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Letter to the Editor

Detection of extended-spectrum beta-lactamase production in Enterobacteriales from patients with suspected urinary tract infections, Tabora region, Rural Tanzania

To the Editor,

Antibiotic resistance (ABR) is an increasing challenge in the treatment of infectious diseases (1). The effects of ABR can be seen as prolonged hospital stay, increased mortality as well as higher health care expenditure (2). The burden of ABR seems to fall mainly on low- and middle-income countries, for example, in Sub-Saharan African region (3). In urban Tanzania, ESBLproducing *E. coli* and *Klebsiella spp*. has been reported to be 39% and 51%, respectively, in patients with urinary tract infections (UTI) (4). Still, the deficit in surveillance systems fails to mount the true dimensions of ABR, especially in rural areas (5). Reliable susceptibility testing needs thoroughly standardized laboratory procedures and well-equipped and -staffed laboratories, something which is uncommon in rural parts of Sub-Saharan Africa (6). The recently introduced β LACTA® test (Bio-Rad Laboratories Inc., Hercules, California, USA) allows quick detection of β-lactamases in clinical isolates and facilitates ESBL surveillance. The cleaved chromogenic component (HMRZ-86) is readily hydrolysed by β-lactamases of ESBL (e.g. CTX-M type, SHV-5 and -18) and carbapenemase type (e.g. Metallo-β-lactamases of IMP-type) and to some extent by hyper production of AmpC-lactamases (e.g. FOX-3, ACC-1) (7, 8). This study investigated the presence of β LACTA® test-positive Enterobacteriales in urinary samples collected in a rural Tanzanian setting at Nkinga Referral Hospital (NRH), Tabora region. We also aimed to assess the utility of the β LACTA® test as a tool to improve surveillance of ABR in a low-resource laboratory.

The Nkinga Referral Hospital Laboratory (NRHL) performs staining techniques for microorganism identification and, to a limited extent, culture and antibiotic susceptibility tests using in-house prepared antibiotic discs (Fig. 1). Urine samples sent as a part of standard care from patients at NRH with a suspicion of UTI during February to June 2016 was included. The β LACTA® test was performed besides standard examination by DN and JM and the results were blinded and not reported on patient level. All cultures during the time period were granted by the project. The culture of urine samples was performed on cystine-lactose-electrolyte deficient (CLED) agar (9). Colony appearance on CLED agar and Gram stain was used to discriminate Enterobacteriaceae spp. The β LACTA® test (BLT) (Bio-Rad Laboratories Inc., Hercules, California, USA) was performed according to the manufacturer's instructions (10). A negative control, using reagents only, was performed in parallel. The study was approved by the NRH board on October 2015.

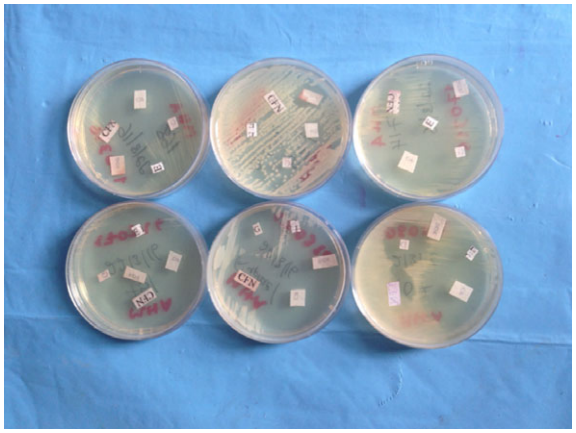

Fig. 1. Routinely performed antibiotic susceptibility test at Nkinga Referral Hospital.

In total, 95 urine samples were received at the laboratory from both inpatients (n = 23) and outpatients (n = 72), see Table 1. Fifty-eight cultures were positive with 7 *Pseudomonas* spp (characteristic appearance on CLED agar) and 51 Enterobacteriaceae spp. β LACTA® test was performed on the 51 isolates characterized as non-pseudomonal Gram-negative rods, of which 19 (37%) were positive. As previous studies have shown, β LACTA® test positivity is mainly caused by ESBL (10) indicating a high level of ESBL-producing bacteria in this area. To the best of our knowledge, this study is the first attempt to predict the ESBL prevalence in rural Tanzania; yet, the results are not surprising as high resistance rates in *E. coli* and *Klebsiella spp*. has been reported for commonly used antibiotics such as ampicillin (68–99%), co-trimoxazole (63–83%), ceftriaxone (21–66%), ciprofloxacin (19–47%) and nitrofurantoin (14–23%) in Tanzania and other parts of eastern Africa (4, 11, 12). Faecal carriage of ESBL-producing

Work was carried out at the Department of Laboratory Medicine, Nkinga Referral Hospital Laboratory, Po. Box 1, Nkinga, Tabora, Tanzania East Africa.

Table 1. Characteristics of collected urine samples

	No.	%
Urine samples (n = 95)		
Gender		
Male	43	45
Female	52	55
Inpatients	23	24
Surgical ward	6	6
Medical ward	11	12
Children's ward	6	6
Outpatients	72	76
Positive Cultures (n = 58)		
Growth amount		
Significant (>10^5 CFU/mL)	40	69
Non-significant (<10^5 CFU/mL)	18	31
No. Species		
Homogenous	45	78
Mixed growth	13	22
Colony appearance on CLED agar		
Yellow colonies	40	69
Non-yellow colonies	11	19
Pseudomonas appearance (Excluded for β LACTA testing)	7	12

Enterobacteriales has also been reported in up to 34% of healthy individuals (13) and high rates of antibiotic-resistant *E. coli* have been found in drinking water sources in Tanzania (14). The number of bacterial isolates was, however, low in this study and the bacteria were not identified to species level. Even though the cost for laboratory analyses was granted by the project, we most likely had selection biases in collection of urine samples. Introduction of the β LACTA® test for ESBL screening could have potential benefits. The test is fast, easy to use and could help to guide treatment as well as preventive actions for spread of resistant organisms in the hospital. Still, it is too expensive for the setting studied (5 USD/test during the study period), especially as the cost would fall on a poor patient group (most diagnostic procedures are paid for by the patient) and the test does not give information on susceptibility of other antibiotics. This study indicates that multidrug resistant Enterobacteriales are common at Nkinga Referral Hospital. New easily handled diagnostic tools like the β LACTA® test could potentially benefit antibiotic resistance surveillance in rural areas provided that the cost of the tests is reduced.

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CONFLICT OF INTEREST

The authors report no conflict of interest.

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