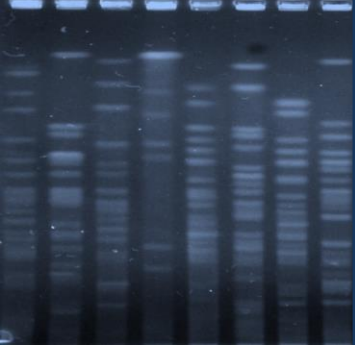
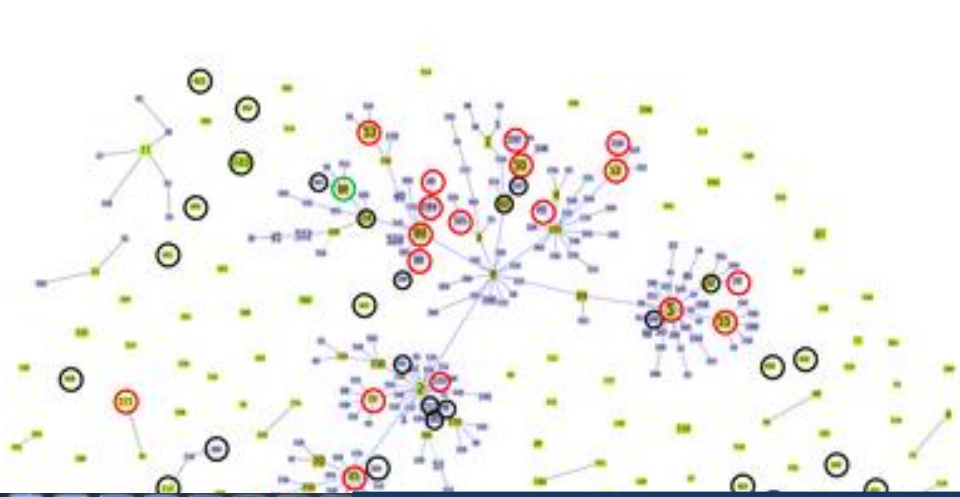




Hastane Salgınlarında Moleküler Epidemiyolojik Yaklaşım

Bariş Otlu
İnönü Üniversitesi Tıp Fakültesi
Tıbbi Mikrobiyoloji Anabilim Dalı



Hastane İnfeksiyonları Tarihi

3. Yüzyıl Roma Şiiri, Bir hastanın Dr. Simmakus'a seslenişi

“Hasta oluyordum ve sen hemen geldin
Yanında yüz öğrenciyle, ah **Simmakus**
Yüz soğuk el bana dokundu
Hiç ateşim yoktu, ah Simmakus, şimdi var”



Hastane İnfeksiyonları Tarihi

- Hastane ve hastane infeksiyonları kavramlarının birlikte geliştiği düşünülebilir.



18.yy' da Hotel Dieu;

- amputasyonlardan ölüm oranı %60
- salgın sırasında lohusalık hummasından ölüm oranı 19/20

1840- El Hijyeninin Önemi → 1976 Standart Enfeksiyon Önlemi

- Hastane enfeksiyonlarının tespiti ve önlenmesi ile ilgili **ilk bilimsel yaklaşım**.



Ignaz Semmelweis

Downloaded from <http://bmjopen.bmj.com/> on March 26, 2018 - Published by group.bmj.com

Open Access

Research

BMJ Open 'And you'll suddenly realise 'I've not washed my hands': medical students', junior doctors' and medical educators' narratives of hygiene behaviours

Penelope Cresswell,¹ Lynn V Monrouxe²

To cite: Cresswell P, Monrouxe LV. 'And you'll suddenly realise 'I've not washed my hands': medical students', junior doctors' and medical educators' narratives of hygiene behaviours. *BMJ Open* 2018;8:e018156. doi:10.1136/bmjopen-2017-018156

► Prepublication history for this paper is available online. To view please visit the journal online (<http://dx.doi.org/10.1136/bmjopen-2017-018156>).

Received 9 June 2017
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Accepted 9 October 2017

ABSTRACT

Objective Compliance to hygiene behaviours has long been recognised as important in the prevention and control of healthcare associated infections, but medical doctors still display some of the lowest rates of compliance of all healthcare workers. We aim to understand compliance to hygiene behaviours by analysing medical students', junior doctors' and medical educators' narratives of these behaviours to identify their respective attitudes and beliefs around compliance and how these are learnt during training. Such an understanding can inform future interventions to improve compliance targeted to areas of greatest need.

Design A qualitative study, using narrative interviews (nine focus groups and one individual interview). Data were analysed thematically using inductive framework analysis.

Setting Teaching hospitals in the UK.

Participants Convenience sample of 25 participants: third-year medical students in their first clinical year (n=13), junior doctors (n=6) and medical educators (n=6).

Results We identified four main themes: (1) knowledge, (2) constraints, (3) role models/culture and (4) hygiene as an added extra. Knowledge varied across participant groups and appeared to influence behaviours; medical students relied on what they have been told by seniors, while medical educators relied on their own knowledge and experience. There was a strong belief that evidence

Strengths and limitations of this study

- This is the first study we are aware of that compares medical students and doctors at different stages of training through qualitative interviews.
- Qualitative narrative interviewing allowed participants to share their stories so the data are grounded in behaviours as well as attitudes and beliefs.
- Opportunistic sampling could lead to population bias and reduced generalisability.
- Data collected from participants in two health boards in a single UK country.

(HCAs) are a priority in healthcare.¹ It is estimated that HCAs cost the UK National Health Service (NHS) in excess of £1 billion every year, posing a significant economic burden.² In terms of human cost, it is thought that over 5000 deaths occur as a direct consequence of HCAs each year.² This is particularly significant as, in many circumstances, HCAs have been shown to be preventable through good hygiene behaviours. An intervention leading to improvements in hygiene practices resulted in a decrease of 31% and

Hastane İnfeksiyonları Tarihi

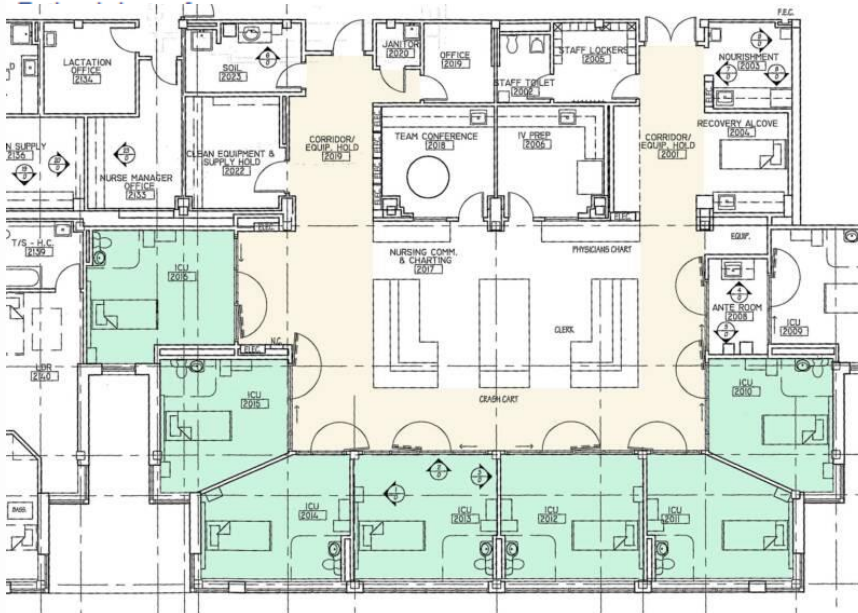
from *Notes on Hospitals* published in 1863

[J Emerg Trauma Shock. 2009 Sep-Dec; 2\(3\): 175–179.](#)

PMCID: PMC2776365

doi: [10.4103/0974-2700.55329](#)

Hospital design for better infection control



from *Notes on Hospitals*

Hastane Enfeksiyonları Tarihi- Yoğun Bakımlar

Intensive care medicine is 60 years old: the history and future of the intensive care unit

Authors: Fiona E Kelly,^A Kevin Fong,^B Nicholas Hirsch^C and Jerry P Nolan^D

Intensive care is celebrating its 60th anniversary this year. The concept arose from the devastating Copenhagen polio epidemic of 1952, which resulted in hundreds of victims experiencing respiratory and bulbar failure. Over 300 patients required artificial ventilation for several weeks. This was provided by 1,000 medical and dental students who were employed to hand ventilate the lungs of these patients via tracheostomies. By 1953, Bjorn Ibsen, the anaesthetist who had suggested that positive pressure ventilation should be the treatment of choice during the epidemic, had set up the first intensive care unit (ICU) in Europe, gathering together physicians and physiologists to manage sick patients – many would consider him to be the ‘father’ of intensive care. Here, we discuss the events surrounding the 1952 polio epidemic, the subsequent development of ICUs throughout the UK, the changes that have occurred in intensive care over the past 10 years and what the future holds for the specialty.



Fig 3. An 8-year-old girl being hand ventilated via a tracheostomy.



Fig 1. Coventry alligator iron lung.

Hastane İnfeksiyonları Tarihi-Yoğun Bakımlar

J. Hyg., Camb. (1969), 67, 525
Printed in Great Britain

525

Infection in intensive care unit

529

Control of cross-infection in an intensive care unit

By D. M. HARRIS, J. M. ORWIN, J. COLQUHOUN AND H. G. SCHROEDER

*From the Department of Bacteriology and the Intensive Therapy Unit,
Royal Hospital, Sheffield*

conditions which rendered them liable to infection. In groups II and III the proportion of patients admitted on account of primary lung disease or major trauma was much lower than in group I.

Table 2 is an attempt to assess the influence of I.P.P.V. on the acquisition of infection. For the three categories of patient considered (all of whom had been subjected to tracheostomy or endotracheal intubation), the incidence of infection was approximately the same whether I.P.P.V. had been used or not.

Considerable attention is

Causative organism	No. of infections	Site of infection			
		Sputum	Tracheostomy	Wound swab	Urine
Coliforms	39	18	6	5	10
<i>Ps. aeruginosa</i>	21	11	7	2	1
<i>Proteus</i> spp.	9	5	1	1	2
<i>H. influenzae</i>	6	5	1	0	0
<i>Strep. pneumoniae</i>	3	3	0	0	0
<i>Staph. aureus</i>	21	10	4	7	0
<i>Strep. faecalis</i>	4	0	1	3	0
<i>Strep. pyogenes</i>	3	0	0	3	0
<i>Candida</i> spp.	14	3	11	0	0
<i>Clostridium welchii</i>	1	0	0	1	0
Totals	121	55	31	22	13



Plan of the Intensive Therapy Unit at the Royal Hospital, Sheffield.



ELSEVIER



ScienceDirect



REVIEW

Acinetobacter: an old friend, but a new enemy

K.J. Towner*

Department of Clinical Microbiology, Nottingham University Hospitals NHS Trust, Queen's Medical Centre, Nottingham NG7 2UH, UK

Available online 22 August 2009

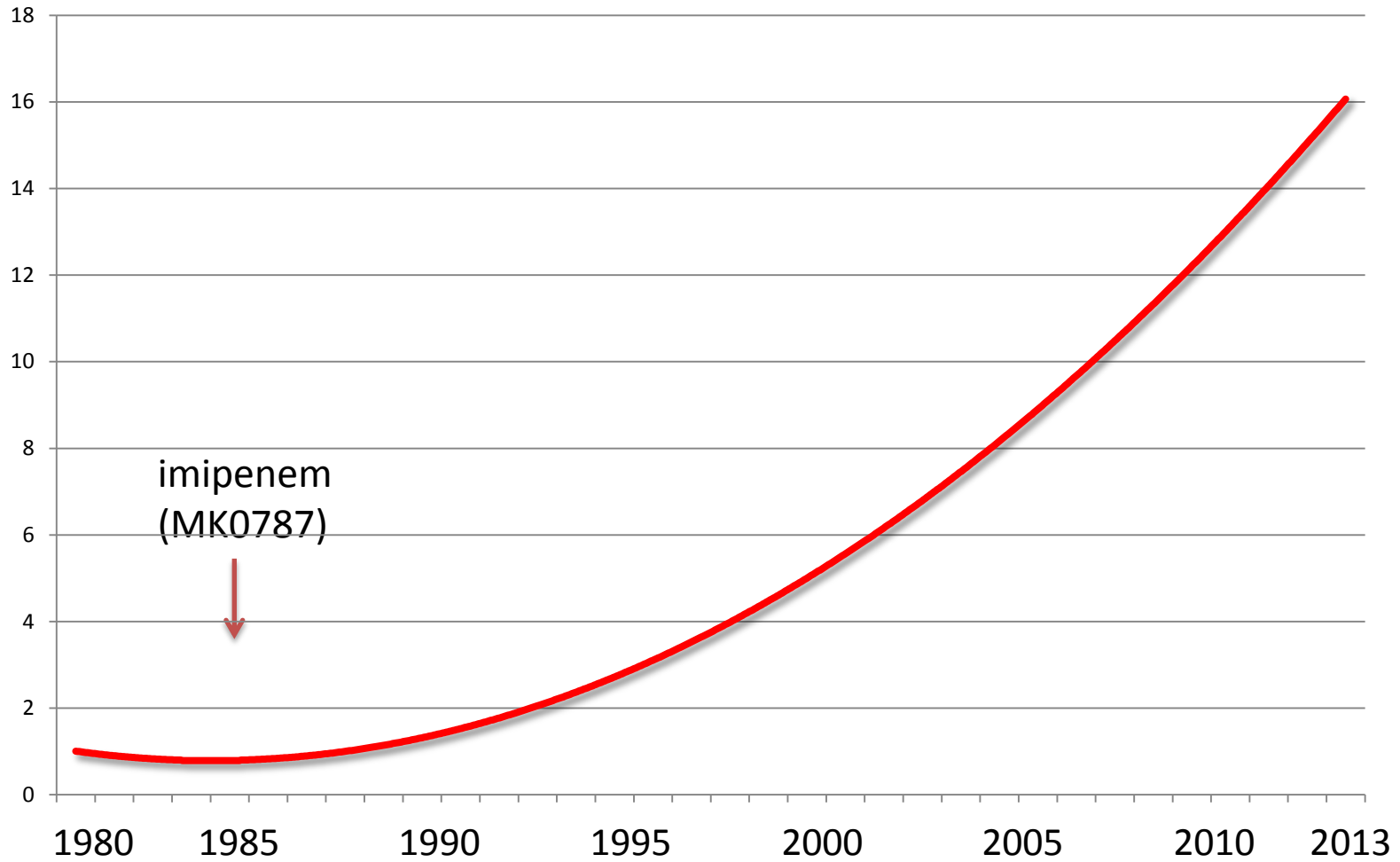
KEYWORDS

Acinetobacter;
Epidemiology;
Infection control
measures;
Nosocomial infection;
Therapeutic options

outbreaks can occur in such units, involving the infection or colonisation of numerous patients by specific epidemic strains of *A. baumannii*. Recently, a particular problem has concerned cross-infection of injured military patients repatriated from combat regions of the world (e.g. Iraq and Afghanistan). Carbapenems have previously been the treatment of choice for infected patients, but increasing reports worldwide now describe *A. baumannii* isolates resistant to all conventional antimicrobial regimens. Data to support therapeutic use of the limited number of new antimicrobial agents (e.g. tigecycline) with in-vitro activity against these pathogens are still very limited. Detailed advice concerning prevention and control of outbreaks caused by multidrug-resistant strains of acinetobacter is available from the UK Health Protection Agency. In addition to antibiotic prescribing policies and audit, these measures focus on reinforcing standard infection control procedures and precautions, with particular attention to thorough cleaning of patient areas to take account of the long-term survival of acinetobacter after drying and inadequate disinfection. Despite these measures, the problem continues to escalate, with many hospitals worldwide now reporting outbreaks caused by multidrug-resistant strains of acinetobacter.

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PubMed'de son 35 yılda 250 den fazla *Acinetobacter spp.* salgını yayın haline gelmiş !!!



Hastane İnfeksiyonları-Sınıflandırma

- Endemik Hastane İnfeksiyonları

Bir hastalığın belirli bir yerde (örneğin bir hastanede) her zaman beklenen görülme düzeyi.

Hastane infeksiyonlarının %90-95

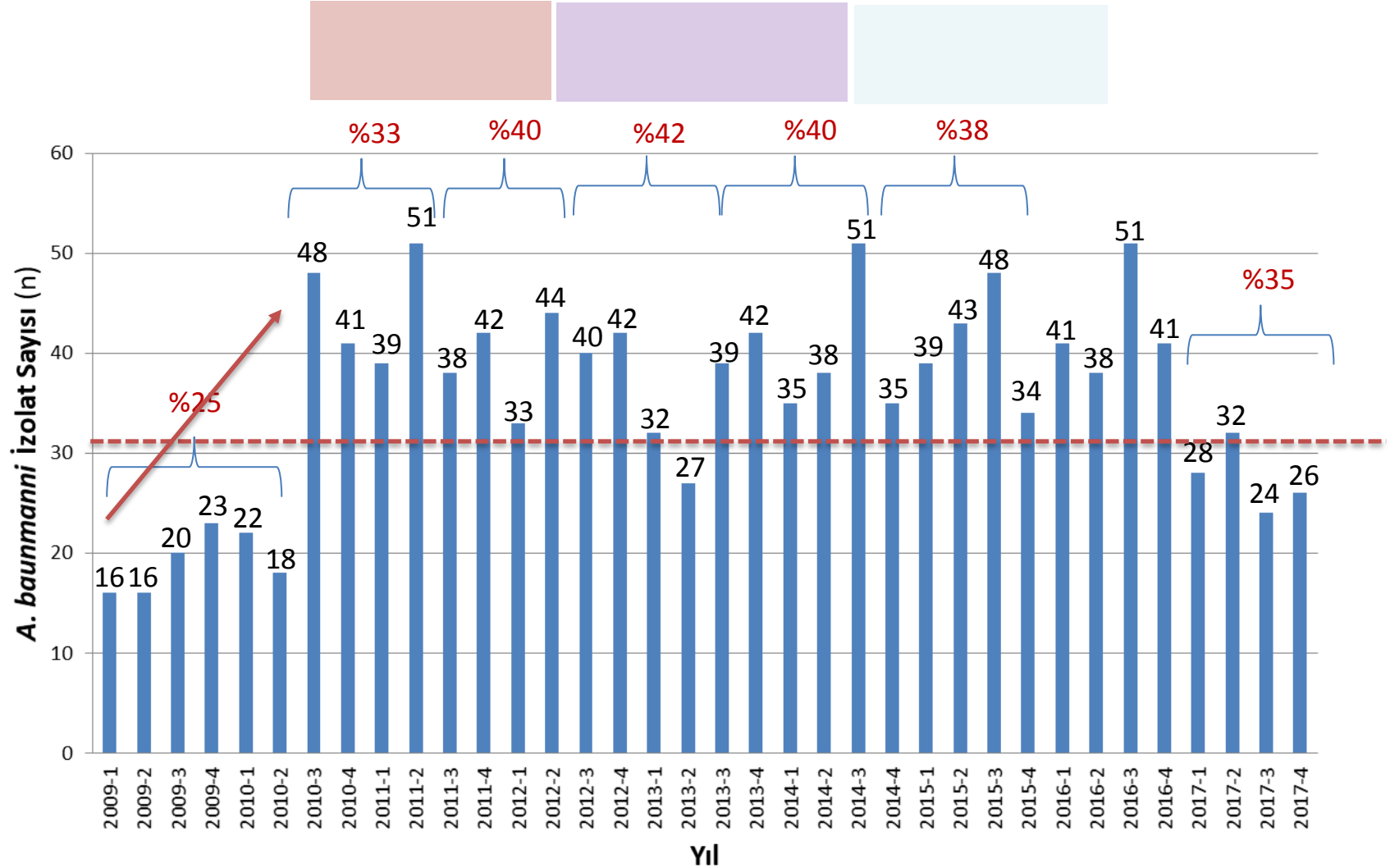
- Epidemik Hastane İnfeksiyonları

Hastane infeksiyonların yaklaşık %5-10

Görülme sıklığı: 1/10.000 yatış

Endemik Hastane İnfeksiyonları

- Yoğun bakımlarda *A. baumannii* oranlarımızın yıllara göre değişimi



Epidemik Hastane İnfeksiyonları-Salgınlar

- Kaos, kargaşa ve tedirginlik başlar.
- Komplo teorileri üretilir.
- Güvenirlilik ve prestij azalır, bir suçlu aranır.



Hastane İnfeksiyonları ve Mikrobiyoloji Laboratuvarı

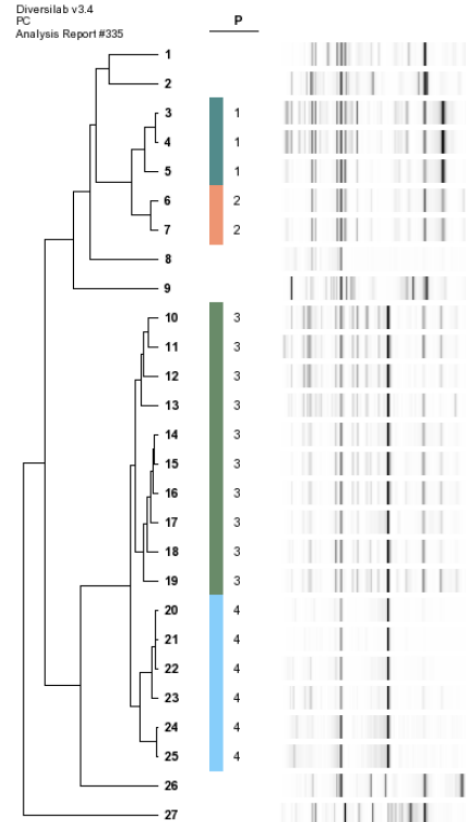
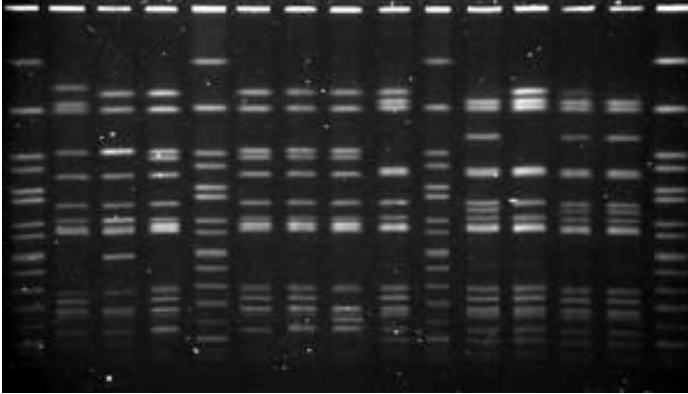
- İnfeksiyona neden olan etkenin belirlenmesi
 - Etkenin hızla tespiti
 - Antimikrobiyal duyarlılığın belirlenmesi
 - Antibiyotik direnç genlerinin tespit edilmesi
 - Virülans genlerinin belirlenmesi
- İzolatlar arasındaki klonal ilişkinin ve bulaş kaynağının araştırılması
 - Hastane salgınlarının tespit edilmesi
 - Salgının kaynağının belirlenmesi
 - Dirençli bakterilerin izlenmesi
 - Re-aktivasyonun yeni bulaştan ayırt edilmesi

**“Moleküler Epidemiyoloji 21. yüzyılın
en önemli ve heyecan verici
araştırma alanlarından biridir”**



Moleküler Epidemiyoloji

- Moleküler epidemiyoloji terimi;
genellikle “moleküler biyolojik yöntemlerle suş tiplendirme”
çalışmalarına verilen ortak bir isim gibi kullanılmaktadır.





Population Biology, Evolution, and Infectious Disease: Convergence and Synthesis

Bruce R. Levin,¹ Marc Lipsitch,¹ Sebastian Bonhoeffer²

Traditionally, the interest of population and evolutionary biologists in infectious diseases has been almost exclusively in their role as agents of natural selection in higher organisms. Recently, this interest has expanded to include the genetic structure and evolution of microparasite populations, the immune response, and the population genetics of medical and public health interventions. In these areas, emphasizing the ways in which these approaches have been contributing to the design and evaluation of interventions.

Moleküler epidemiyolojinin sınırlarını;

- etken mikroorganizmanın tespiti
- bulaşma yollarının araştırılması
- virülanslarından sorumlu genlerin belirlenmesi ve
- izolatlar arası **klonal ilişkilerin araştırılmasını** kapsayan geniş bir yelpazede çizilmiştir.

Moleküler Epidemiyoloji



Moleküler Epidemiyoloji

- Hastane infeksiyonuna neden olan etkenlerin izini sürebilmek için;
 - izolatların moleküler karakterizasyonu yapılmalı ve
 - izolatlar arasındaki klonal ilişkiler genotiplendirme yöntemleriyle araştırılmalıdır.



İzolatlarda Moleküler Karakterizasyonu

- İzole edilen mikroorganizmaların moleküler karakterizasyonu için;
 - antimikrobiyal direnç genleri
 - virülans genleri
 - mobil genetik elemanları
 - plazmid
 - transpozon
 - İntegron
 - hücre duvar yapıları araştırılmalıdır.

İzolatların Moleküler Karakterizasyonu

- Yoğun bakım ünitesinden izole edilen; zaman ve mekan açısından kümeleşme gösteren *27 P. aeruginosa* izolatu.



Karbapenem **dirençli 16 izolatta direnç genleri** in-house PCR ile araştırıldı.

- OXA-48 - OXA-23 - OXA-2 - VIM - KPC
- NDM - GES - SMP - GIM - SIM
- **OXA-10 +**



Karbapenemaz ?

İzolatların Moleküler Karakterizasyonu

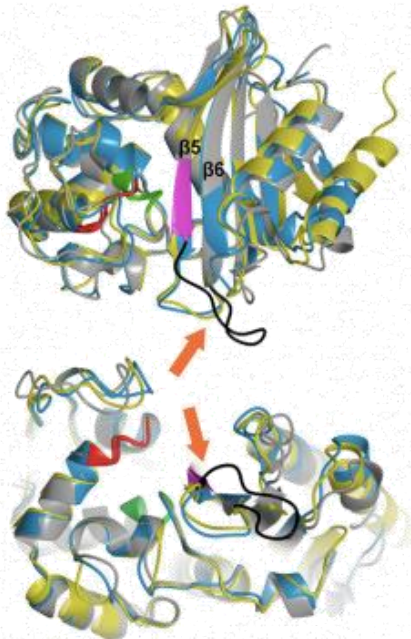
- Orijinal OXA-10 gen ürününün alfa yapısı beta'ya döndüğünde OXA-48 benzeri karbapenemaz benzeri etkin gösteriyor.

PNAS

Evolution to carbapenem-hydrolyzing activity in noncarbapenemase class D β -lactamase OXA-10 by rational protein design

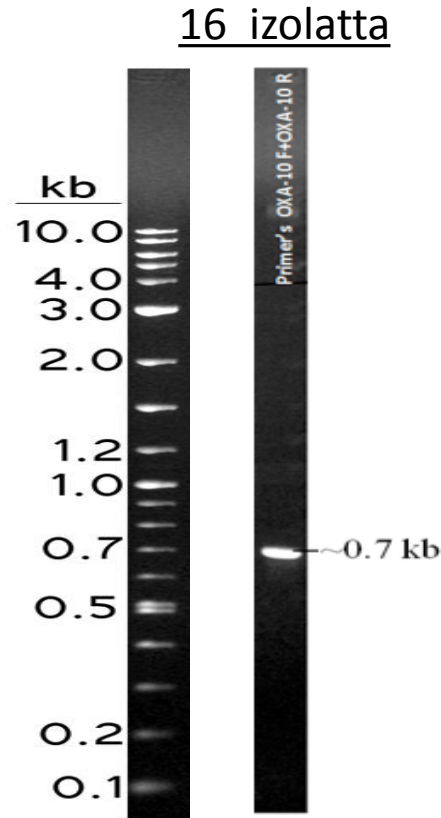
Filomena De Luca^a, Manuela Benvenuti^b, Filippo Carboni^b, Cecilia Pozzi^b, Gian Maria Rossolini^{a,c}, Stefano Mangani^{b,d}, and Jean-Denis Docquier^{a,1}

A



İzolatların Moleküler Karakterizasyonu

- OXA-10 geninin PCR ile gösterilmesi ve dizi analizi



Dizi analizi

```
tcttggaaac aagagttctc tgccgaagcc gtcaatgggt tcttcgtgct ttgtaaaagt  
agcagtaaat cctgogctac caatgactta gctcgtgcat caaaggaata tcttccagca  
tcaacattta agatocccaa cgcaattatc ggcttagaaa ctggtgcat aaagaatgag  
catcaggttt tcaaatggga cggaaagcca agagccatga agcaatggga aagagacttg  
accttaagag gggcaataca agtttcagct gttcccgat ttcaacaaat cgcagagaaa  
gttgcggaag taagaatgca gaaatacctt aaaaaattt cctatggcaa ccagaatata  
agtgttgcca ttgacaaatt ctggttgaaa ggccagctta gaatttcgc agttaatcaa  
gtggagtctc tagagtctct atatttaaat aaattgtcag catctaaga aaaccagcta  
atagtaaaa aggttttgg aacggaggcg gcacctgaat atctagtgea ttcaaaaaat  
ggtttttctg gtgtgggaac tgagtcaaat cctggtgtcg catggtgggt tgggtgggtt  
gagaaggaga cagaggttta ctttttcgcc tttaacatgg atatagacaa cgaaagt
```

- Class I Integron üzerinde taşıyor.

☐	Klebsiella pneumoniae strain CGM-HK78 plasmid pLK78, complete sequence	1214	1214	100%	0.0	100%	KJ440075.1
☒	Escherichia coli strain NF27595(C113) class I integron, partial sequence	1214	1214	100%	0.0	100%	KM111261.1
☐	Pseudomonas aeruginosa beta-lactamase OXA-10 (oxa-10) gene, partial cds	1214	1214	100%	0.0	100%	KM051379.1
☒	Providencia rettgeri strain NF13072521(432) class I integron aminoglycoside 2'-adenylyltransferase (aadB), chloramphenicol acetyltransferase (cat), beta-lactamase (g	1214	1214	100%	0.0	100%	KJ488889.1
☐	Escherichia coli strain NF130801020(188) class I integron dihydrofolate reductase (dhfrA1), aminoglycoside 6'-N-acetyltransferase (aacA4), and beta-lactamase (oxa-10)	1214	1214	100%	0.0	100%	KJ488893.1
☒	Citrobacter freundii strain S1-1 class I integron, partial sequence	1214	1214	100%	0.0	100%	KJ420610.1
☐	Uncultured bacterium clone S-31a class I integron OXA-10 and putative aminoglycoside 3'-adenylyltransferase genes, complete cds	1214	1214	100%	0.0	100%	KF525298.1
☐	Klebsiella pneumoniae plasmid InoAC-L56, complete sequence	1214	1214	100%	0.0	100%	JX442976.1
☐	Escherichia coli plasmid pNDM-HU01 genes for qacEdelta1, aadA, blaNDM-1, blaR, trpF, ISCR1 transposase, sul1, partial cds, strain: M5260	1214	1214	100%	0.0	100%	AB769140.1
☐	Enterobacter cloacae plasmid p068-50545A class I integron, partial sequence	1214	1214	100%	0.0	100%	KC678185.1
☐	Pseudomonas aeruginosa PA96 plasmid pQZ176, complete sequence	1214	1214	100%	0.0	100%	KC643497.1
☒	Pseudomonas aeruginosa strain 1083 integron OXA-10 beta-lactamase (oxa-10), aac(6)-Ib aminoglycoside modifying enzyme (aacA4), VIM-2 beta-lactamase (vim-2), a	1214	1214	100%	0.0	100%	KC527014.1
☐	Citrobacter freundii strain Iona 4 plasmid pCF1-2, complete sequence	1214	1214	100%	0.0	100%	JN215524.1
☐	Escherichia coli strain M17 class I integron class I integrase (int1) gene, partial cds, metallo-beta-lactamase GIM-1 (blaGIM-1), aminoglycoside 6'-N-acetyltransferase (g	1214	1214	100%	0.0	100%	JX566705.1
☒	Bacterium AK-MB35 class I integron containing blaIMP-9, aacA4, oxa-10 and aadA2 genes, isolate MB 35	1214	1214	100%	0.0	100%	FN178517.2
☐	Aeromonas hydrophila plasmid pR148, complete sequence	1214	1214	100%	0.0	100%	JX141473.1
☐	Klebsiella pneumoniae strain Kpn-2741 plasmid p2741 insertion sequence IS6100, partial sequence; insertion sequences IS26-1, IS26-2, and IS1999, complete sequence	1214	1214	100%	0.0	100%	NG_041455.1
☐	Proteus mirabilis strain EY991 plasmid pPM91-VEB insertion sequence IS6100 TnpA IS6100 (hnpA) gene, complete cds; insertion sequence IS26-1, complete sequence	1214	1214	100%	0.0	100%	NG_041554.1
☒	Aeromonas hydrophila strain CS2a class I integron, partial sequence	1214	1214	100%	0.0	100%	JQ837987.1
☐	Pseudomonas aeruginosa strain YMC/Anscr PAE A15 class I integron, partial sequence	1214	1214	100%	0.0	100%	JQ829829.1
☐	Providencia stuartii plasmid pMR0211, complete sequence	1214	1214	100%	0.0	100%	JN687470.1
☐	Providencia stuartii strain Pb373 plasmid InoAC class I integron, complete sequence	1214	1214	100%	0.0	100%	NG_041443.1
☒	Pseudomonas aeruginosa partial class I integron In493	1214	1214	100%	0.0	100%	AM392422.3

İzolatların Moleküler Karakterizasyonu

- Class I Integronun karakterizasyonu

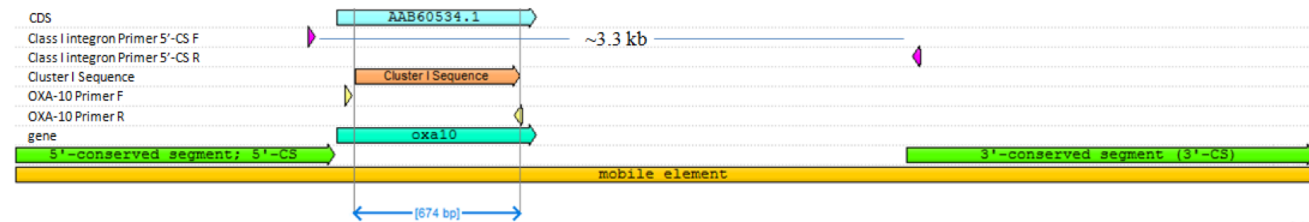
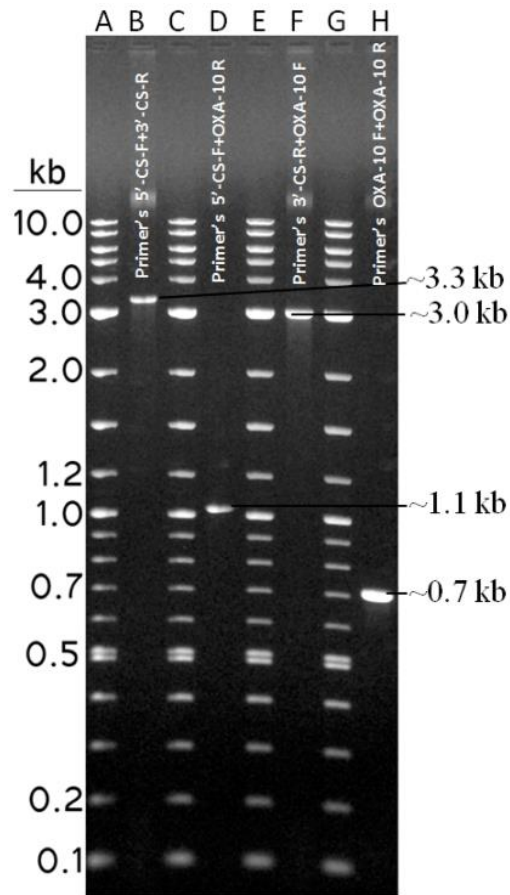


Figure 4. Schematic view of the genetic structures associated with 674 bp *blaOXA-10* gene on Class I integron. Map of Class I integron made with UGENE software (<http://ugene.unipro.ru>) from GenBank JQ629929.1

Figure 5 . PCR Gel image of integron analysis. B: PCR amplification of the variable region of Class I integron, with the specific primers belong to 5'-CS and 3'-CS regions'; D: PCR amplification of the variable region of Class I integron, with the specific primers belong to 5'-

İzolatların Moleküler Karakterizasyonu

- Karbapenemaz aktivitesinin yeniden araştırılması.
 - Karbapenemaz (-)



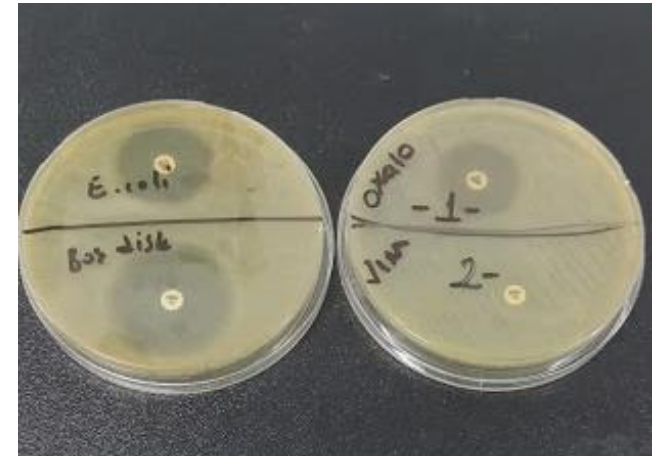
RESEARCH ARTICLE

The Carbapenem Inactivation Method (CIM), a Simple and Low-Cost Alternative for the Carba NP Test to Assess Phenotypic Carbapenemase Activity in Gram-Negative Rods

Kim van der Zwaluw*, Angela de Haan, Gerlinde N. Pluister, Hester J. Bootsma, Albert J. de Neeling, Leo M. Schouls

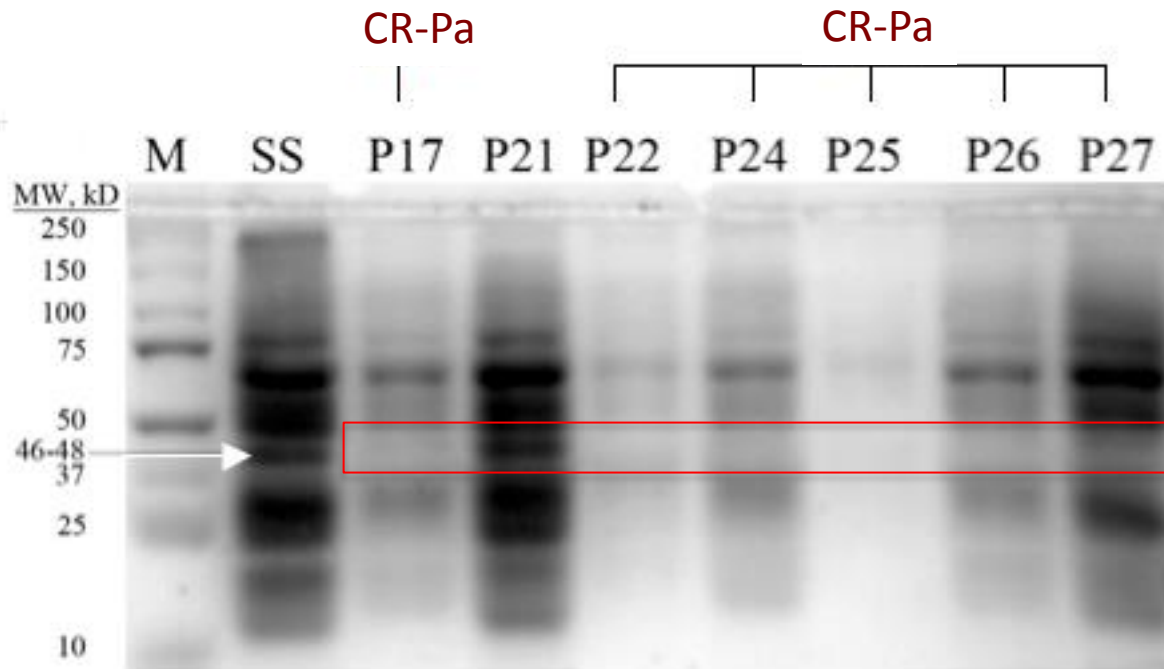
Centre for Infectious Diseases Research, Diagnostics and Screening (IDS), National Institute for Public Health and the Environment (RIVM), Bilthoven, The Netherlands

* Kim.van.der.Zwaluw@rivm.nl



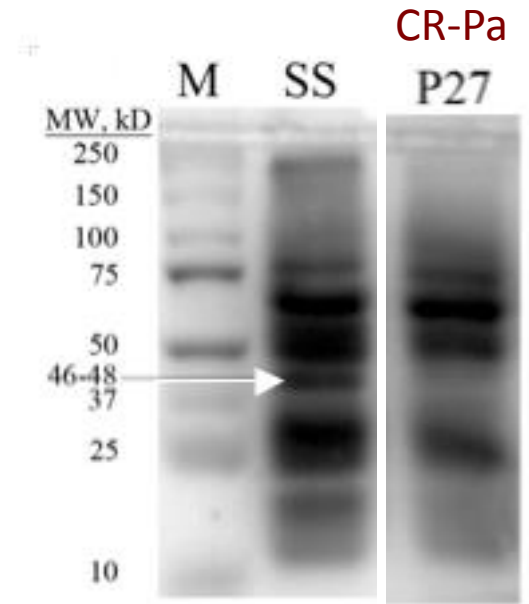
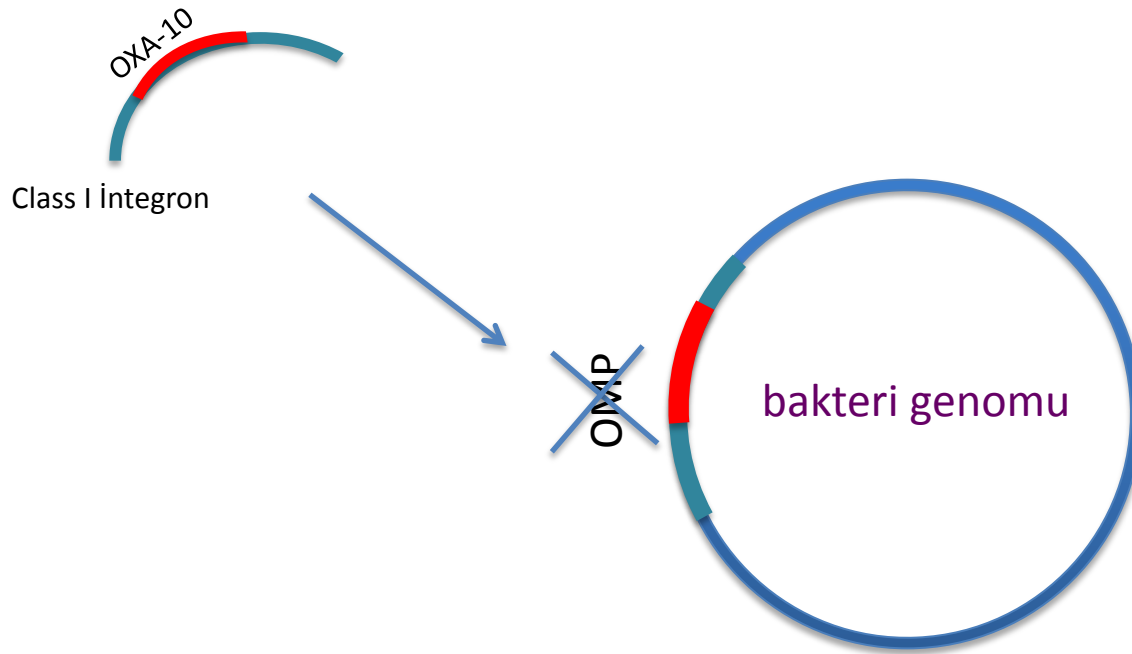
İzolatların Moleküler Karakterizasyonu

- Karbapenem direncinin **nedeni OXA-10** olamaz!
- Hücre duvarından **karbapenem geçişini** etkileyecek bir değişim var mı?
 - **SDS-PAGE**



İzolatların Moleküler Karakterizasyonu

- Sadece OXA-10 taşıyan Class I İntegrona sahip izolatlarda 47 kDa OMP eksiklik var.



İzolatların Moleküler Karakterizasyonu

- Suşla ilgi tüm veriler ve sekans bilgisinin **GenBank** kaydı yapıldı.

Pseudomonas aeruginosa beta-lactamase OXA-10 (oxa-10) gene, partial cds

GenBank: KM051379.1

[FASTA](#) [Graphics](#)

Go to: ☐

LOCUS KM051379 657 bp DNA linear BCT 23-SEP-2014
DEFINITION Pseudomonas aeruginosa beta-lactamase OXA-10 (oxa-10) gene, partial cds.
ACCESSION KM051379
VERSION KM051379.1 GI:685505748
KEYWORDS .
SOURCE Pseudomonas aeruginosa
ORGANISM [Pseudomonas aeruginosa](#)
Bacteria; Proteobacteria; Gammaproteobacteria; Pseudomonadales; Pseudomonadaceae; Pseudomonas.
REFERENCE 1 (bases 1 to 657)
AUTHORS Otlu,B., Demirdag,K., Yakupogullari,Y., Ozguler,M. and Giray Kurt,A.
TITLE Molecular Characterization of the Carbapenem-Resistant, OXA-10 Beta-Lactamase-Producing Pseudomonas aeruginosa: An Outbreak Clone that Caused High Mortality in the Patients with Ventilator Associated Pneumonia
JOURNAL Unpublished
REFERENCE 2 (bases 1 to 657)
AUTHORS Otlu,B., Demirdag,K., Yakupogullari,Y., Ozguler,M. and Giray Kurt,A.
TITLE Direct Submission
JOURNAL Submitted (19-JUN-2014) Medical Microbiology, Inonu University Faculty of Medicine, Malatya 44100, Turkey
COMMENT ##Assembly-Data-START##
Sequencing Technology :: Sanger dideoxy sequencing
##Assembly-Data-END##
FEATURES Location/Qualifiers
source 1..657



- Bu dirençli izolatların/salgınların **maliyeti ne oldu?**

enases are significant challenges in treatment of apapenem production in carbapenem-resistant *P*.
pital was aimed. A total of 84 carbapenem-resistant
d' (CIM) was used for phenotypic detection of
vIM, *bla*_{OXA-48}, and *bla*_{GES} genes was investigated by
identified by sequence analysis. Arbitrarily primed
g the isolates. The presence of high-risk clones in
esorption/ionization—time of flight mass
s carbapenemase producers by CIM tests, while PCR
rell. *bla*_{VIM} gene was found in two isolates and *bla*_{GES}
e carbapenemases were VIM-1, VIM-2, and GES-5.
MALDI-TOF MS analysis, none of the carbapenemase-
resence of VIM-1, VIM-2, and GES-5 type
st time in our hospital, GES-5 being reported for the
spread of carbapenemases and contribute to

İzolatların Moleküler Karakterizasyonu

- Bu dirençli izolatların/salgınların maliyeti ne oldu?
- 29 hastanın etkilendiği NDM-1'i taşıyan *K. pneumoniae* salgının maliyeti yaklaşık 804.263 dolar

EMERGING INFECTIOUS DISEASES®

ISSN: 1080-6059

CDC > EID journal > Volume 23 > Number 9—September 2017

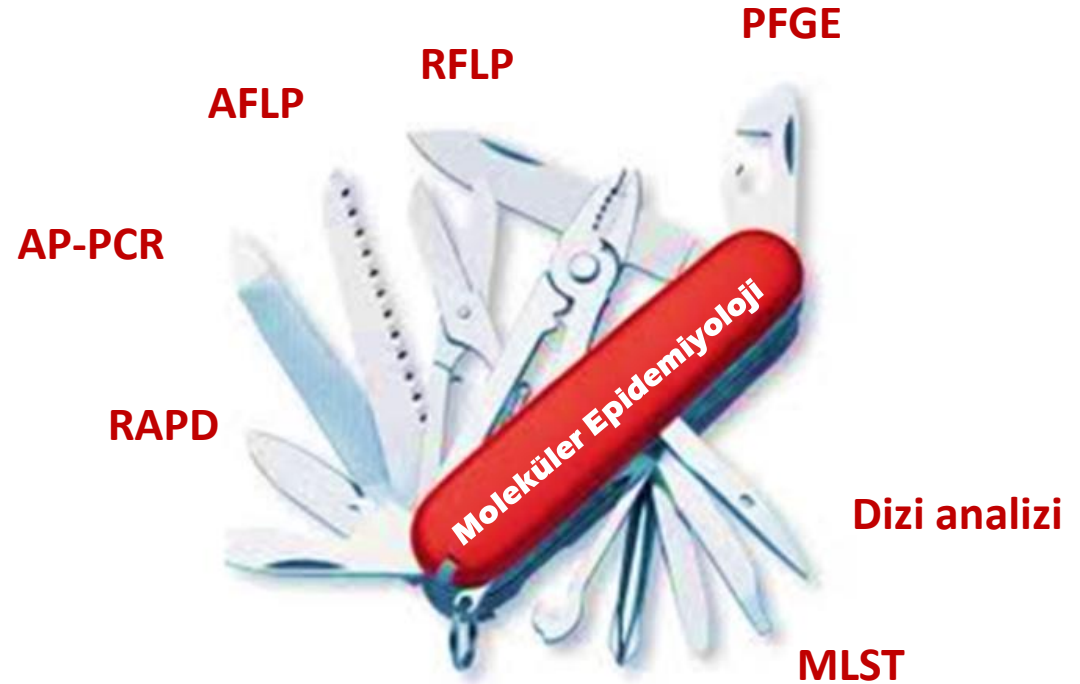
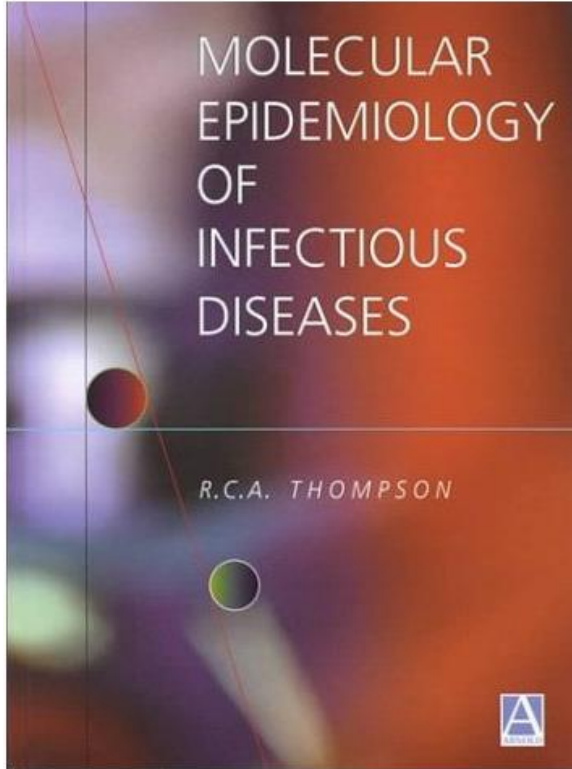


Risk altındaki 2964 kişinin >%95'i taranmış

Total outbreak costs stratified by type of cost, Jeroen Bosch hospital, the Netherlands, Oct 2015–Jan 2016*

Other outbreak control costs			
Infection prevention experts	Estimated 2,336 h for internal advice and guidance†	105,356	85,655
Patients in isolation	280 patients, averaged at 5.2 d of hospitalization, at \$31.40/d or €25.53/d (6)	45,718	37,172
Staff meetings	23 staff meetings with on average 21 participants × 0.75 h × \$1,525/h†	26,306	21,390
Communication	320 h for internal and patient-related communication spent by several communication employees†	17,696	14,387
Costs for mailings		10,701	8,700
Subtotal outbreak control costs		205,777	167,304
Total costs		804,263	653,801

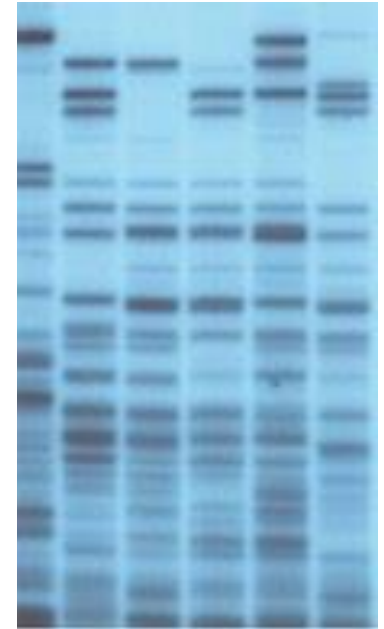
İzolatlar Arasındaki Klonal İlişkilerin Araştırılması



Moleküler Epidemiyoloji İçin Yöntemler

- Hastane infeksiyonları ile ilişkili mikroorganizmaların genotiplendirmesinde; kromozomal DNA polimorfizmine dayalı olan yöntemler, hızlı ve güvenilir olmalarıyla ön plana çıkmaktadırlar.

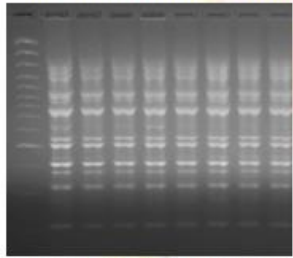
- *Pulsed field gel electrophoresis (PFGE)*
- *PCR-restriction fragment length polymorphism (PCR-RFLP)*
- *Random amplified polymorphic DNA (RAPD)*
- *Arbitrarily primed polymerase chain reaction (AP-PCR)*
- *Repetitive extragenic palindromic element-PCR (rep-PCR)*
- *Amplified ribosomal DNA restriction analysis (ARDRA)*



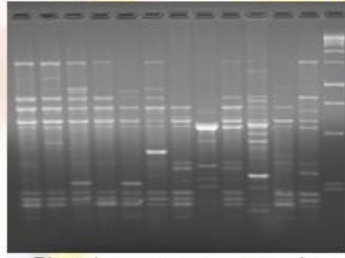
Arbitrarily Primed- PCR (AP-PCR) / Rep-PCR

- Moleküler epidemiyolojik çalışmalar için kullanılan bir hızlı PCR temeli DNA fingerprinting yöntemidir.
- Kısa primerler kullanılarak DNA'daki benzer/tekrarlayan bölgelerin amplifikasyonu yapılır.

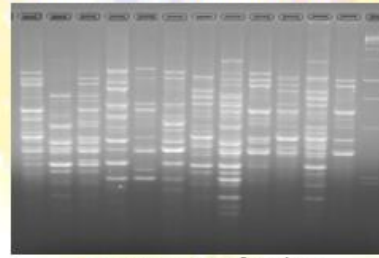
- Elektroforezde her mikroorganizma için tiplendirmeyi sağlayacak sayıda (5-21) DNA bantı elde edildi.



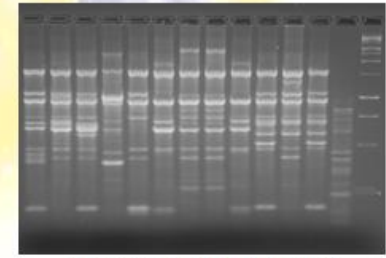
Acinetobacter baumannii



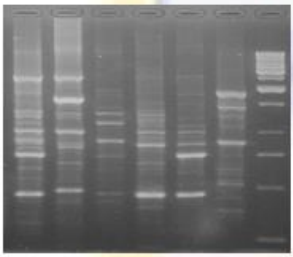
Streptococcus pneumoniae



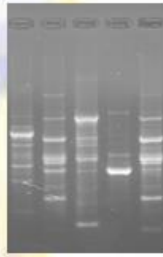
Enterococcus faecium



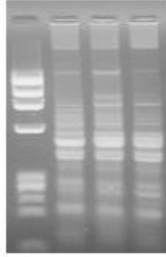
Streptococcus pyogenes



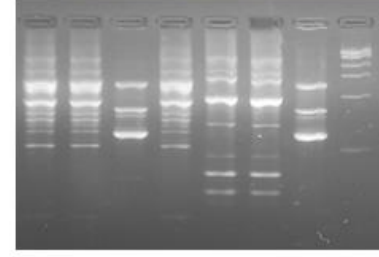
Aeromonas hydrophila



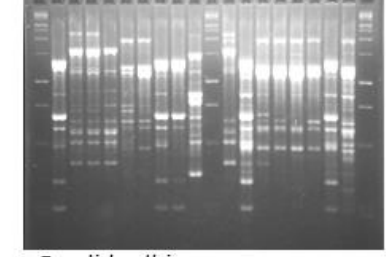
P. aeruginosa



S. maltophilia



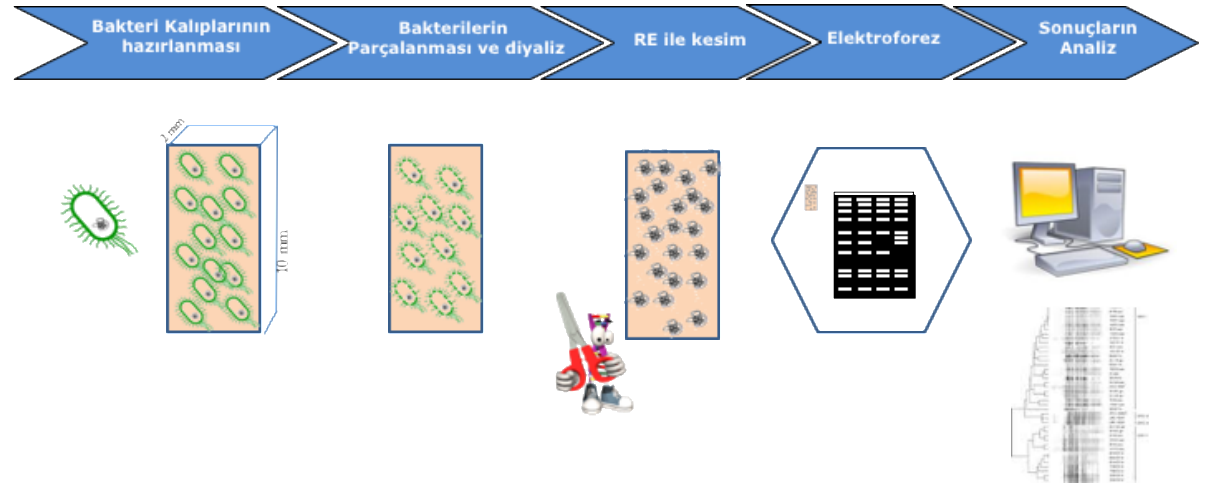
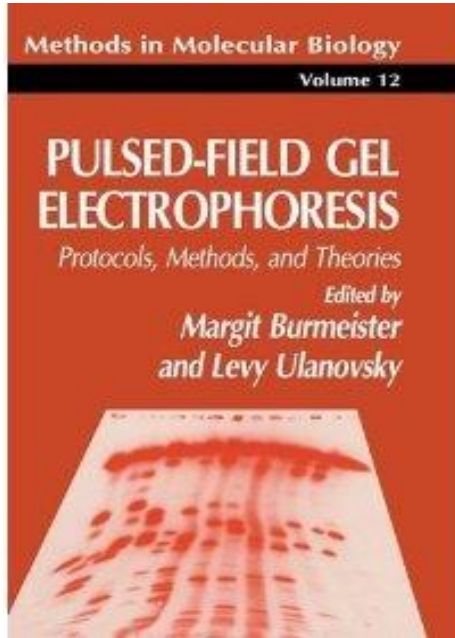
Cryobacterium meningosepticum



Candida albicans

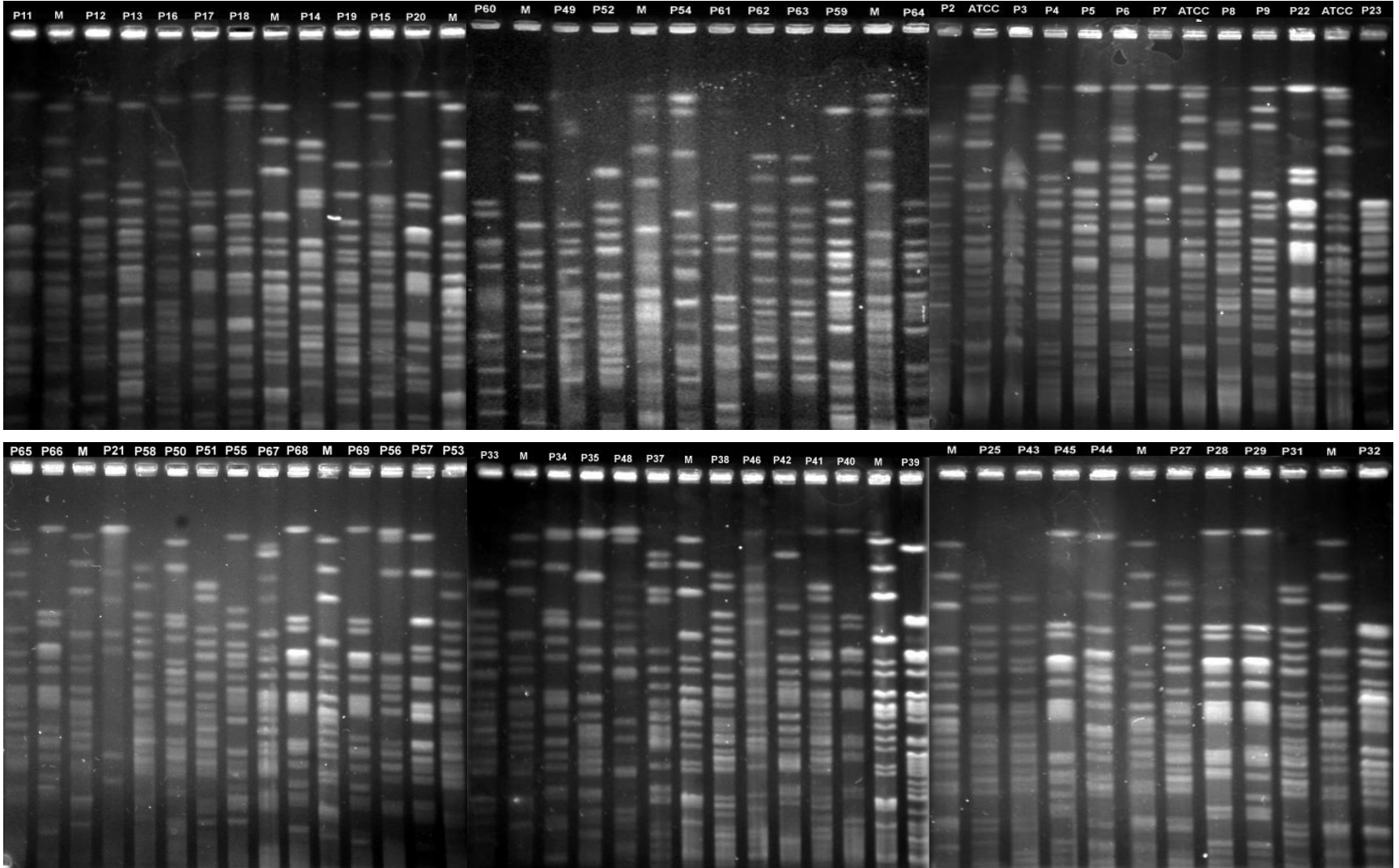
Pulsed Field Gel Electrophoresis (PFGE)

- Bir çok mikroorganizma için halen «**altın standart**» genotiplendirme yöntemidir.



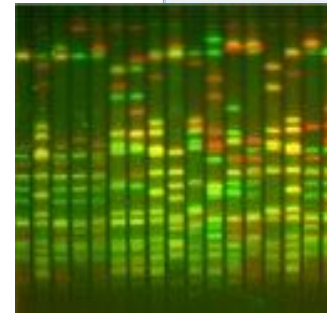
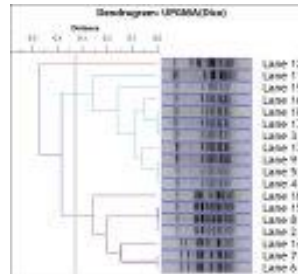
Pulsed Field Gel Electrophoresis (PFGE)

- Sonuçların analizi, benzerlik yüzdesi, bant sayısı ve büyüklüğündeki



Pulsed Field Gel Electrophoresis (PFGE)

- Sonuçların analizi ve yorumlanması
 - Bu amaçla en çok kullanılan bilgisayar programları;
 - Gel-Compar (Applied Maths, Kortrijk, Belgium)
 - Dendron (Solltech, Oakdale, Iowa)
 - Diversity Database Fingerprinting Software (Bio-Rad)
 - Gene Profiler (Scanlytics, Fairfax, Va)
 - Phoretix 1-D (Nonlinear USA, Durham, N.C.)
 - Quantar Pro (KeyGene Products, Wageningen, Netherlands)
 - Taxotron (Taxolab, Institut Pasteur, Paris, France)



Pulsed Field Gel Electrophoresis (PFGE)

- Sonuçların analizi ve yorumlanması



Salgın Araştırmaları

- Retrospektif salgın araştırmaların faydası sınırlıdır.

NEW MICROBIOLOGICA, 31, 401-408, 2008

Infection

Correspondence

Epidemiological Characteristics of Fatal *Candida krusei* Fungemia in Immunocompromised Febrile Neutropenic Children

In early 1990s, fluconazole was introduced as a prophylactic antifungal agent after stem cell transplantation (SCT). At the same time, *Candida krusei* emerged as a chief fungal pathogen among the patients with SCT [1, 2]. In our SCT center, which has a 15-bed capacity unit dedicated for children with malignant diseases and 5-bed unit for SCT patients, low-dose fluconazole is routinely included in the posttransplant prophylaxis regimen (starting at day 7). In recent years, we observed mortality due to fungal sepsis in patients undergoing allogeneic SCT and chemotherapy. Here, we report the characteristics of the patients having mortal *C. krusei* fungemia and the molecular typing results of the *C. krusei* strains in order to develop more effective policies for treatment and prevention of fungal infections.

The febrile neutropenic episodes of the 47 children (median age: 9.33 ± 4.75 years) were prospectively evaluated from March 2001 to January 2003. For each patient, daily leukocyte counts and body temperatures were measured. Fever was defined as a single oral temperature of $\geq 38.3^\circ\text{C}$ or a temperature of $\geq 38.0^\circ\text{C}$ for ≥ 1 h. Neutropenia was defined as a neutrophil count of < 500 cells/mm³, or a count of $< 1,000$ cells/mm³ with a predicted decrease to < 500 cells/mm³ (Guidelines from the Infectious Disease Society of America, 1997). Patients who had both fever and neutropenia were recognized as febrile neutropenic. Nine patients developed severe fungal infection during neutropenia. Three of those having bloodstream infection with the *C. krusei* were analyzed in detail, in this study.

Surveillance cultures were taken from blood, throat, stool, urine and catheter exit sites of the patients in the SCT unit when they had febrile episode and/or weekly on a routine basis. Automated blood culture system (Becton Dickinson, pediatric vials, USA) and BACTEC MYCO/F LYTC (Becton Dickinson) were used for blood cultures. To search exogenous sources of *C. krusei* sepsis, 51 specimens from the hands of 32 health care workers (9 doctors, 11 nurses, 4 laboratory workers, and 8 cleaning staff) and 19 environmental swabs from commonly used areas around the patients (nurse desk, room and toilet doors, refrigerator doors, telephones, etc) were analyzed.

antifungal susceptibility. Molecular typing of the nine *C. krusei* strains of the three patients (three blood, two stool, one urine, one throat, one vagina, and one sputum isolates) was performed by pulsed field gel electrophoresis (PFGE) following the protocol described previously [3].

A total of 156 febrile episodes were recorded in the 47 patients within the 712 febrile neutropenic days. Bacteria were isolated in 28 (52%) and fungi in 26 (48%) of the 54 clinical specimens. It was noted that the patients with prolonged and deep neutropenia (hematological malignancy patients) are under an increased risk for fungal infections [1, 4, 5]. In agreement with these findings, we observed systemic fungal infections with high mortality in recent years. Of the 47 patients with febrile neutropenia, 14 had fungal isolation from various body sites and 9 (19%) died due to fungal infection. Eight of these fatal cases had non-*Candida albicans* infection (Table 1). Distribution of *Candida* spp. among the five non-fatal cases was as follows: *C. krusei* (1), *C. kefyr* (1), *C. albicans* (2), *C. famata* (1). In an Italian tertiary hospital, it was found that 60% of the candidemia episodes were related to non-*albicans* *Candida* and high mortality rates were observed particularly for hematological (77%) and transplant patients (50%) [6]. In another tertiary care hospital in

Infection 2008; 36: 88-91
DOI 10.1007/s10250-007-4246-1

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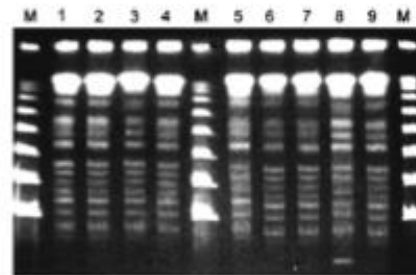
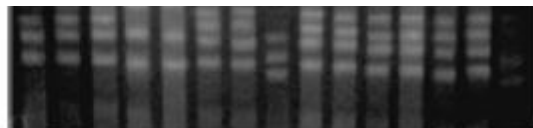


Figure 1. PFGE profile of the 9 *C. krusei* strains. M molecular weight marker. Lanes 1 and 2 patient 1's blood and stool isolates; lanes 3-6, patient 2's throat, stool, blood, and sputum isolates, respectively; lanes 7-9 patient 3's blood, vagina, and throat isolates, respectively.

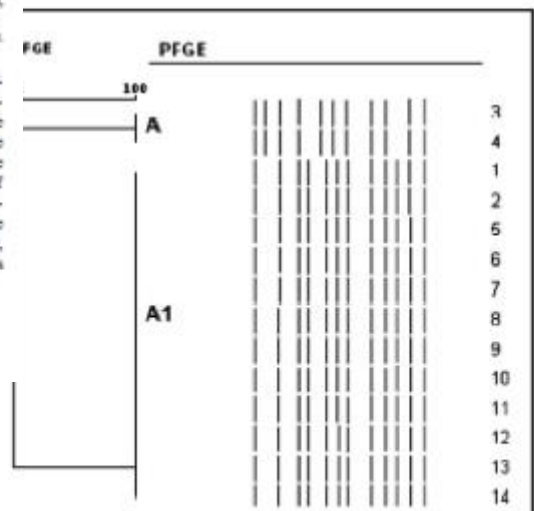
breaks lasting 1 or 2 days or ineffective doses due to difficulties to obtain liposomal amphotericin B. Amphotericin B treatment was started at tenth day of the febrile neutropenia for two patients and at the first day of the febrile neutropenia and sepsis for the third patient. These patients died on days 8, 21, and 23 after the diagnosis of sepsis. Since there were no concomitant bacterial infections in these patients having many risk factors, the mortality was attributed due to *C. krusei* sepsis. However, it is possible that the high mortality in these patients was

ted

CA, 30, 131-137, 2007

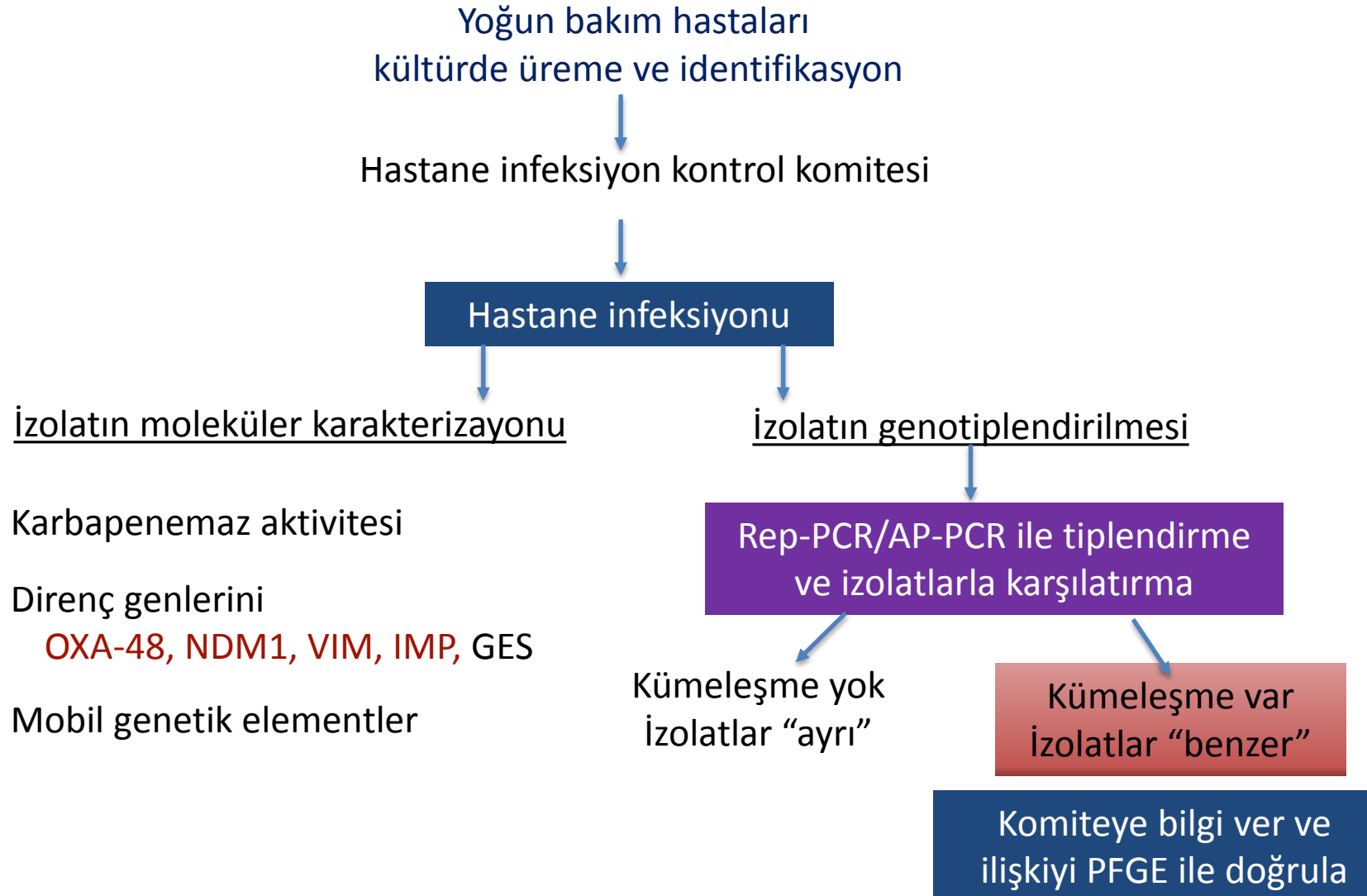
C. aeruginosa
infection
: A pulsed-
ed

AD, Y. Yasemin Ersoy, MD, *



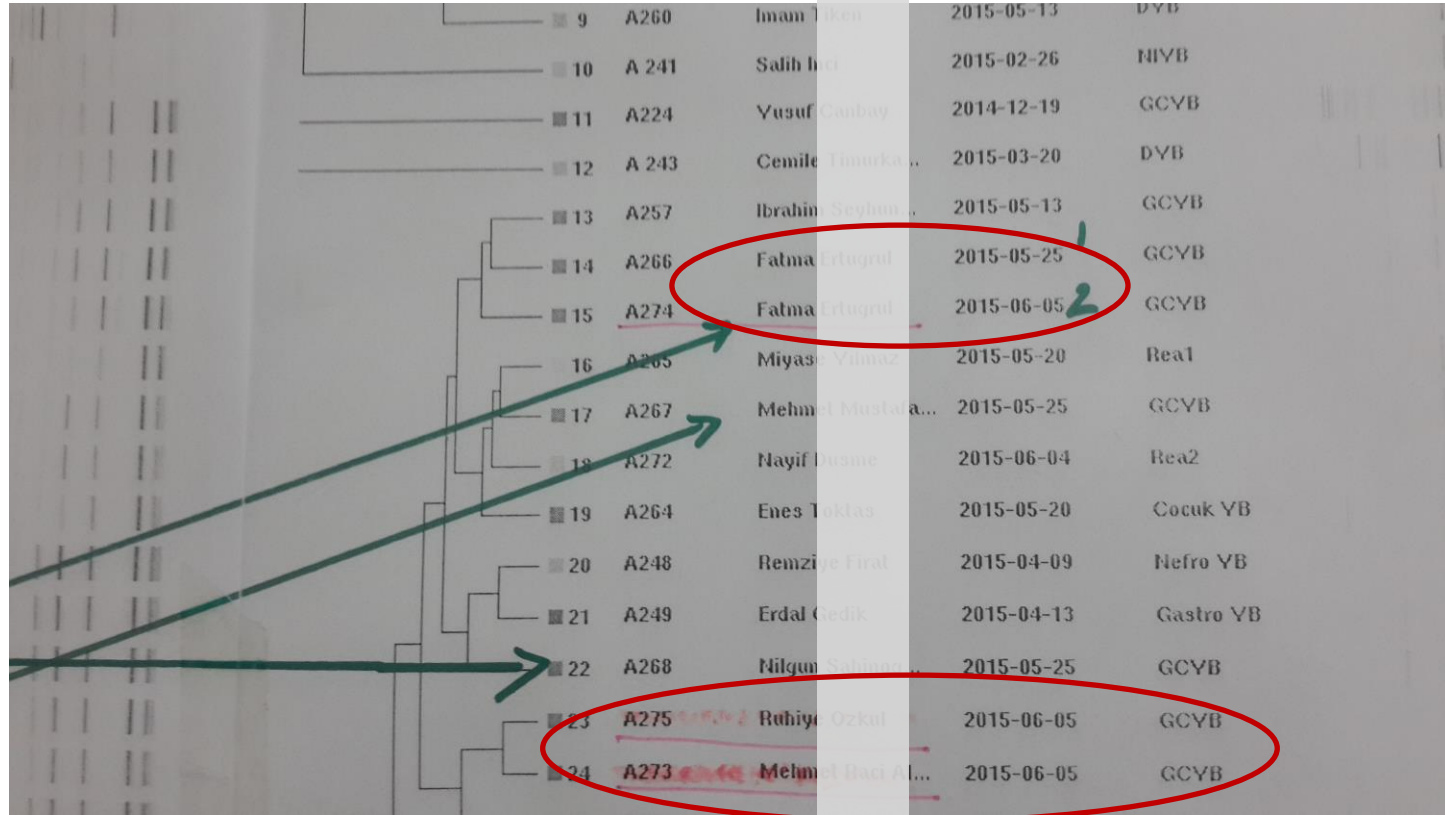
Gerçek Zamanlı Salgın Araştırması

- 2013 yılından itibaren gerçek zamanlı (prospektif) izlenimi



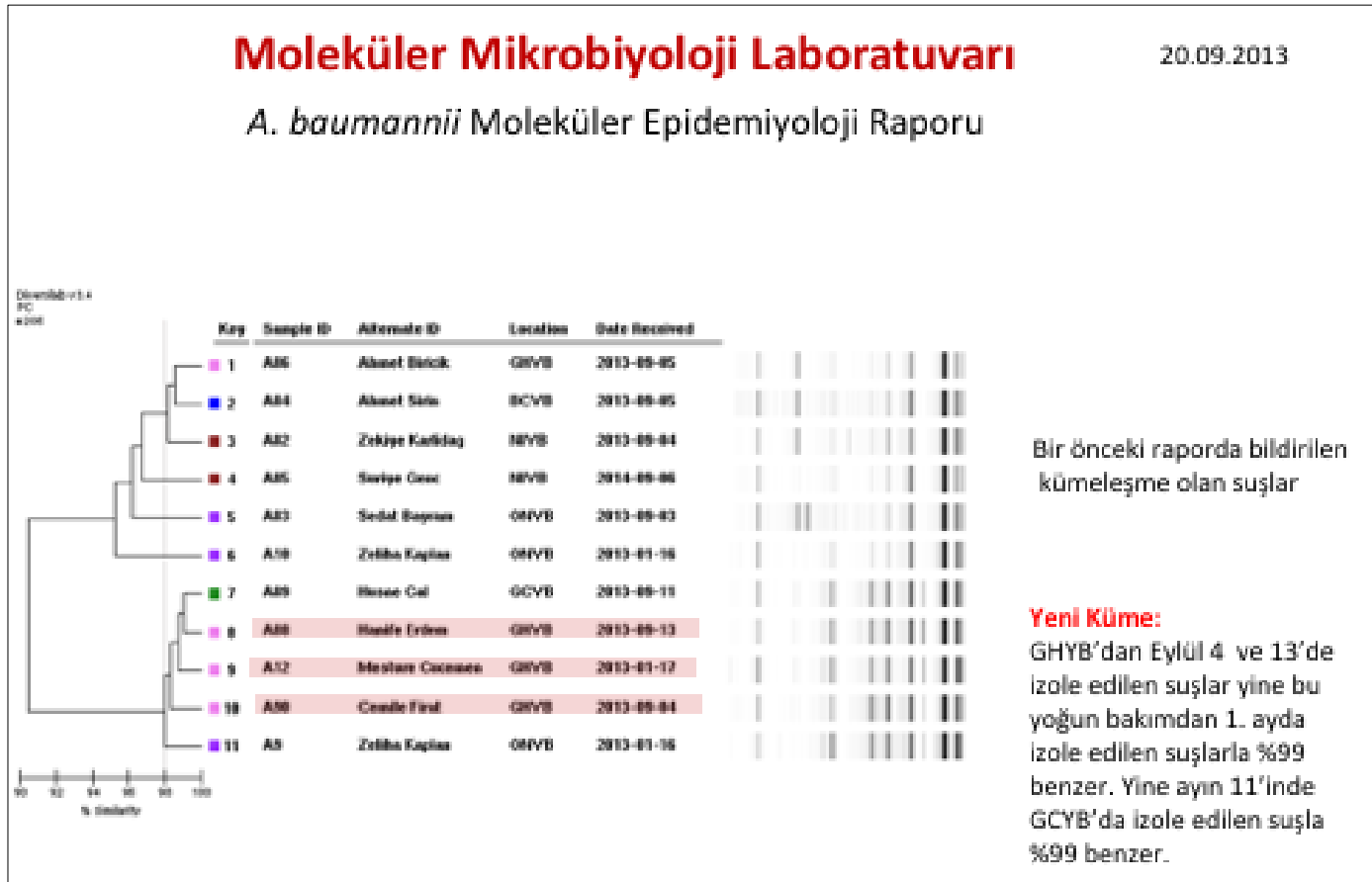
Gerçek Zamanlı Salgın Araştırması

- Hastane infeksiyonlarının gerçek zamanlı (prospektif) izlenimi



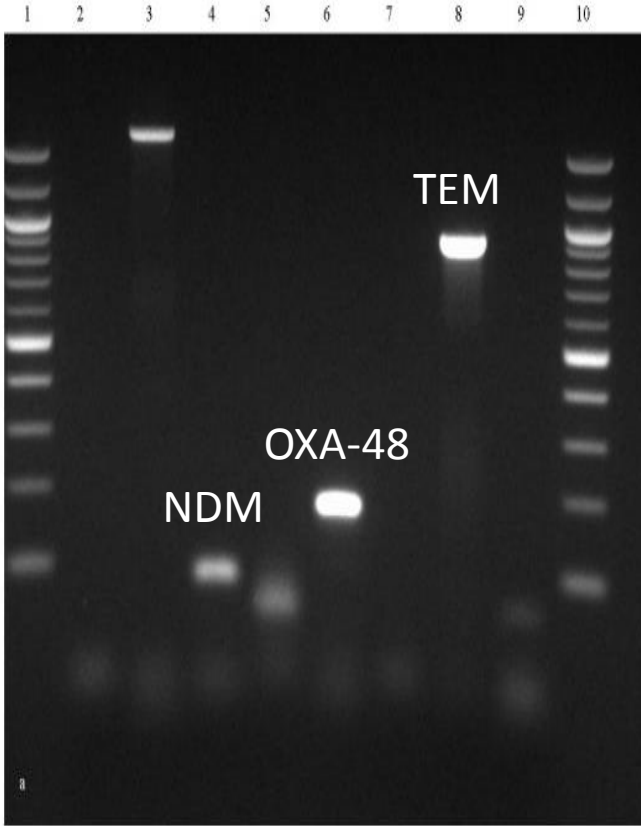
Gerçek Zamanlı Salgın Araştırması

- Hastane infeksiyonlarının **gerçek zamanlı (prospektif)** izlenimi.
- Sonuçları HİKK ne raporladık.



Gerçek Zamanlı Salgın Araştırması

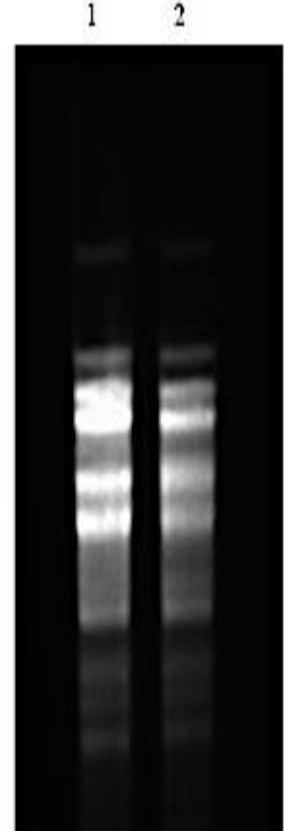
- *Providencia rettgeri*, yanık ünitesinden izole edildi OXA-48 ve NDM-1 karbapenemaz birlikte üretimi



7 ay sonra aynı merkezde bir
izolat daha
aynı direnç paterni
OXA-48 ve NDM-1 birlikteliği.

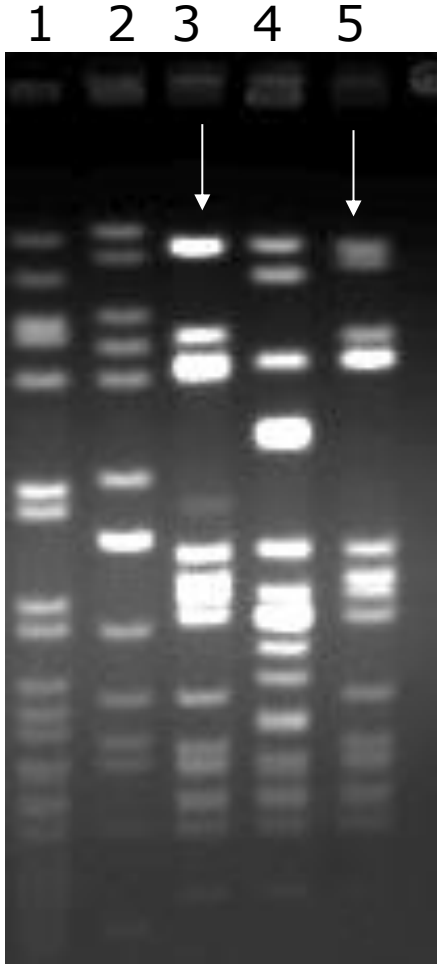


HİKK bilgi verildi
gerekli önlemleri aldı



Gerçek Zamanlı Salgın Araştırması

- 17 Mayıs 2017'de **beş *S. aureus* izolasyonu** oldu. Bir salgın başlangıcı şüphesi ile bu **suşlarda klonal ilişki araştırıldı.**



3- Yanık servisi, yara enfeksiyonu izolatu

5- Ortopedi polikliniği, osteomyelit debridmanı kültürü izolatu

iki hastaya aynı gün plastik cerrahlar tarafından yara debridmanı uygulandığı ortaya çıktı.

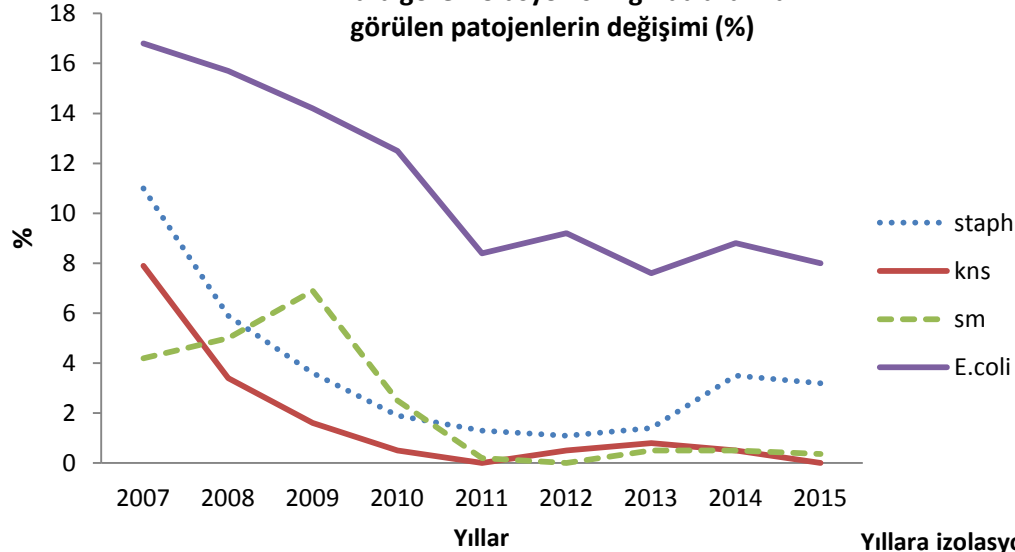


HIKK bilgi verildi
gerekli önlemleri aldı

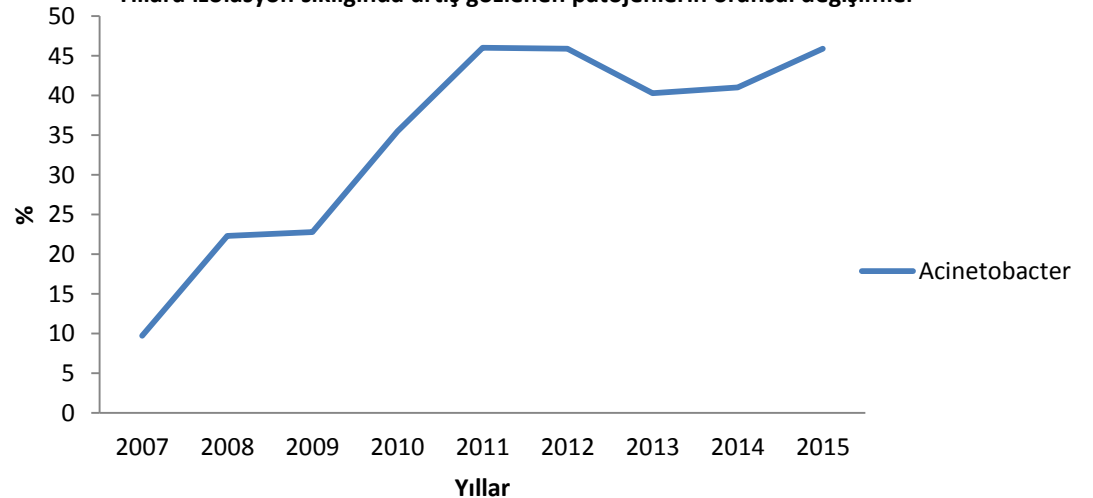
Bir sorun var?

- HİKK alınan tüm önlemlere rağmen *A. baumannii* önlenemiyor.

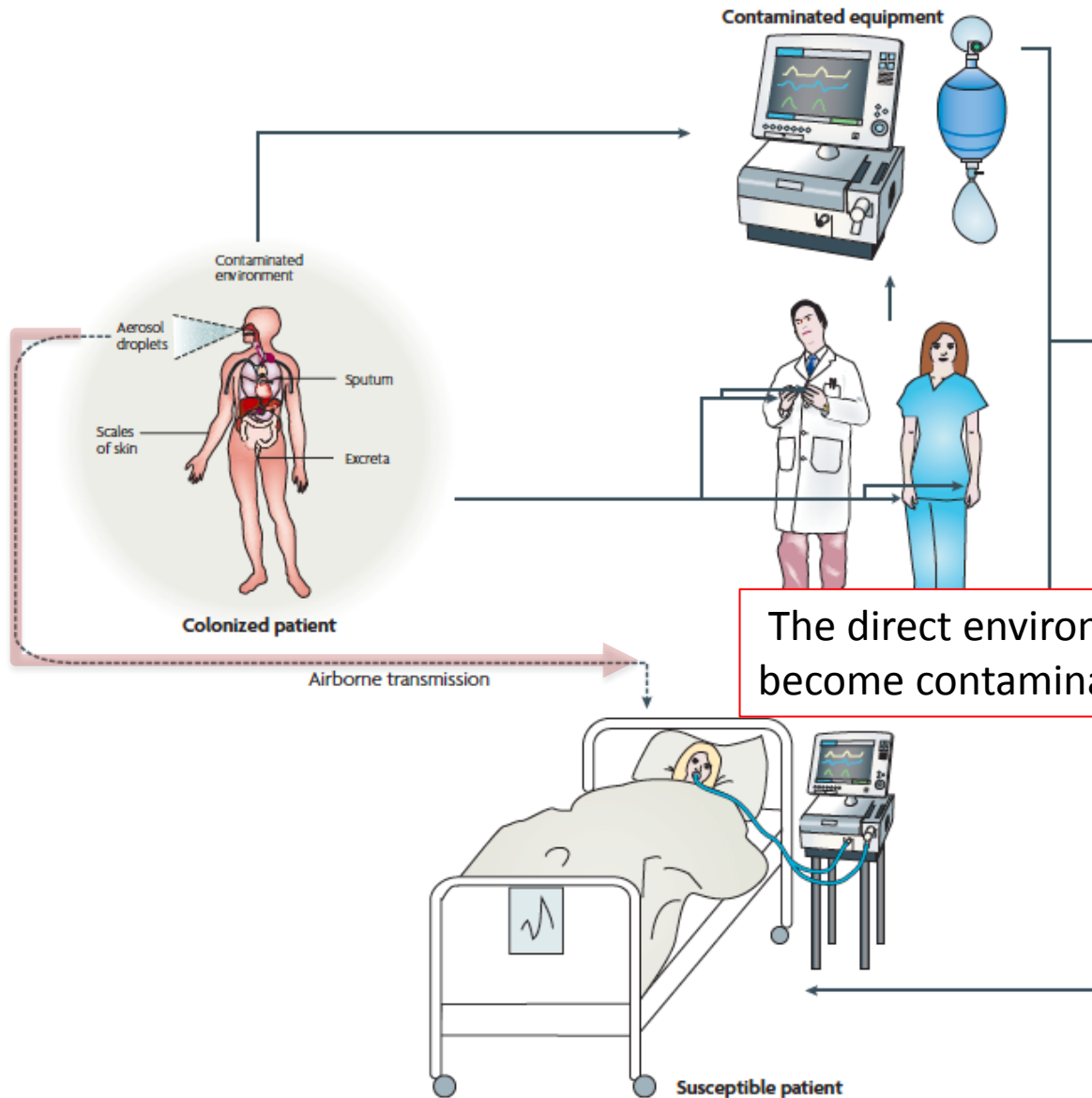
Yıllara göre izolasyon sıklığında azalma
görülen patojenlerin değişimi (%)



Yıllara izolasyon sıklığında artış gözlenen patojenlerin oransal değişimler



A. baumannii ile kontamine hastane çevresi



A. baumannii hava yolu ile bulaşır mı?

Aerosolization of *Acinetobacter baumannii* in a Trauma ICU*

L. Silvia Munoz-Price, MD^{1,2,4}; Yovanit Fajardo-Aquino, MD⁴; Kristopher L. Arheart, EdD^{2,5}; Timothy Cleary, PhD⁶; Dennise T. Brown, PhD⁷; Robert A. Archer, PhD⁸; Robert A. Archer, PhD⁸; Jesabel I. Rivera, BS⁷; Jessica A.

INFECTION CONTROL AND HOSPITAL EPIDEMIOLOGY APRIL 2006, VOL. 27, NO. 4

ORIGINAL ARTICLE

Epidemiology of *Acinetobacter baumannii* in a University Hospital in Turkey

Halis Akalin, MD; Cüneyt Özakin, MD; Suna Gedikoğlu, MD

Objective: To establish the presence of air cor
netobacter baumannii in the trauma ICU.

Design: Point prevalence microbiological surv

Settings: A 1,500-bed public teaching hospita
area.

Patients: Trauma ICU patients.

Measurements: Pulsed field electrophoresis
environmental and clinical isolates to determin
any isolates from the air with clinical isolates.

Main Results: Out of 53 patient areas cultur
their air positive for *A. baumannii*. The presen
nii-positive patient (underneath the plate) v
positive air cultures for *A. baumannii* (11 of 21
[0%]; $p < 0.0001$). However, we were not abl
in air contamination based on the presence

OBJECTIVE. Molecular epidemiologic surveillance of *Acinetobacter baumannii* by polymerase chain reaction-randomly amplified polymorphic DNA analysis in a university hospital for 3 consecutive study periods.

RESULTS. Twelve different *Acinetobacter baumannii* genotypes (A-L) were detected. Although only 2 genotypes were detected during the first period and genotype A appeared to be the most common genotype, genotype D was included in these genotypes during the second study period. Genotype A completely disappeared during the third period. Although the presence of genotype C and the genotype D continued during the third period, 9 new genotypes were detected during this period. Genotype A appeared to be the most common genotype in the hospital (detected in 19 different clinics). The distribution of genotypes in clinical samples correlated with patient traffic between them. Some genotypes were found in both clinical and environmental samples. Seventeen different antibiotypes were detected, according to antibiotic susceptibility profiles.

CONCLUSIONS. Environmental contamination, airborne transmission, patient transfer, and cross-contamination play important roles in epidemics caused by *A. baumannii* in our hospital. The distribution of genotypes can change over time, so antibiotyping is not appropriate for the epidemiological analysis of *A. baumannii* infection.

Infect Control Hosp Epidemiol 2006; 27:404-408

A. baumannii hava yolu ile bulaşır mı?



Contents lists available at [ScienceDirect](#)

American Journal of Infection Control

journal homepage: www.ajicjournal.org



Major Article

Is airborne transmission of *Acinetobacter baumannii* possible: A prospective molecular epidemiologic study in a tertiary care hospital



Yusuf Yakupogullari MD ^a, Baris Otlu PhD ^{a,*}, Yasemin Ersoy MD ^b, Cigdem Kuzucu MD ^a,
Yasar Bayindir MD ^b, Uner Kayabas MD ^b, Turkan Togal MD ^c, Canan Kizilkaya PhD ^d

Table 2
Genetic relationships of air and clinical isolates

ID	Air Sampling Area	ICU	Genotype No	S-Date	DNA Band Pattern		S-Date	Relationship
					Air Isolates	Patient Isolate		
1	Patient-A-1m	II	1	3/9/2013			3/8/2013	Clonal relation with Patient-A's clinical isolate (DL>95%)
2	ICU center							
3	Patient-C Bs-1m	2	2	3/19/2013			1/23/2013	Clonal relation with Patient-B's clinical isolate (DL>96%)
4	Patient-C Bs-2m							
5	Patient-C Bs-3m	I	2a	3/19/2013			3/17/2013	Clonal relation with Patient-C's clinical isolate (DL>99%)
6	ICU center							
7	Nurse desk	II	3	3/20/2013			3/19/2013	Clonal relation with Patient-D's clinical isolate (DL>98%)
8	Storage corridor							
9	ICU entrance	II	4	4/19/2013			4/17/2013	Clonal relation with Patient-E's clinical isolate (DL>99%)
10	Patient-D Bs-3m							
11	Nurse desk	II	5	5/20/2013		None		No clonal relation
12	Patient-E Bs-2m							
13	Patient-E Bs-1m	II	5a	5/20/2013		None		No clonal relation
14	ICU center							
15	Nurse desk	I	6	6/20/2013			7/10/2013	Clonal relation with Patient-F's clinical isolate (in ICU-I; DL>98%)
16	ICU center							
17	ICU entrance	II	7/C-S	6/24/2013		None	10/1/2013	Clonal relation with Patient-G's clinical isolate (in ICU-II; DL>97%)
18	Nurse desk	I	8	7/10/2013		None		No clonal relation
19	ICU center	I	9	8/7/2013		None		No clonal relation
20	ICU entrance	I	9a	8/7/2013		None		No clonal relation
21	Patient-H Bs-1m	I	10	9/3/2013			8/31/2013	Clonal relation with Patient-H's clinical sample (DL>99%)
22	Patient-H Bs-2m							
23	Nurse desk	II	11	9/11/2013		None		No clonal relation
24	ICU center							
25	Storage room	II	12/C-S	11/20/2013		None		No clonal relation
26	Patient-I Bs-1m	II	13	11/20/2013			11/18/2013	Clonal relation with Patient-I's clinical sample (DL>96%)

NOTE. Similarity cut off value ≥95% clonally related strains, 95%-97% subgroup, ≥98%-100% same genotype, and <95% no relation. Bs, bedside; C-S, carbapenem-susceptible; DL, DL pattern; S-Date, sampling date.

MALDI-TOFF ile Moleküler Epidemiyoloji

- Kütle spektrometresi ile **tanı gücünün yanın da klonal ilişki analizi.**



Journal of
Proteomics & Bioinformatics

Castañero et al., J Proteomics Bioinform 2016, 9:9
DOI: 10.4172/jpb.1000411

Review Article

Open Access

Is MALDI-TOF Mass Spectrometry a Valuable New Tool for Microorganisms Epidemiology?

Silvia Vega Castañero¹, Fernando Sánchez Juanes², Laura Ferreira³, Jose Manuel González Buitrago^{2,4,5} and Juan Luis Muñoz Bellido^{2,6-8*}

¹Microbiology Unit, Asistencial Complex Segovia, Spain

²Institute of Biomedical Research Salamanca (IBSAL-CSIC), Spain

³MSD Sharp & Dohme GmbH, Salamanca, Spain

⁴Clinical Analysis, Salamanca University Welfare Complex, Spain

⁵Department of Biochemistry and Molecular Biology, University of Salamanca, Spain

⁶Microbiology Unit, Salamanca University Welfare Complex, Spain

⁷Department of Biomedical Sciences and Diagnostics, University of Salamanca, Spain

⁸Consolidated Research Unit Infectious and Tropical Diseases, University of Salamanca, Spain

Abstract

Matrix-assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF MS) is now accepted as a fast and reliable method for bacterial identification. It has been shown a very accurate method for identifying bacteria and yeast and moulds from culture, but also from blood cultures and directly from some samples, such as urine. Some authors have proposed the application of the protein profiles obtained by MALDI-TOF MS for bacterial typing and hierarchical clustering with epidemiological purposes.

Here we revise and discuss the information published in the last years on the possible usefulness of MALDI-TOF MS for this purpose, the approaches proposed for different microorganisms and the future perspectives of this technology in the area of infectious diseases epidemiology.

MALDI-TOFF ile Moleküler Epidemiyoloji

RESEARCH ARTICLE

Matrix-Assisted Laser Desorption/Ionization Time of Flight Mass-Spectrometry (MALDI-TOF MS) Based Typing of Extended-Spectrum β -Lactamase Producing *E. coli* – A Novel Tool for Real-Time Outbreak Investigation

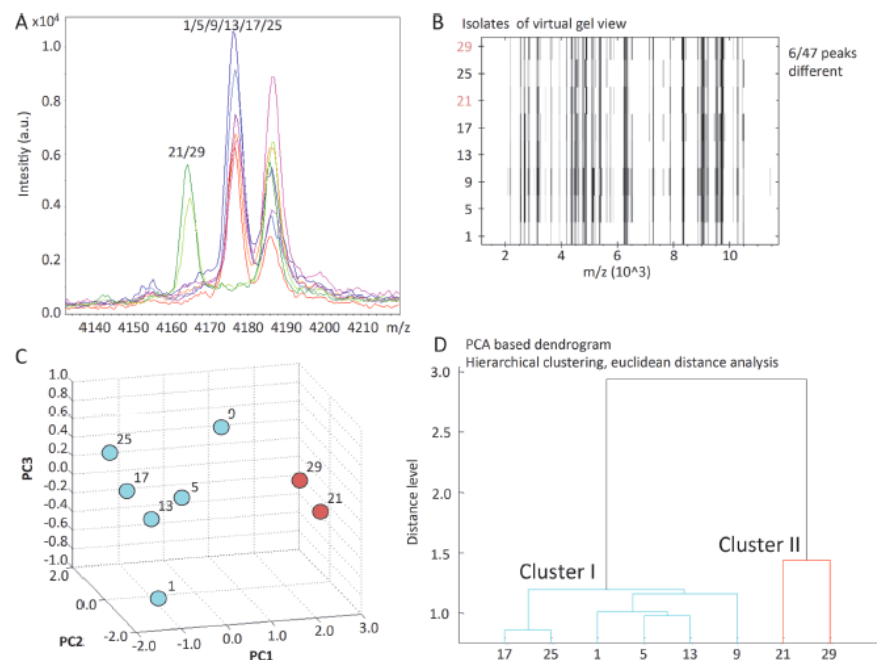
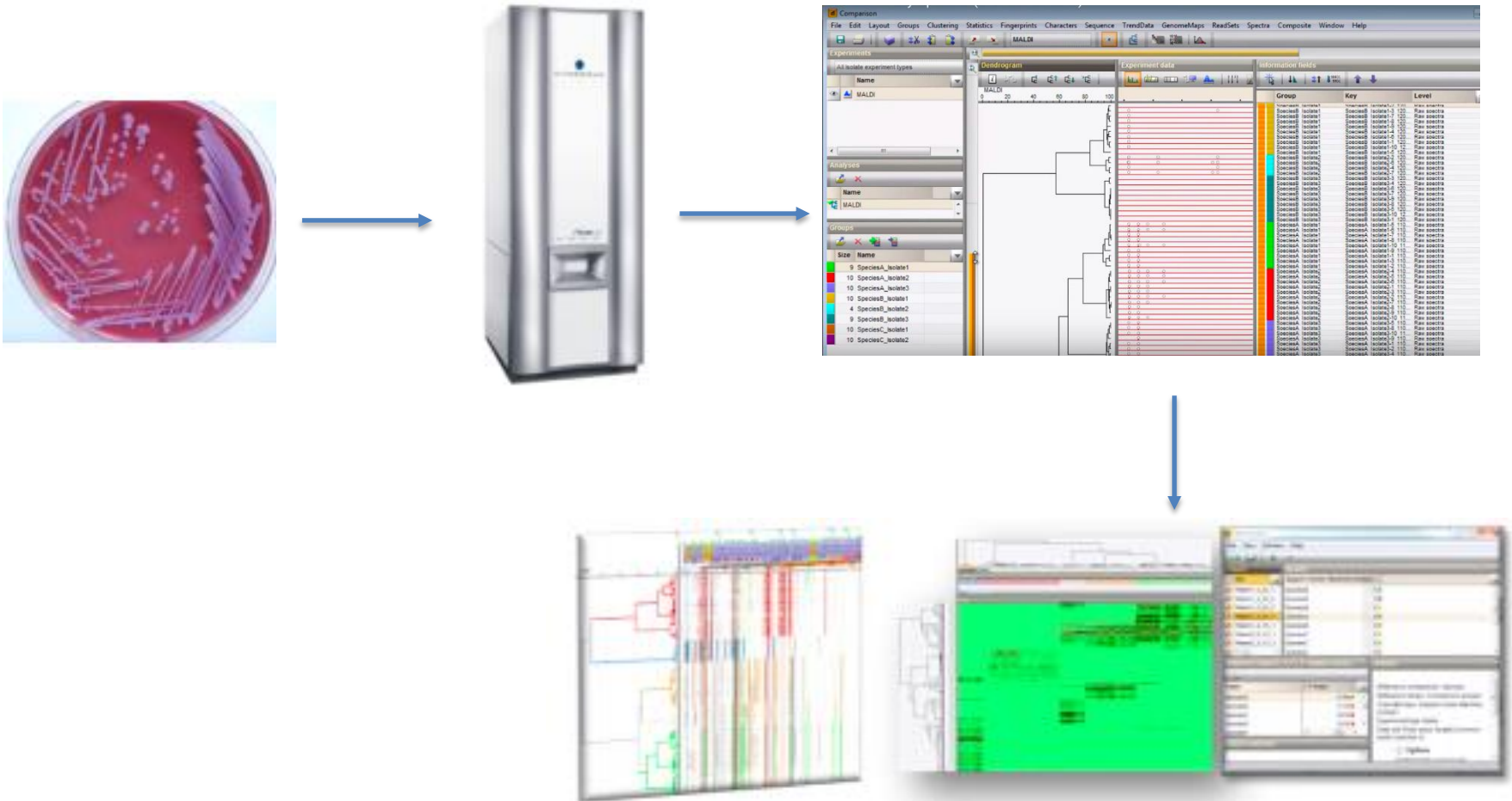


Fig 1. MALDI-TOF MS based typing. A. Representative example of mass spectrum differences between ESBL *E. coli*. Isolates are indicated by color code. Isolates 1, 5, 9, 13, 17 and 25 (outbreak cluster) show a clear distinguished peak at position 4175 m/z, whereas isolates 21 and 29 show a shift to position 4165 m/z (non-related cluster). This corresponds to change of about 10 Da. Various other areas of peak shifts have been identified between outbreak and non-related ESBL *E. coli* clusters. B. MALDI-TOF MS based virtual gel view. MS generated peaks of the protein expression profile are shown for every bacterial isolate. Red labels highlight the non-related isolates. C. Three-dimensional principal component analysis. Based on the Euclidean distance analysis algorithm a three dimensional plot was generated. PC1 and PC2 showed the highest discriminatory potential and indicated a cluster for the outbreak associated isolates, whereas the non-related isolates are less similar. D. PCA-based dendrogram generated by MALDI-TOF MS. The PCA based dendrogram uses a hierarchical clustering algorithm with a Euclidean distance analysis.

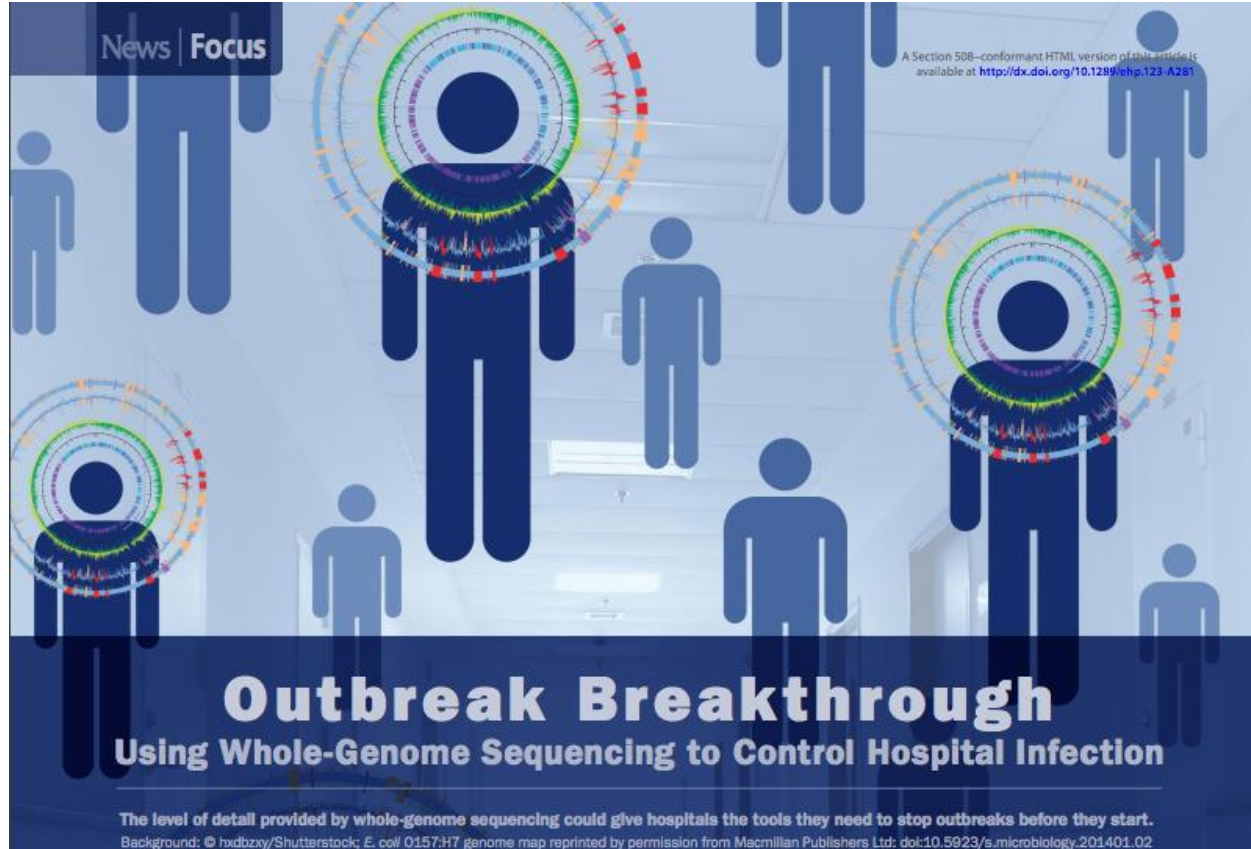
MALDI-TOFF ile Moleküler Epidemiyoloji

- Günlük bakteri tanımlamasını takiben, klonal analizi.



Yeni Nesil Dizileme Sistemleri

- YNS dizileme sistemlerinin **ucuzlaması ve kullanımlarının kolaylaşması**

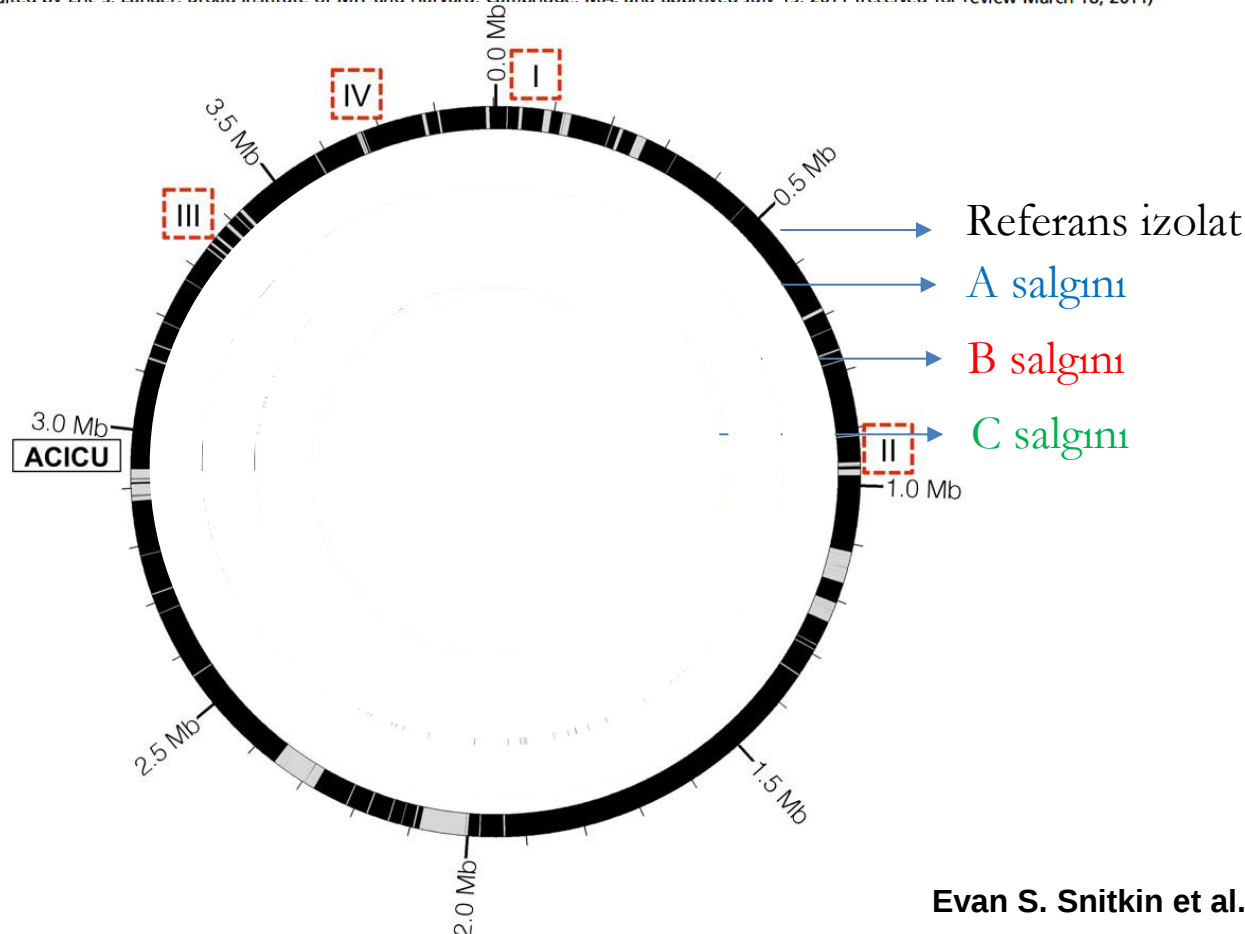


Genome-wide recombination drives diversification of epidemic strains of *Acinetobacter baumannii*

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Broad Active Surveillance Enabled by Pathogenica's HAI BioDetection Kit

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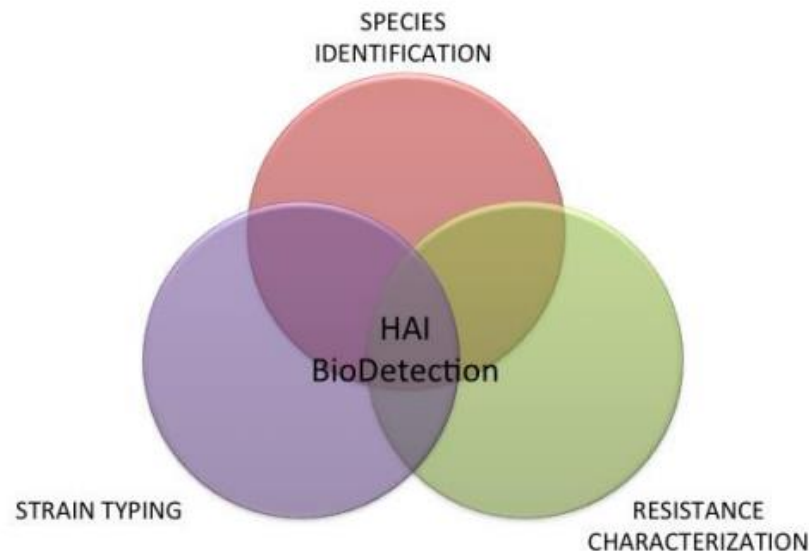


Figure 1: In addition to active surveillance, HAI BioDetection is suitable for strain-typing and antimicrobial resistance characterization.

RESEARCH HIGHLIGHT

Stopping outbreaks with real-time genomic epidemiology

Patrick Tang^{1,2*} and Jennifer L Gardy^{1,3}

Abstract

One of the most successful public health applications of next-generation sequencing is whole-genome sequencing of pathogens to not only detect and characterize outbreaks, but also to inform outbreak management. Using genomics, infection control teams can now track, with extraordinarily high resolution, the transmission events within outbreaks, opening up possibilities for targeted interventions. These successes are positioning the emerging field of genomic epidemiology to replace traditional molecular epidemiology, and increasing our ability to limit the spread of multidrug-resistant organisms.

introduction and onward transmission of HAIs in their settings. While surveillance and screening, in combination with molecular genotyping, can indicate the presence of a nosocomial outbreak, conventional molecular epidemiology methods lack sufficient resolution to reveal the origins and transmission dynamics of these outbreaks - information integral to implementing appropriate and effective infection control strategies.

Over the past few decades, a series of molecular epidemiology methods, including pulsed field gel electrophoresis and multi-locus sequence typing, have been developed to estimate phylogenetic relationships between bacterial isolates - each one trying to improve upon the speed, accuracy, reproducibility, ease of use or discriminatory power of previous methods. However, the introduction

Eldeki tüm imkanlar kullanılmalı

Diagnostic Report: *Acinetobacter baumannii*

Isolate number	113	Date isolated	2012-09-04
Origin	Throat swab	Pat. ID	XXXX-XXGiXXX

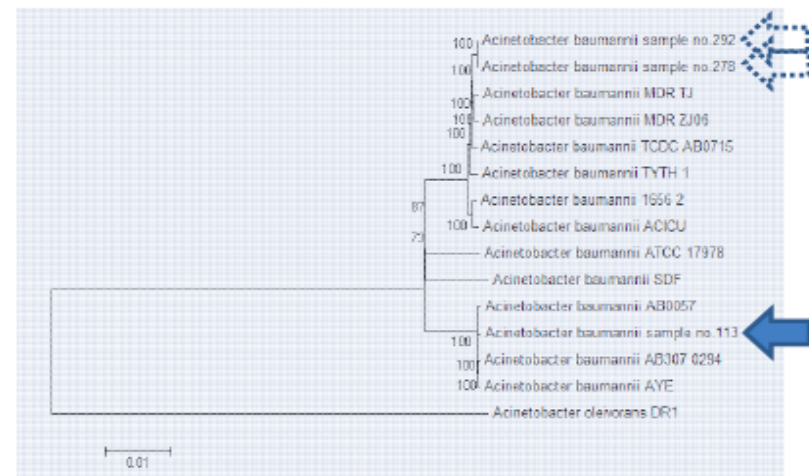
MLST type

1

Phylogeny

Resistance genes	Antibiotic group	Location
<i>bla</i> _{oxa-72}	Beta-Lactam	Plasmid 1
<i>bla</i> _{oxa-66}	Beta-Lactam	Chromosome
<i>aadA1</i>	Aminoglycoside	Chromosome
<i>sul1</i>	Sulfonamid	Chromosome

Virulence genes detected	Location
Type IV secretion system	Plasmid 2



Assessment

CAVE: Strain with two carbapenemase genes detected. Classified as **multiresistant**. Phylogeny relation traced to known outbreak strain AB0057. No significant relation to other clinical isolates in recent time frame.

Hygiene: Single patient isolation recommended.

Treatment options: Tetracyclin, Tigecyclin, Colistin (inhalative)

Neler yaptık?

- Hastane infeksiyon kontrol komitesine **anlık bilgi verdik** ve **haftalık toplantılarına katıldık**.
- Hastane infeksiyon kontrol komitesinin **elini güçlendirdik**.



Ne yapmak gerekiyor ?

- Hastane infeksiyonları,
sık görülen komplikasyonlar olduğundan bu enfeksiyonlar sürekli olarak takip edilmeli ve salgınların önlenmesi için stratejiler geliştirilmelidir.

