TABLE 1. Classifications of Observed Hand Disinfection Tests and Group-Specific Results[a]

A. Hand Hygiene Demonstrations					
Variable	Test 1	Test 2	Test 3	Test 4	Test 5
Time, s	32	12	29	31	32
Coverage, % of skin area	100	50	90	50	60
Classification	Correct	Incorrect	Correct	Incorrect	Incorrect
B. Participant Assessments					
Group	Correct Classifications	Incorrect Classifications			
Live	40	1			
Video	33	7			

[a]Total observations, n = 81.

The addition of an easy-to-use qualitative component to hand hygiene compliance observations and consecutive training efforts is important, given that <10% of all hand disinfections were performed correctly in an observational study by Tschudin-Sutter et al,[1] who observed the 6-step technique. Appropriate hand-surface coverage was reached in only 7.9% of hand hygiene procedures observed by Park et al,[2] despite a high rate of compliance with the correct indications. Shah et al[3] performed a video observation of hand washing. Of 1,081 recordings, 403 (37.3%) were excellent, 521 (48.2%) were acceptable, and 157 (14.5%) were unacceptable.

A limitation of our study is the lack of bacterial counts, but the results of Riley et al,[4] who showed no correlation between hand coverage and bacterial counts with a 6-step technique compared to a 3-step approach, had not been published at the time of our experiment.[4] Another limitation is the small number of participants and the experimental setting of this proof-of-principle study. However, we believe that based on our results, the addition of dichotomous subjective quality assessment using the parameters time and skin coverage during live observation by experienced infection control staff is feasible and could be a valuable addition to conventional hand hygiene observation.

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Emergence of OXA-72-producing *Acinetobacter baumannii* Belonging to High-Risk Clones (CC15 and CC79) in Different Brazilian States

To the Editor—Carbapenem resistance limits treatment options and causes major therapeutic problems; it has been continuously reported worldwide among *Acinetobacter baumannii* isolates. Carbapenem resistance in *A. baumannii* is frequently associated with Ambler class D carbapenemase, mainly *bla*OXA-23. Until now, there have been only a few reports of other oxacilinases, such as *bla*OXA-72, in Brazil.[1] Multilocus sequence typing (MLST) seems to be a reliable tool for investigating population structure and global *A. baumannii* epidemiology. In Brazil, most carbapenem-resistant *bla*OXA-23–producing *A. baumannii* have been associated with clonal complexes CC79 and CC15.[2] To the best of our knowledge, our report here is the first report of the epidemic clonal complex CC15 associated with *A. baumannii* carrying *bla*OXA-72. Furthermore, we describe the spread of

A. baumannii high-risk clones (CC79 and CC15) carrying *bla*OXA-72 gene in 3 Brazilian states.

As part of a surveillance study performed by our research group (Laboratório de Pesquisa em Resistência Bacteriana – LABRESIS), we evaluated 94 *A. baumannii-calcoaceticus* isolates from 4 Brazilian states (São Paulo, Rio de Janeiro, Rio Grande do Sul, and Paraná) identified between April and October 2013. The isolates were screened for oxacilinases genes (ie, OXA-23, OXA-24/40, OXA-51, OXA-58, and OXA-143) by polymerase chain reaction (PCR). Class 1 and 2 integrons were detected by PCR for the integrase gene.[3] Among these 94 isolates from 3 states (São Paulo, Paraná and Rio Grande do Sul), 11 isolates (11.7%) presented positive results for the *bla*OXA-24/40, which was identified by sequencing as the variant *bla*OXA-72 (ABI 3500 Genetic Analyzer). Species identification was performed using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF) and a *gyrB* multiplex PCR, and all *bla*OXA-72–producing isolates were identified as *A. baumannii*.

Clonal diversity of the *A. baumannii* OXA-72 producers was investigated using repetitive-sequence-based PCR (REP-PCR).[4] Results were interpreted according to a dendrogram constructed using BioNumerics v6.5 software (bioMèrieux, Marcy-l'Étoile, France). REP-PCR analysis showed the presence of 3 clonal groups (A, B, and C; Table 1).

The OXA-72 *A. baumannii* producers were typed using the MLST scheme from the Pasteur Institute (http://www.pasteur.fr). Analyses of the allele sequences and sequence type (ST) were performed via the *A. baumannii* MLST Institute Pasteur website (http://pubmlst.org/abaumannii/). The relationships among the new and existing STs were evaluated using the eBURST program to determinate the clonal relation of *A. baumannii* carrying *bla*OXA-72 among different states. MLST demonstrated that the isolates belonged to 5 different sequence types: ST79, ST180, ST730, and the new ST890 and ST891 (Table 1). According to the MLST data analysis and the eBURST algorithm, the isolates belonged to the epidemic clonal complexes CC15 (ST180 and ST890) and CC79 (ST79 and ST730), and the ST891 was a singleton. CC15 was restricted to the state of Paraná, and CC79 was present in Rio Grande do Sul and São Paulo states. Class 1 integrons were detected in all isolates belonging to CC15, and all isolates of CC79 presented class 2 integrons (Table 1).

Acinetobacter baumannii is characterized by a remarkable capability to acquire antibiotic resistance determinants, intra- and interhospital outbreaks, and national and international clonal dissemination. Worldwide dissemination of multidrug-resistant *A. baumannii* OXA-23–producers has been associated with specific clones, such as international clone 1 ($CC1^{IP}$), international clone 2 ($CC2^{IP}$), and $CC15^{IP}$.[5] In Brazil, the extensive dissemination of CC79 and CC15 have been associated with *bla*OXA-23 production in different states.[1,2] Furthermore, Stietz et al[6] reported that ST79 is also frequently observed among OXA-23–producing *A. baumannii* isolates in Argentina, which demonstrates the dispersal of this CC79 not only in Brazil but also in South America countries.[6] The same study demonstrated the spread of *bla*OXA-72 related to the epidemic clone CC79.[6]

TABLE 1. Molecular Typing Analysis of *bla*OXA-72–Producing *A. baumannii* in Different Brazilian States

Isolate	Location	REP-PCR	ST[a]	CC[b]	*Int*
15POA	Rio Grande do Sul	A	730	79	*Int2*
16POA	Rio Grande do Sul	A	730	79	*Int2*
6SP	São Paulo	A	79	79	*Int2*
7SP	São Paulo	A	79	79	*Int2*
1PR	Paraná	B	180	15	*Int1*
7PR	Paraná	B	180	15	*Int1*
8PR	Paraná	B	180	15	*Int1*
9PR	Paraná	B	180	15	*Int1*
15PR	Paraná	B	180	15	*Int1*
17PR	Paraná	B	891	Singleton	*Int1*
5PR	Paraná	C	890	15	*Int1*

NOTE. REP-PCR, repetitive-sequence–based polymerase chain reaction; CC, clonal complex; ST, sequence type; MLST, multilocus sequence typing; *Int*, integron.
[a]Sequence type by Institute Pasteur MLST scheme.
[b]MLST clonal complex defined by Institute Pasteur.

Acinetobacter baumannii belonging to CC15 have had evolutionary success, and they usually exhibit multidrug-resistant phenotypes, which has facilitated their rapid clonal expansion in recent years. This clonal complex has been related to OXA-23 outbreaks in several European countries, including Italy, Spain, Greece, and Turkey, as well as an outbreak in South America.[5] To the best of our knowledge, this is the first description of CC15 associated with OXA-72 production reported in the literature.

The screening for integrase genes demonstrated the presence of class 1 and 2 integrons associated with CC15 and CC79, respectively (Table 1). Interestingly, Martins et al[7] showed the same distribution of class 1 and 2 integrons, related to specific CCs such as CC15 and CC79.[7] In a previous study, we reported a high prevalence of class 2 integrons in southern Brazil. Indeed, these results might be explained by the association of class 2 integrons to CC79, which are prevalent in South America.[8]

The OXA-72 enzyme was first identified in *A. baumannii* from Thailand in 2004. Later, this enzyme was reported in *Acinetobacter* spp. clinical isolates from China, South Korea, Taiwan, Italy, Spain, and France.[9] Until now, there have only been a few reports of *A. baumannii* carrying the *bla*OXA-72 gene in Brazil.[1] In the present study, we have described the association of *A. baumannii* carrying *bla*OXA-72 with the epidemic clones CC15, CC79, and a new ST (ST 891). Camargo et al[2] described *bla*OXA-72 producers of CC79 in Brazil. In contrast, the data presented in our study have demonstrated not only the association of *bla*OXA-72 producers to CC79 but also to CC15. The ability of these particular clones to acquire antibiotic resistance should trigger

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efforts to prevent the spread of OXA-72–producing isolates as occurred with *bla*OXA-23.[10] These data indicate the potential for this gene to spread to different countries and distinct geographical regions.

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A Silent Epidemic of Colistin- and Carbapenem-Resistant Enterobacteriaceae at a Turkish University Hospital

To the Editor—We read with great interest the manuscript emphasizing increasing resistance to colistin and tigecycline in Enterobacteriaceae.[1] Hence, we present the epidemiology of colistin- and carbapenem-resistant (CoCR) *Klebsiella pneumoniae* (CoCR-KP) and *Escherichia coli* (CoCR-*E. coli*) isolated from various clinical samples from January 1 through July 30, 2015, at a 700-bed tertiary care university hospital. We also report synergy testing results of antibiotic combinations that could be used for the treatment of the infections caused by CoCR isolates.

A total of 19 isolates (6 *E. coli*, 13 *K. pneumoniae*) from 17 patients were included in the study. All *E. coli* and 3 *K. pneumoniae* isolates were recovered from rectal swab samples collected during a point prevalance program performed for detection of CR-KP colonization in accordance with Centers for Disease Control and Prevention methods. Ten *K. pneumoniae* isolates were obtained from urine (n = 7), blood (n = 1), central venous catheter (n = 1), and peritoneal fluid (n = 1) samples. The identification of the isolates was made by matrix-assisted laser desorption/ionization–time of flight mass spectrometry (VITEK MS; bioMérieux) and by analytical profile index (API20E; bioMérieux). Antimicrobial susceptibility testing against carbapenem, colistin, and tigecycline was performed by Etest (bioMérieux) and against amikacin, gentamicin, cefuroxime, ceftazidime, cefepime,