# Disoxaril Mutants of Coxsackievirus B1: Phenotypic Characteristics and Analysis of the Target VP1 Gene

Ivanka Nikolova[a], Angel S. Galabov[a,*], Rumena Petkova[b], Stoyan Chakarov[c], and Boris Atanasov[d]

[a] The Stephan Angeloff Institute of Microbiology, Bulgarian Academy of Sciences, 26, Georgi Bonchev Street, 1113 Sofia, Bulgaria. Fax: +359-2/870 0109. E-mail: galabov@microbio.bas.bg
[b] Scientific Technological Service, Ltd., Sofia, Bulgaria
[c] Department of Biochemistry, Faculty of Biology, Sofia University, Sofia, Bulgaria
[d] Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, Sofia, Bulgaria

* Author for correspondence and reprint requests

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Disoxaril inhibits enterovirus replication by binding to the hydrophobic pocket within the VP1 coat protein, thus stabilizing the virion and blocking its uncoating. Disoxaril-resistant (RES) mutants of the Coxsackievirus B1 (CVB1/RES) were derived from the wild disoxaril-sensitive (SOF) strain (CVB1/SOF) using a selection approach. A disoxaril-dependent (DEP) mutant (CVB1/DEP) was obtained following nine consecutive passages of the disoxaril-resistant mutant in the presence of disoxaril. Phenotypic characteristics of the disoxaril mutants were investigated. A timing-of-addition study of the CVB1/DEP replication demonstrated that in the absence of disoxaril the virus particle assembly stopped. VP1 RNA sequences of disoxaril mutants were compared with the existing Gen Bank CVB1 reference structure. The amino acid sequence of a large VP1 196–258 peptide (disoxaril-binding region) of CVB1/RES was significantly different from that of the CVB1/SOF. Crucially important changes in CVB1/RES were two point mutations, M213H and F237L, both in the ligand-binding pocket. The sequence analysis of the CVB1/DEP showed some reversion to CVB1/SOF. The amino acid sequences of the three VP1 proteins are presented.

*Key words:* Coxsackievirus B1, Disoxaril Mutants, VP1 Amino Acid Sequence