Pharmaceutical chemistry

Antibacterial Antibiotics

Synthetic antibacterial agents

quinolones, Aminoacridines, Rifamycins, Nitroimidazoles and nitrofurantoin

AL-Mustaqbal university
College of pharmacy

Lecture 6

Agents that act on nucleic acid transcription and replication

Quinolones and fluoroquinolones

Several organic **compounds obtained by chemical synthesis** on the basis of model compounds have useful antibacterial activity for the treatment of local, systemic, and/or **urinary tract infections.**

Some chemical **classes of synthetic antibacterial** agents include the **sulfonamides,** certain nitroheterocyclic compounds (e.g., **nitrofurans, metronidazole**), and the **quinolones**.

Quinolones

The quinolones comprise a series of synthetic antibacterial agents patterned after **Nalidixic acid**, a naphthyridine derivative introduced for the treatment of urinary tract infections in 1963.

Isosteric heterocyclic groupings in this class include the :

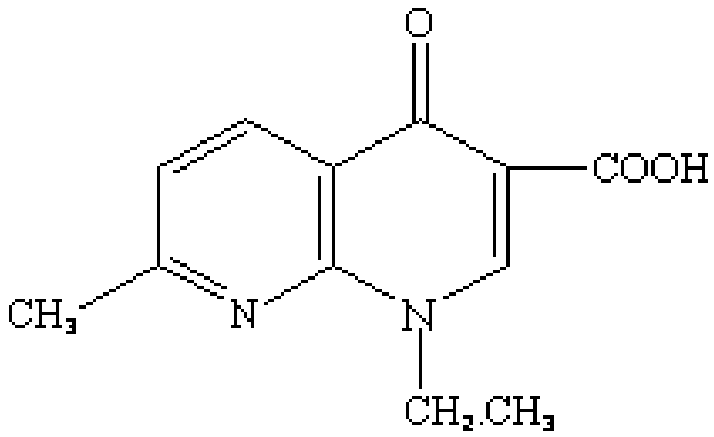
a)quinolones (e.g., **Norfloxacin, Ciprofloxacin, Lomefloxacin),**

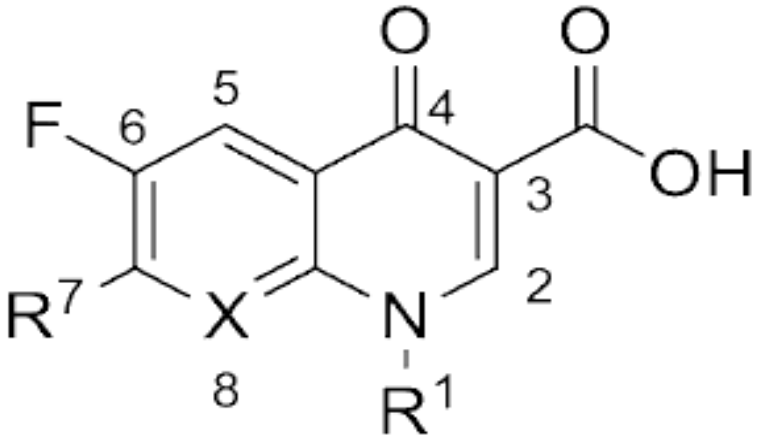
b) **naphthyridines** (e.g., Nalidixic acid, enoxacin), and the

c) cinnolines (e.g., **cinoxacin**).

Up to the present time, the clinical usefulness of the quinolones has been largely confined to the treatment of urinary tract infections.

A - Quinolones





Parent drug: nalidixic acid

Naphthyridine derivative

Quinolones

and fluoroquinolones

quinine derivative

Classification

- Quinolones (**1st generation**)

Highly protein bound

Mostly used in UTIs

- Fluoroquinolones (**2nd, 3rd and 4th generation**)

Modified 1st generation quinolones

Not highly protein bound

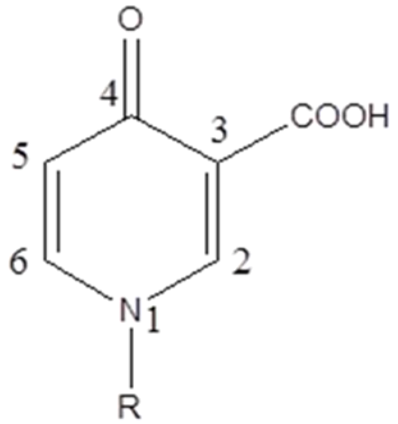
Wide distribution to urine and other tissues; limited CSF penetration. (cerebrospinal fluid)

Generation	Drug Names	Spectrum
1st	nalidixic acid cinoxacin	Gram- but not Pseudomonas species
2nd	norfloxacin ciprofloxacin enoxacin ofloxacin	Gram- (including Pseudomonas species), some Gram+ (S. aureus) and some atypicals
3rd	levofloxacin sparfloxacin moxifloxacin gemifloxacin	Same as 2nd generation with extended Gram+ and atypical coverage
4th	*trovafloxacin	Same as 3rd generation with broad anaerobic coverage

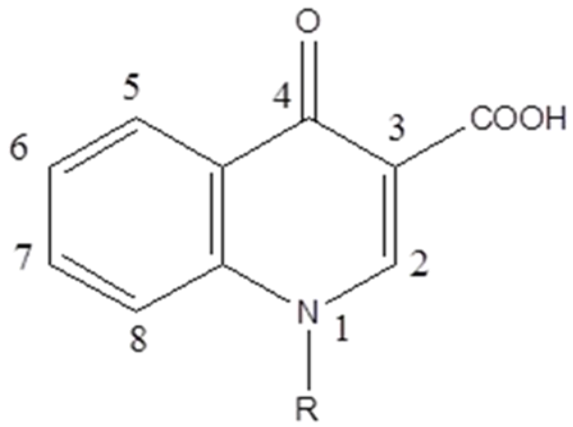
Structure–activity studies

Structure–activity studies have shown that :

1. The 1,4-dihydro-4-oxo-3-pyridinecarboxylic acid moiety is essential for antibacterial activity. The pyridone system must be annulated with an aromatic ring.

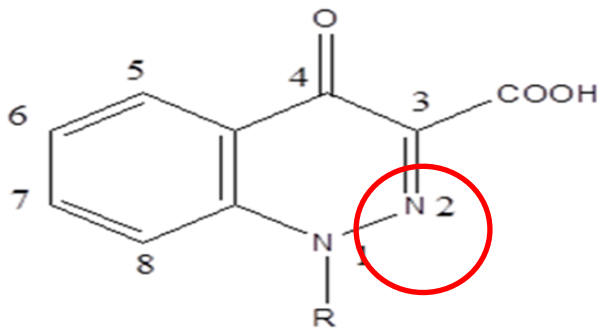


Substituted 1,4-dihydro-4-oxo-3-pyridinecarboxylic acid

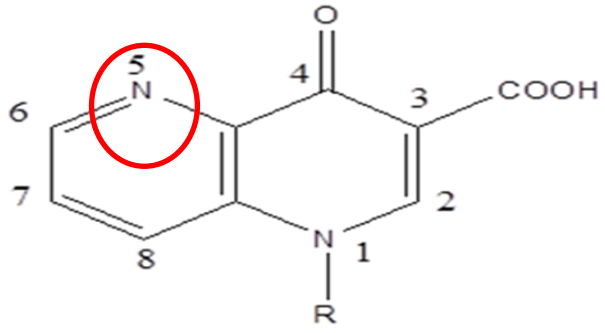


Substituted quinolone 3-carboxylic acid

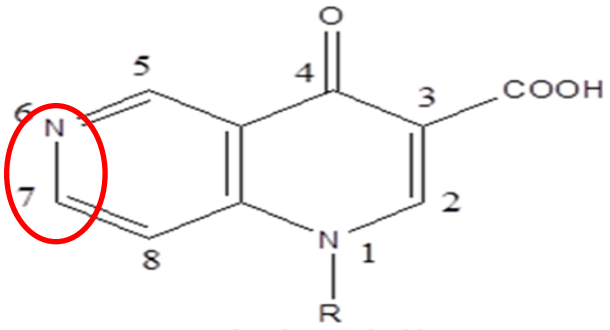
2. Isosteric replacements of nitrogen for carbon atoms at positions 2 (cinnolines), 5 (1,5-napthyridines), 6 (1,6-naphthyridines), and 8 (1,8-naphthyridines) are consistent with retention of antibacterial activity.



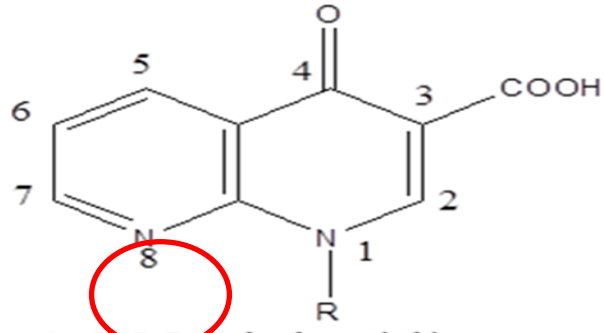
Cinnolines



1,5-Naphthyridines

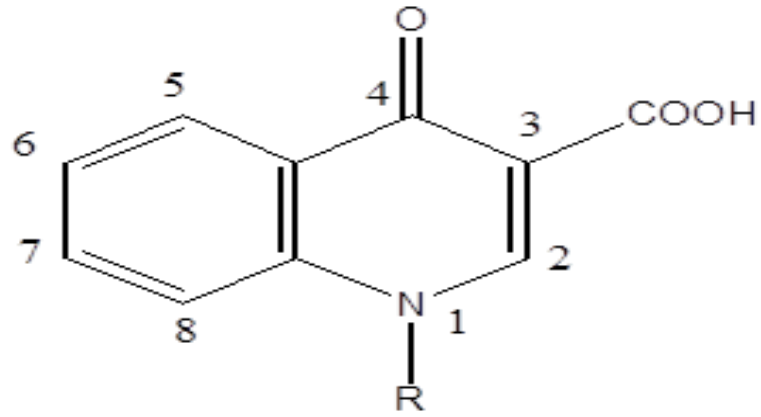


1,6-Naphthyridines



1,8-Naphthyridines

Structure–activity studies

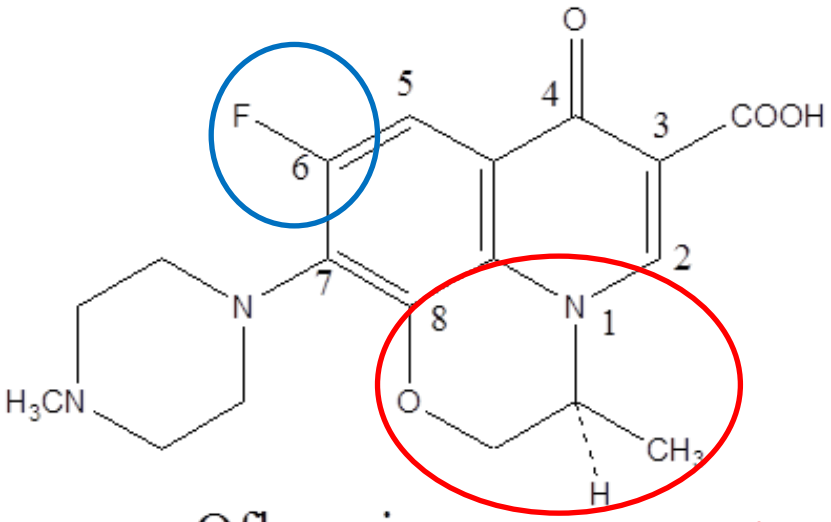


3. substituents at position 2 greatly reduces or abolishes activity,

4. Positions 5, 6, 7(especially), and 8 of the annulated ring may be substituted with good effects. For example, piperazinyl and 3-aminopyrrolidinyl

5. substitutions at position 7 have been shown to convey enhanced activity on members of the quinolone class against P. aeruginosa.

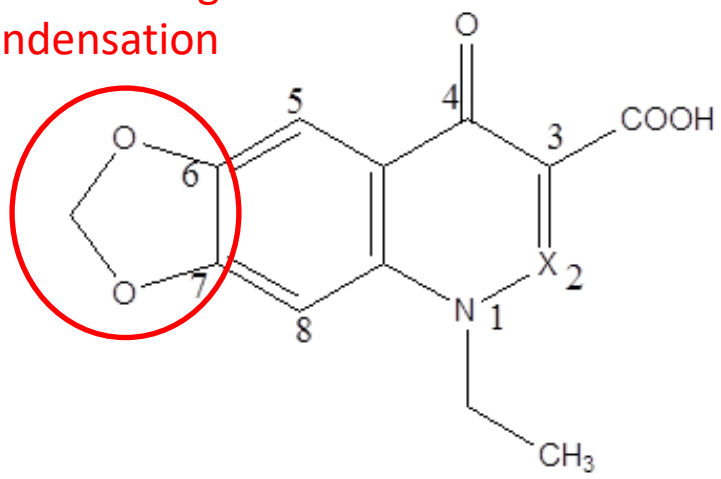
6. Fluorine atom substitution at position 6 is also associated with significantly enhanced antibacterial activity.



Ofloxacin
Levofloxacin (-)S

Ring condensation

Ring condensation



X: N = Cinoxacin
X: CH = Oxolinic acid

Structure–activity studies

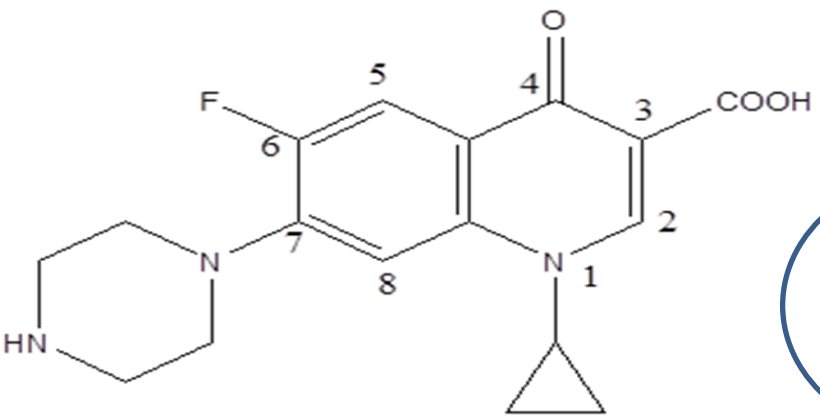
7. Alkyl substitution at the 1-position is essential for activity, with lower alkyl (methyl, ethyl, cyclopropyl) compounds generally having progressively greater potency.

8. Aryl substitution at the 1-position is also consistent with antibacterial activity, with a 2,4-difluorophenyl group providing optimal potency.

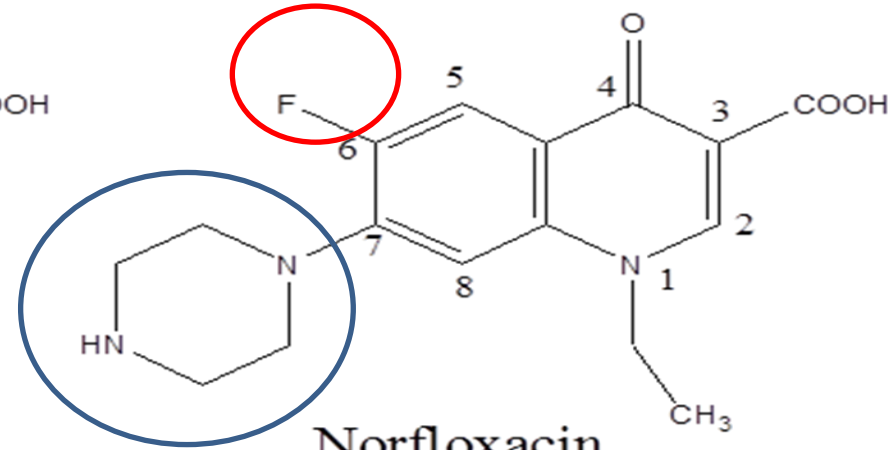
9. Ring condensations at the 1,8-, 5,6-, 6,7-, and 7,8-positions also lead to active compounds.

Structure–activity studies

10. Newer members of the class possessing 6-fluoro and 7-piperazinyl substituents exhibit an extended spectrum of activity that includes effectiveness against additional Gram-negative pathogens

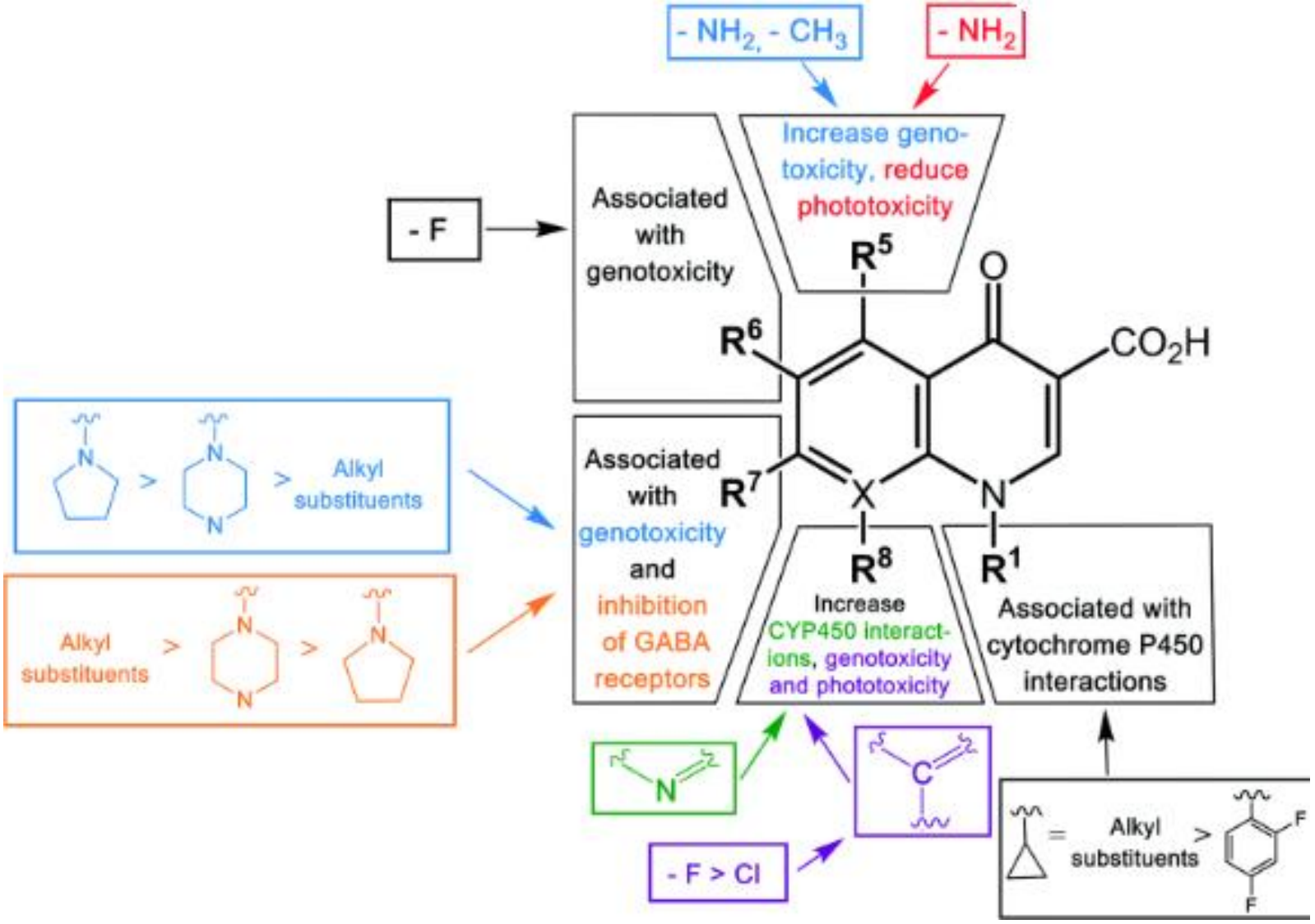


Ciprofloxacin

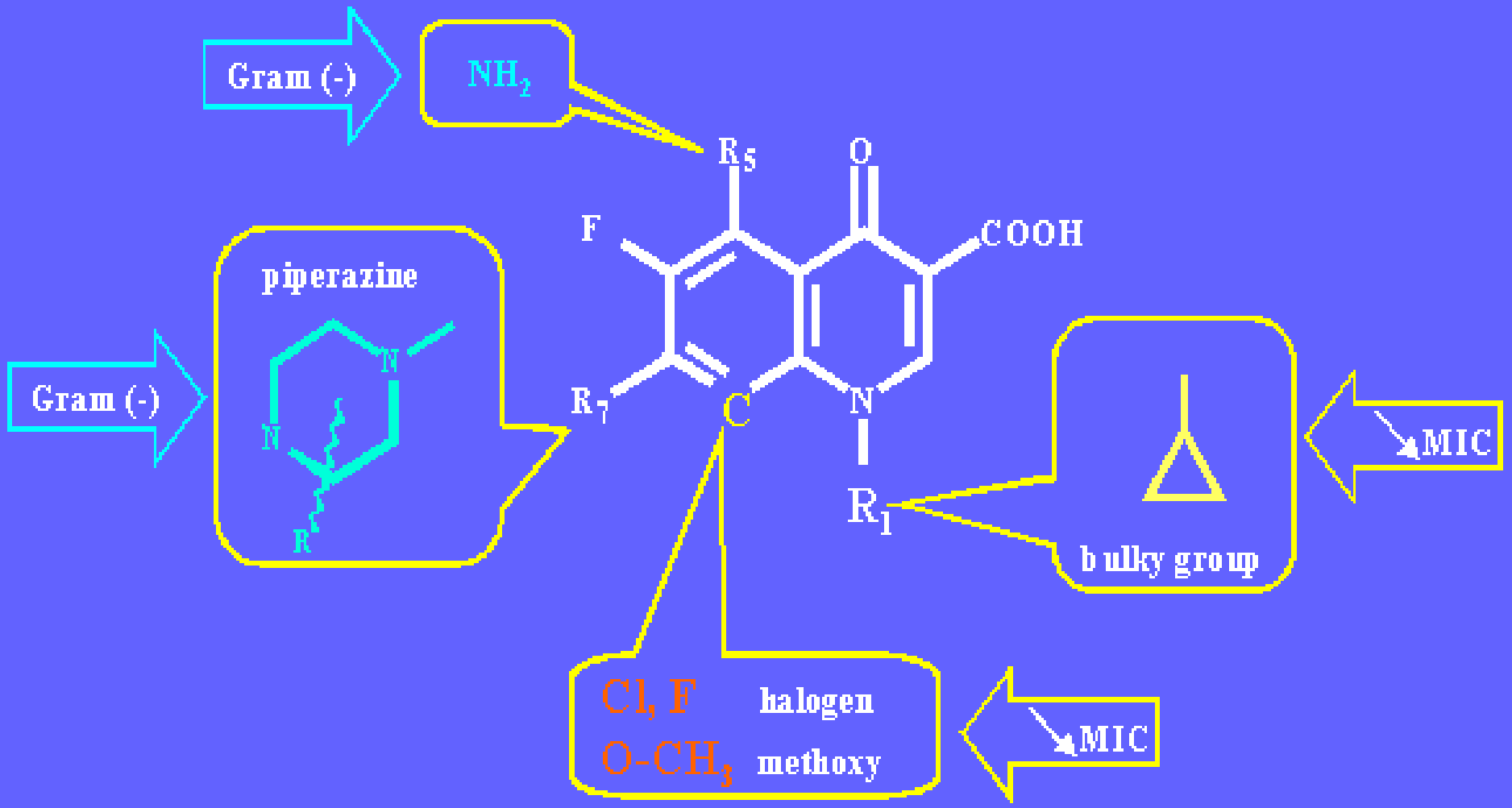


Norfloxacin

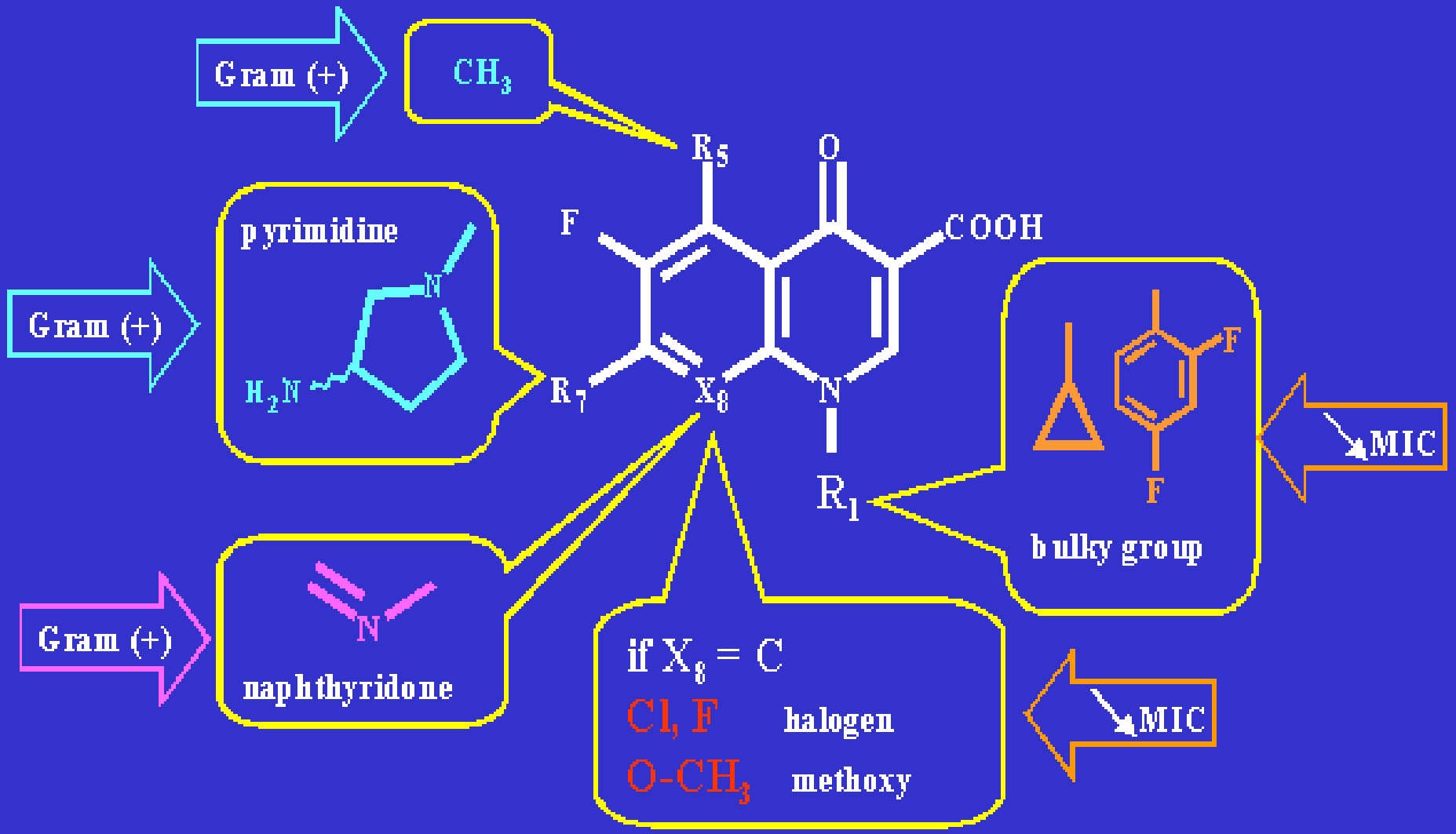
Structure–activity studies



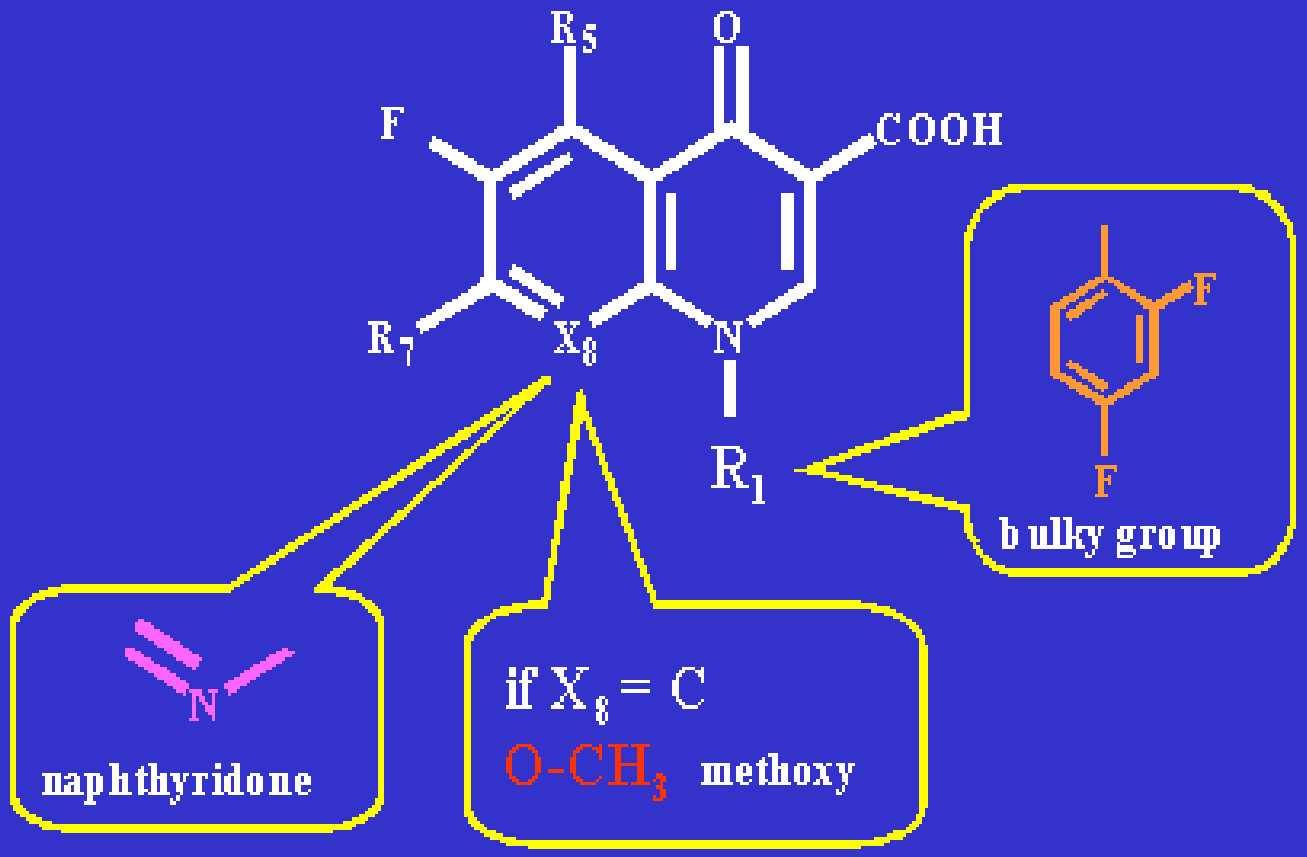
1. maintenance of anti - Gram (-) activity



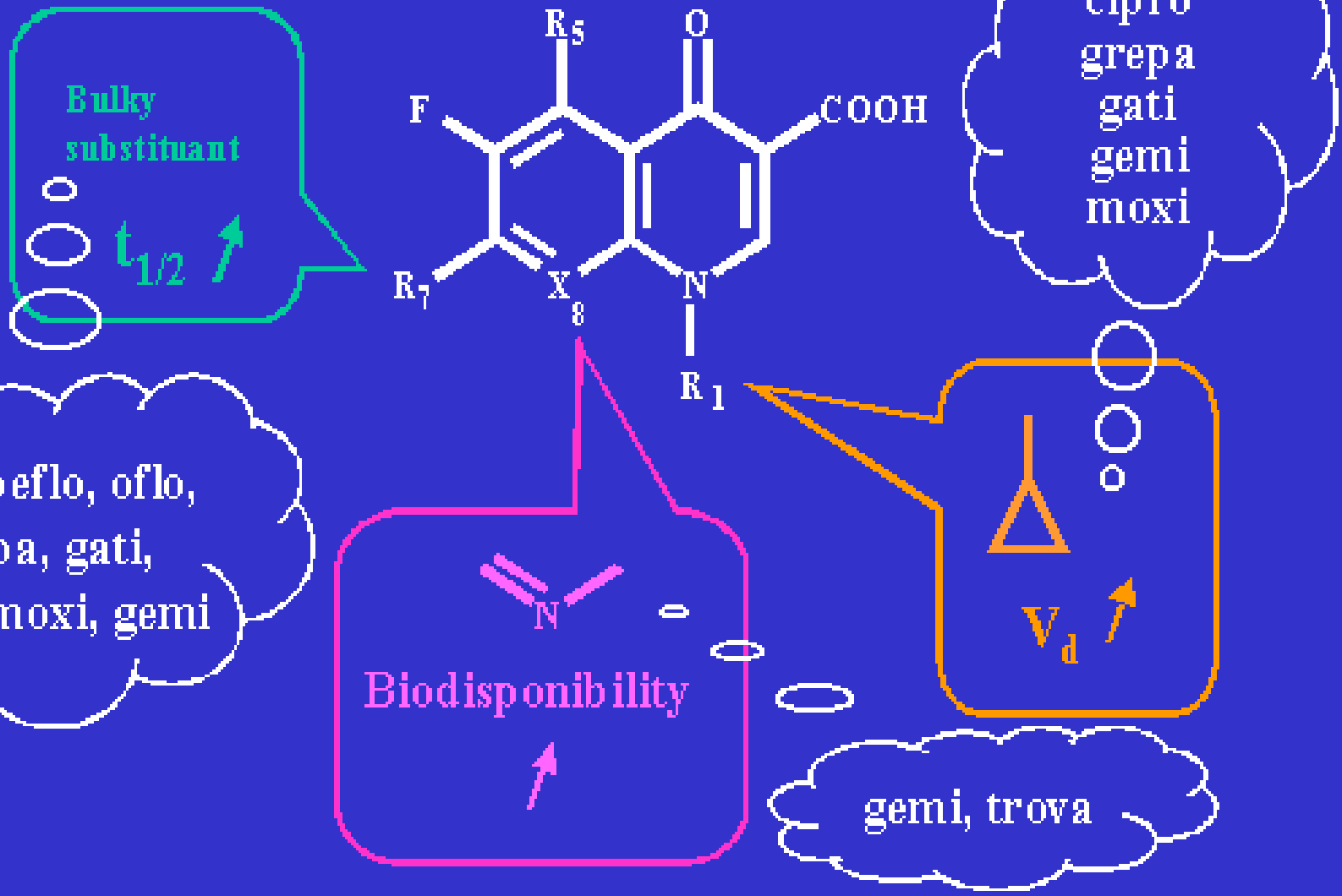
2. improving Gram (+) activity (*S. pneumoniae*)



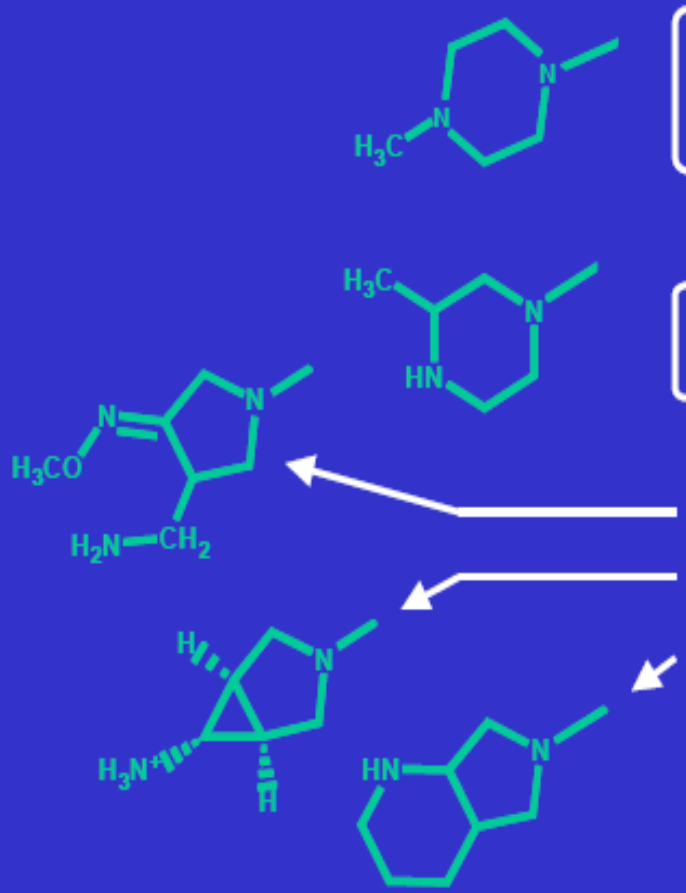
3. obtaining activity against anaerobes ...



SAR of pharmacokinetic parameters



SAR of main pharmacokinetic parameters: how to get a long half life

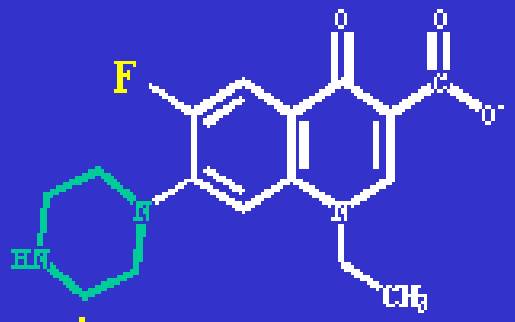


	$t_{1/2}$ (h)	no. of daily administrations
oflo	5 - 7	2 x*
peflo	10	2 x*
flero	9 - 13	1 x
grepa	10 - 12	1 x
gati	13	1 x
gemi	8	1 x
trova	10	1 x
moxi	12	1 x
other FQ	3 - 6	2 x

* higher MIC...

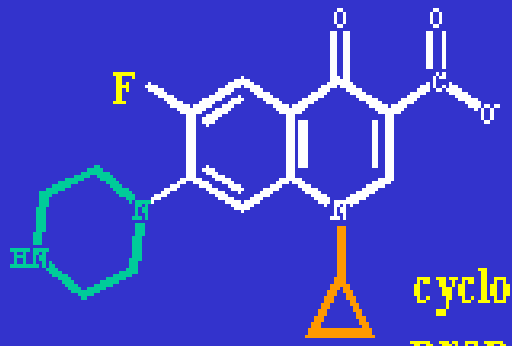
"1st generation" fluoroquinolones

norfloxacin

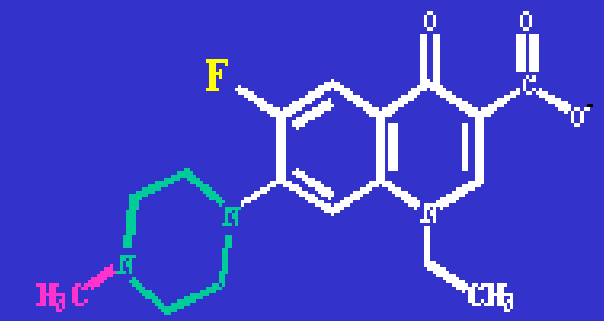


piperazine

ciprofloxacin

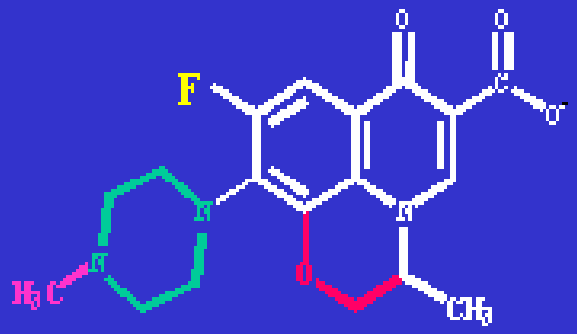


cyclo
propyl



methyl

pefloxacin

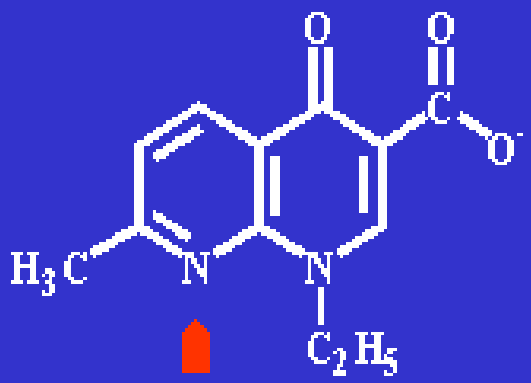


ofloxacin

morpholine

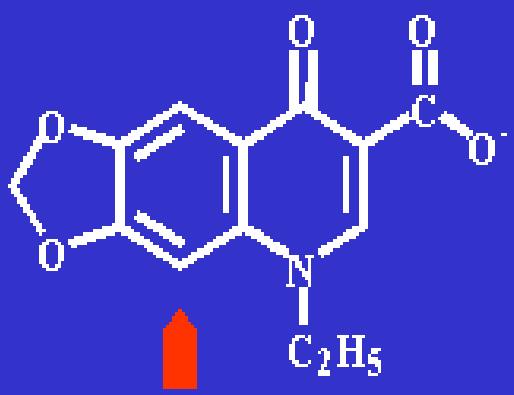
From nalidixic acid to the 1st fluoroquinolone (1 of 4)

nalidixic acid



1. modify naphthyridone into quinolone

oxolinic acid *



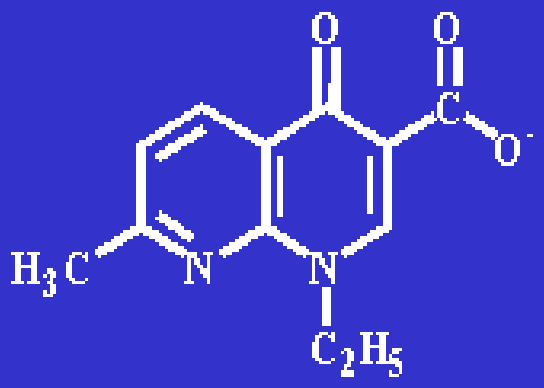
shows reduced protein binding...

* Ger. pat. to Warner Lambert, 1967

* quinoleine

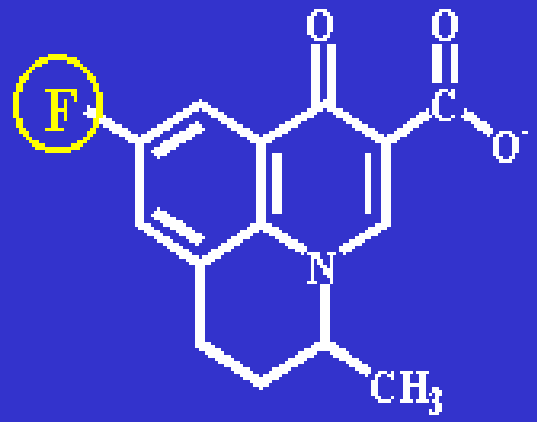
From nalidixic acid to the 1st fluoroquinolone (1 of 4)

nalidixic acid



2. discovery of flumequine *

flumequine *



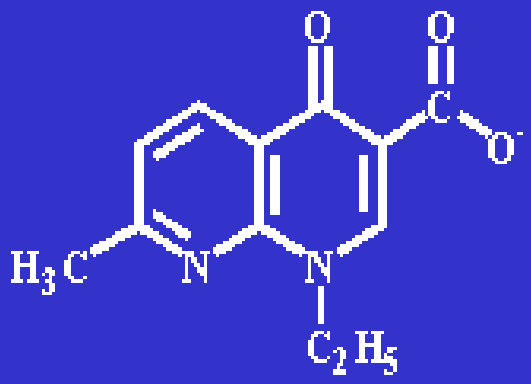
shows weak but broad Gram(-) activity

* Ger pat. to Rikker Lab s, 1973

* benzo-quinolizine

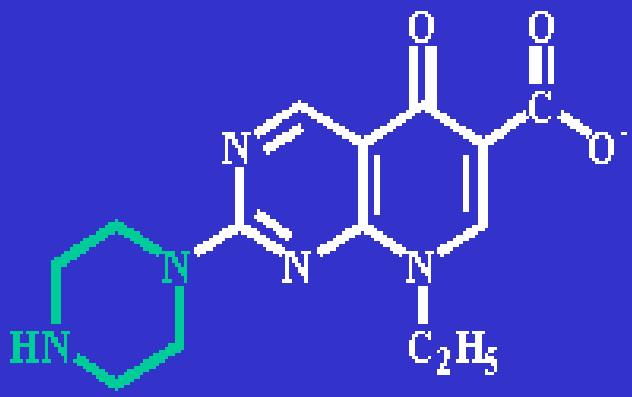
From nalidixic acid to the 1st fluoroquinolone (1 of 4)

nalidixic acid



3. introduce a piperazine *

pipemidic acid *



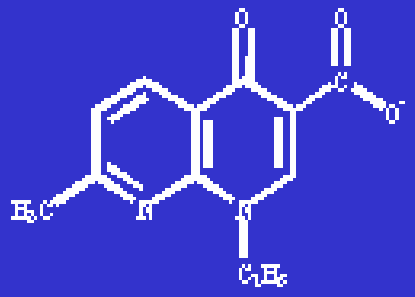
shows longer half-life...

* Ger. Pat. to Roger Bellon, 1974

* pyrido-2-3-pyrimidine

From nalidixic acid to the 1st fluoroquinolone (1 of 4)

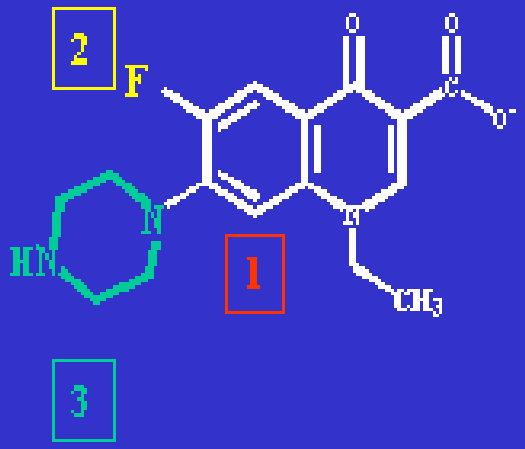
nalidixic acid



combine all 3 features *...

1978

norfloxacin *



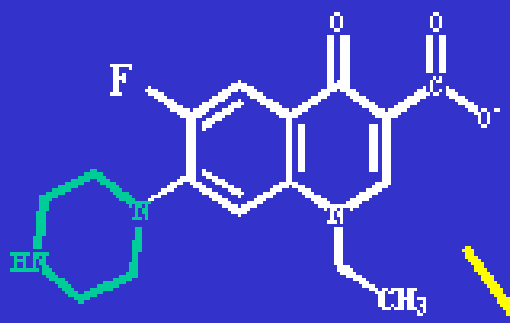
broader Gram(-) activity
less protein binding (50%)
longer half-life (3-4h)

* Belgian patent 863,429, 1978 to Kyorin

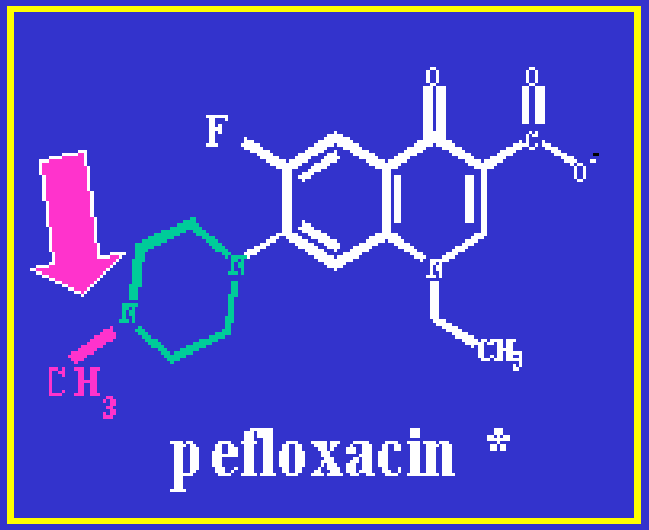
* 6-fluoro-7-pyrimidino-quinoleine

From norfloxacin to the other 1st generation fluoroquinolones: pefloxacin

norfloxacin



Add a methyl to still increase half-life

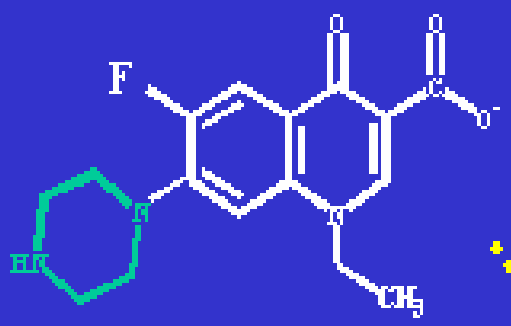


pefloxacin *

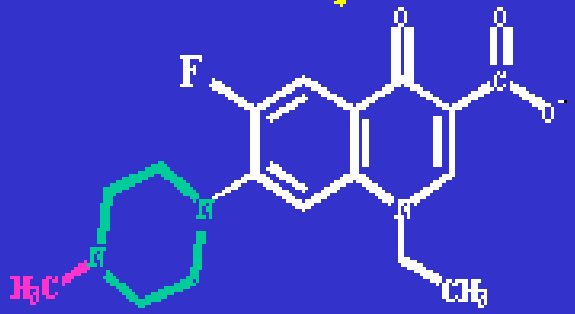
* Ger. pat. 2,840,910 to Roger Bellon/Dainippon,1979

From norfloxacin to the other 1st generation fluoroquinolones: ofloxacin

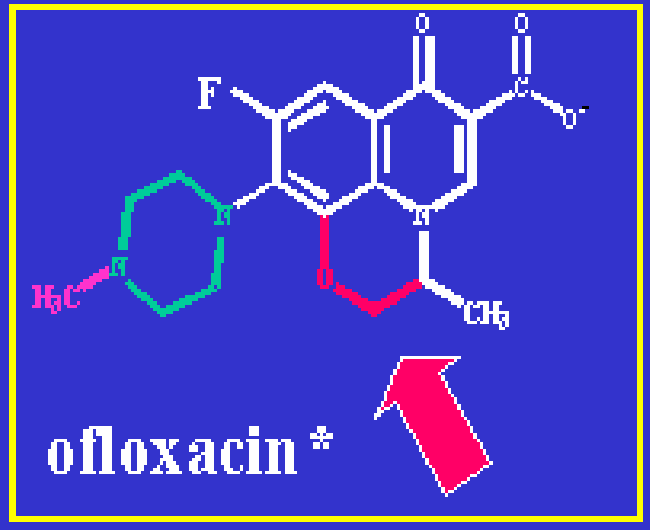
norfloxacin



tricyclic compound
(as in flumequine but
morpholine ring)



pefloxacin

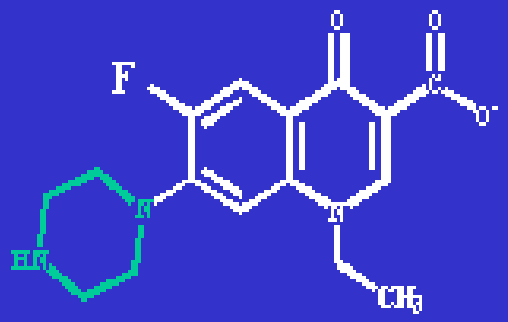


ofloxacin *

* Eur. pat. Appl. 47,005 to Daiichi, 1982

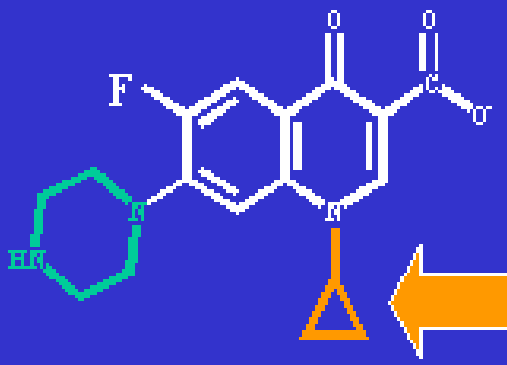
From norfloxacin to the other 1st generation fluoroquinolones: ciprofloxacin

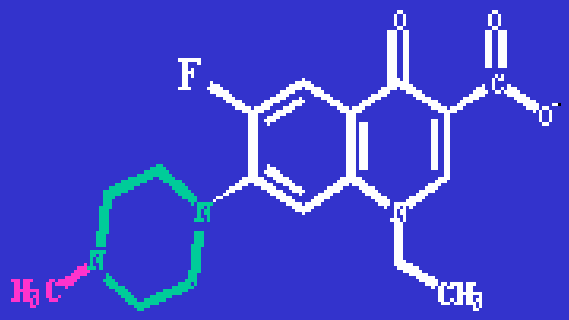
norfloxacin



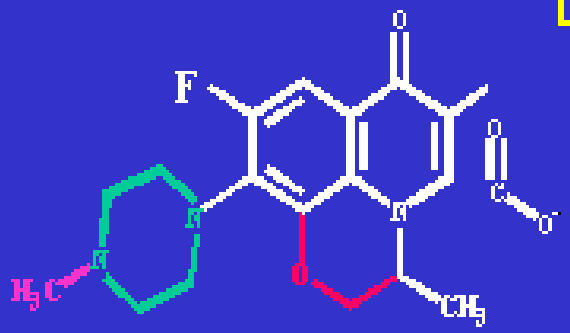
cyclopropyl to increase potency

ciprofloxacin *





pefloxacin

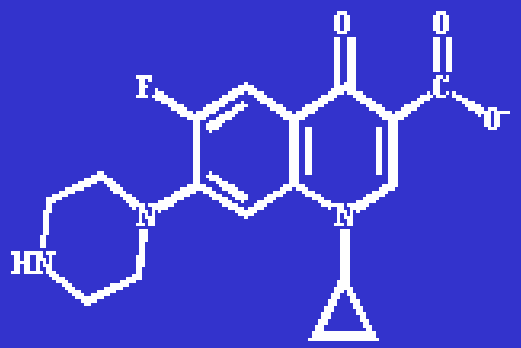


ofloxacin

* Ger. pat. 3,142,854 to Bayer AG, 1983

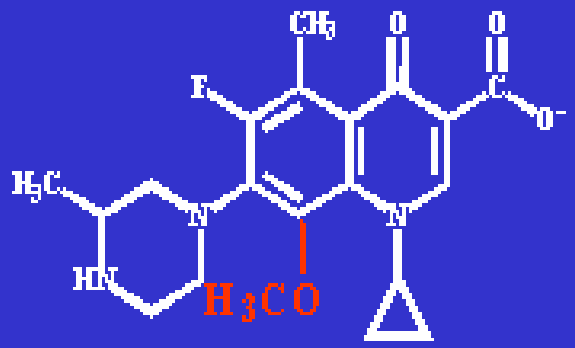
Fluoroquinolones with a C8-methoxy

I



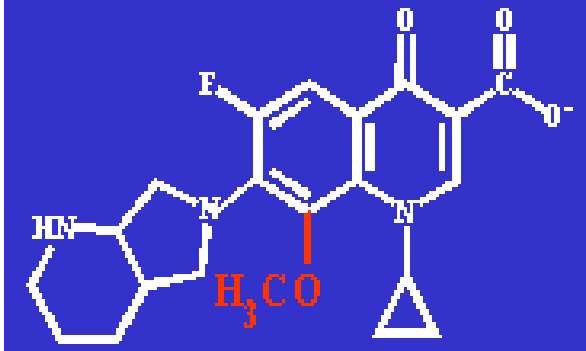
ciprofloxacin

II



gatifloxacin

III



moxifloxacin

Second generation fluoroquinolone

Ciprofloxacin, a second generation fluoroquinolone, interrupts DNA replication by inhibiting both topoisomerase II and IV. Topoisomerase II is an enzyme that reduces the amount of supercoiling of the DNA double-stranded helix during the replication process, while the unlinking of the two daughter strands of DNA is a result of topoisomerase IV. Therefore,

Ciprofloxacin has bactericidal and bacteriostatic properties against both Gram-negative and Gram-positive bacterial pathogens.

Ciprofloxacin is differentiated from the quinolone class of antibiotics by the fluorinated carbon atom located at C6 in the aromatic ring. This substitution helps increase the specificity of the drug for topoisomerase by a factor of ten.

Ciprofloxacin is classified as a quinolone antibiotic; a class of broad-spectrum anti-bacterial drugs.

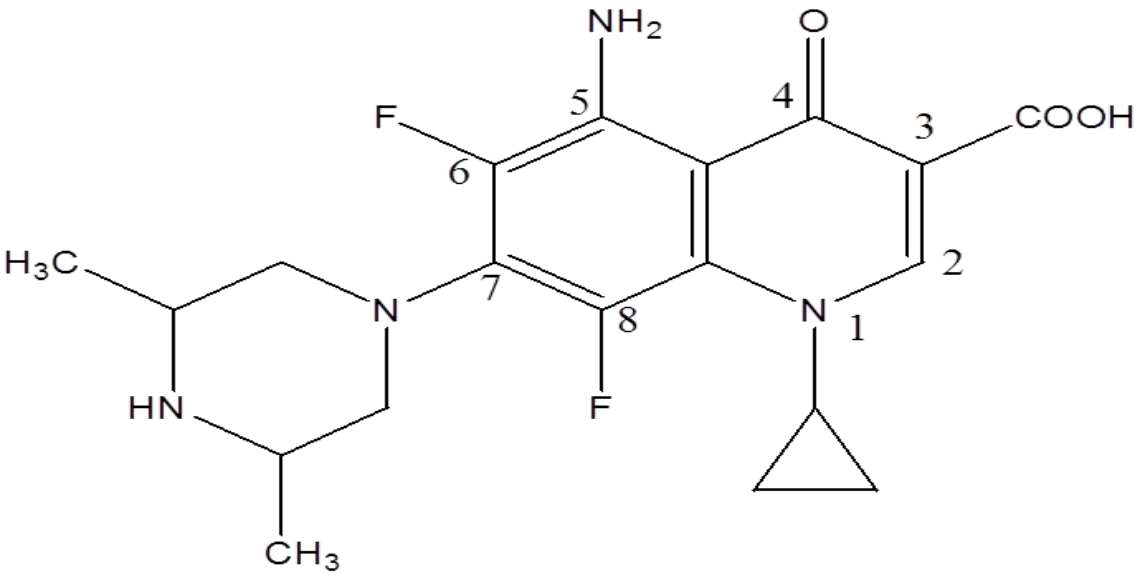
Quinolone interaction with DNA was primarily thought to be through Van der Waals forces and p-p stacking. However, an alternate binding method has recently been uncovered leading to the revision of this initial mechanism of action.

Ciprofloxacin intercalated in DNA - Hydrogen bond hydrophobic and Pi-Pi interaction

Third Generation

Sparfloxacin (Zagam) is a fluoroquinolone antibiotic used in the treatment of bacterial infections. Sparfloxacin inhibits bacterial DNA gyrase, thereby inhibiting DNA replication and transcription. It has a controversial safety profile.

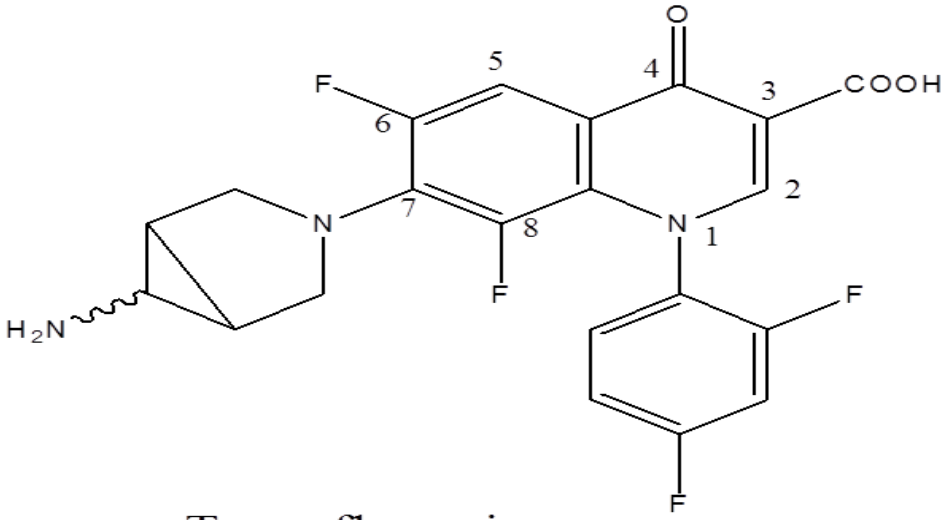
It was patented in 1985 and approved for medical use in 1993. Zagam is no longer available in the United States.



Sparfloxacin

Fourth Generation

Trovafloxacin (sold as Trovan by Pfizer and Turvel by Laboratorios Almirall) is a broad spectrum antibiotic that inhibits the uncoiling of supercoiled DNA in various bacteria by blocking the activity of DNA gyrase and topoisomerase IV. It was withdrawn from the market due to the risk of hepatotoxicity. It had better Gram-positive bacterial coverage and less Gram-negative coverage than the previous fluoroquinolones.



Trovafloxacin

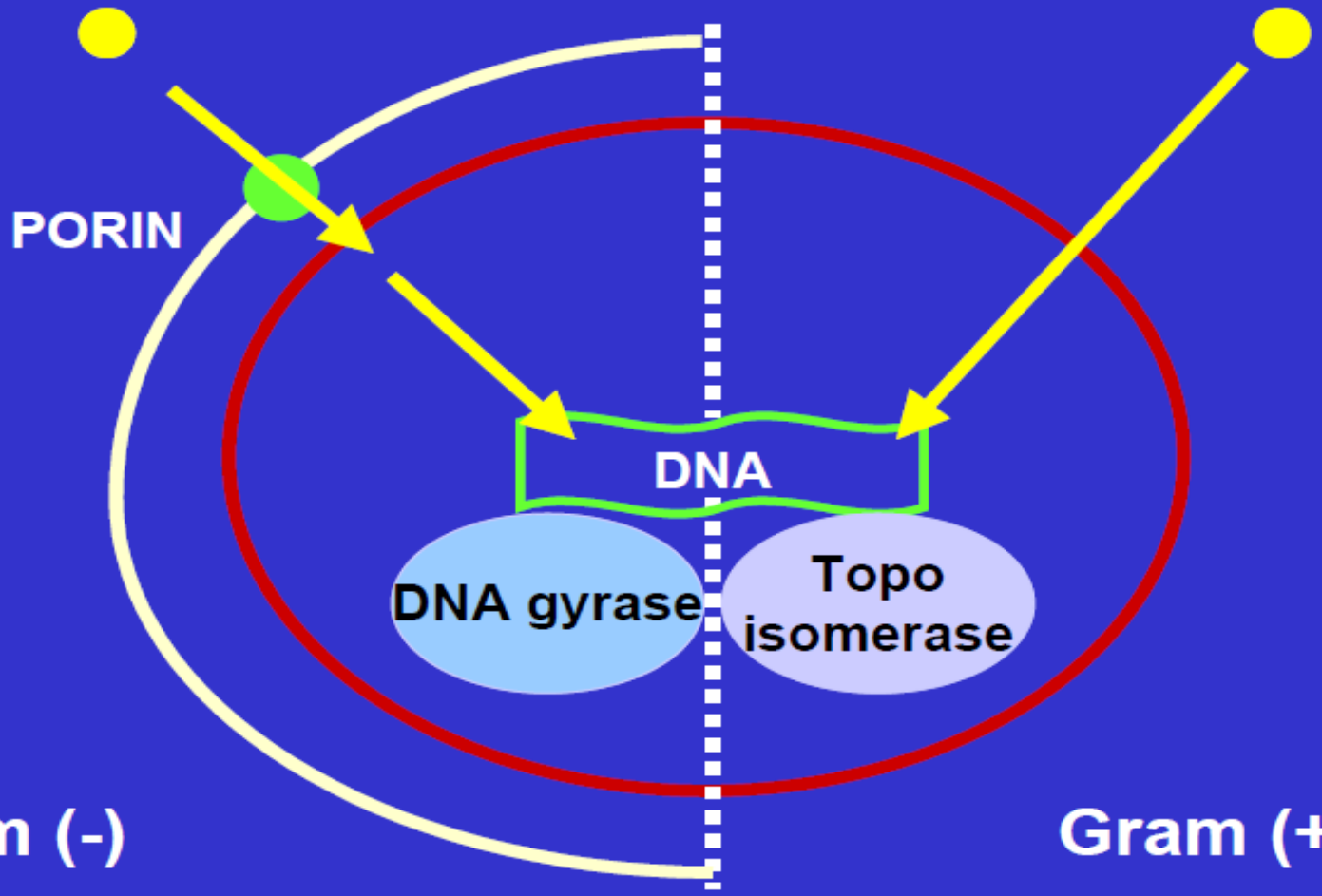
Mechanism of action

The bactericidal action of nalidixic acid and its congeners is known to result from the inhibition of DNA synthesis. This effect is believed to be caused by the inhibition of bacterial DNA gyrase (topoisomerase II), an enzyme responsible for introducing negative supercoils into circular duplex DNA.

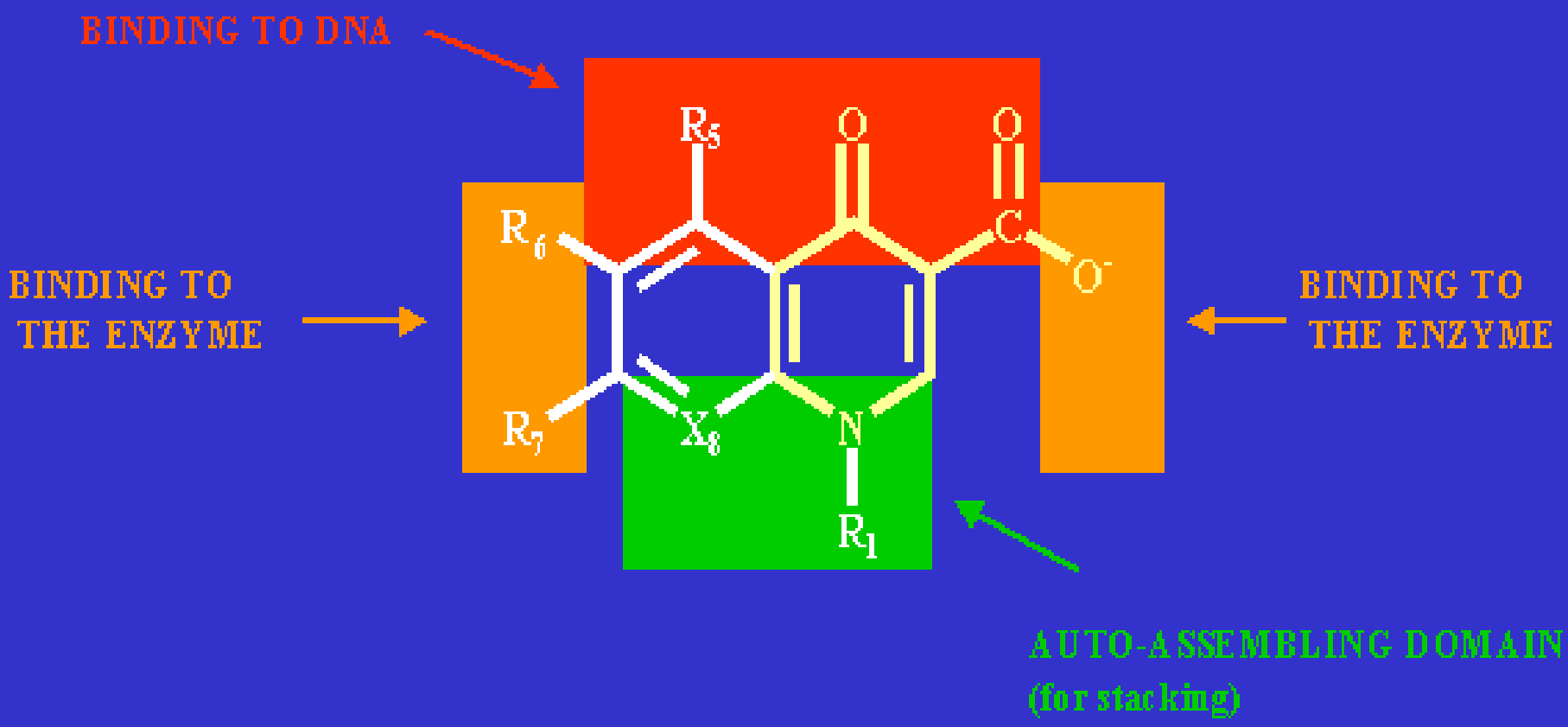
The highly polar quinolones are believed to enter bacterial cells through densely charged porin channels in the outer bacterial membrane. Mutations leading to altered porin proteins can lead to decreased uptake of quinolones and cause resistance.

- In Gram-positive bacteria, the stabilized complexes are between DNA and topoisomerase IV ,with the drugs showing a 1000-fold selectivity for the bacterial enzyme over the corresponding enzyme in human cells.
- In Gram-negative bacteria, the main target for fluoroquinolones is the complex between DNA and a topoisomerase II enzyme called DNA gyrase .
- It has the same role as topoisomerase IV in reverse and is required when the DNA double helix is being supercoiled after replication and transcription.

Mechanism of action of fluoroquinolones: the basics...



The pharmacophore common to all fluoroquinolones



The antibacterial quinolones can be divided into two classes on the basis of their dissociation properties in physiologically relevant conditions.

- The first class, represented by nalidixic acid, oxolinic acid and cinoxacin, possesses only the 3-carboxylic acid group as an ionizable functionality. The pKa values for the 3-carboxyl group in nalidixic acid and other quinolone antibacterial drugs fall in the range of 5.6 to 6.4

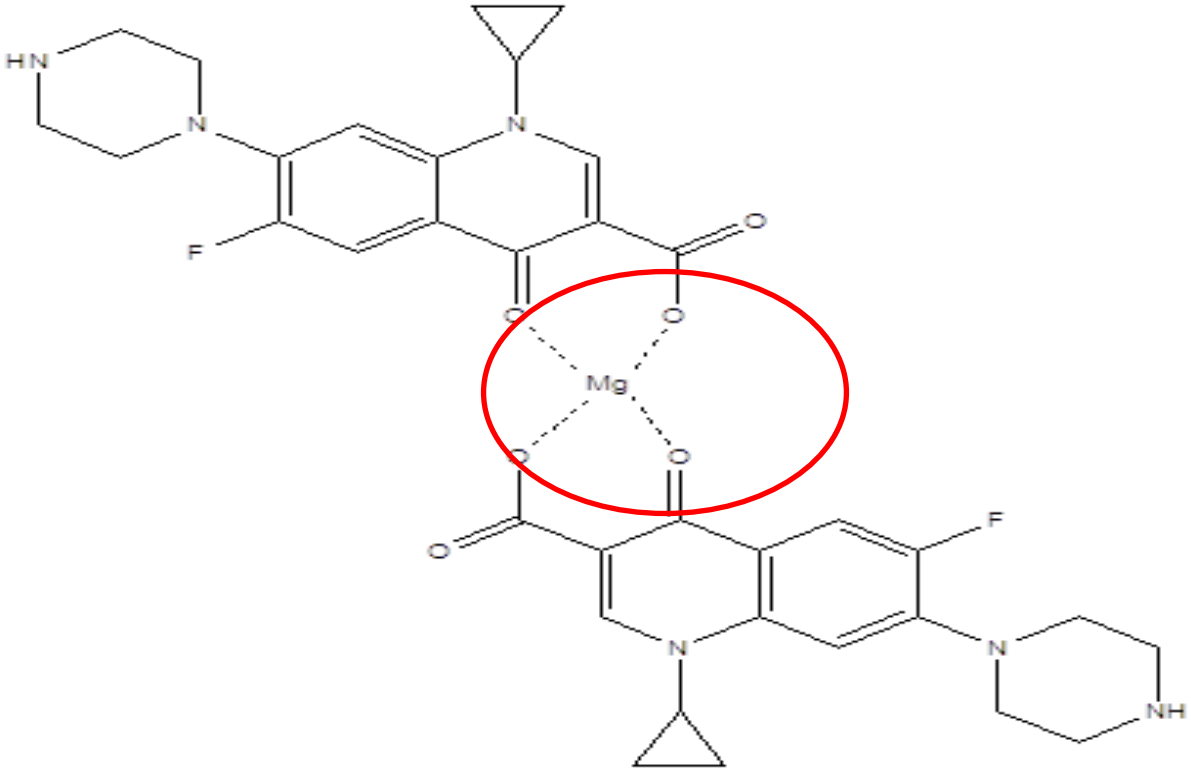
These comparatively high pKa values relative to the pKa of 4.2 for benzoic acid are attributed to the acid weakening effect of hydrogen bonding of the 3-carboxyl group to the adjacent 4-carbonyl group.

- The second class of antibacterial quinolones embraces the broad-spectrum fluoroquinolones (namely, norfloxacin, enoxacin, ciprofloxacin, ofloxacin, lomefloxacin, and sparfloxacin), all of which possess, in addition to the 3-carboxylic acid group, a basic piperazino functionality at the 7-position and a 6-fluoro substituent. The pKa values for the more basic nitrogen atom of the piperazino group fall in the range of 8.1 to 9.3

The excellent chelating properties of the quinolones provide the basis for their incompatibility with antacids, hematinics, and mineral supplements containing divalent or trivalent metals.

The quinolones may form 1:1, 2:1, or 3:1 chelates with metal ions such as Ca2, Mg2, Zn2, Fe2, Fe3, and Bi3. The stoichiometry of the chelate formed depends on various factors, such as the relative concentrations of chelating agent (quinolone) and metal ion present, the valence (or charge) on the metal ion, and the pH.

How fluoroquinolones cause metal complexation



This occurs with cations such as Ca2+, Zn2+, Fe2+, Fe3+, Bi3+. That's why there is an interaction between quinolones and mineral containing drugs.

Aminoacridines

Aminoacridine agents, such as the yellow-coloured **proflavine** , are topical antibacterial agents which were used particularly during World War II to treat deep surface wounds.

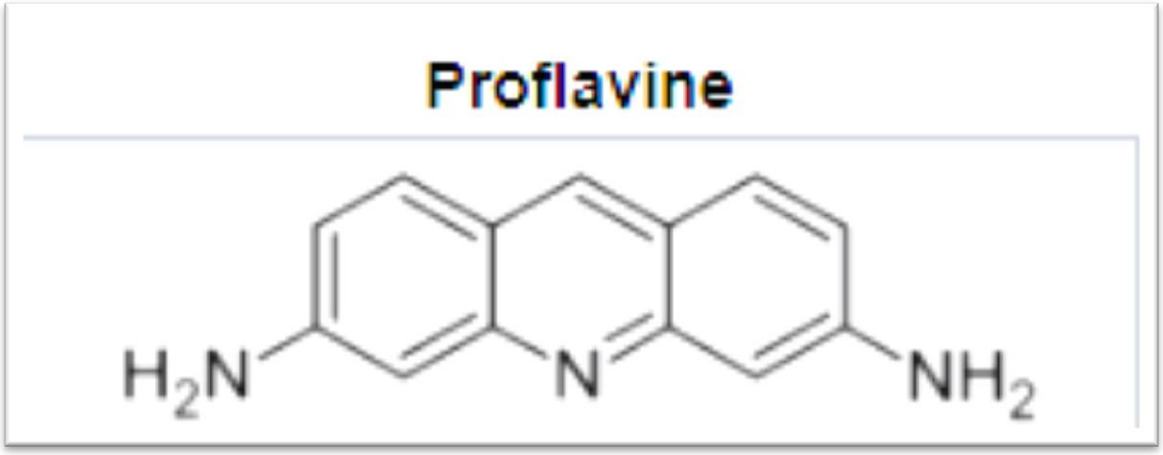
The best agents are completely ionized at pH 7 and they interact directly with bacterial DNA by intercalation.

Proflavine, also called proflavin and diaminoacridine, is an acriflavine derivative, a disinfectant bacteriostatic against many gram-positive bacteria

It has been used in the form of the dihydrochloride and hemisulfate salts as a topical antiseptic, and was formerly used as a urinary antiseptic.

Proflavine is also known to have a mutagenic effect on DNA by intercalating between nucleic acid base pairs.

It differs from most other mutagenic components by causing basepair-deletions or basepair-insertions and not substitutions. In the presence of light, proflavine can induce double-stranded breaks in DNA.



Rifamycins

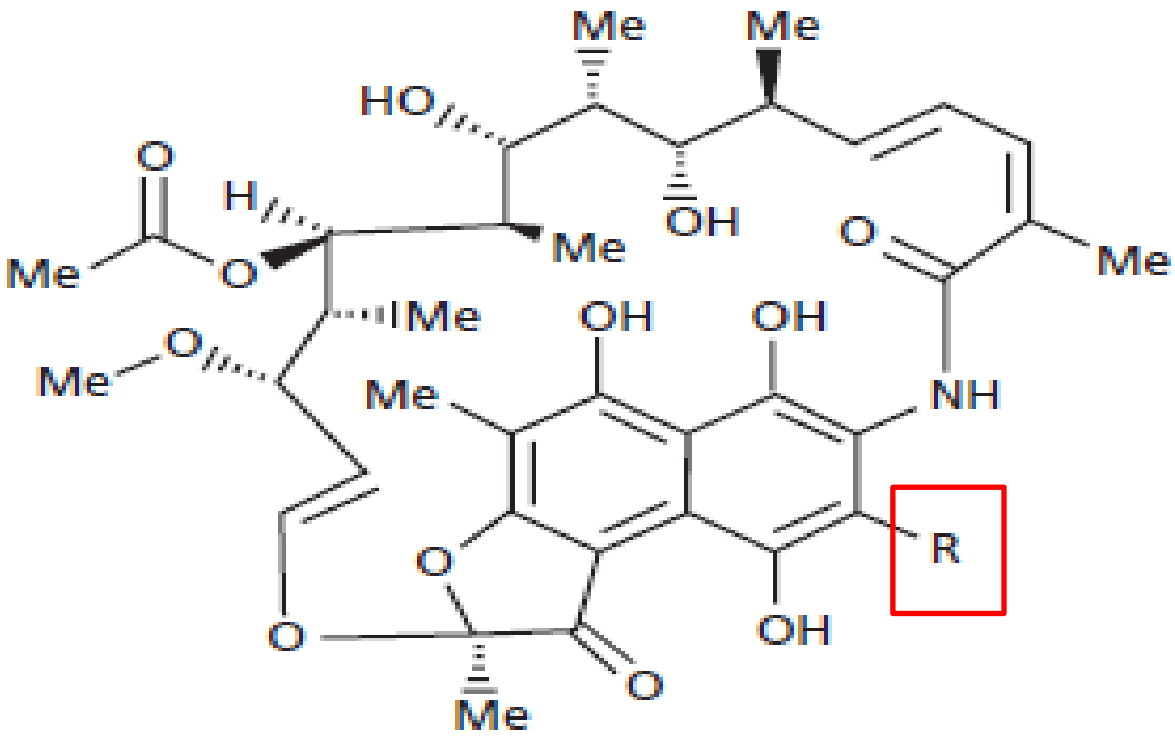
Rifampicin is a semi-synthetic rifamycin made from rifamycin B —an antibiotic which was isolated from Streptomyces mediterranei in 1957. It inhibits Gram-positive bacteria and works by binding non-covalently to DNA-dependent RNA polymerase and inhibiting the start of RNA synthesis.

The DNA-dependent RNA polymerases in eukaryotic cells are unaffected because the drug binds to a peptide chain not present in the mammalian RNA polymerase. It is, therefore, highly selective.

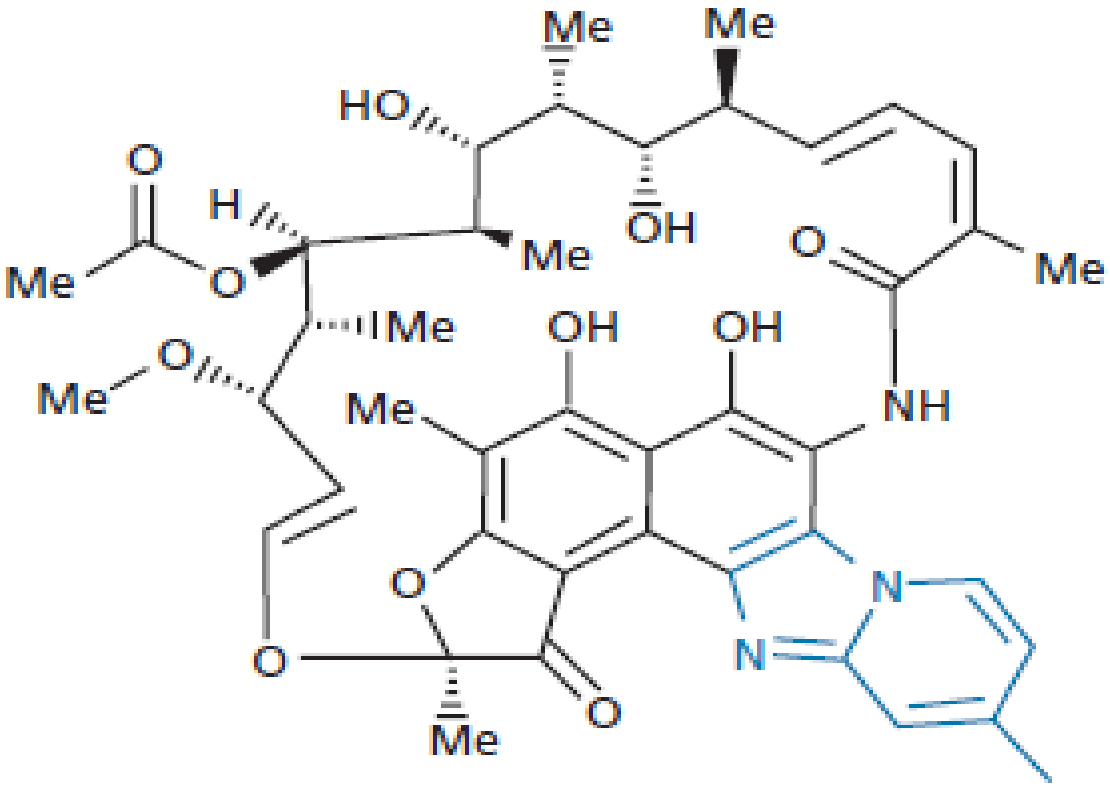
The flat naphthalene ring and several of the hydroxyl groups are essential for activity and the molecule exists as a zwitterion, giving it good solubility both in lipids and aqueous acid.

Rifaximin is another semisynthetic analogue that was approved in 2004 for the treatment of diarrhoea and E. coli infection.

Rifamycins



R =	H	Rifamycin B
R =	CH=N–N NH	Rifampicin



Rifaximin

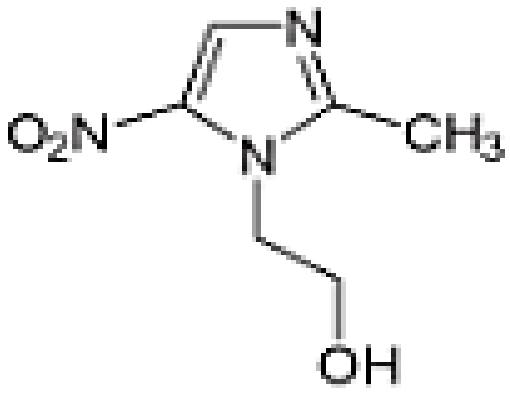
Nitroimidazoles and nitrofurantoin

Metronidazole is a nitroimidazole structure which was introduced in 1959 as an anti-protozoal agent, but began to be used as an antibacterial agent in the 1970s.

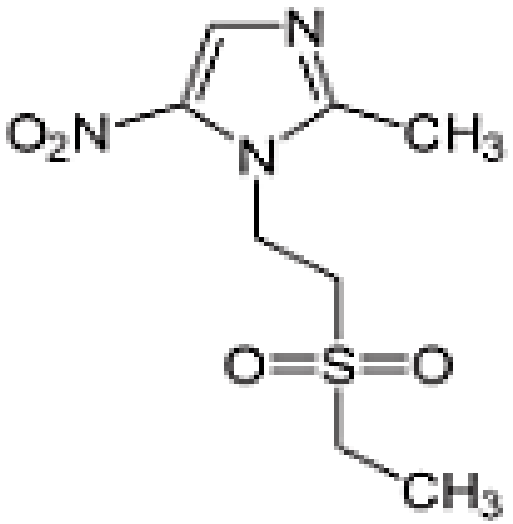
The nitro group is reduced when the drug enters the bacterial cell, which lowers the concentration of metronidazole within the cell and sets up a concentration gradient down which more drug can flow.

The reduction mechanism also proves toxic to the cell as free radicals are formed which act on DNA.

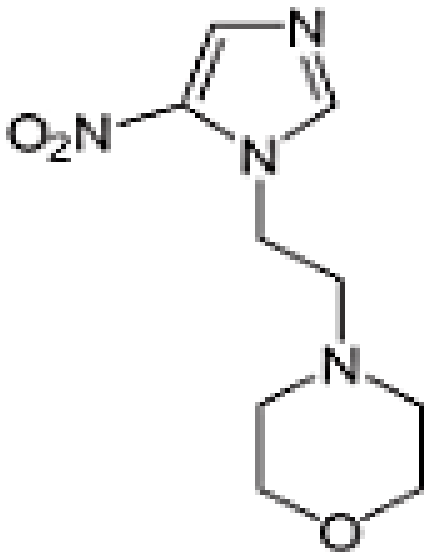
Nitrofurantoin also undergoes reduction within bacterial cells to form radical species that act on DNA.



Metronidazole



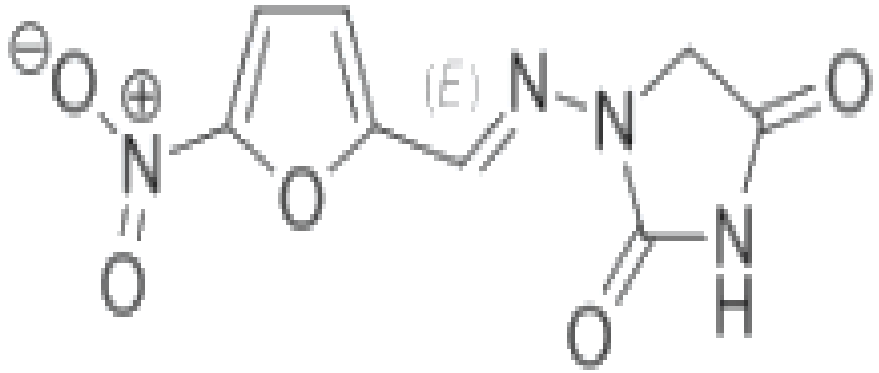
Tinidazole



Nimorazole

Three nitroimidazole antibiotics: metronidazole, tinidazole, and nimorazole

Nitrofurantoin

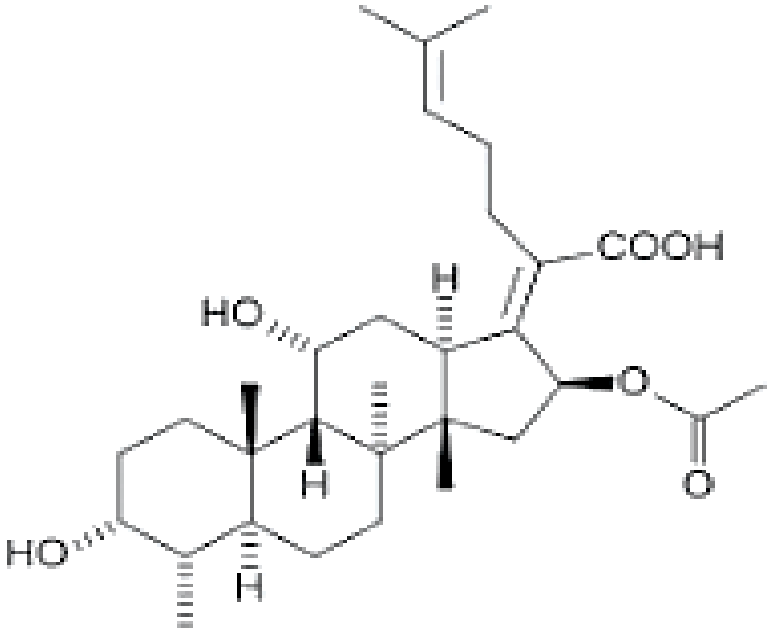


Miscellaneous agents

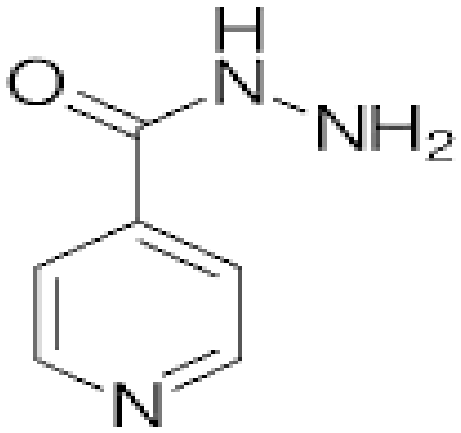
Fusidic acid acts as a bacterial protein synthesis inhibitor

It is a topical antibacterial agent that is used in eye drops and skin creams. It can penetrate intact and damaged skin, so it is useful for the treatment of boils.

It has also been used to eradicate MRSA colonies carried in the nasal passages of hospital patients and health workers.



Isoniazid **i**s the most widely used drug for the treatment of tuberculosis and is part of a four-drug cocktail which is the first choice treatment for the initial phase of the disease.



Isoniazid