NG·BIOTECH LABORATOIRES

NG-Test® CARBA-5 Literature

Boutal, Hervé *et al*. "A multiplex lateral flow immunoassay for the rapid identification of NDM-, KPC-, IMP- and VIM-type and OXA-48-like carbapenemase-producing Enterobacteriaceae." *The Journal of antimicrobial chemotherapy* vol. 73,4 (2018): 909-915. doi:10.1093/jac/dkx521

Objectives: The global spread of carbapenemase-producing Enterobacteriaceae represents a substantial challenge in clinical practice and rapid and reliable detection of these organisms is essential. The aim of this study was to develop and validate a lateral flow immunoassay (Carba5) for the detection of the five main carbapenemases (KPC-, NDM-, VIM- and IMP-type and OXA-48-like). **Methods**: Carba5 was retrospectively and prospectively evaluated using 296 enterobacterial isolates from agar culture. An isolated colony was suspended in extraction buffer and then loaded on the manufactured Carba5. **Results**: All 185 isolates expressing a carbapenemase related to one of the Carba5 targets were correctly and unambiguously detected in <15 min. All other isolates gave negative results except those producing OXA-163 and OXA-405, which are considered low-activity carbapenemases. No cross-reaction was observed with non-targeted carbapenemases, ESBLs, AmpCs or oxacillinases (OXA-1, -2, -9 and -10). Overall, this assay reached 100% sensitivity and 95.3% (retrospectively) to 100% (prospectively) specificity. **Conclusions**: Carba5 is efficient, rapid and easy to implement in the routine workflow of a clinical microbiology laboratory for confirmation of the five main carbapenemases encountered in Enterobacteriaceae.

Volland, Hervé *et al*. "Improvement of the Immunochromatographic NG-Test Carba 5 Assay for the Detection of IMP Variants Previously Undetected." *Antimicrobial agents and chemotherapy* vol. 64,1 e01940-19. 20 Dec. 2019, doi:10.1128/AAC.01940-19

Abstract Here, we evaluated the immunochromatographic assay NG-Test Carba 5v2 (NG-Biotech), with improved IMP variant detection on 31 IMP producers, representing the different branches of the IMP phylogeny, including 32 OXA-48, 19 KPC, 12 VIM, 14 NDM, and 13 multiple carbapenemase producers (CPs), 13 CPs that were not targeted, and 13 carbapenemase-negative isolates. All tested IMP variants were accurately detected without impairing detection of the other carbapenemases. Thus, NG-Test Carba 5v2 is now well adapted to countries with high IMP prevalence and to the epidemiology of CP-Pseudomonas aeruginosa, where IMPs are most frequently detected.

Yoon, Jung *et al*. "Application of a multiplex immunochromatographic assay for rapid identification of carbapenemases in a clinical microbiology laboratory: performance and turn-around-time evaluation of NG-test Carba 5." *BMC microbiology* vol. 21,1 260. 29 Sep. 2021, doi:10.1186/s12866-021-02309-9

Background: Prompt and accurate identification of carbapenemase production is essential for appropriate treatment and infection control. NG-Test Carba 5 (termed herein "Carba 5"; NG Biotech, Guipry, France) is a multiplex immunochromatographic assay for the rapid phenotypic identification of five major carbapenemases (KPC, NDM, VIM, IMP, and OXA-48-like) from bacterial isolates. This study aimed to evaluate the diagnostic performance of Carba 5 and its impact on the turn-around-time in a clinical microbiology laboratory. **Results**: Carba 5 was retrospectively evaluated using 78 carbapenemase producers and 23 non-carbapenemase producers confirmed by PCR and sequencing. The performance and time required for carbapenemase identification were prospectively evaluated using 47 carbapenem resistant Enterobacteriaceae isolates, and the results were compared to those obtained using Xpert Carba-R (Cepheid, Sunnyvale, CA, USA). For the bacterial isolates included in retrospective and prospective evaluation, the Carba 5 assay correctly identified 147 isolates except one isolate with a sensitivity of 99.13% (95% CI 95.25-99.98%) and specificity of 100% (95% CI 89.42-100%). The Carba 5 assay missed one VIM-1 among 13 VIM producers. The assay showed a sensitivity of 92.31% (95% CI 63.97-99.81%) for detecting VIM and 100% for detecting KPC, NDM, OXA-48-like, and IMP. Compared to the Xpert Carba-R assay, Carba 5 exhibited 100% agreement and was more time-efficient (median time 24 min vs. 1 h 11 min). **Conclusions**: The Carba 5 assay has potential as an alternative to molecular methods for detecting major carbapenemases from bacterial isolates in a clinical microbiology laboratory. Compared to the Xpert Carba-R, Carba 5 turns out to be more affordable and time-efficient while showing a comparable performance, and may accelerate therapeutic and infection control decisions.

Khalifa, Hazim O *et al*. "Comparative Evaluation of Five Assays for Detection of Carbapenemases with a Proposed Scheme for Their Precise Application." *The Journal of molecular diagnostics : JMD* vol. 22,9 (2020): 1129-1138. doi:10.1016/j.jmoldx.2020.05.012

Abstract The escalating problem of the dissemination of carbapenemase-producing bacteria (CPB) has gained worldwide attention. The prompt diagnosis of CPB and precise identification of carbapenemases are imperative to enable specific antibiotic therapy and control the spread of these bacteria. The present study was designed to assess the performance of five important assays for the detection of carbapenemases. The modified carbapenem inactivation method (**mCIM**), **CARBA-5, GeneXpert Carba-R, BD MAX Check-Points CPO**, and **GeneFields CPE** assays were evaluated with an international collection of 159 bacterial isolates, including 93 CPB and 66 non-CPB isolates. The overall accuracy/sensitivity/specificity for carbapenemase detection were 100% (95% CI, 97.7%-100%)/100% (95% CI, 96.1%-100%)/100% (95% CI, 94.6%-100%) for mCIM, 98.7% (95% CI, 95.5%-99.9%)/97.9% (95% CI, 92.5%-99.7%)/100% (95% CI, 94.6%-100%) for CARBA-5, 96.9% (95% CI, 92.8%-99%)/95.7% (95% CI, 89.4%-98.8%)/98.5% (95% CI, 91.8%-99.9%) for GeneXpert Carba-R, 94.3% (95% CI, 89.5%-97.4%)/90.3% (95% CI, 82.4%-95.5%)/100% (95% CI, 94.6%-100%) for BD MAX Check-Points CPO, and 86.2% (95% CI, 79.8%-91.1%)/77.4% (95% CI, 67.6%-85.5%)/98.5% (95% CI, 91.8%-100%) for GeneFields CPE. Interestingly, mCIM and CARBA-5 assays showed 100% accuracy/sensitivity/specificity for detection of the target genes. Furthermore, all the other assays showed comparable high accuracy (96.9% to 100%), sensitivity (100%), and specificity (96.4% to 100%) for the detection of the target genes. On the basis of these results, a new scheme was proposed for their efficient application. These results confirmed the high sensitivity of the evaluated assays, and the proposed scheme is reliable and improves the overall sensitivity and specificity of the assays.

Kanahashi, Toru *et al*. "Comparison of the Xpert Carba-R and NG-Test CARBA5 for the detection of carbapenemases in an IMP-type carbapenemase endemic region in Japan." *Journal of infection and chemotherapy : official journal of the Japan Society of Chemotherapy* vol. 27,3 (2021): 503-506. doi:10.1016/j.jiac.2020.11.001

Introduction: The real-time PCR assay Xpert Carba-R and the lateral flow immunoassay NG-Test CARBA5 were developed to detect 5 types of carbapenemase genes (blaIMP, blaKPC, blaVIM, blaOXA-48, and blaNDM). **Methods**: We compared the diagnostic performance, turn-around time, and cost of these assays. Carbapenemase genes were defined using the Carba NP test, modified Carbapenem Inactivation Methods (mCIM), multiplex PCR, and whole-genome sequencing. We included clinical Enterobacterales isolates (n = 36) and nonfermenting gram-negative bacilli isolates (n = 17) collected from 16 acute-care hospitals in the Kinki region of Japan. **Results**: Twenty-six of these 53 isolates were positive according to both of the Carba NP test and mCIM and, contained the following carbapenemase genes: blaIMP-1 (n = 3), blaIMP-6 (n = 1), blaIMP-19 (n = 12), blaIMP-26 (n = 1), blaIMP-41 (n = 2), blaIMP-66 (n = 2), blaNDM-1 (n = 3), and blaVIM-2 (n = 2). All of the remaining 27 isolates were negative according to the Carba NP test, mCIM, and multiplex PCR. The specificities of both assays were 100%. The sensitivity of the Xpert Carba-R assay was as low as 53.8% and that of the NG-Test CARBA5 was 92.3% because the former failed to detect all isolates with blaIMP-19 (n = 12) and the latter failed to detect isolates with blaIMP-66 (n = 2). Both assays can easily be performed in less than 5 min. **Conclusions**: The NG-Test CARBA5 assay was superior with regard to assay time and cost per sample. We propose the use of the NG-Test CARBA5 assay in clinical laboratories where IMP-type carbapenemases are endemic.

Takissian, Julie *et al*. "NG-Test Carba 5 for Rapid Detection of Carbapenemase-Producing Enterobacterales from Positive Blood Cultures." *Antimicrobial agents and chemotherapy* vol. 63,5 e00011-19. 25 Apr. 2019 doi:10.1128/AAC.00011-19

Abstract The immunochromatographic assay, NG-test Carba 5 (NG Biotech), has been evaluated for detection of carbapenemase-producing *Enterobacterales* (CPE) from spiked blood cultures (*n*_205). It detected and discriminated in less than 30 minutes KPC, IMP, VIM, NDM, and OXA-48-like producers with a sensitivity and specificity of 97.7% and 96.1%, respectively. Thus, it might help the rapid optimization of treatment of bloodstream infections due to CPE.

Boattini, Matteo *et al*. "Fast-track identification of CTX-M-extended-spectrum-β-lactamase- and carbapenemase-producing Enterobacterales in bloodstream infections: implications on the likelihood of deduction of antibiotic susceptibility in emergency and internal medicine departments." *European journal of clinical microbiology & infectious diseases: official publication of the European Society of Clinical Microbiology* vol. 40,7 (2021): 1495-1501. doi:10.1007/s10096-021-04192-8

Abstract - This study aims at presenting a reliable fast-track diagnostics for the detection of CTX-M ESBL- (CTX-M-p) and carbapenemase-producers (CA-p) directly from blood cultures (BCs) of patients with Enterobacterales (EB) bloodstream infections (BSIs) admitted in emergency and internal medicine departments and its contribution in estimation of in vitro antibiotic susceptibility. A fast-track workflow including MALDI-TOF species identification and two lateral flow immunochromatographic assays for the detection of CTX-M-p and CA-p directly from BCs was performed in parallel with conventional routine, and results were compared. A total of 236 BCs of patients suffering from EB BSI were included. Accuracy of the fast-track workflow ranged from 99.6 to 100%. Among E. coli isolates, CTX-M-p (20.5%) were susceptible to ceftolozane-tazobactam (C/T, 97%), ceftazidime-avibactam (CZA, 100%), and piperacillin-tazobactam (TZP, 84.8%), whereas CTX-M-and-main-carbapenemases-non-producer (CTX-M-CA-np, 79.5%) isolates were susceptible to all the antibiotics tested. Among K. pneumoniae isolates, CTX-M-p (23.3%) were poorly susceptible to TZP (40%) but widely susceptible to C/T (90%), CZA (100%), and amikacin (90%), whereas CTX-M-CA-np (55.8%) were also susceptible to cefepime. CA-p K. pneumoniae (20.9%) were susceptible to CZA (88.9%). All the species other than E. coli and K. pneumoniae were CTX-M-CA-np and were widely susceptible to the antibiotics tested except for isolates of the inducible and derepressed AmpC- or AmpC/ESBL-p species. Rapid identification of species and phenotype together with knowledge of local epidemiology may be crucial to determine the likelihood of deduction of in vitro antibiotic susceptibility on the same day of positive BC processing.

Vasilakopoulou, Alexandra *et al*. "Detection of KPC, NDM and VIM-Producing Organisms Directly from Rectal Swabs by a Multiplex Lateral Flow Immunoassay." *Microorganisms* vol. 9,5 942. 27 Apr. 2021, doi:10.3390/microorganisms9050942

Abstract - We report a preliminary evaluation of the NG-Test CARBA 5 immunochromatographic assay for detecting carbapenemases directly from rectal swabs on the same day of sampling. Thirty fecal swabs were examined for carbapenemase-producing organisms (CPOs) by conventional culture, PCR, and NG-Test CARBA 5. Each sample was tested by the immunochromatographic assay five times, including direct testing and incubation in trypticase soy broth for 1, 2, 3, and 4 h. Twenty patients yielded CPOs by culture. Immunochromatographic and PCR results were concordant and detected the same 25 carbapenemases (11 KPC, 8 VIM, and 6 NDM). In five cases, we detected co-carriage of KPC and VIM. Compared with PCR, the sensitivity of NG-Test CARBA 5 for the detection of KPC, VIM, and NDM was 80% without incubation, 88% with one hour, 92% with two, and 100% with three hours incubation, while specificity was 100% for all time points. All samples containing adequate fecal content were detected by NG-Test CARBA 5 concordantly with PCR, without incubation. NG-Test CARBA 5 is a reliable test that rapidly detects the presence of carbapenemases at the same day of sampling, directly from rectal swabs. It thus provides early information to guide antimicrobial treatment and infection control interventions.

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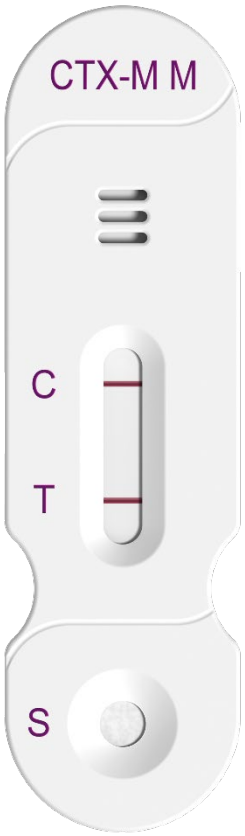
NG-Test® CTX-M MULTI Literature



Bernabeu, Sandrine *et al*. "A Lateral Flow Immunoassay for the Rapid Identification of CTX-M-Producing Enterobacterales from Culture Plates and Positive Blood Cultures." *Diagnostics (Basel, Switzerland)* vol. 10,10 764. 28 Sep. 2020, doi:10.3390/diagnostics10100764

Abstract We have developed a lateral flow immunoassay (LFIA), named NG-Test CTX-M MULTI (NG-Test), to detect group 1, 2, 8, 9, 25 CTX-M producers from agar plates and from positive blood cultures in less than 15 min. The NG-Test was validated retrospectively on 113 well-characterized enterobacterial isolates, prospectively on 102 consecutively isolated ESBL-producers from the Bicêtre hospital and on 100 consecutive blood cultures positive with a gram-negative bacilli (GNB). The NG-Test was able to detect all CTX-M producers grown on the different agar plates used in clinical microbiology laboratories. No false positive nor negative results were observed. Among the 102 consecutive ESBL isolates, three hyper mucous isolates showed an incorrect migration leading to invalid results (no control band). Using an adapted protocol, the results could be validated. The NG-Test detected 99/102 ESBLs as being CTX-Ms. Three SHV producers were not detected. Among the 100 positive blood cultures with GNB tested 10/11 ESBL-producers were detected (8 CTX-M-15, 2 CTX-M-27). One SHV-2-producing-E. cloacae was missed. The NG-Test CTX-M MULTI showed 100% sensitivity and specificity with isolates cultured on agar plates and was able to detect 98% of the ESBL-producers identified in our clinical setting either from colonies or from positive blood cultures.

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