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AbaR-Type Genomic Islands in Non-*baumannii Acinetobacter* Species Isolates from South Korea

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To investigate the presence and structure of AbaR-type genomic islands (GIs) in non-*Acinetobacter baumannii* isolates, a total of 155 non-*baumannii Acinetobacter* isolates from a South Korean hospital were analyzed. GIs were found in three *Acinetobacter nosocomialis* and two *Acinetobacter seifertii* isolates. Their structures were similar to those in *A. baumannii* isolates from Asian countries, including South Korea. The existence of AbaR-type GIs in non-*baumannii Acinetobacter* isolates is believed to be due to interspecies transfer of GI.

Recently, *Acinetobacter* spp. have emerged as important opportunistic nosocomial pathogens. In addition to *Acinetobacter baumannii*, which is the most important species in clinical settings, especially in intensive care units, other *Acinetobacter* spp., such as *A. nosocomialis* and *A. pittii*, are frequently isolated in hospitals (1, 2). A resistance island, termed AbaR1, was identified by whole-genome sequence comparisons with a multidrug-resistant (MDR) *A. baumannii* strain, AYE (3). AbaR1 is integrated into the ATPase gene (now called *comM*) and contains a large cluster of antimicrobial and heavy metal resistance genes. Some studies have revealed diverse related AbaR resistance islands in the same region of *comM* in *A. baumannii* isolates (4–8). Although most AbaR resistance islands have been reported in *A. baumannii* isolates, recently, AbaR4 was reported in *A. nosocomialis* isolates from South Korea and Thailand (9). However, the prevalence and characteristics of AbaR-type genomic islands (GIs) in non-*baumannii Acinetobacter* isolates are not well known.

In the present study, the prevalence of AbaR-type GIs was investigated among non-*baumannii Acinetobacter* isolates from a South Korean hospital. In addition, the structure of AbaR-type GIs found in non-*baumannii Acinetobacter* isolates was analyzed.

In a previous study (10), 155 non-*baumannii Acinetobacter* strains were isolated from patients with bloodstream infections admitted to a tertiary care hospital in South Korea between August 2003 and February 2010. Species identification of the isolates using *rpoB* and 16S rRNA gene sequences revealed 93 *A. nosocomialis*, 28 *Acinetobacter seifertii* (formerly *Acinetobacter* genomic species "close to 13TU"), 15 *A. pittii*, three *Acinetobacter calcoaceticus*, six *Acinetobacter bereziniae*, four *Acinetobacter* genomic species 16, three *Acinetobacter ursingii*, two *Acinetobacter parvus*, and one *Acinetobacter junii* strain. *In vitro* antimicrobial susceptibility testing was also performed using a broth microdilution method, according to CLSI guidelines, in a previous study (10, 11). All of these 155 isolates were used in the present study.

Transposon insertion into *comM* was investigated using previously published primers for all *Acinetobacter* isolates (12, 13). Amplification of intact *comM* (982 bp) indicated that no GI interrupted *comM*, while no amplification indicated the possibility of GI interrupting *comM*. The integration of GI was confirmed by using two primer sets amplifying AbaR-*comM* (RH927 and RH797) and *comM*-AbaR (RH916 and RH928) (12). The structure of AbaR-type GIs in non-*baumannii Acinetobacter* isolates was identified by sequential PCR amplification (amplicon sizes, 4 to 5 kb) and sequencing using additional 20 primers.

Among the 155 non-*baumannii Acinetobacter* isolates, GIs were identified in five isolates, of which three were *A. nosocomialis* and two were *A. seifertii*. On the other hand, the *comM* gene was yielded by primers used in this study in *A. pittii*, *A. calcoaceticus*, *A. junii*, *A. parvus*, *A. ursingii*, *A. bereziniae*, and *Acinetobacter* genomic species 16. Among the five GI-positive non-*baumannii Acinetobacter* isolates, three and four were resistant to imipenem and meropenem, respectively (Table 1). All GI-positive non-*baumannii Acinetobacter* isolates were resistant to ciprofloxacin, piperacillin-tazobactam, and ampicillin-sulbactam. *A. nosocomialis* strain H06-681 was resistant to all antimicrobial agents, excluding imipenem, polymyxins, and tigecycline. Only *A. seifertii* strain C066 was resistant to colistin.

The structure of AbaR-type GIs in three *A. nosocomialis* and two *A. seifertii* isolates was determined (Fig. 1). *A. nosocomialis* H06-681 and *A. seifertii* C066 carried Tn*6022* with a deletion of *tniD* (Tn*6022*Δ*tniD*), although they belong to different species (Fig. 1). Although a bla_{OXA-23}-like gene was identified with IS*Aba1* in *A. nosocomialis* H06-681, it was not detected within the GI. Thus, the bla_{OXA-23}-like gene was assumed to be located in another region of the chromosome of *A. nosocomialis* H06-081, along with IS*Aba1*. On the other hand, no bla_{OXA-23}-like gene was identified in *A. seifertii* C066. *A. nosocomialis* strains H09-1045 and E09-34, which showed very high MICs for carbapenems (Table 1), harbored AbaR4, which is composed of Tn*6022* with Tn*2006* (14). In *A. seifertii* strain C044, Tn*6166* was also identified (Fig. 1). However, the Tn*6166* identified in this isolate, which lacked *tniD* and Tn*2006*, included *tetA*(B), *tetR*(B),

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TABLE 1 AbaR-type genomic island and antimicrobial resistance in non-*baumannii Acinetobacter* isolates harboring genomic islands

Species	Isolate	Type	MIC (mg/liter) of[a]:													
			IMP	MEM	TET	CIP	RIF	AMK	CPM	CTR	CAZ	P/T	A/S	PB	COL	TIG
A. nosocomialis	H06-681	Tn*6022*Δ*tniD*	2	**16**	**>64**	**>64**	**4**	**>128**	**>64**	**>128**	**>64**	**>256/4**	**64/32**	1	2	4
	H09-1045	AbaR4	**>64**	**>64**	**64**	**64**	2	32	**64<**	16	8	**>256/4**	**64/32**	2	2	4
	E09-34	AbaR4	**>64**	**>64**	**>64**	**8**	**4**	32	**64<**	16	8	**>256/4**	**>64/32**	2	2	4
A. seiffertii	C044	Tn*6166*	1	1	**>64**	**>64**	**4**	**>128**	16	**>128**	**>64**	**256/4**	**64/32**	1	2	2
	C066	Tn*6022*Δ*tniD*	**16**	**32**	8	**>64**	2	16	**>64**	**>128**	**>64**	**>256/4**	**64/32**	2	**8**	4

[a] IMP, imipenem; MEM, meropenem; TET, tetracycline; CIP, ciprofloxacin; RIF, rifampin; AMK, amikacin; CPM, cefepime; CTR, ceftriaxone; CAZ, ceftazidime; P/T, piperacillin-tazobactam; A/S, ampicillin-sulbactam; PB, polymyxin B; COL, colistin; TIG, tigecycline. MICs in bold indicate resistance. The breakpoints of resistance are from CLSI guidelines (11) for most antimicrobial resistance (resistance defined as ≥16 mg/liter for both imipenem and meropenem). The criteria recommended by the CLSI for staphylococci were applied for rifampin (resistance defined as ≥4 mg/liter), and the criteria of the U.S. Food and Drug Administration (FDA) for *Enterobacteriaceae* were used for tigecycline (resistance defined as ≥8 mg/liter) (18).

CR2, *strB*, *strA*, and orf4b instead and did not interrupt the *comM* gene.

Since the discovery of AbaR1 in *A. baumannii* strain AYE in 2006 (3), it has been known that the resistance island may play a significant role in the antimicrobial resistance of *A. baumannii*. Since then, several of its variants have been identified and used in epidemiological studies (4–8, 12, 15–17).

One of the most interesting findings in this study was that GIs were identified in five isolates of *A. nosocomialis* and *A. seifertii*, which belong to the *A. calcoaceticus/A. baumannii* (ACB) complex or *A. baumannii* complex. None of them are clonal, judging from 16S rRNA and *rpoB* gene sequences. Non-*baumannii Acinetobacter* species of *A. baumannii* complex, such as *A. nosocomialis*, *A. pittii*, and *A. seifertii*, are increasingly reported to cause human infections with the introduction of molecular identification tools (2). The AbaR-type GIs of five non-*baumannii Acinetobacter* isolates identified in this study were shared with those of *A. baumannii* isolates. Tn*6022*Δ*tniD*, which was detected in *A. nosocomialis* H06-681 and *A. seifertii* C066, contains a Tn*6022* backbone and is the simplest GI (14, 16). The same GI structure found in different non-*baumannii Acinetobacter* species may be evidence that horizontal transfer of GIs occurred several times. AbaR4, which was identified in two isolates of *A. nosocomialis*, has been reported in *A. baumannii* sequence type 75 (ST75) isolates from South Korea, in *A. nosocomialis* strain Th01-06 from Thailand, and in many *A. baumannii* isolates from South Korea (9, 15). In addition, the Tn*6166* structure of C044 is identical to that described by Nigro and Hall (6). These data imply that the GIs are

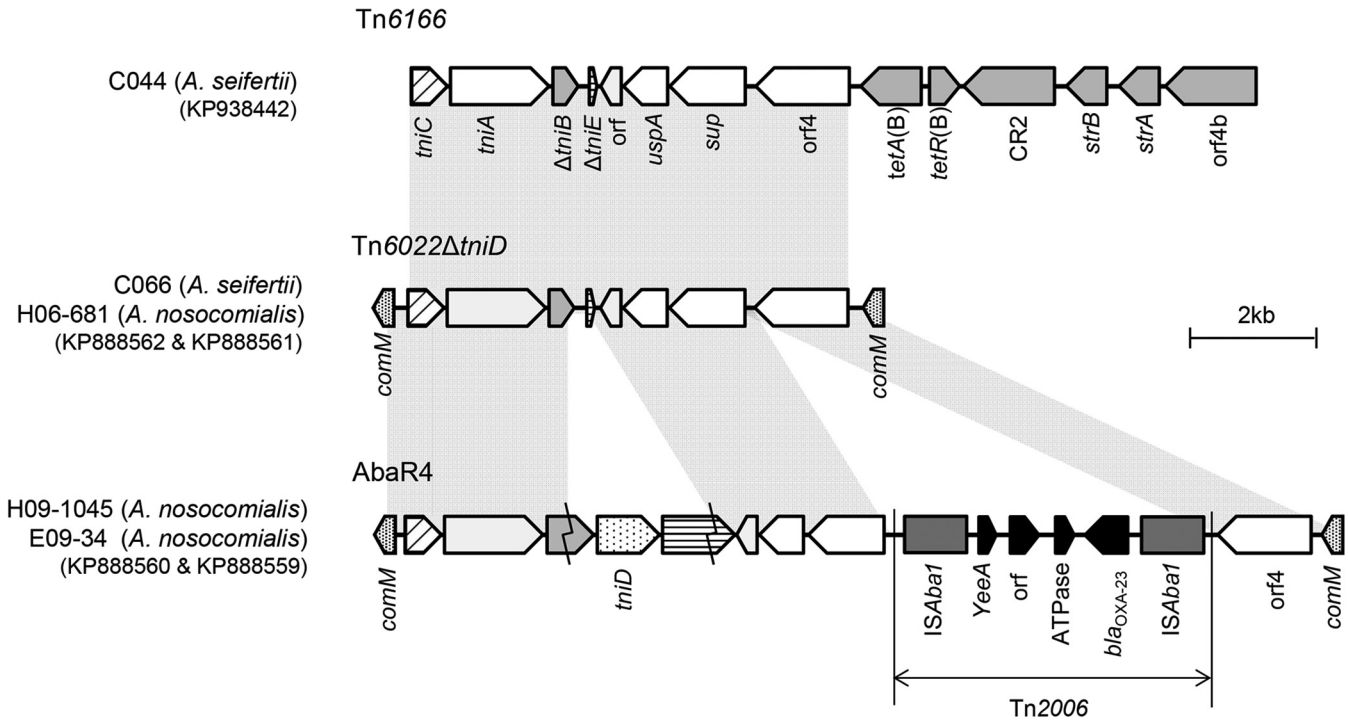

FIG 1 Structure of GIs in five non-*baumannii Acinetobacter* isolates from Korea. Tn*6166* of C044 is different from Tn*6022*Δ*tniD* of C066 and H06-681 in that six genes, *tetA*(B), *tetR*(B), *CR2*, *strB*, *strA*, and orf4b, are present at the 5′ end of orf4. In AbaR4 of H09-1045 and E09-34, *tniD* was intact and Tn*2006*, including bla_{OXA-23}, was incorporated, comparing it with Tn*6022*Δ*tniD*.

possibly transferred among *Acinetobacter* species and suggest the increased frequency of AbaR-type GI in non-*baumannii Acinetobacter* isolates in the future.

In this study, we identified the AbaR-type GIs in five non-*baumannii Acinetobacter* isolates, and their structures were determined. The structure of GIs in non-*baumannii Acinetobacter* isolates suggests the interspecies transfer of GIs.

Nucleotide sequence accession numbers. Sequences determined in this study have been deposited in GenBank under accession no. KP938442, KP888559, KP888560, KP888561, and KP888562.

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