizar los genes que producen resistencia a los antibióticos betalactámicos en enterobacterias, aisladas de cinco marcas de ensalada comercial para consumo humano en México. En total, se recolectaron veinticinco muestras, cultivadas en placas de agar sangre, las bacterias fueron identificadas bioquímicamente y se realizaron pruebas de susceptibilidad antimicrobiana. Los genes propios de la familia fueron identificados mediante PCR en punto final y se confirmaron los genes específicos con la secuencia completa de genomas (SCG) mediante secuencia de nueva

generación (SNG). Se identificaron doce cultivos positivos cuya distribución microbiológica fue la siguiente: 8.3% para *Enterobacter aerogene* ( $n = 1$ ), 8.3% para *Serratia fonticola* ( $n = 1$ ), 16.7% para *Serratia marcescens* ( $n = 2$ ), 16.7% para *Klebsiella pneumoniae* ( $n = 2$ ), and 50% ( $n = 6$ ) para *Enterobacter cloacae*. Los resultados del PCR en punto final mostraron 11 colonias positivas para blaBIL (91.7%), 11 para blaSHV (91.7%), 11 para blaCTX (97.7%), 12 para blaDHA (100%), cuatro para blaVIM (33.3%), dos para blaOXA (16.7%), dos para blaIMP (16.7%), una para blaKPC (8.3%), y una para el gen blaTEM (8.3%); todas la muestras fueron negativas para los genes blaROB, blaCMY, blaP, blaCFX y blaLAP. El análisis de secuencia reveló un genotipo específico para *Enterobacter cloacae* (blaSHV-12, blaCTX-M-15, blaDHA-1, blaKPC-2); *Serratia marcescens* (blaSHV-1, blaCTX-M-3, blaDHA-1, blaVIM-2); *Klebsiella pneumoniae* (blaSHV-12, blaCTX-M-15, blaDHA-1); *Serratia fonticola* (blaSHV-12, blaVIM-1, blaDHA-1); y *Enterobacter aerogene* (blaSHV-1, blaCTX-M-1, blaDHA-1, blaVIM-2, blaOXA-9). Nuestros resultados indican que las bacterias resistentes a la betalactamasa han adquirido integrones con diferente número de genes que proveen panresistencia a los antibióticos betalactámicos, incluyendo penicilinas, oxacilinas, cefalosporinas, monobactamas, carbapenems e imipenemas.

### **Abstract.**

Beta-lactam resistant bacteria, which are commonly resident in tertiary hospitals, have emerged as a worldwide health problem because of ready-to-eat vegetable intake. We aimed to characterize the genes that provide resistance to beta-lactam antibiotics in Enterobacteriaceae, isolated from five commercial salad brands for human consumption in Mexico City. In total, twenty-five samples were collected, grown in blood agar plates, and the bacteria were biochemistry identified and antimicrobial susceptibility testing was done. The carried family genes were identified by endpoint PCR and the specific genes were confirmed with whole genome sequencing (WGS) by Next Generation Sequencing (NGS). Twelve positive cultures were identified and their microbiological distribution was as follows: 8.3% for *Enterobacter aerogene* ( $n = 1$ ), 8.3% for *Serratia fonticola* ( $n = 1$ ), 16.7% for *Serratia marcesens* ( $n = 2$ ), 16.7% for *Klebsiella pneumoniae* ( $n = 2$ ), and 50% ( $n = 6$ ) for *Enterobacter cloacae*. The endpoint PCR results showed 11 colonies positive for blaBIL (91.7%), 11 for blaSHV (91.7%), 11 for blaCTX (97.7%), 12 for blaDHA (100%), four for blaVIM (33.3%), two for blaOXA (16.7%), two for blaIMP (16.7%), one for blaKPC (8.3%), and one for blaTEM (8.3%) gen; all samples were negative for blaROB, blaCMY, blaP, blaCFX and blaLAP gene. The sequencing analysis revealed a specific genotype for *Enterobacter cloacae* (blaSHV-12, blaCTX-M-15, blaDHA-1, blaKPC-2); *Serratia marcescens* (blaSHV-1, blaCTX-M-3, blaDHA-1, blaVIM-2); *Klebsiella pneumoniae* (blaSHV-12, blaCTX-M-15, blaDHA-1); *Serratia fonticola* (blaSHV-12, blaVIM-1, blaDHA-1); and, *Enterobacter aerogene* (blaSHV-1, blaCTX-M-1, blaDHA-1, blaVIM-2, blaOXA-9). Our results indicate that beta-lactam-resistant bacteria have acquired integrons with a different number of genes that provide pan-resistance to beta-lactam antibiotics, including

penicillins, oxacillins, cefalosporins, monobactams, carbapenems, and imipenems.

### **Bibliografía.**

Vázquez, R., Solano, S., León, B., Thompson, M., Guerrero, T., Gómez, E., Martínez, D., & González, J. (2018). Characterization of Gene Families Encoding Beta-Lactamases of Gram-Negative Rods Isolated from Ready-to-Eat Vegetables in Mexico City. *High-Throughput*, 7(4), 36. Disponible en [10.3390/ht7040036](https://doi.org/10.3390/ht7040036).