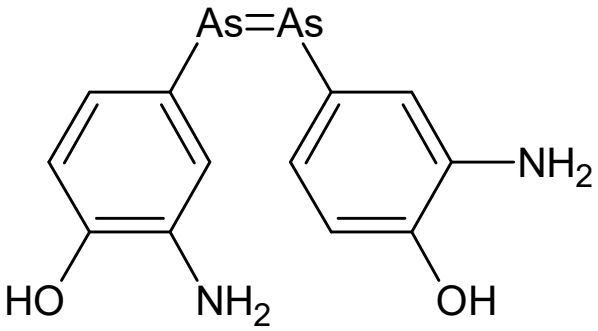
How much did Fleming discover penicillin?
„World“ and „our Czech“ penicillins...

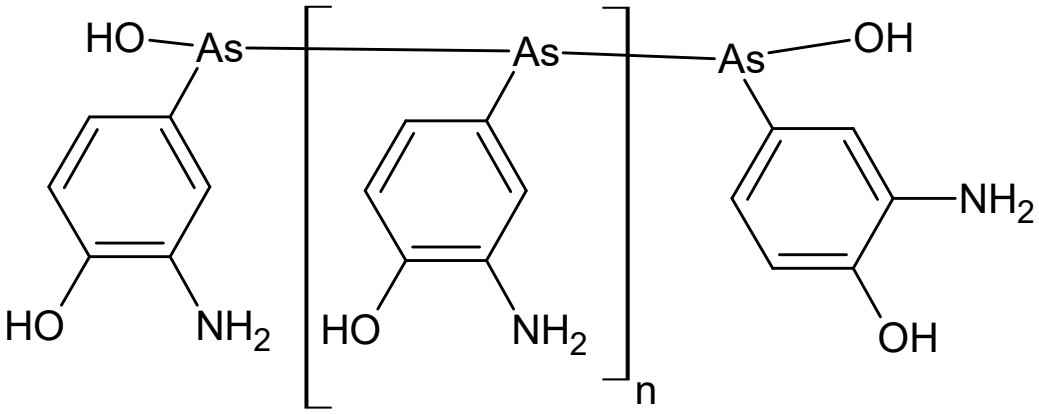
The period before penicillin...

arsphenamine (Ehrlich 1910)
Salvarsan

- for treatment of syphilis
- first antibacterial chemotherapeutic



Structure proposed by Ehrlich



Structure as it is recognized today

Penicilin(s)
History started

(World)
1928 – **Alexander Fleming** – isolated a liquid concentrate inhibiting growth of bacteria from a broth of a mould of *Penicillium* species and published it in 1929:
***A. Fleming, Br. J. Exp. Pathol.,* 10, 226 (1929)**

PROFESSOR ALEXANDER FLEMING

ON THE ANTIBACTERIAL ACTION OF CULTURES OF A PENICILLIUM, WITH SPECIAL REFERENCE TO THEIR USE IN THE ISOLATION OF *B. INFLUENZÆ*.

ALEXANDER FLEMING, F.R.C.S.

From the Laboratories of the Inoculation Department, St Mary's Hospital, London.

Received for publication May 10th, 1929.

WHILE working with staphylococcus variants a number of culture-plates were set aside on the laboratory bench and examined from time to time. In the examinations these plates were necessarily exposed to the air and they became contaminated with various micro-organisms. It was noticed that around a large colony of a contaminating mould the staphylococcus colonies became transparent and were obviously undergoing lysis (see Fig. 1).

Subcultures of this mould were made and experiments conducted with a view to ascertaining something of the properties of the bacteriolytic substance which had evidently been formed in the mould culture and which had diffused into the surrounding medium. It was found that broth in which the mould had been grown at room temperature for one or two weeks had acquired marked inhibitory, bactericidal and bacteriolytic properties to many of the more common pathogenic bacteria.

A. Fleming, Br. J. Exp. Pathol., 10, 226 (1929).

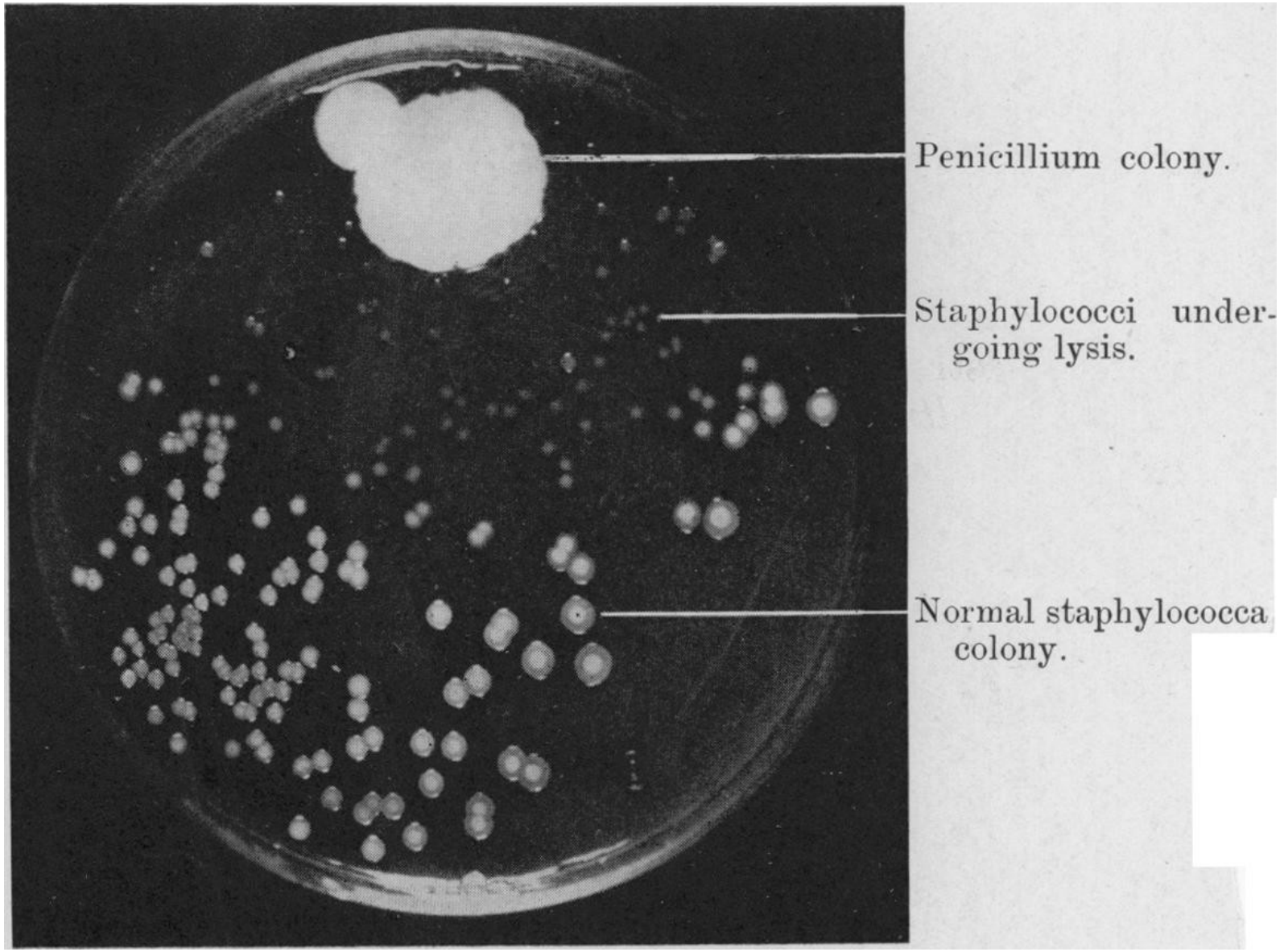


FIG. 1.-Photograph of a culture-plate showing the dissolution of staphylococcal colonies in the neighbourhood of a penicillium colony.

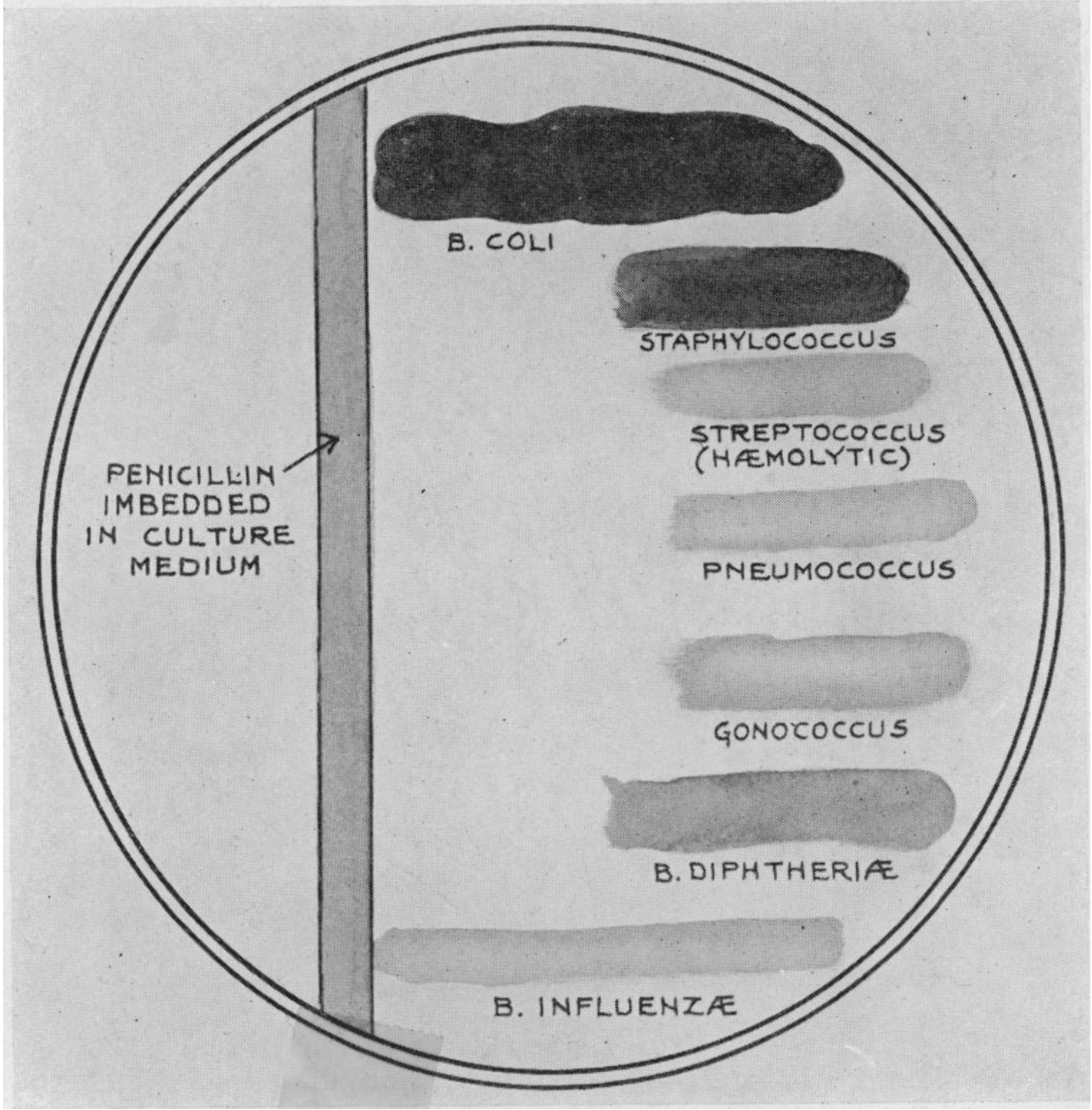

B. COLI
STAPHYLOCOCCUS
STREPTOCOCCUS (HÆMOLYTIC)
PNEUMOCOCCUS
GONOCOCCUS
B. DIPHTHERIÆ
B. INFLUENZÆ
PENICILLIN IMBEDDED IN CULTURE MEDIUM

TABLE III.—*Inhibitory Power of Penicillin on Different Bacteria.*

	Dilution of penicillin in broth.											
	1/5.	1/10.	1/20.	1/40.	1/80.	1/100.	1/200.	1/400.	1/800.	1/1600.	1/3200.	Control.
Staphylococcus aureus	0	0	0	0	0	0	0	0	±	++	++	++
„ *epidermidis*	0	0	0	0	0	0	0	0	±	++	++	++
Pneumococcus	0	0	0	0	0	0	0	0	0	++	++	++
Streptococcus (hæmolytic)	0	0	0	0	0	0	0	0	0	±	++	++
„ *viridans* (mouth)	0	0	0	0	0	0	±	++	++	++	++	++
„ *fæcalis*	++	++	++	++	++	++	++	++	++	++	++	++
B. anthracis	0	0	+	+	++	++	++	++	++	++	++	++
B. pseudo-tuberculosis rodentium	+	+	++	++	++	++	++	++	++	++	++	++
B. pullorum	+	+	++	++	++	++	++	++	++	++	++	++
B. dysenteriæ	+	++	++	++	++	++	++	++	++	++	++	++
B. coli	++	++	++	...	...	...	...	...	...	...	...	++
B. typhosus	++	++	++	...	...	...	...	...	...	...	...	++
B. pyocyaneus	++	++	++	...	...	...	...	...	...	...	...	++
B. proteus	++	++	++	...	...	...	...	...	...	..	...	++
V. choleræ	++	++	++	...	...	...	...	...	...	...	...	++

	1/60.	1/120.	1/300.	1/600.	Control.
B. diphtheriæ (3 strains)	0	±	++	++	++
Streptococcus pyogenes (13 strains)	0	0	0	++	++
„ „ (1 „)	0	0	±	++	++
„ *fæcalis* (11 „)	++	++	++	++	++
„ *viridans* at random from fæces (1 strain)	0	0	0	++	++
„ „ „ „ (2 strains)	0	0	±	++	++
„ „ „ „ (1 strain)	0	±	++	++	++
„ „ „ „ (1 „)	+	++	++	++	++
„ „ „ „ (1 „)	++	++	++	++	++
„ „ at random from mouth (1 „)	0	±	++	++	++
„ „ „ „ (2 strains)	0	0	++	++	++
„ „ „ „ (1 strain)	0	0	0	++	++

0 = no growth; ± = trace of growth; + = poor growth; ++ = normal growth.

PROPERTIES OF THE ANTIBACTERIAL SUBSTANCE.

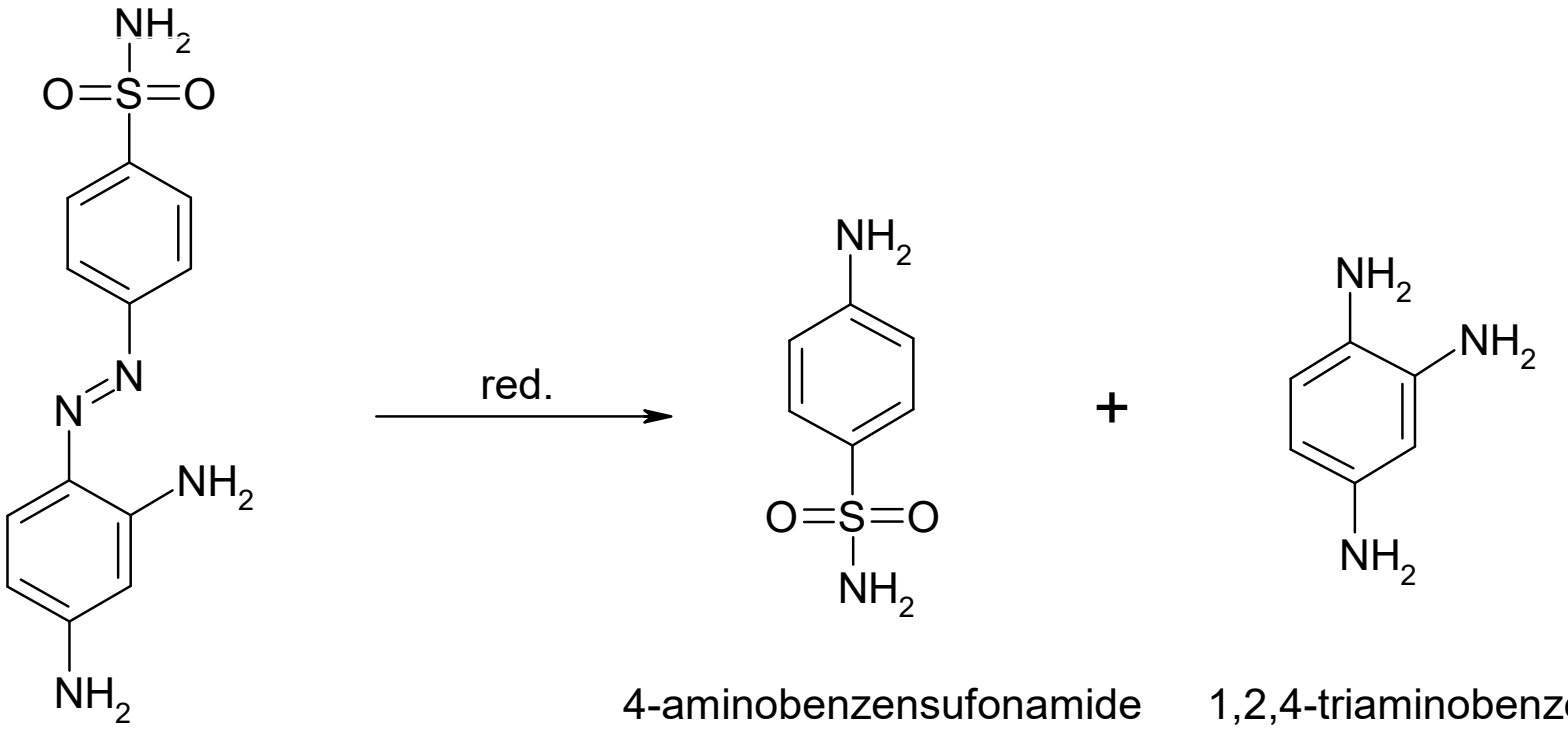
Effect of heat.—Heating for 1 hour at 56° or 80° C. has no effect on the antibacterial power of penicillin. Boiling for a few minutes hardly affects it (see Table II). Boiling for 1 hour reduces it to less than one quarter its previous strength if the fluid is alkaline, but if it is neutral or very slightly acid then the reduction is much less. Autoclaving for 20 minutes at 115° C. practically destroys it.

Effect of filtration.—Passage through a Seitz filter does not diminish the antibacterial power. This is the best method of obtaining sterile active mould broth.

Solubility.—It is freely soluble in water and weak saline solutions. My colleague, Mr. Ridley, has found that if penicillin is evaporated at a low temperature to a sticky mass the active principle can be completely extracted by absolute alcohol. It is insoluble in ether or chloroform.

...and this was everything what was known about the chemical nature of the substance in 1929...

„Rule of sulfonamides“ 1935 - 1943



4-aminobenzensufonamide
sulfanilamide

1,2,4-triaminobenzene

4-(2,4-diaminophenylazo)benzenesulfonamide

Prontosil rubrum
1932: synthesis by Mietsch and Klarer; successfully tested by Domagk against streptococci

1935: Jacques and Therése Tréfoulé: the holder of activity is sulfanilamide (*Prontosil album)*

Sulfonamides

- effect is **bacteriostatic**, only in combination with 2,6-diaminopyrimidines (trimetoprim) **bactericidal**

Spectrum of effect:

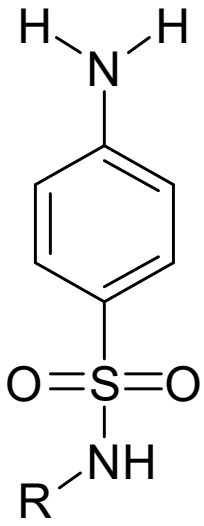
broad, G^+ as well as G^-

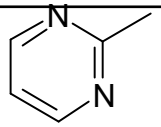
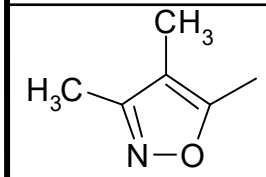
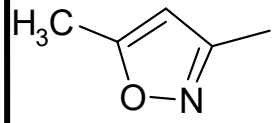
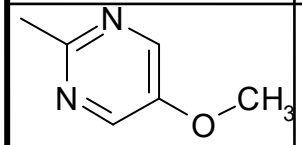
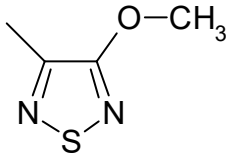
- but not enough covering G^+ bacilli in wounds...

Sulfonamides

- the most of used compounds are sulfonamides substituted with a nitrogenous heterocycle on N[1]

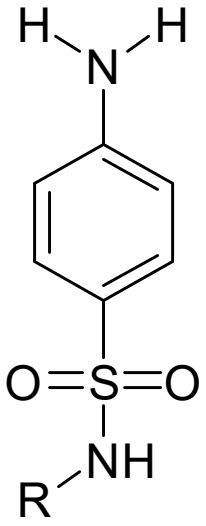
Overwiev of structures of commonly used compounds

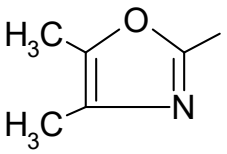
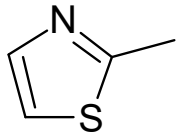
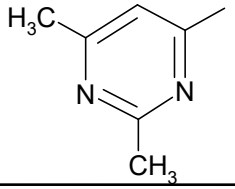
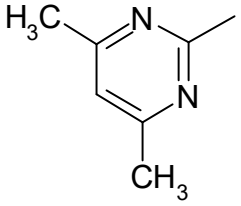
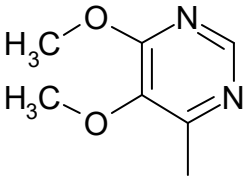


R	INN name/official name	Notice	Preparation authorized in the CR
	sulfadiazine *Sulfadiazinum PhEur*	a.u.v.	Norodine® 24 a.u.v. inj.
	sulfafurazol (syn. sulfizoxazole [USAN])		Sulfisoxazol® tbl.
	sulfamethoxazole	in combination with trimetoprim - cotrimoxazol	Biseptol®, Co-trimoxazol AL®…
	**sulfamethoxydiazine (syn. sulfameter [USAN)**	also leprostatic	
	sulfametrole	in combination with trimetoprim - lidaprim	

Sulfonamides

Overwiev of structures of commonly used compounds - continued



R	INN name/official name	Notice	Preparation authorized in the CR
	sulfamoxole	in combination with trimethoprim - supristol	
	sulfathiazole *Sulfathiazolum PhEur*		Sulfathiazol Neo® ung. Argosulfan®2% (Ag salt)
	sulfisomidine		Aristamid® gel
	sulfadimidine *Sulfadimidinum PhEur*	a.u.v. treatment of coccidiosis	Sulfadimidin Bioveta® a.u.v. plv. sol.
	sulfadoxine *Sulfadoxinum PhEur*		.

Penicilin(s)
History continued

1939 - 1943 Fleming, Florey, Chain & Johnson – larger scale production, isolation and constitution of penicillin(s)
Chain, E., Florey, H. W., Gardner, A. D., Heatley, N. G., Jennings, M. A., Orr-Ewing, J., and Sanders, A. G., Lancet, 226 (1940)

ORIGINAL ARTICLES

FURTHER OBSERVATIONS ON PENICILLIN

E. P. ABRAHAM,* D. PHIL. OXFD
A. D. GARDNER, D.M. OXFD, F.R.C.S.
E. CHAIN,* PH.D. CAMB.
N. G. HEATLEY,† PH.D. CAMB.
C. M. FLETCHER,‡ M.B. CAMB., M.R.C.P.
M. A. JENNINGS,* B.M. OXFD
H. W. FLOREY, M.B. ADELAIDE, F.R.S.

(The Sir William Dunn School of Pathology and the Radcliffe Infirmary, Oxford)

THE work on penicillin briefly reported by Chain and others (1940) is here presented in greater detail, and its further development to the stage of human therapy is described.

Growth of Penicillin-producing Mould

The mould will grow and produce penicillin on a of development may be greater or less than that described, depending largely on the depth of the medium. A systematic study of the factors influencing penicillin-production was begun, but it could not be completed owing to the very numerous and often interdependent variables, and to the fact that the assay-method then in use could only detect large differences of titre. The following conclusions, however, could be drawn :

1. Penicillin production seems to take place over a wide range of oxygen tension. (The mould will not grow anaerobically.)
2. The mould grows satisfactorily at 24° C. At lower temperatures growth is delayed and as harvesting of the medium is carried out in the incubator higher temperatures have not been studied, 24° C. being about the upper limit of comfort. Fleming (1929) in his original description stated that the mould would not grow at 37° C. and this has been confirmed.
3. Crude attempts to change the pH of the medium or to maintain it at a constant value have not resulted in a noticeable increase in yield of penicillin, nor has the incorporation of ten times the normal amount of phosphate buffer.

Culture vessels and sowing.—After many types of containers had been tried a satisfactory ceramic vessel was eventually designed,[1] the shape and dimensions of which can be seen in fig. 2. The vessels are glazed only on the inside; this both reduces the cost and renders them easier to handle and less liable to slide when stacked one on top of the other. The inset of fig. 2 shows a convenient way in which they can be stacked for autoclaving, sowing and so on; each plug is well separated from the other but no bench space is lost, and should the medium boil in the autoclave the plugs are unlikely to be wetted. One litre of medium fills the vessels to a depth of about 1·7 cm. When a batch of vessels is first set up the medium (containing 10% of yeast-extract) is sterilised in the vessels, which are then inoculated with a few drops of a spore-suspension[2] and incubated at 24° C. Apart from an occasional test the vessels are not touched until the medium is ready to be harvested.

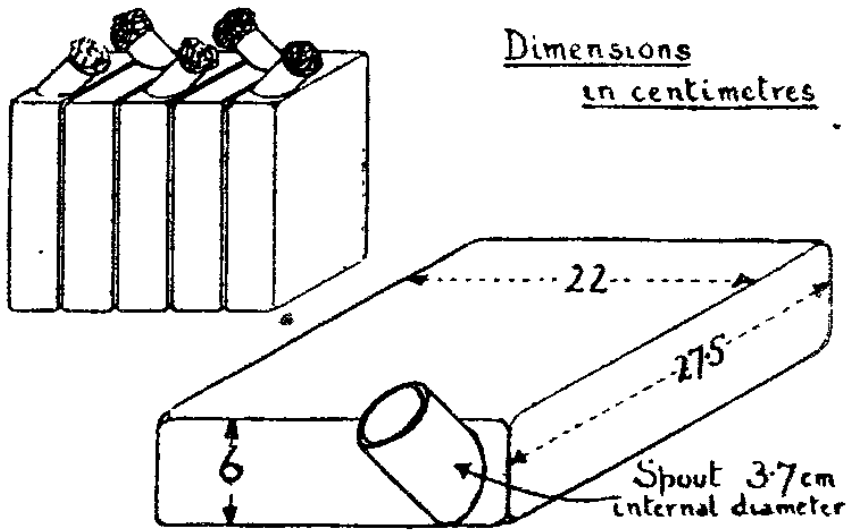


FIG. 2—Earthenware culture vessel.
Above.—Vessels stacked for autoclaving.

Arrangements for withdrawing and replacing medium.—The penicillin-con-

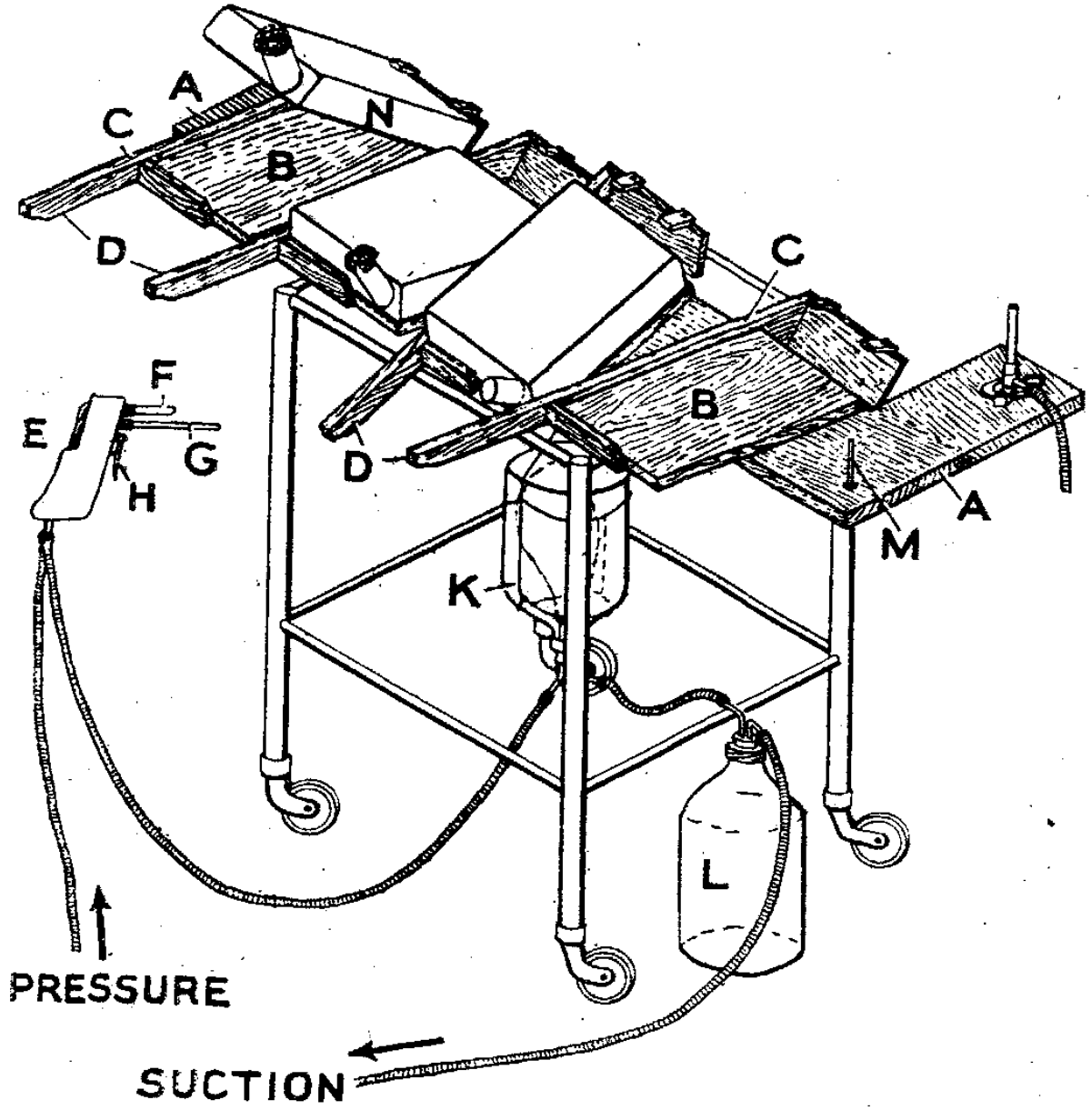


FIG. 3—Changing trolley.

A semi-automatic apparatus for continuous production of penicillin-enriched broth

Processing of the penicillin-enriched broth

- extraction from its solution with pH adjusted to 2 with diethyl ether or pentyl acetate
- re-extraction into water
- purification by a column chromatography at Brockmann´s alumina (Al_2O_3)

1. A dark brownish-orange layer whose depth is inversely proportional to the amount of charcoal used for the decolorisation and which may be absent altogether. This layer contains some penicillin.

2. A light yellow layer containing most of the penicillin but none of the pyrogen.

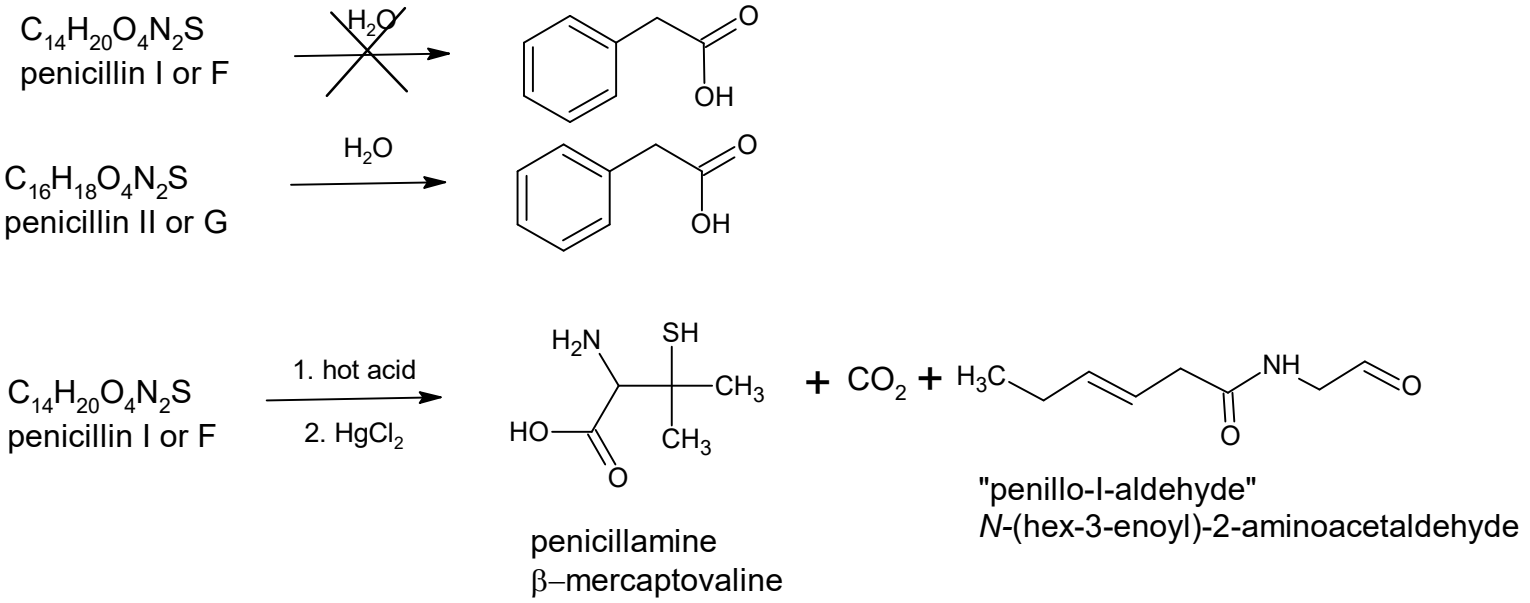
3. An orange layer which contains some penicillin and some or all of the pyrogen.

4. A brownish or reddish-violet layer which contains practically no penicillin. The violet pigment disappears on exposure to light.

- re-extracted to diethyl ether and then to diluted NaOH solution
- the composition or constitution of penicillin stil not known
- successfully on both animals and people

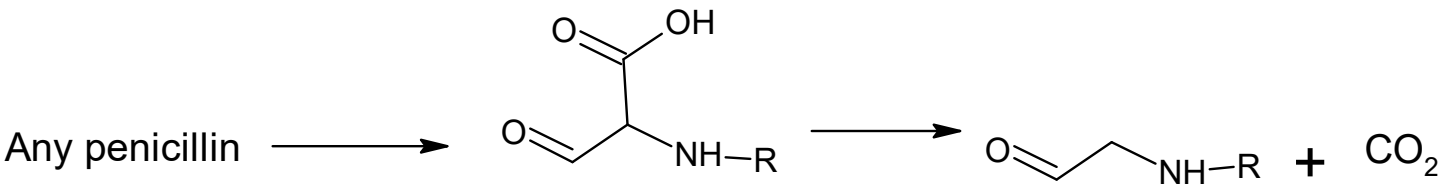
Elucidation of composition and structure of penicilline

- during 2nd World War; communication among many British and American laboratories had to be secured
- better purity by purification by chromatography on silica
- found that it is a weak acid containing N and S
- 1st isolated compounds and their decomposition products, which helped to structure elucidation:



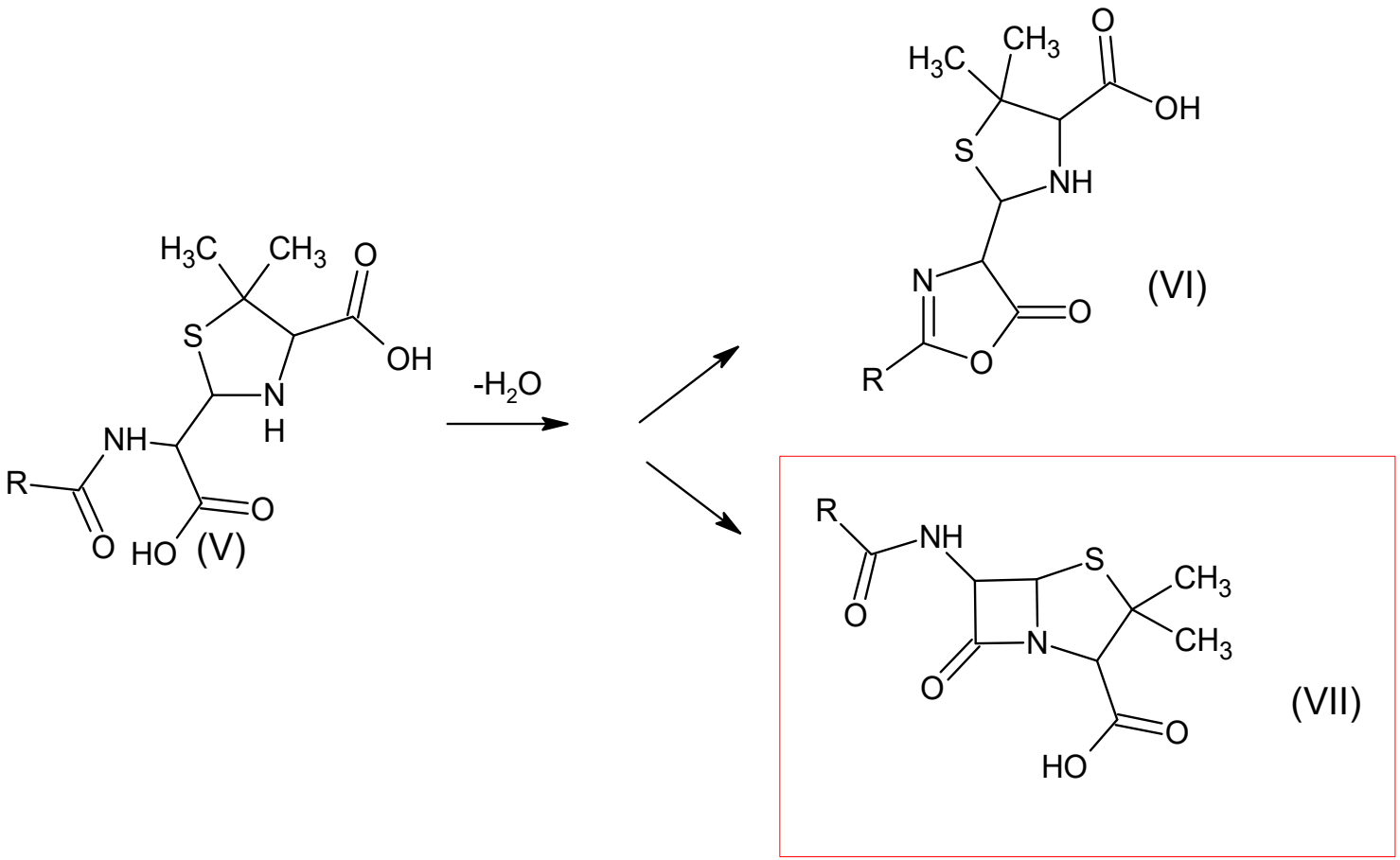
Elucidation of composition and structure of penicilline (continued)

The ease with which carbon dioxide was eliminated from the penicillin molecule suggested that it came from a carboxyl group in the β-position to the carbonyl group of the aldehyde fragment.



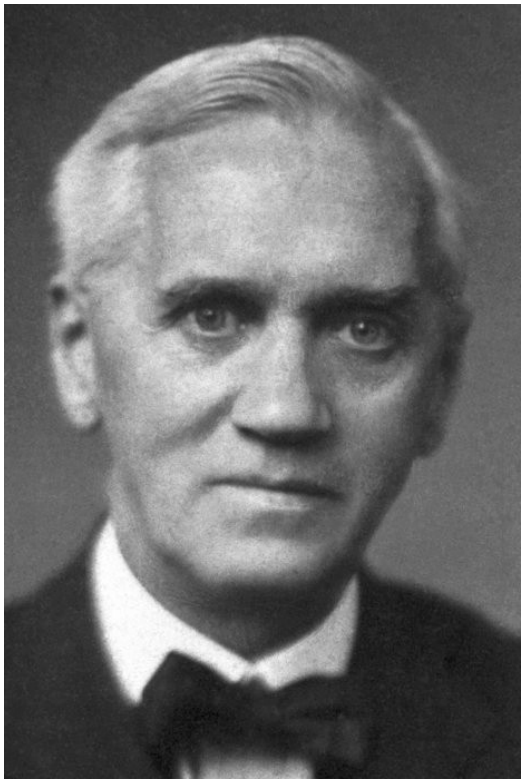
The product obtained by inactivating penicillin with dilute alkali had the properties of a thiazolidine, for it broke down into penicillamine, an aldehyde and carbon dioxide on the addition of mercuric chloride. It could therefore be assigned the structure (V), a structure that was later established beyond doubt in the Merck Laboratories. The structure of penicillin would then be found by the elimination of one molecule of water in an appropriate manner from (V):

Elucidation of composition and structure of penicilline (continued)



- no basicity adviced structure VII
- finally decided in spring 1945 by usage X-ray crystallography

October 25th, 1945 - Nobel prize for physiology and medicine for Alexander Fleming, Ernest Boris Chain and Howard Walter Florey "for the discovery of penicillin and its curative effect in various infectious diseases"



Czech penicillin

- 2nd World War:
- Czech territory occupied by Nazi Germany („Protectorate“)
- Czech universities closed; some experts found jobs in industry
- Benjamin Fragner Medicines Factory in Prague, Dolní Měcholupy
  - 1943 – a group consisting of
    - Málek - microbiology,
    - Frágner – coordination of the team
    - Miloš Herold – growing of moulds
    - Ivo Hais – chromatography on buffered silica (with J. Koštíř)
    - and others
- was secretly developing penicillin
    - (they did not want to give their results to Germans)
    - succeeded to develop a preparation called Mykoin BF 510, which corresponded approximately to the first amorphous penicillin preparation produced in England

Herold M., Matelová V., Nečásek J. A Review of Research and the Development of the Technology of Penicillin in Czechoslovakia. Folia Microbiologica 4, 351 – 359 (1959)

Czech penicillin

- clinical efficacy of Mykoin was demonstrated by several published case studies

Blecha J., Štol J.” Mykoinem BF 510 vyléčený případ stafylokokového empyému hrudníku. Čas. lék. čas. 84, 699 (1945)

Budín B., Čupík J., Málek I.: Léčba mykoinem BF 510 v praxi. Čas. lék. čes. 84, 690 (1945).

REPUBLIKA ČESKOSLOVENSKÁ

ÚŘAD PRO VYNÁLEZY

Třída 30 h, 6.

Vydáno 19. března 1955.

PATENTNÍ SPIS č. 83295 a

Doc. Dr. JOSEF KOŠTÍŘ, PRAHA a MUDr IVO HAIS, PRAHA.

Způsob získávání antibiotik z jejich nečistých roztoků.

Přihlášeno 21. června 1945. Platnost patentu od 1. dubna 1952.

Heading of a Czechoslovak patent devoted to acquiring of penicilline fraction by a chromatography on a buffered silica column

Further fate of penicillin production in Czechoslovakia

- the end of 1945: selected experts from Mykoin team (I. Málek, Z. Kabátek, M. Herold) sent to University of Toronto to study production of penicillin
- after return in 1946 and 1947, preliminary experiments using *Penicillium chrysogenum Q176* made at Dept. of microbiology, Fac. of Medicine, Charles University, under leading of I. Málek
- 1947: plant Biogena at Roztoky near Prague linked with the Institute for Antibiotic Research founded for the development and production of penicillin
- new co-workers adopted: A. Břečka, J. Zajíček, B. Sikyta and others
- original Toronto procedure improved (phenylacetamide replaced with ammonium phenylacetate etc.), % penicillin G in the mixture increased
  - paper chromatography used for estimation of % of penicillin G in the mixture
- since 1962, production of penicillin V (phenoxymethylpenicillin) started

ČESKOSLOVENSKÁ SOCIALISTICKÁ REPUBLIKA

ÚŘAD PRO PATENTY A VYNÁLEZY

Třída 30 h, 6
6 a, 14

Vydáno 15. října 1962
Vyloženo 15. dubna 1962

PATENTNÍ SPIS č. 105312

Právo k využití vynálezu přísluší státu podle § 3 odst. 6 zák. č. 34/1957 Sb.

Doc. inž. MILOŠ HEROLD, doktor věd, PRAHA, VLASTA MATELOVÁ, prom. biol., ROZTOKY u Prahy, inž. ANTONÍN BENDA, BANSKÁ BYSTRICA, ANTONÍN BŘEČKA, prom. biol., ROZTOKY u Prahy, dr. JAROSLAV DAŠEK, BANSKÁ BYSTRICA a dr. JAN NEČÁSEK, PRAHA

Způsob výběru kmenů Penicillium chrysogenum, produkujících penicilin V

Přihlášeno 15. července 1961 (PV 4404-61)
Platnost patentu od 15. července 1961

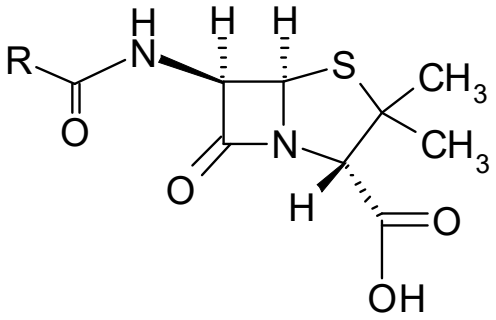
Heading of a Czechoslovak patent accompanying the beginning of production of the first orally active penicillin in Czechoslovakia

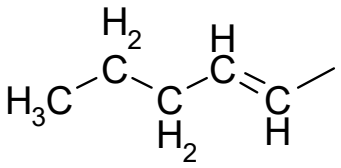
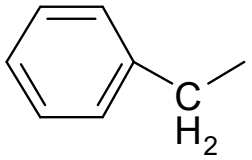
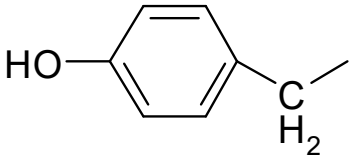
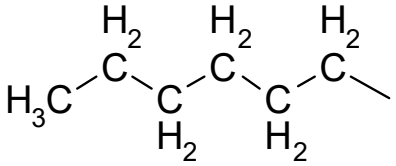
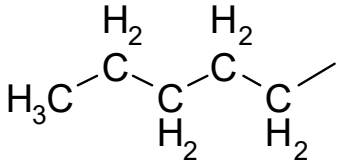
Further fate of penicillin production in Czechoslovakia

- 1953: a new enterprise Biotika in Slovenská Lupča in Central Slovakia founded
- 1956: production of penicillin started there (moved from Roztoky near Prague)
  - penicillin V as calcium and potassium salts produced there up to now

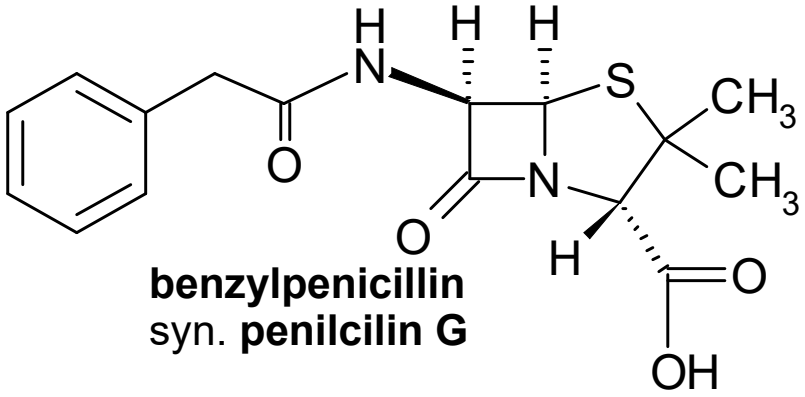
Penicillins

The initial „amorphous penicillin“ was a mixture of several compounds:



R	Penicillin	
	pentenyl-	F (or I)
	benzyl-	G (or II)
	*p*-hydroxybenzyl-	X
	heptyl-	K
	amyl-	dihydro-F

Penicillins
Benzylpenicillin and its problems

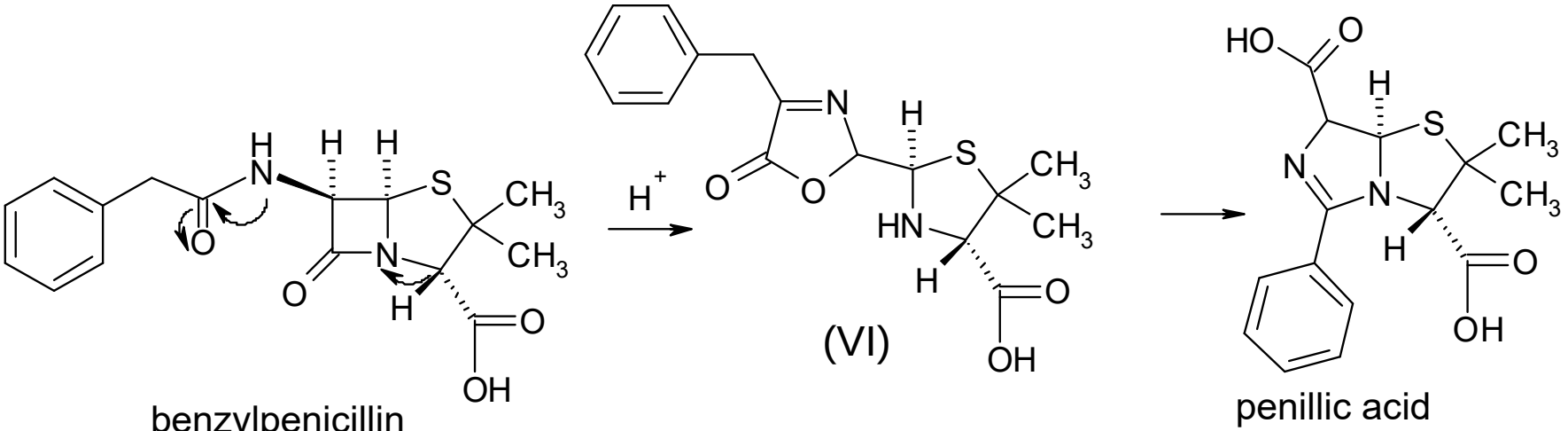


benzylpenicillin
syn. **penilcilin G**

- production of enriched benzylpenicillin by the mold enabled by addition of phenylacetic acid (or its ammonium salt, or phenylacetamide) into its broth

Problems:

- weak binding to plasma proteins ⇒ fast excretion ⇒ frequent administration is necessary
- instability in acid media of stomach (see reaction scheme) ⇒ impossibility of p.o. application



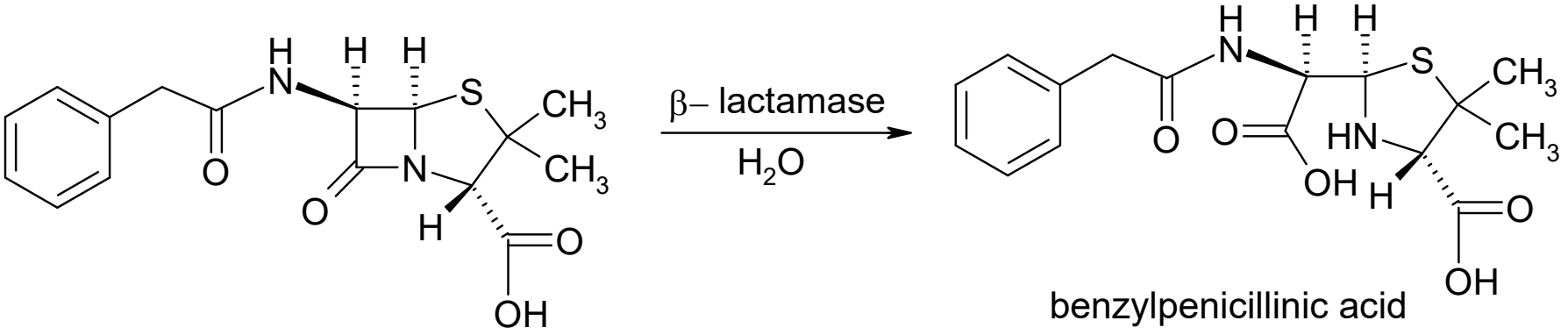
benzylpenicillin

(VI)

penillic acid

Penicillins
Benzylpenicillin and its problems

3. Sensitivity to penicillinases (β-lactamases – enzymes catalysing hydrolytic cleavage of the β-lactame ring) – see the scheme

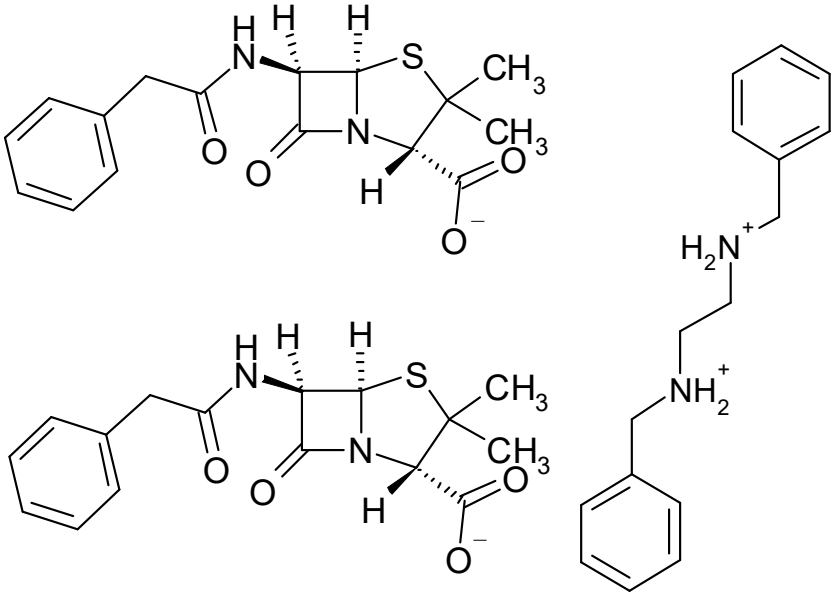


4. Rel. narrow spectrum – only G^+ strains (*Streptococcus, Staphylococcus, Clostridium, Neisseria, Corynebacteriun, Bacilus anthracis …)*

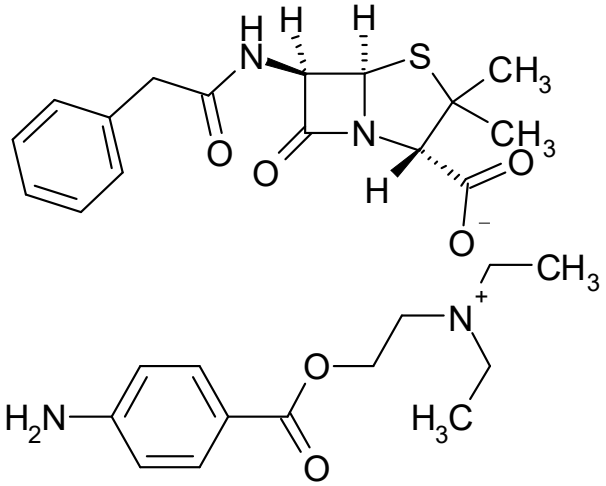
5. Inducing allergies – anaphylactic shock – caused by 6-aminopenicillanic acid as the impurity – resolved by better purification (chromatography)

Penicillins
Resolving of benzylpenicillin problems

Ad 1. (necessity of frequent application) – poorly soluble salts with organic bases



benzathine benzylpenicillin
Pendepon® inj. sic.

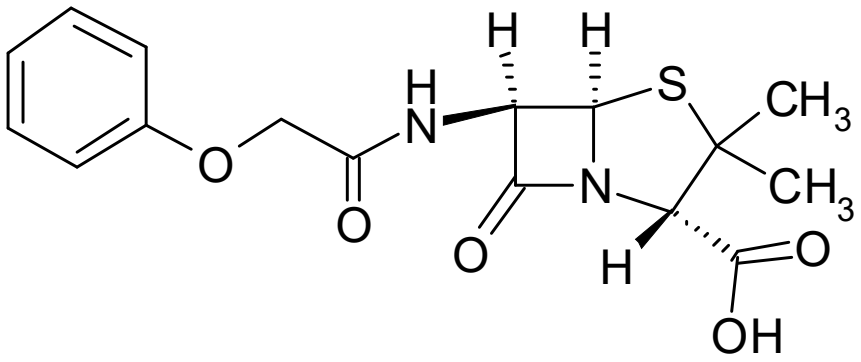


procaine benzylpenicillin
Prokain Penicilin G® Biotika inj. sic.

- depot (= long acting) forms for i.m. injections

Penicillins
Resolving of benzylpenicillin problems

Ad 2. – ↑ of stability in acid media



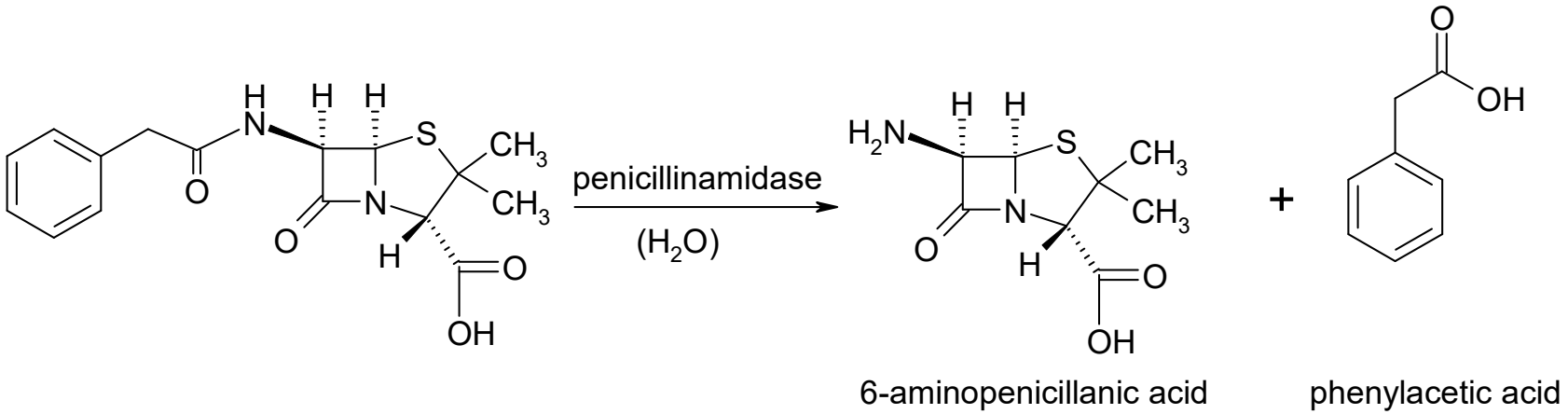
phenoxymethylpenicillin
syn. **penicillin V**
•acquired by addition of phenoxyacetic acid into the broth of the production strain
•suitable for p.o. administration
V-Penicilin®, Ospen®

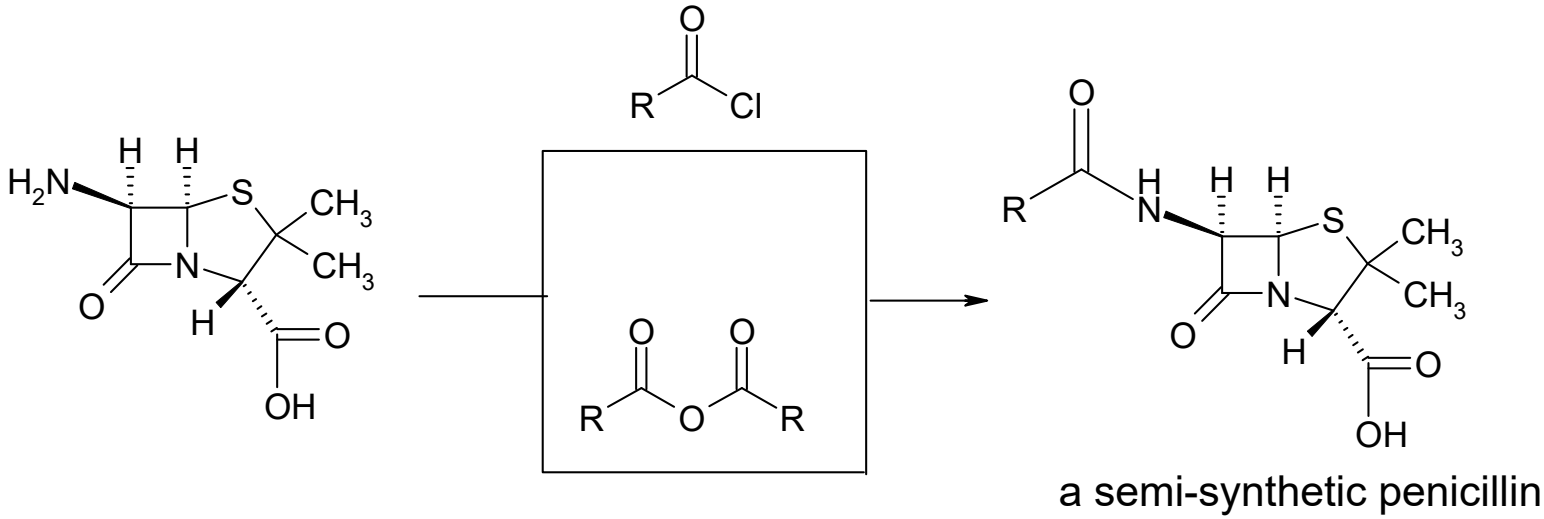
Penicillins

Overall resolving of benzylpenicililn problems – **semi-synthetic penicillins**

·**penicillinamidase (penicillinacylase) –** hydrolyzes acyclic amide bond, not β-lactame ring

·used a microbe which produces it (e.g. *E. coli*)

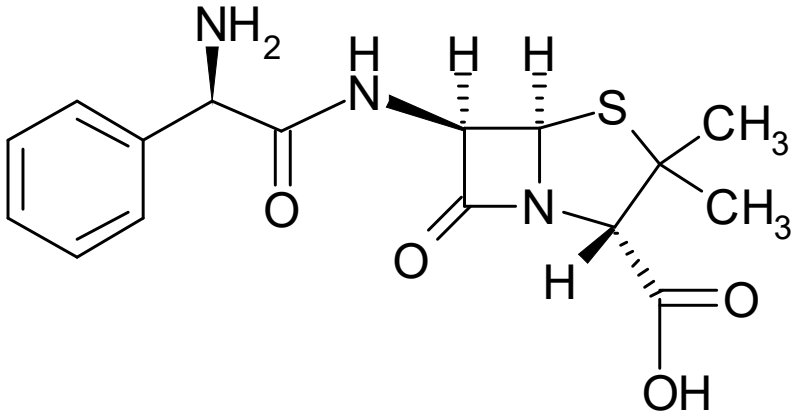




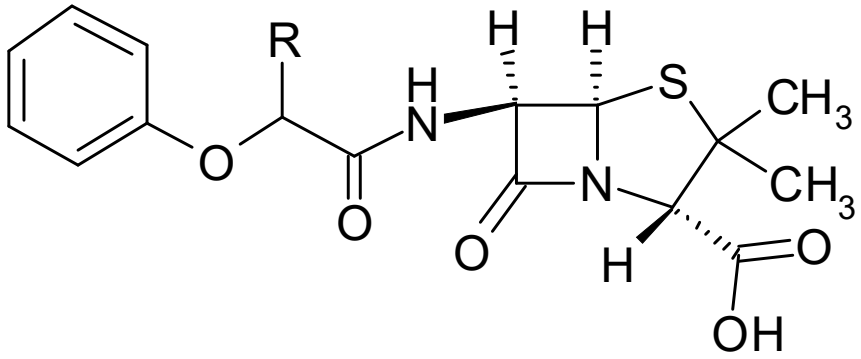
Penicillins

Mostly semi-synthetic penicillins stable in acid media

·stability against acids is increased by electron-donor substituents in N-acyl side chain (I+ or M+ effect)



ampicillin
Ampicilin ® cps., inj sic.

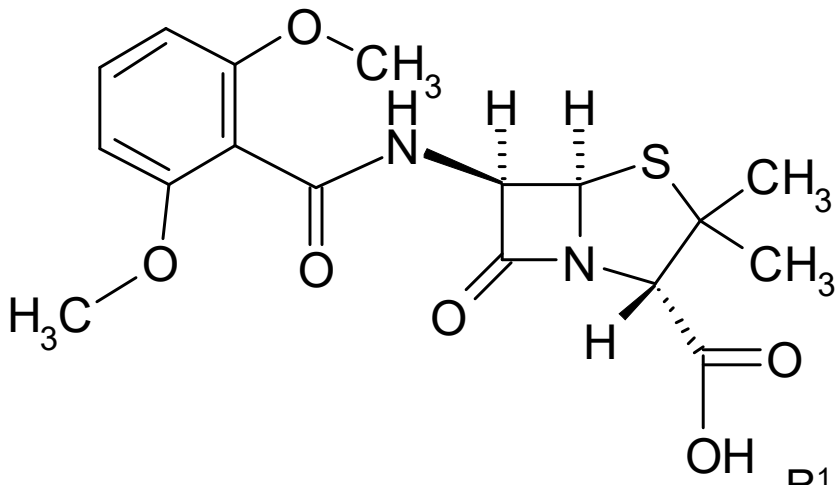


R = -H **phenoxymethylpenicillin**
V-Penicilin® tbl., Ospen tbl. obd.
R = -CH_3 **phenethicillin**
R = -CH_2CH_3 **propicillin**

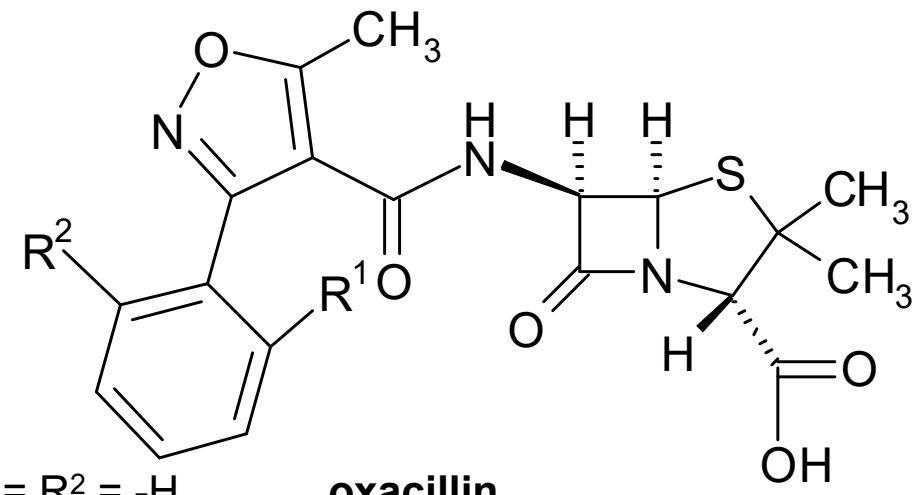
Penicillins

Semi-synthetic penicillins resistant to β-lactamases

•formed by acylation of amino group of 6-aminopenicillanic acid with bulky acyl rest; the lactame ring is then sterically hindered (⇒ protected)



meticillin



$R^1 = R^2 = -H$ **oxacillin**
Prostaphlin® ~~cps.~~, inj. sic.

$R^1 = -Cl$, $R^2 = -H$ **cloxacillin**

$R^1 = R^2 = -Cl$ **dicloxacillin**

$R^1 = -Cl$, $R^2 = -F$ **flucloxacillin**
syn. floxacillin [USAN]

•resistant also to acid media; the resistance increases
oxacillin < cloxacillin < dicloxacillin = flucloxacillin

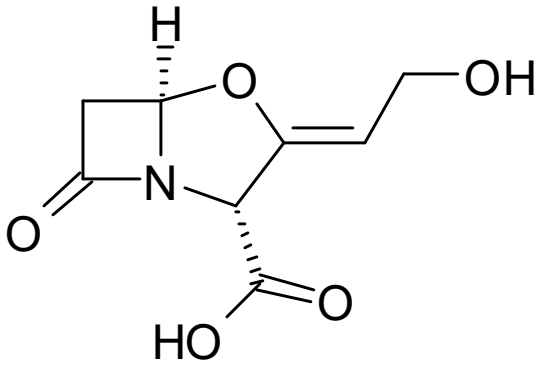
Penicillins

An alternative approach to ↑ of resistance to β-lactamases:

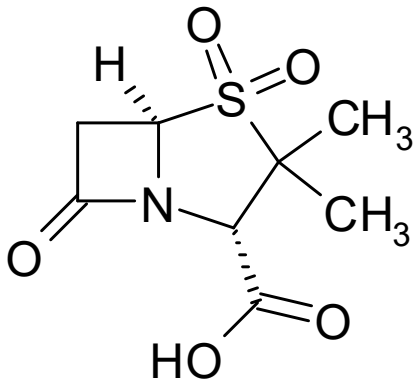
β-lactamases inhibitors

•compounds with β-lactam ring which binds to the enzyme active site with greater affinity and block this site

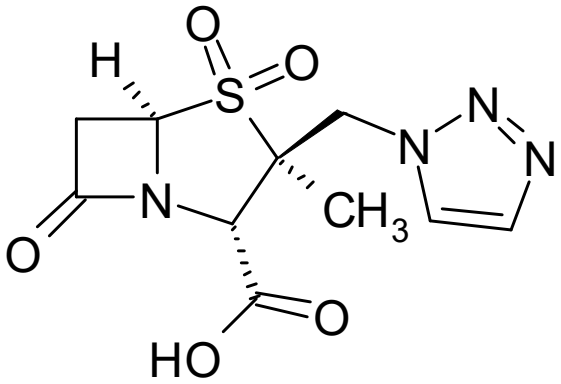
•used in combination with penicillins



clavulanic acid
•isolated from *Streptomyces clavuligerus*
+ amoxicillin (= Amoxiklav®, Augmentin®)
+ ticarcillin (= Timentin® inj. sic.)



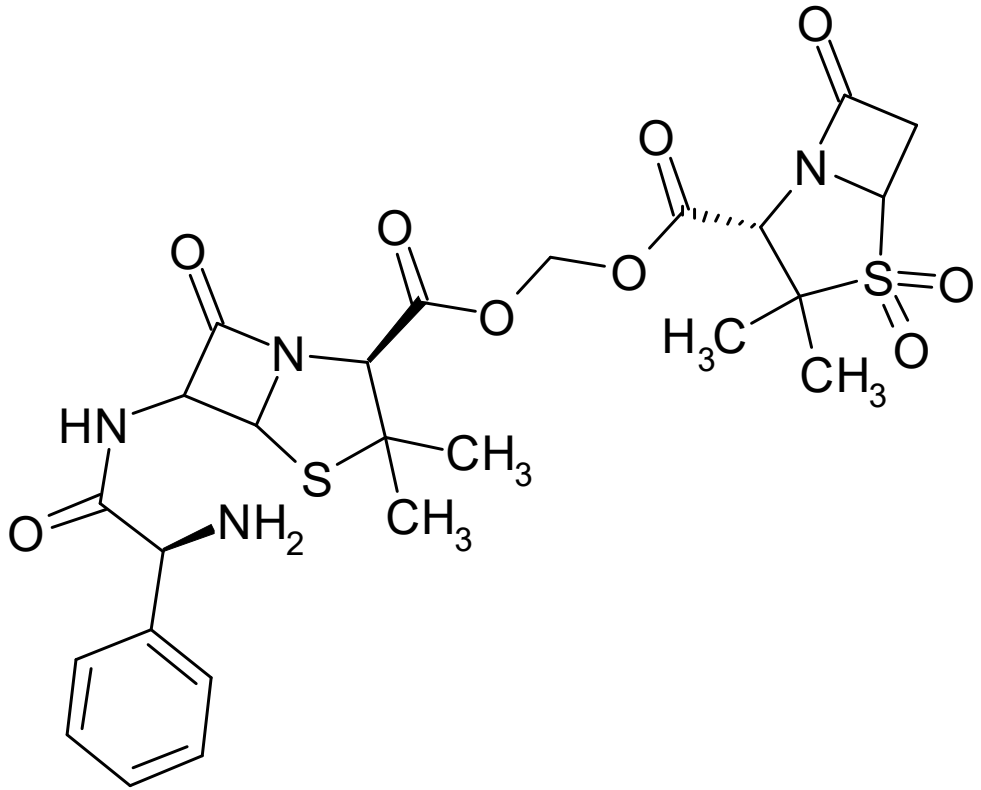
4,4-dioxopenicillanic acid
sulbactam
Betrion®
+ ampicillin (= Ampisucillin® inj. plv. sol.)



tazobactam
+ piperacillin (= Tazocin® inj. sic.)

Penicillins

A combination of a penicilline with a β-lactamase inhibitor in one molecule



•a mixed ester of ampicillin and sulbactam with methanediol

•a prodrug of both components

sultamicillin

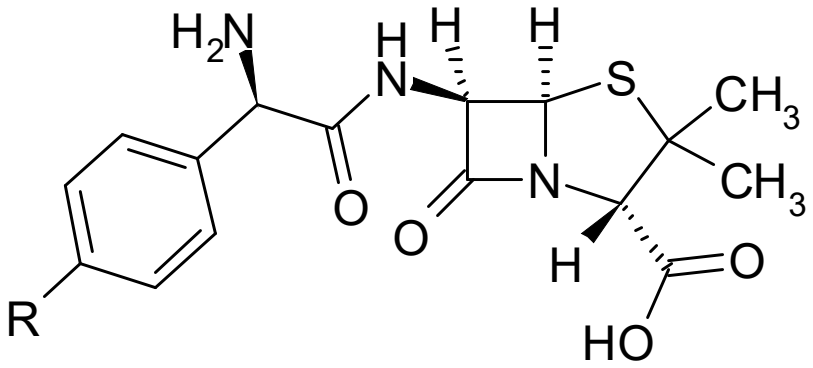
Bitamon® inj. sic., Unasyn® tbl. obd.

Penicillins

Penicillins with broadened spectrum

Ad 4. – introduction of a hydrophilic substituent to β-position of the acyl attached to amino group of 6-aminopenicillanic acid ⇒ **broadening of the antibacterial spectrum of penicillins also to G⁻ strains**

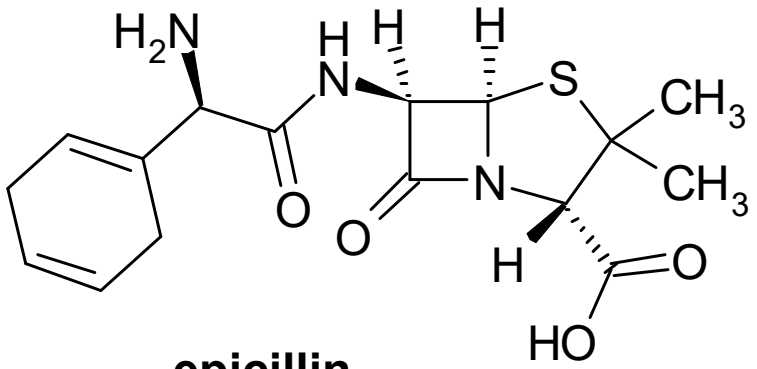
Compouns with free primary amino group



R = -H **ampicillin**

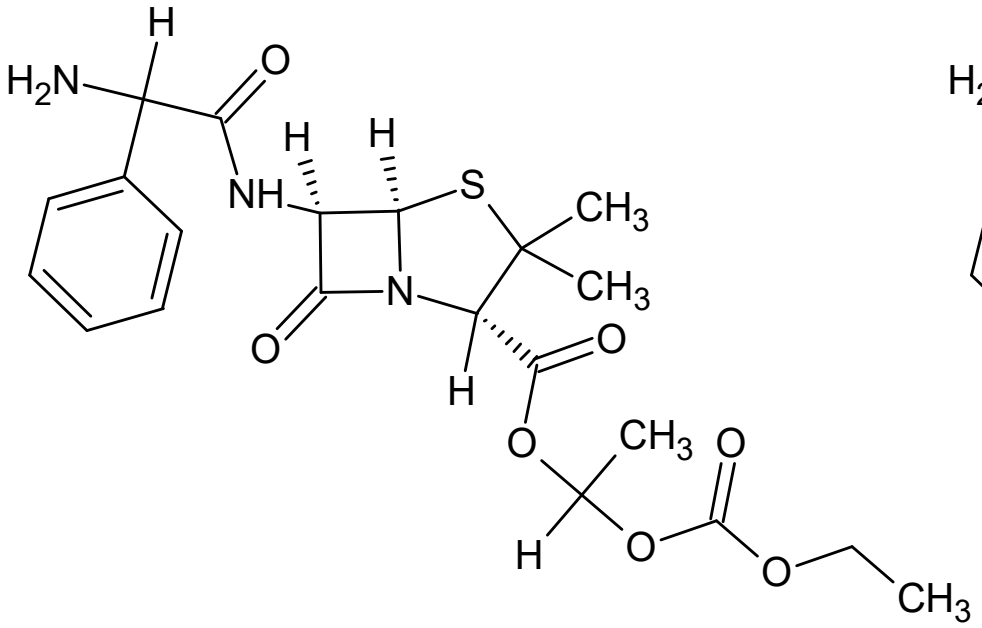
R= -OH **amoxycillin**

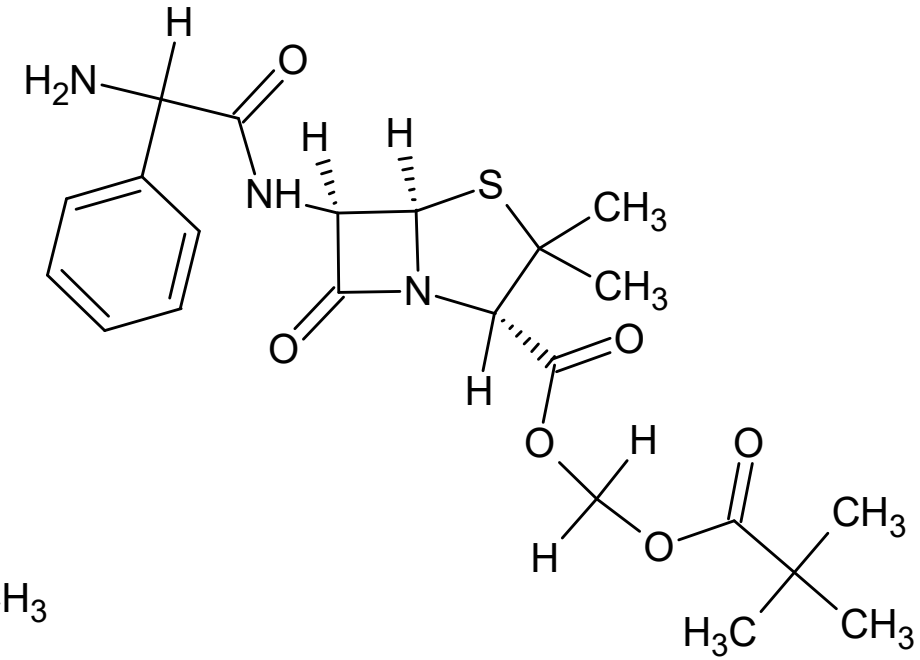
Amoclen®, Amopen®



epicillin

Penicillins with broadened spectrum
Ampicillin prodrugs





•hydrolyzed *in vivo* to ampicillin
•achieve significantly higher blood and tissue levels and attains peak blood levels more rapidly than equimolar doses of oral ampicillin
•more frequently used in veterinary (horses) than in human medicine
•models for design of prodrugs of cephalosporins

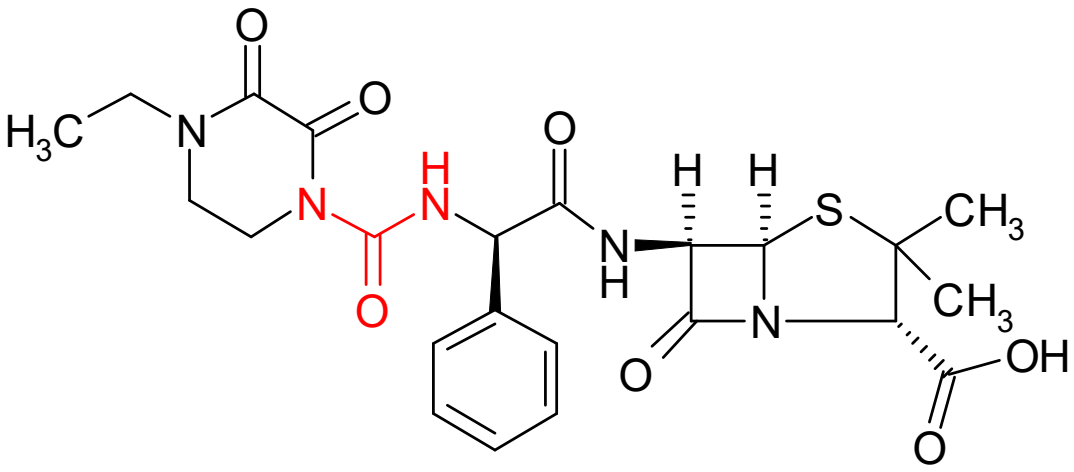
bacampicillin
ampicillin 1-(ethoxycarbonyloxy)ethylester

pivampicillin
ampicillin pivaloyloxymethylester
•successful in acute exacerbations of chronic bronchitis

Penicillins with broadened spectrum: ureidopenicillins

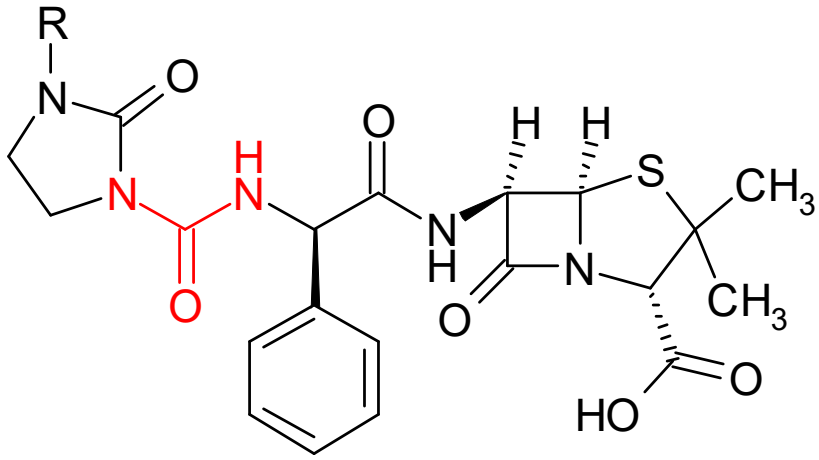
Compounds in which the amino group in β-position of the acyl is a part of urea moiety = **ureidopenicillins =** „anti-pseudomonas“ penicillins

•their spectrum includes *Pseudomonas aeruginosa*



piperacillin

Tazocin ® inj. plv. sol.(+ tazobactam)



R = H- **azlocillin**

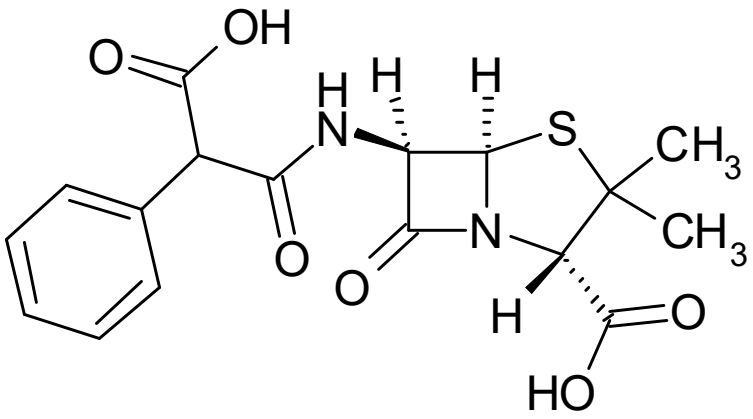
R= CH_3SO_2- **mezlocillin**

•serious infections including *otitis media,* CNS infections …

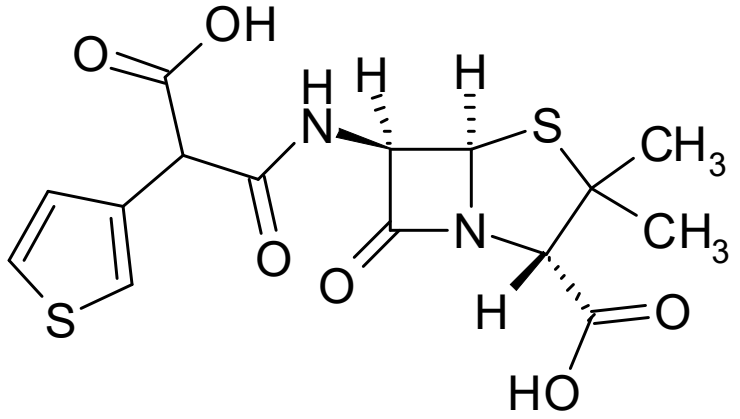
Penicillins with broadened spectrum:

•compounds with the additional carboxyl in β-position of the acyl attached to amino group in position 6

•in fact substituted malonic acids monoamides



carbenicillin



ticarcillin

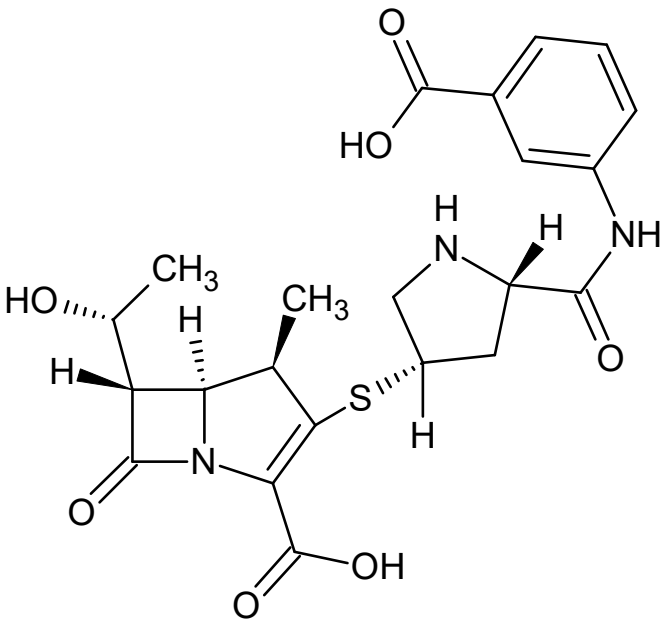
Timentin® inj. sic. (+ clavulanic acid)

•infections of bones and junctures (*Staphylococcus aureus*), gynecological & abdominal infections …

•ring analogy (benzene – thiophene)

Penems

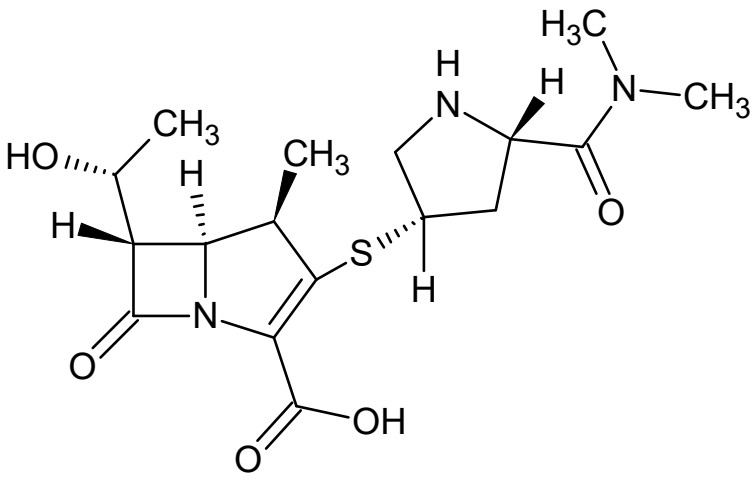
Carbapenems



ertapenem

Invanz ® plv. inf.

- pneumonias
- intraabdominal infections
- acute gynecological infections
- infections of diabetic foot



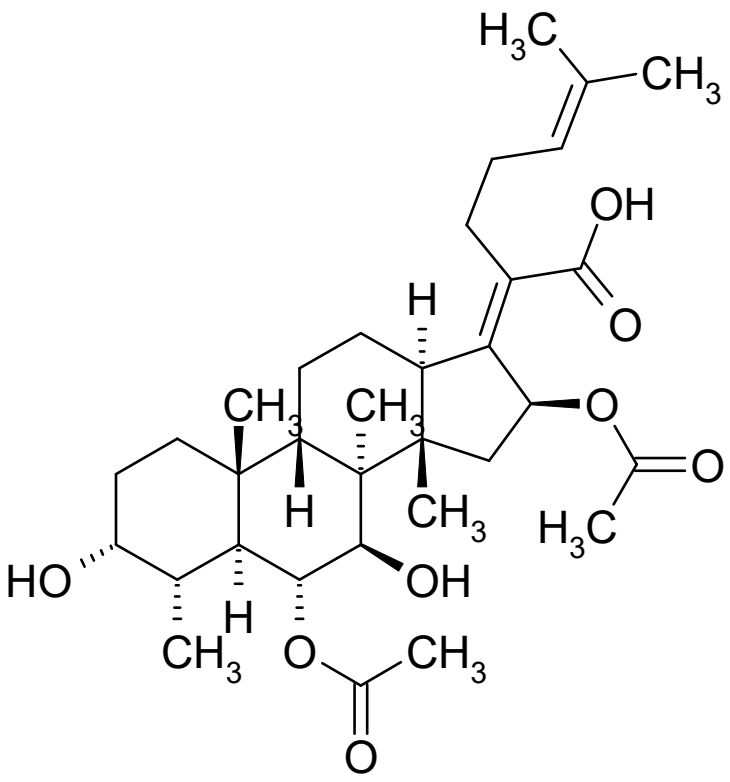
meropenem

Archifar ® plv. inf.

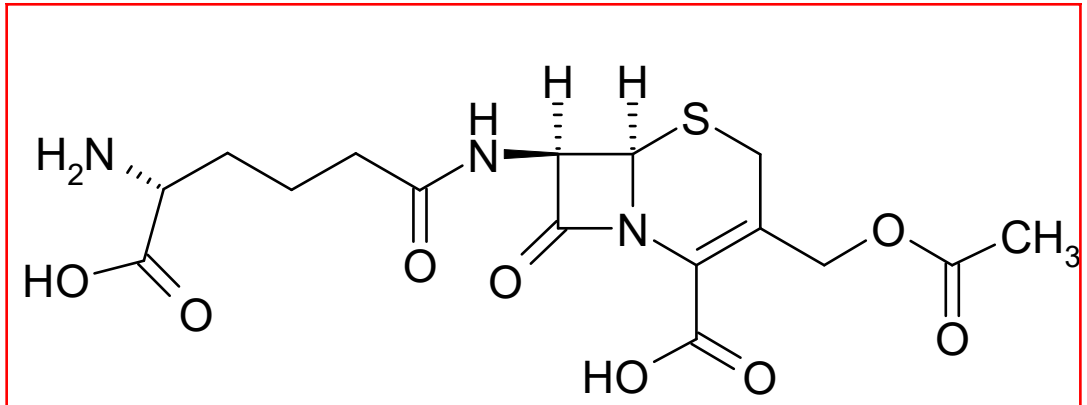
- pneumonias, bronchopulmonary infections in cystic fibrosis
- meningitides
- complicated infections of urinary tract

Cephalosporins

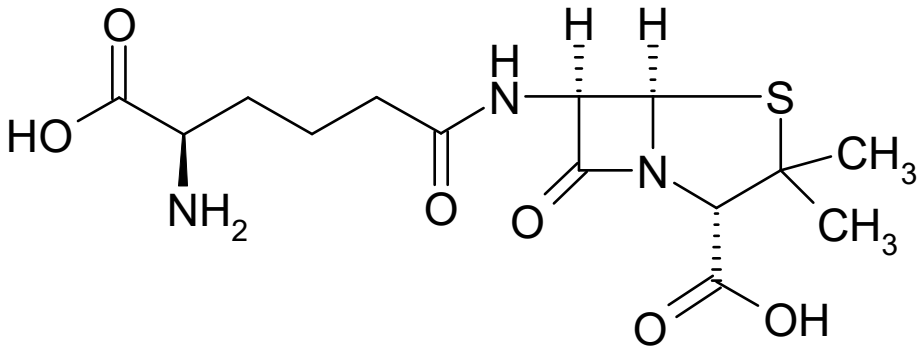
• fungi *Cephalosporium spp.* (1948)



cephalosporin P_1



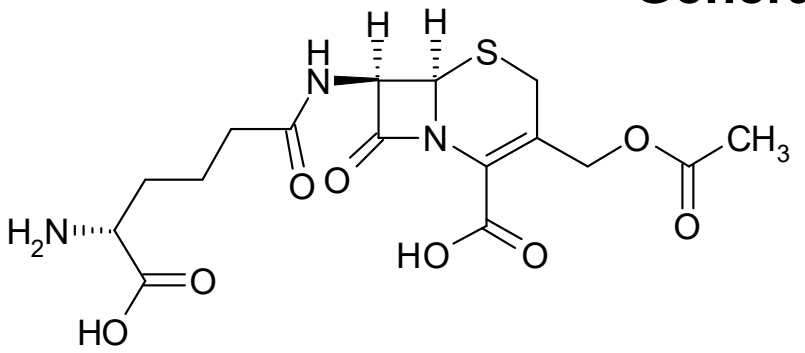
cephalosporin C



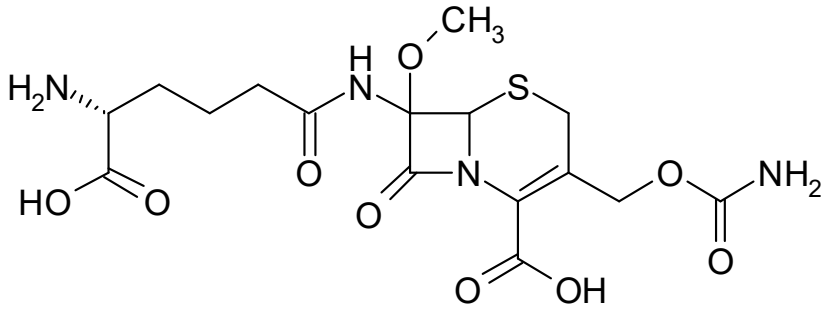
cephalosporin N

… and other various structures

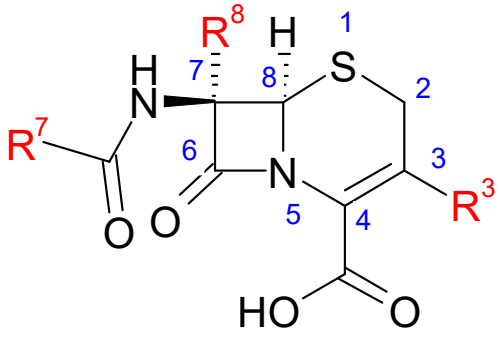
Cephalosporins
General structure



cephalosporin C
•isolated from *Cephalosporium spp.*

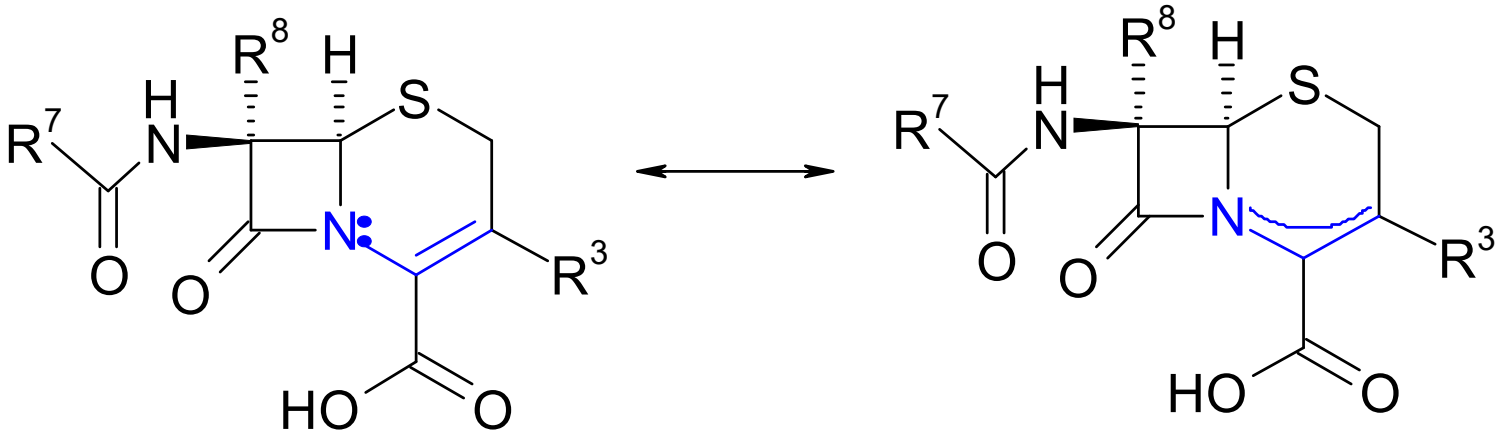


cephamycin C
•isolated from *Streptomyces lactadurans*



Cephalosporins

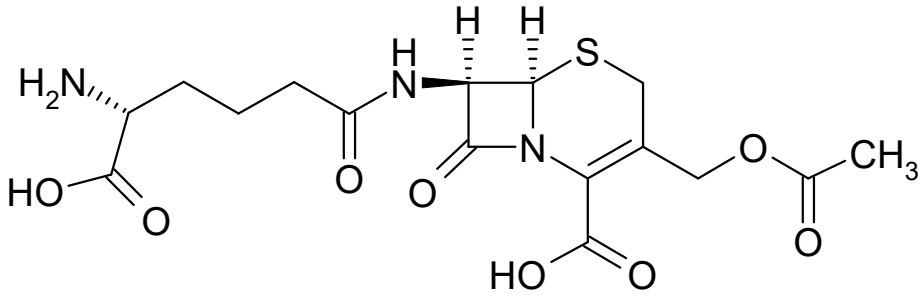
Properties



- electron pair on N5 is linked to conjugation with double bond ⇒ ↓ of electron density on N5 ⇒ ↓ of nucleophilicity of N5 ↑ stability in acid media
- also ↑ resistance to β-lactamases (cefalosporinases)

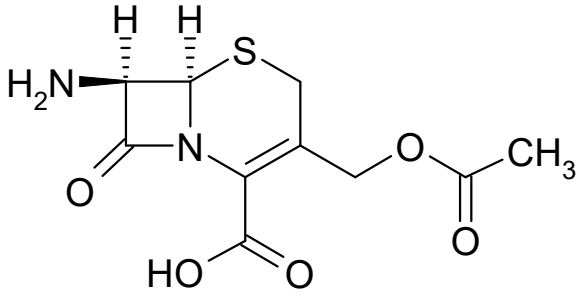
Cephalosporins

Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid.

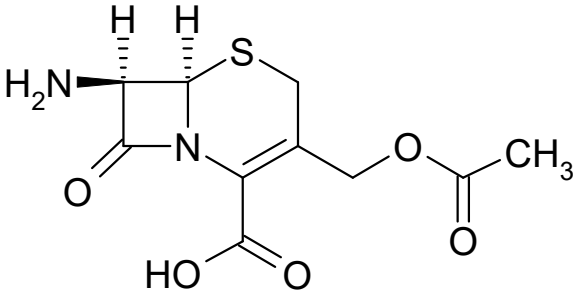


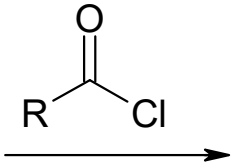
cephalosporin C

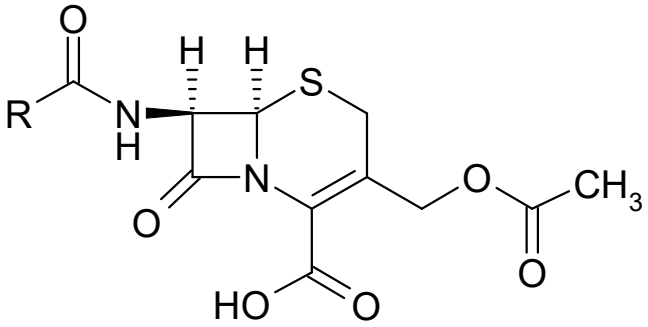
NOCl
or enzymes*



7-aminocefalosporanic acid







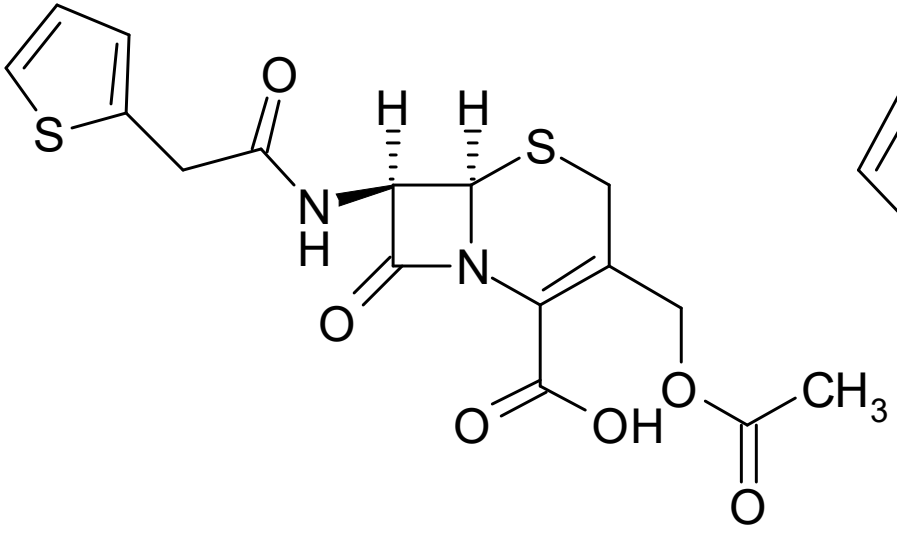
a semi-synthetic cephalosporin, or an intermediate

*glutarylacylase + D-amino acid oxidase

Cephalosporins

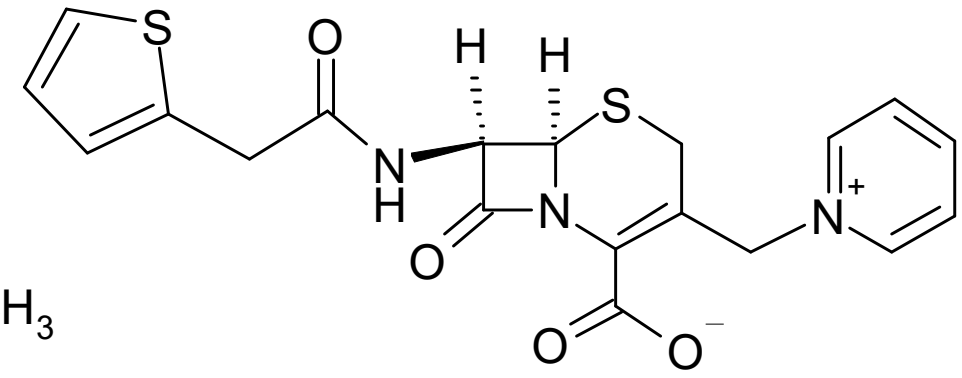
Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid

1st generation: for parenteral administration only (not absorbed from GIT)



cephalotin

Cefalotin® Biotika inj. sic.

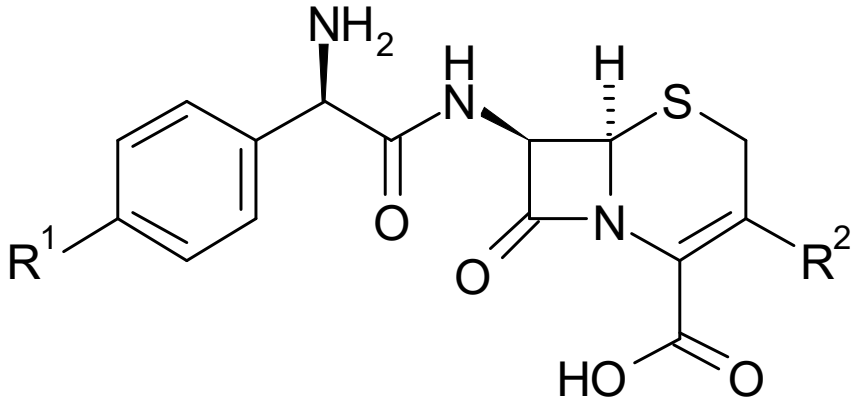


cefaloridin

Cephalosporins

Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid

2nd generation: for oral administration

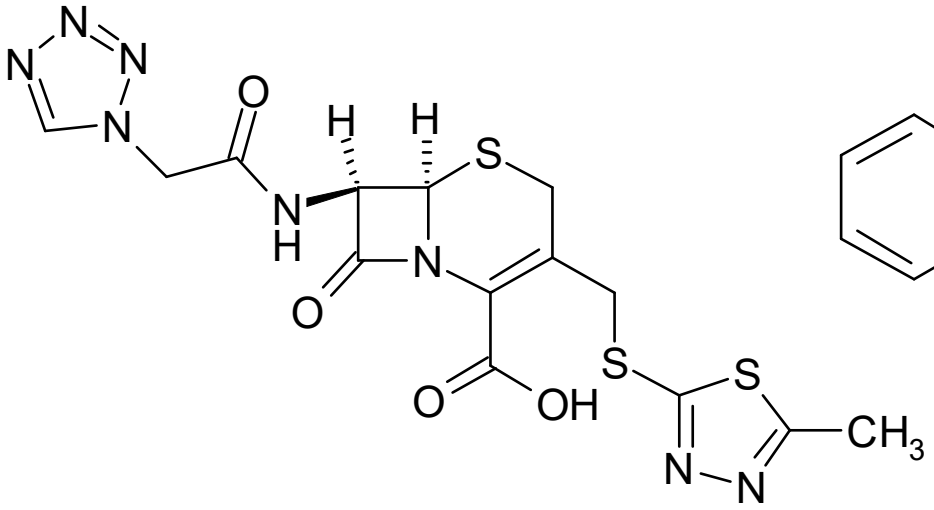


R^1= -H, R^2= $-CH_3$	**cefalexin**	Cefaclen® cps.
R^1= -OH, R^2= $-CH_3$	**cefadroxil**	Biodroxil® tbl. obd.
R^1= -H, R^2=Cl	**cefaklor**	Ceclor® cps.

Cephalosporins

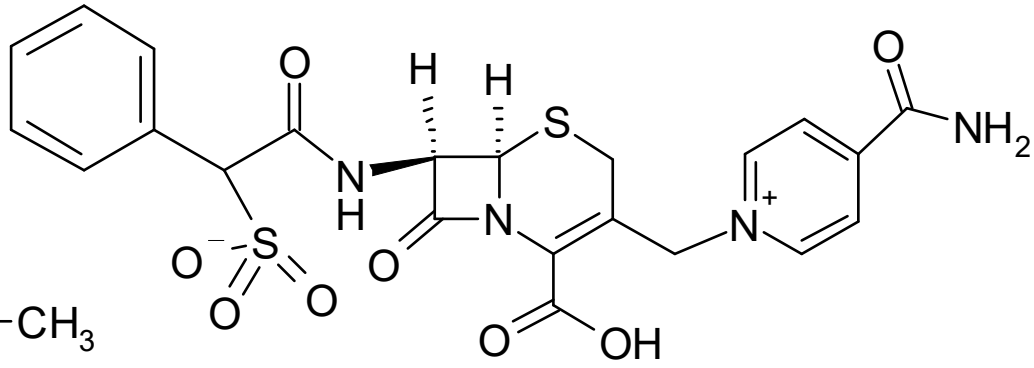
Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid

2nd generation: for parenteral use but with ↑ effect to G^-, ↑ resistance to β-lactamases



cefazolin

Kefzol® inj. sic.



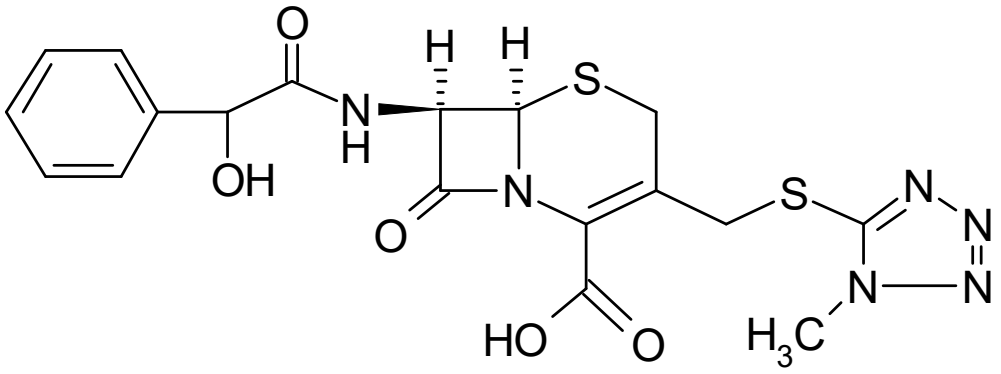
cefsulodin

·*Pseudomonas*

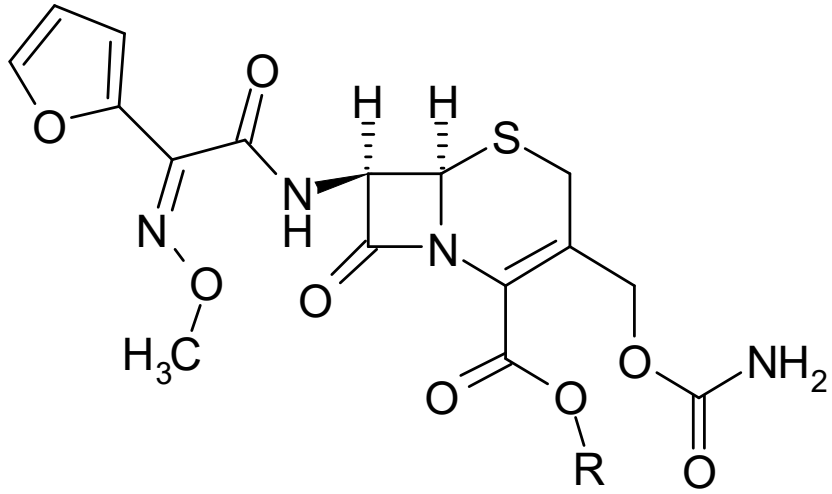
Cephalosporins

Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid

2nd generation: for both parenteral and p.o. administration, very resistant to ✔-lactamase

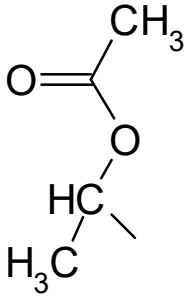


cefamandole



R = H- **cefuroxime**

Ceroxim® tbl.

R = 

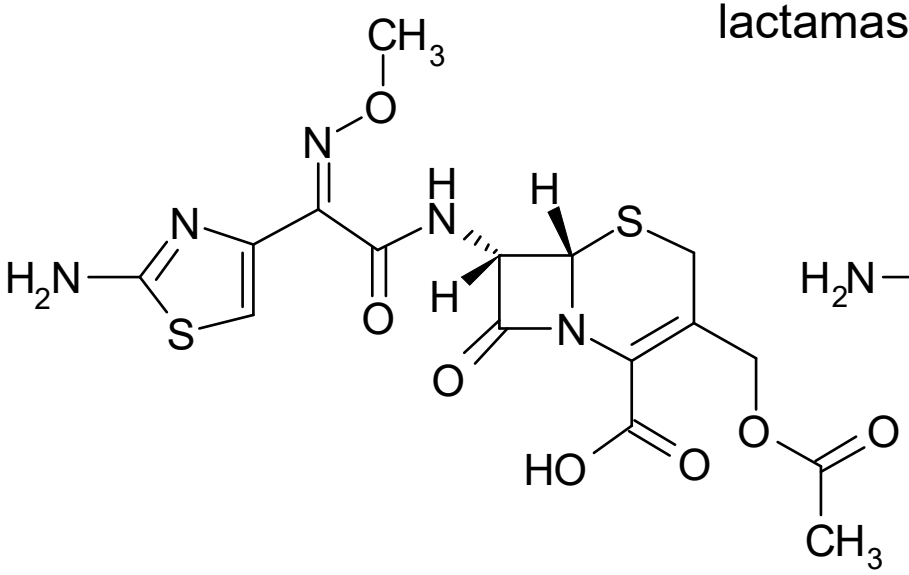
cefuroxime axetil

Zinnat® tbl. obd.

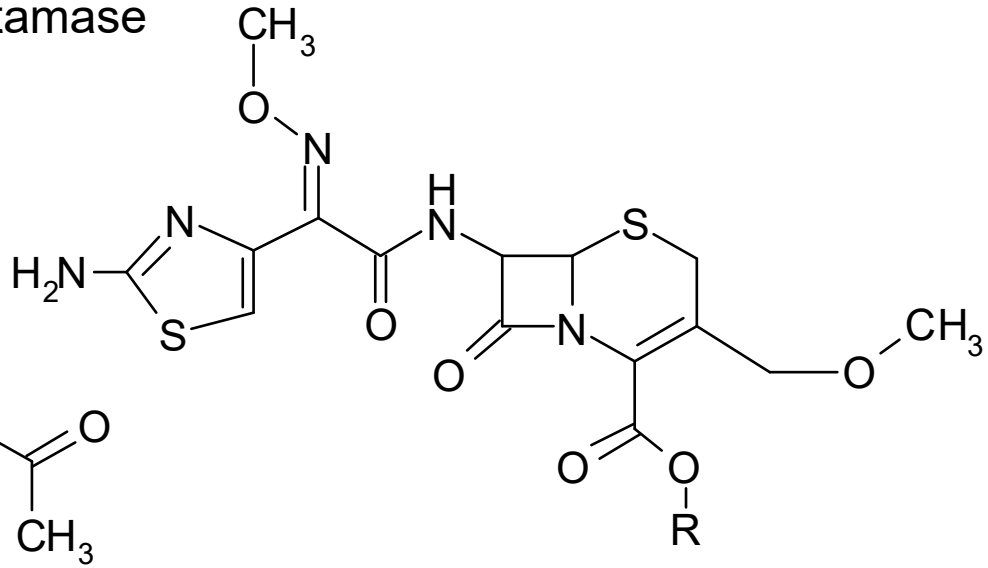
Cephalosporins

Compounds related to cephalosporin C, i.e. N-acylderivatives of 7-aminocephalosporanic acid

3rd generation: for both parenteral and p.o. administration, very resistant to ✔-lactamase

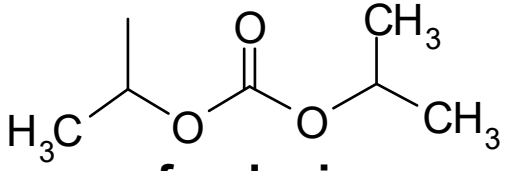


cefotaxime
Claforan® inj. sic.



R = H- **cefpodoxime**

R =

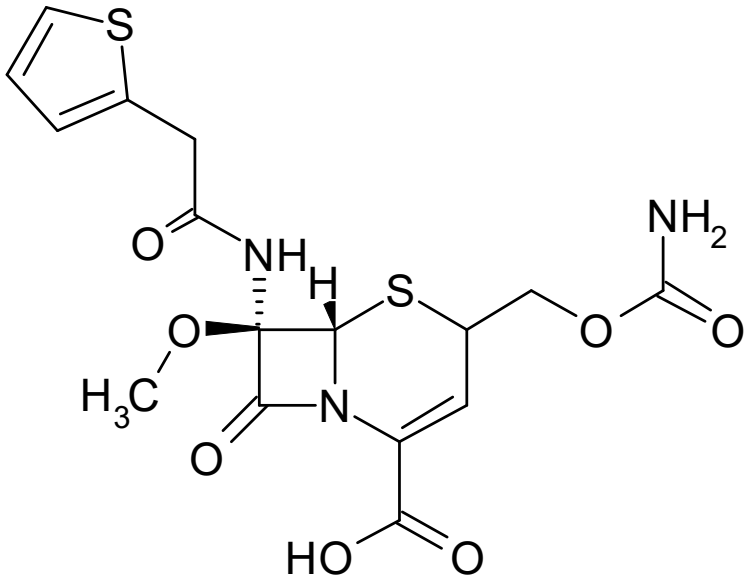


cefpodoxime proxetil
Orelox® tbl. obd.

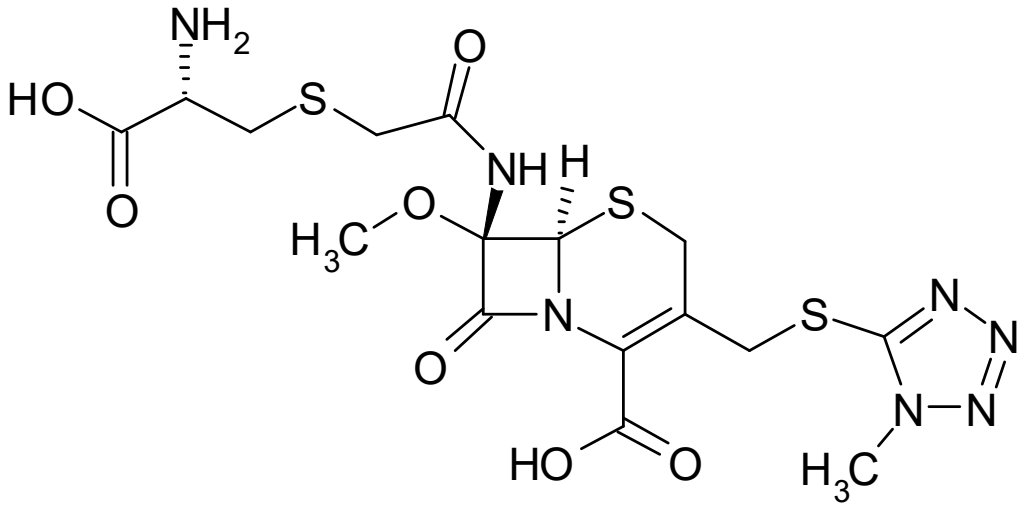
Cephalosporins

Compounds related to cephamycin C, i.e. N-acylderivatives of 7-methoxy-7-aminocephalosporanic acid

„New class“ – for both parenteral and p.o administration – resistant to β-lactamase

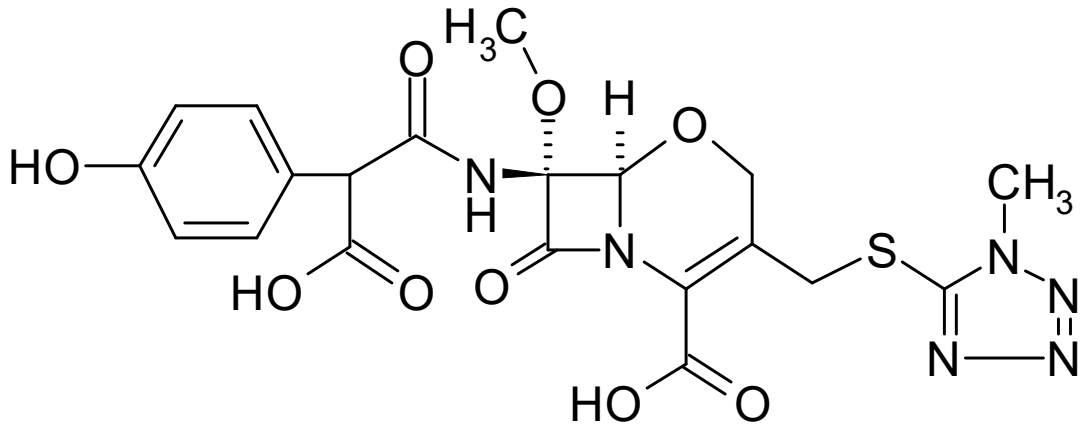


cefoxitin
Mefoxin® inj. sic.



cefminox

Cephalosporin analogues



moxalactam

•dihydrooxazine derivative related to 4th generation of cephalosporins

•developed especially for treatment of meningitis (crosses the blood-brain barrier) and anaerobic infections

Macrolides

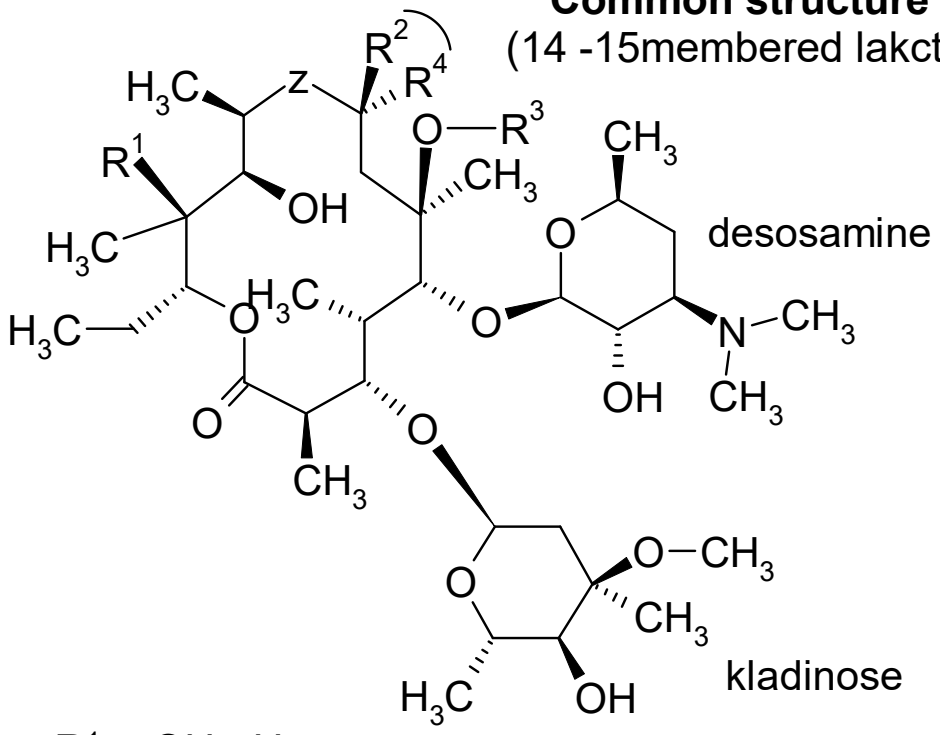
·makrocyclic lactones with 10 – 40membered ring with 1 aminomonosaccharide and 1 „neutral“ monosacharide which can have an additional aminosaccharide attached

·1st group (with larger ring)- natamycine, nystatine, amphothericine B – see antimycotics

·2nd group – **erythromycine group** (erythromycine and its analogues, spiramycine, tylosine)

Common structure of narrower group of erythromycine

(14 -15membered lakctone ring - erythromycine and analogues)



R^1= -OH, -H

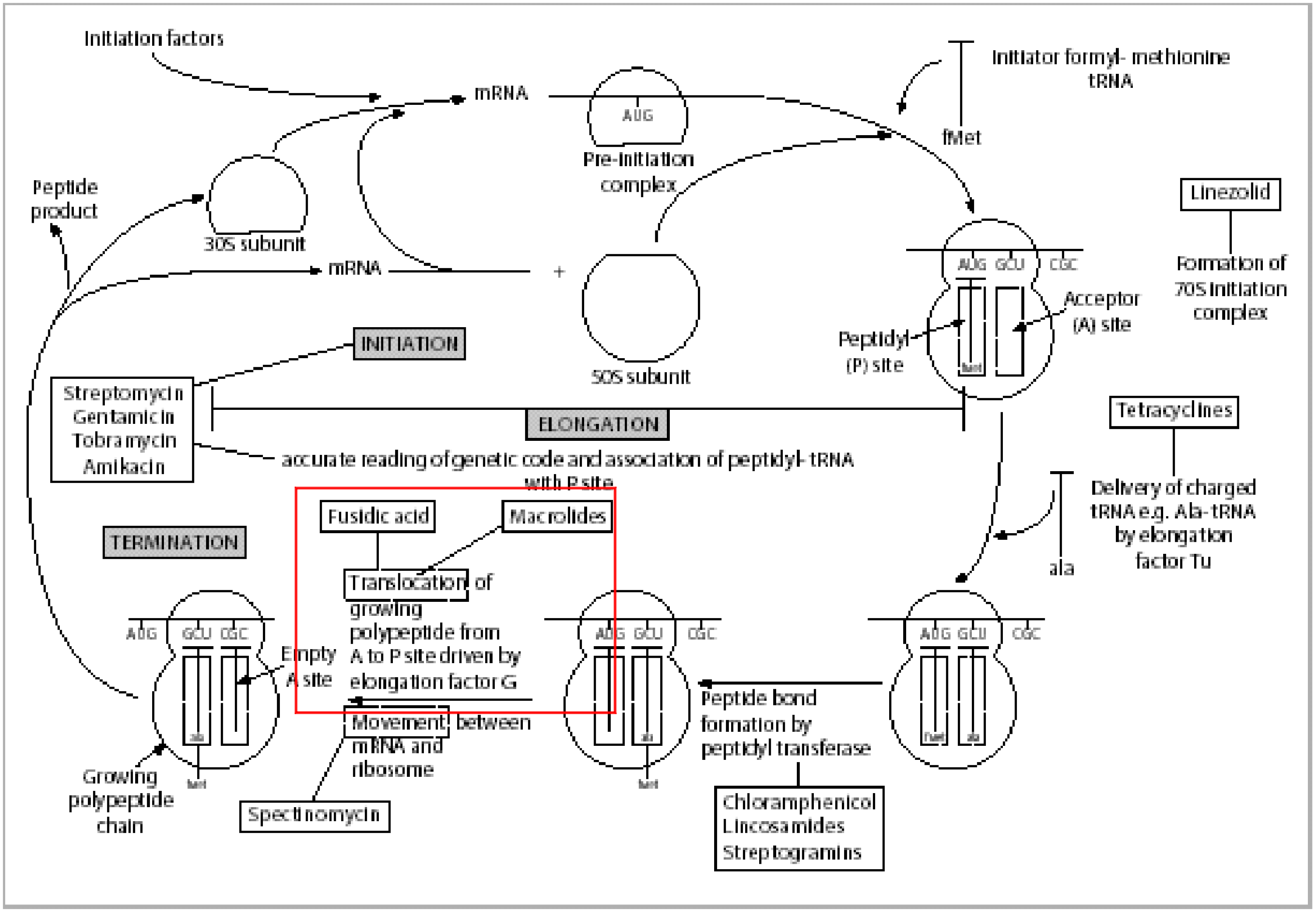
Z = ⇧C=O, $-CH_2N(CH_3)-$, ⇧C=N-O-CH_3, ⇧C=N$OCH_2OCH_2CH_2OCH_3$

R^2= -H, -F

R^3= -H, -CH_3

R^4= -CH_3 or R^2 + R^4= oxirane

Macrolides
Site & mechanism of action



Macrolides

Site and mechanism of action

- **Proteosynthesis inhibition**
- act at 50S ribosome subunit
- inhibit the translocation of growing peptide from acceptor (A) to peptide (P) site
- **bacteriostatic effect**

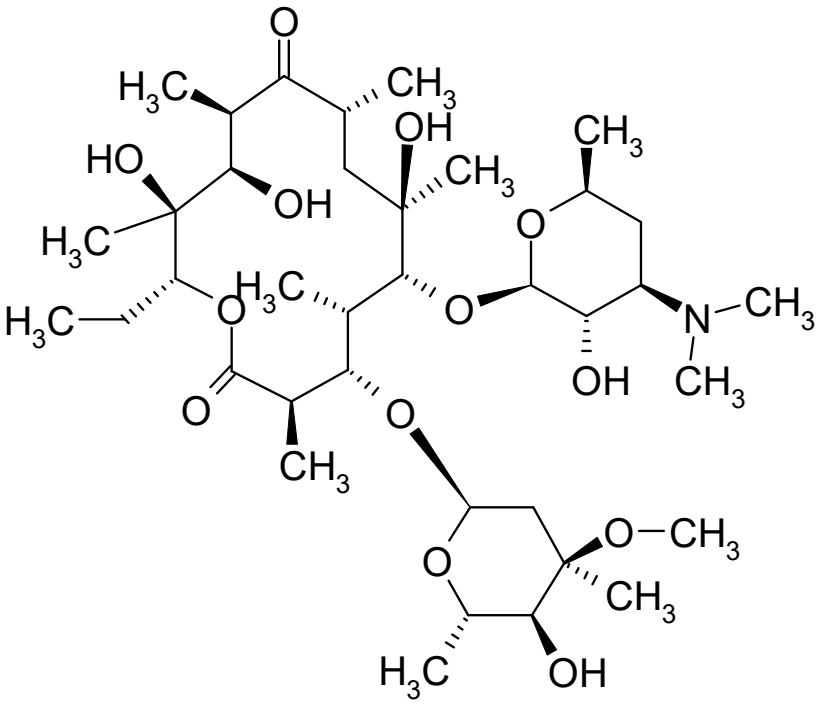
Spectrum:

- both G^+ and G^-

Neisseria, Haemophillus, Brahmanella, Legionella …

Macrolides

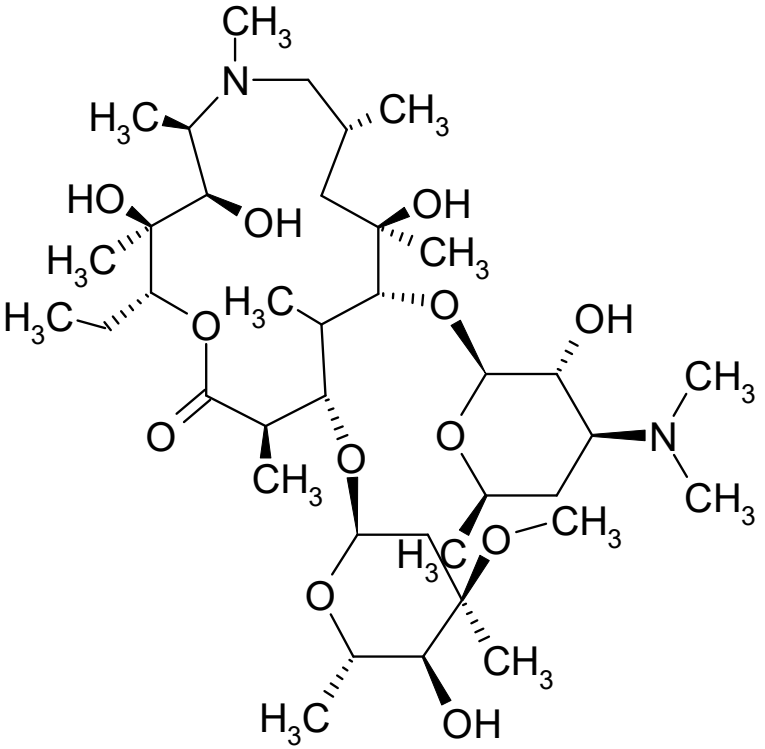
Erythromycine and its analogues



erythromycine

- isolated 1952 from *Streptomyces erythreus*
- poor biological availability ⇒ lipophilic salts (stearate, ethylsuccinate …)
- external form (lotions …) – treatment of *acne vulgaris*

Porphyrocin® tbl.



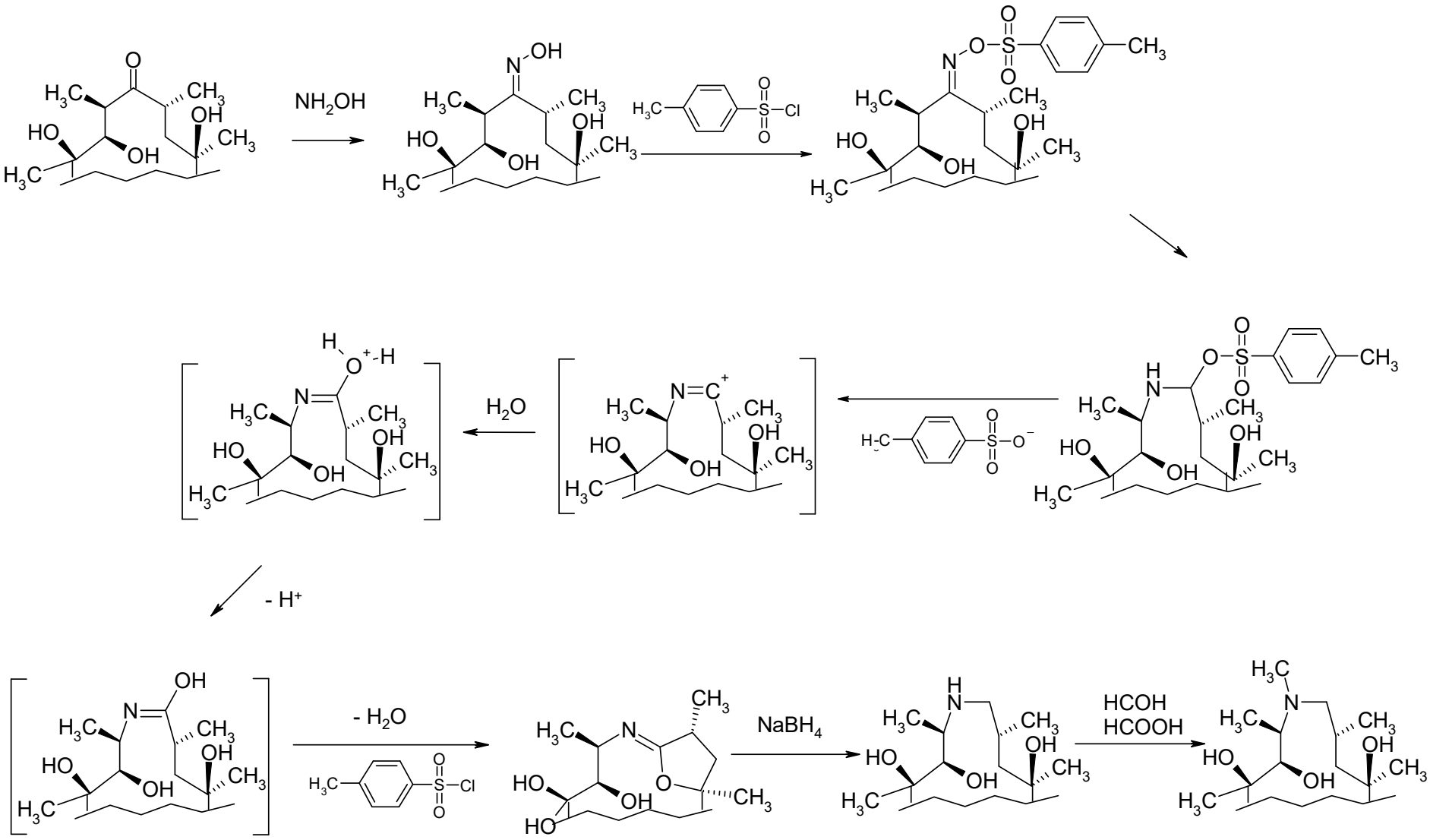
azithromycine

- semi-synthetic compound

Sumamed® tbl. obd.

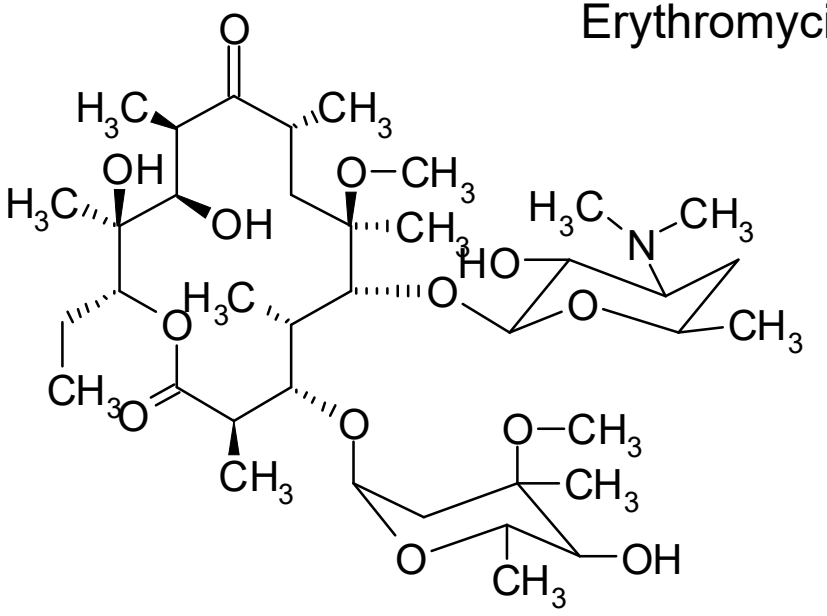
Macrolides

Synthesis of azithromycine from erythromycine

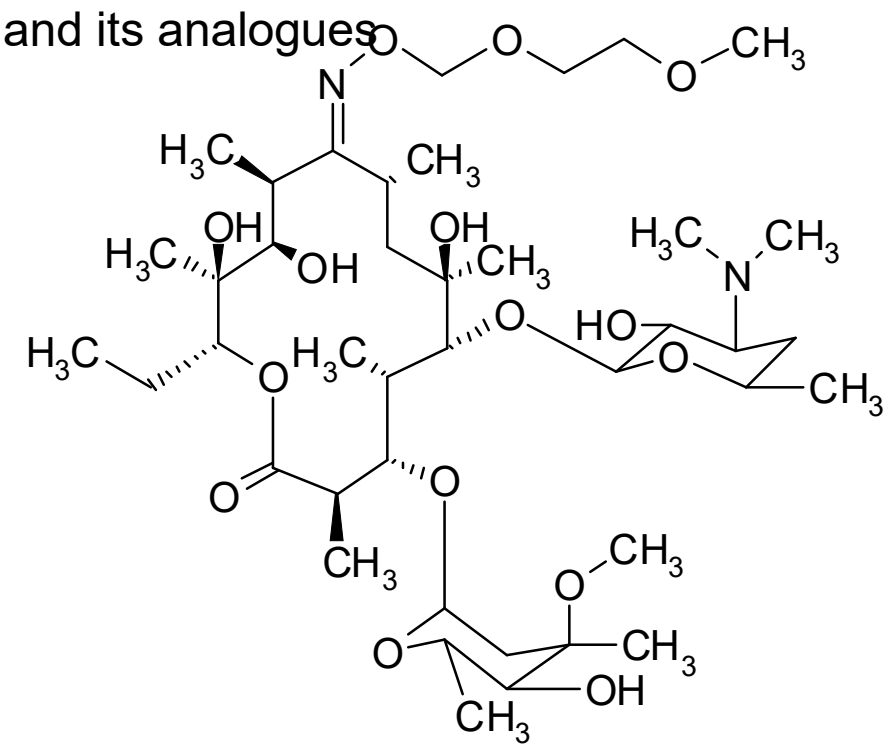


Macrolides

Erythromycine and its analogues



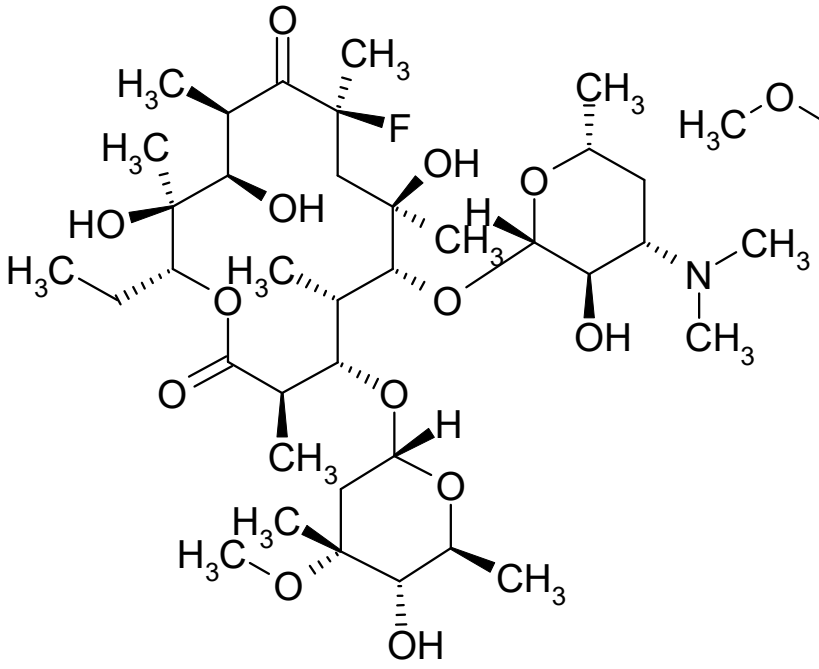
6-*O*-methylerythromycine
clarithromycine
·also some strains of *Mycobacterium avium*
Klacid® tbl. obd.



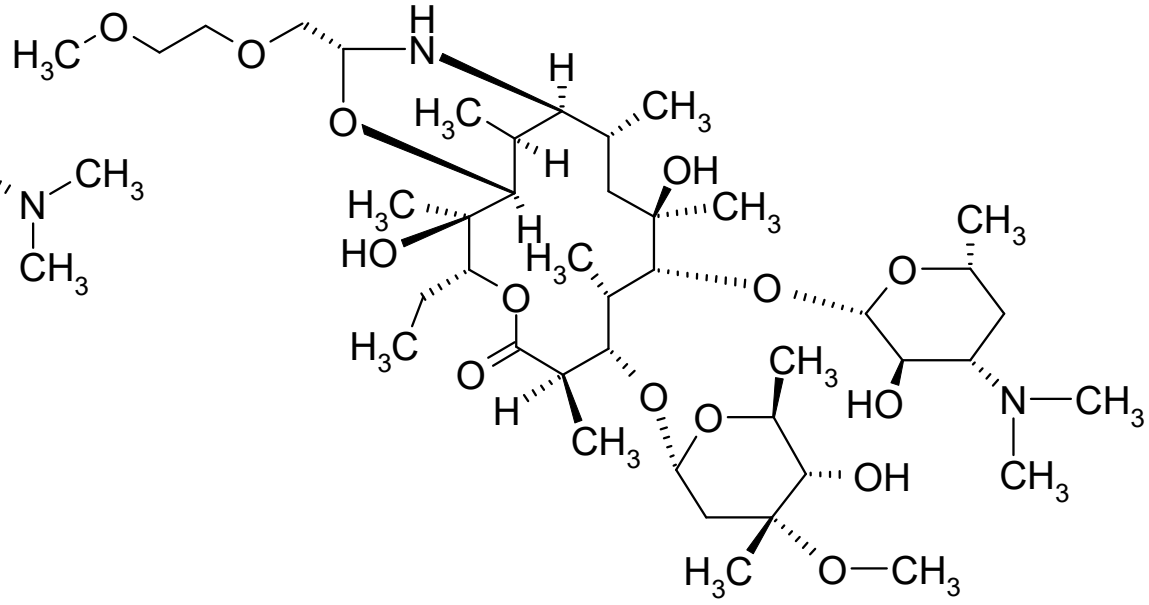
roxithromycine
Rulid® tbl.

Macrolides

Erythromycine and its analogues



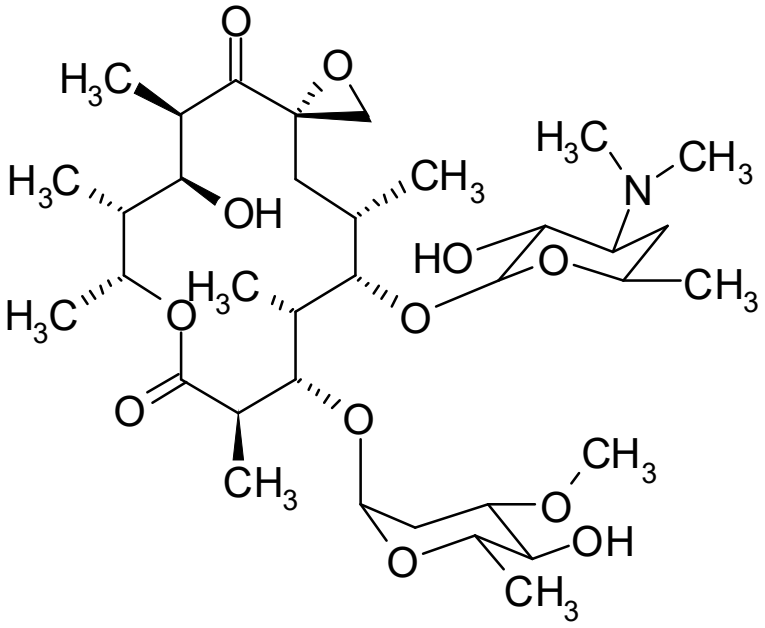
8-fluoroerythromycine
flurithromycine



dirithromycin

Macrolides

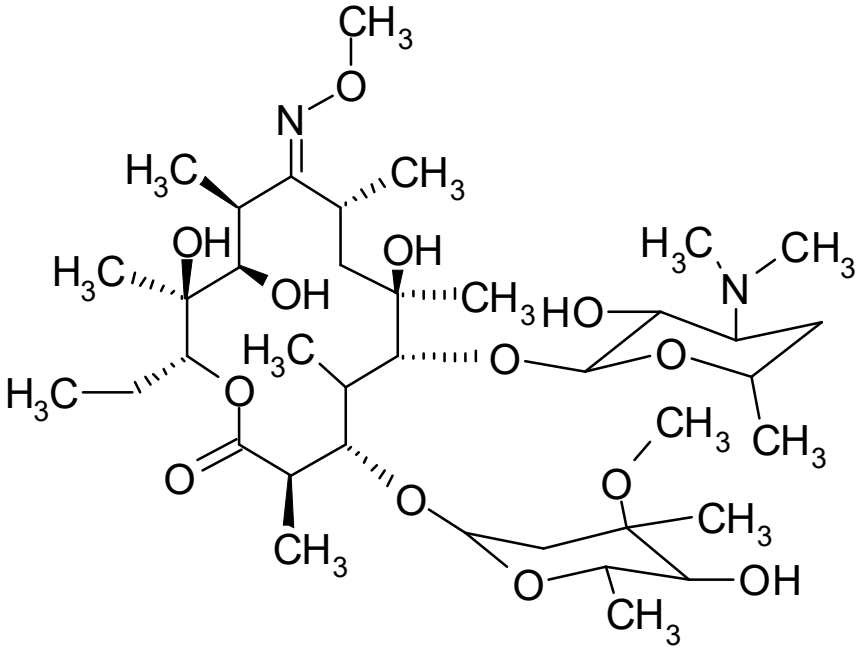
Erythromycine and its analogues



L-oleandrose

oleandomycine

·isolated 1954 from *Streptomyces antibioticus*

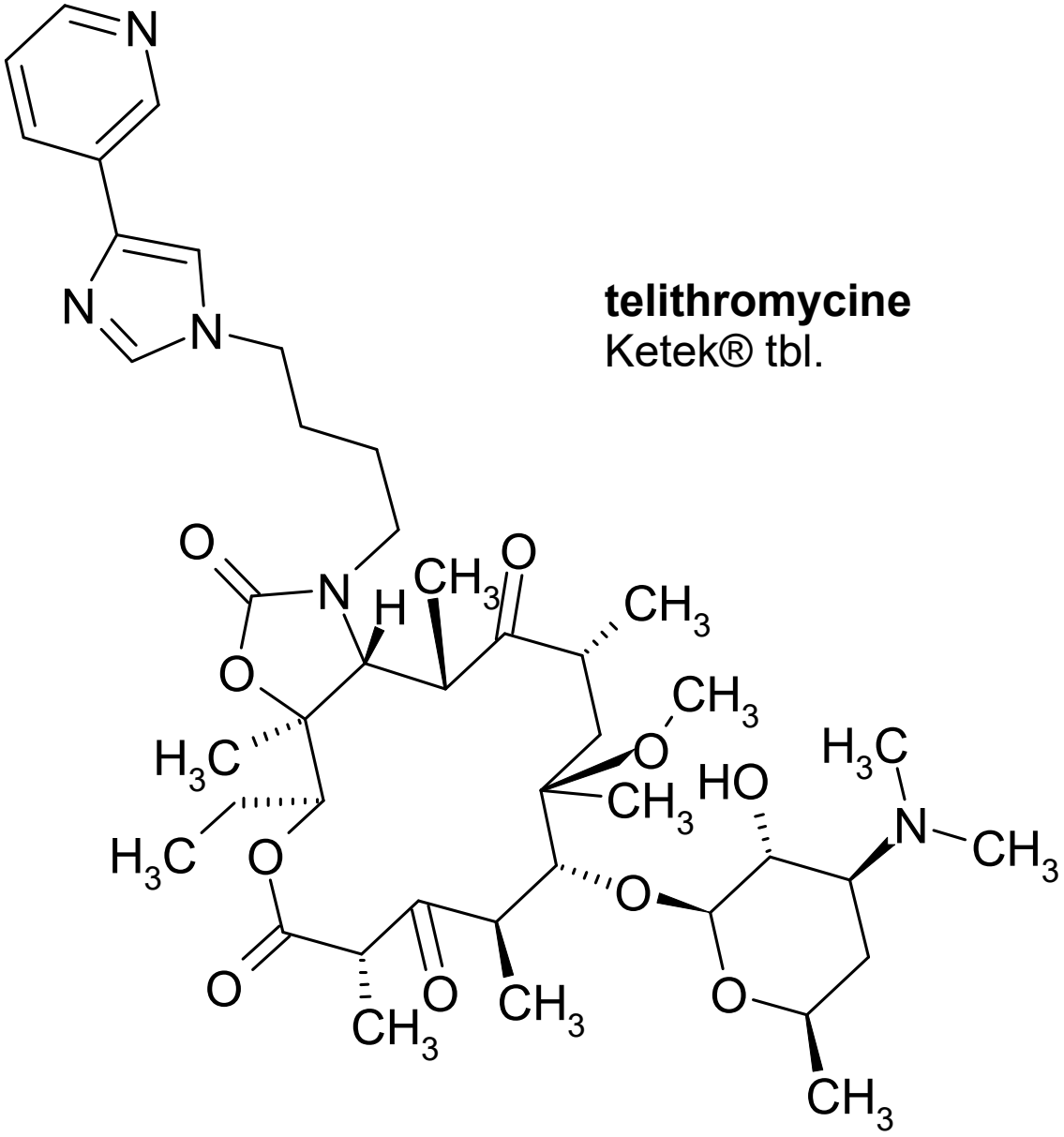


lexithromycine

„More free“ erythromycine analogues: **Ketolides**

•2 keto-moieties on lactone ring (+ 1 ester carbonyl + 1 cabamate carbonyl)

•good biological availability

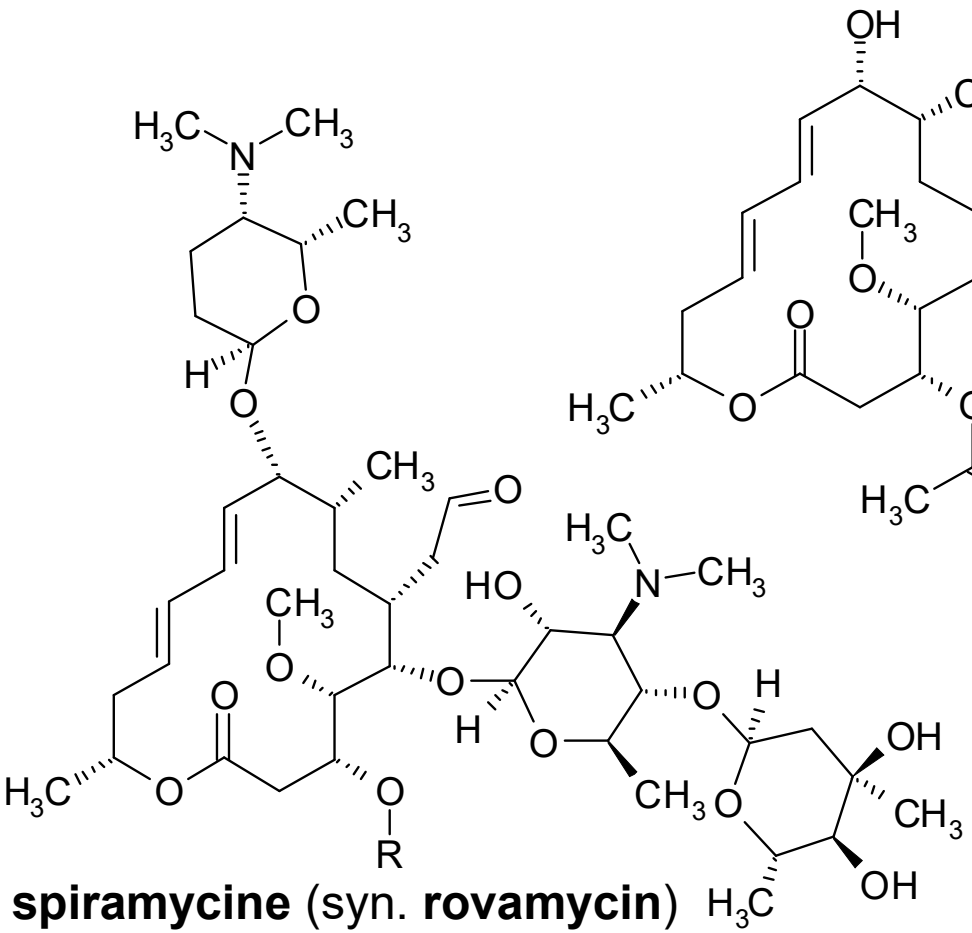


telithromycine

Ketek® tbl.

Macrolides

Compounds with 16membered lactone ring unsaturated in positions 10 and 12



spiramycine (syn. **rovamycin**)

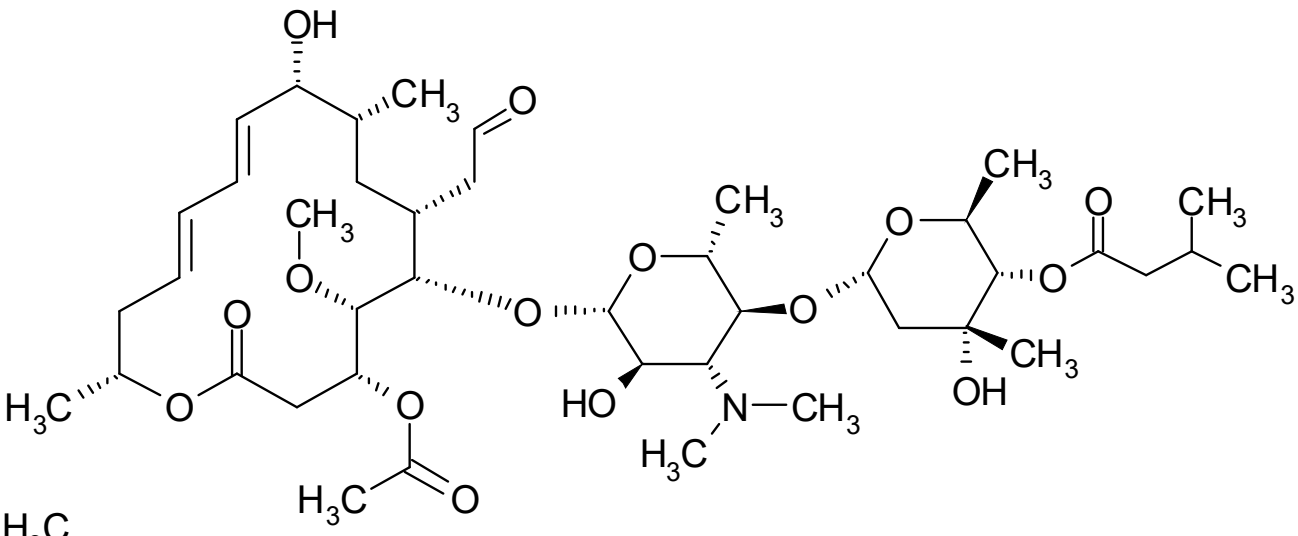
·isolated 1954 from *Streptomyces ambofaciens*

R = HCO- spiramycine A = spiramycine I

CH_3CO- spiramycine B = spiramycine II

CH_3CH_2CO- spiramycine C = spiramycine III

Rovamycine® tbl. obd.



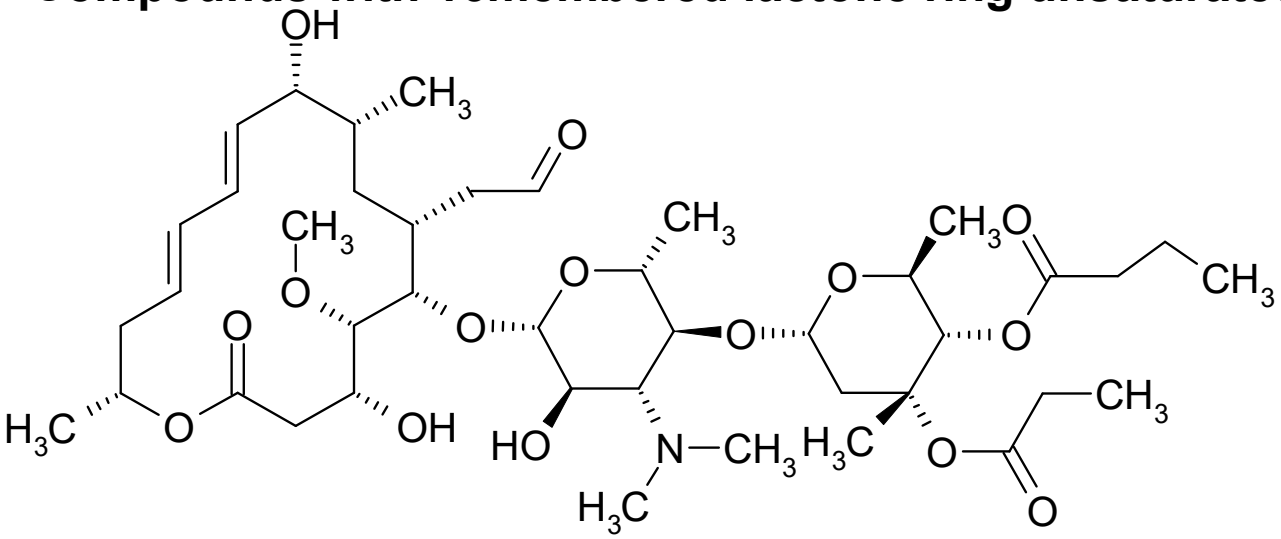
josamycin

·also *Mycoplasma*

Wilprafen® tbl. obd., Alpucine® premix a.u.v.

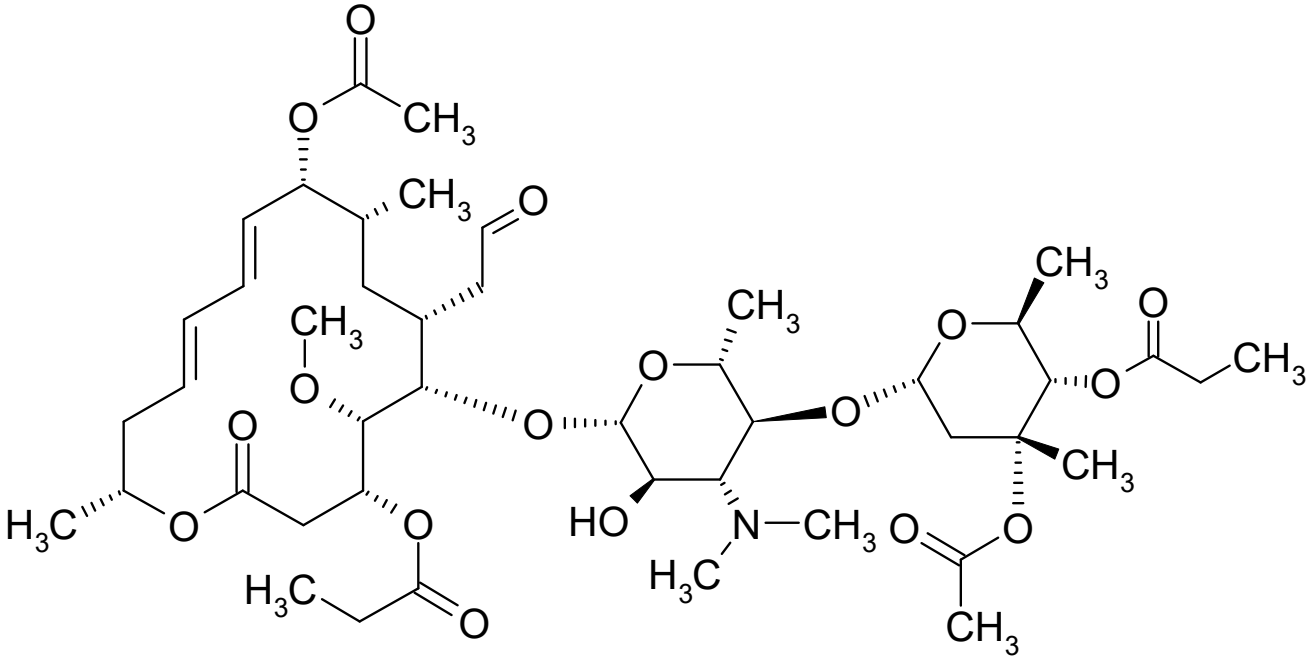
Macrolides

Compounds with 16membered lactone ring unsaturated in positions 11 and 13



rokitamycin

miokamycin



Aminoglycosides

- aminosaccharide glycosides produced by strains of *Streptomyces* genus
- Streptomycin group
- Neomycin group
- Kanamycin and gentamycin group

Mechanism of action

- **protheosynthesis inhibition**
- they avoid accurate reading of the genetic code and binding of peptidyl-tRNA to the peptide binding site
- **effect bacteriostatic – bactericidal**

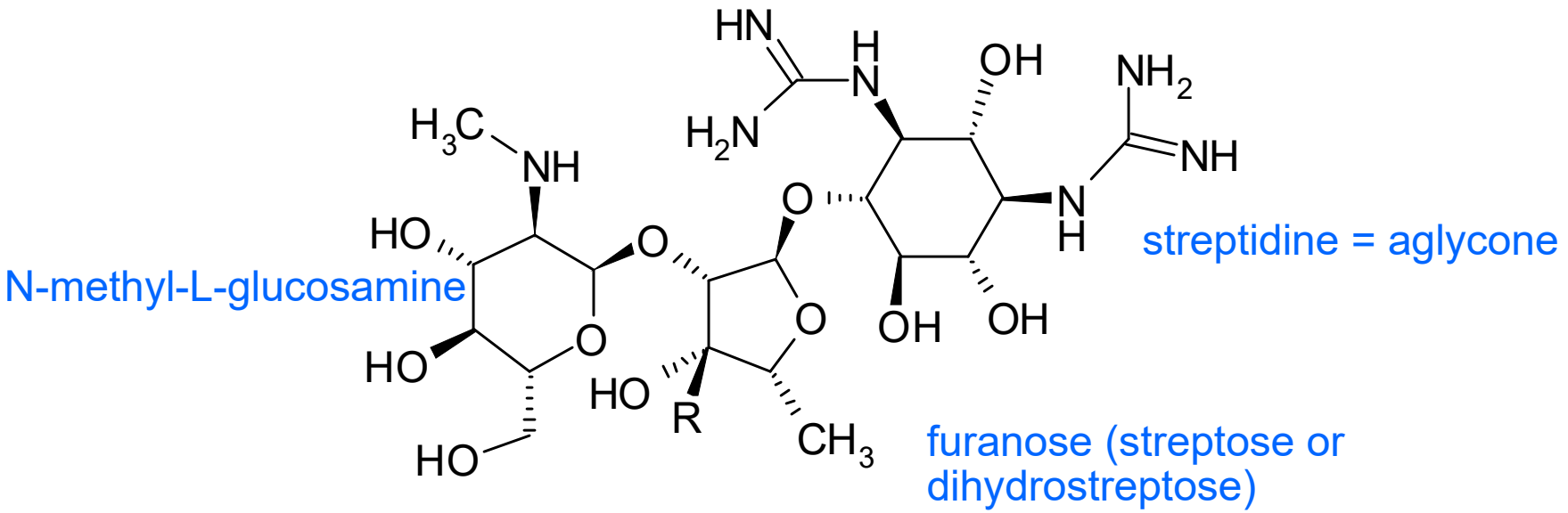
Spectrum

$G^+ < G^-$

Bacillus anthracis, Bordetella pertussis, Brucella, Corynebacterium diphteriae, E. coli, Enterobacter, Haemophillus, Mycobacterium tuberculosis…

Aminoglycosides

1. Streptomycin group



R = -CHO **streptomycin**

- isolated 1944 from *Streptomyces fradiae*
- used to *M. tuberculosis* in combination with other tuberculostatics
- bactericidal

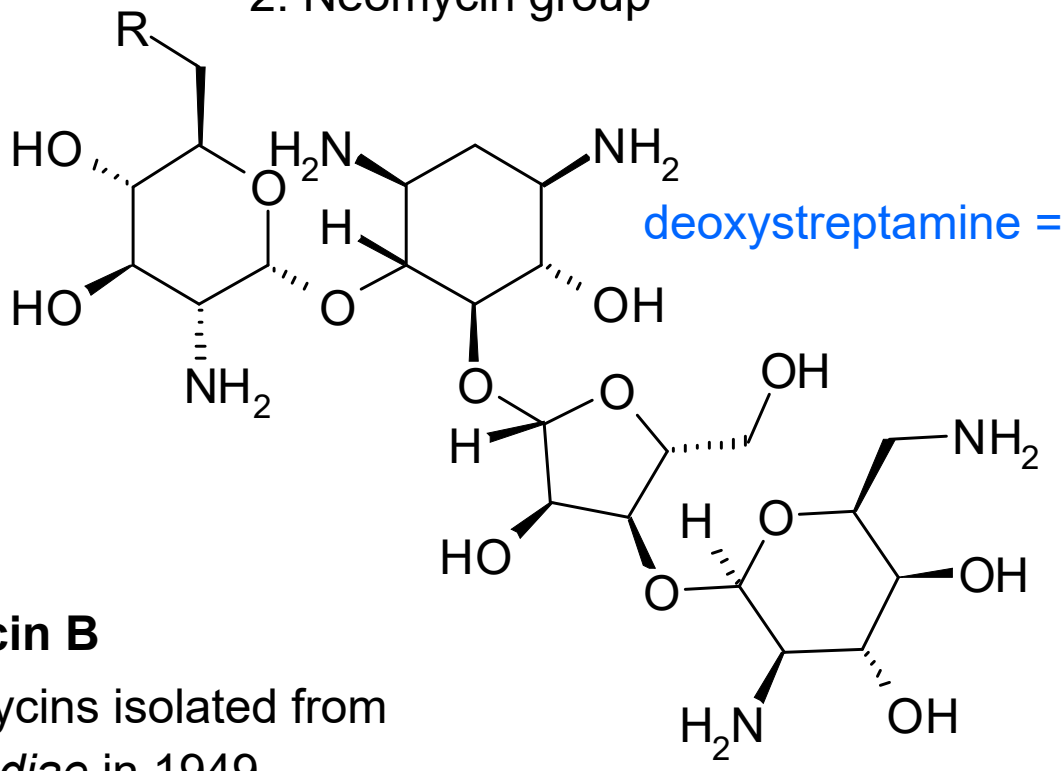
Streptomycin „Grünenthal“® inj. sic., Streptowerfft® a.u.v

R = -CH_2OH **dihydrostreptomycin**

Depomycine® a.u.v. inj. (+ benzylpenicillin)

Aminoglycosides

2. Neomycin group



deoxystreptamine = aglycon

R= -NH_2 **neomycin B**

- mixture of neomycins isolated from *Streptomyces fradiae* in 1949

Framykoin® ung., Pamycon® plv. (+ bacitracin)

R = -OH **paromomycin**

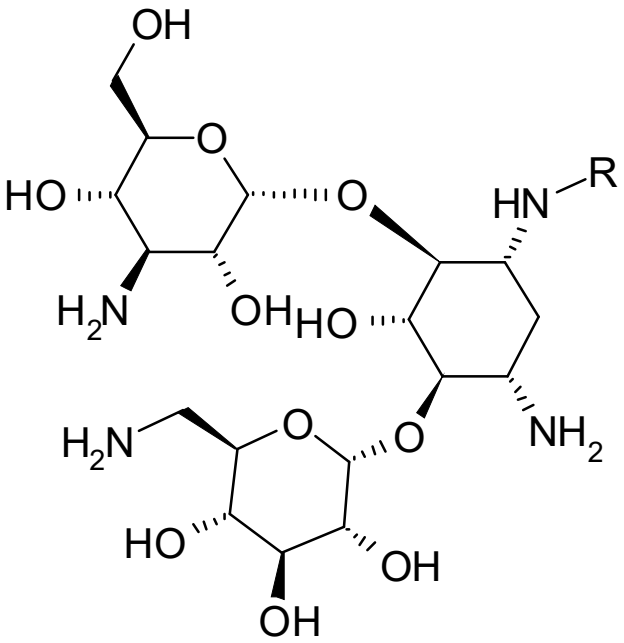
- not absorbed from GIT
- used for *Entamoeba histolytica*

Humatin® cps.

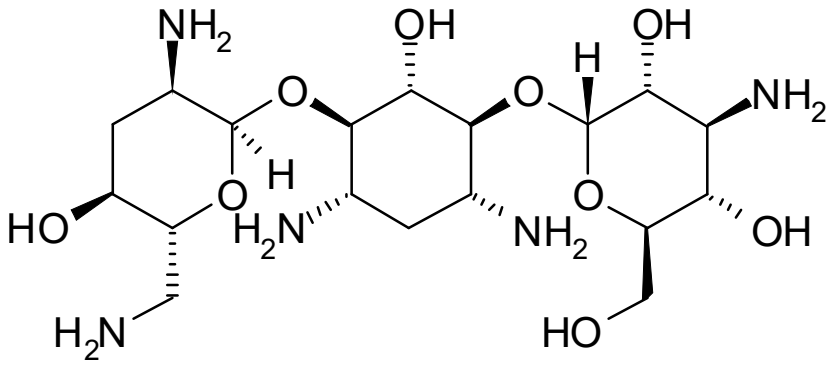
Aminoglycosides

3. Group of kanamycin and gemtamycin

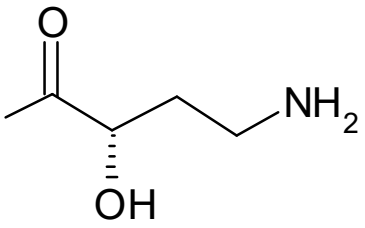
Kanamycin subgroup



R = -H **kanamycin**
Kanacol® a.u.v. inj.



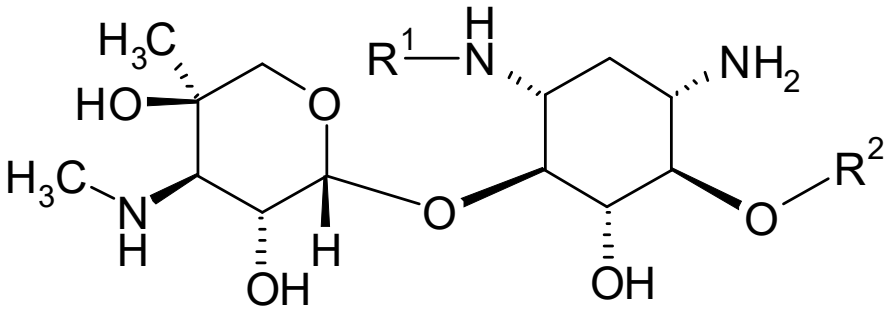
tobramycin
Tobi Nebuliser Solution® inh. sol.
•treatment of chronic pulmonary infection caused by *Pseudimonas* in patients with cystic fibrosis

R= 

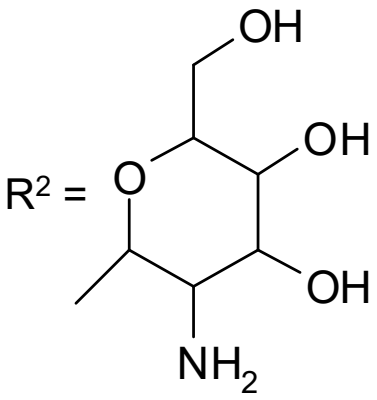
amikacin
Amikin® inj.

Aminoglycosides

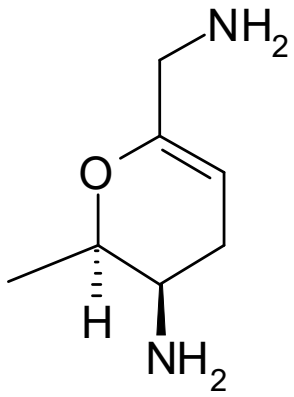
3. Group of kanamycin a gemtamycin
Subgroup of gentamycin



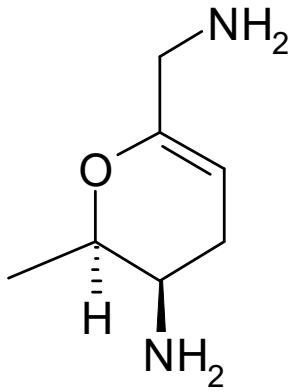
R^1= H- H- CH_3CH_2-



gentamycin
Garasone® gtt. opht.
(+betamethason)
Diagen® a.u.v.



sisomycin



netilmycin
Netromycine® inj.
•serious infections,
sepsis …