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Antibiotikaresistens og nye antibiotika - er det mulig å tenke nytt?

Engelsk:

The ZinChel Project - Overcoming β-Lactam Resistance

The Zinchel Project
October 2015

Pål Rongved

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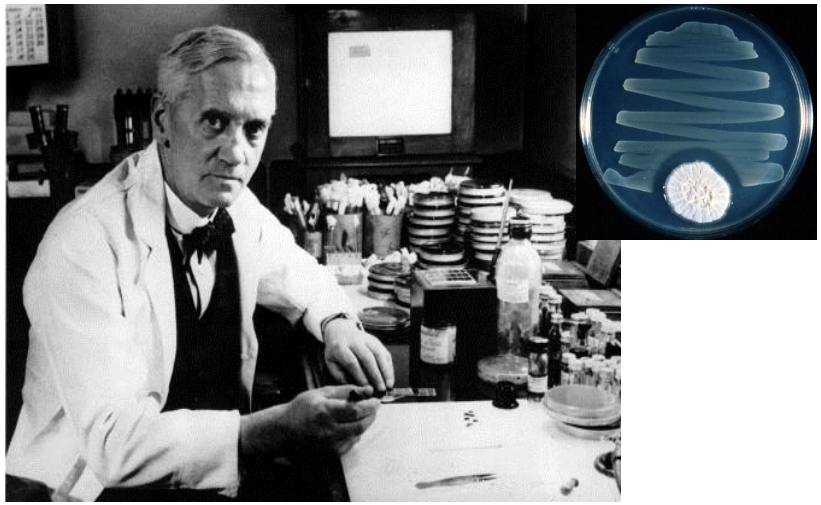
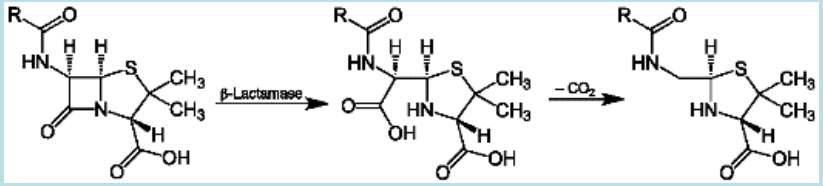
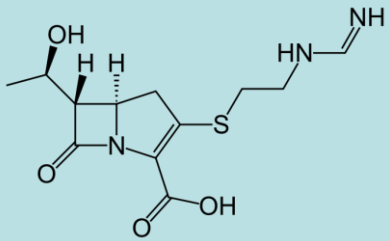
Drug Discovery within Antibiotics

Alexander Fleming: penicillin G in 1928
- the 1945 Nobel Price in medicine

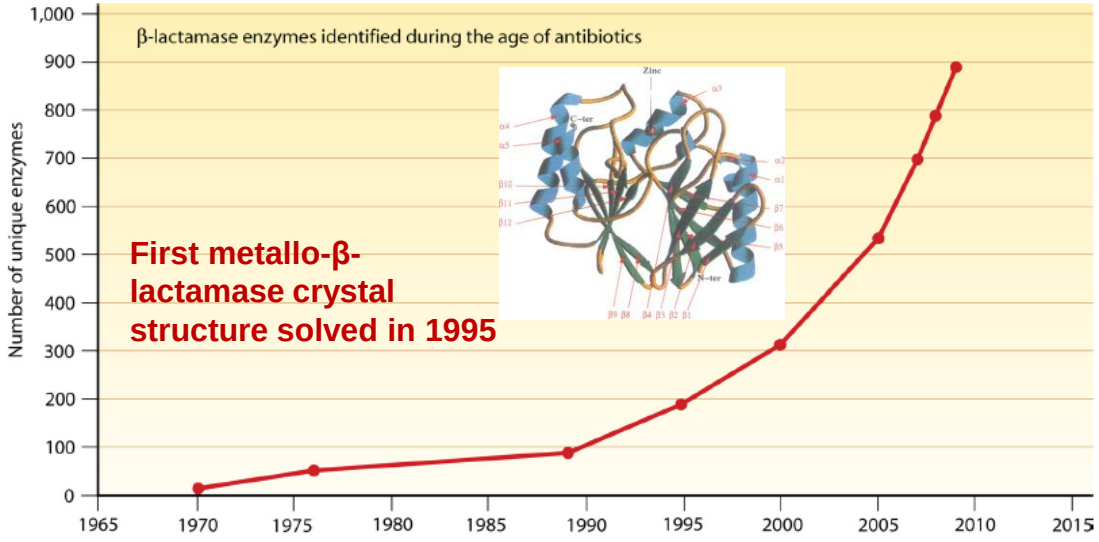


1940 – first penicillinase discovered
1942 – first penicillin became «available»





Imipenem. The first carbapenem. FDA approved 1985.



Timeline of Antibiotic Resistance

Nearly as quickly as life-saving antibiotics are created, new drug-resistant infections appear

ANTIBIOTICS INTRODUCED

ANTIBIOTIC RESISTANCE IDENTIFIED

Treats pneumonia; meningitis; skin, bone, joint, stomach, blood, and heart valve infections.	*penicillin	1943	1959	tetracycline-R Shigella
Treats pneumonia; acne; infections of respiratory tract, genitals, urinary systems and stomach ulcers.	tetracycline	1950	1962	methicillin-R Staphylococcus
Treats bronchitis; diphtheria; Legionnaires' disease; whooping cough; pneumonia; rheumatic fever; venereal disease, and many others.	erythromycin	1953	1965	penicillin-R pneumococcus
Treats staph infections.	methicillin	1960	1968	erythromycin-R Streptococcus
Treats infections of lung, skin, bone, joint, stomach, blood, and urinary tract.	gentamicin	1967	1979	gentamicin-R Enterococcus
Treats colitis.	vancomycin	1972	1987	ceftazidime-R Enterobacteriaceae
Treat infections of lung, skin, bone, joint, stomach, blood and gynecological and urinary tracts.	imipenem and ceftazidime	1985	1988	vancomycin-R Enterococcus
Treats pneumonia, chronic bronchitis and sinus, urinary tract, kidney, prostate, and skin infections.	levofloxacin	1996	1996	levofloxacin-R pneumococcus
Treats pneumonia, and infections of the skin and blood.	linezolid	2000	1998	imipenem-R Enterobacteriaceae
			2001	linezolid-R Staphylococcus
			2002	vancomycin-R Staphylococcus
Injection is used to treat some types of skin infections and pneumonia.	ceftaroline	2010	2011	ceftaroline-R Staphylococcus

*Penicillin-resistant Staphylococcus appeared in 1940, three years before widespread use of the drug.

Source: Centers for Disease Control and Prevention.

Credits: SwitchYard Media and Food & Environment Reporting Network

YEARLY ANTIBIOTIC-RESISTANT INFECTIONS

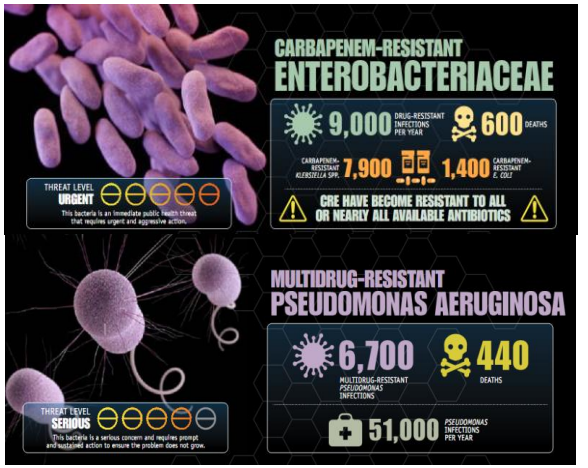
More than 2 million people in the U.S. are sickened every year with antibiotic-resistant infections. At least 23,000 die as a result.

ANTIBIOTICS IN AGRICULTURE

Of all antibiotics sold in the U.S. each year, 80 percent by weight are used in agriculture, primarily to fatten and protect animals.

BEFORE ANTIBIOTICS:

- **5 women died** out of every 1,000 who gave birth.
- **Three out of 10** who contracted pneumonia died.
- **One out of every 10** people who got a skin infection from a scrape, a cut, or scratching a bite lost a limb



Hospitals – immuno deficiency…

Source: M. McKenna, 2013, Fern's AG Insider

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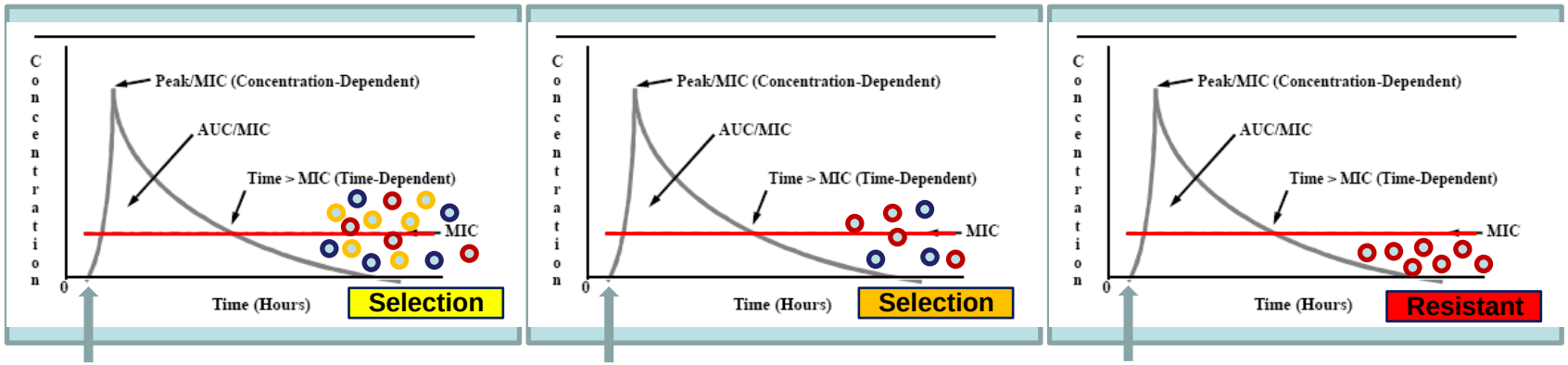
Antibiotics and Resistance

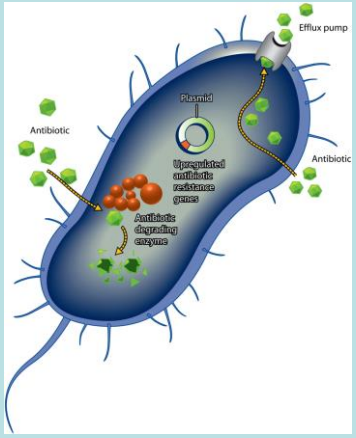


1st dose **2nd dose** **3rd dose**

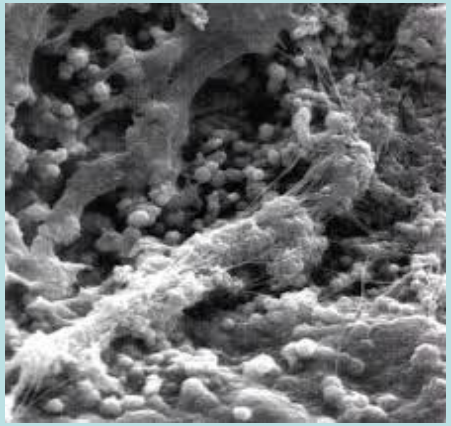
Level of resistance: High / Medium / Low

MIC: minimum effective concentration

Enzymes, efflux pumps, gene transfer

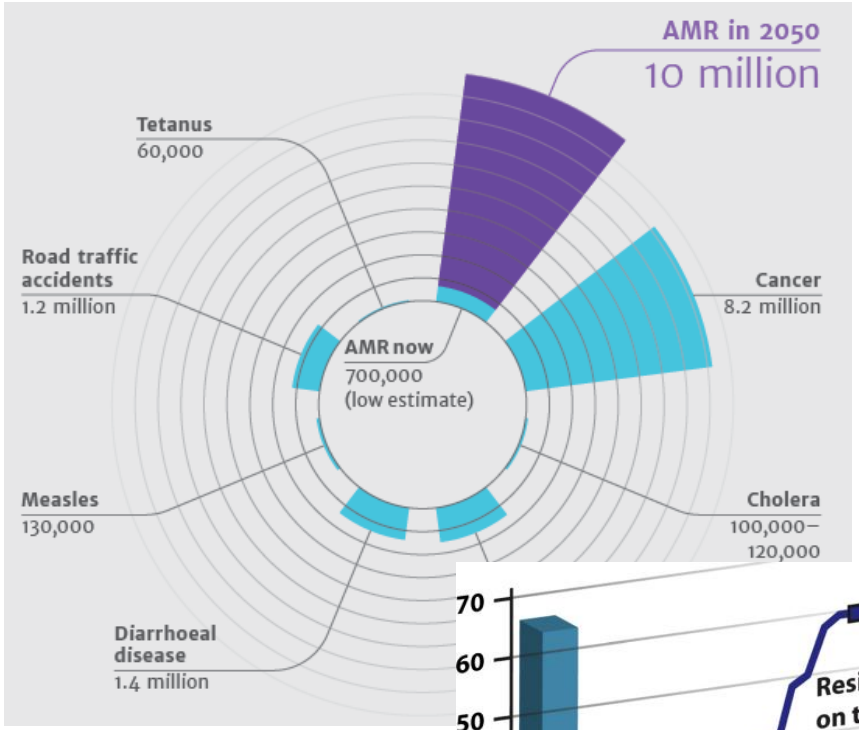


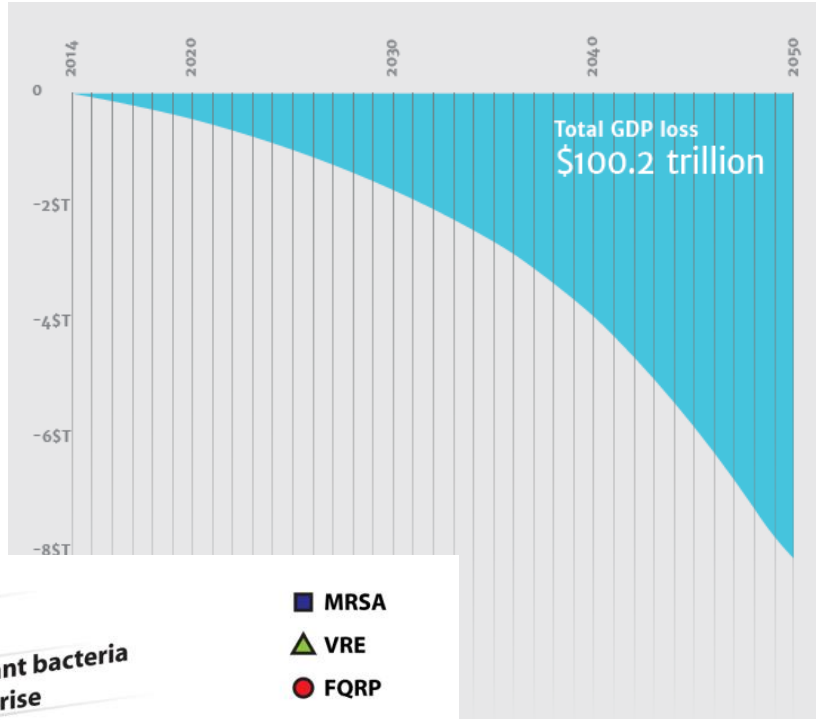
Biofilm

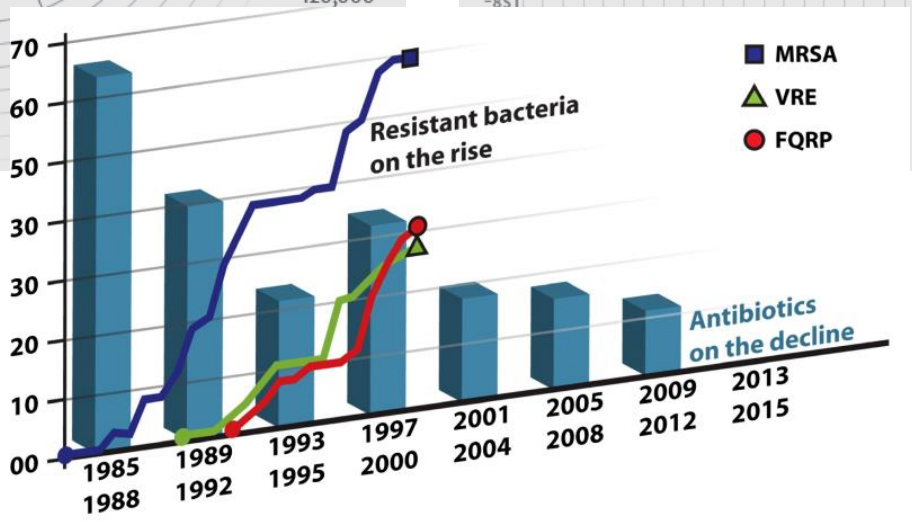


The Review on Antimicrobial Resistance
Chaired by Jim O'Neill
December 2014

If no new antibiotics..



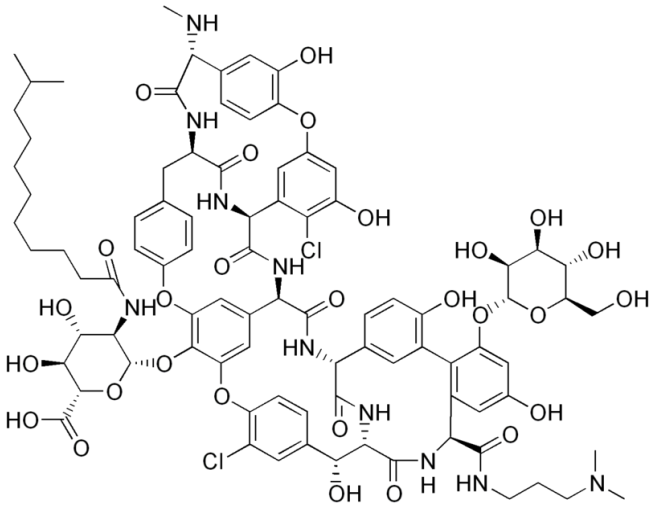




What new Antibiotics Marketed?

In 20 years: only three genuinely first-in-class AB to the market (US):

- **2014: Dalbavancin (Dalvance, Durata Terapeutics) – lipoglycopeptide – against *gram-positive***
- **2012: Bedaquilin (Sirturo, Janssen) – diarylquinoline – against *tuberculosis***
- **2011: Fidaxomicin (Dificid, Cubist/Merck) – macrocyclic – against *clostridium difficile***



Dalbavancin
(EU: Xydalba, Actavis)

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β-Lactamases

- β-lactamases: β-lactam-cleaving enzymes.
- Capable of cleaving all β-lactam antibiotics
- Zinc is essential for catalytic activity.
- 2015: no clinical inhibitor of metallo-β-lactamase on the market.

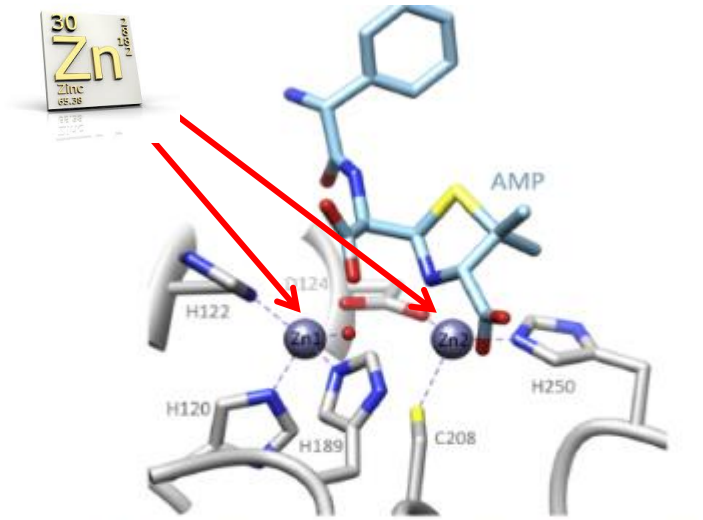
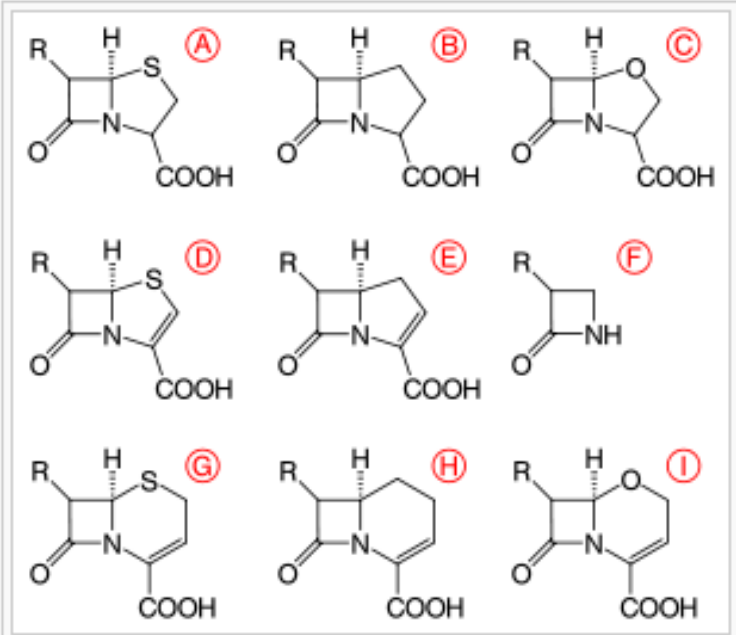


Figure 5. Primary zinc ligands of NDM-1. A structure of NDM-1 (grey) bound to hydrolyzed ampicillin (light blue) shows the primary zinc ligands conserved throughout the B1 family of metallo-beta-lactamases. Notably, Zn_2 bridges between Cys208 and the product carboxylate. The figure was prepared using coordinates from protein databank accession code 3Q6X[23] and the molecular visualization program UCSF Chimera.[24]



The β-lactam core structures. **(A)** A penam. **(B)** A carbapenam. **(C)** An oxapenam. **(D)** A penem. **(E)** A carbapenem. **(F)** A monobactam. **(G)** A cephem. **(H)** A carbacephem. **(I)** An oxacephem.

- 2008 – New Dehli metallo-β-lactamase discovered in Sweden.
- Active against all β-lactam antibiotics on the market.
- First patient with NDM-1-related disease died in 2010.

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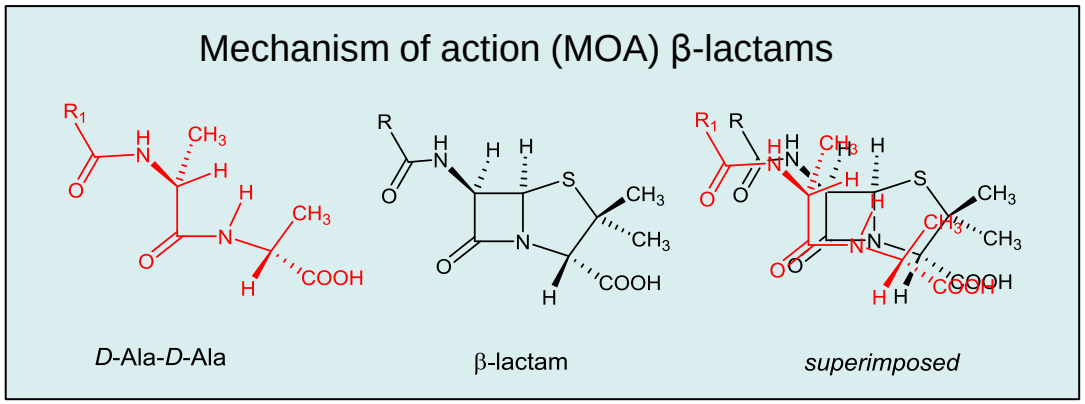
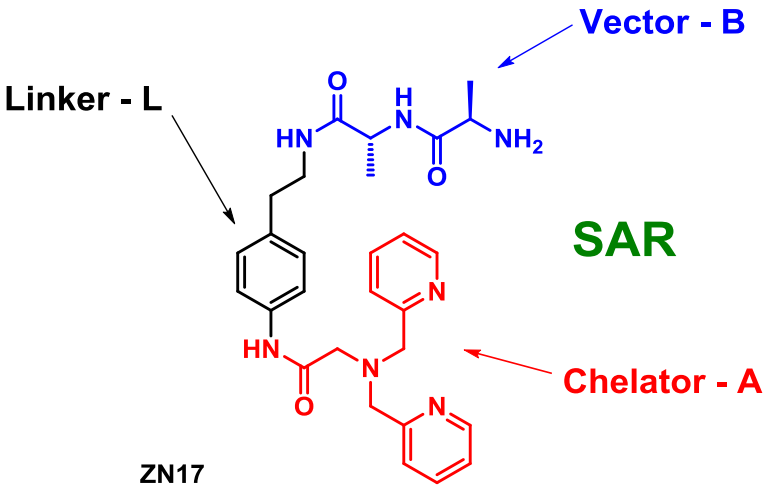
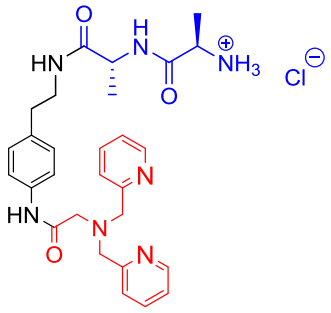
ZinChel project – a new strategy against resistant bacteria

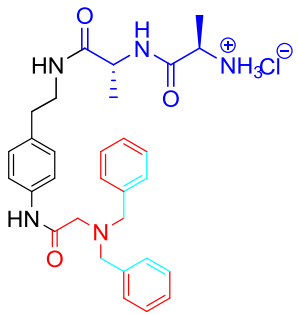
Based on chemistry – rational design

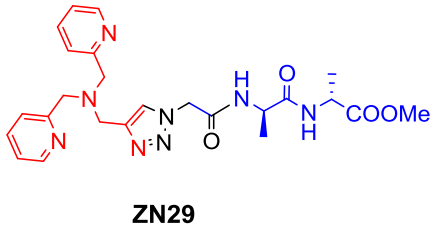


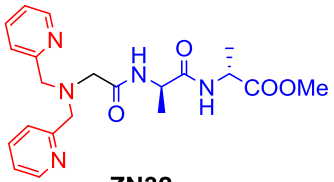


General binding to penicillin-binding proteins (PBPs)?







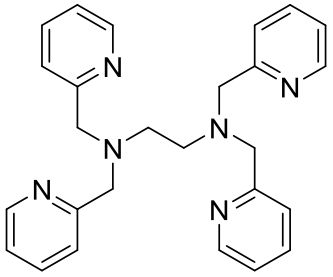


Structure gram negative bacteria are ongoing. They revealed the necessity of all the elements **A**, **L** and **B** in the general structure below, exemplified by the lead candidate **ZN17.**

Results – Enzyme Inhibition and Whole Cell

Enzyme inhibition pure enzyme

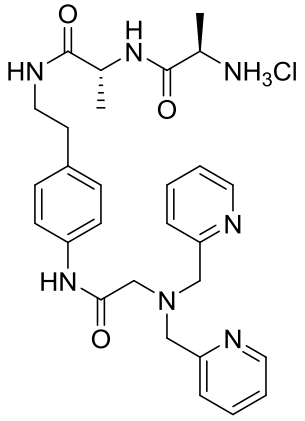
	IC_{50} value VIM-2 µM	IC_{50} value VIM-7 µM	IC_{50} value GIM-1 µM	IC_{50} value NDM-1 µM
TPEN	11.69	103.2	0.61	0.30
ZN17	22.8	54.48	0.40	0.52



TPEN

Whole cell assay

	% inhibition at 250 µM VIM-2	% inhibition at 250 µM GIM-1
TPEN	98.90	38.45
ZN17	98.25	69.20
Positive control	90.63	18.07
Netative control	0	0



ZN17

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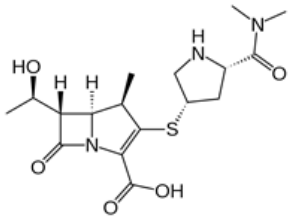
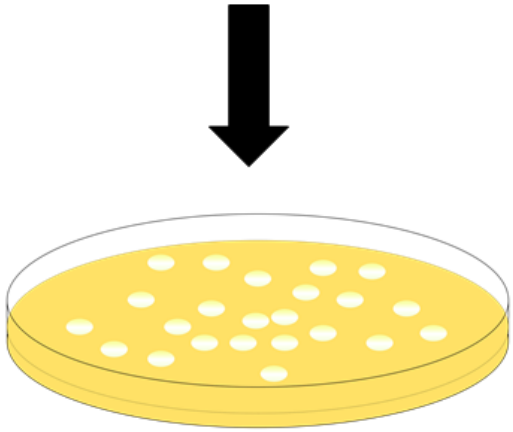
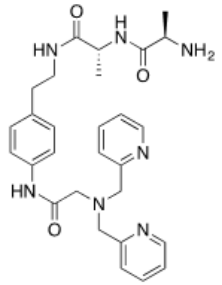
Results –
Testing Clinically Relevant Resistant Bacteria

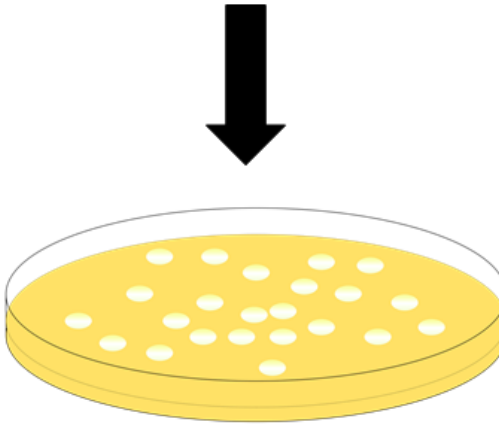
Meropenem



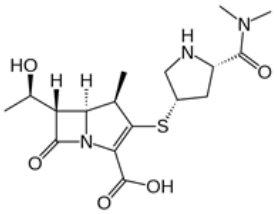


ZN17





Meropenem + ZN17



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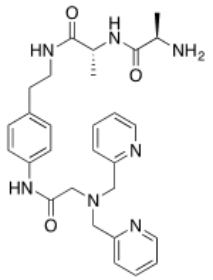

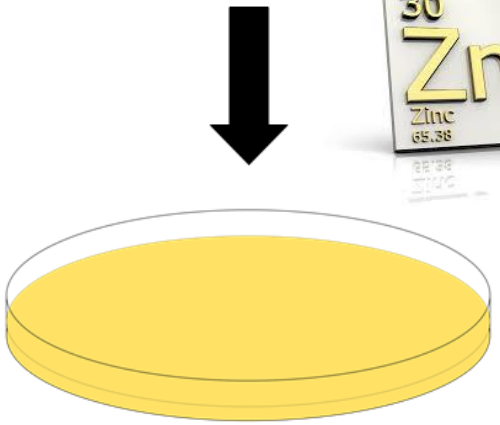

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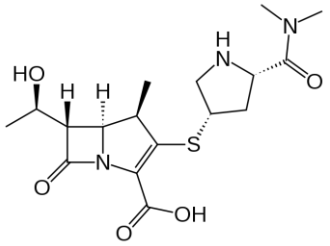
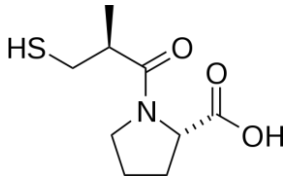
Results – Clinical Isolates

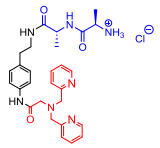
Ørjan Samuelsen, K-Res, UNN



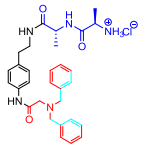
Meropenem



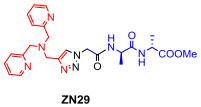
Captopril



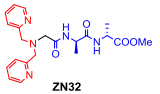
ZN17



ZN42



ZN29



ZN32

	Gram-negative bacteria harboring NDM-1			Toxicity in human cancer cells (IC_{50}, µM)		
Ref. no	50692172	K66-45	K71-77	Pancreas cancer		Breast cancer
Species	P. aeruginosa	K. pneumoniae	E. coli	MDA-MB-231	MiaPaCa	Colo357
MEM	32-128	32-64	1-8	n.a.	n.a.	n.a.
MEM + ZN17	≤0,5	≤0,5	≤0,064	127.2 ± 91.6	118.4 ± 31.5	117.9
MEM + ZN 29	2	≤0,5	≤0,064	152.2 ± 17.1	115.7 ± 8.9	186.4
MEM + ZN32	16	8-16	0,5-1	183.6 ± 45.8	97.7 ± 6.3	169.1
MEM + ZN42	64	128	32	15.4 ± 10.0	12.7 ± 4.3	20.4 ± 5.1
MEM + Captopril	32-64	64	2-4	n.a.	n.a.	n.a.

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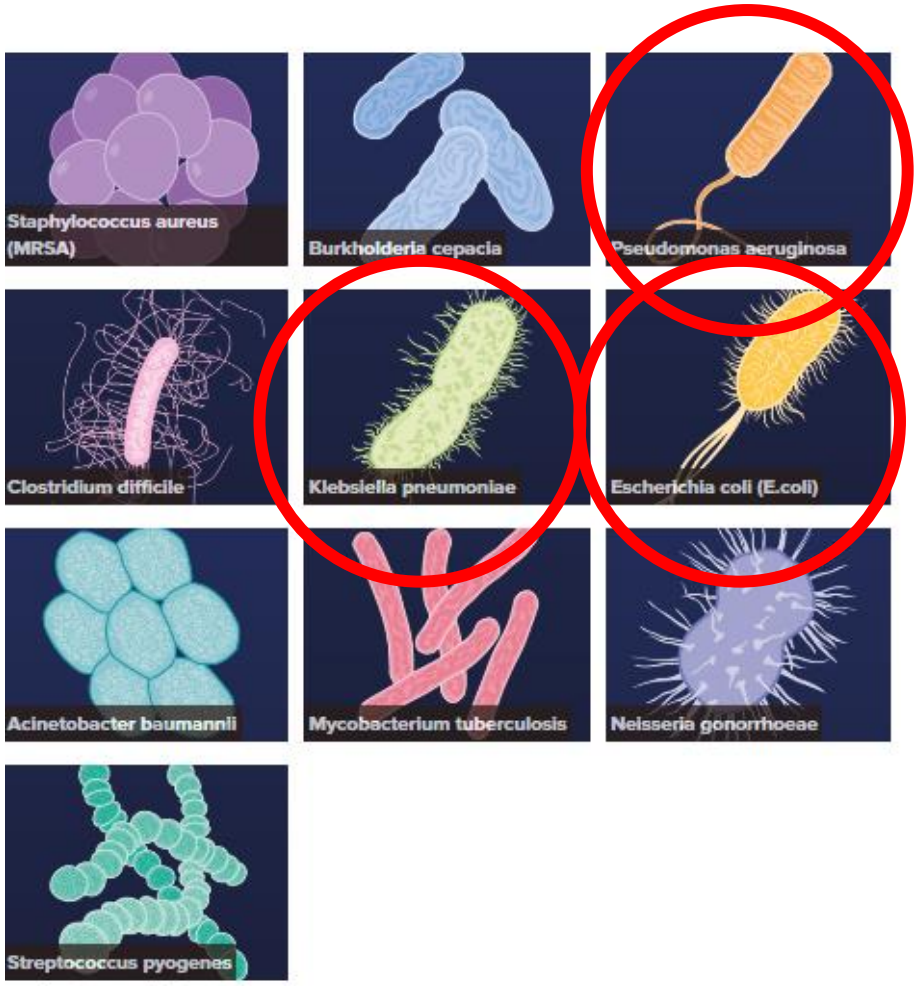
The most Threatening Resistant Bacteria



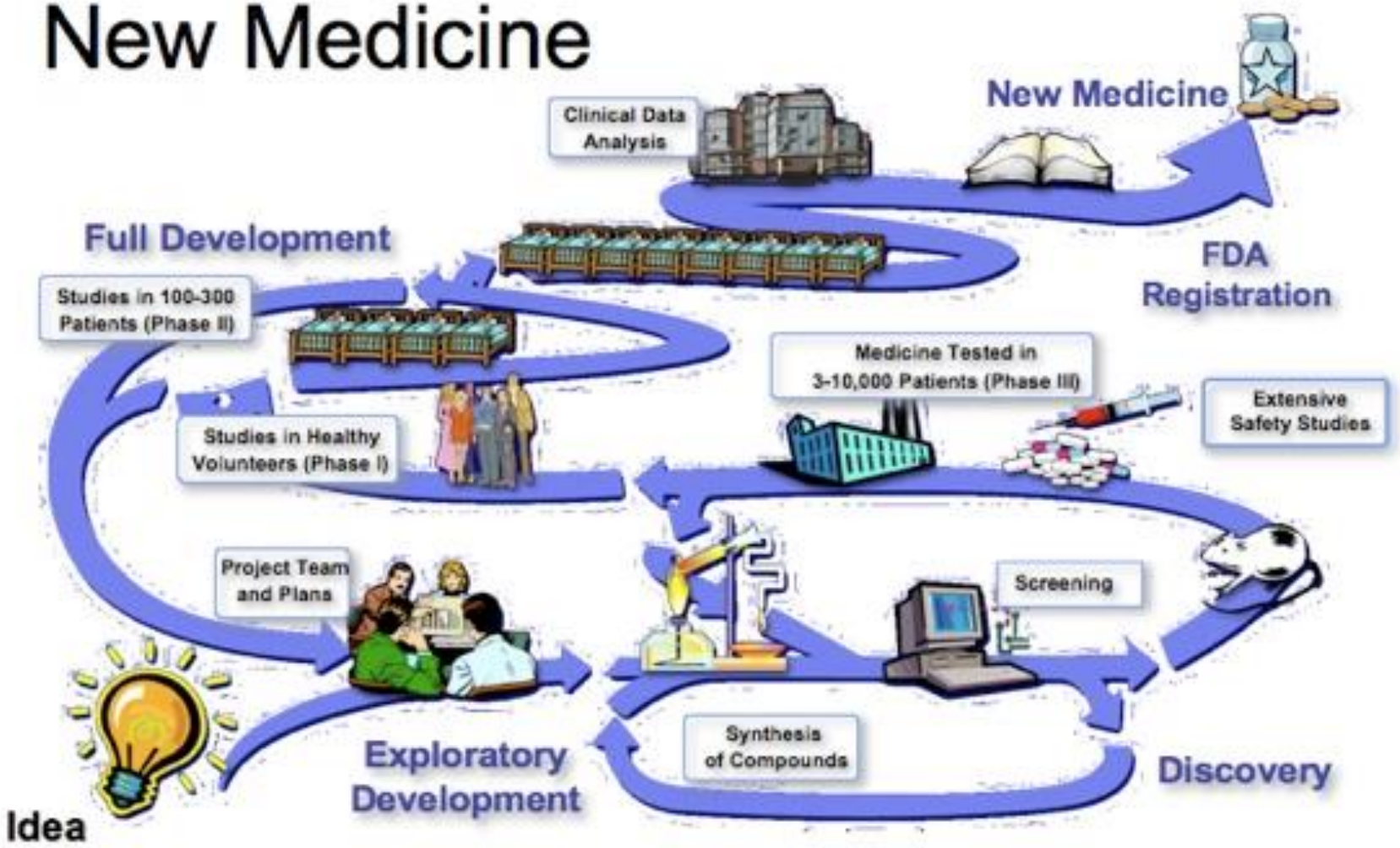
Zinchel: Bold Hypotheses:

1. **Resistance seem to develop faster in mono-target antibacterial technologies.**
2. **Zinchel is a multi-target concept – data indicates PBP-binding.**
3. **ZinChel disturbs a basic physiological parameter – the Zinc homeostasis.**
4. **In spite of 3., the compounds seem non-toxic to human cells – more data needed!**

Investigation of resistance development potential key study in project plan

Source: Nesta, October 2015: http://www.nesta.org.uk/news/antibiotic-resistant-bacteria-10-most-dangerous

The Long Road to a New Medicine



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Summary

- Only three genuinely new classes of antibiotics has been introduced to the market in 20 years (R.J. Anderson)
- More people are going to die from infections than cancer in 2050 if no new technologies are brought to the market.
- Industry is reluctant because of rapid development of resistance.
- Health authorities/governements world wide must (and will) take action both financially and in other ways.
- Our research group at School of Pharmacy/UiO has discovered a genuinely new adjuvant technology, dramatically reducing resistance towards carbapenems.
- The scope of application for other antibacterial drug classes is very wide.
- The project is not based on natural products but on medicinal chemistry rational design.
- The project has attracted attention and financing from both The NRC and two pharmaceutical companies.

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Acknowledgements - People

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MSc student Ørjan Apeland
PhD student Anthony Prandina
Associate Professsor Lars Petter Jordheim

S	Organic synthesis
Y	
N	Nano technology
F	Formulation Physical chemistry
A	Analysis
S	Synergy

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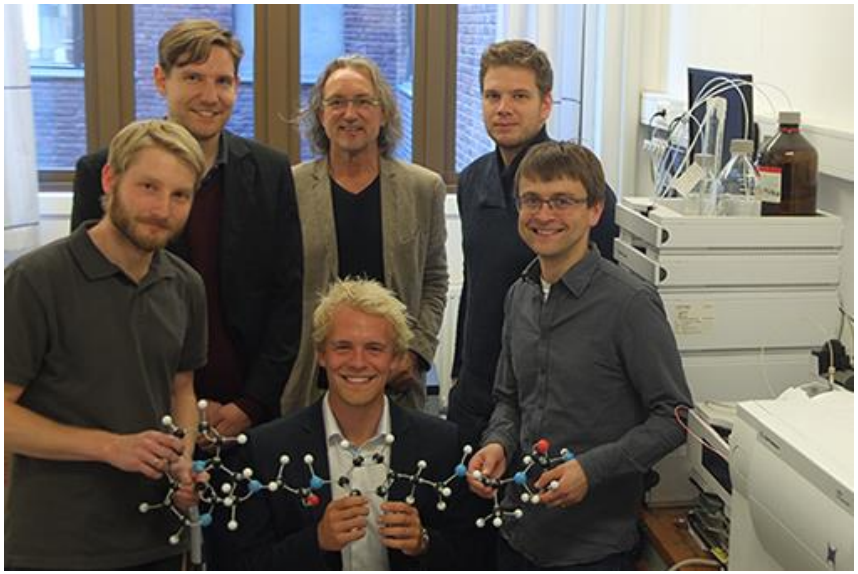
Associate Professors
- Annette Bayer
- Dr Hanna-Kirsti Leiros



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Forskningsrådet

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