

Bakterie z grupy „HACEK”- chorobotwórczość i diagnostyka

Dorota Olszańska

Zakład Diagnostyki Mikrobiologicznej i Immunologii Infekcyjnej

Kierownik: Prof. dr hab. n. med. Elżbieta Tryniszewska

Uniwersytecki Szpital Kliniczny w Białymstoku



Bakterie z grupy „HACEK”

- **H** – *Haemophilus parainfluenzae*
- **A** – *Aggregatibacter actinomycetemcomitans*, *A. aphrophilus*, *A. paraaphrophilus*, *A. segnis*
- **C** – *Cardiobacterium hominis*, *C. vulvarum*
- **E** – *Eikenella corrodens*
- **K** – *Kingella kingae*, *K. denitrificans*



Bakterie z grupy „HACEK” - występowanie

- ▣ Mikrobiota fizjologiczna górnych dróg oddechowych
 - ▣ Śluzówki jamy ustnej
 - ▣ Dziaśła
 - ▣ Kamień nazębny
- ▣ mikrobiota przewodu pokarmowego
- ▣ Mikrobiota dróg moczowo-płciowych



Chorobotwórczość grupy „HACEK”

- ▣ bakteriemie
- ▣ Infekcyjne zapalenie wsierdzia 1-3% przypadków
- ▣ Agresywne zapalenie przyzębia (*A. actinomycetemcomitans*)
- ▣ Infekcje skóry
- ▣ Zapalenie kości i stawów
- ▣ Zapalenie nerwu wzrokowego
- ▣ Infekcje wewnątrzbrzuszne
- ▣ Ropnie otrzewnowe
- ▣ Ropnie mózgu (szczególnie *A. aphrophilus*)

Haemophilus parainfluenzae

- ▮ Mikrobiota fizjologiczna jamy ustnej i górnych dróg oddechowych
- ▮ rzadka przyczyna etiologiczna :
 - ▮ Infekcji górnych dróg oddechowych
 - ▮ Bakteriemii
 - ▮ bakteryjnego zapalenia wsierdza
- ▮ Bakterie wymagające pokarmowo , potrzebują czynnika V
- ▮ Hodowla na podłożach czekoladowych w 5 % CO₂ , 24 godziny
- ▮ Identyfikacja na karcie VITEK NH, MALDI-TOF
- ▮ Antybiogramy wg rekomendacji EUCAST dla H. influenzae

Haemophilus influenzae

Expert Rules and Intrinsic Resistance Tables

EUCAST Clinical Breakpoint Tables v. 9.0, valid from 2019-01-01

EUCAST breakpoints have been defined for *H. influenzae* only. Clinical data for other *Haemophilus* species are scarce. MIC distributions for *H. parainfluenzae* are similar to those for *H. influenzae*. In the absence of specific breakpoints, the *H. influenzae* MIC breakpoints can be applied to *H. parainfluenzae*.

MIC determination (broth microdilution according to ISO standard 20776-1)

Medium: Mueller-Hinton broth + 5% lysed horse blood and 20 mg/L β-NAD (MH-F broth)

Inoculum: 5x10⁵ CFU/mL

Incubation: Sealed panels, air, 35±1°C, 18±2h

Reading: Unless otherwise stated, read MICs at the lowest concentration of the agent that completely inhibits visible growth.

Quality control: *Haemophilus influenzae* ATCC 49766. For agents not covered by this strain and for control of the inhibitor component of beta-lactam inhibitor combinations, see EUCAST QC Tables.

Disk diffusion (EUCAST standardised disk diffusion method)

Medium: Mueller-Hinton agar + 5% defibrinated horse blood and 20 mg/L β-NAD (MH-F)

Inoculum: McFarland 0.5

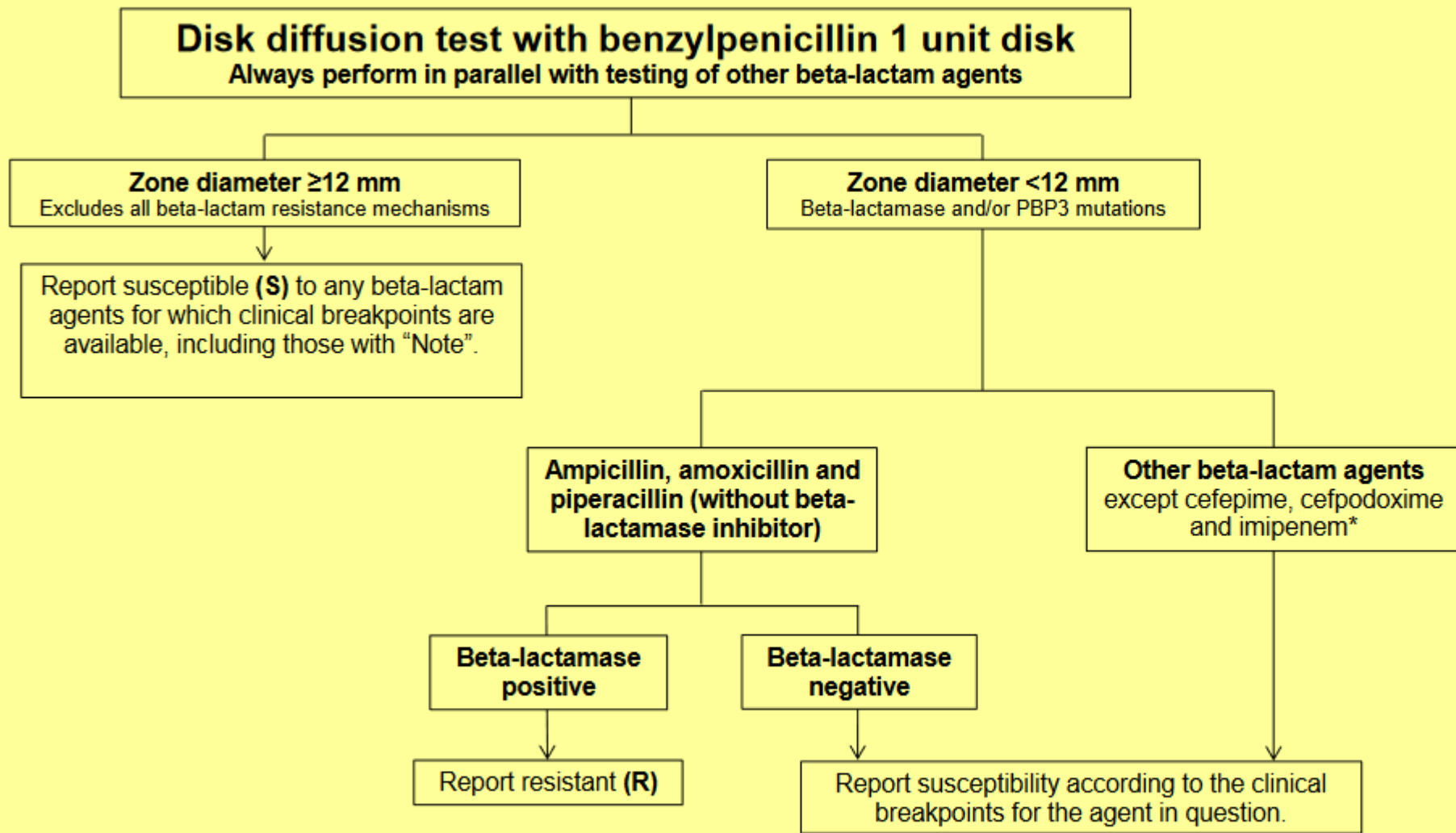
Incubation: 5% CO₂, 35±1°C, 18±2h

Reading: Unless otherwise stated, read zone edges as the point showing no growth viewed from the front of the plate with the lid removed and with reflected light.

Quality control: *Haemophilus influenzae* ATCC 49766. For agents not covered by this strain and for control of the inhibitor component of beta-lactam inhibitor-combination disks, see EUCAST QC Tables.

Penicillins ¹	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Benzylpenicillin	IE	IE			IE	IE		<p>Numbered notes relate to general comments and/or MIC breakpoints. Lettered notes relate to the disk diffusion method.</p> <p>1/A. The benzylpenicillin 1 unit disk screen test shall be used to exclude beta-lactam resistance mechanisms. When the screen is negative (inhibition zone ≥12 mm) all beta-lactam agents for which clinical breakpoints are available, including those with "Note", can be reported susceptible without further testing. When the screen is positive (inhibition zone <12 mm), see flow chart below for interpretation.</p> <p>1. Breakpoints are based on intravenous administration.</p> <p>2. Beta-lactamase positive isolates can be reported resistant to ampicillin, amoxicillin and piperacillin without inhibitors. Tests based on a chromogenic cephalosporin can be used to detect the beta-lactamase.</p> <p>3. For susceptibility testing purposes, the concentration of sulbactam is fixed at 4 mg/L.</p> <p>4/C. Susceptibility can be inferred from amoxicillin-clavulanic acid.</p> <p>5. For susceptibility testing purposes, the concentration of clavulanic acid is fixed at 2 mg/L.</p> <p>6. For susceptibility testing purposes, the concentration of tazobactam is fixed at 4 mg/L.</p> <p>7/E. Susceptibility inferred from ampicillin or amoxicillin.</p> <p>B. ATU relevant only if the benzylpenicillin 1 unit disk screen is positive (inhibition zone <12 mm).</p> <p>D. Susceptibility can be inferred from ampicillin.</p>
Benzylpenicillin (screen) ¹	NA	NA		1 unit	12 ^A	Note ^A		
Ampicillin ²	1	1		2	16 ^A	16 ^A	16-19 ^B	
Ampicillin-sulbactam	1 ^{3,4}	1 ^{3,4}		10-10	Note ^{A,C}	Note ^{A,C}		
Amoxicillin iv ²	2	2			Note ^{A,D}	Note ^{A,D}		
Amoxicillin oral ^{2, HE}	2	2			Note ^{A,D}	Note ^{A,D}		
Amoxicillin-clavulanic acid iv	2 ⁵	2 ⁵		2-1	15 ^A	15 ^A	14-16 ^B	
Amoxicillin-clavulanic acid oral ^{HE}	2 ⁵	2 ⁵		2-1	15 ^A	15 ^A	14-16 ^B	
Piperacillin ²	IE	IE			IE	IE		
Piperacillin-tazobactam	0.25 ⁵	0.25 ⁵		30-6	27 ^A	27 ^A	24-27 ^B	
Ticarcillin	IE	IE			IE	IE		
Ticarcillin-clavulanic acid	IE	IE			IE	IE		
Temocillin	IE	IE			IE	IE		
Phenoxyethylpenicillin	IE	IE			IE	IE		
Oxacillin	-	-			-	-		
Cloxacillin	-	-			-	-		
Dicloxacillin	-	-			-	-		
Flucloxacillin	-	-			-	-		
Mecillinam (uncomplicated UTI only)	-	-			-	-		

Screening for beta-lactam resistance in *H. influenzae*



*For cefepime, cefpodoxime and imipenem, if resistant by both screen and agent disk diffusion test, report resistant. If resistant by screen test and susceptible by agent disk diffusion test, determine the MIC of the agent and interpret according to breakpoints.

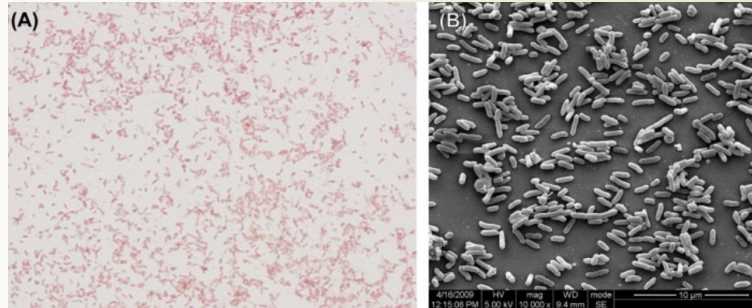


Aggregatibacter actinomycetemcomitans, aphrophilus, paraaphrophilus, segnis

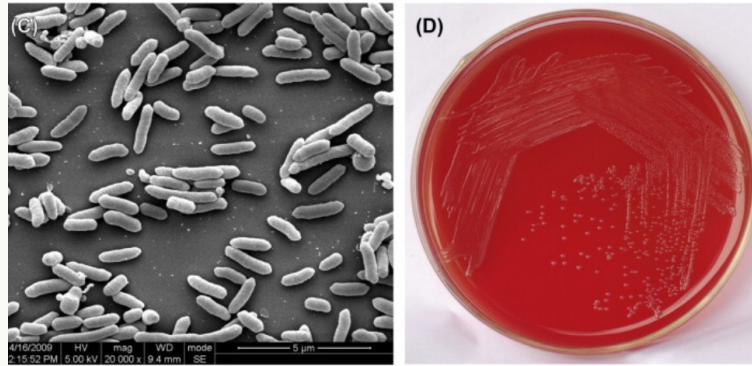
- ▣ Rodzaj utworzony w 2006 roku należy do Rodziny Pasteurellaceae
- ▣ Względnie beztlenowe Gram (-) ziarniako-pałeczki, oksydazo i katalazo dodatnie
- ▣ Wolno rosną wymagają ponad 7 dni hodowli
- ▣ Hodowla na podłożach czekoladowych i agarze krwawym BHI w 5% CO₂, 37°C
- ▣ Identyfikacja karta Vitek NH, Maldi-TOF

Aggregatibacter actinomycetemcomitans

A- Preparat grama

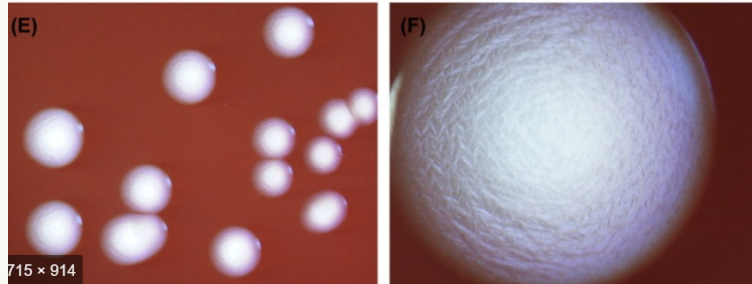


B / C- komórki w mikroskopie SEM



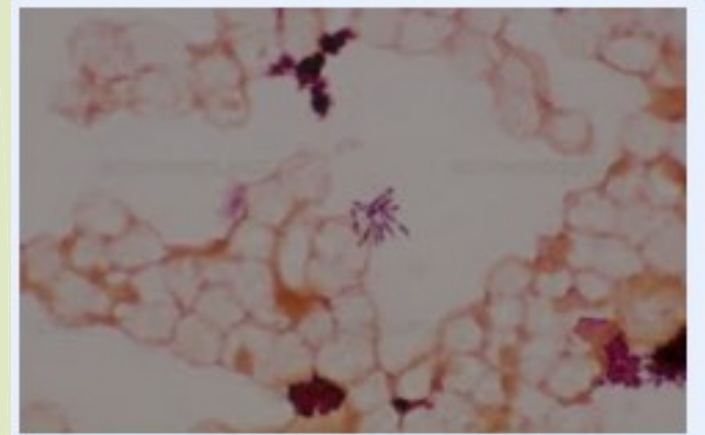
D - Kolonie na agarze krwawym BHI

E/F - kolonie stereomikroskop



Cardiobacterium hominis, *C. valvarum*

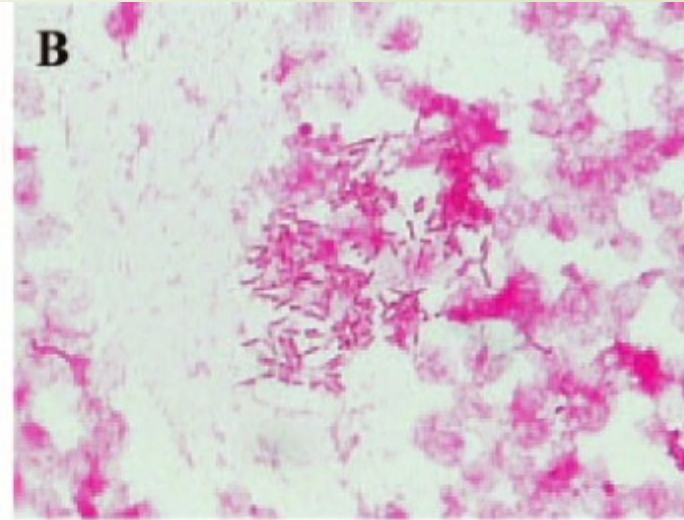
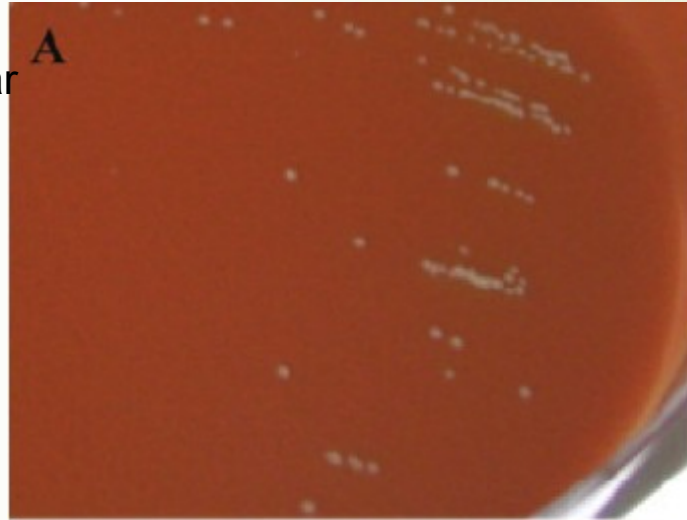
- Gram (-) pałeczka, pleomorficzna, kapnofilna, Rodzina Cardiobacteriaceae
- Katalazo-ujemne, oksydazo-dodatnie, β -hemolityczne
- Hodowla na agarze krwawym lub czekoladowym w 5 % CO₂, 37°C
- Czas hodowli 3-5 dni zaczynają pojawiać się hodowle dodatnie
- Po 14 dniach raportujemy hodowle ujemne



Science Source - Cardiobacterium ho...
sciencesource.com

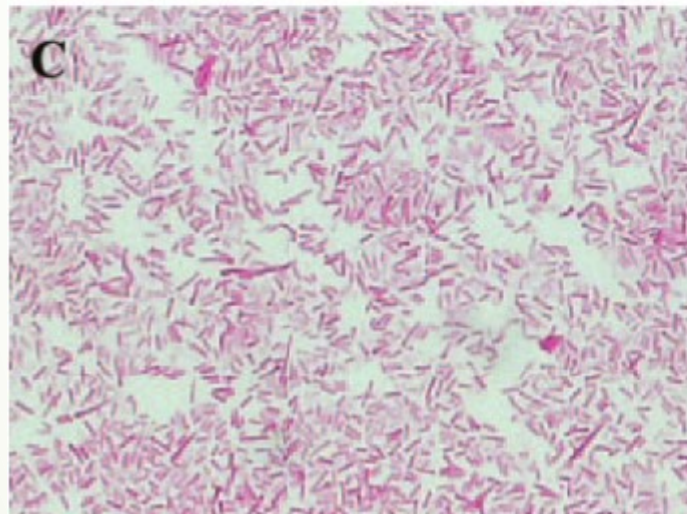
Cardiobacterium vulvarum

A- 4 doby, 5 % CO₂, 37°C agar czekoladowy

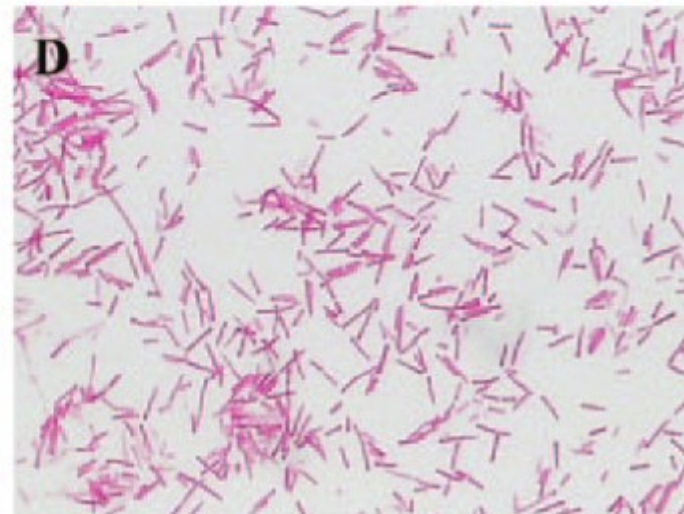


B- z hodowli krwi

B,C, D – preparaty grama



C- z agaru z krwią owcy





D- z agaru czekoladowego

A case of *Cardiobacterium valvarum* endocarditis with cerebral hemorrhage after MVR, TVP and vegetation removal operation

Authors

[Authors and affiliations](#)

Lijia Ni, Xiaoying Xie, Nengyong Ouyang, Baiji Chen, Dongye Wang, Xiaoqiang Liu, Xiquan Wu, Jiajian Guo, Hongyu Li,

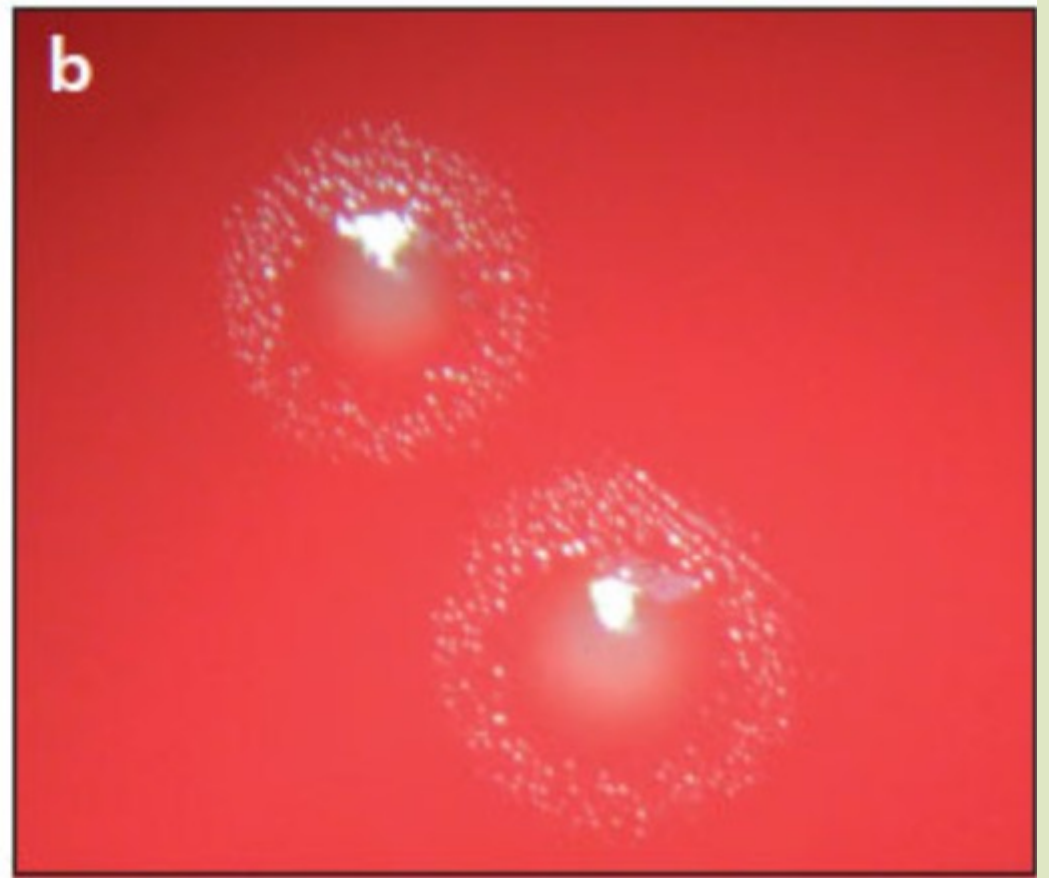
Yandan Yao , Songyin Huang 

- ▮ Infekcje są bardzo rzadkie
- ▮ Od 2004 roku opisano w literaturze światowej 13 przypadków

Eikenella corrodens

- W 1958 roku odkryta przez M.Eikena
- Rodzina Neisseriaceae
- Gram(-) ziarniakopoleczka, wymagająca pokarmowo, względnie beztlenowa
- Hodowla na agarze z krwią lub czekoladowym 5 % CO₂, 37°C, 2 dni
kolonie małe, szare, potem stają się żółte wrastające w podłoże
- Wydziela charakterystyczny zapach, (hypochlorid)
- Oksydazo dodatnia, katalazo ujemna
- Identyfikacja na kartach Vitek NH, MALDI-TOF
- Lekowrażliwość wg norm EUCAST dla bakterii niezwiązanych z gatunkiem

Eikenella corrodens



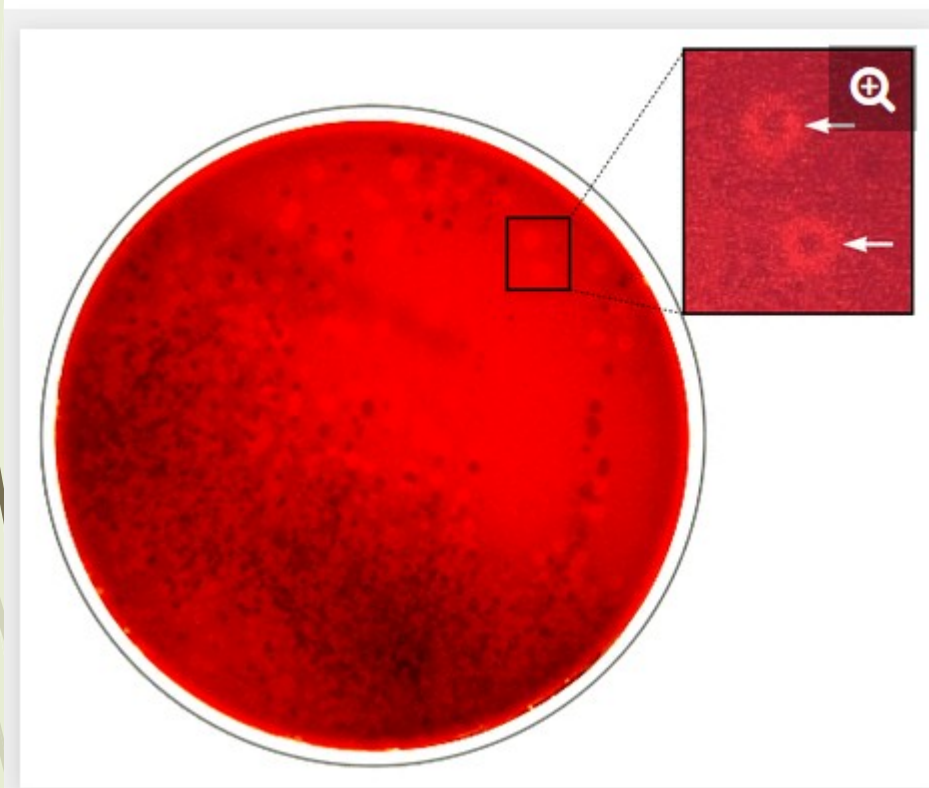
Inwazyjne kolonie 0.5-1 mm po 48 godzinach z poszarpanym brzegiem



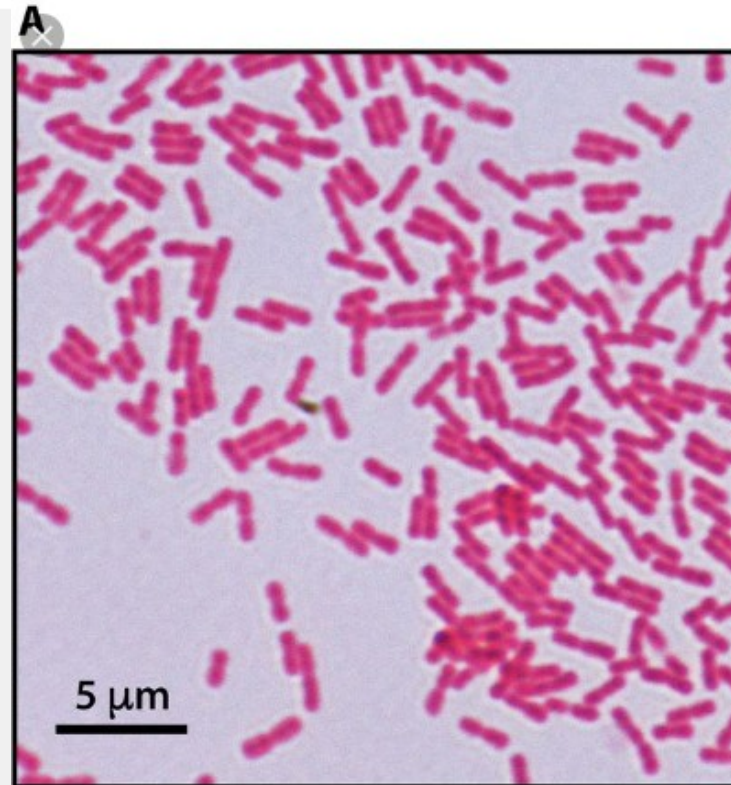
Kingella kingae

- Gram(-) ziarniakopoleczka, wymagająca pokarmowo, względnie beztlenowa
- Rodzina Neisseriaceae
- Kolonizuje 10-28 % dzieci do 4 roku życia, 1% dorosłych
- Hodowla na agarze z krwią, tworzy β -hemolizę
lub agrze czekoladowym 5 % CO₂, 37°C, 2 dni
- Oksydazo dodatnia, katalazo ujemna
- Identyfikacja na kartach Vitek NH, MALDI-TOF

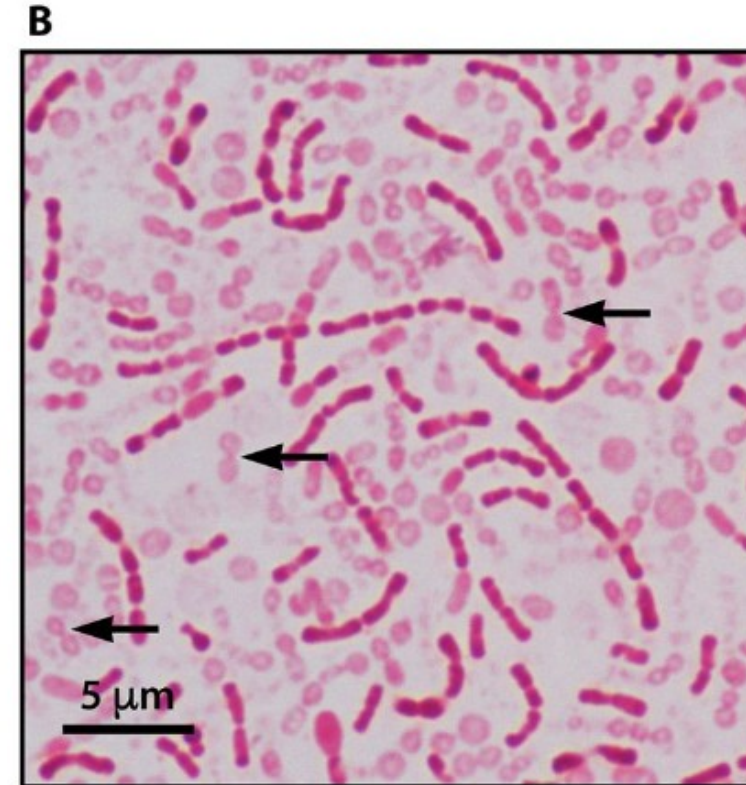
Kingella kingae, *K. nevegensis*



β -hemoliza wokół *K. kingae* z nosogardła



A- preparat grama *K. kingae*



B- autoliza *K. nevegensis* po 36 godz. hodowli

Expert Rules and Intrinsic Resistance Tables

MIC determination (broth microdilution according to ISO standard 20776-1)

Medium: Mueller-Hinton broth + 5% lysed horse blood and 20 mg/L β-NAD (MH-F broth)

Inoculum: 5x10⁵ CFU/mL

Incubation: Sealed panels, air, 35±1°C, 18±2h. Isolates with insufficient growth after 16-20h incubation are reincubated immediately and inhibition zones read after a total of 40-44h incubation.

Reading: Unless otherwise stated, read MICs at the lowest concentration of the agent that completely inhibits visible growth.

Quality control: *Haemophilus influenzae* ATCC 49766. For agents not covered by this strain, see EUCAST QC Tables.

Disk diffusion (EUCAST standardised disk diffusion method)

Medium: Mueller-Hinton agar + 5% defibrinated horse blood and 20 mg/L β-NAD (MH-F)

Inoculum: McFarland 0.5

Incubation: 5% CO₂, 35±1°C, 18±2h. Isolates with insufficient growth after 16-20h incubation are reincubated immediately and inhibition zones read after a total of 40-44h incubation.

Reading: Unless otherwise stated, read zone edges as the point showing no growth viewed from the front of the plate with the lid removed and with reflected light.

Quality control: *Haemophilus influenzae* ATCC 49766. For agents not covered by this strain, see EUCAST QC Tables.

Penicillins ¹	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Benzylpenicillin	0.03	0.03		1 unit	25	25		1. Beta-lactamase positive isolates can be reported resistant to benzylpenicillin and to ampicillin and amoxicillin without inhibitors. Tests based on a chromogenic cephalosporin can be used to detect the beta-lactamase. Beta-lactam resistance mechanisms other than beta-lactamase production have not yet been described for <i>K. kingae</i> . 2. Susceptibility can be inferred from benzylpenicillin susceptibility. 3/B. The intrinsic activity of clavulanic acid in <i>K. kingae</i> is such that the organism is inhibited by 2 mg/L clavulanic acid. Therefore no breakpoints for amoxicillin-clavulanic acid can be given. A. Infer susceptibility from benzylpenicillin susceptibility.
Ampicillin	0.06 ²	0.06 ²			Note ^A	Note ^A		
Amoxicillin	0.125 ²	0.125 ²			Note ^A	Note ^A		
Amoxicillin-clavulanic acid	Note ³	Note ³			Note ^B	Note ^B		

Cephalosporins	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Cefotaxime	0.125	0.125		5	27	27		Numbered notes relate to general comments and/or MIC breakpoints. Lettered notes relate to the disk diffusion method.
Ceftriaxone	0.06	0.06		30	30	30		
Cefuroxime iv	0.5	0.5		30	29	29		

Carbapenems	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Meropenem	0.03	0.03		10	30	30		Numbered notes relate to general comments and/or MIC breakpoints. Lettered notes relate to the disk diffusion method.

Fluoroquinolones	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Ciprofloxacin	0.06	0.06		5	28	28		Numbered notes relate to general comments and/or MIC breakpoints. Lettered notes relate to the disk diffusion method.
Levofloxacin	0.125	0.125		5	28	28		

Kingella kingae

Expert Rules and Intrinsic Resistance Tables

EUCAST Clinical Breakpoint Tables v. 9.0, valid from 2019-01-01

Macrolides and lincosamides	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Azithromycin	0.25 ¹	0.25 ¹			Note ^A	Note ^A		1. Susceptibility can be inferred from erythromycin susceptibility. A. Infer susceptibility from erythromycin susceptibility.
Clarithromycin	0.5 ¹	0.5 ¹			Note ^A	Note ^A		
Erythromycin	0.5	0.5		15	20	20		
Clindamycin	-	-			-	-		

Tetracyclines	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Doxycycline	0.5 ¹	0.5 ¹			Note ^A	Note ^A		1/A. Isolates susceptible to tetracycline are also susceptible to doxycycline, but some resistant to tetracycline may be susceptible to doxycycline. An MIC method should be used to test doxycycline susceptibility of tetracycline resistant isolates if required.
Tetracycline	0.5	0.5		30	28	28		

Miscellaneous agents	MIC breakpoints (mg/L)			Disk content (µg)	Zone diameter breakpoints (mm)			Notes
	S ≤	R >	ATU		S ≥	R <	ATU	
Rifampicin	0.5	0.5		5	20	20		1. Trimethoprim:sulfamethoxazole in the ratio 1:19. Breakpoints are expressed as the trimethoprim concentration.
Trimethoprim-sulfamethoxazole ¹	0.25	0.25		1.25-23.75	28	28		

Wytyczne ESC dotyczące leczenia infekcyjnego zapalenia wsierdza o etiologii „HACEK” z 2015 roku

- ▣ Ceftriakson 2 g/dobę 4 tygodnie w IZW na zastawkach natywnych
6 tygodni w IZW na zastawkach sztucznych
- ▣ Ampicylina 12g/dobę (w 6 dawkach)
+ Gentamycina 3 mg/kg w 2-3 dawkach
- ▣ Ciprofloksacin 400 mg co 8-12 godz. lub 750 mg co 12 godzin

dziękuję za uwagę

