# Rapid bench-side detection of carbapenemase genes in Gram negative bacteria by LAMP assay

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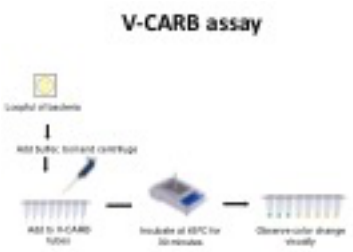
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Understanding genetic mechanisms of carbapenem resistance among Gram-negative bacteria (GNB) could help tailor antibiotic therapy and lead to improved outcomes, particularly for critically ill patients. We developed a novel, low cost, bench side rapid molecular assay, V-CARB, based on the Loop mediated isothermal amplification (LAMP) technology that simultaneously identifies the 4 major carbapenemase genes: bla_NDM, bla_OXA-48, bla_OXA-23 and bla_VIM. The assay takes one hour and gives a clear visual readout. 188 clinical GNB isolates, were screened by the V-CARB assay for bla_NDM, bla_OXA-48,bla_OXA-23and bla_VIM genes and by the Kirby Bauer's Disc diffusion susceptibility test (DDST). For isolates with mismatched results, the Carbapenem Inhibition assay (CIM) was undertaken.

41/188 (21.8%) isolates were positive for at least one gene by the V-CARB assay. Of these, 14 were detected as susceptible by DDST. 8/14 were true positives and 6/14 were false positives. Further, 5 isolates detected as resistant by DDST were not identified by the V-CARB assay. Of these, 2 expressed unidentified carbapenemases, 1 was carbapenemase-independent and 2 were false negatives. With DDST as a gold standard, the LAMP assay had a sensitivity of 95%, negative predictive value of 98.61%, specificity of 95.95% and positive predictive value of 86.36%.

The different isolates tested included *Acinetobacter* (12), *E. coli* (108), *Klebsiella* (52) and *Pseudomonas* (16). The LAMP assay detected 9 NDM, 2 VIM, 16 OXA- 23 and 21 OXA-48 positive isolates. NDM was found across the species, except *Pseudomonas*. OXA-48 was more common among *Klebsiella pneumoniae* while OXA-23was found exclusively in Acinetobacter. VIM was detected in *Pseudomonas* and *Klebsiella* isolates. The V-CARB assay for carbapenemase genes developed by our team is a highly sensitive, specific, rapid and simple bench-side assay. The availability of the results in a clinically useful time will help clinicians select an appropriate antibiotic at the earliest.

**Image:**




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