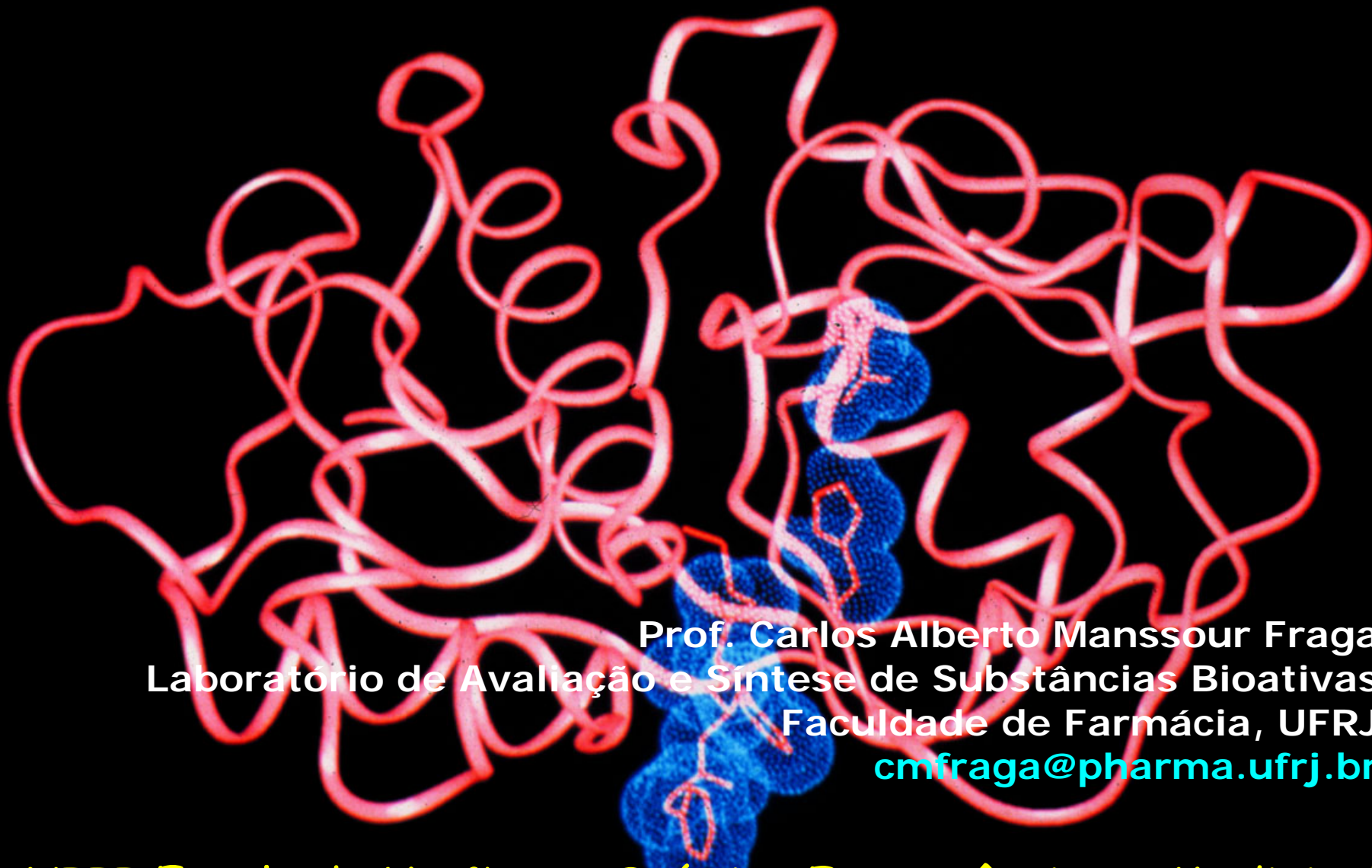


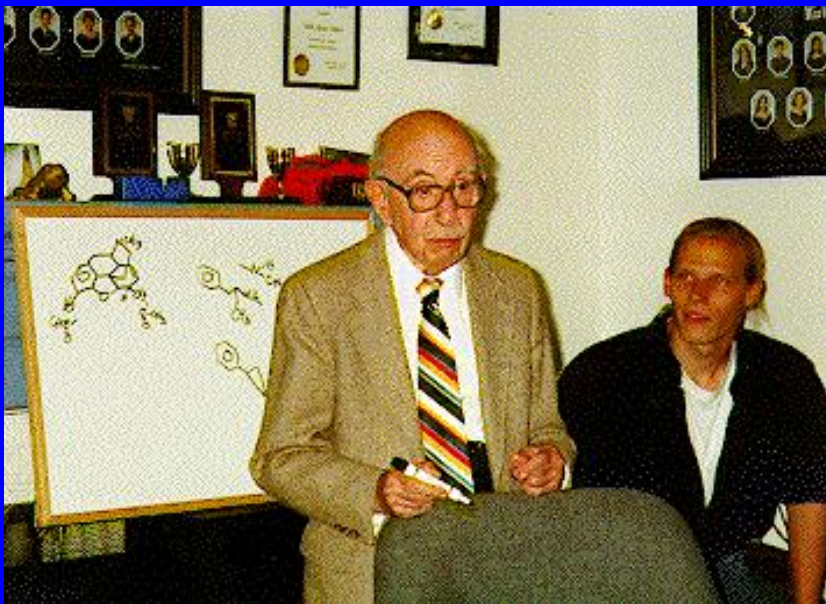
# *História da Descoberta de Fármacos*



Prof. Carlos Alberto Manssour Fraga  
Laboratório de Avaliação e Síntese de Substâncias Bioativas  
Faculdade de Farmácia, UFRJ  
[cmfraga@pharma.ufrj.br](mailto:cmfraga@pharma.ufrj.br)

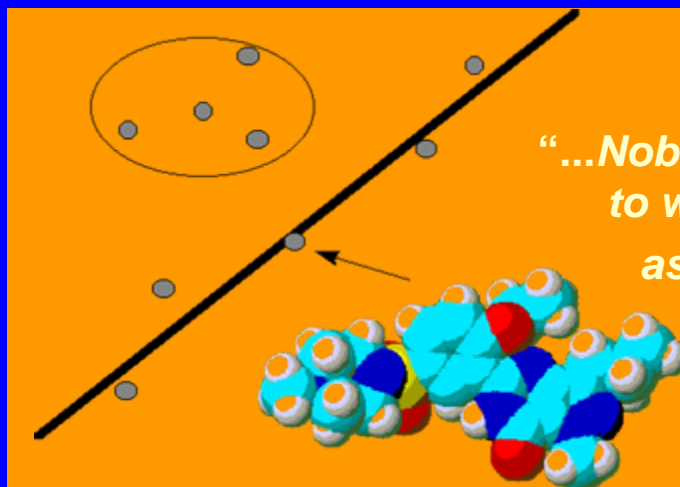
XIII Escola de Verão em Química Farmacêutica e Medicinal  
Rio de Janeiro, 05-09 de Fevereiro de 2007

# O Paradigma da Química Medicinal



“Medicinal Chemistry remains a challenging science, which provides profound satisfaction to its practitioners. It intrigues those of us who like to solve problems posed by nature. It verges increasingly on biochemistry and all the physical, genetic and chemical riddles in animal physiology which bear on medicine. Medicinal Chemists have a chance to participate in the fundamentals of prevention, therapy and understanding of diseases and thereby to contribute to a healthier and happier life.”

(Alfred Burger, 1990)



*“...Nobody in the world is condemned to work with as many variables as the medicinal chemist...”*

(Corwin Hansch, 1993)

Burger's Medicinal Chemistry and Drug Discovery, Vol.1, Wolff, M. E., Ed., 5th Ed., 1994.

***“INTERDISCIPLINARIDADE”***

# Definição da IUPAC



“**Medicinal chemistry** is a chemistry-based discipline, also involving aspects of biological, medical and pharmaceutical sciences. It is concerned with the invention, discovery, design, identification and preparation of biologically active compounds, the study of their metabolism, the interpretation of their mode of action at the molecular level and the construction of structure-activity relationships.”

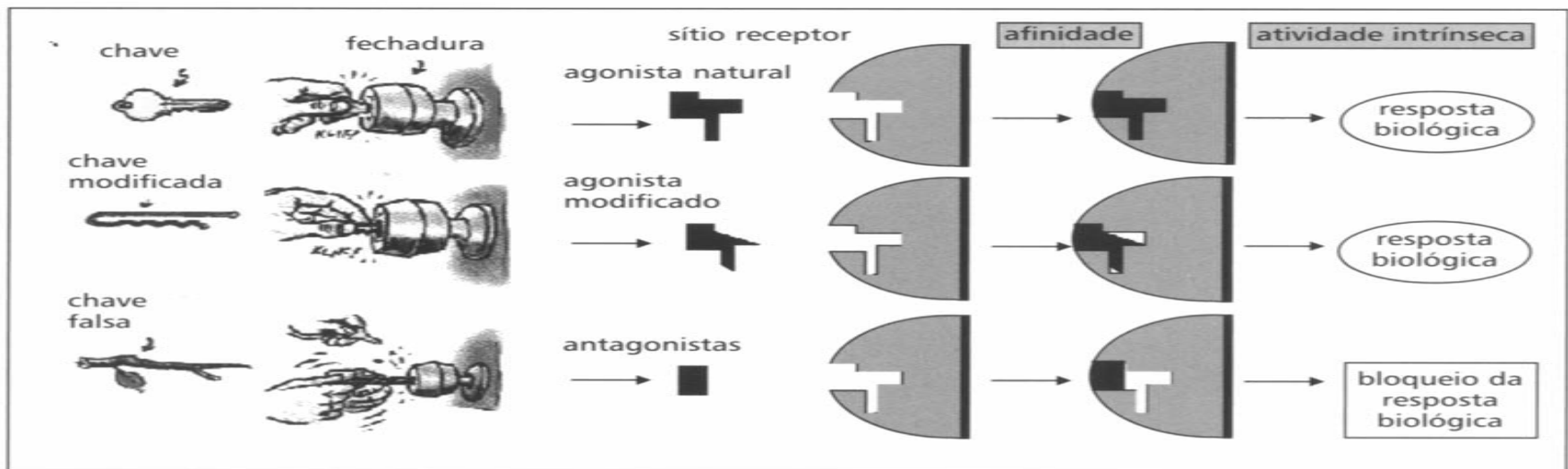
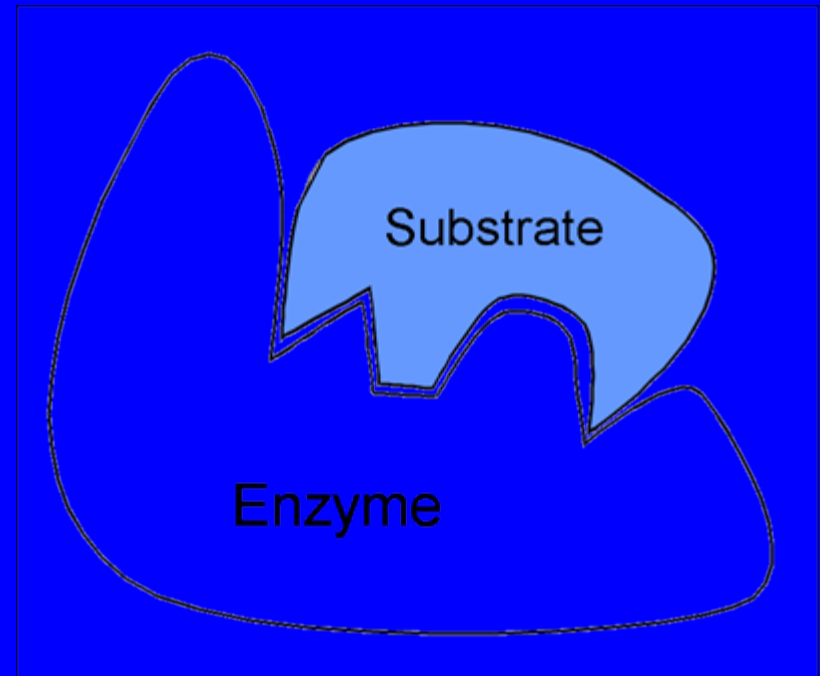
**C.G. Wermuth, C.R. Ganellin, P. Lindberg and L.A. Mitscher  
(1998) Glossary of Terms Used in Medicinal Chemistry**

# Modelo-Chave Fechadura (102 anos)



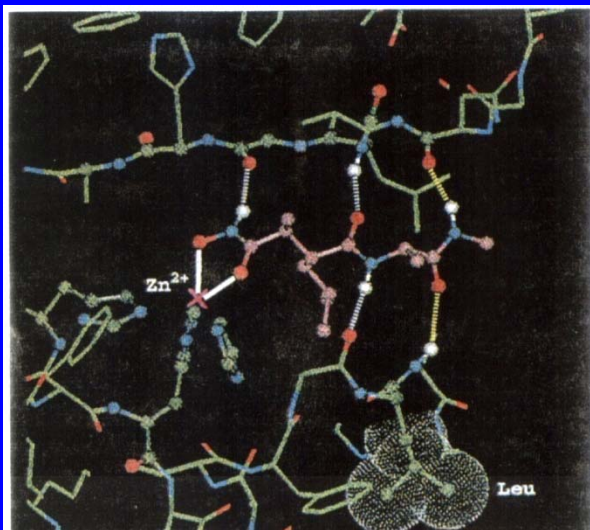
**Emil Fischer**  
(1852-1919)  
(Prêmio Nobel, 1902)

Fischer, E. H. "Dictionary of Scientific Biography", Charles Scribner's Sons: 1970-1990; Vol. 5, pp. 1-5.

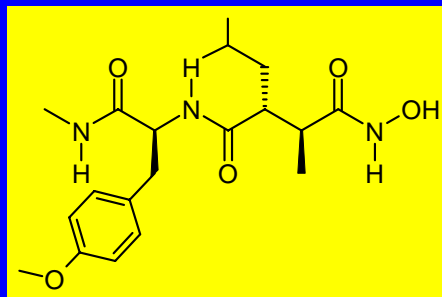




# Teoria do Encaixe Induzido



Koshland Jr., D. E.; Neméthy, G.; Filmer, D. (1966) *Biochemistry* 5, 365.

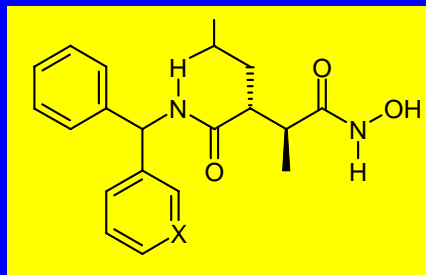
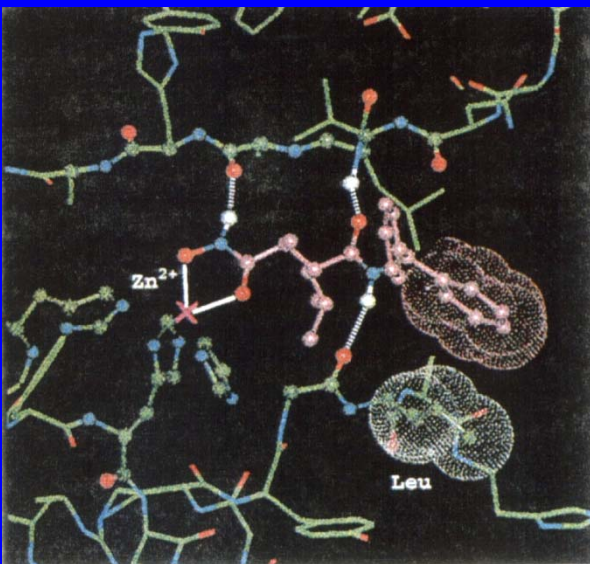


Seleção da  
conformação do ligante  
(reconhecimento)

Ligante

Receptor

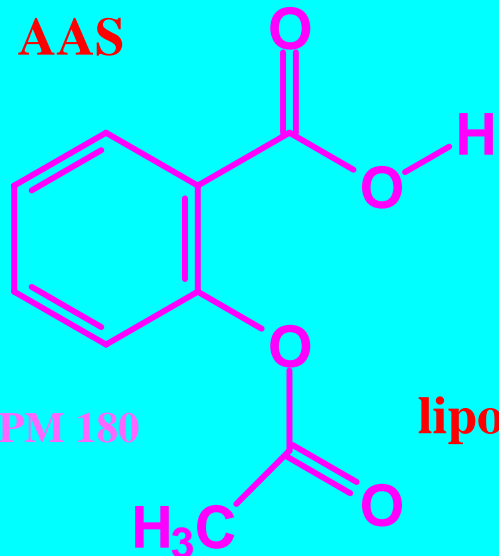
Modificação do  
ambiente molecular  
(sítio receptor)



Rockwell, A.; Melden, M.; Copeland, R. A.; Hardman, K.; Decicco, C. P.; DeGrado, W. F.;  
*J. Am. Chem. Soc.* 1996, 118, 10337

*xenobiótico*

**AAS**



PM 180

**lipossolúvel**

**Coeficiente de partição**

0,100g  
0,55 mM

**p.o.**

**absorção**

**Posologia:** concentração  
tempo de meia-vida  
metabólito ativo (?)  
metabólito tóxico (?)  
outras atividades (?)

**Fatores Farmacocinéticos**

**BIOFASE**

**ADME**

**Rins**

**URINA**

**eliminação**

**hidrossolúvel**

**Enzimas oxidativas**  
**Citocromo P-450**

**Fármaco:**  
ativo  
inativo

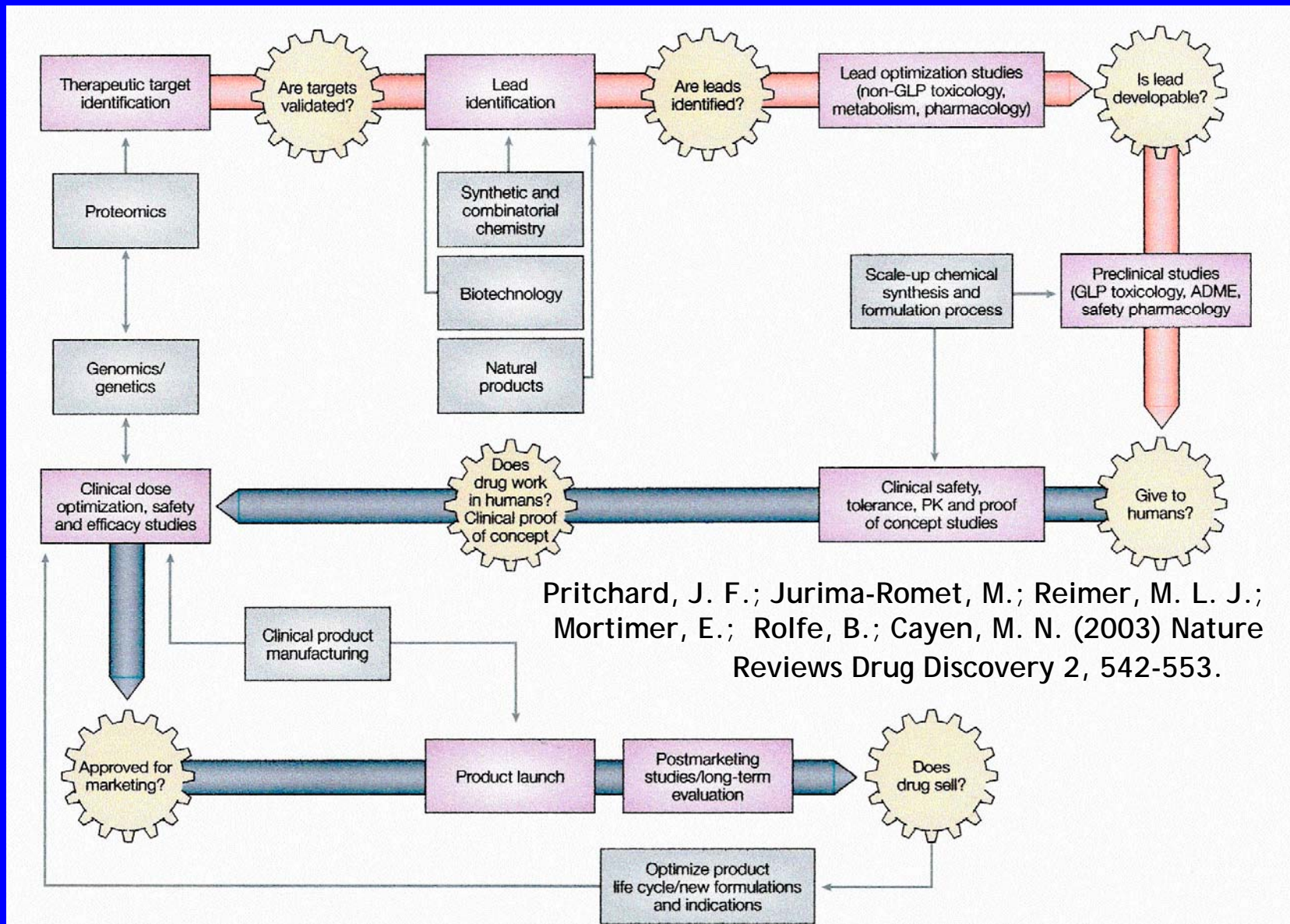
**Bioativação**  
**Biotransformação**

**Fígado**

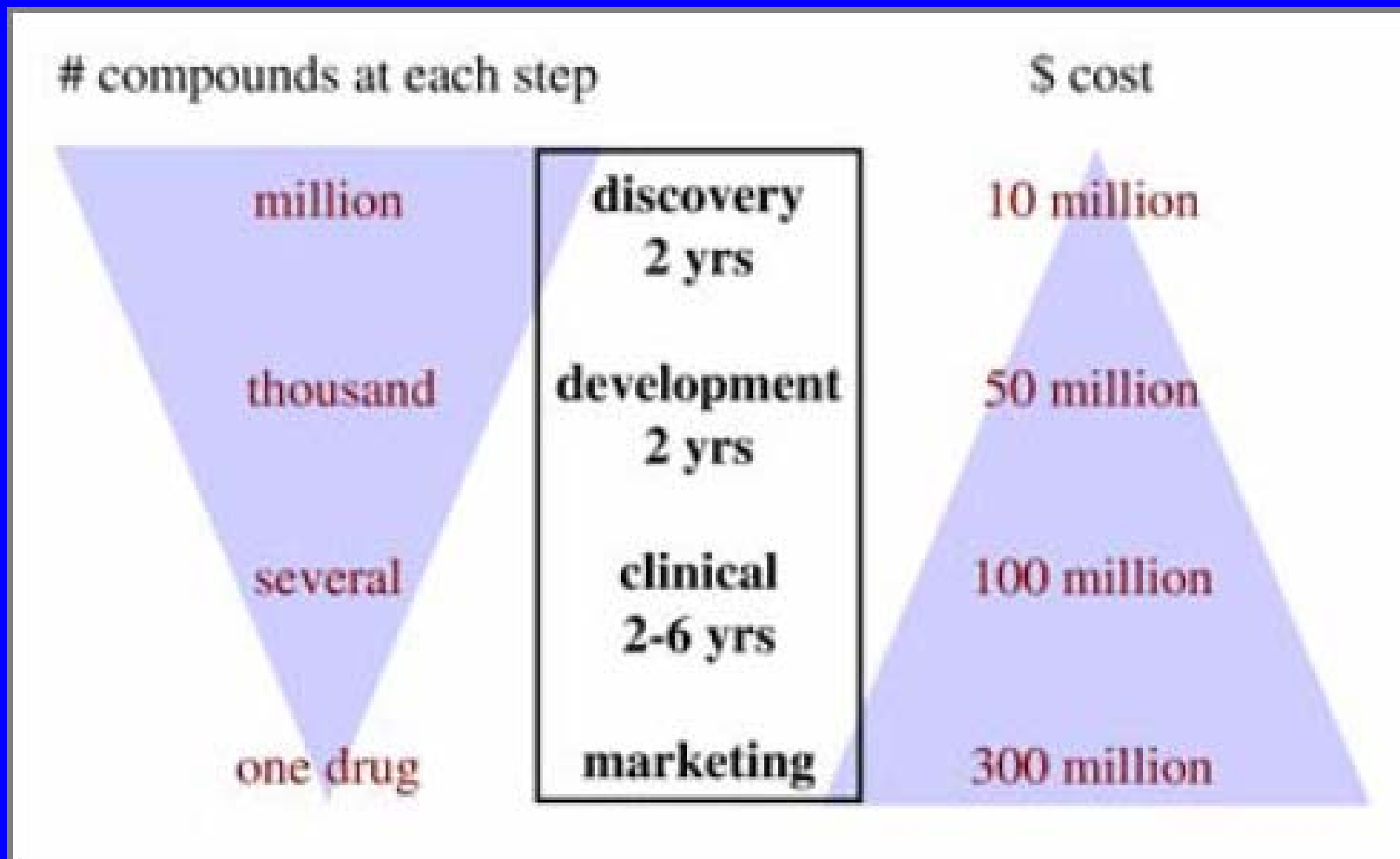
**Retículo microssomal**

**Sangue**

# Estratégias e a Sequência do Processo de Descoberta e Desenvolvimento de Novos Fármacos



# Pirâmide da Descoberta de Fármacos

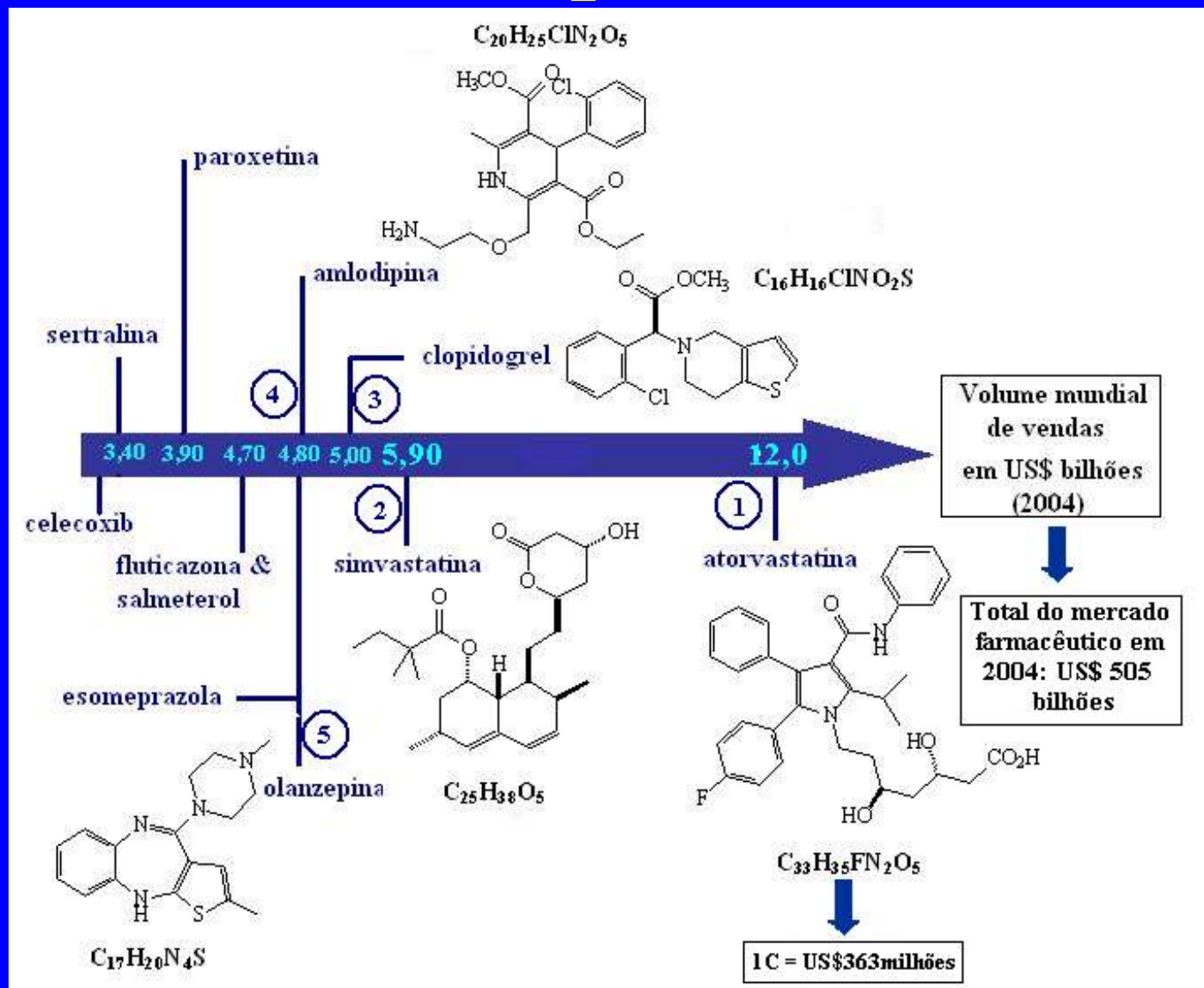


Lipper, R. A. (1999) *Modern Drug Discovery* 2 (1), 55-60.

Rawlins, M. D. (2004) *Nature Reviews Drug Discovery* 3, 360-364.

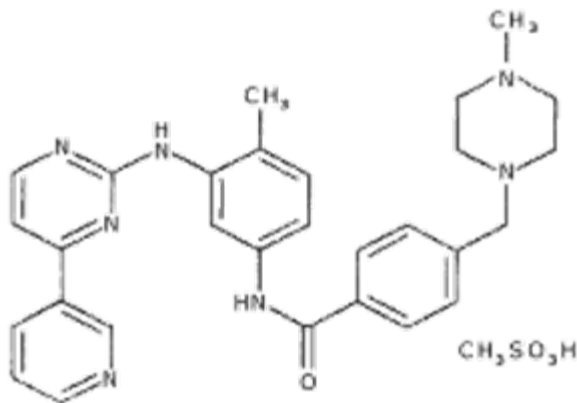


# A Recompensa !!!!!



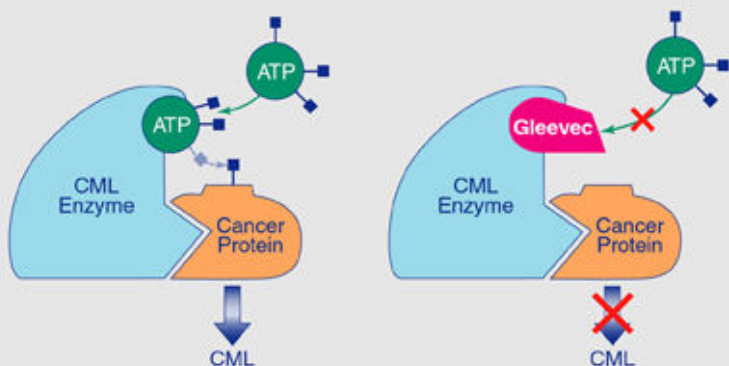
Dez principais fármacos em vendas (2004)

# “Blockbusters”



Imatinib (Gleevec-Novartis)  
Leucemia Mielóide Crônica

## Gleevec: HOW IT WORKS



Company	Potential Blockbuster Drug(s)	Estimated 2008 Sales From Pipeline (\$mil)
AstraZeneca	Exanta	\$972
AtheroGenics	AGI-1067	1,310
Aventis	Genasense	937
Bristol-Myers Squibb	Erbitux	804
Eli Lilly	Cymbalta	987
Genentech	Avastin	1,335
GlaxoSmithKline	Bonviva**	700
Novartis	Prexige	784
Pfizer	Inspra, Caduet, pregabalin, Indiplon, lasofoxifene	5,463
Roche	Avastin, Bonviva**	2,035

Prices as of Oct. 7. EPS: Earnings per share. \*Annualized; projected over the next three to five years. \*\*Joint development. Sources: FT Interactive, Reuters Fundamentals and Thomson First Call via FactSet Research Systems; Datamonitor

# Possíveis Origens para Novos Candidatos à Fármacos

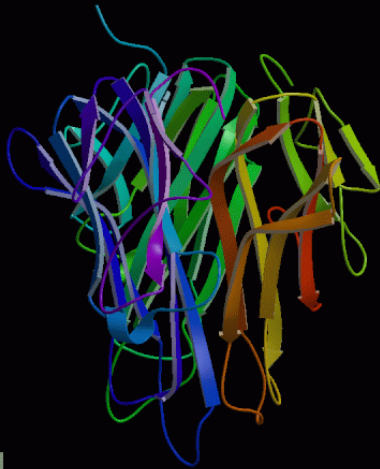
- Substâncias de Baixo Peso Molecular
  - Produtos Naturais
    - Caldo Fermentativos
    - Extrato de Plantas
    - Fluidos Animais (*e.g.*, veneno de cobra)
  - Substâncias de Origem Sintética
    - Química Medicinal
    - Química Combinatorial
- Substâncias de Elevado Peso Molecular
  - Produtos Naturais
  - Produtos Recombinares
  - Produtos Quiméricos

# Produção de Proteínas e Peptídeos Recombinantes (Processos Biotecnológicos)

Nome Genérico	Nome Fantasia	Uso Terapêutico
Insulina Humana	Humulin (Lilly) Novolin (Novo Nordisk)	Insulin dependent diabetes
Hormônio do crescimento	Protopin (Genentech) Humatrope (Lilly) Nutropin (Genentech)	Deficiência do Hormônio do Crescimento em crianças;
Vacina Hepatite B	Engerix-B (SmithKline Beecham) Recombivax HB (MSD)	Prevenção da Hepatite B
Interferon alfa-2a	Roferon-A (Hoffman-	Sarcoma de Kaposi (AIDS)
Muromonab-CD3	Orthoclone OKT 3 (Ortho)	Auxiliar na terapia da rejeição à transplantes de coração e rins



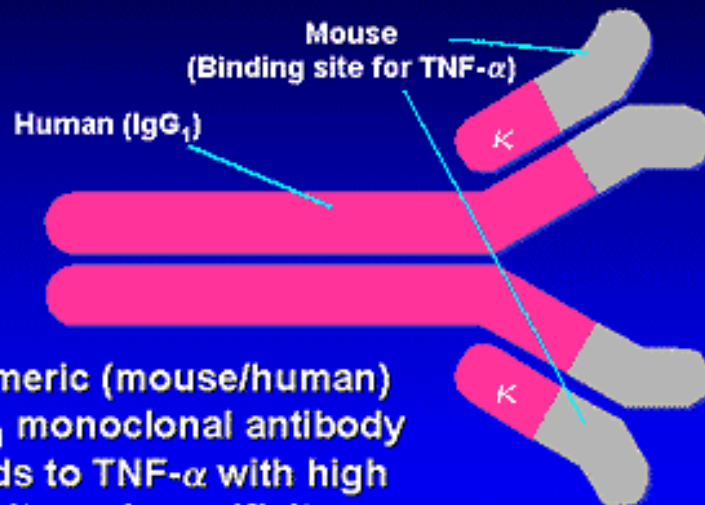
# Anticorpos Monoclonais Quiméricos



Cytokines  
Wd



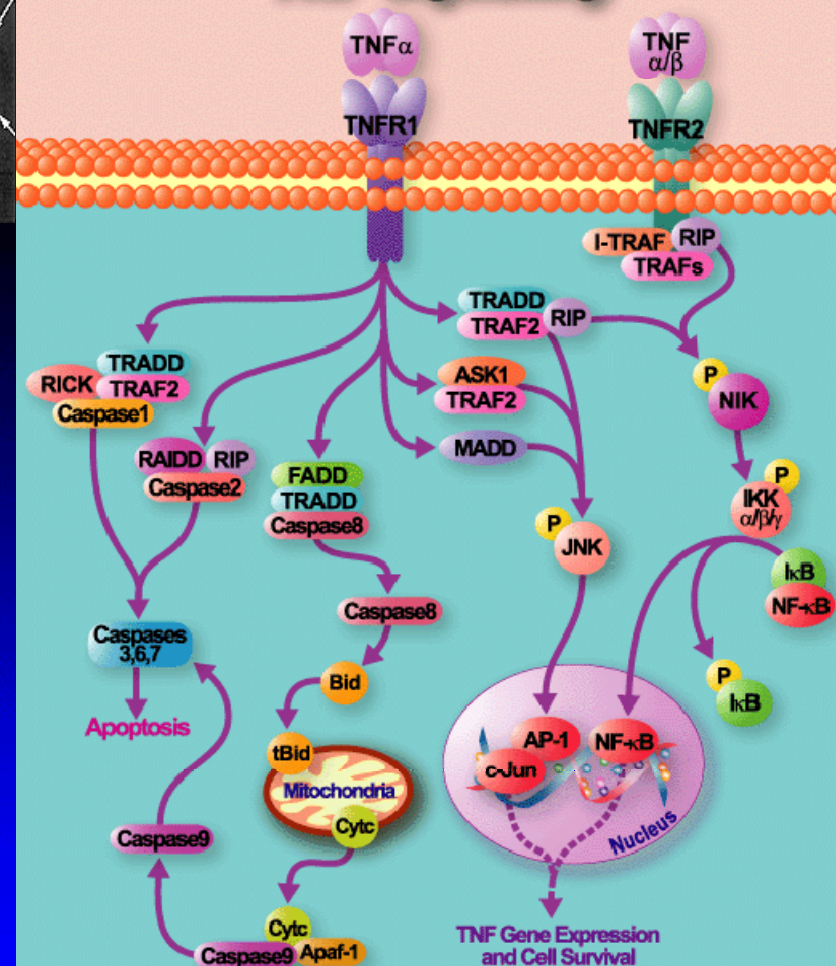
## Infliximab



- Chimeric (mouse/human) IgG<sub>1</sub> monoclonal antibody
- Binds to TNF- $\alpha$  with high affinity and specificity

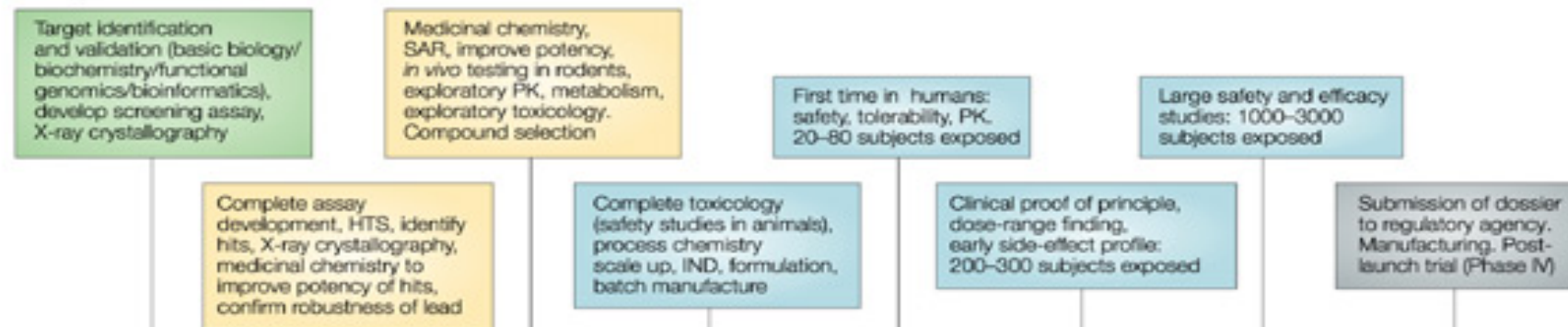
Knight DM, et al. *Mol Immunol.* 1993;30:1443-1453.

## TNF Signaling





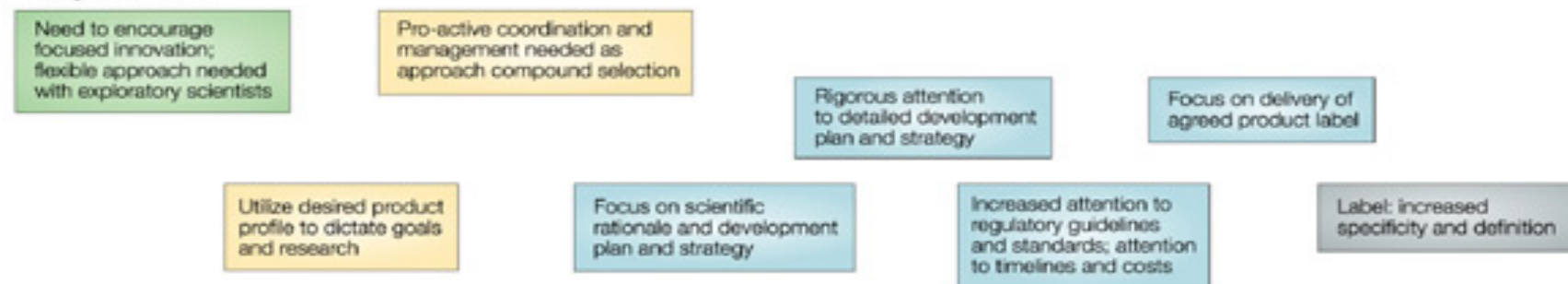
## Description of activities



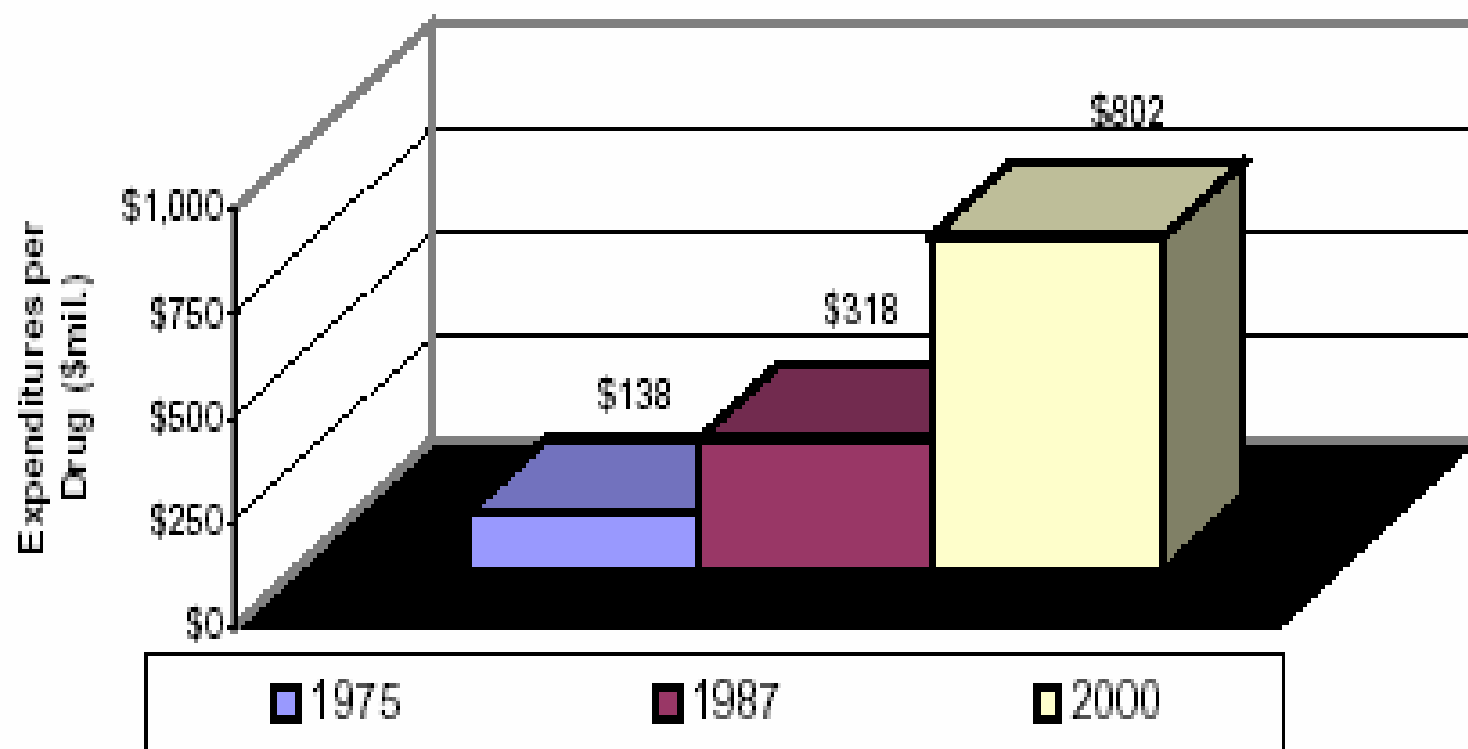
## Process for competitively selected projects



## Management issues

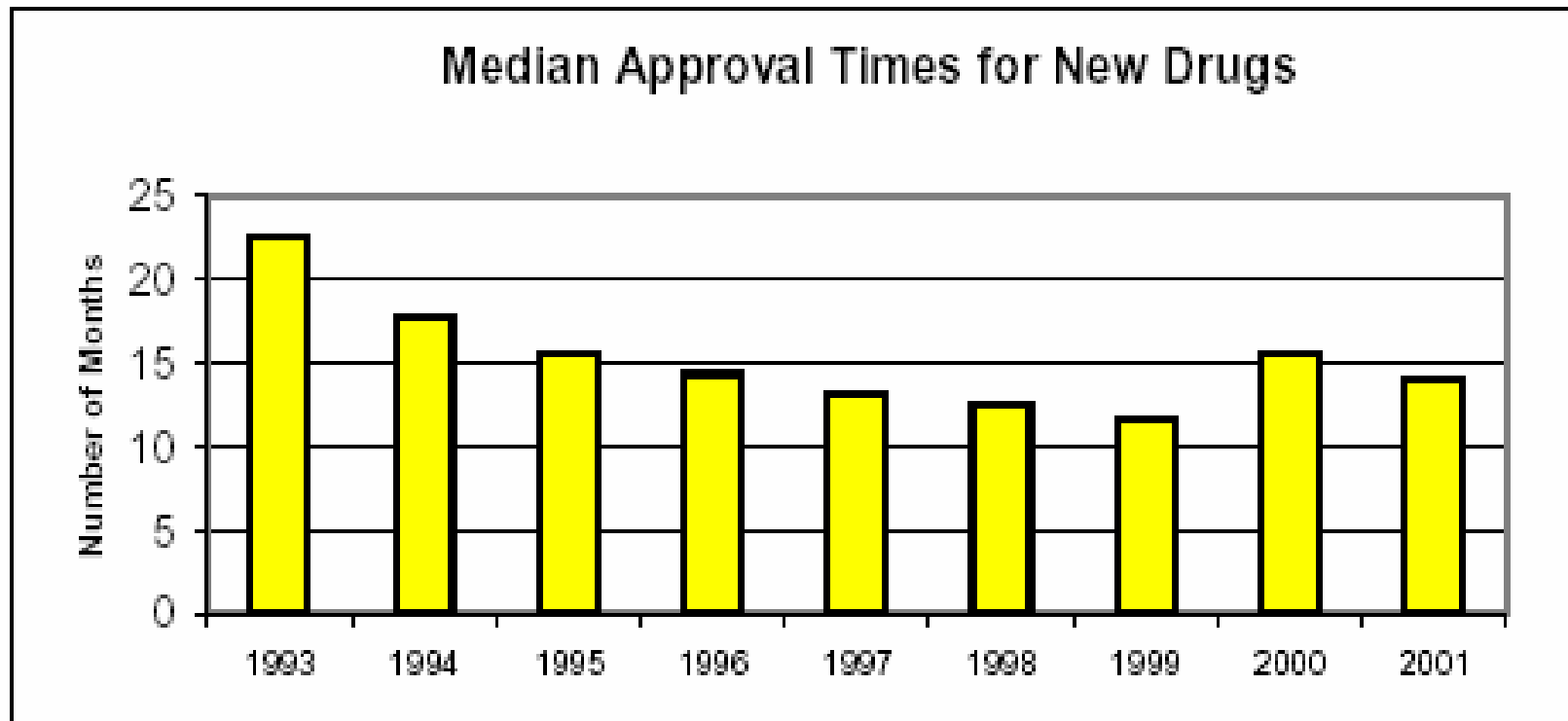


# Custo Atual para o Desenvolvimento de Fármacos (???)



Source: *Journal of Health Economics* 22 (2003): 151-185

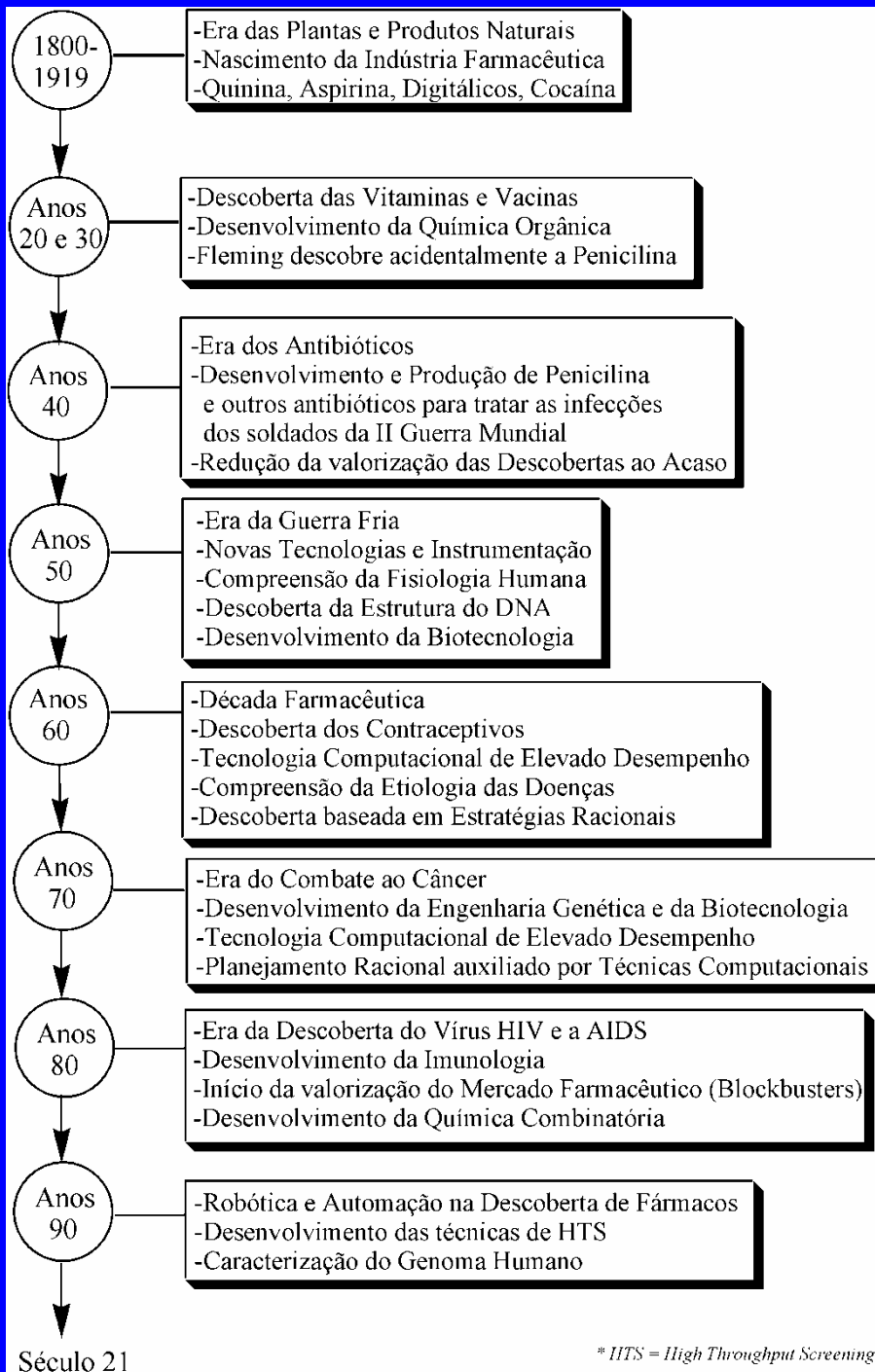
# Tempo Médio para Aprovar um Fármaco



Source: U.S. Food and Drug Administration



# Evolução Temporal do Processo de Descoberta de Fármacos



Fraga, C. A. M. & Barreiro, E. J. (2005)  
Química Nova 26, Suplemento, in press.

# A DESCOBERTA DE FÁRMACOS

A busca por substâncias terapeuticamente úteis era centrada na investigação de plantas que cresciam em jardins de instituições religiosas

Uma importante mudança de paradigma ocorreu no momento em que os alcalóides foram isolados e moléculas simples como o ácido acetil salicílico foram sintetizadas



Pen ts'ao

Shen Nung



*Dichroa febrifuga*



*Rauwolfia serpentina*



Theophrastus

5100 a.C.

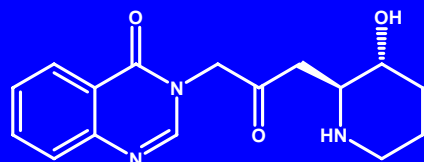
Gh'ang Shan

1000 a.C.

hindus

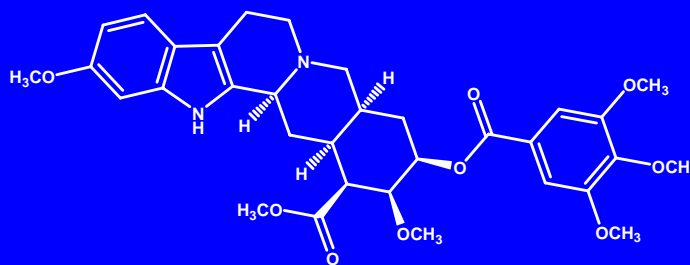
300 a.C.

opióides



(1)

*febrifugina*



(2)

*reserpina*



*papoula*

Chen, K.K. (1925) J. Am. Pharm. Assoc., 14, 189.

Jang, C.S., *et al.* (1946) Science, 103, 59.

Grob, D. (1955) J. Chronic Diseases, 1, 229.

Silverman, R.B. (2004) The Organic Chemistry of Drug Design and Drug Action, Elsevier, San Diego, 10.



## Colchicum autumnale



## Cinchona

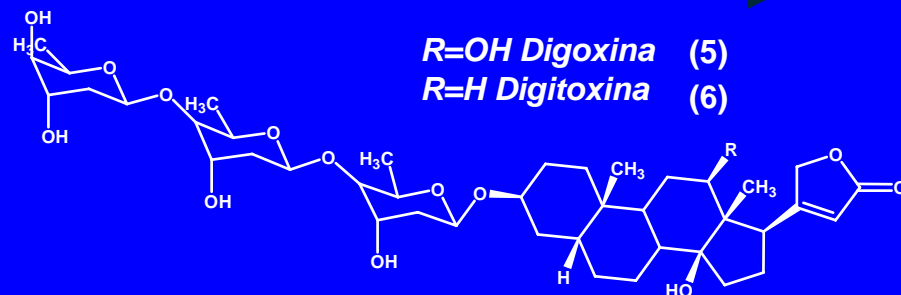
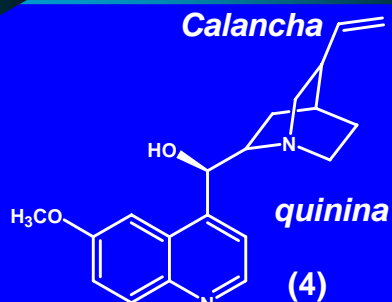
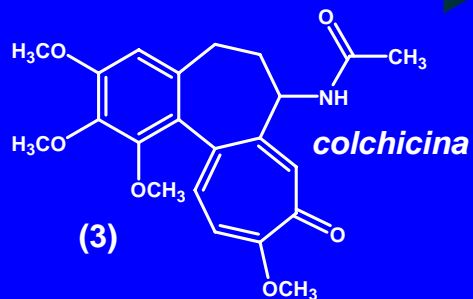


## *Digitalis purpurea*

500

1633

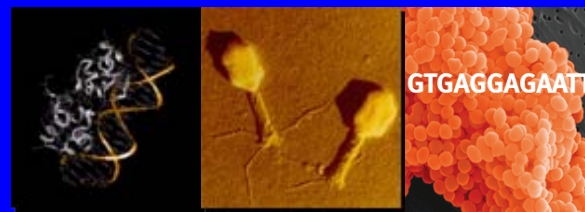
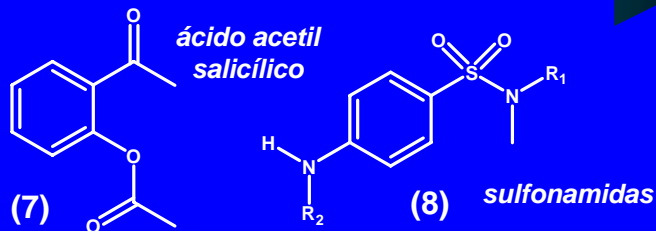
1785



## Início dos fármacos sintéticos e da Química Medicinal

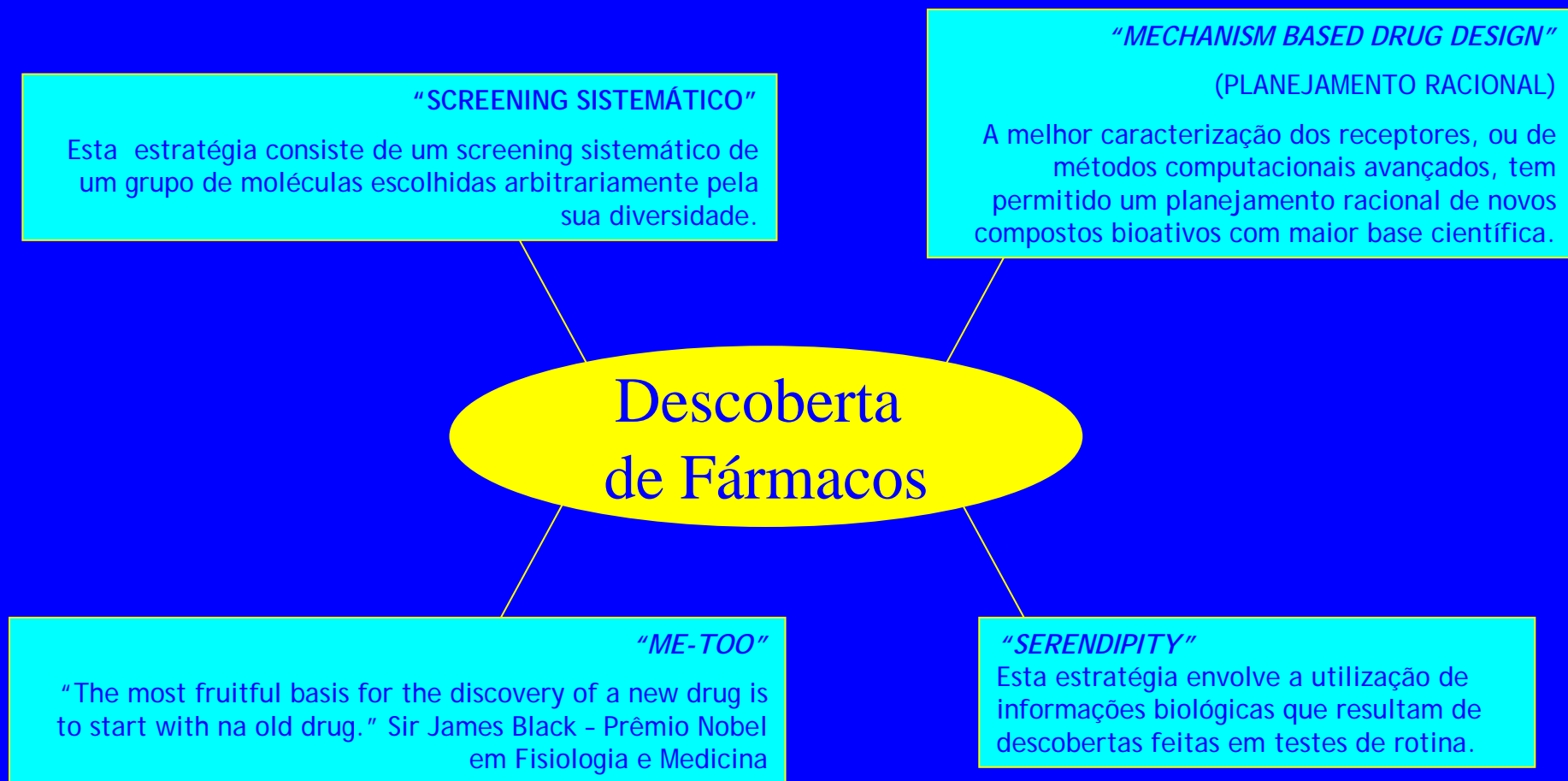
1900

## Futuro



Tracy, J.W. & Webster, L.T. (1993) em *The Pharmacological Basis of Therapeutics*, McGraw Hill, Nova Tork, 965.  
Hoffman, B.F. & Bigger, T. (1991) em *As Bases Farmacológicas da Terapêutica*, Guanabara Kogaan, Rio de Janeiro, 536.  
Newman, D.J.; Cragg, G.M.; Sneader, K.M. (2004) *J. Nat. Prod.*, 66, 1022.  
Silverman, R.B. (2004) *The Organic Chemistry of Drug Design and Drug Action*, Elsevier, San Diego, 10.  
Burger, A. (1980) *Burger's Medicinal Chemistry*, Willey, Nova York, 23.

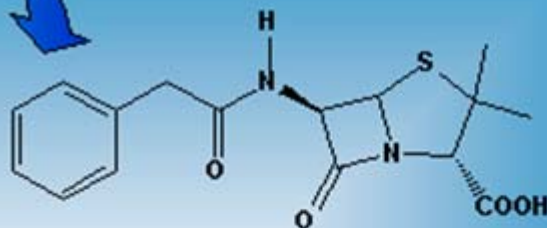
# Estratégias Empregadas na Descoberta de Novos Fármacos



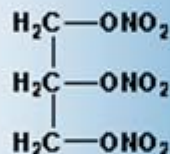
*"... in the recent years the paradigms of drug research changed significantly..."*  
*Kubinyi, H., 1995*

Fraga, C. A. M. & Barreiro, E. J. (2005)  
Química Nova 26, Suplemento, in press.

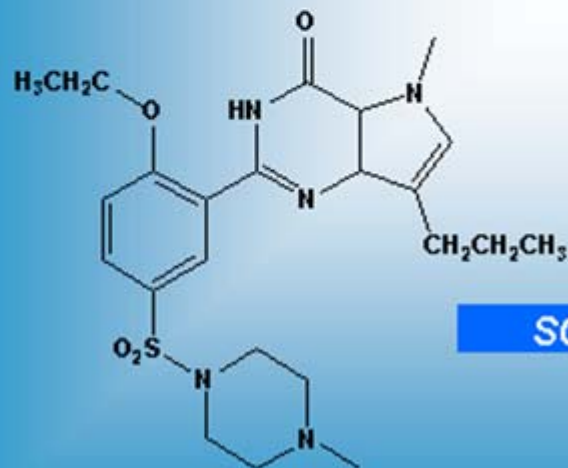




**Penicilina G**



**trinitroglicerina**



**Sildenafil**

**SOSA**

**Nova classe de  
fármacos úteis no  
tratamento da  
disfunção erétil**

## "SERENDIPITY"

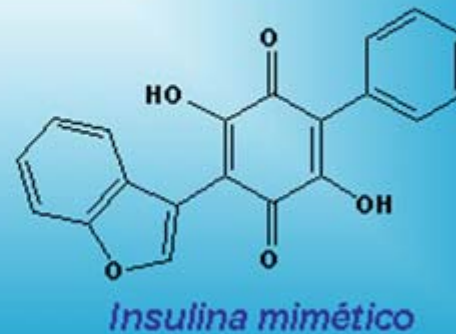
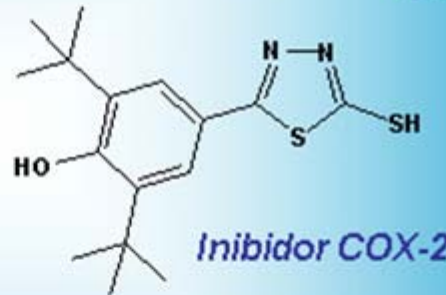
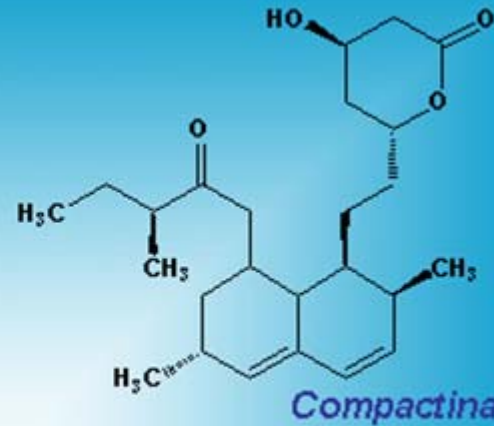
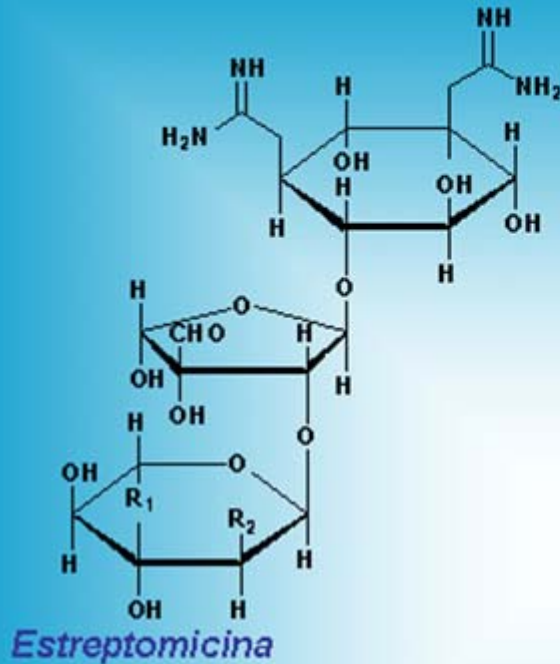
A word coined by Horace Walpole, who says (Let. to Mann, 28 Jan. 1754) that he had formed it upon the title of the fairy-tale 'The Three Princes of Serendip', the heroes of which '**were always making discoveries, by accidents and sagacity,** of things they were not in quest of"

Wermuth, C.G. (2004)  
J. Med. Chem., 47,  
1303.

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

Terret, N.K. (1996)  
Bioorg. Med. Chem.  
Lett., 6, 1819.

# "Screening Sistemático"



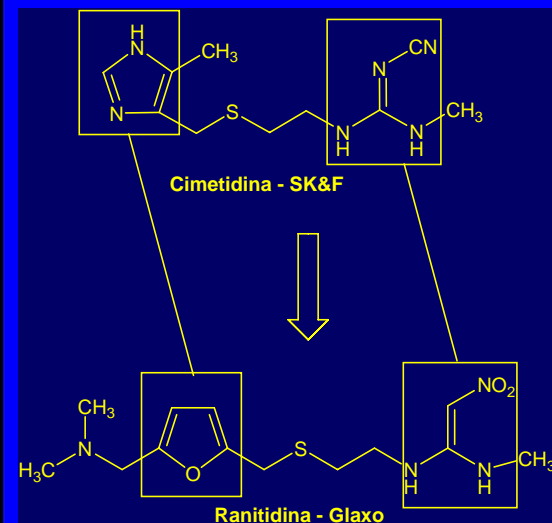
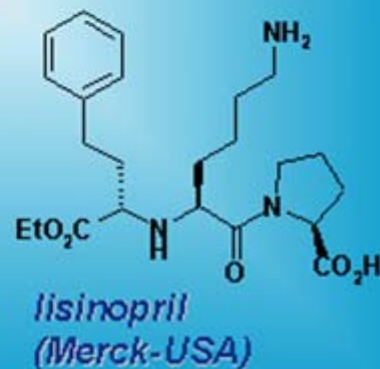
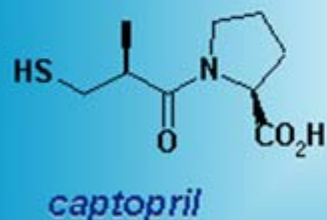
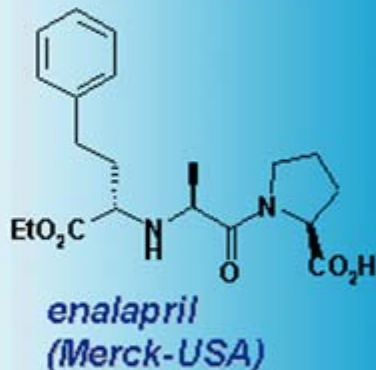
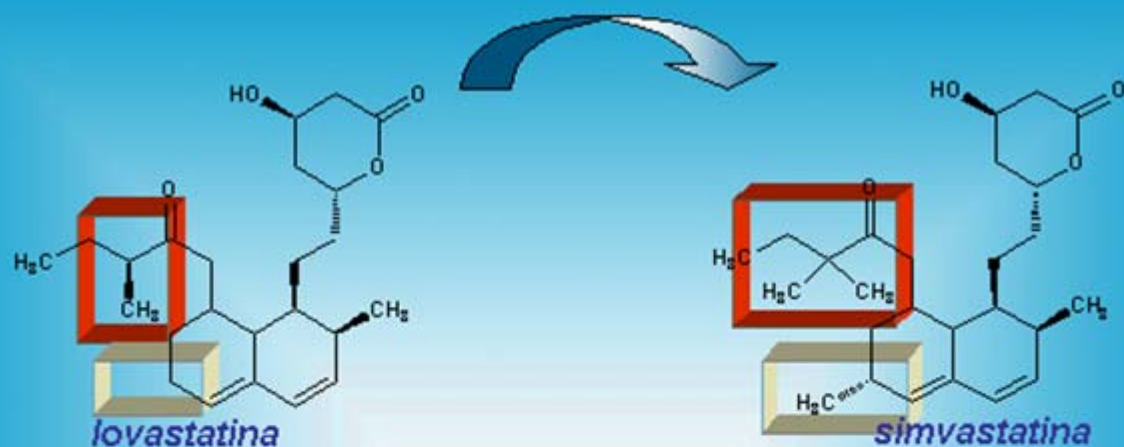
Nakamura, C.E. &  
Abeles, R.H. (1985)  
*Biochemistry*, 24,  
1364.

Liu, K. *et al.* (2000) *J. Med. Chem.*, 43, 3487.

Doman, T.N. *et al.*  
(2002) *J. Med. Chem.*,  
45, 2213.

Drews, J. (2003) *Drug  
Discovery Today*, 8,  
411.

# "Me-Too's"



Wermuth, C.G. (2004)  
J. Med. Chem., 47,  
1303.

Raju, T.N.K. (2000)  
Lancet, 355, 121.

Cushman, D.W. &  
Ondetti, M.A. (1991)  
Hypertension, 17, 589.

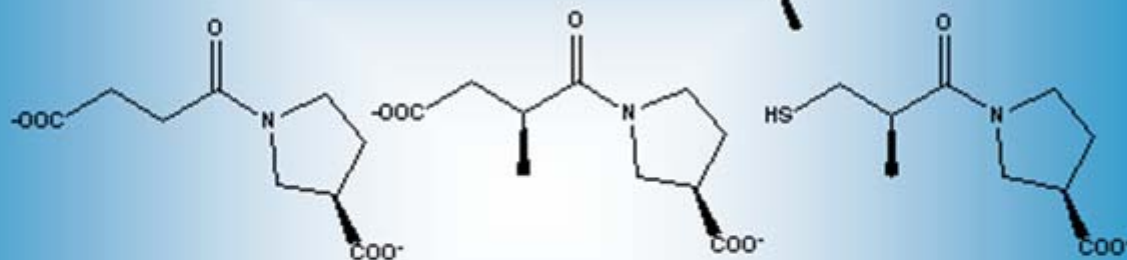
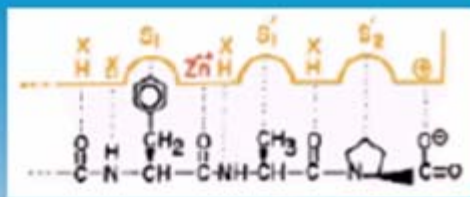
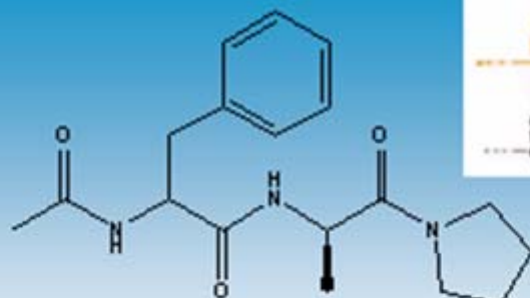
Tobert, J.A. (2003)  
Nat. Rev. Drug Disc. 2,  
517.



# Planejamento Baseado no Mecanismo de Ação



Jararaca bothrops

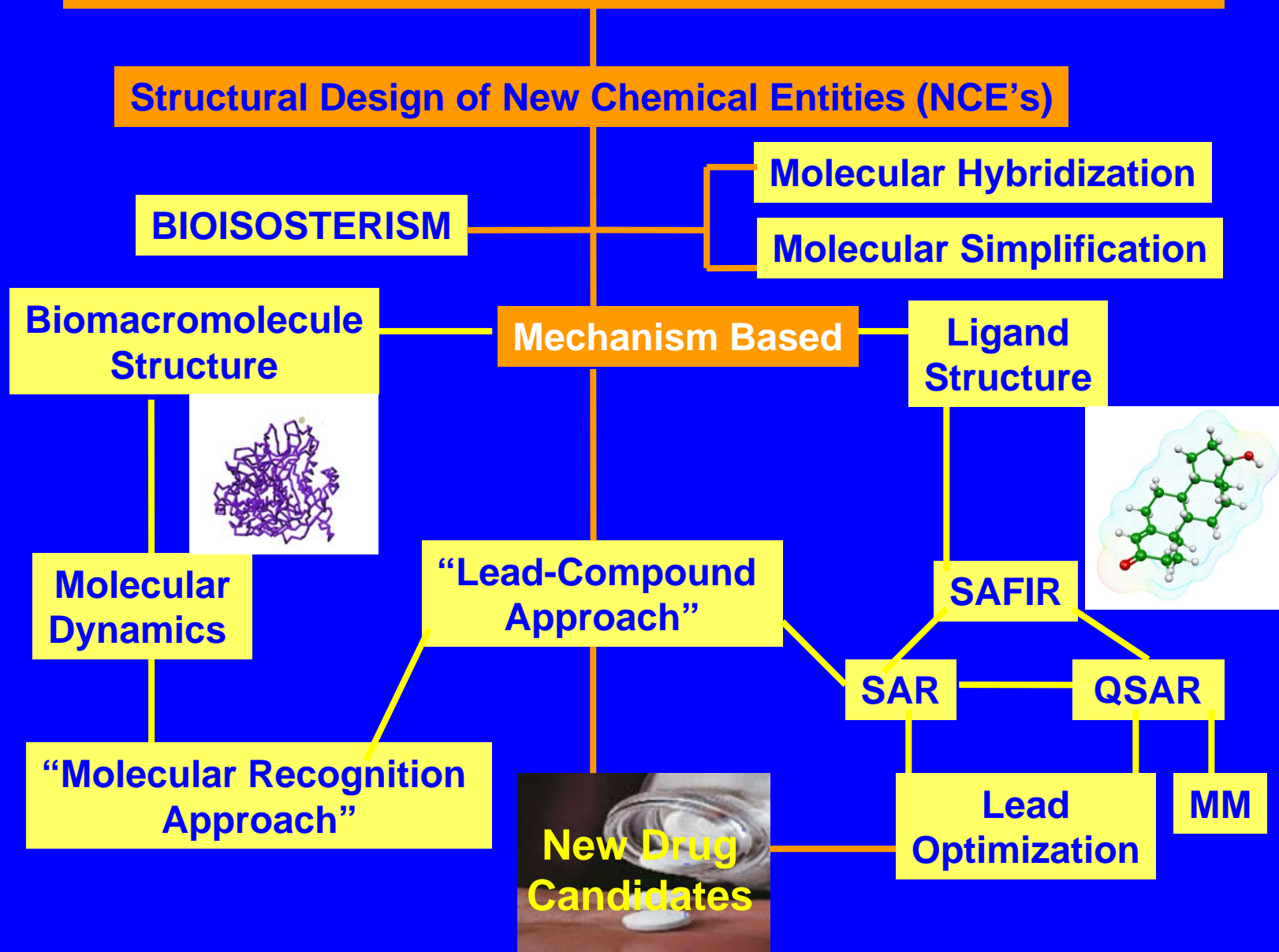


**Racionalização de  
novos compostos  
com maior base  
científica**

*“O processo moderno da descoberta de fármacos, baseado no mecanismo de ação, visa a identificação de novos compostos bioativos através do desenho planejado de sua arquitetura molecular, ou a identificação de um bioligante eficiente, cuja arquitetura molecular é planejada com base na estrutura de um biorreceptor conhecido, empregando estratégias de complementaridade molecular”*

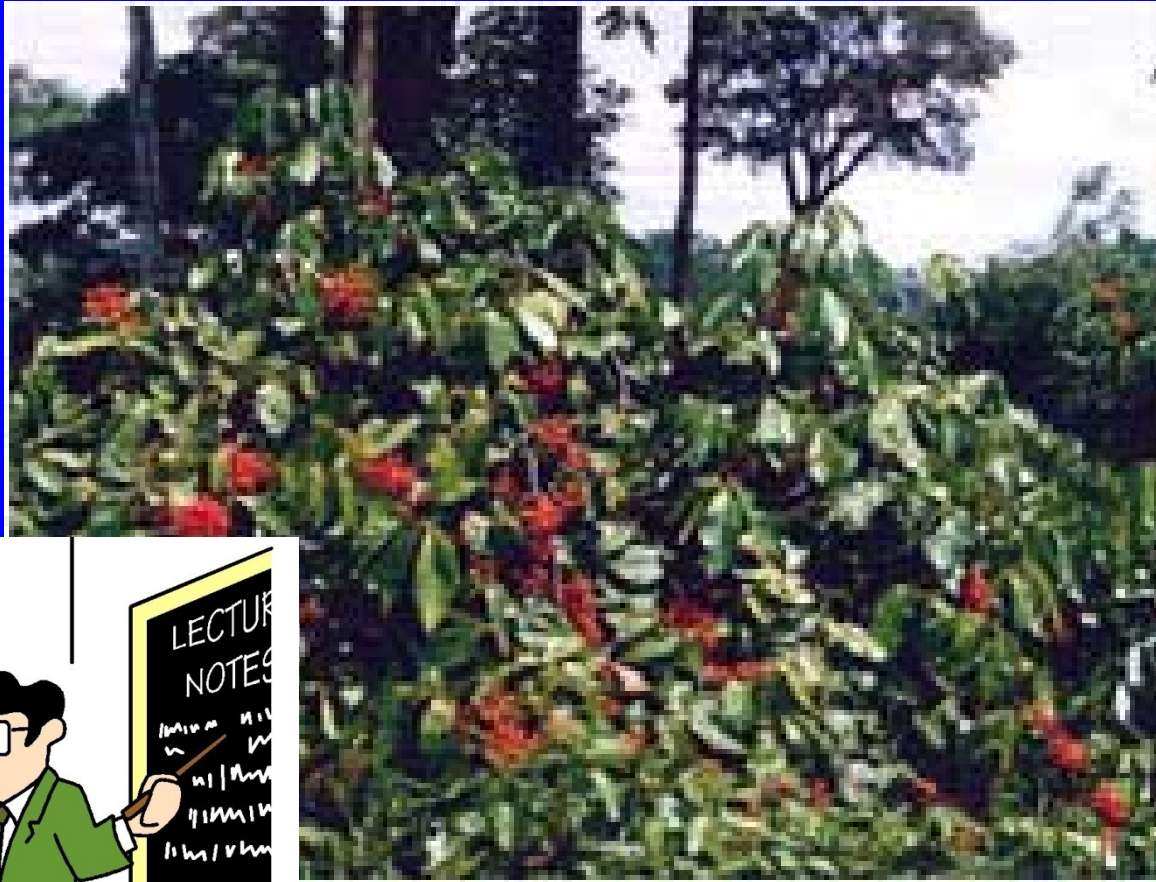
Fraga, C. A. M. & Barreiro, E. J. (2001) Química Medicinal, ArtMed, Porto Alegre, p.17.

## Rational Drug Discovery Strategies





# Os Principais Fármacos vieram da Natureza.....



*Paullinia cupuana*  
João F. Batendorf  
(1669)

**Indígenas da Amazônia:**

Estimulante

Diurético

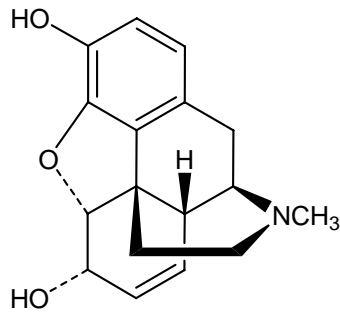
Antipirético

Enxaqueca

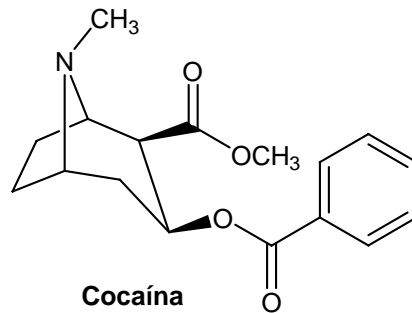
Inibidor do Appetite



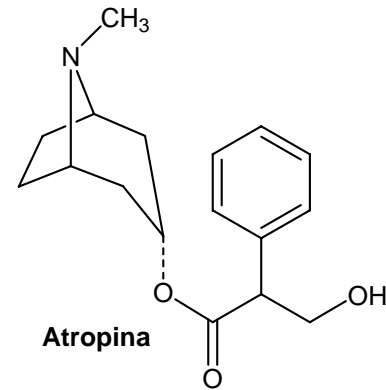
# No início.....Os Produtos Naturais...



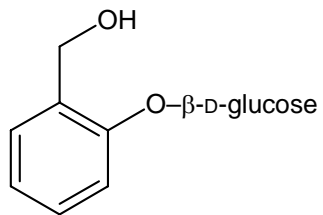
**Morfina**



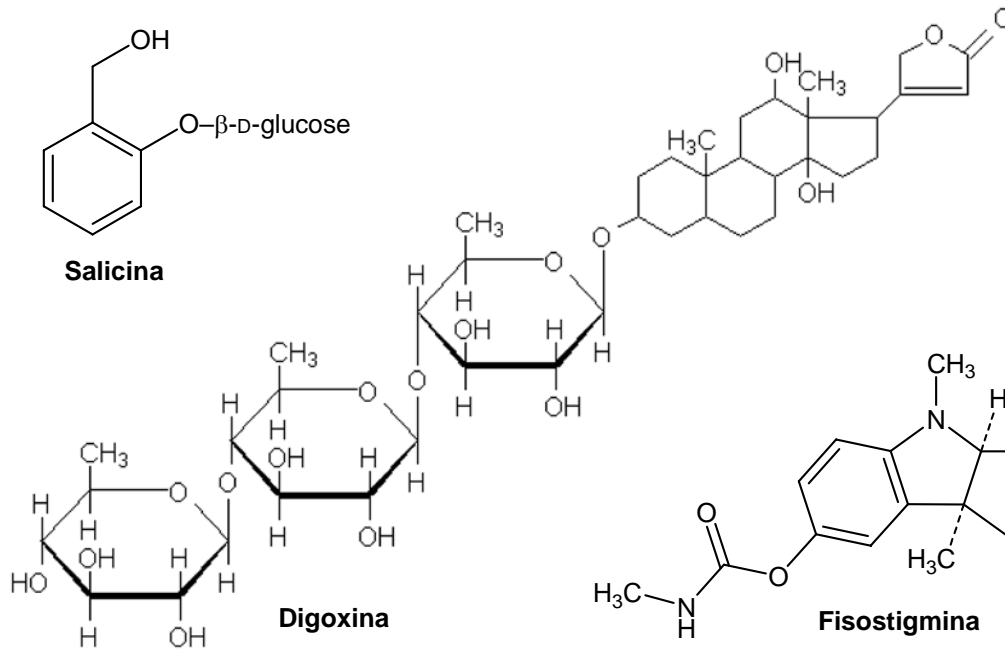
**Cocaína**



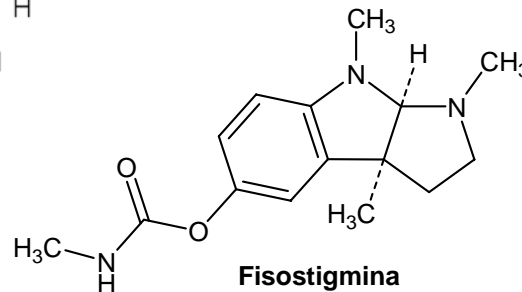
**Atropina**



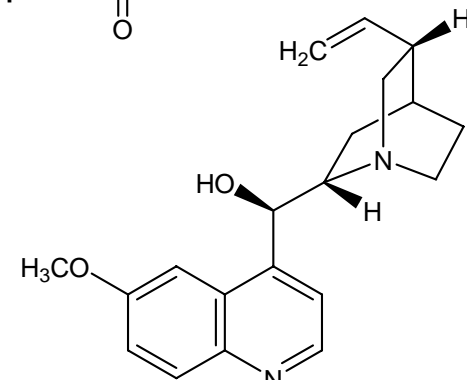
**Salicina**



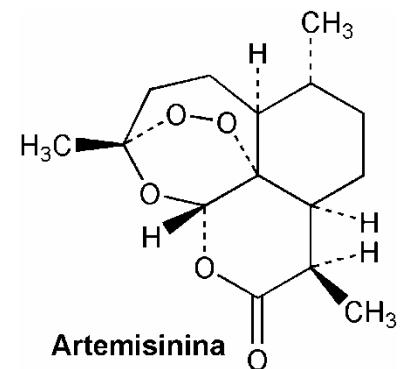
**Digoxina**



**Fisostigmina**



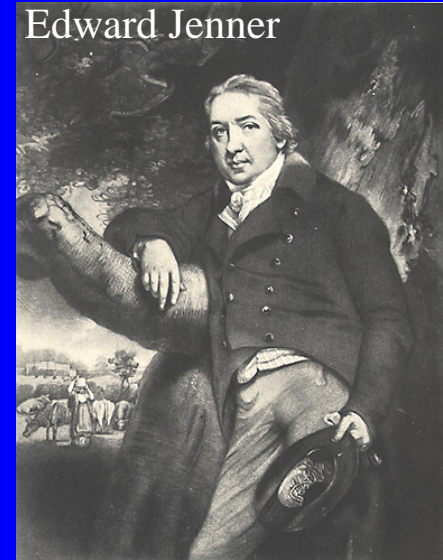
**Quinina**



**Artemisinina**

# O Nascimento da Quimioterapia Moderna

- O tratamento com substâncias químicas é chamada quimioterapia. A Quinina foi uma das primeiras substâncias químicas introduzidas no tratamento de doenças (malária).
- 1796: Jenner faz experimentos com formas atenuadas do vírus da varíola (VACINAS).
- 1861: Louis Pasteur: Relação microorganismo/patologia/antimicrobianos.
- 1910: Paul Erlich descobre o salvarsan, um derivado arsenical, que foi efetivo no tratamento da sífilis.



# A História da Antibioticoterapia

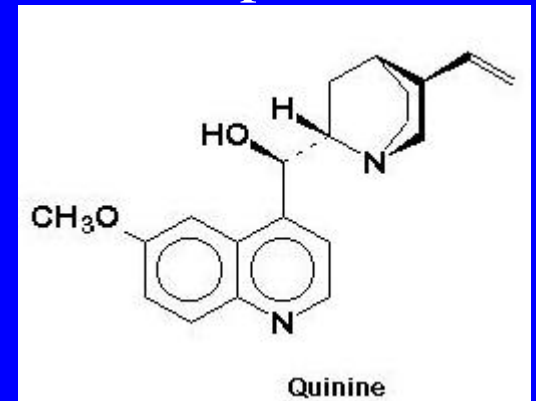
- Século 16: Índios Peruanos usavam o extrato da casca da árvore Chinchona para tratar a malária.
- Conta uma lenda espanhola que um soldado, sofrendo de um acesso de malária no meio da selva, bebeu a água amarronzada de uma pequena lagoa onde árvores de quinina haviam caído. Ele então foi dormir, e quando acordou sua febre havia desaparecido. O soldado concluiu que a água responsável pela cura era um "chá" feito do tronco e casca das árvores embebidos na água.
- Em 1633 um jesuíta chamado Padre Calancha descreveu as propriedades de cura da árvore na Crônica de Santo Agostinho: *" Uma árvore cresce, que eles chamam de árvore da febre, na região de Loxa, cuja casca tem cor de canela. Quando transformada em pó, juntando-se uma quantidade equivalente ao peso de duas moedas de prata, e oferecida ao paciente como bebida, ela cura febre e ... tem curado miraculosamente em Lima"*
- Em 1645, o padre Bartolomeu Tafur levou algumas cascas para Roma, onde seu uso espalhou-se entre os clérigos. Em 1654 a casca peruana foi introduzida na Inglaterra.



*Cinchona officinalis*

# Descoberta da Quinina

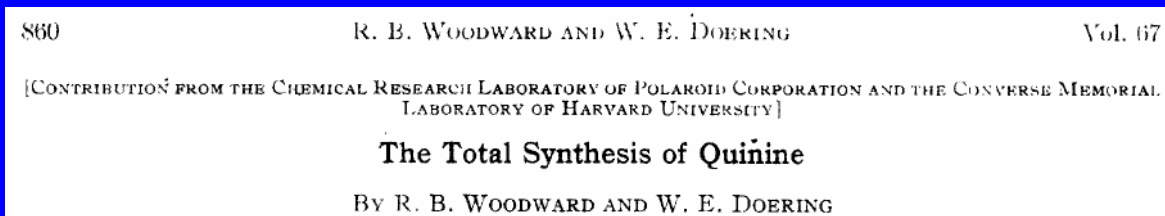
- 1739: Carl Linnaeus: *Cinchona officinalis*
- 1820: Joseph Pelletier e Joseph Caventou (Elucidação Estrutural)
- 1880: Colômbia sozinha exportou 6 milhões de libras para a Europa (Casca de Chinchona)
- 1930: Plantações holandesas em Java produziam 22 milhões de libras de casca, equivalente a 97% da quinina mundial.
- 1940: Segunda Guerra: o exército alemão apoderou-se de toda a reserva de quinina da Europa quando invadiu Amsterdã. Quando os japoneses invadiram a Indonésia em 1942 os EUA e seus aliados ficaram quase sem fornecimentos de quinina.





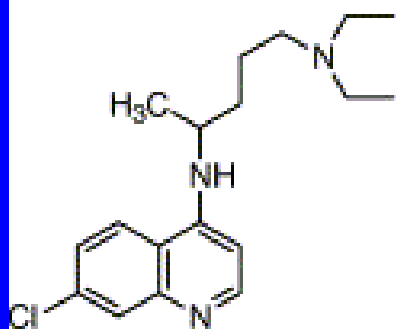
# A Era dos Análogos Sintéticos

- 1944: Robert Woodward e William Doering sintetizaram a quinina a partir do coal-tar (J. Am. Chem. Soc.)



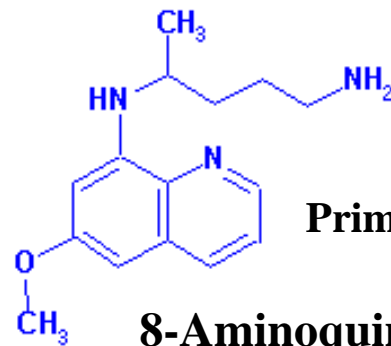
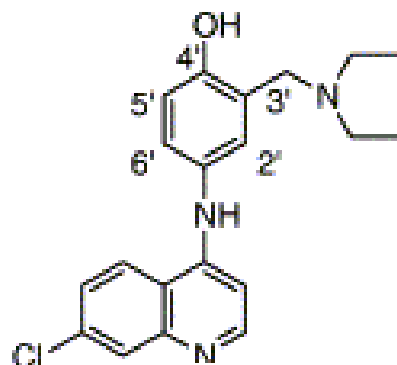
- Walter Reed Institute (USA): Novos Antimaláricos Sintéticos

(a) Chloroquine



4-Aminoquinolinas

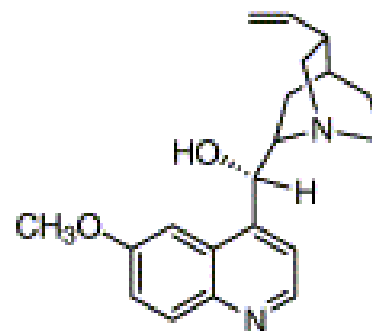
(b) Amodiaquine



Primaquina

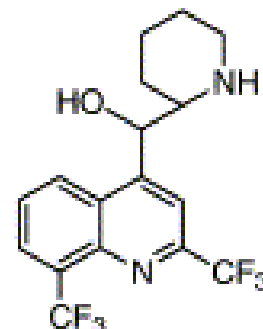
8-Aminoquinolinas

(c) Quinine



Quinolinometanóis

(d) Mefloquine



1971

TRENDS in Parasitology

# Malária

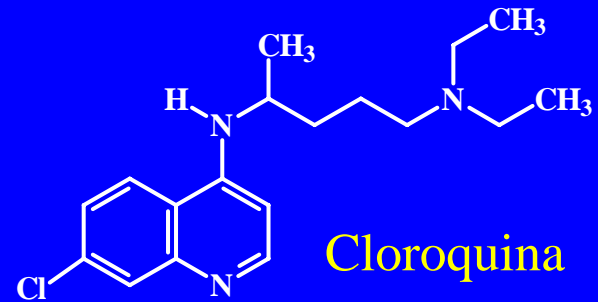
Doença parasitária causada pelo protozoário do gênero *Plasmodium*.  
Em humanos:

- P.vivax*
- P.malariae*
- P.ovale*
- P.falciparum*
- Transmissão : Picada da fêmea do mosquito *Anopheles*
- 1955 (OMS)- Lançamento do programa de erradicação da malária em regiões endêmicas- 250 milhões de casos de malária/ano.
  - uso de inseticidas: DDT
  - antimaláricos clássicos: quinina e cloroquina

Molyneux, D.H.; Floyd, K.; Barnish, G. and Frère, E.M. (1999) *Parasitology Today*, 15, 238-240.  
Hagan, P.; Chauhan, V. (1997) *Parasitology Today*, 13, 290-294.  
Watkins, W.M.; Marsh, K. (1997) *J. Pharm. Pharmacol.*, 49 (suppl. 2), 9-12.  
Rollason, P.V. (1997) *J. Pharm. Pharmacol.*, 49 (suppl. 2), 13-16.

# Espalhamento de cepas de *Plasmodium falciparum* cloroquina-resistentes

Source: Cambridge University



# O Ressurgimento da Malária e seu Impacto Social

## 1970- A volta da Malária

- Descontinuidade do programa da OMS
- Falta de incentivo monetário dos governos
- Migração em massa da população infectada
- Resistência do vetor (*Anopheles*) aos inseticidas

## PROBLEMÁTICA!!!

Resistência do *Plasmodium* aos Antimaláricos

### *ESTATÍSTICA ALARMANTE!!!*

*5617 crianças mortas/dia*

*234/h*

*4/min*

*Malária no RJ: 2001 registrados 70 casos*

*2002 foram registrados 34 casos*

### *Países mais atingidos:*

**Índia**

**Brasil**

Sri Lanka

Vietname

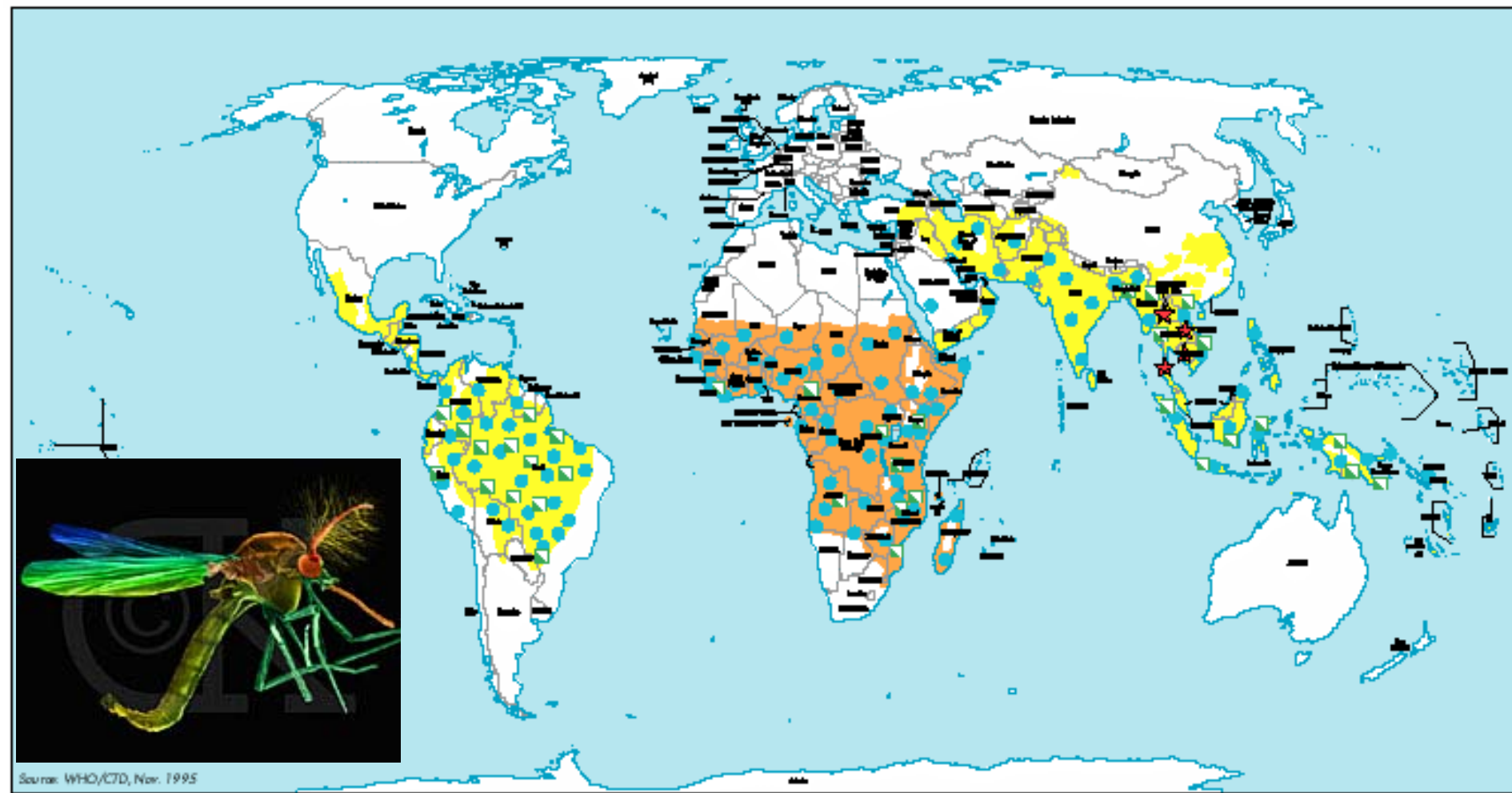
Colômbia

# DISTRIBUIÇÃO GEOGRÁFICA DA MALÁRIA

<http://www.worldbank.org/>

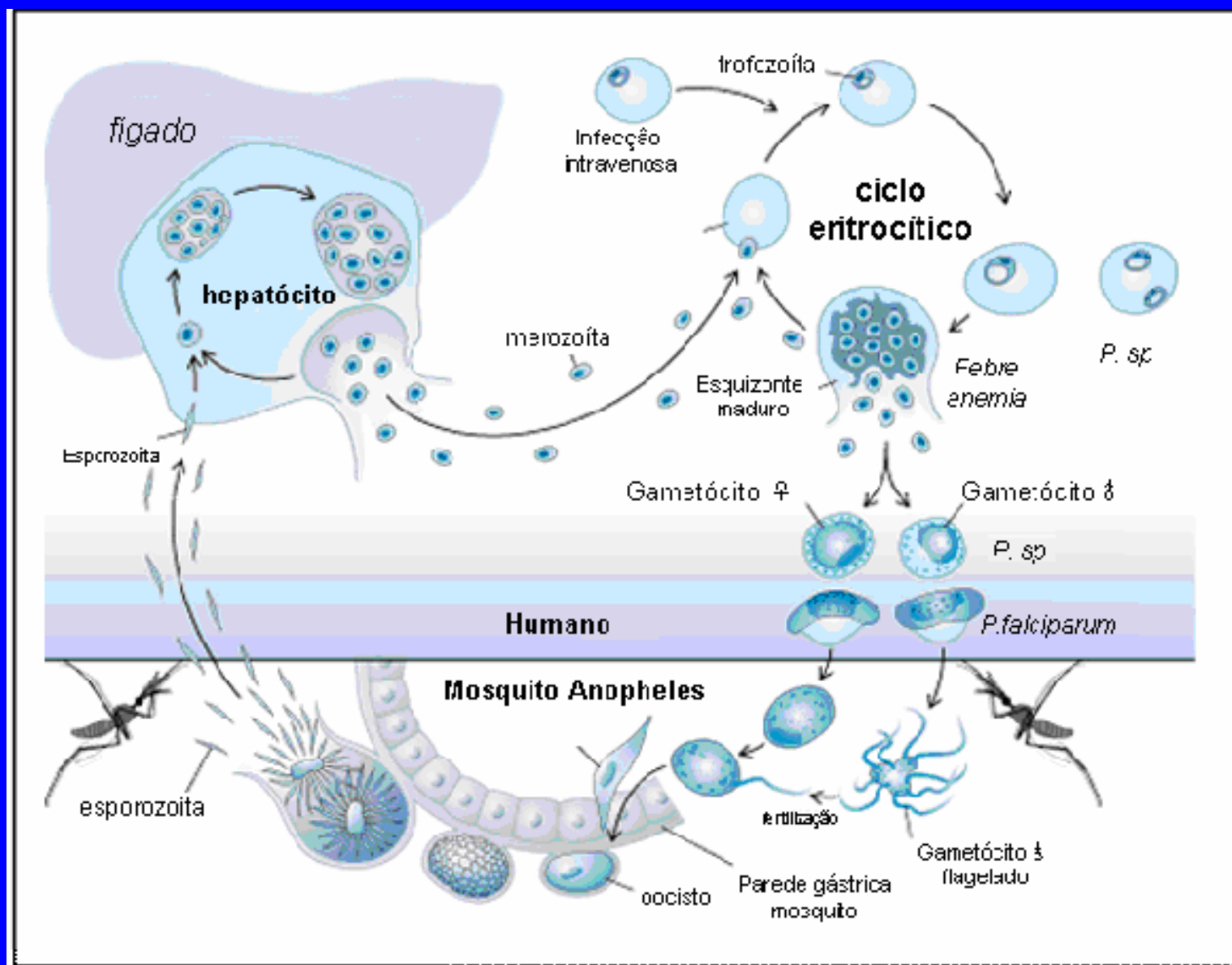
- 90% dos casos e morte
- casos de malária remanescente e morte

- resistência a cloroquina
- resistência a associação pirimetamina-sulfadoxina
- resistência cruzada



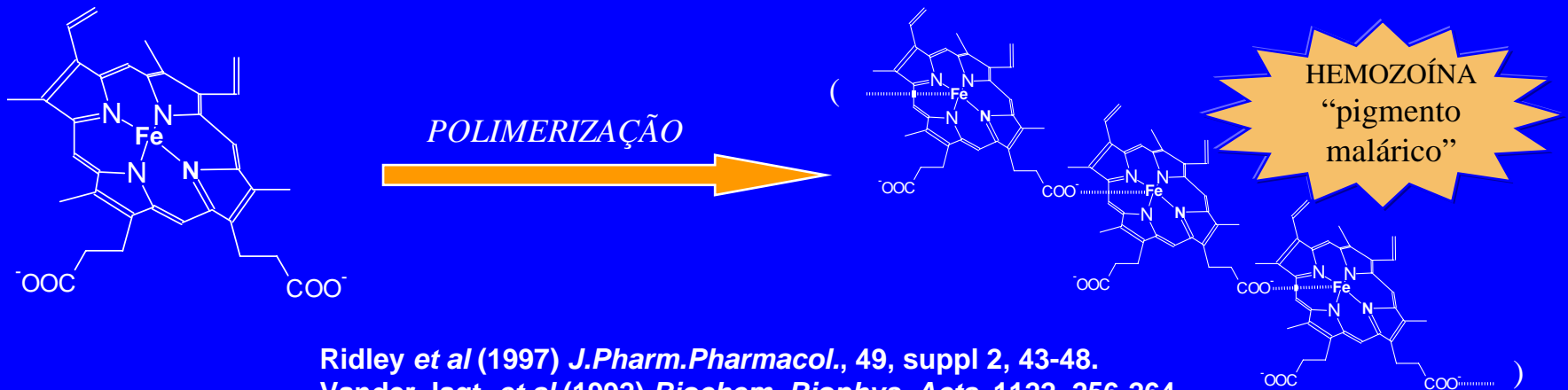
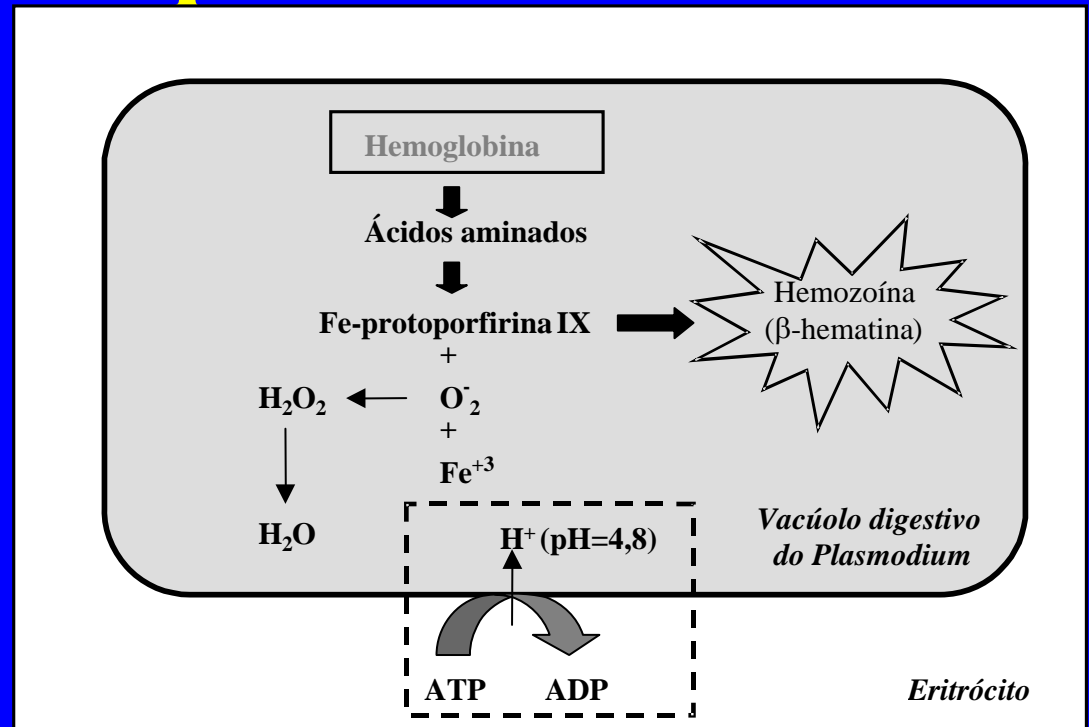


# O Ciclo Parasitário do *Plasmodium* (Olliaro, 1999)



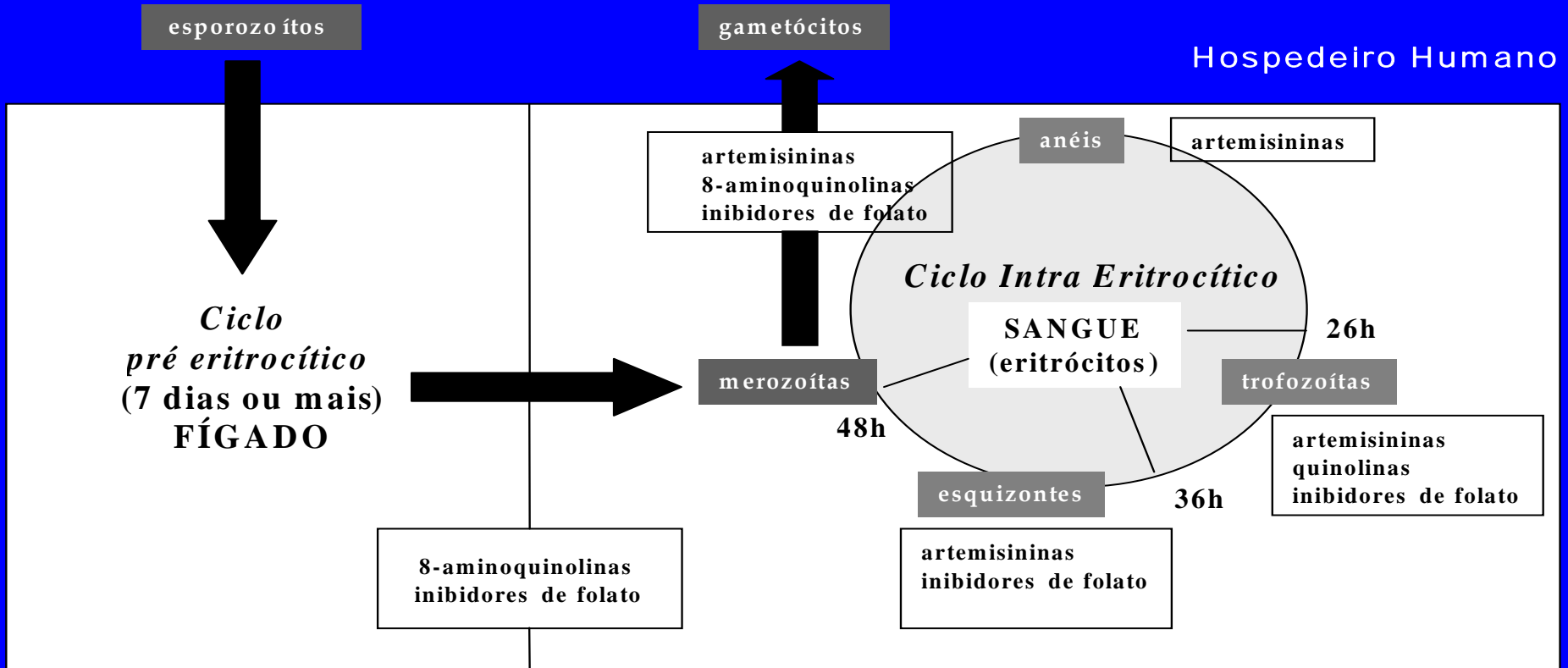
# Quimioterapia da Malária

**Vacúolo Digestivo do Plasmodium**  
degradação da hemoglobina e o mecanismo de detoxificação (polimerização)



Ridley et al (1997) *J.Pharm.Pharmacol.*, 49, suppl 2, 43-48.  
Vander Jagt et al (1992) *Biochem. Biophys. Acta*, 1122, 256-264.  
Zarchin et al (1986) *Biochem.Pharmacol.*, 35, 2435-2442.

# Alvo dos Principais Antimaláricos no Ciclo Parasitário do *P.falciparum*

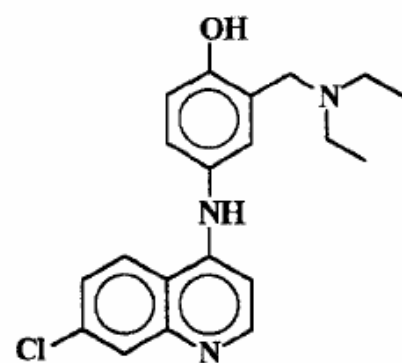


Olliaro, P. (2001) *Pharmacology & Therapeutics*, 89, 207-219.

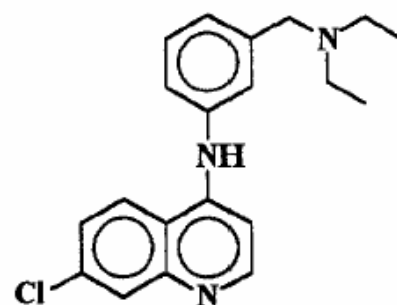
Foley, M. & Tilley, L. (1998) *Pharmacology & Therapeutics*, 79, 55-87

$$\frac{[\text{Drug}]_v}{[\text{Drug}]_o} = \text{Vacuolar drug accumulation ratio (VAR)}$$

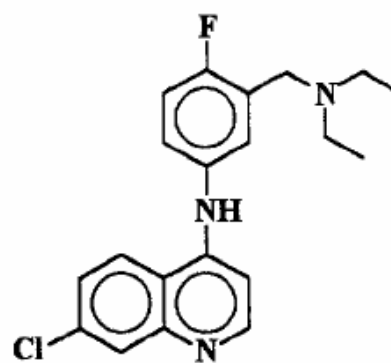
$$\text{CAR} = \text{VAR} \times \text{Fractional cell volume occupied by acid vacuoles} \quad (5)$$



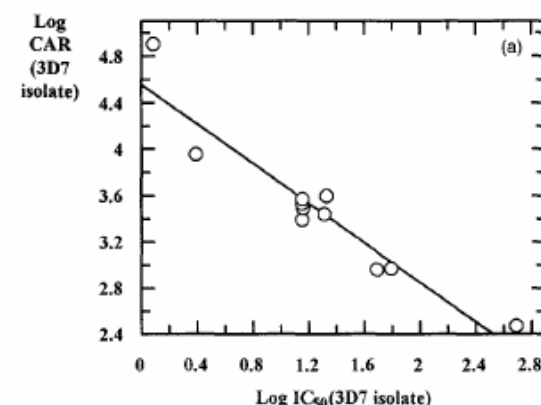
amodiaquine (AQ)



4'-dehydroxyamodiaquine  
(4deOH-AQ)



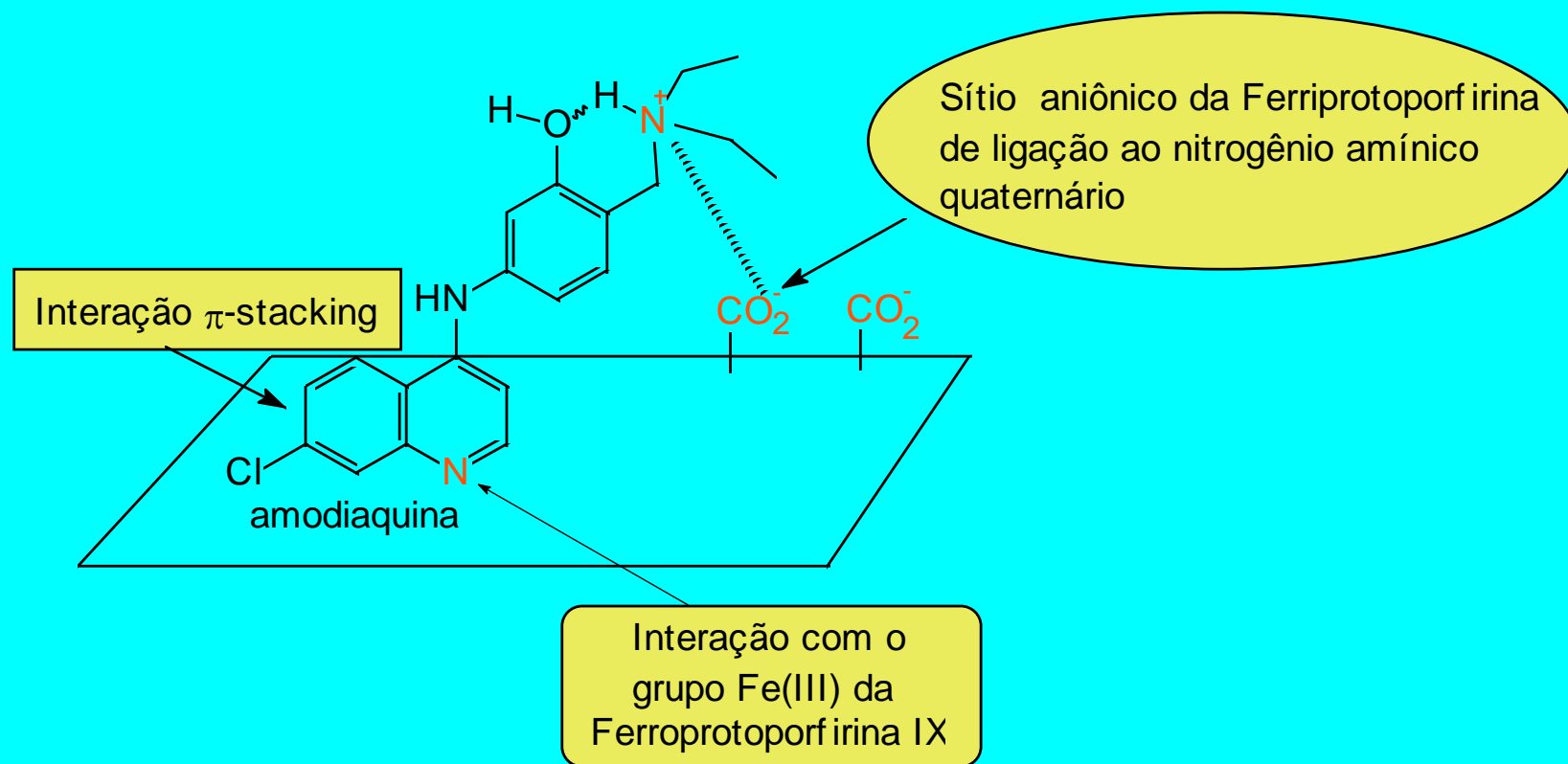
4'-dehydroxy-4'-fluoroamodiaquine  
(4F-AQ)



	Dissociation constants		Protonation equilibria (at pH 5.0)*		Protonation equilibria (at pH 7.4)*		Log D	
	pKa 1	pKa 2	% Monoprotic	% Diprotic	% Monoprotic	% Diprotic	pH 5.0	pH 7.4
AQ	8.14	7.08	0.753	99.175	52.183	32.432	-1.4	2.61
4deOH-AQ	9.18	7.26	0.540	99.453	56.373	41.995	-1.3	2.44
4F-AQ	8.70	7.26	0.526	99.454	53.367	41.860	-0.6	2.19

Hawley et al. (1996) The Role of Drug Accumulation in 4-Aminoquinoline Antimalarial Potency. *Biochem. Pharmacol.*, 52, 723-733.

# Modelo mecanístico proposto por Koh e colaboradores para a ação antimalárica de derivados quinolínicos



Koh et al (1994) *Eur.J.Med.Chem.*, **29**, 107-113.

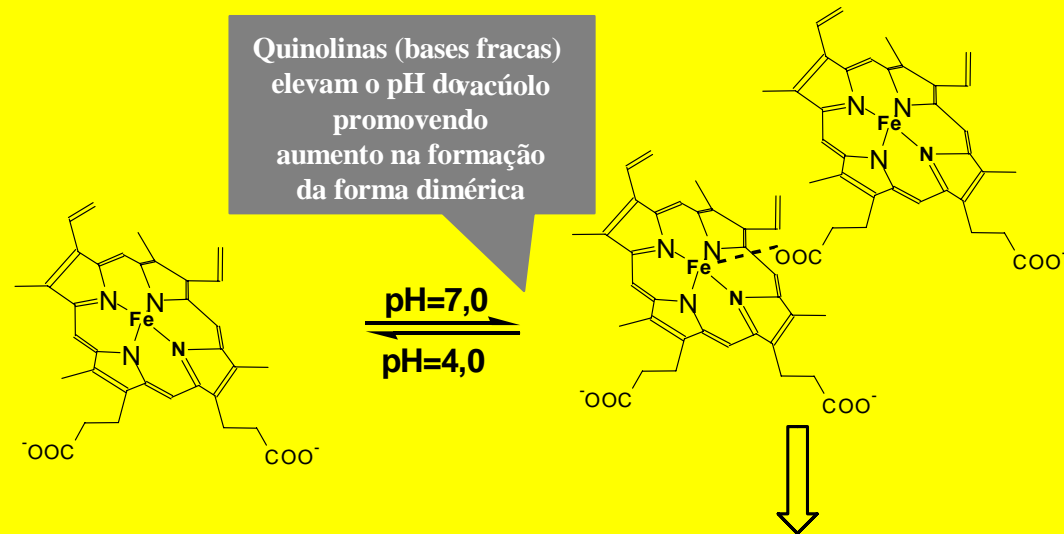


# Modelo mecanístico para a ação antimalárica derivados quinolínicos: cloroquina e mefloquina

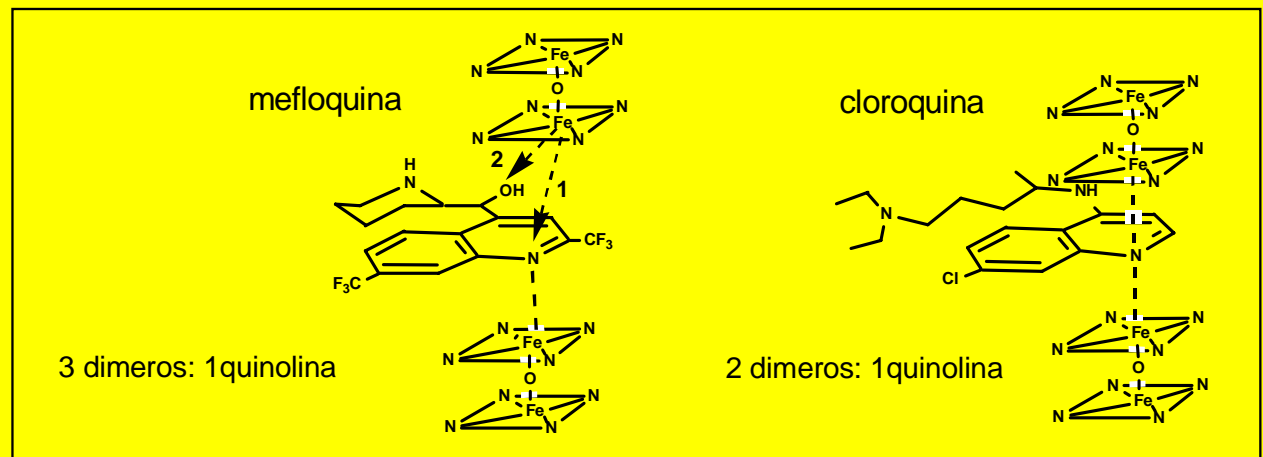
Vippagunta, S.R. et al (1999)  
*J. Med. Chem.*, 42, 4630-4639.

Dorn, A. et al (1998)  
*Biochem. Pharm.*, 55, 737-747.

Moreau, S. et al (1985)  
*Biochem. Biophysica Acta*, 840, 107-116.



Complexação do fármaco com a forma dimérica



# A Resistência do *Plasmodium* aos antimaláricos clássicos

## Teorias para explicar a resistência:

- 1) Inativação do fármaco pelo *Plasmodium*
- 2) Internalização reduzida do fármaco
- 3) Alteração do metabolismo do *plasmodium* de forma a eliminar etapas suscetíveis ao quimioterápico

HIPÓTESE MAIS ACEITA

GLICOPROTEÍNA HOMÓLOGA 1 = Pgh 1 ou *pfmdr-1*



**RESISTÊNCIA** = mutação genética da proteína  
seguida de seleção das espécies mutantes

# Efeitos tóxicos associados à alguns antimaláricos quinolínicos

## EFEITOS COLATERAIS

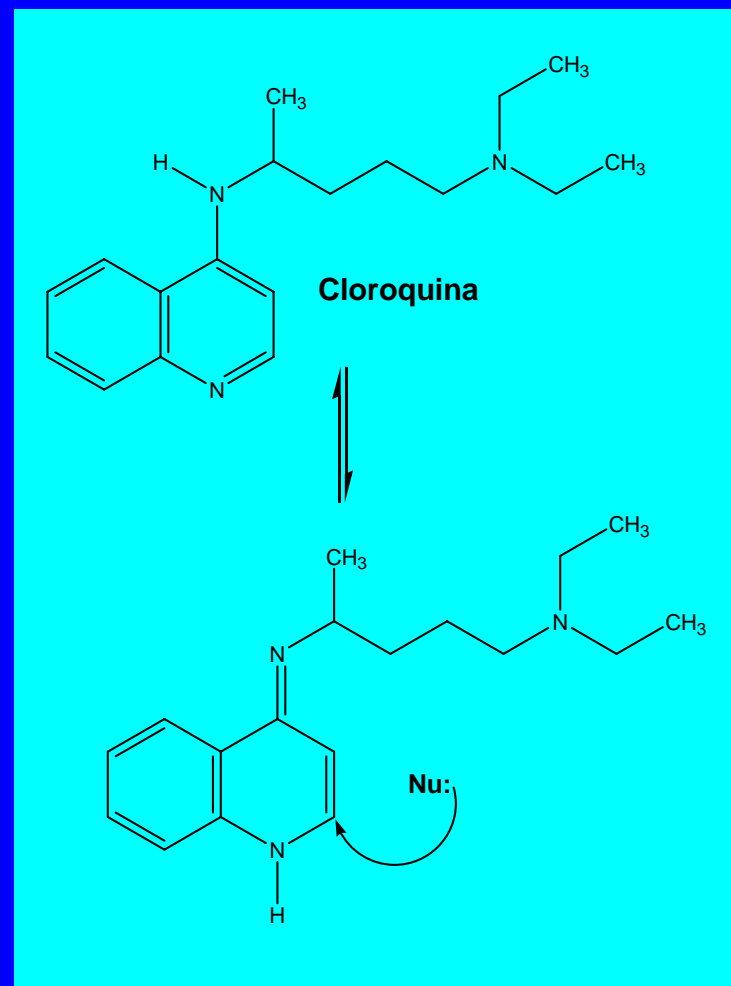
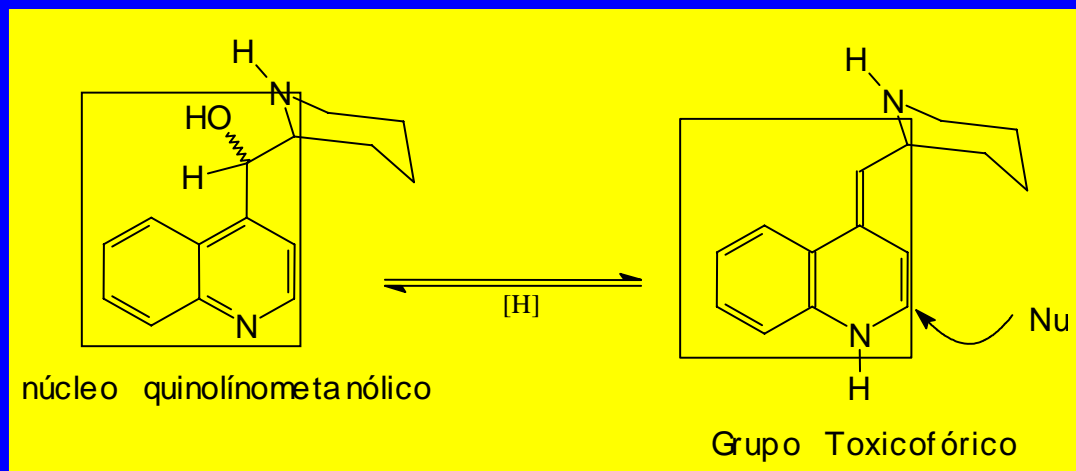
Neurotoxicidade

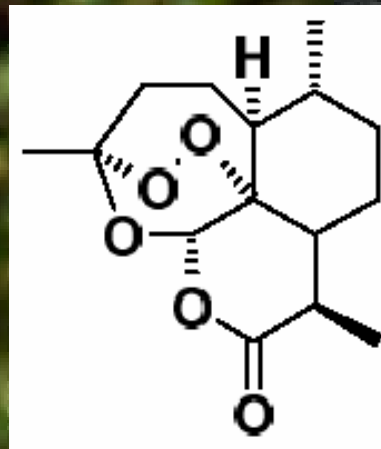
Arritmias

Febre

Depressão respiratória

Cegueira





*Artemisia annua*

“Qinghao”

Artemisinina (Qinghaosu)

Isolamento: 0,3-0,5 % (1%)

$IC_{50} = 1 \text{ ng/mL}$

168 AC: Primeira referência de uso como medicamento. Tumba de Mawangdui Han  
340 DC: Ge Hong preconiza seu uso no tratamento da febre  
Li Shizen: Compêndio de Matéria Médica (Bencao Gangmu) → Malária

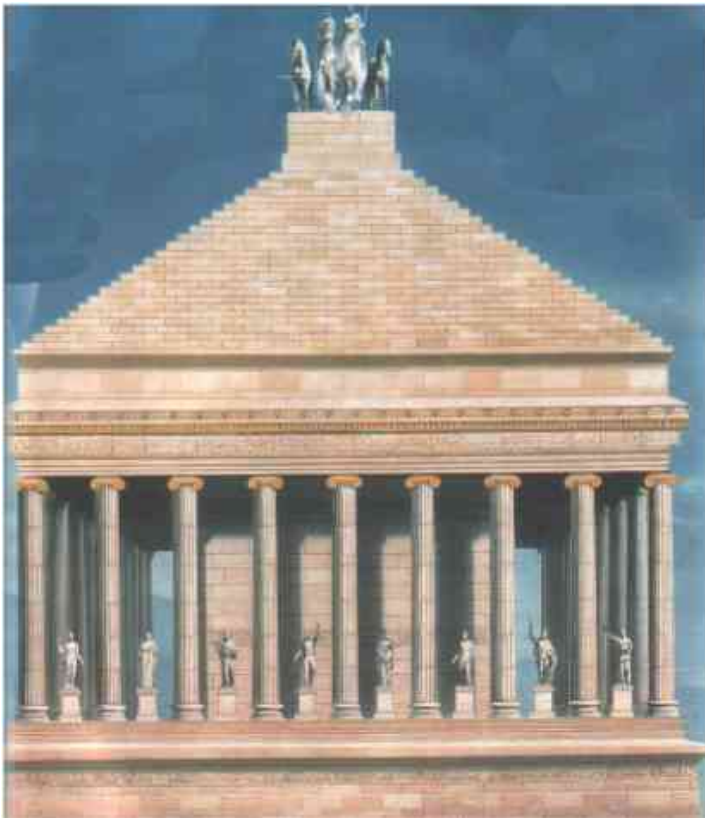


## > Mausolo

Nacionalidade: Persa

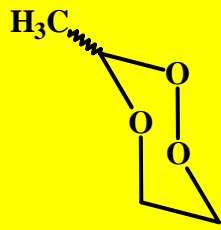
- 352 a.C.

A la muerte del sátrapa persa de Caria Mausolo, su esposa Artemisia mandó construir una sepultura en Halicarnaso, considerada una de las maravillas del mundo. Desde ese momento las sepulturas monumentales reciben el nombre de mausoleos. ([www.artehistoria.com](http://www.artehistoria.com))

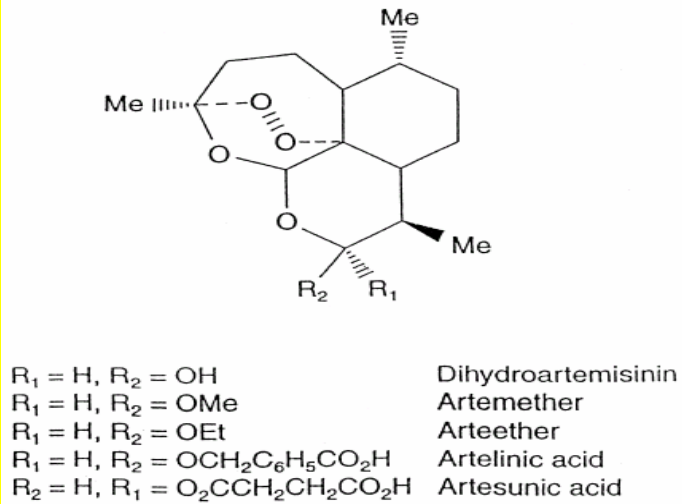




# Análogos da Artemisinina



Inativo



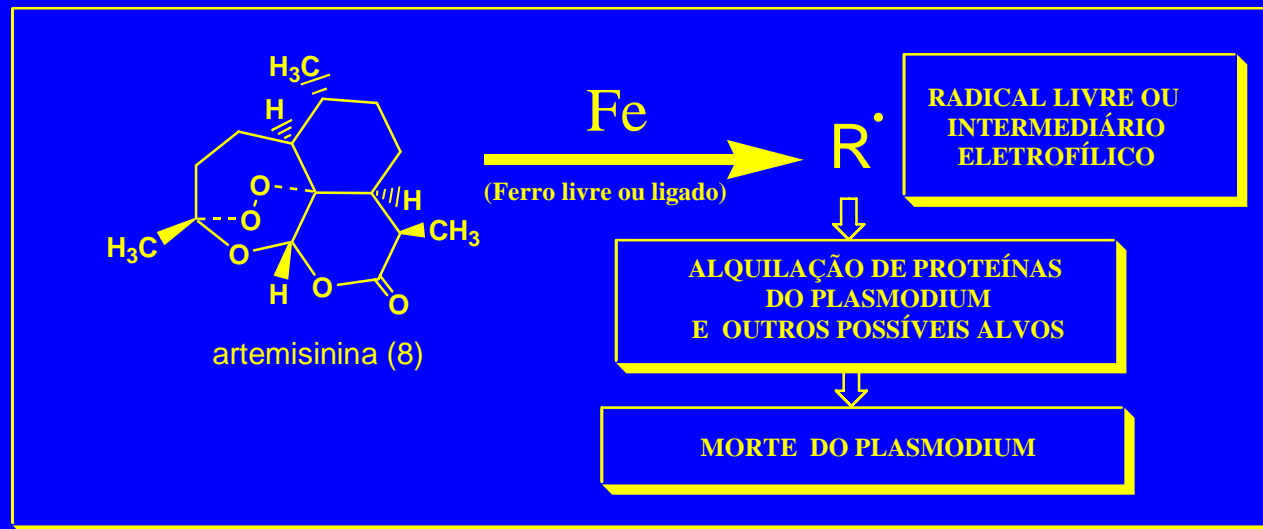
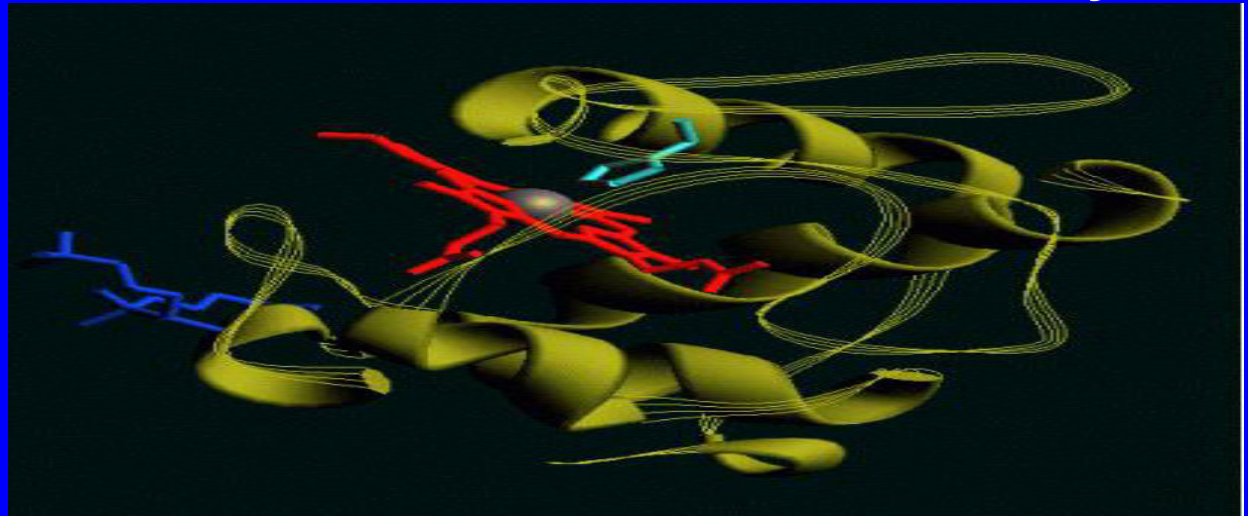
Cpd	R	R'	Activity, IC <sub>50</sub> ng/ml	
Dihydroartemisinin	OH,H	H, OH	0.51	0.25
β-Arteether	OEt	H	0.49	0.54
	H	OEt	0.51	0.93
β-Artemether	OMe	H	0.40	0.50
	H	OMe	0.36	0.55
Sodium artesunate	H	O <sub>2</sub> CCH <sub>2</sub> CH <sub>2</sub> CO <sub>2</sub> -Na <sup>+</sup>	1.66	2.18
Artelinic acid	OCH <sub>2</sub> C <sub>6</sub> H <sub>5</sub> CO <sub>2</sub> H	H	1.38	4.07

# Análise Farmacocinética Comparativa (Análogos da Artemisinina)

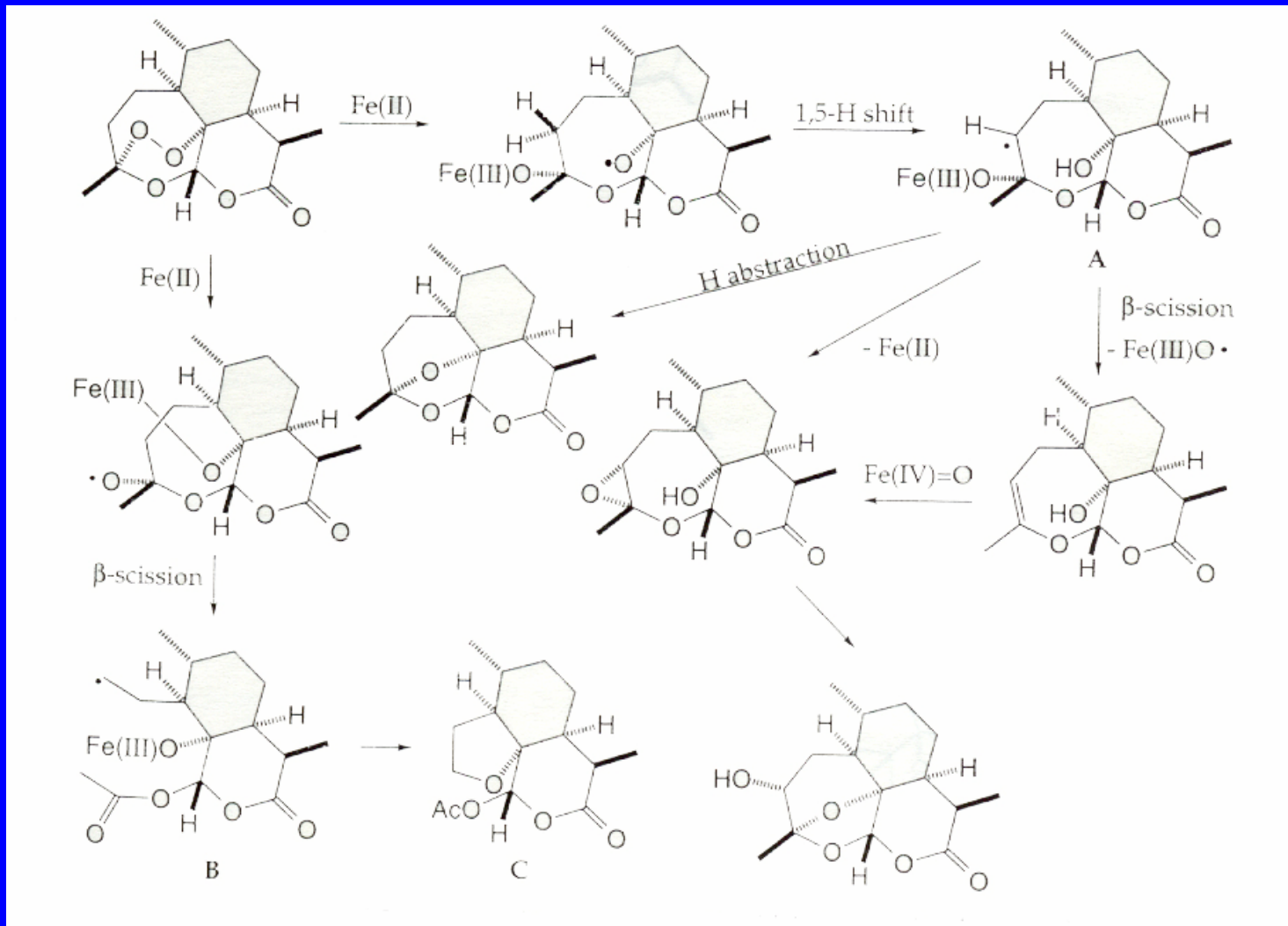
	Absorption	Bioavailability (%)	Protein Binding	First-pass Effect	Oral (t <sub>1/2</sub> )	Intravenous (t <sub>1/2</sub> )	intramuscular (t <sub>1/2</sub> )
<b>Artemisinin</b>	Rapid & incomplete	8-10	80 (%)	Extensive	2-5h		
<b>Artemether</b>	Rapid & incomplete	54		Extensive	3-11h		8-11 h
<b>Arteether</b>		34			> 20h		23h
<b>Artesunate</b>	Rapid & incomplete	82		Extensive	< 1h	45 min	
<b>Dihydro-artemisinin</b>		85			3.1h		

# Fármacos que atuam na fase eritrocítica do ciclo do *P.falciparum*

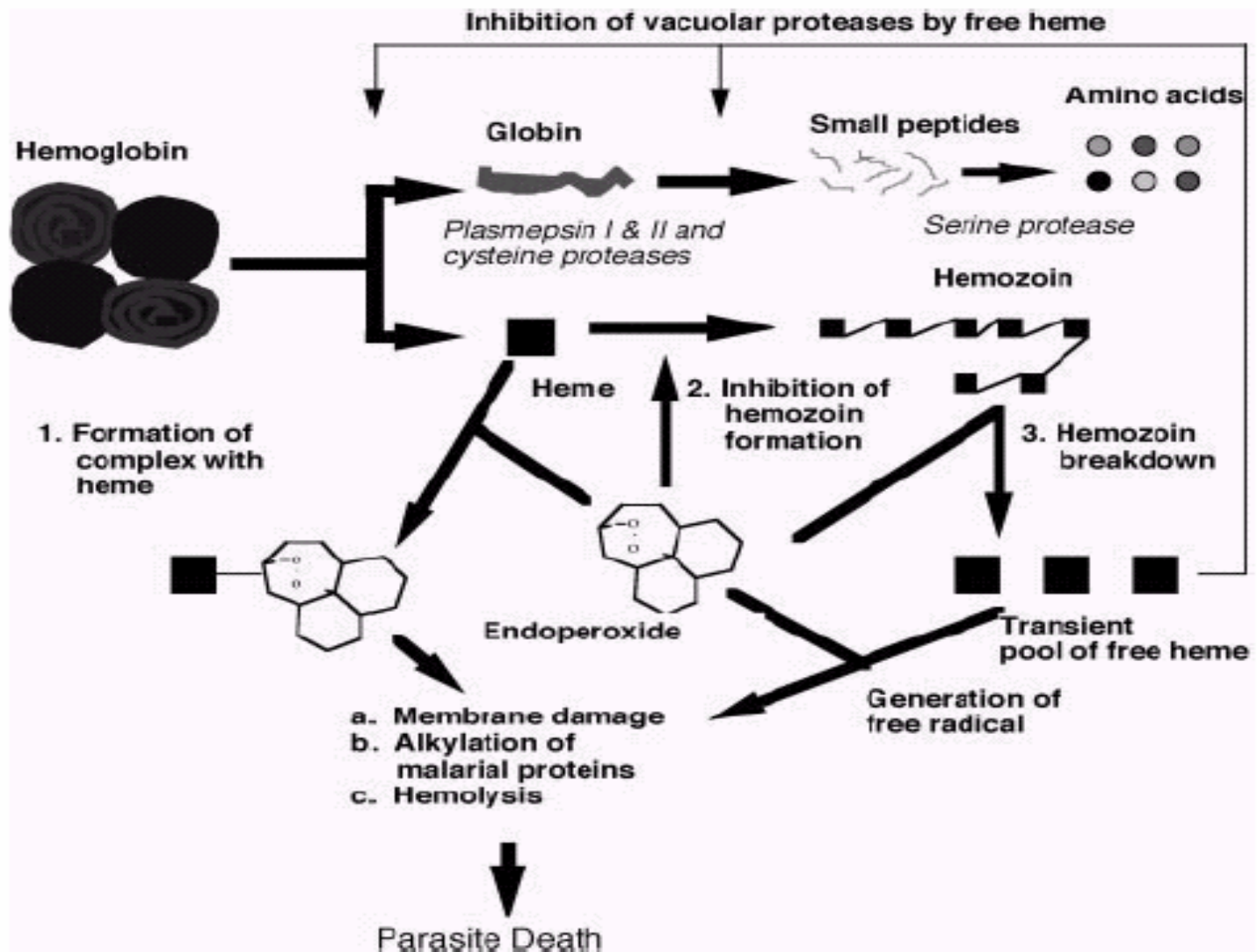
## Derivados artemisinínicos e seu mecanismo de ação



# Mecanismo de Ação da Artemisinina (Inibição de CYP)



# Mecanismo de Ação da Artemisinina





# Quimioterapia Moderna



**Prêmio Nobel  
Fisiologia/Medicina  
(1908)**

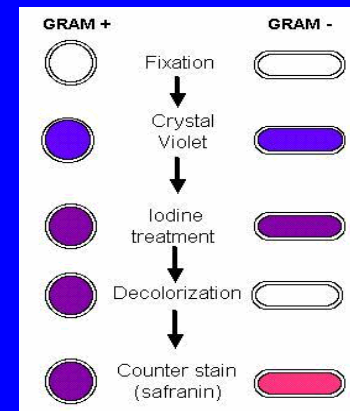
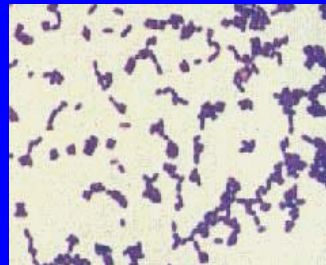
P. Ehrlich, 1909

“The discoveries of those uncivilized peoples represented the sum of limitless testing of thousands of natural materials. By contrast with their selection of medicines by pure chance, we have to find first certain compounds, for example some arsenic derivatives, which show at least a low degree of therapeutic effect. Once this is done through more or less laborious tests, the purely empirical screening is replaced by preparing chemical variations, homologs and other derivatives whose efficacy has to be tested. But even at best chemical drugs are not magic bullets, and will not always hit only the center of the target, that is the disease-causing organisms. Moreover, nothing is as simple as to ascertain the lethal or the maximal well-tolerated dose, and the curative dose in a given animal species. In humans the determination of dosages is infinitely more difficult as one has to start with low doses and increase them gradually until they become therapeutically active. This is further aggravated by the occurrence of congenital or acquired idiosyncrasies from most medicines... and it cannot justly be demanded that a decision be made within a few months as to the merits or demerits of such new agents.”

"Magic Bullets"  
"Magic Bullets"

# A Evolução da Quimioterapia

- Confluência da Microbiologia: Teoria da Infecção (Koch, Pasteur); Desenvolvimento da Indústria Química de Corantes na Alemanha;
- Imunologia começa a ser vista como um tipo de “quimioterapia”
- Desenvolvimento de testes diagnósticos com corantes específicos para microorganismos;
- Teste de substâncias de uso diagnóstico (corantes) como Antibióticos (Trypanosoma sp. / Febre do Sono)



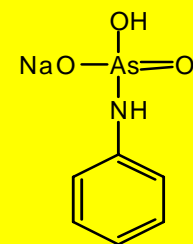
**Gram**

# Ehrlich concebeu e reconheceu muitos dos elementos-chaves da descoberta de fármacos

- Reconhecimento do uso de substâncias químicas na terapia;
- Química Sintética como ferramenta para se obter muitos derivados congêneres

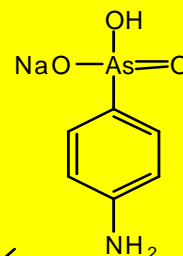
- Metabolismo: Conceito de Metabólito Ativo;
- Resistência dos microorganismos: Hereditariedade

Arsenic Acid + Aniline



Reported Structure  
Atoxyl

Atividade  
Antitripanossomal  
in Ratos

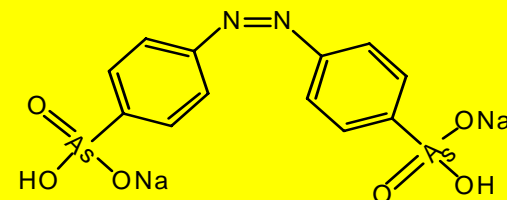
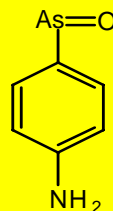


Actual Structure

Reduction

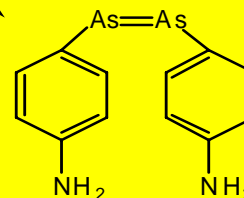
Diazotization

In Vivo Metabolism



Azo-Dyes

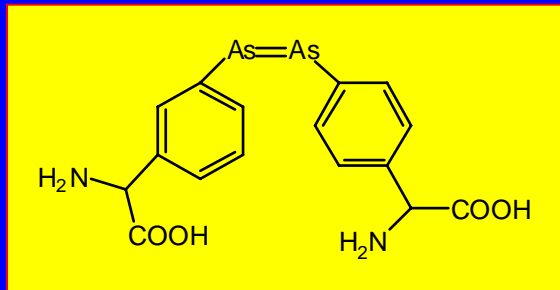
Reduction



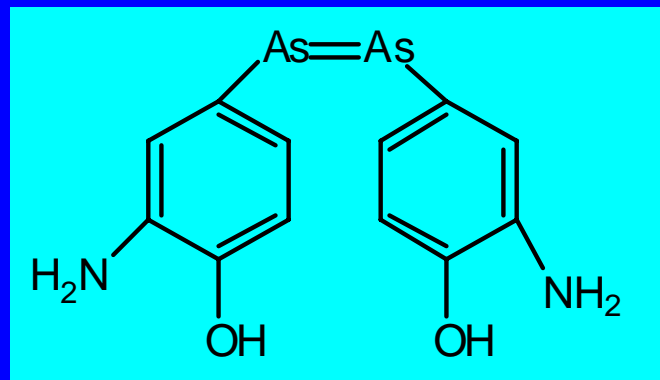
Substituted Anilines and other Chemical Derivatizations

# Arsephenamine (Composto 606)

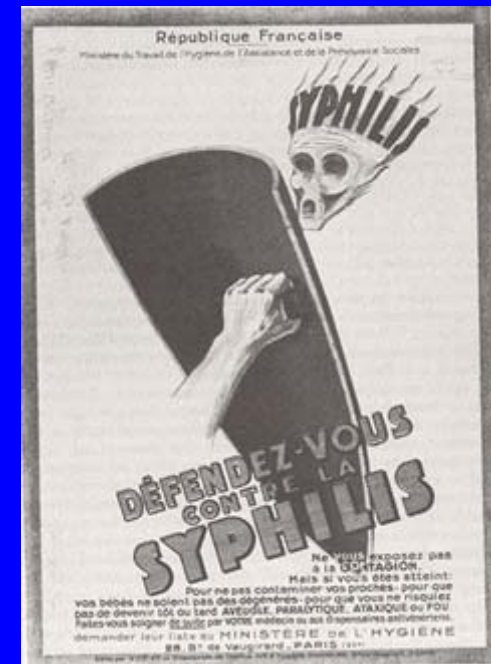
- ◆ Mais de 300 compostos foram inicialmente testados contra tripanossomas mas a cepa utilizada foi resistente a muitos deles.
- ◆ Então, um novo pós-doutor ingressou no grupo trazendo a expertise no modelo de espiroquetas causadoras da Sífilis em ratos. Os mesmos compostos foram re-ensaiados e o composto "606" mostrou uma potente atividade inibitória das espiroquetas.
- ◆
- ◆ Este composto, chamado Salvarsan foi então empregado clinicamente no tratamento da Sífilis por muitos anos.



Composto "418" Anti-trypanosomal  
Ativo em ratos mas não em humanos



Salvarsan: Primeiro Fármaco  
Anti-espiroqueta empregado  
no tratamento da Sífilis





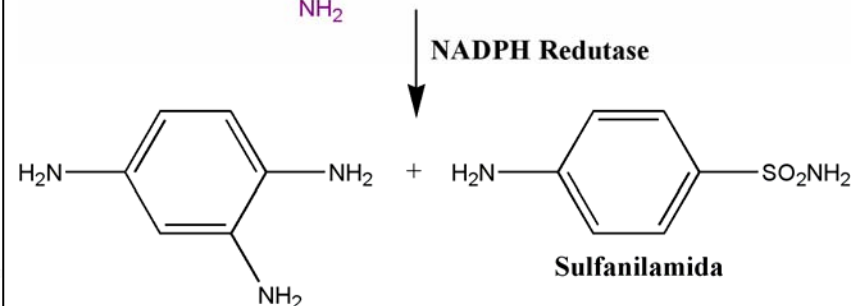
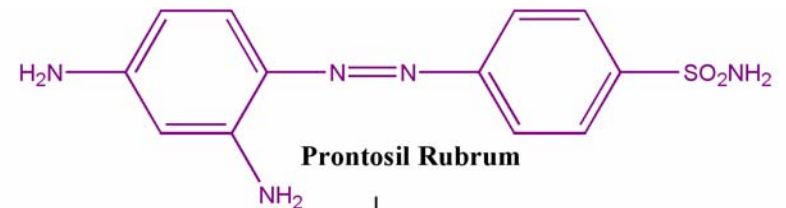
# Sulfas Antibacterianas

- 1935 – Gerhard Domagk
- Nobel Fisiologia 1939
- – Prontosil
- – Sulfa Drugs



*S. aureus*

Gelmo (1908)



Teridina + Paba

Inibido por Sulfonamidas

Acido Dihidropteróico

Glutamato

Acido Dihidrofólico

NADPH

NADP

Inibido por Trimetopim

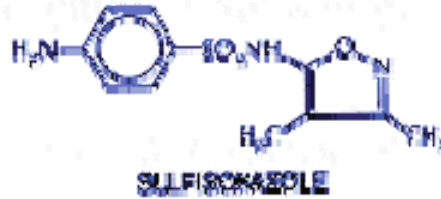
Acido Tetrahidrofólico

Cofactores FAH4

Timidina → DNA

Purinas → RNA DNA

Metionina Glicina t-met-t-RNA → PROTEINAS





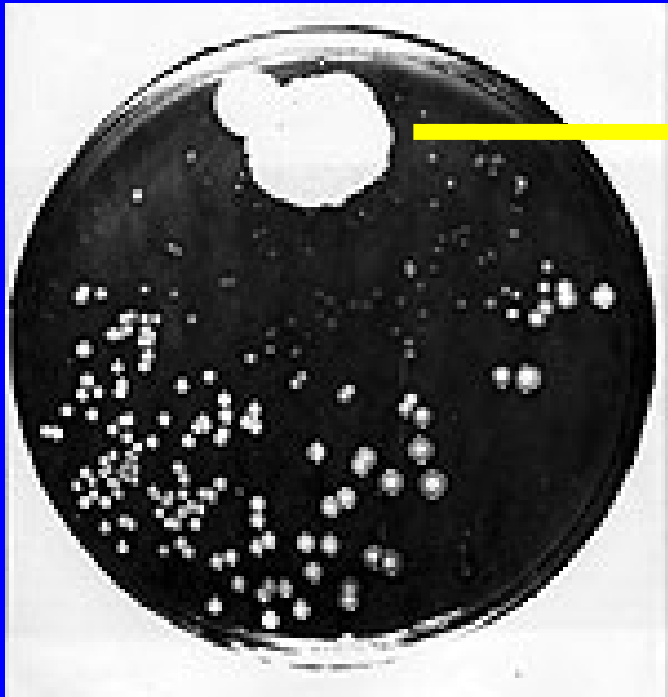
# Contribuições do Acaso (**The Chance Discovery**)



Londres, St. Mary's Hospital, 1928



Alexander Fleming



## “Serendipity”

A word coined by Horace Walpole, who says (Let. to Mann, 28 Jan. 1754) that he had formed it upon the title of the fairy-tale ‘The Three Princes of Serendip’, the heroes of which ‘**were always making discoveries, by accidents and sagacity**, of things they were not in quest of”



"Chance favours the prepared mind..."



H. Florey



E. Chain

1932 - Isolation

1939 – Universidade de Oxford  
(Howard Florey e Ernst Chain)

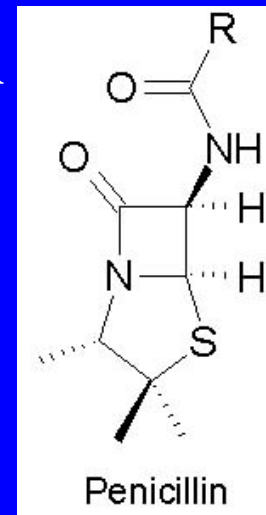
1941- Primeiros ensaios em humanos

1941- Pesquisa foi transferida para os EUA  
(USDA labs) em Peoria, Illinois  
Summer

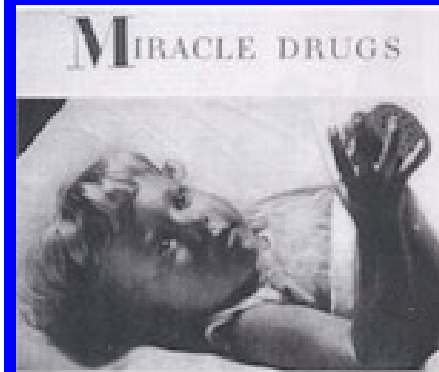
1943 - *Penicillium chrysogenum*

1944 - Caracterização Estrutural

1945 - Prêmio Nobel

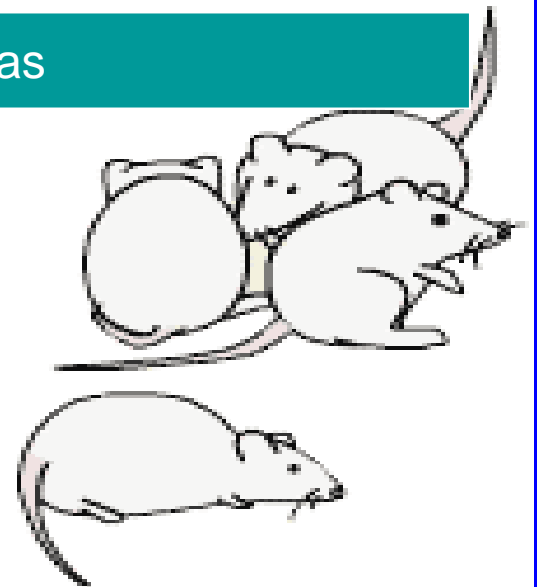
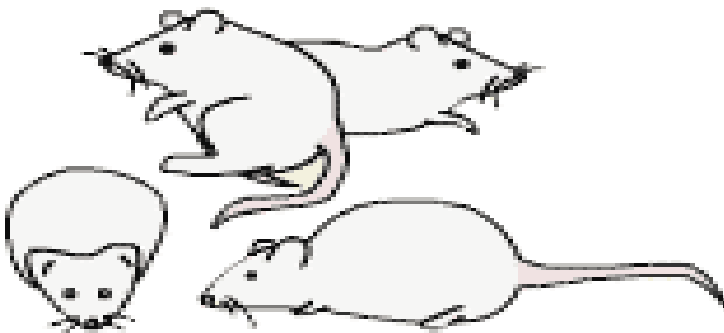


R = Benzila



# Primeiro Teste Terapêutico

Ratos Inoculados com Bactérias



c/ Penicilina

s/ Penicilina

# O Milagre



**Os Ratos Inoculados com Penicilina Sobreviveram!!!!**

# Penicilina ganha o mundo!!!








- Publicação: Florey, Chain, *et. al. Lancet*, 24 de Agosto de 1940;
- Naquele momento a 2ª Guerra Mundial avançava pela Europa, com muitos feridos e inúmeros mortos, como resultado de infecções bacterianas;
- Clamor pela descoberta de Novos Antibacterianos!
- Primeiro Ensaio de Triagem Clínica: Fevereiro de 1941 (Voluntários).



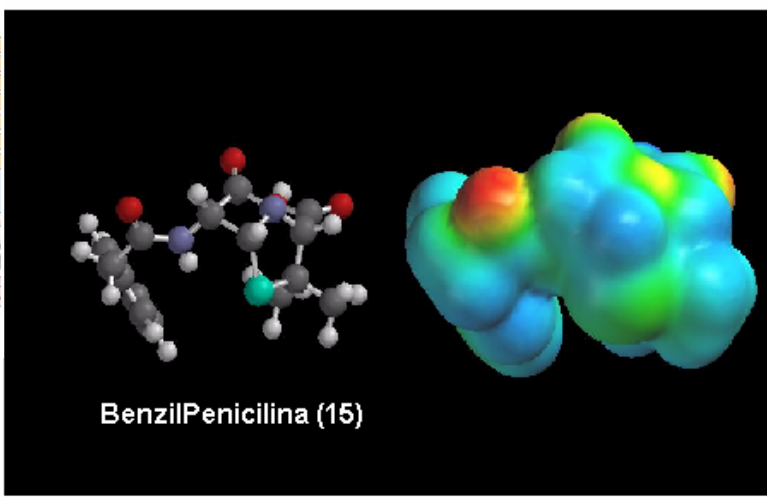
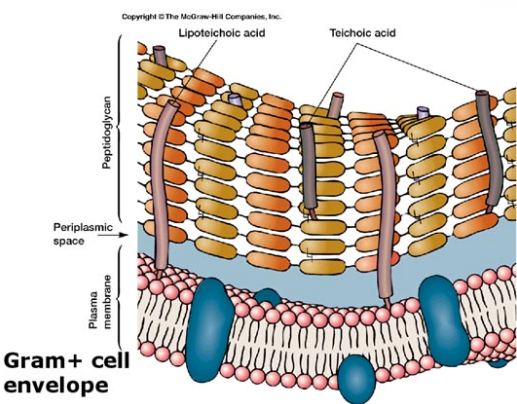
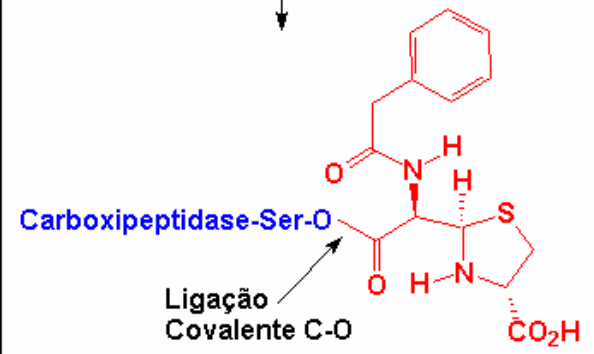
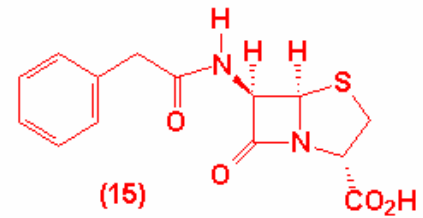
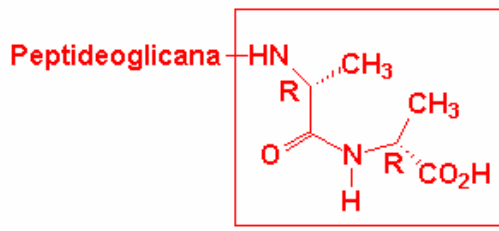
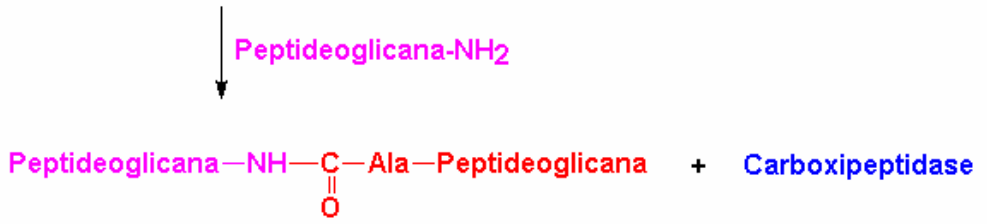
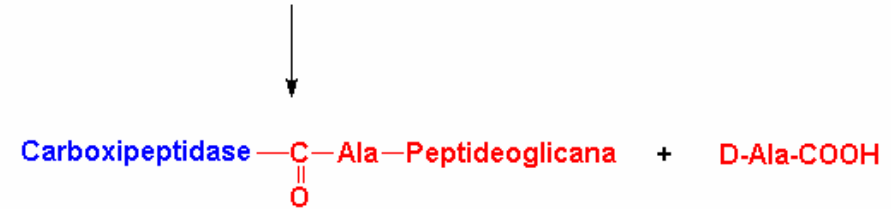
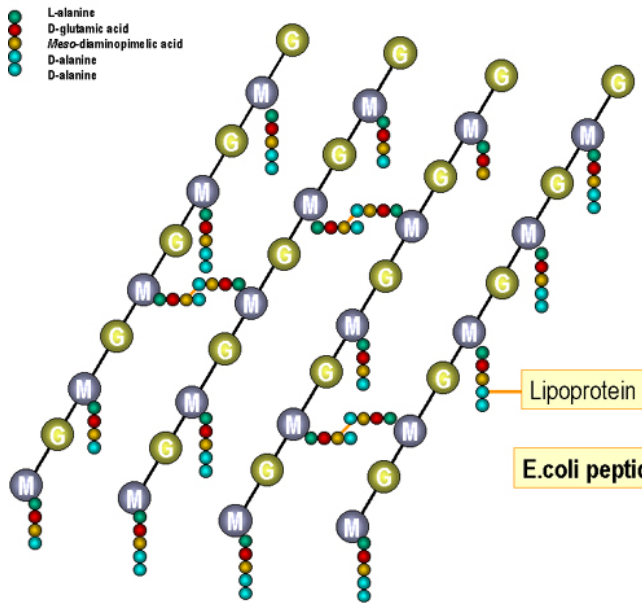


# Proteínas Ligantes de Penicilina

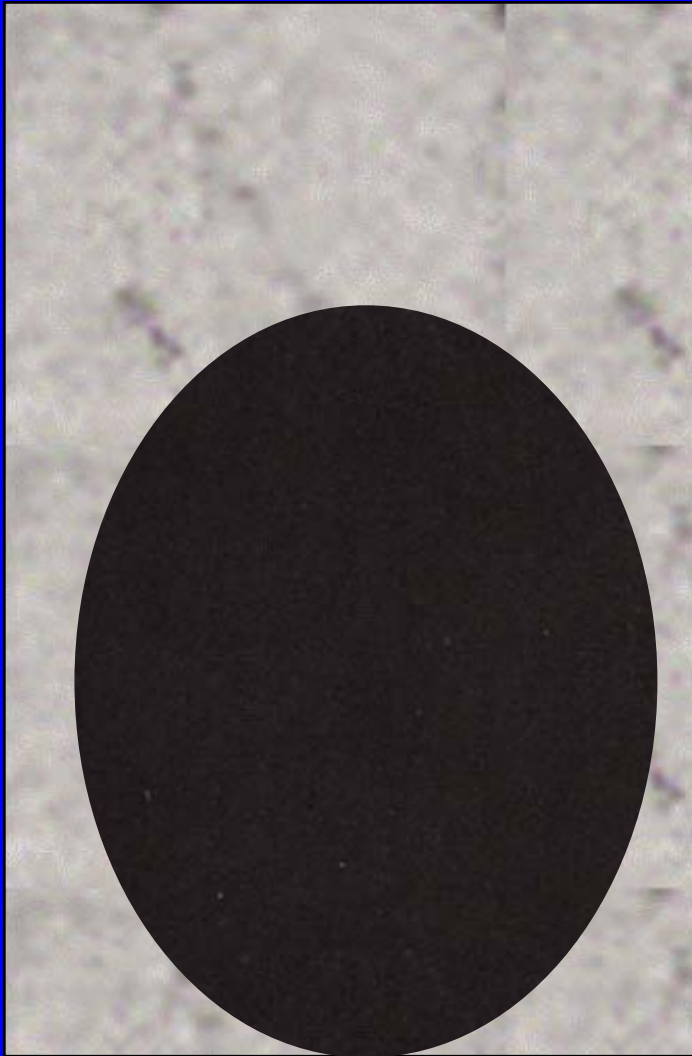
## Identificação por Gel de Eletroforese

Apparent molecular weight	PBP		% of total PBP	Function
91,000	1a		8.1	Transpeptidases involved in peptidoglycan synthesis during elongation
87,000	1b			
66,000	2		0.7	Required for maintenance of "rod" shape
60,000	3		1.9	Required for septum formation
49,000	4		4.0	D-alanine carboxypeptidases
42,000	5		64.7	
40,000	6		20.6	

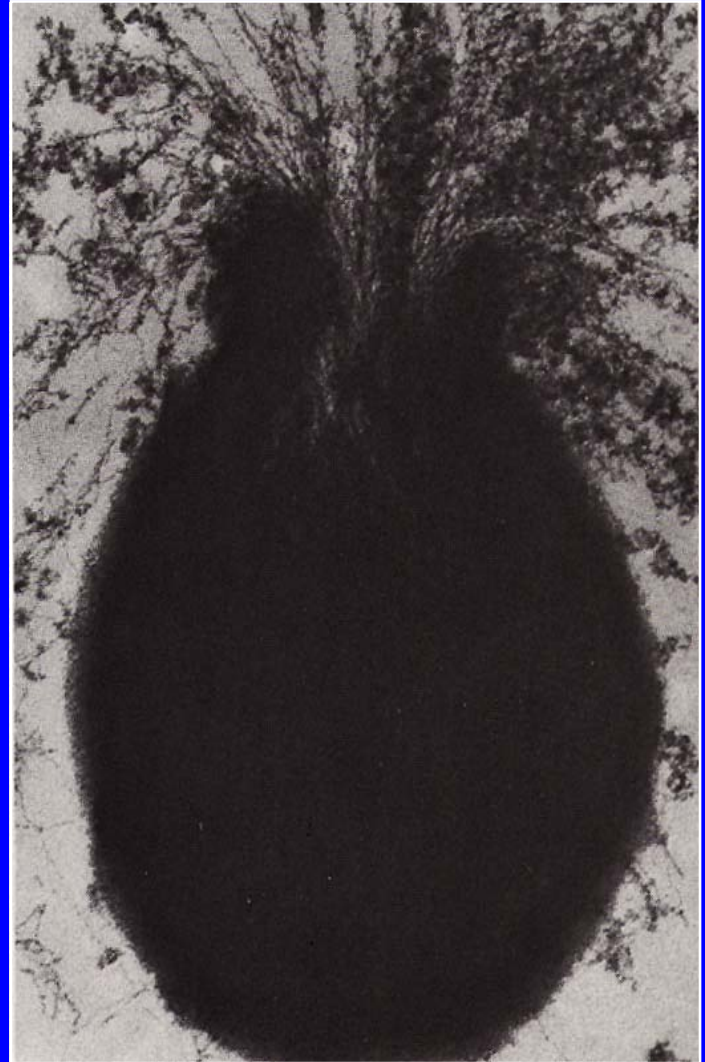
# Mecanismo de Ação das Penicilinas



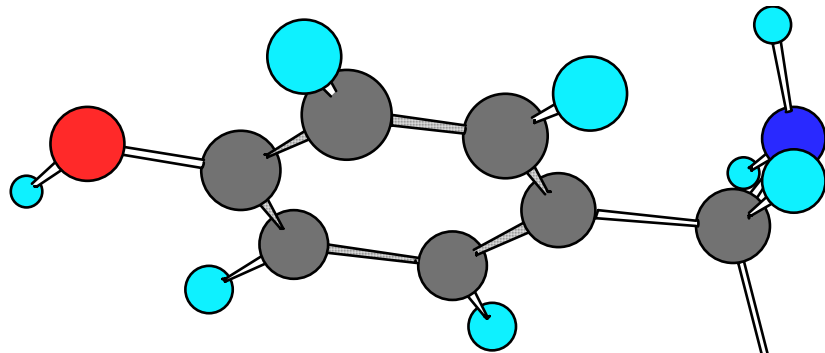
# STAPHYLOCOCCUS



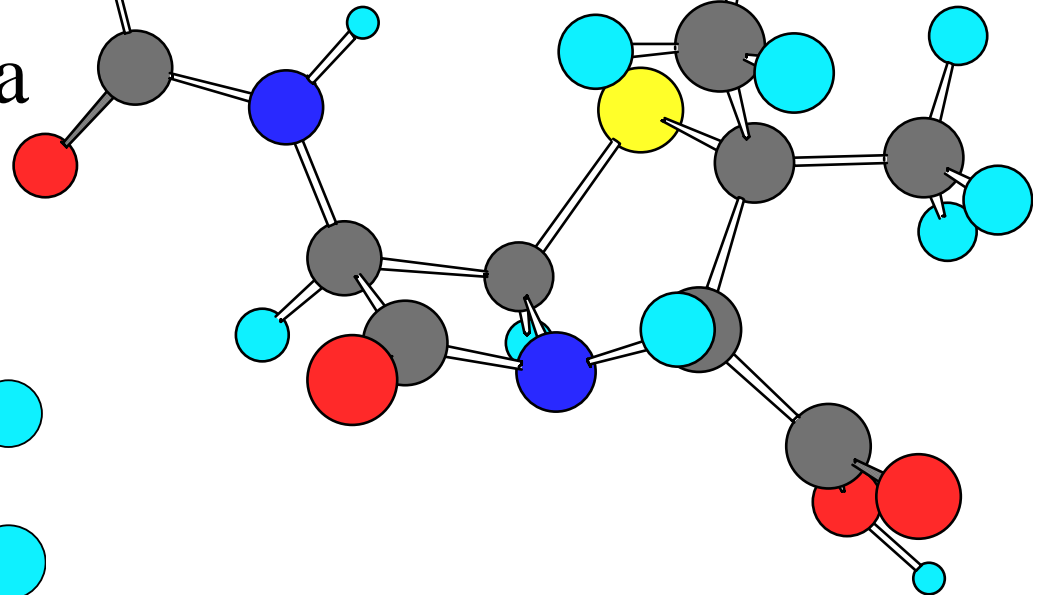
# STAPHYLOCOCCUS TRATADO C/ PENICILINA



# Amoxicilina vs D-Ala-D-Ala

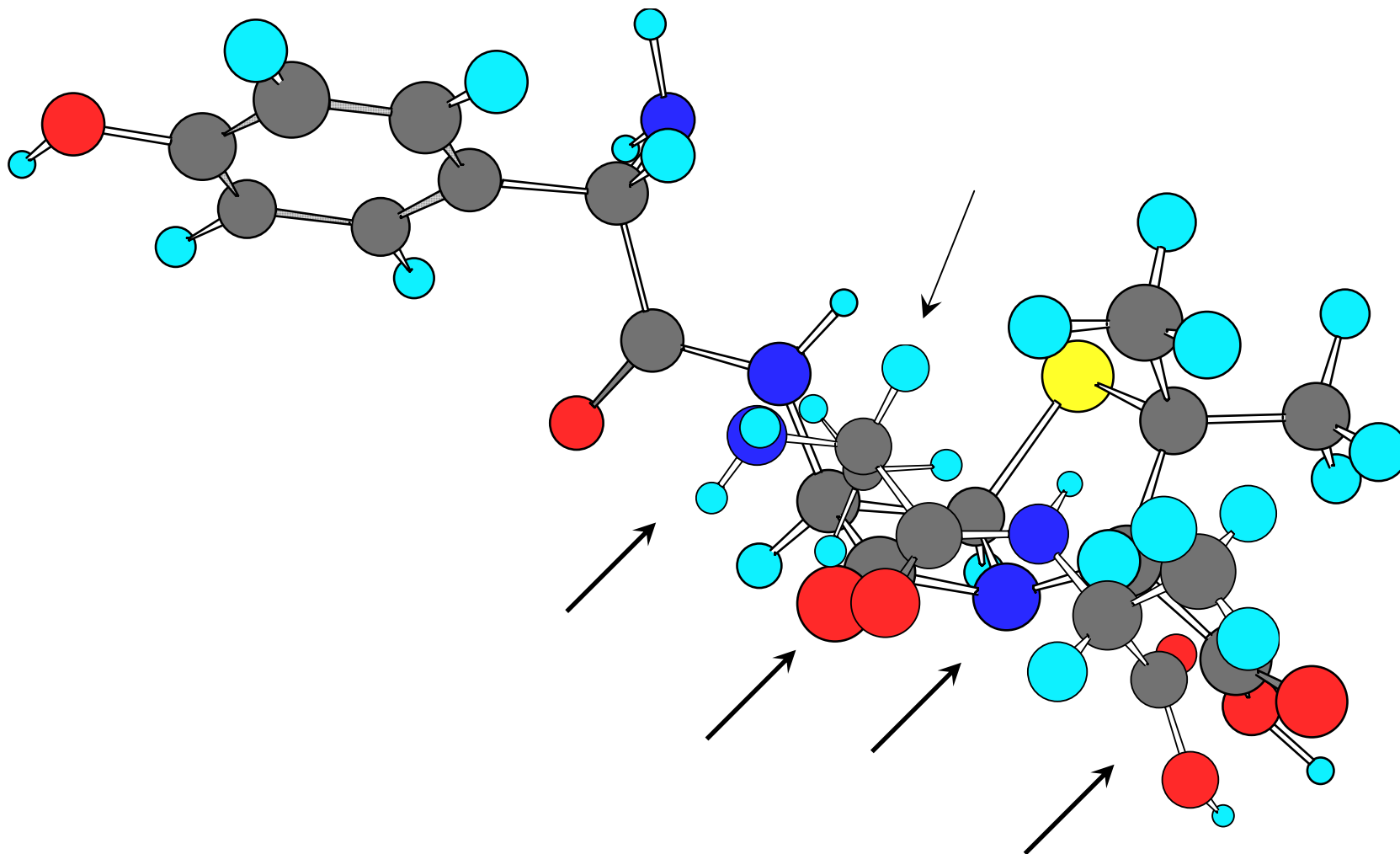


Amoxicilina



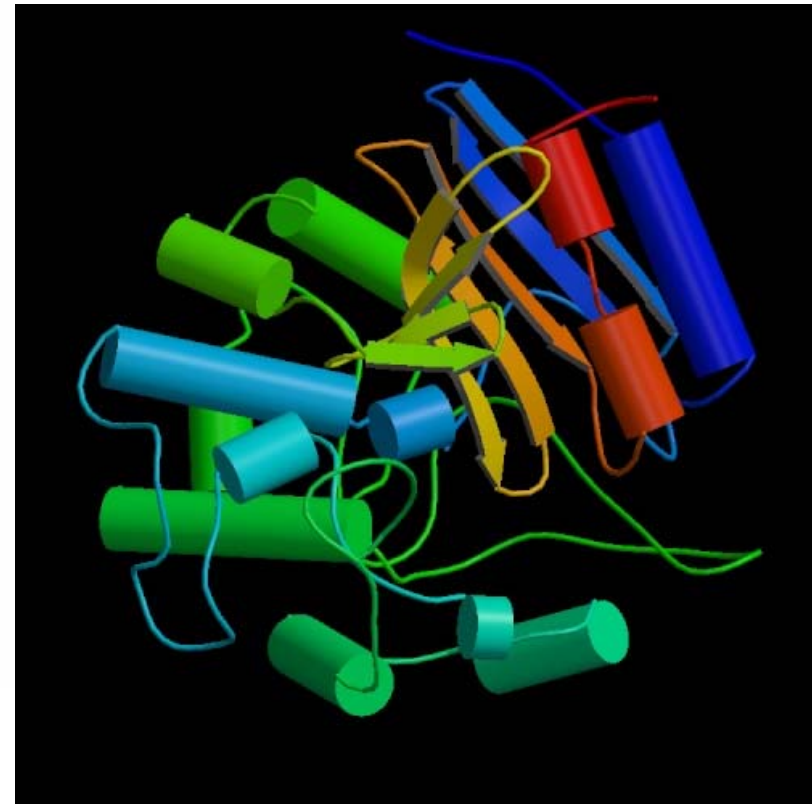
D-Ala-D-Ala

# Sobreposição da Amoxiciclina com D-Ala-D-Ala



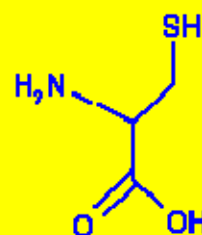
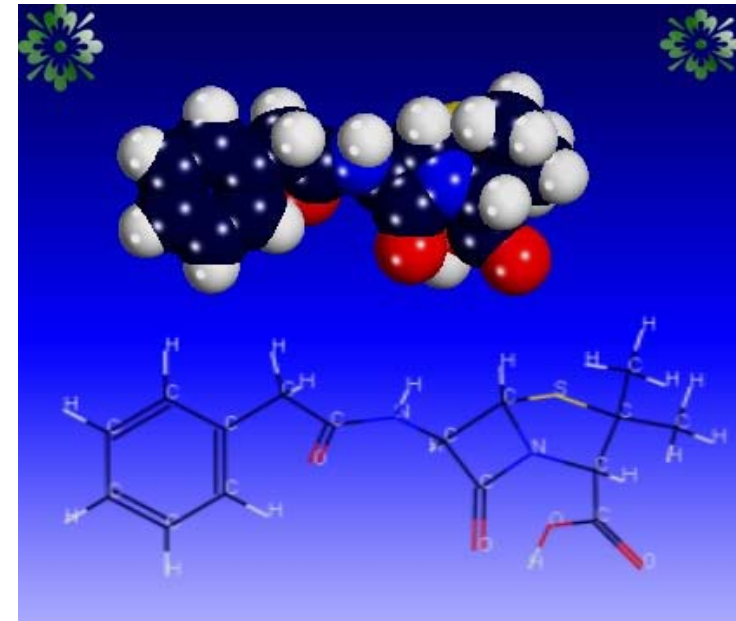
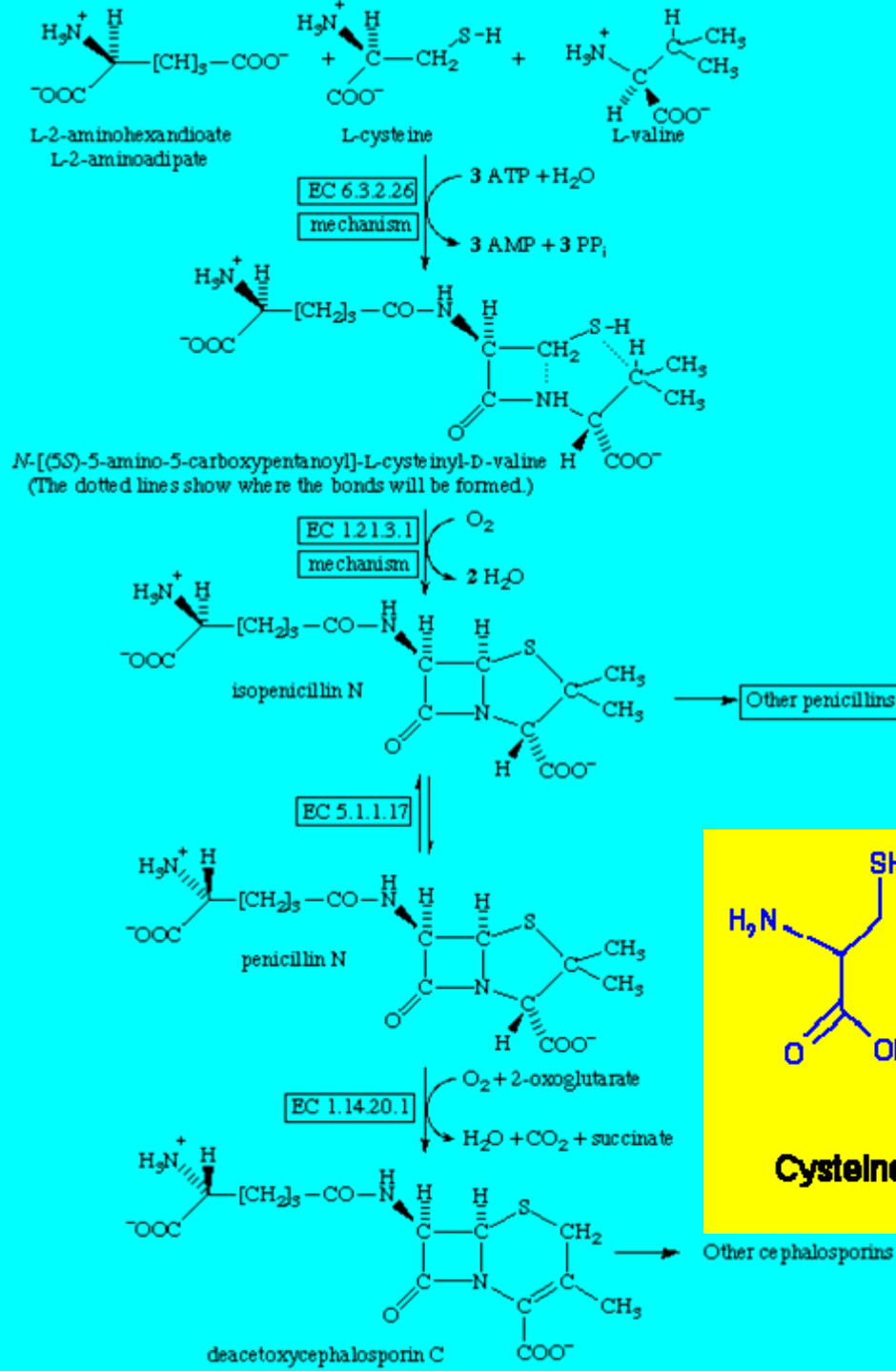
# Estrutura da D-Ala-D-Ala-Carboxipeptidase

PDB: [3pte](#)

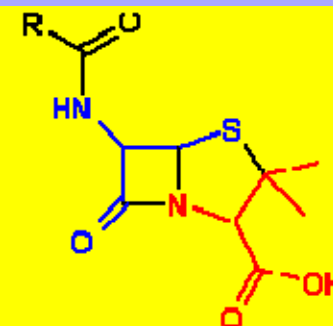




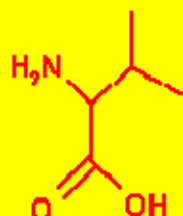
# Bioss ntese das Penicilinas



Cysteine



β-lactam structure

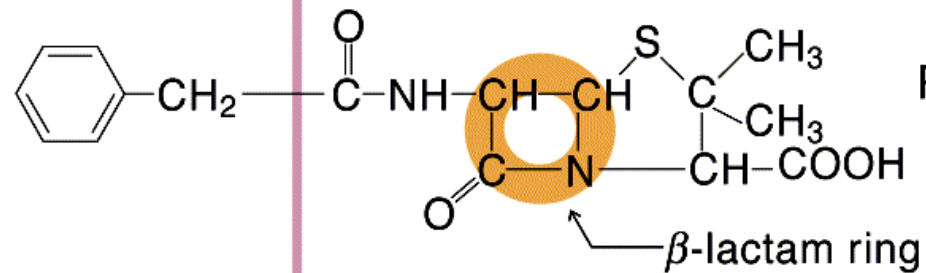


Valine

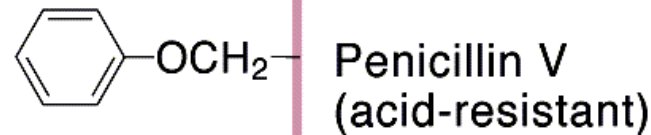
# Família de Penicilinas

## Side Chain

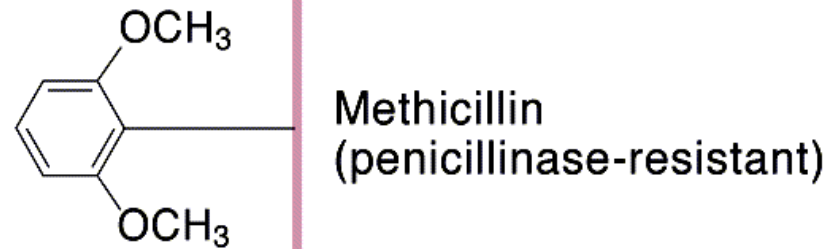
## Basic Structure



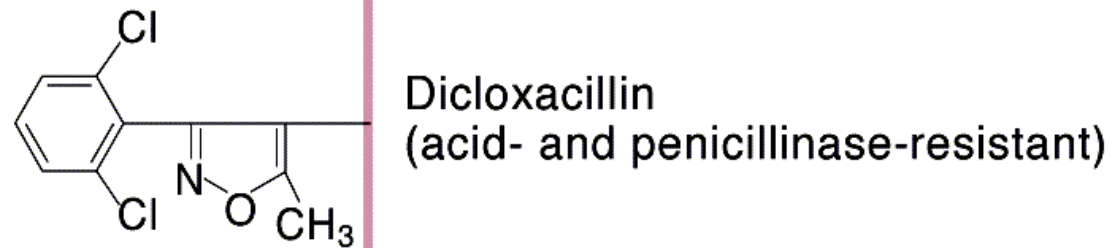
Penicillin G



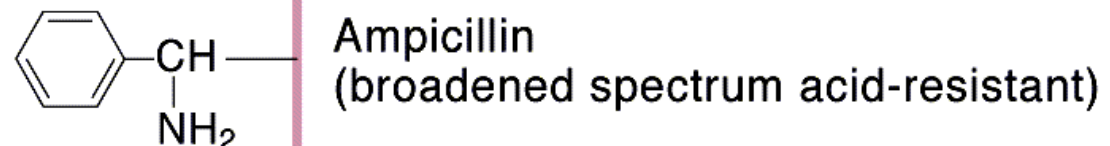
Penicillin V  
(acid-resistant)



Methicillin  
(penicillinase-resistant)



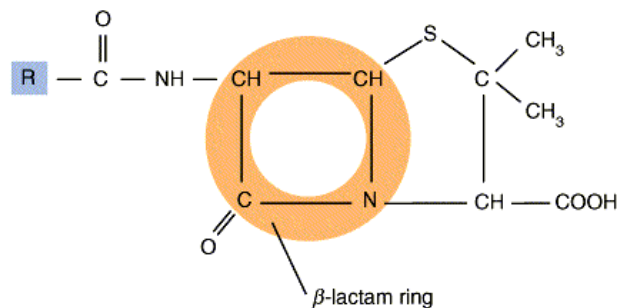
Dicloxacillin  
(acid- and penicillinase-resistant)



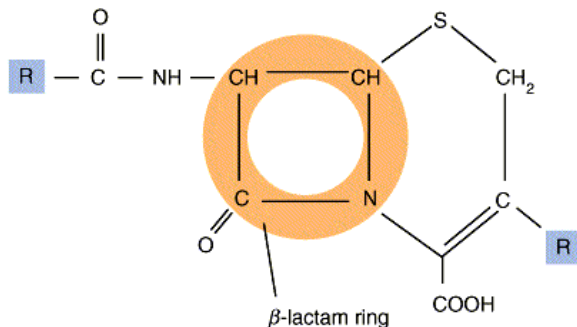
Ampicillin  
(broadened spectrum acid-resistant)

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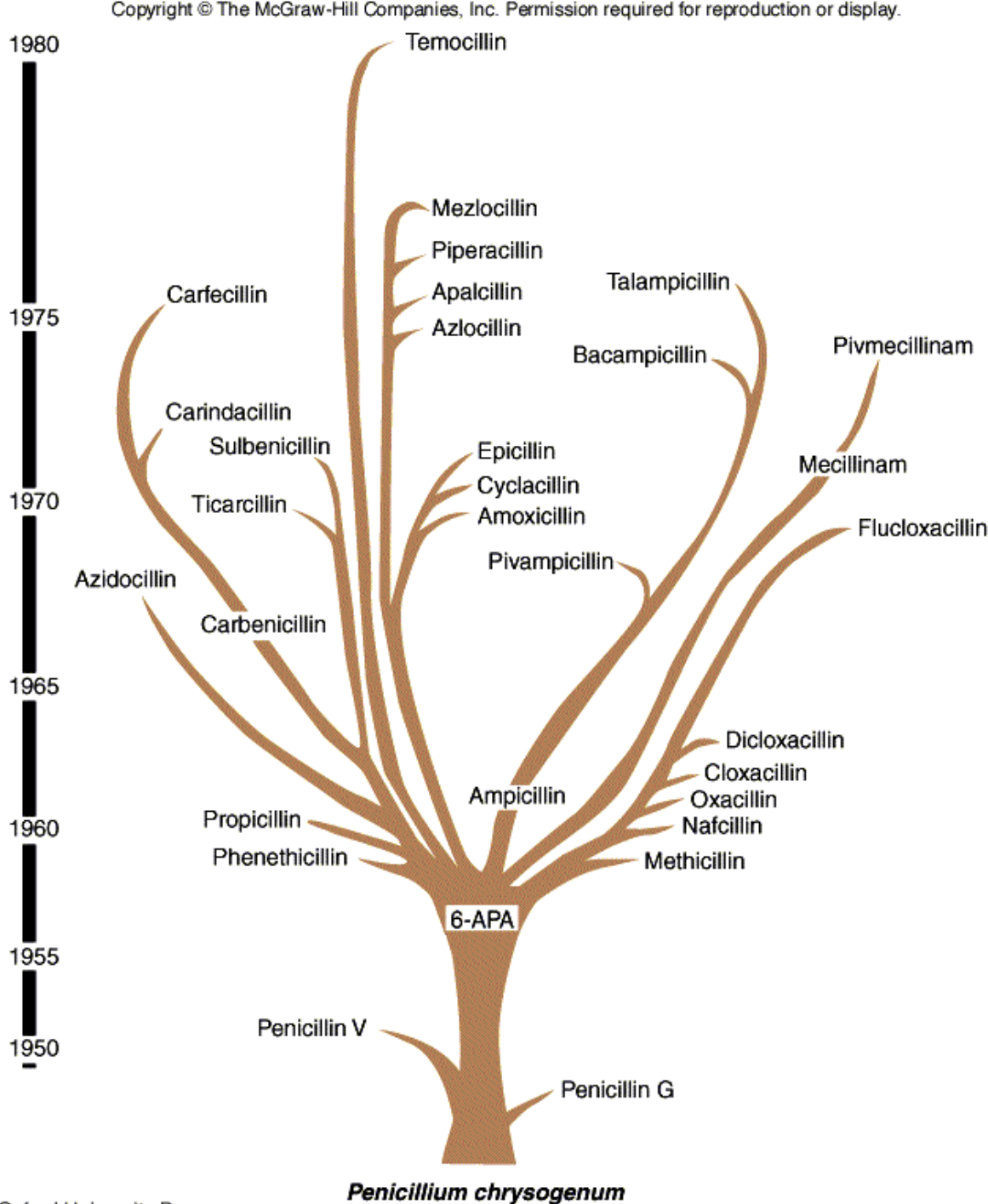
### (a) Penicillin



### (b) Cephalosporin

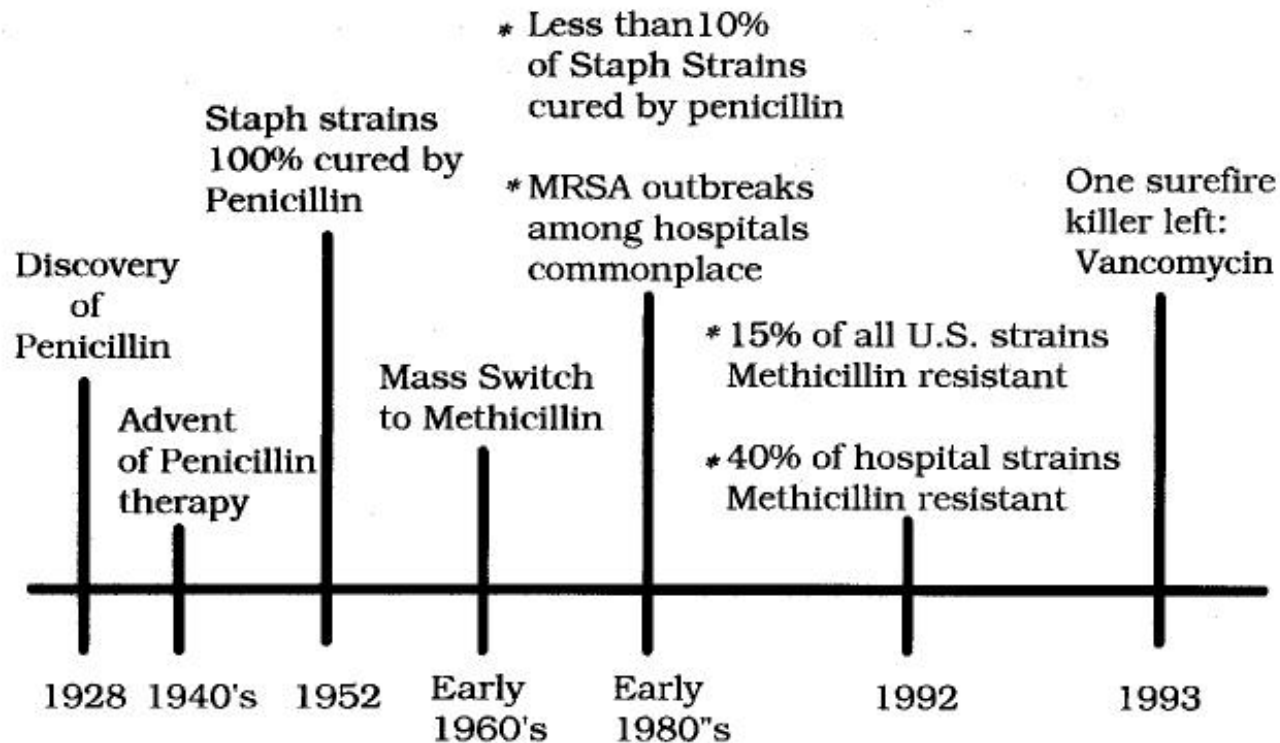


# Evolução Temporal das Penicilinas

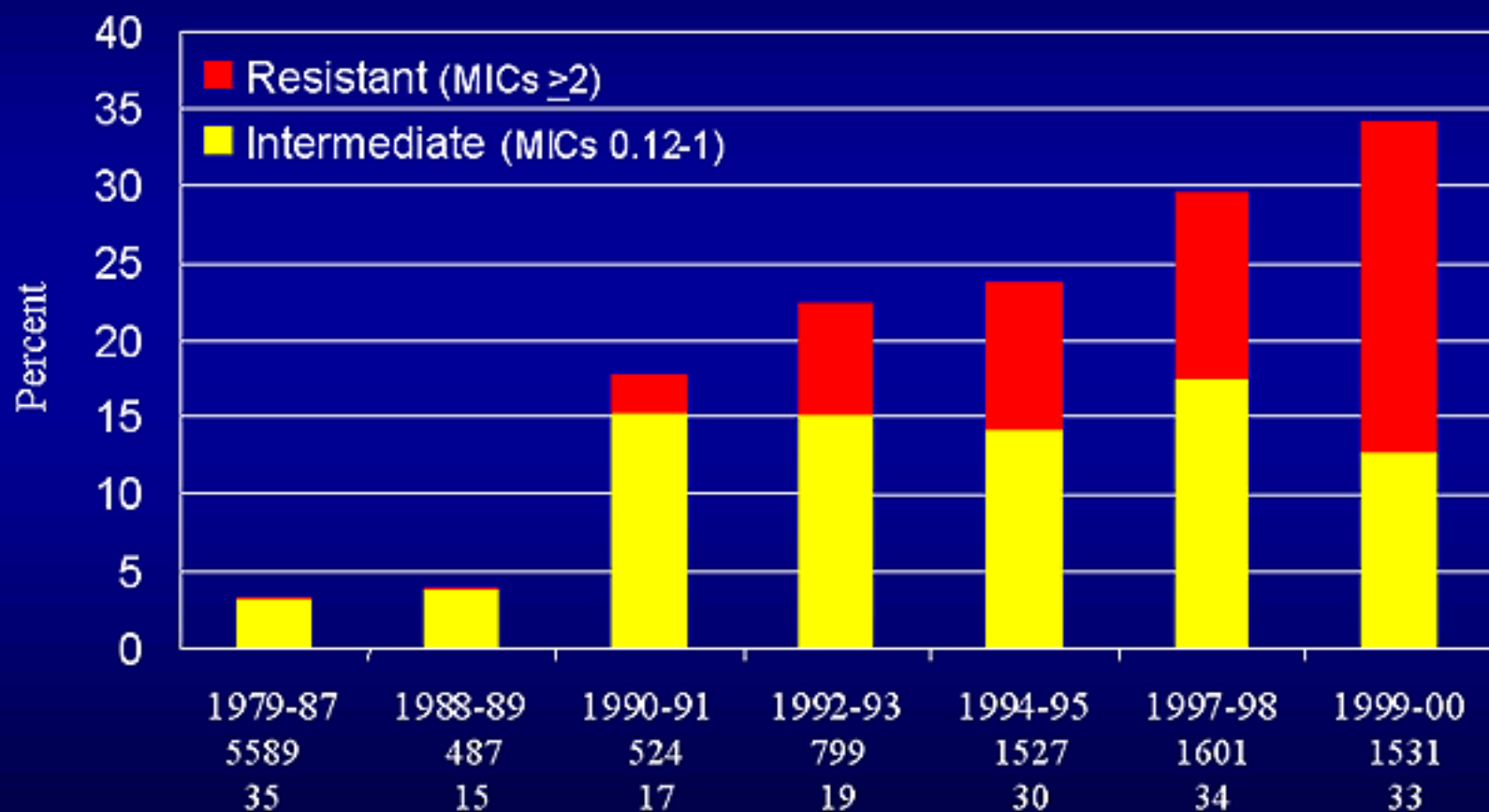


# Resistência às Penicilinas

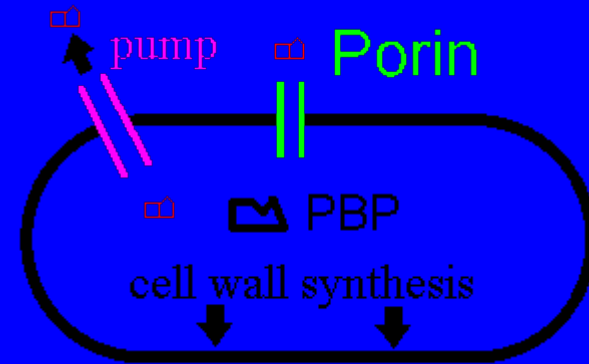
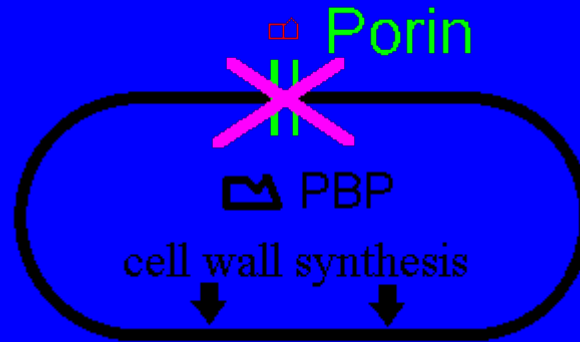
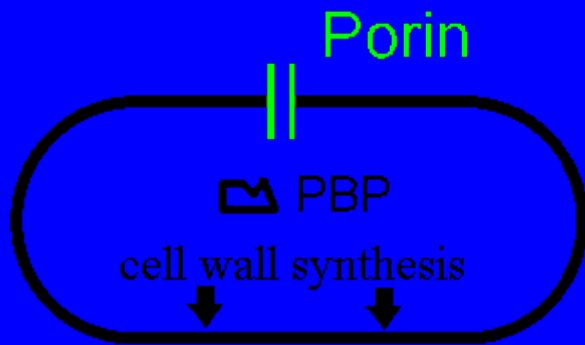
## Emergence of Antibiotic Resistant Staphylococci



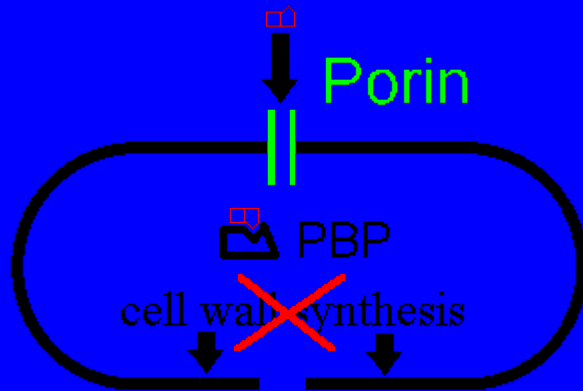
## Penicillin Resistance with *Streptococcus pneumoniae* in the United States



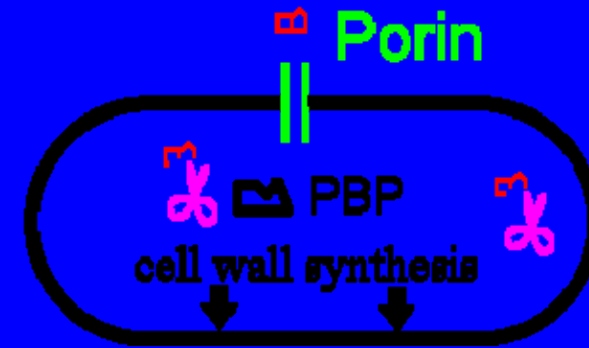
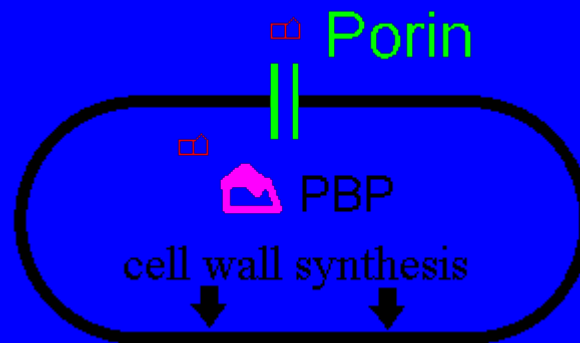
# Mecanismos de Resistência



Beta Lactam



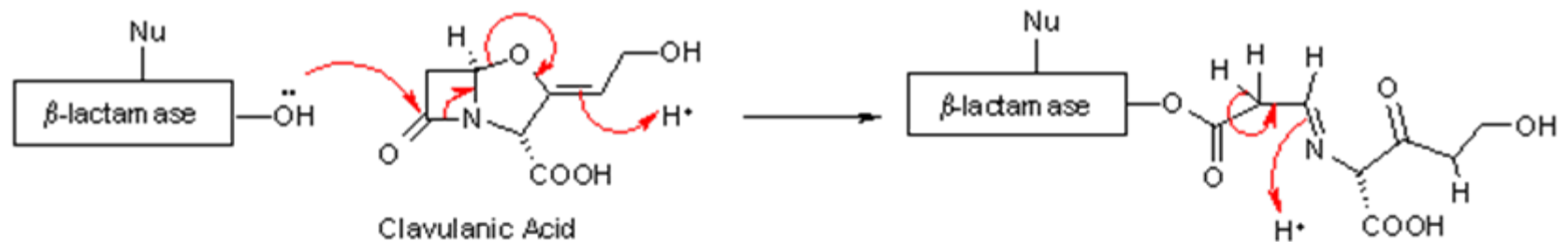
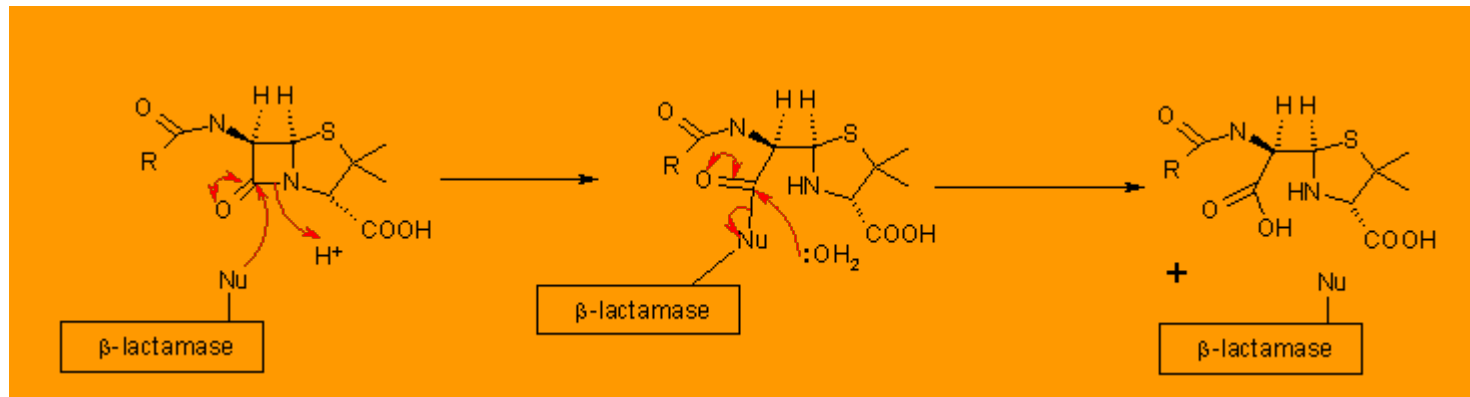
Lysis



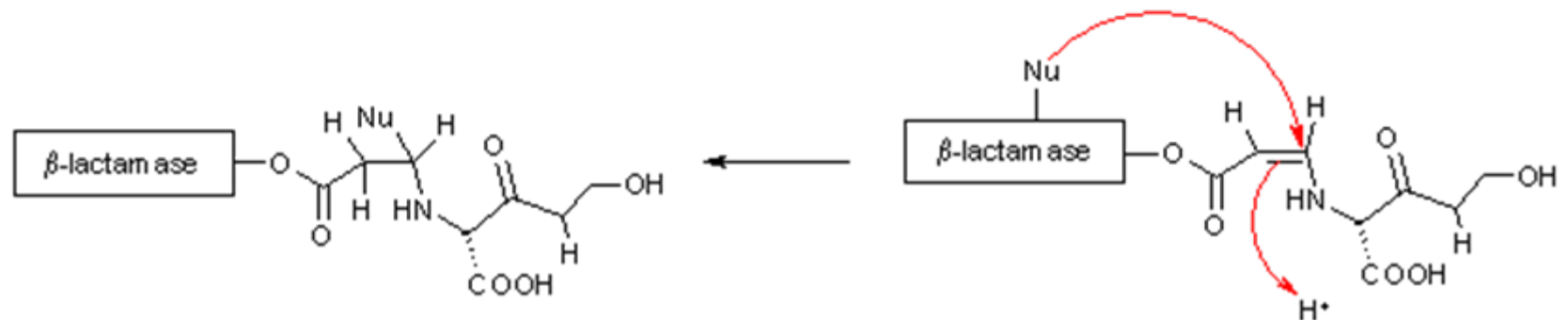
$\beta$ -lactamases



# $\beta$ -Lactamases x Ácido Clavulânico



Inibidor Irreversível



# Antibióticos de Origen Natural

Some clinically important antibiotics			
Antibiotic	Producer organism	Activity	Site or mode of action
Penicillin	<i>Penicillium chrysogenum</i>	Gram-positive bacteria	Wall synthesis
Cephalosporin	<i>Cephalosporium acremonium</i>	Broad spectrum	Wall synthesis
Griseofulvin	<i>Penicillium griseofulvum</i>	Dermatophytic fungi	Microtubules
Bacitracin	<i>Bacillus subtilis</i>	Gram-positive bacteria	Wall synthesis
Polymyxin B	<i>Bacillus polymyxa</i>	Gram-negative bacteria	Cell membrane
Amphotericin B	<i>Streptomyces nodosus</i>	Fungi	Cell membrane
Erythromycin	<i>Streptomyces erythreus</i>	Gram-positive bacteria	Protein synthesis
Neomycin	<i>Streptomyces fradiae</i>	Broad spectrum	Protein synthesis
Streptomycin	<i>Streptomyces griseus</i>	Gram-negative bacteria	Protein synthesis
Tetracycline	<i>Streptomyces rimosus</i>	Broad spectrum	Protein synthesis
Vancomycin	<i>Streptomyces orientalis</i>	Gram-positive bacteria	Protein synthesis
Gentamicin	<i>Micromonospora purpurea</i>	Broad spectrum	Protein synthesis
Rifamycin	<i>Streptomyces mediterranei</i>	Tuberculosis	Protein synthesis

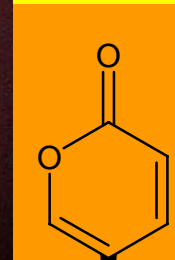
# Digitálicos: *Scilla maritima*

© 1999 Ralf Rebmann

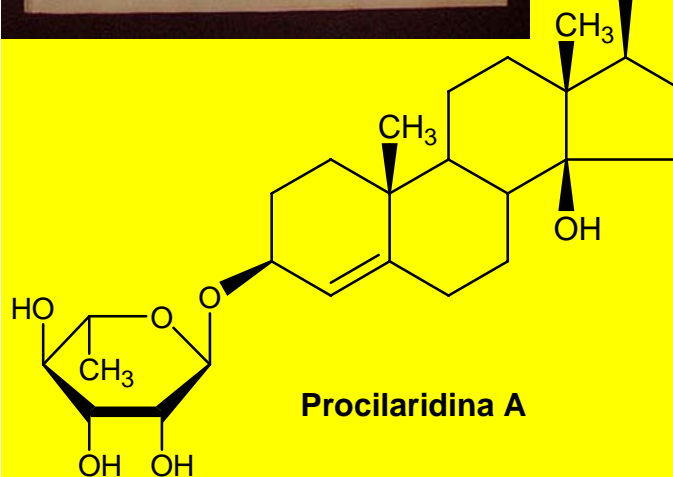


**AC - A cila ficou conhecida como remédio pelos antigos egípcios;**

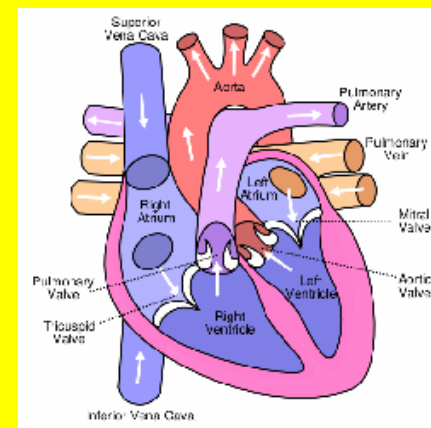
**~1 DC – Os romanos empregaram-na como diurético, tônico cardíaco, emético e veneno para ratos;**



➔ **Bufodienolido**



**Procillaridina A**



# O gênero Digitalis: FOXGLOVES



*Digitalis purpurea*

1250: A digital ou dedaleira foi mencionada nas escrituras dos médicos galeses

1542: Fuchsis descreve botanicamente a digital (Foxglove), e a denominou *Digitais purpurea*;

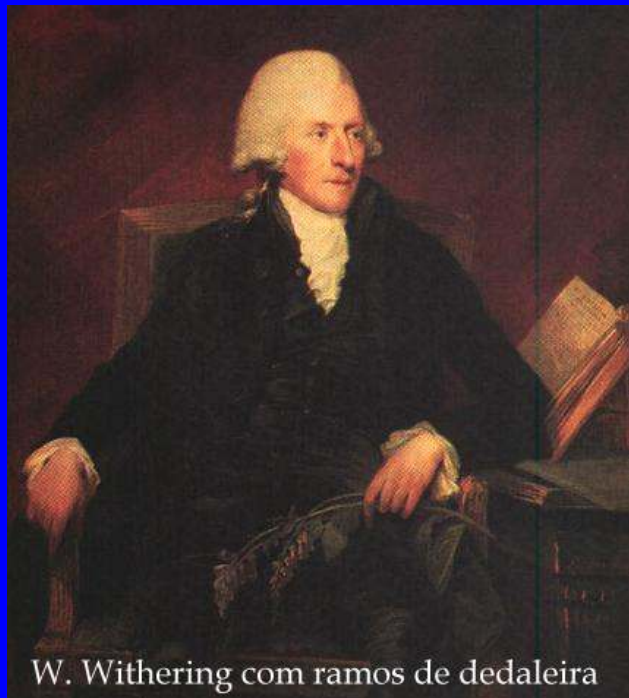
1785: Withering publicou o famoso livro: **“An Account of the Foxglove and Its Medical Uses”**, no qual indicava o emprego da digital no tratamento de condições edematosas;

1250: Ferriar atribuiu uma ação cardíaca as substâncias digitálicas

1835: Homolle preparou pela primeira vez um extrato purificado das folhas de *Digitalis purpurea*;

1872: Frazer atribuiu natureza glicosídica aos princípios ativos dos digitálicos;

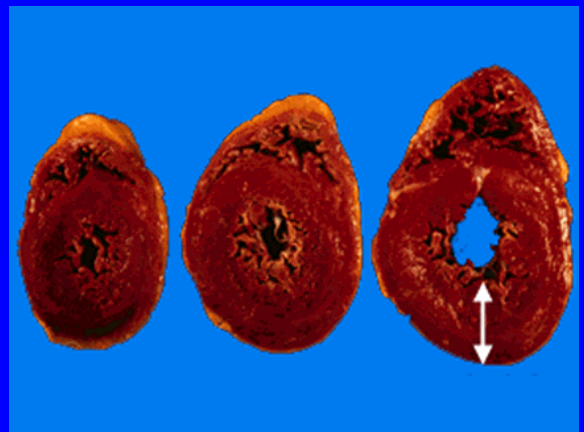
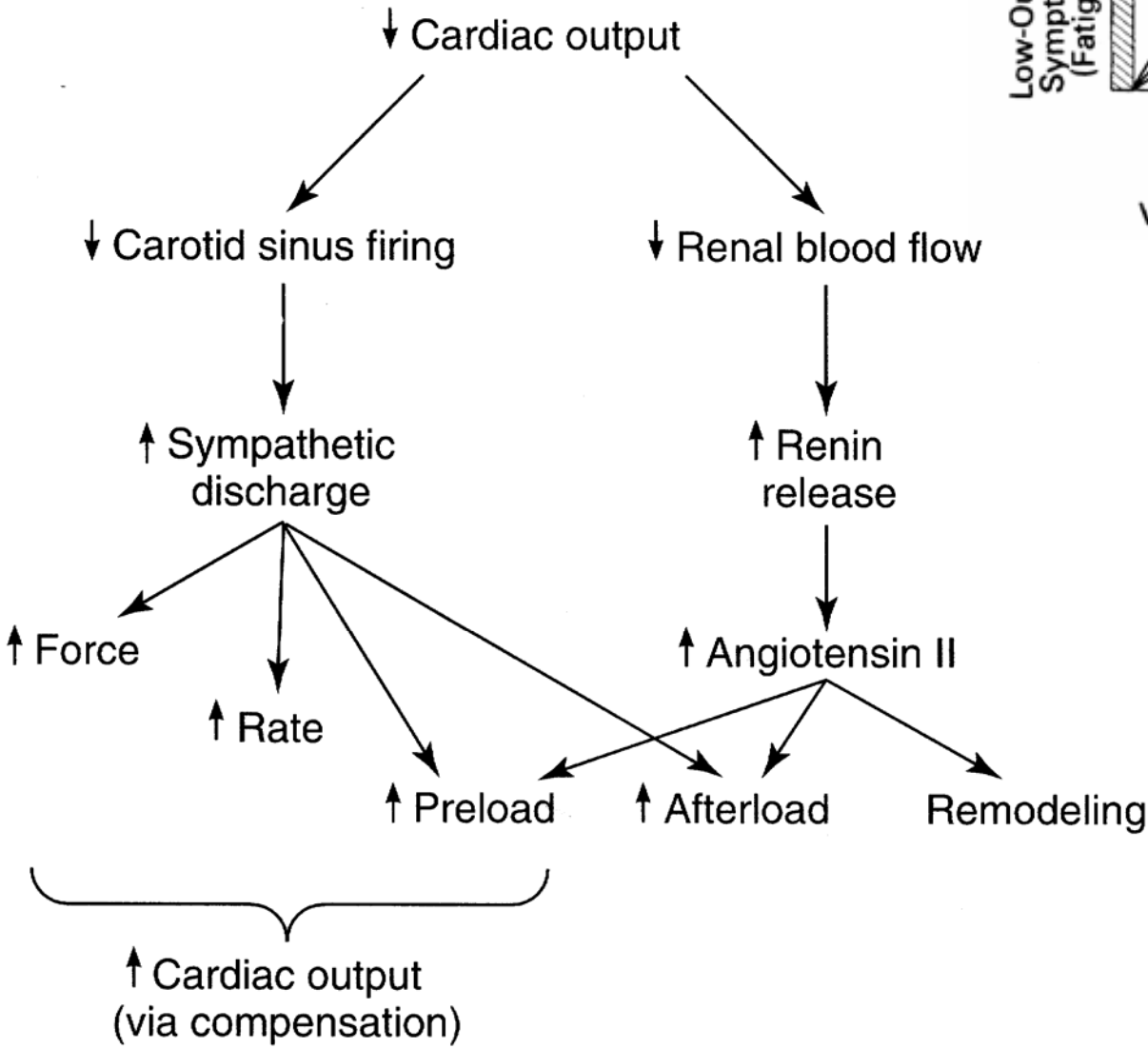
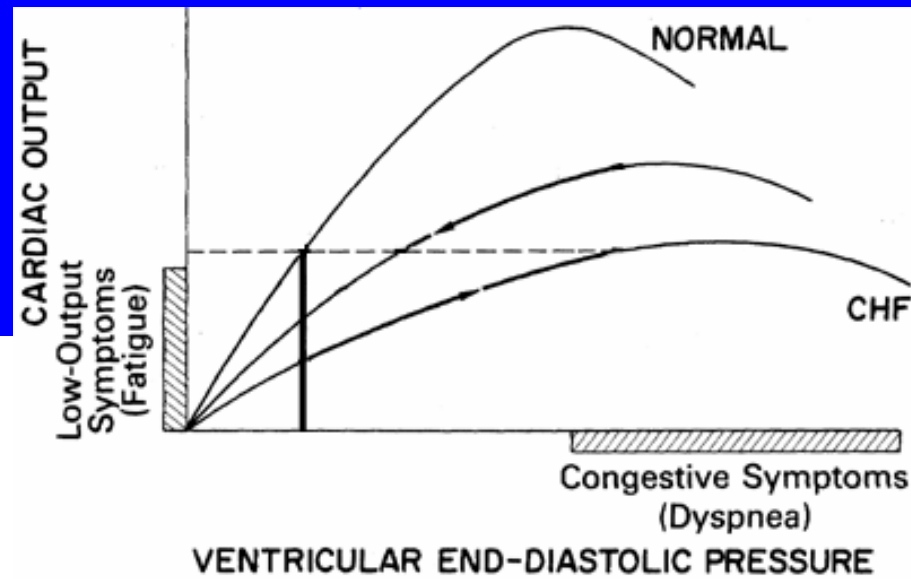
1875: Schiedeberg isolou a digoxitoxina da digital;



W. Withering com ramos de dedaleira



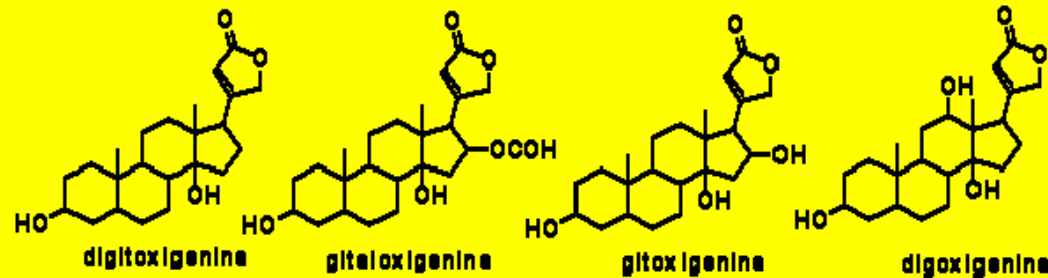
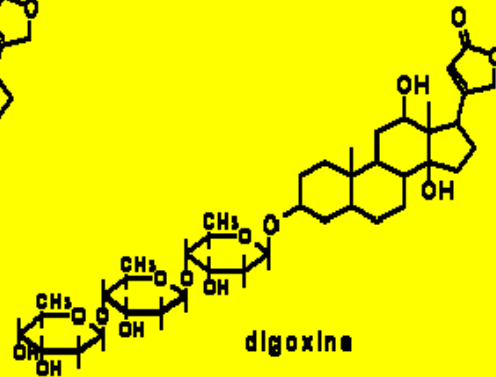
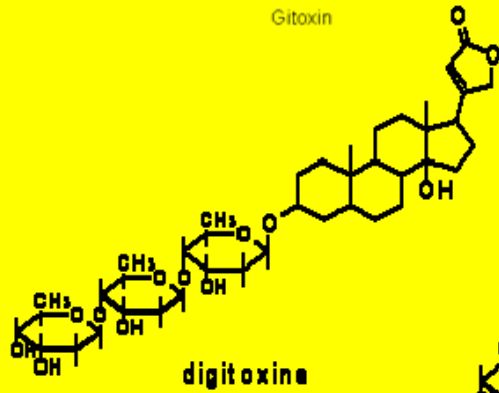
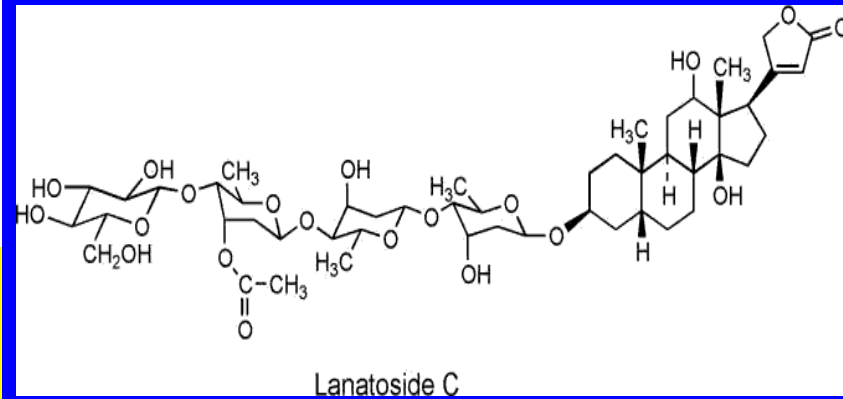
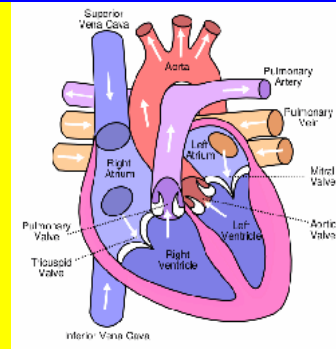
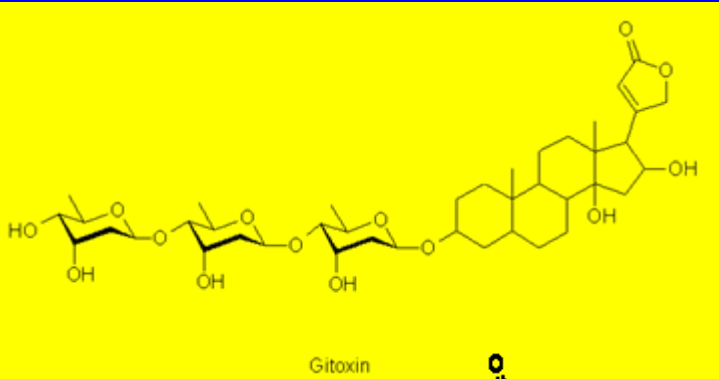
# Insuficiência Cardíaca Congestiva



Principais Sinais  
Clínicos:

- ♦ Hipertrofia Cardíaca
- ♦ Edema
- ♦ Fadiga

# Glicosídeos Naturais x Fármacos



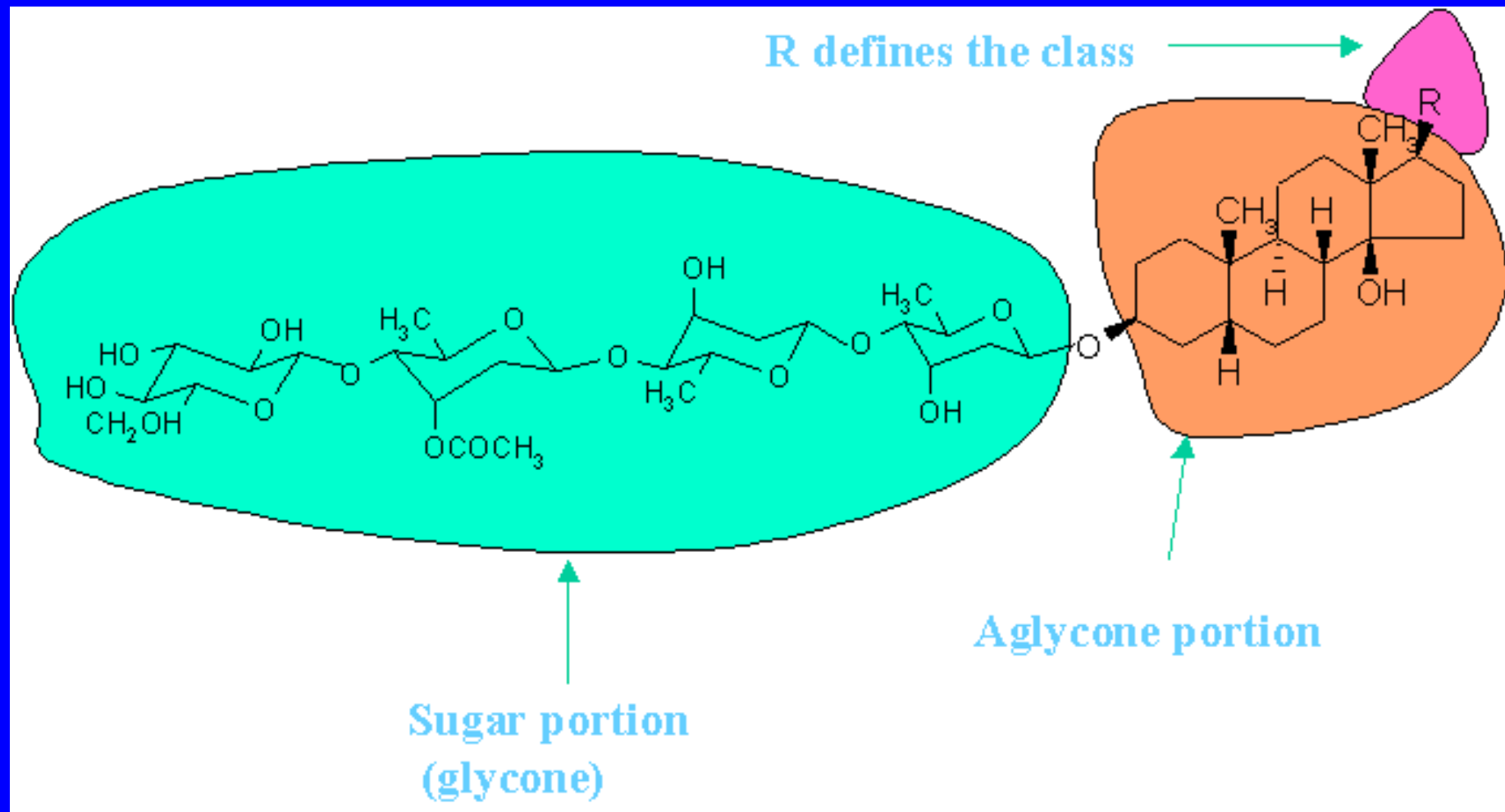
Alguns cardiotônicos de *Digitalis*



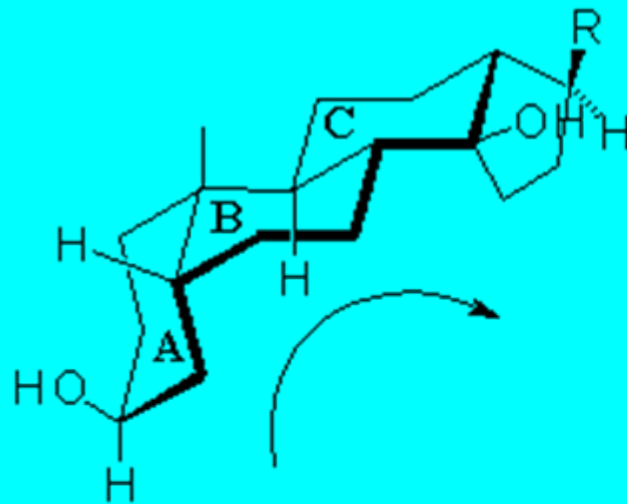
*Digitalis lanata*



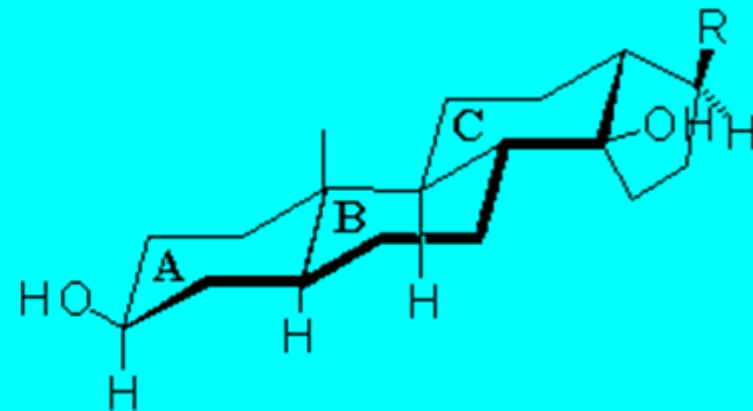
# Estrutura Geral dos Glicosídeos Cardiotônicos



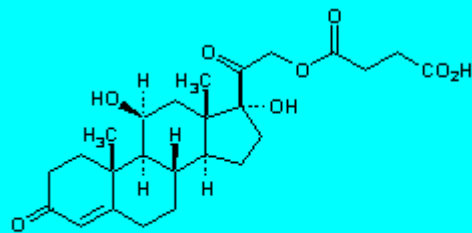
# Estereoquímica dos Glicosídeos Cardiotônicos



**cis-trans-cis ativo**  
**forma de "U" invertido**

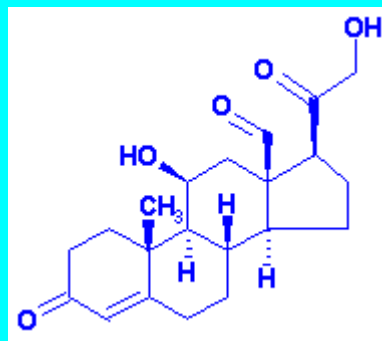


**trans-trans-cis inativo**

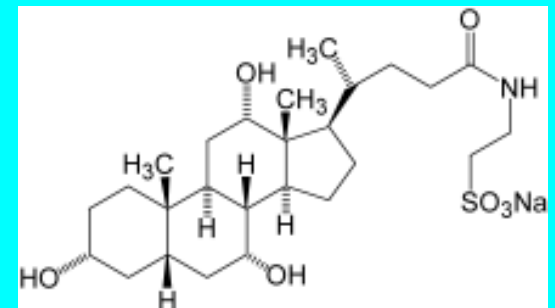


11 $\beta$ ,17,21-Trihydroxypregn-4-ene-3,20-dione 21-(hydrogen succinate)

**Hidrocortisone**

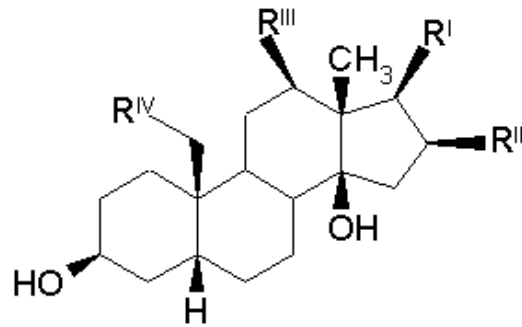


**Aldosterone**

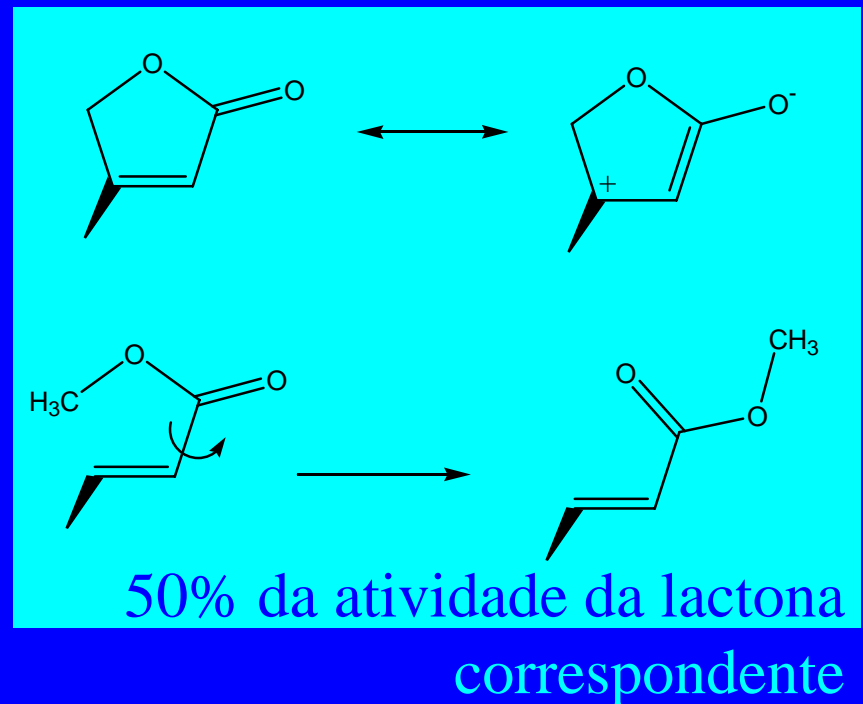
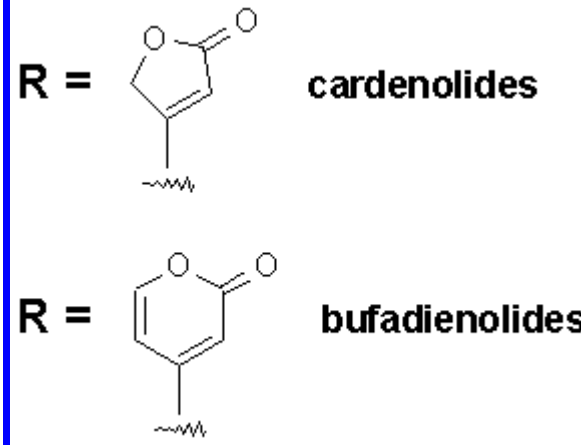


**Ácido Taurocólico**

# Papel do Grupo Lactona no Reconhecimento Molecular



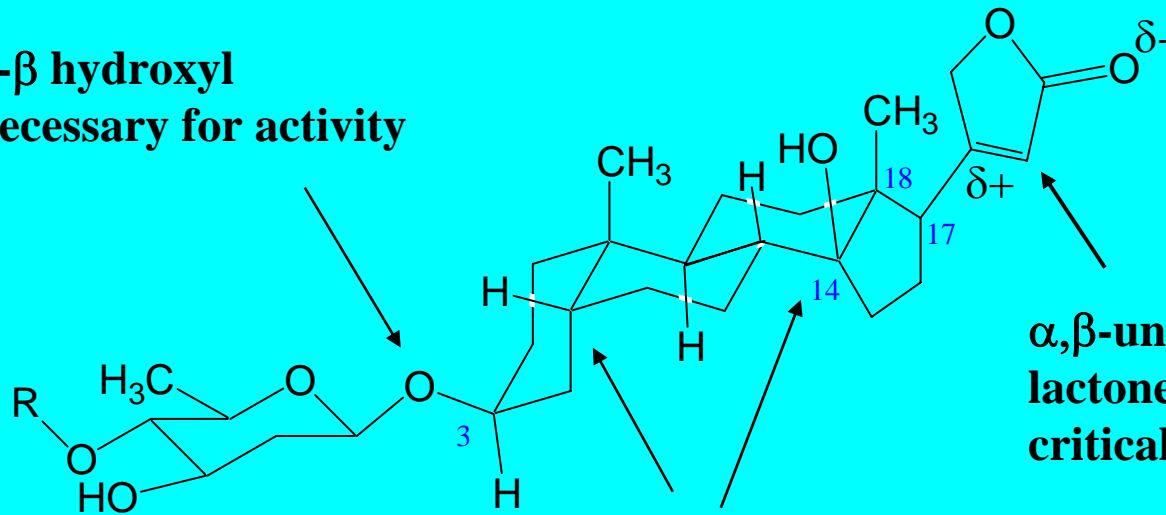
	$R^I$	$R^{II}$	$R^{III}$	$R^{IV}$
Digitoxigenin		H	H	H
Digoxigenin		H	OH	H
Gitoxigenin		OH	H	H
Ouabagenin		H	H	H
Strophanthidin		H	H	=O
Bufalin		H	H	H



# SAR dos Glicosídeos Cardiotônicos

**14-β hydroxyl  
enhances activity  
*cis* to 18-CH<sub>3</sub>**

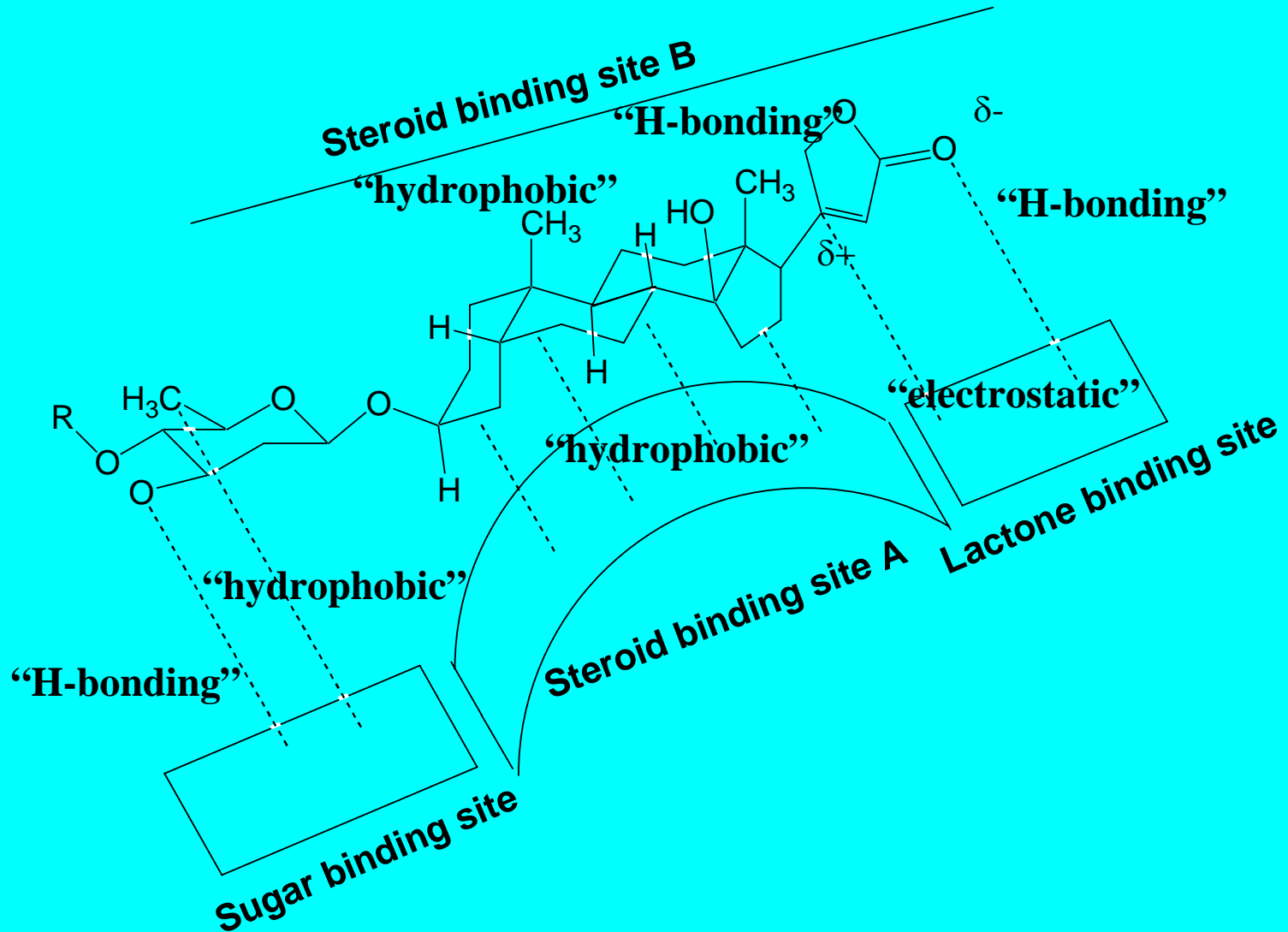
### 3-β hydroxyl necessary for activity



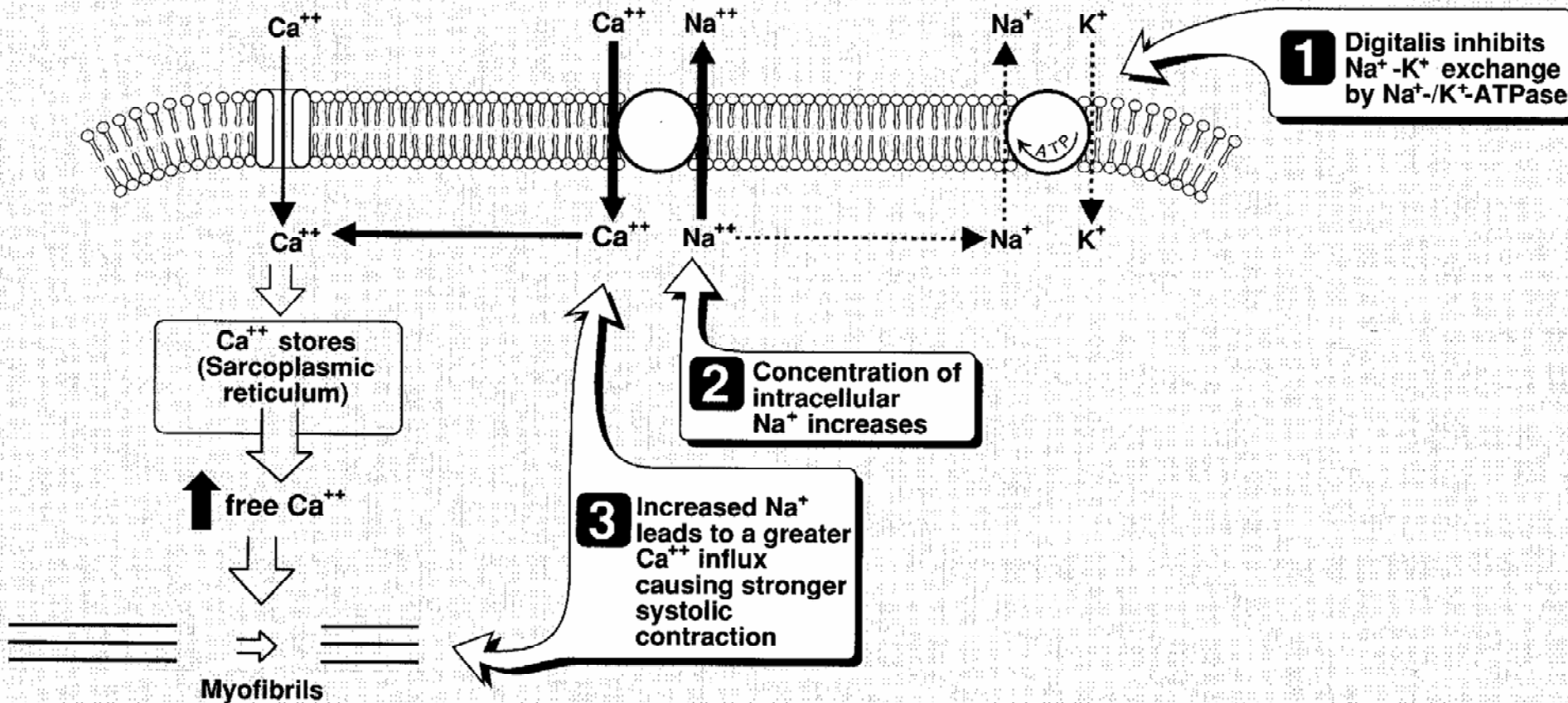
**$\alpha,\beta$ -unsaturated  
lactone  
critical for activity**

**A/B cis**  
**C/D cis**  
**for max activity**

# Modo de Ligação dos Glicosídeos Cardiotônicos



# Mecanismo de Ação do Glicosídeos Digitálicos

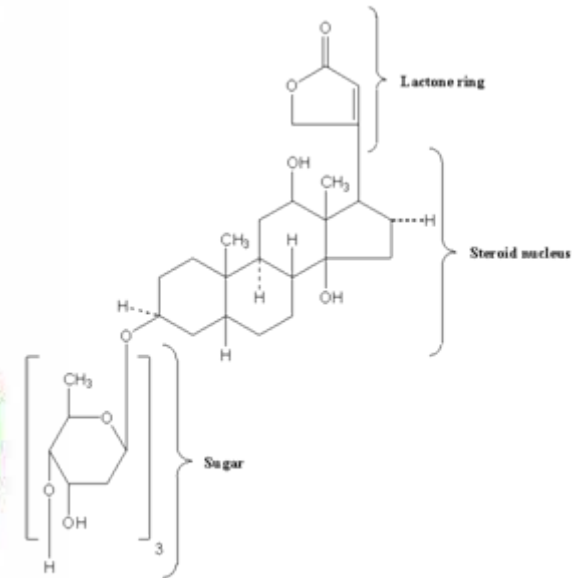
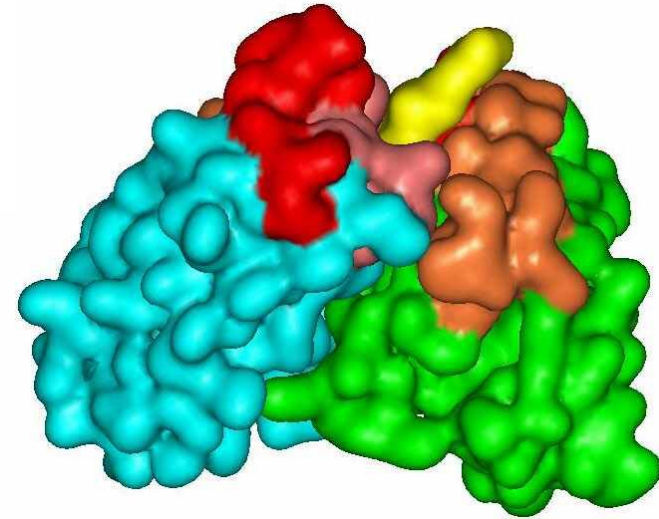


**Figure 16.5**

Mechanism of action of cardiac glycosides, or digitalis.

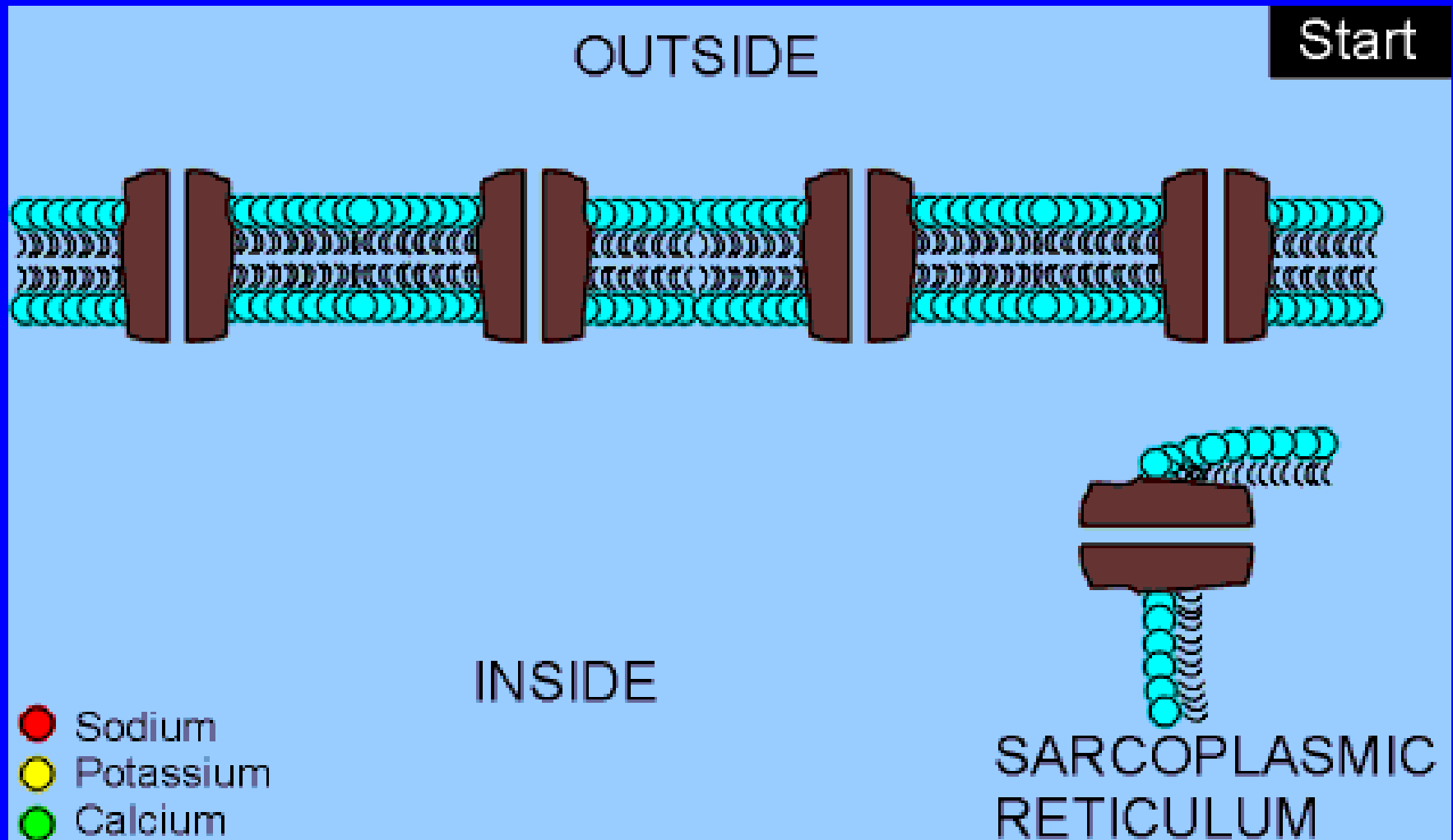


# Interação dos Glicosídeos com a ATPase $\text{Na}^+\text{-K}^+$



Digoxin

# Mecanismo da Ação Inotrópica dos Glicosídeos Cardiotônicos



# Toxicidade dos Glicosídeos Cardiotônicos



Mecanismo  
de  
Ação  
Dependente

Digoxina:

$LD_{50}$  (mg/Kg) =  
4,8 (*iv*); 6,2 (*ip*); 8,2 (*po*) [RATOS]

Digitoxina:

$LD_{50}$  (mg/Kg) = 0,18 (*po*) [GATOS]

Gitoxina:

$LD_{50}$  (mg/Kg) = 6,4 (*ip*) [RATOS]

Aspirina:

$LD_{50}$  (g/Kg) = 1,5 (*po*) [RATOS]

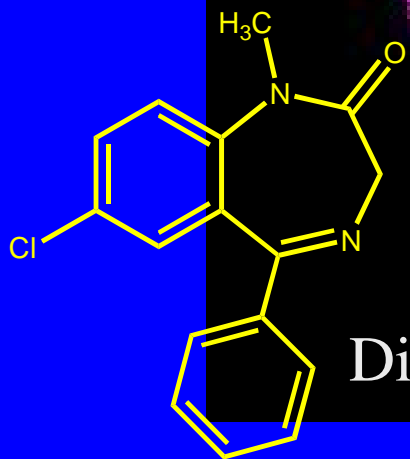
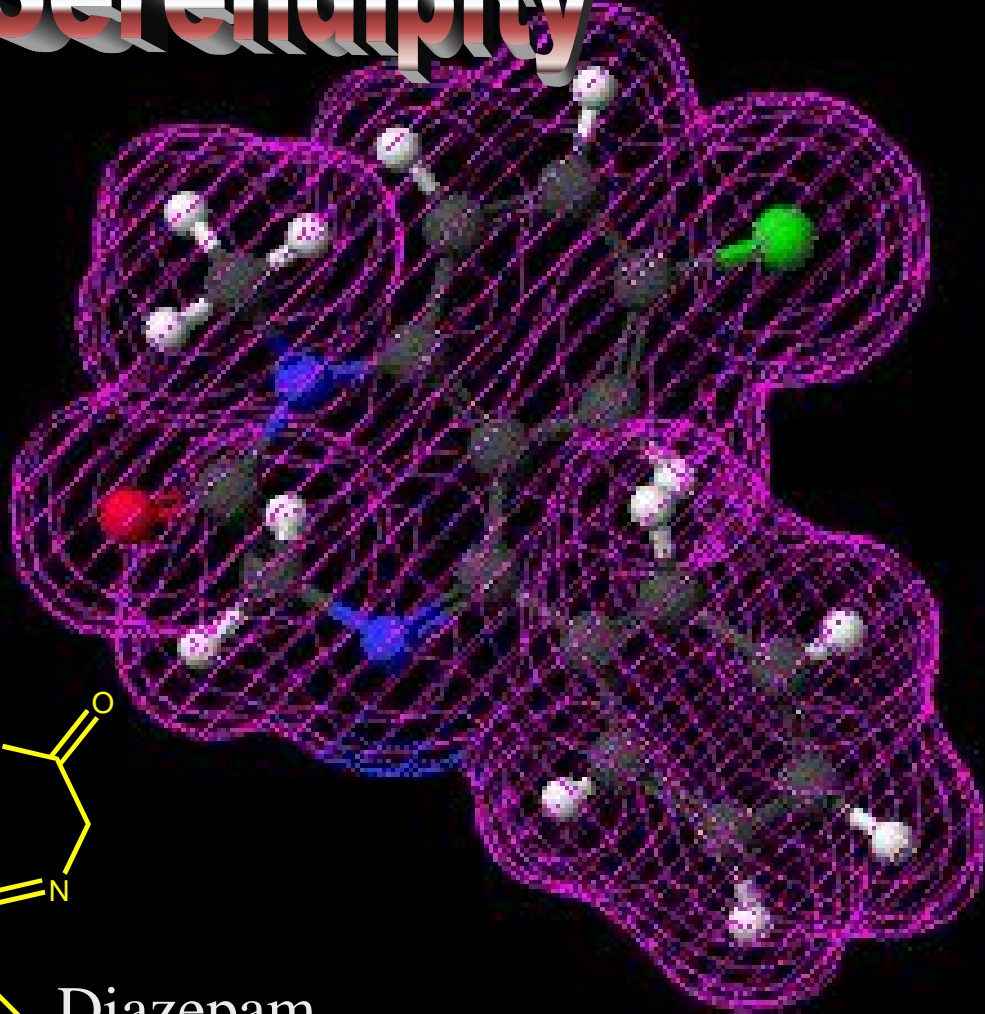
Índice Terapêutico ~ 2

# A História dos Fármacos Benzodiazepínicos



Leo Sternbach  
Roche, 1959

"Serendipity"



Diazepam

US\$ 21.000.000.000,00/ano



# O início do problema...

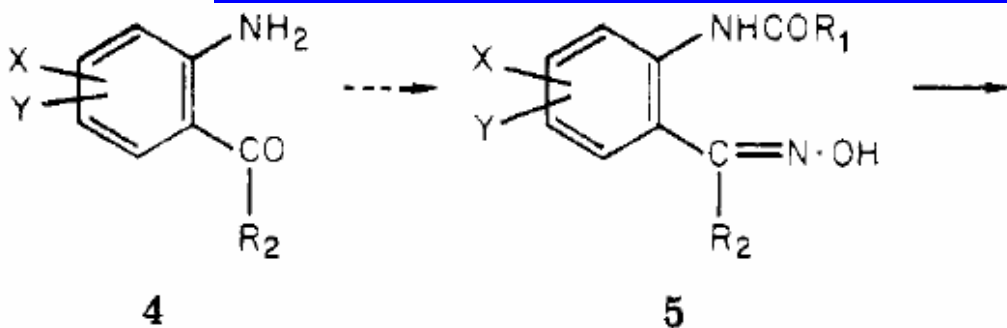
Desenvolvimento de Novos Tranquilizantes  
(Roche, 1950)

Modificação de Protótipos já Existentes  
x Novas Classes Químicas

Pós-Doutorado (Univ. Cracóvia, 1930) –  
Corantes e Intermediários Sintéticos

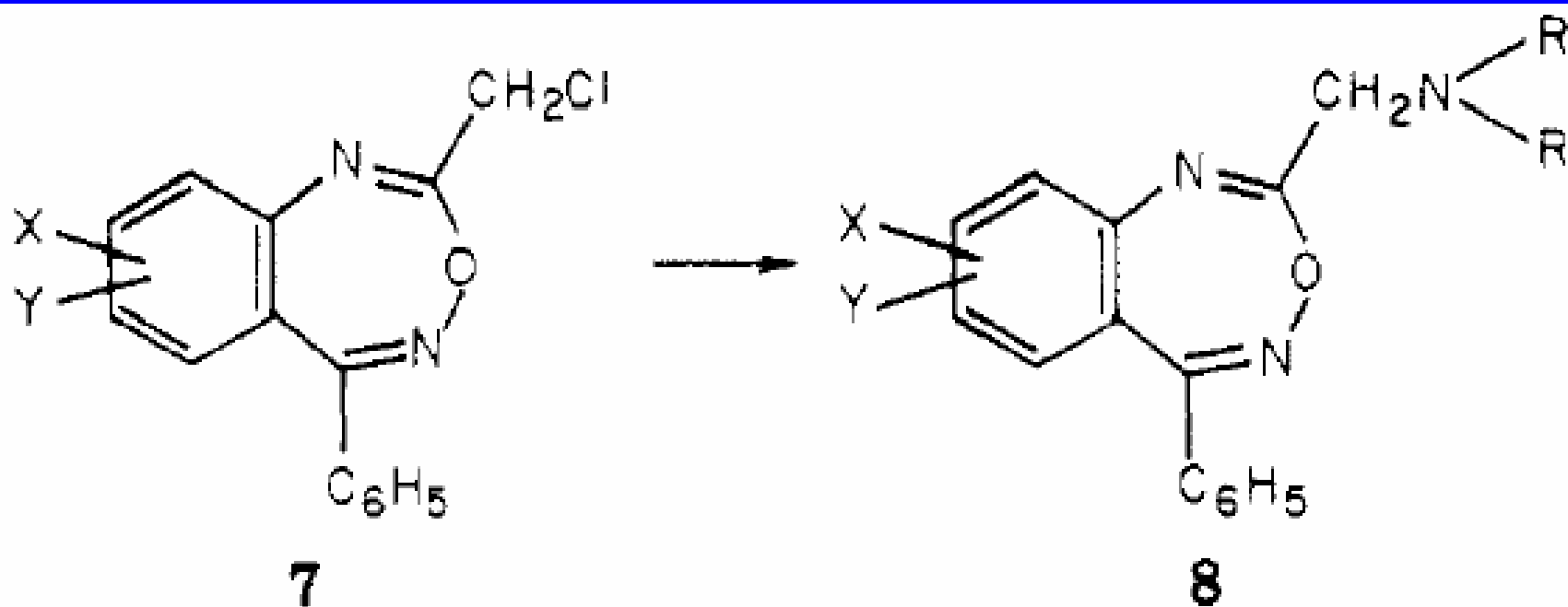
Heptoxdiazines (Estrutura química  
caracterizada em 1924)

- Meprobamato (Miltown)
- Clorpromazina (SKF)
- Fenobarbital (Rhodia)



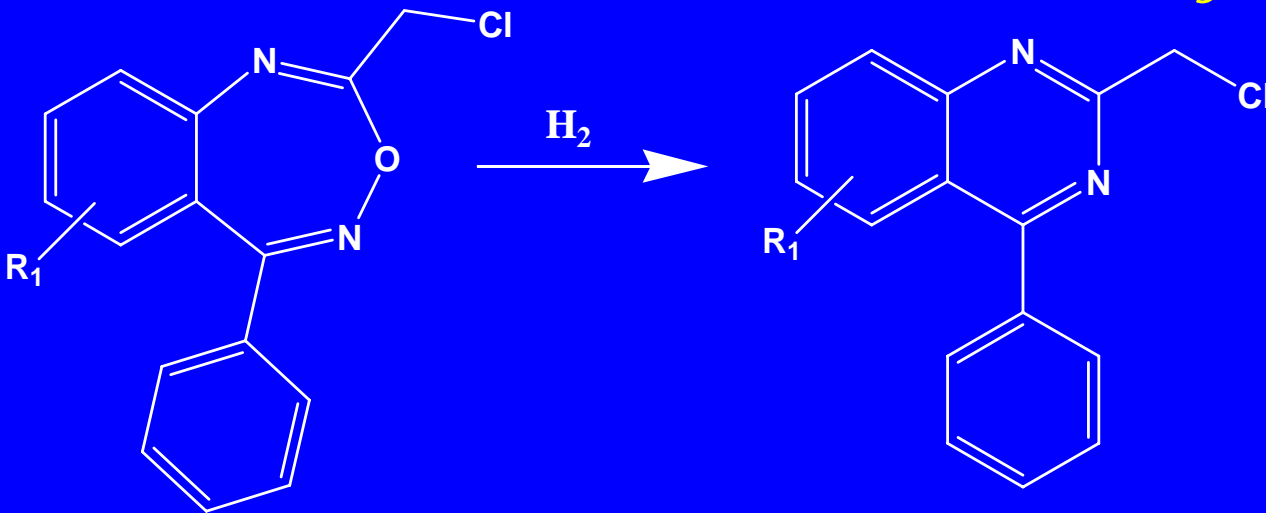
- Pouco Explorada
- Acessível
- Permitir Múltiplas Variações
- Desafio Químico
- Potencial Atividade Biológica

# Definição da Estratégia



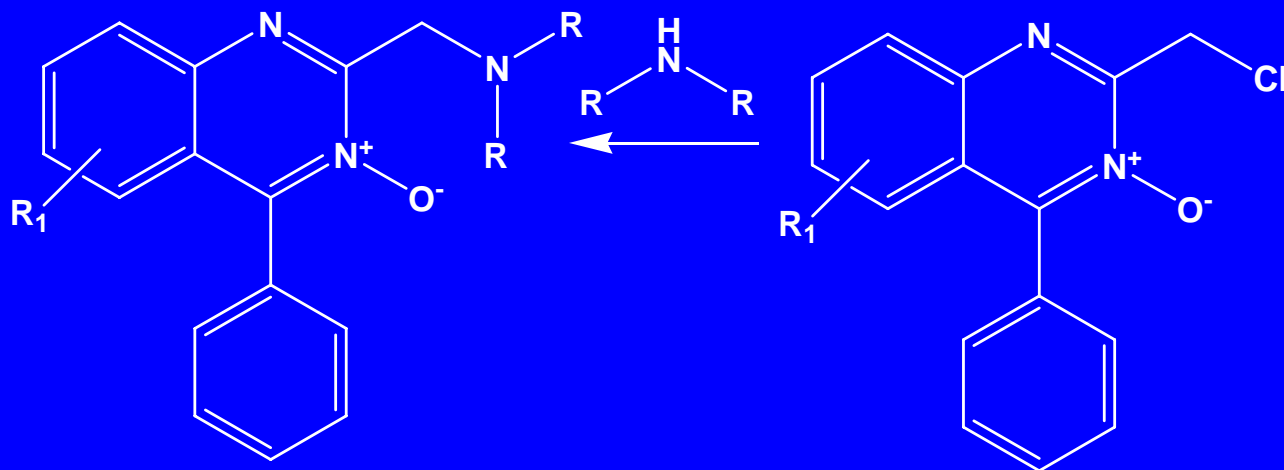


# Confirmação da Estrutura das Heptoxadiazinas



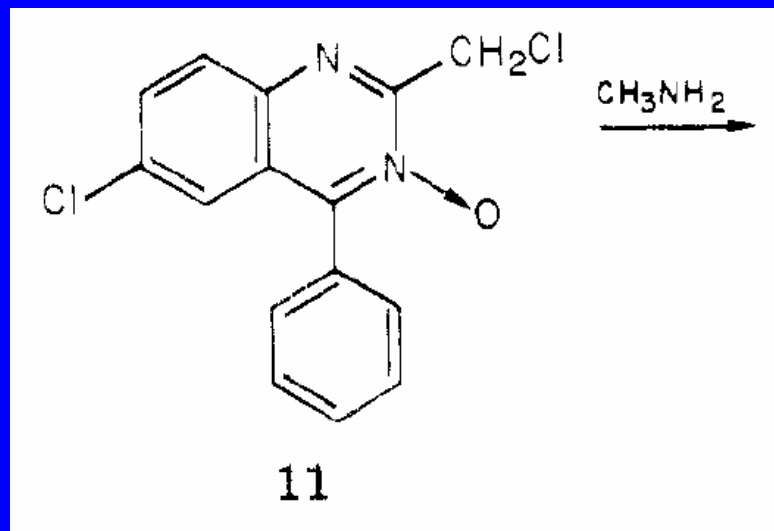
Propriedades  
Farmacológicas:  
Aminas-N-óxido

ou os produtos  
hidrogenados  
correspondentes  
foram inativos



Quinazolin-N-Óxidos

- 1955 (Segunda Metade): Trabalho com as Quinazolininas foi interrompido.
  - Isolamento, Purificação e Degradação de Vários Antibióticos
- 1957: Limpeza do Laboratório



Earl Reeder (Identifica dois produtos)

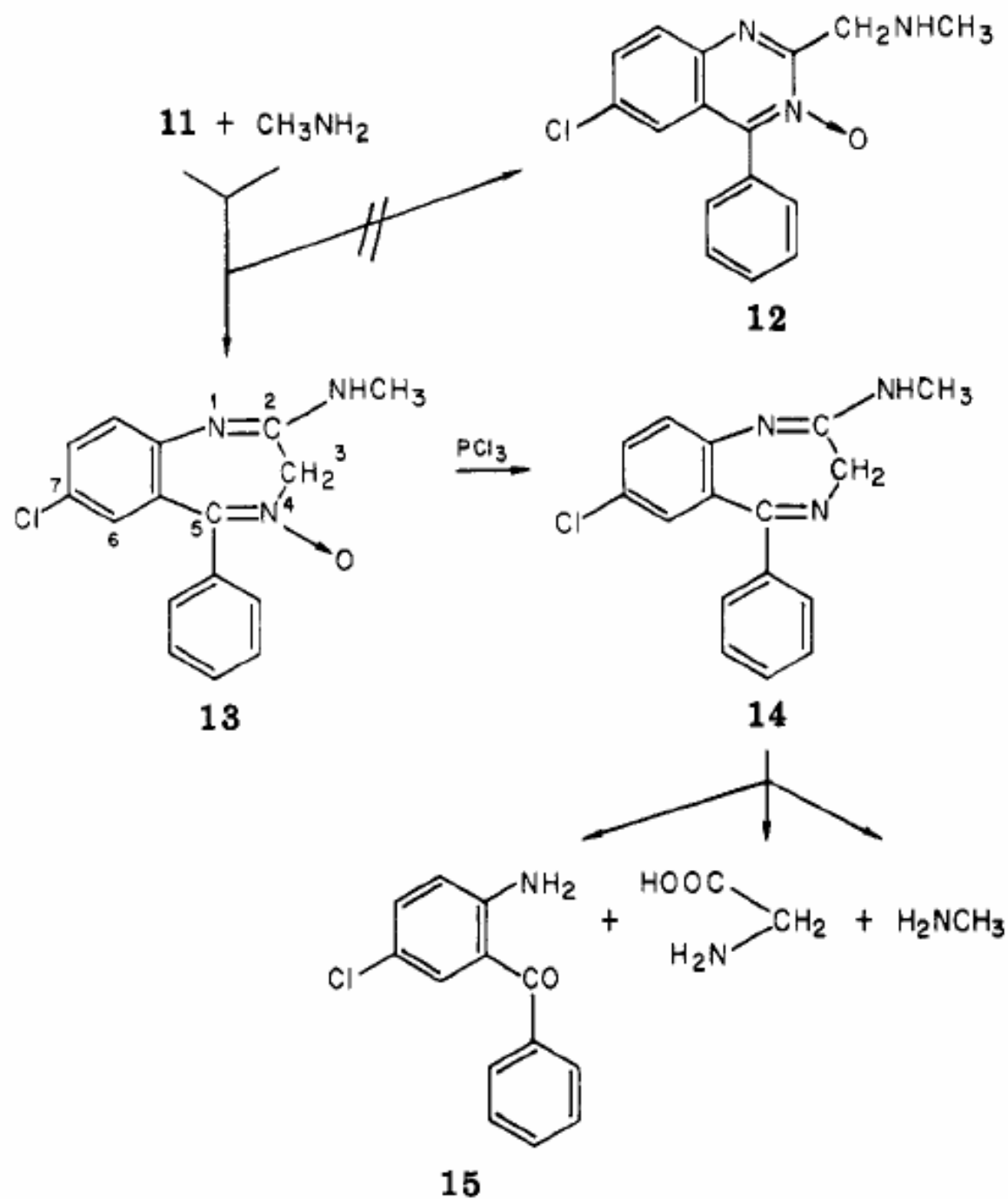
- 1957 (Maio): Dr. Lowell Randall, Farmacologista

Table I. Pharmacological Properties<sup>a</sup> of “New Compound”, Meprobamate, Chlorpromazine, and Phenobarbital

compd	inclined screen	foot shock	cat	anticonvulsant tests		
				pentylene- tetrazole	electroshock	
					max	min
new compound	100	40	2	18	92	150
meprobamate	250	250	100	150	200	167
chlorpromazine	17	20	2.5	42	150	600
phenobarbital	120	80	10	75	18	90

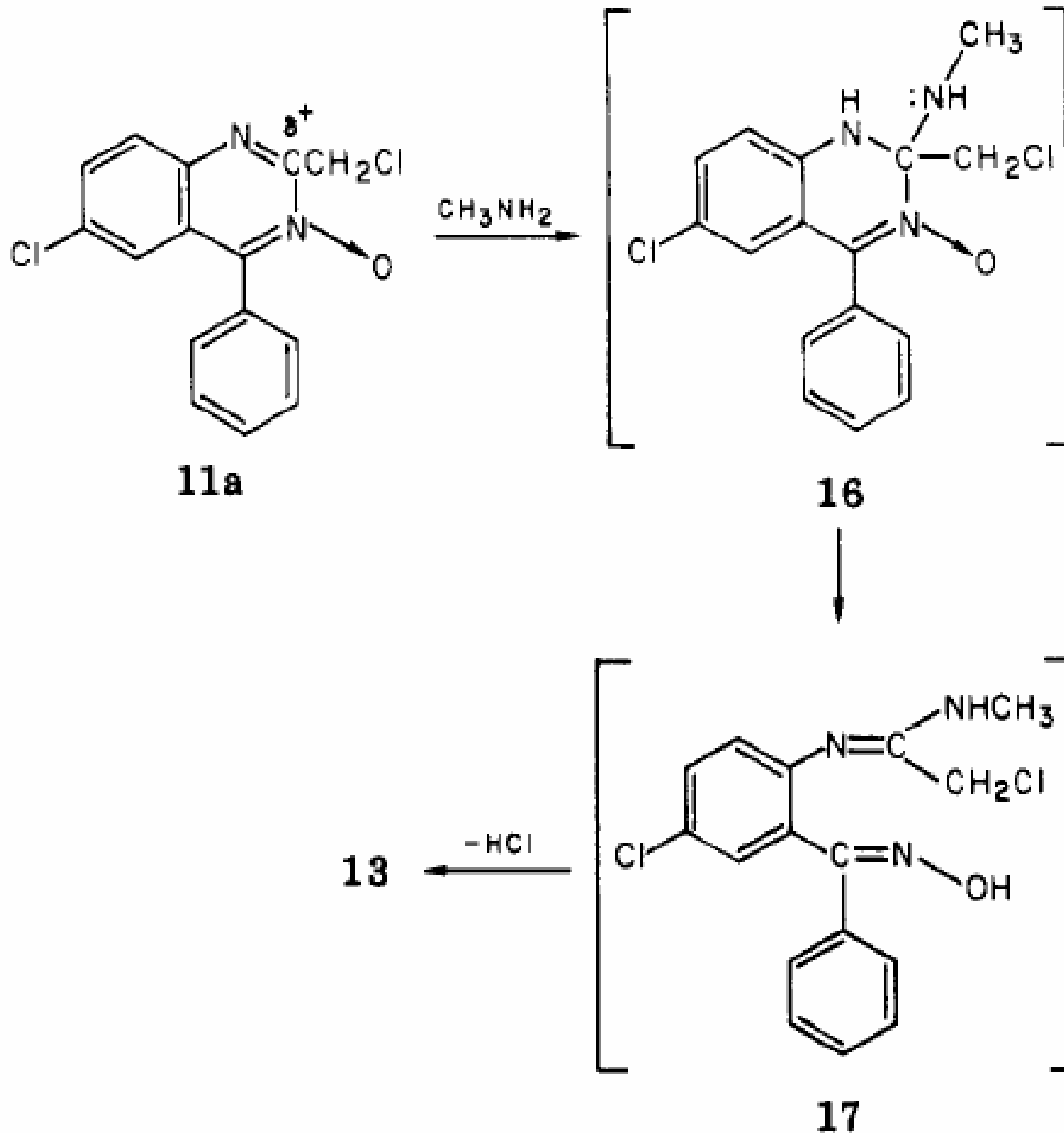
<sup>a</sup> Dose (mg/kg) of orally administered drug required to achieve the desired effect.

Scheme I

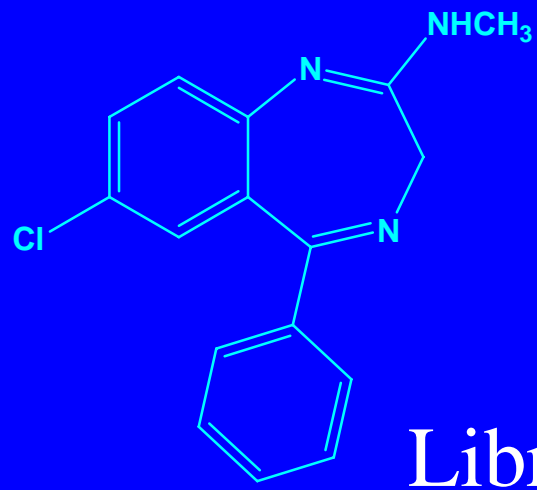


Caracterização  
Estrutural  
do  
“Novo Composto”

# Mecanismo de Formação dos Derivados Benzodiazepínicos



# Caracterização Farmacológica dos Produtos de Transformação do Clordiazepóxido (13)



Librium<sup>R</sup>

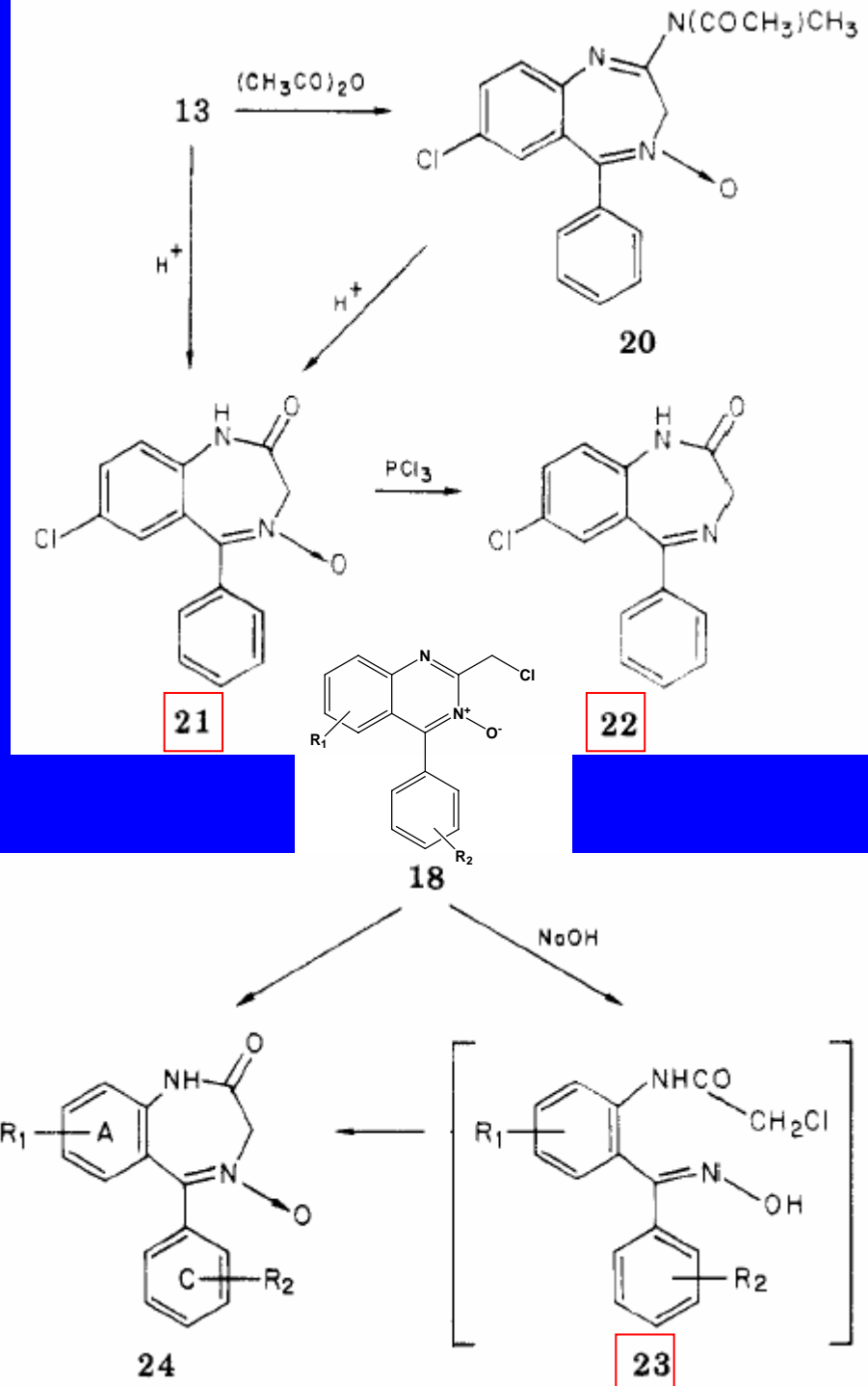
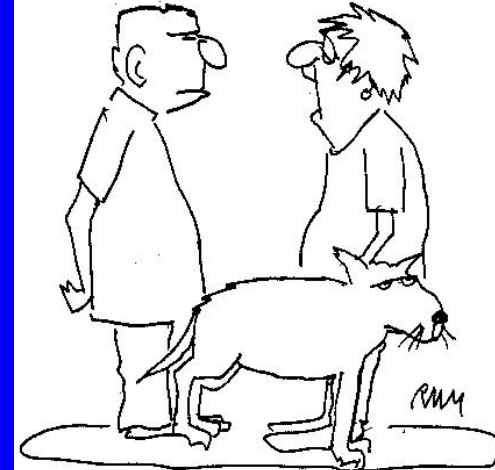
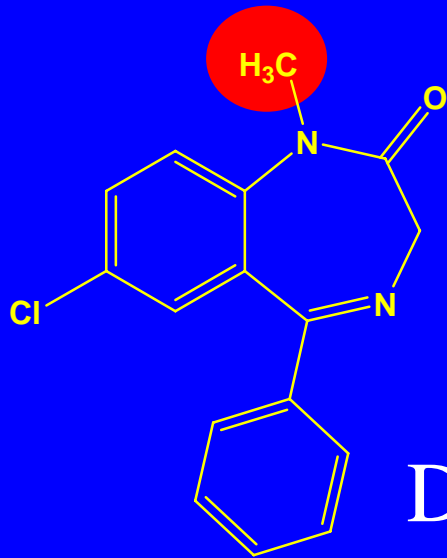


Table II. Pharmacological Activity<sup>a</sup> of Clordiazepoxide and Its Transformation Products

compd	in- clined screen	foot shock	cat	anticonvulsant tests		
				pen- tylene tetra- zole	electroshock	
					max	min
13	100	40	2	18	92	150
21	100	20	2	15	150	150
22	75	40	1	6	52	400
23	75	20	1	6	25	61

<sup>a</sup> Dose (mg/kg) of orally administered drug required to achieve the desired effect.

# E assim nasceu o diazepam.....



"I was giving him his Valium, and the vial fell in the toilet, so I'm going to need a lot more!"

## Diazepam (Valium<sup>R</sup>)

Table III. Comparison of the Pharmacological Activity<sup>a</sup> of Chlordiazepoxide with That of Diazepam

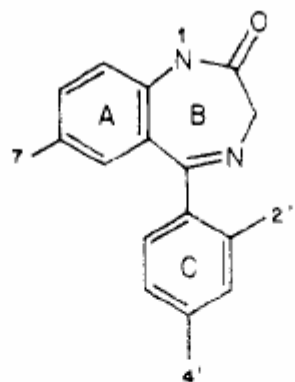
compd	inclined screen	foot shock	cat	anticonvulsant tests		
				pentylene- tetrazole	electroshock	
					max	min
chlordiazepoxide	100	40	2	18	92	150
diazepam	30	10	0.2	1.4	6.4	64

<sup>a</sup> Dose (mg/kg) of orally administered drug required to achieve the desired effect.

- Mercado Farmacêutico:
- Librium<sup>®</sup> foi lançado em 1959
- Valium<sup>®</sup> foi lançado logo em seguida (1963)

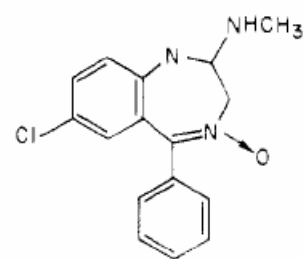
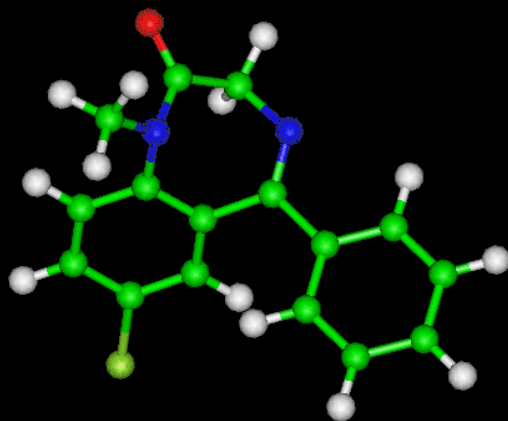


# SAR dos Benzodiazepínicos

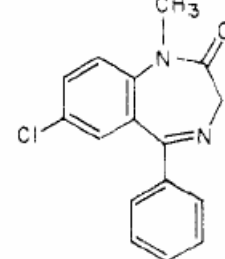


32

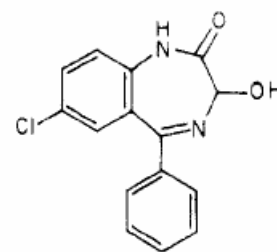
<sup>a</sup> Ring A: (position 7) generally, increased by electron-withdrawing groups, e.g., halogens, NO<sub>2</sub>, and CF<sub>3</sub>, and decreased by electron-releasing groups such as CH<sub>3</sub> and OCH<sub>3</sub>; decreased by any substituents in any positions other than 7. Ring B: increased by a methyl group at position 1; decreased by larger substituents; *tert*-butyl derivative is completely inactive. Ring C: increased by halogens at the 2' position (e.g., Cl and F); very strongly decreased by a substituent at the 4' position.



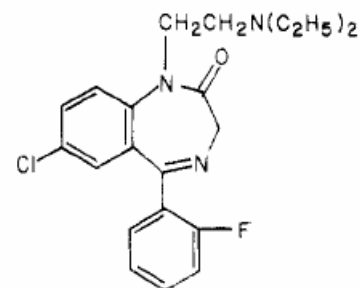
chlordiazepoxide<sup>a</sup>  
(Librium, 1960)



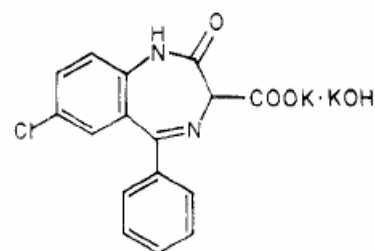
diazepam  
(Valium, 1963)



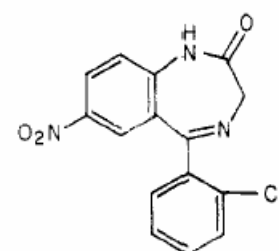
oxazepam  
(Serax, 1965)



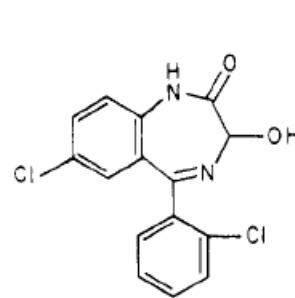
flurazepam<sup>b</sup>  
(Dalmane, 1970)



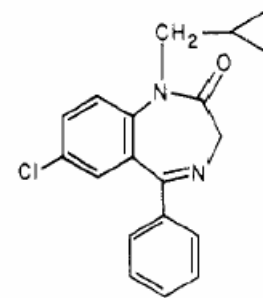
clorazepate  
(Tranxene, 1972)



clonazepam  
(Clonopin, 1975)



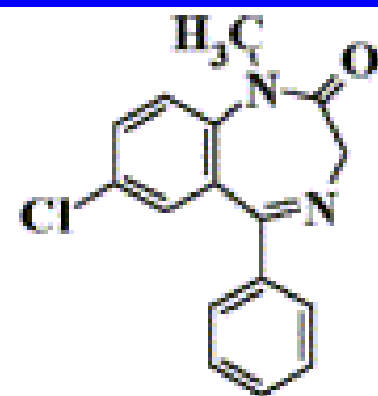
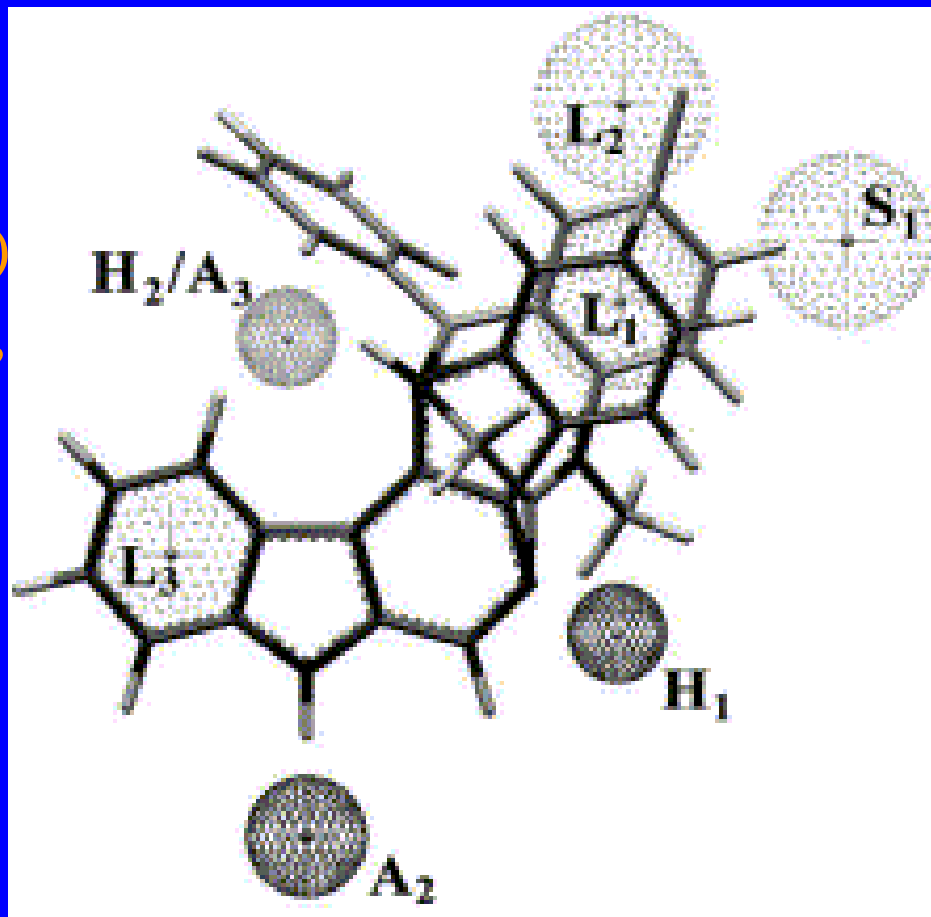
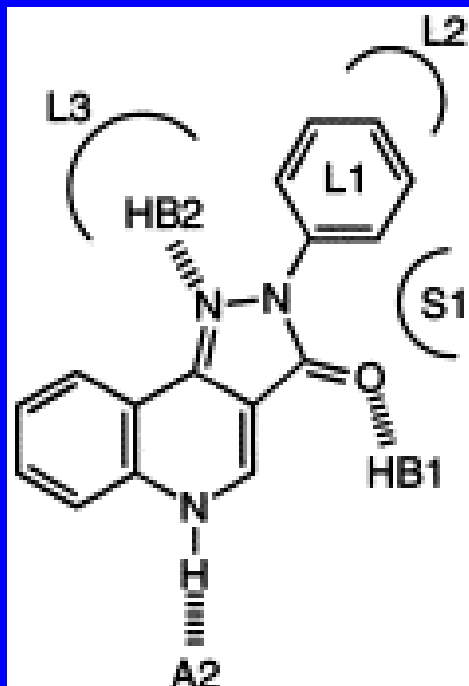
lorazepam  
(Ativan, 1977)



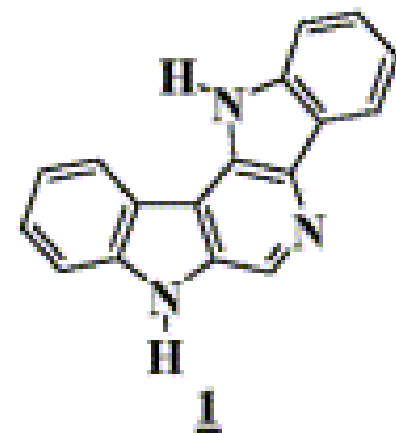
prazepam  
(Verstran, 1977)

# Modelos de Afinidade para o Receptor BZDP

Cook, J. M. (1998)  
*J. Med. Chem.* **41**,  
 2537–2552.



Diazepam

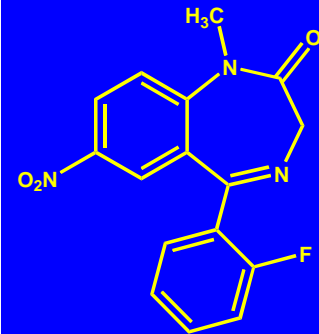
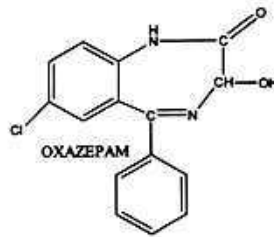
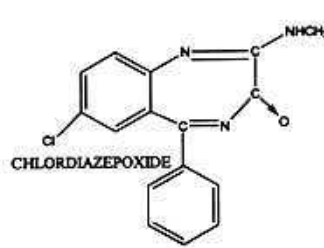
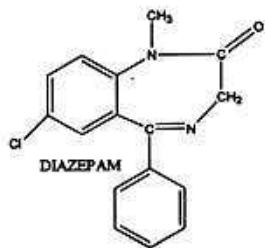


A2 = Aceptor de Ligação Hidrogênio  
 H1 = Doador de Ligação Hidrogênio  
 H2/A3 = Doador/Aceptor de "H"  
 L1-L-3 = Sítios Lipofílicos  
 S1 = Sítio Estérico/Repulsivo

BENZODIAZEPINA (DOSE)

ALTA

BAIXA



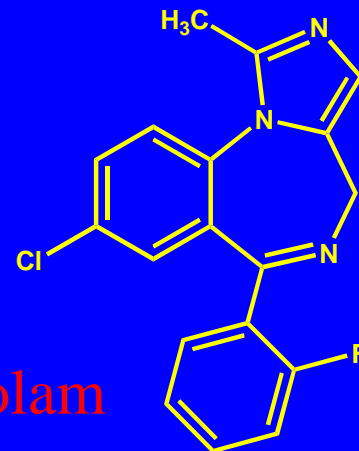
MORTE

ANESTESIA  
GERAL

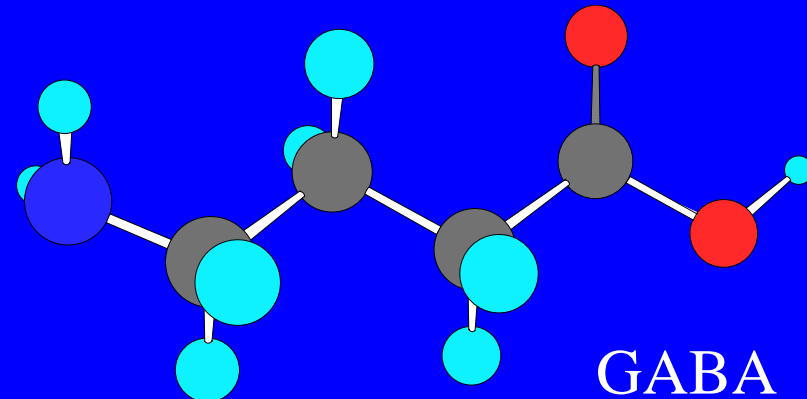
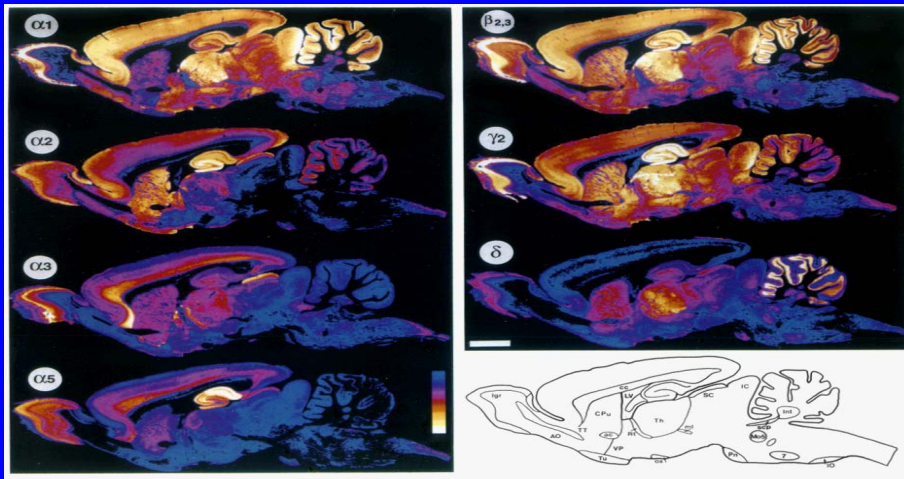
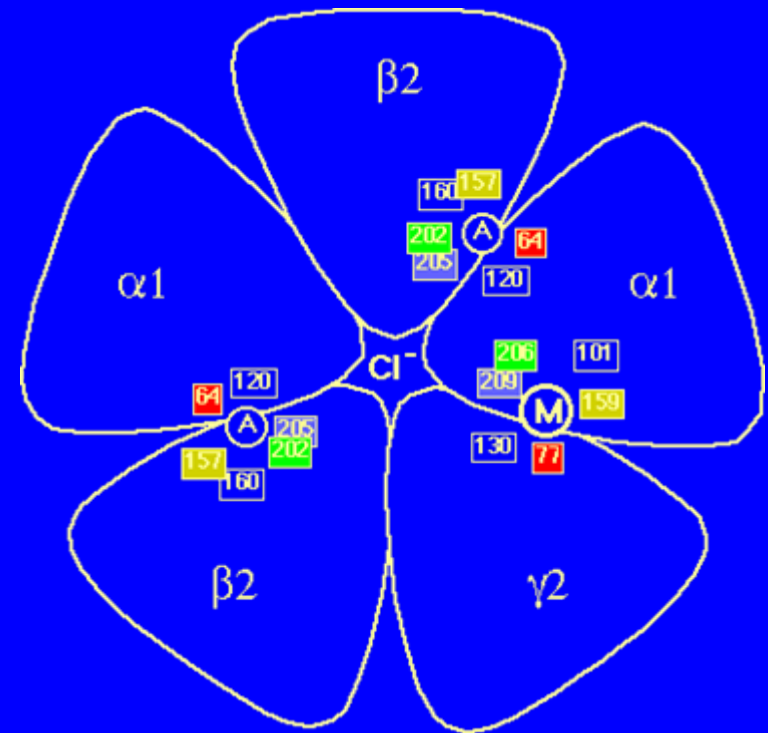
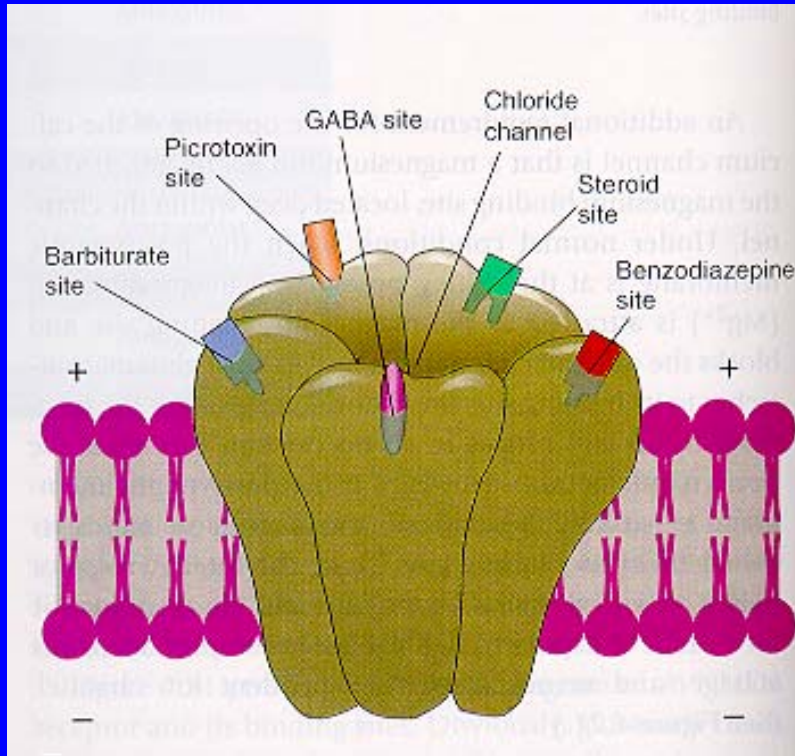
EFEITOS  
SEDATIVOS

EFEITOS  
ANTI-  
CONVULSIVANTES

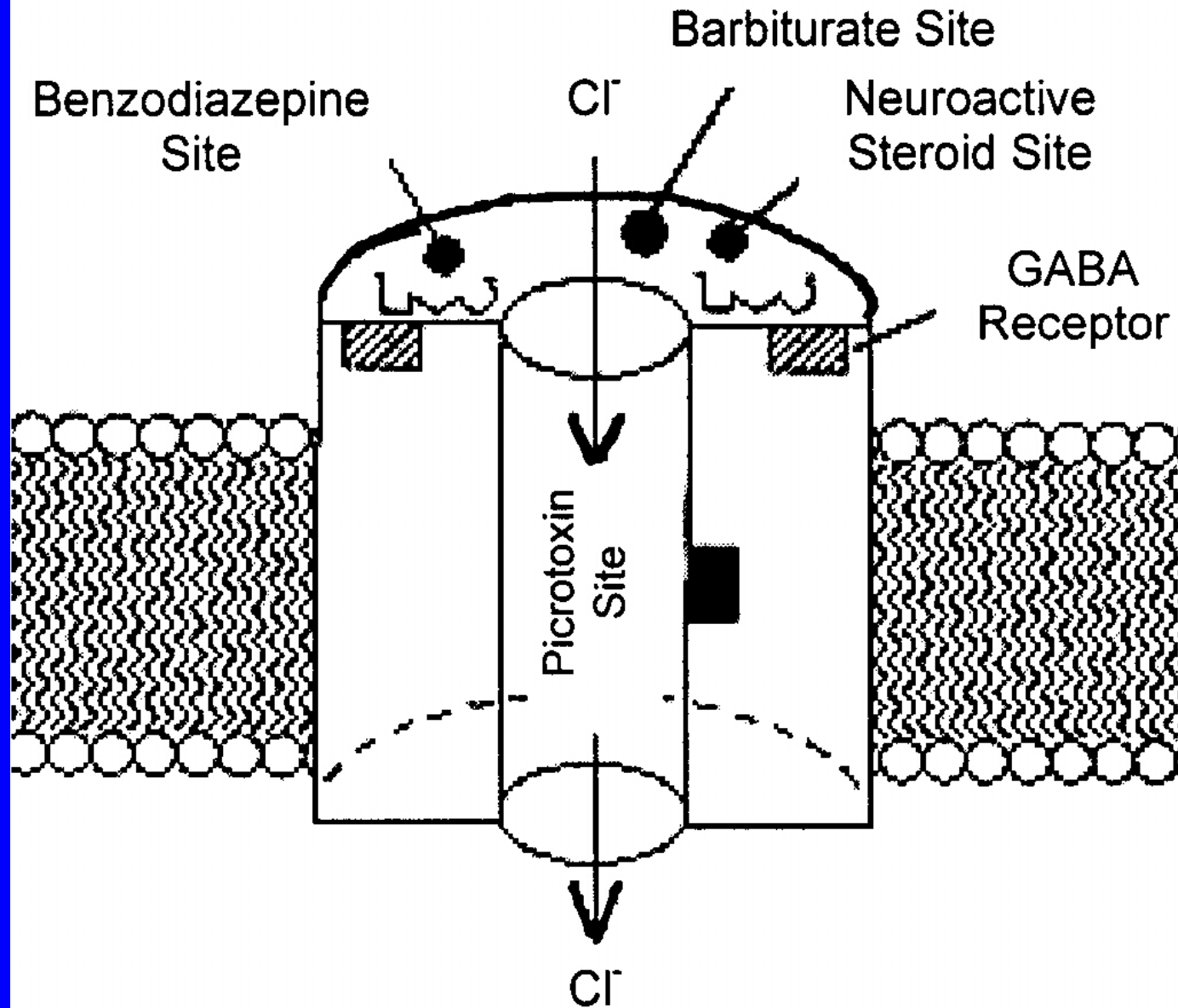
EFEITOS  
ANSIOLÍTICOS



# Receptor Benzodiazepínico

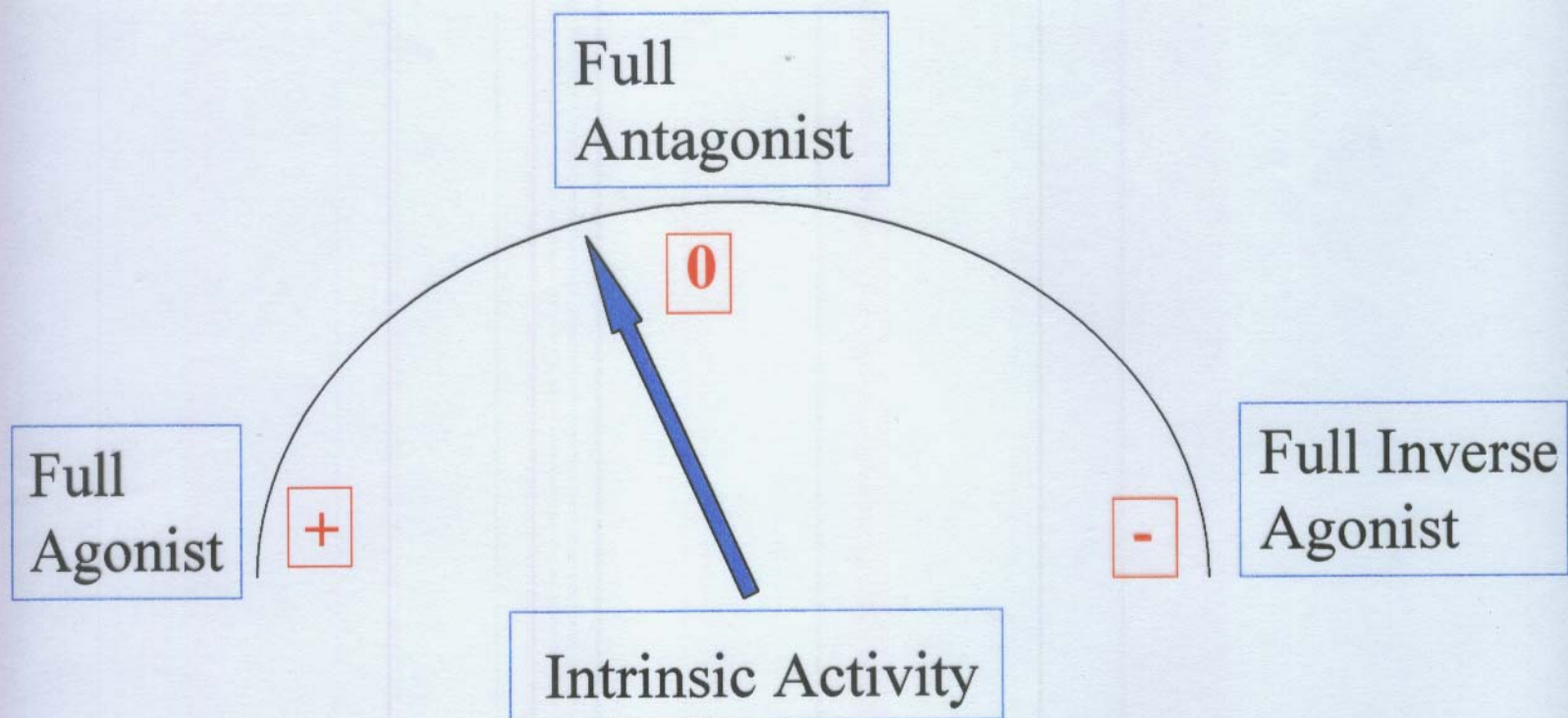


# GABA<sub>A</sub> Receptor Complex



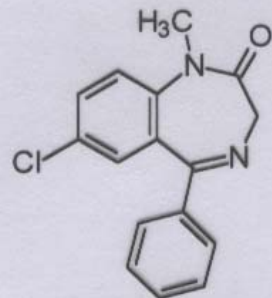


# Atividade Intrínseca de BZDP

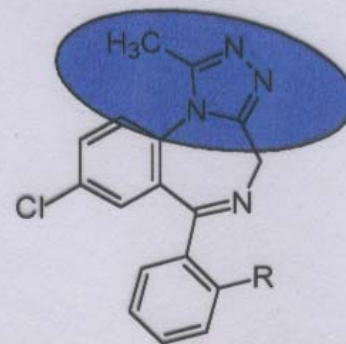


Full Agonist	Full Antagonist	Full Inverse Agonist
Sedative	None	Stimulant
Amnestic	None	Enhance Cognition
Anxiolytic	None	Anxiogenic
Anticonvulsant	None	Convulsant
Ataxia	None	Not Determined
Muscle Relaxant	None	Not Determined
Alcohol Potentiation	None	Reversal of Alcohol Effects





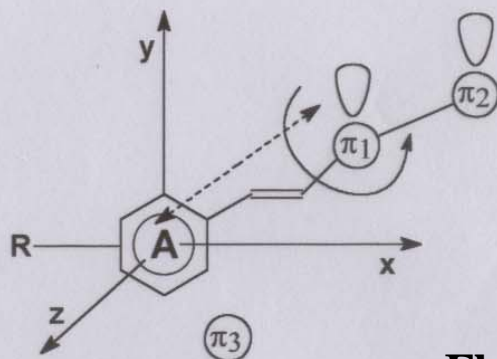
**DIAZEPAM**



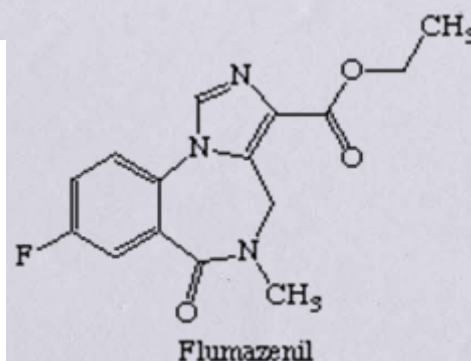
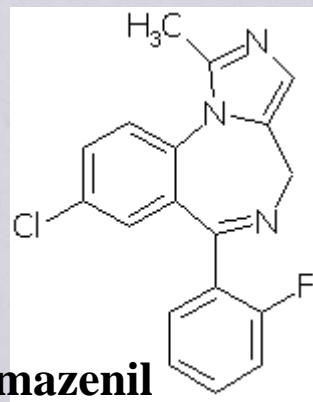
**TRIAZOLOBENZODIAZEPINAS**

Compound <sup>a)</sup>	R	Inclined Screen	Foot Shock	Cat	Anticonvulsant tests		
					pentylene-tetrazole	electroshock	
						max	min
<b>Diazepam</b>	---	30	10	0,2	1,4	6,4	64
<b>Alprazolam</b>	H	2	2,5	0,5	0,3	80	---
<b>Triazolam</b>	Cl	50	0,5	0,05	0,06	40	---

# Modelo para a Atividade Intrínseca de ligantes BZDP



**Flumazenil**




**A** = aromatic or heteroaromatic ring present in XY plane

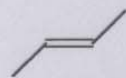
$\pi_1$  = Proton-accepting atom or group, mid A to  $\pi_1$  distance  $\sim 4,0-5,5$  Å

$\pi_2$  = Auxiliary proton-accepting atom or group, mid A to  $\pi_2$  distance  $\sim 7,0$  Å  $\rightarrow$  **ANTAGONIST ACTIVITY**

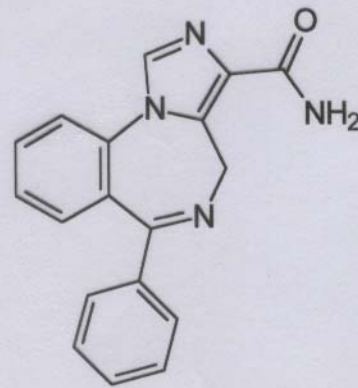
**R** = A ring substituent not required for binding, can affect intrinsic activity

$\pi_3$  = Out of XY plane, aromatic or heteroaromatic substituent may fulfil hydrophobic and/or steric requirement, not allowed for **inverse agonists**

 = indicates flexibility of  $\pi_1$  group

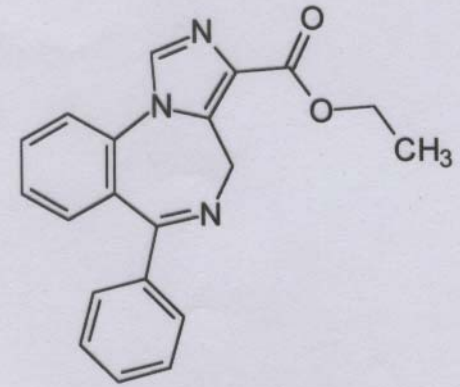
 = indicative of structural skeleton fixing the relationship between A to  $\pi_1$  should be at least partially unsaturated, may be part of aromatic or heteroaromatic ring

# Como Explicar ?



**AGONIST**

$IC_{50} = 45 \text{ nM}$



**ANTAGONIST**

$IC_{50} = 7.2 \text{ nM}$



# História da Aspirina

1500 B.C.: Egípcios gravaram uma coleção de prescrições médicas, a qual incluía uma receita usando uma infusão de folhas de *murta* secas (a qual contém ácido salicílico) para aliviar dor nas costas.

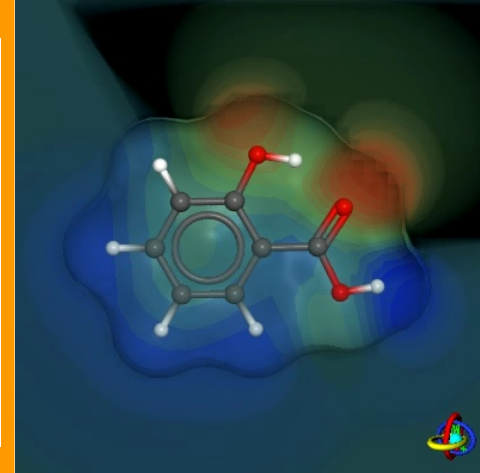
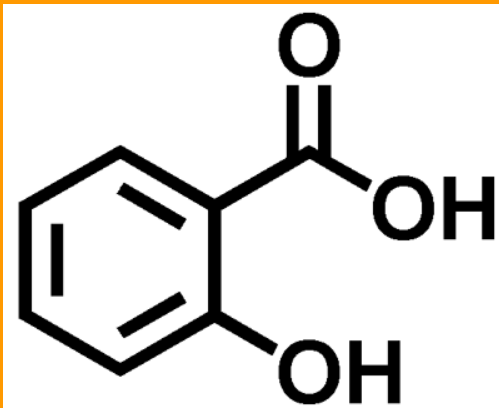
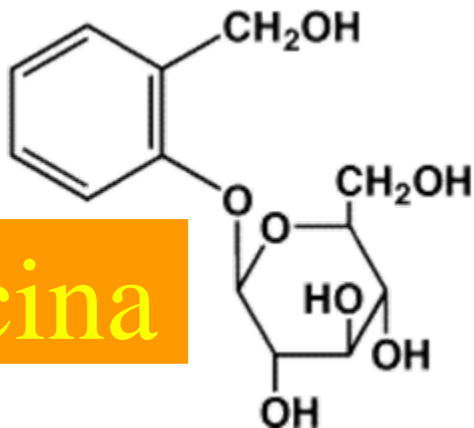


200 B.C.: Hippocrates, um médico Grego, prescreve o uso das folhas e cascas de *salgueiro* (o qual, como a *murta*, também contém ácido salicílico) para aliviar a febre e a dor, incluindo as dores do trabalho de parto.



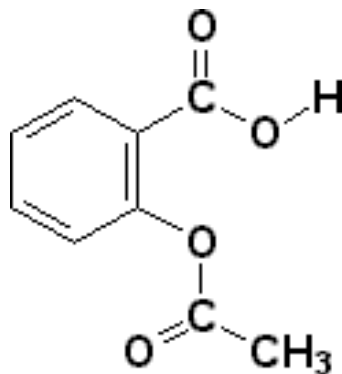
100 A.C.: Cirurgião grego Dioscorides menciona em seus escritos o uso de folhas do *salgueiro* para aliviar a dor.

# Salicina

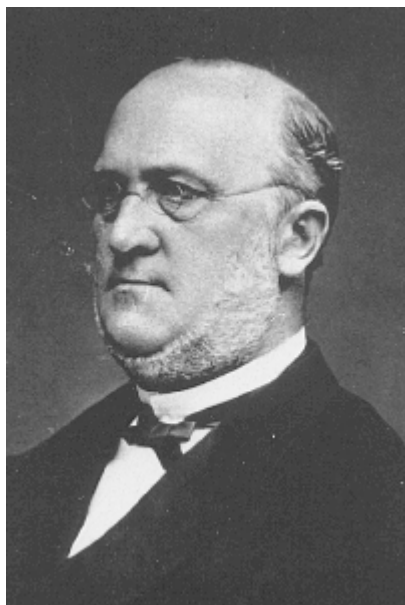


- Idade Média: Europeus param de usar remédios à base de casca de salgueiro, uma vez que o suprimento de cascas é utilizado para fazer vime. Uso do salgueiro para fins medicinais proibida em alguns lugares.
- Após 1500: Nativos da América do Norte aprenderam a fazer remédios para dor à base de salicilatos de cascas de *bétula*.
- 2/07/1793: Edward Stone, um clérigo inglês, relatou para a Royal Society of London, seus experimentos de sucesso envolvendo o uso das cascas do salgueiro para reduzir a febre em 50 de seus pacientes. Enquanto isto, em terras Européias a *quinina* era usada para o tratamento da dor.
- 1828: Johann Büchner em Munique, Alemanha isolou a *SALICINA* pura das cascas de salgueiro. Salicina era a substância da casca do salgueiro responsável pelo alívio das dores. O nome *Salicina* foi derivado de *Salix*, nome latino para *Salgueiro*.
- 1835: Karl Lowig faz ácido salicílico a partir das flores de arbustos do gênero *Spiraea*.
- 1838: Raffaele Piria converte a salicina em Ácido Salicílico. Esta foi a primeira vez que o ácido salicílico foi obtido a partir das cascas do salgueiro em laboratório.

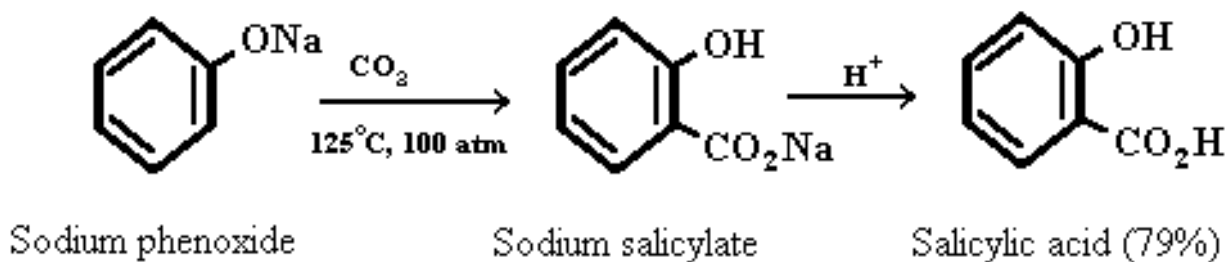
# AAS



- 1853: Charles Frederic Gerhardt first sintetiza pela primeira vez ácido acetilsalicílico, mas falha na compreensão da sua estrutura molecular e sua importância para a humanidade. Seu ácido salicílico não era puro e por isto de uso restrito.



- 1859: H. von Glim também descreve a preparação do ácido acetilsalicílico, também de forma impura.
- Enquanto isto, Herman Kolbe descobre como sintetizar o ácido salicílico a partir do alcatrão. O método que ele empregou ainda é chamado “*Síntese de Kolbe*”.
- 1869: Karl-Johann Kraut repete os experimentos das etapas anteriores e dá as primeiras informações acuradas sobre a estrutura molecular do ácido acetilsalicílico, o éster do ácido salicílico. Como o próprio admitia, suas amostras também não estavam puras.
- 1874: Ácido Salicílico foi preparado industrialmente pela primeira vez usando o método de Kolbe em Dresden, Alemanha. Ele foi vendido como analgésico, mas provocava grave irritação estomacal.



The Kolbe Synthesis



# Ácido Acetilsalicílico

- 1897: Felix Hoffmann, químico das indústrias Bayer na Alemanha, preparou a primeira amostra pura de AAS. Se inicia a história do AAS como conhecida até hoje.
- 1899: Ensaios clínicos indicam os promissores efeitos curativos do AAS. Bayer batiza o AAS de ASPIRINA e a companhia começa a distribuí-la para os médicos (NA FORMA DE PÓ) administrarem em seus pacientes.



- 1900: Bayer introduz o primeiro comprimido de aspirina solúvel em água. Este processo reduz o custo à metade.
- 1915: Aspirina começa a ser utilizada sem prescrição médica.



# Toxicidade da Aspirina

<u>Composto</u>	<u>LD<sub>50</sub> (mg/kg)</u>
-----------------	--------------------------------

Ibuprofeno	1255
------------	------

Naproxeno	1234
-----------	------

Aspirina	1100
----------	------

Ácido Salicílico	500
------------------	-----

Acetaminofeno	338
---------------	-----

Nicotina	230
----------	-----

Cloridrato de Morfina	226
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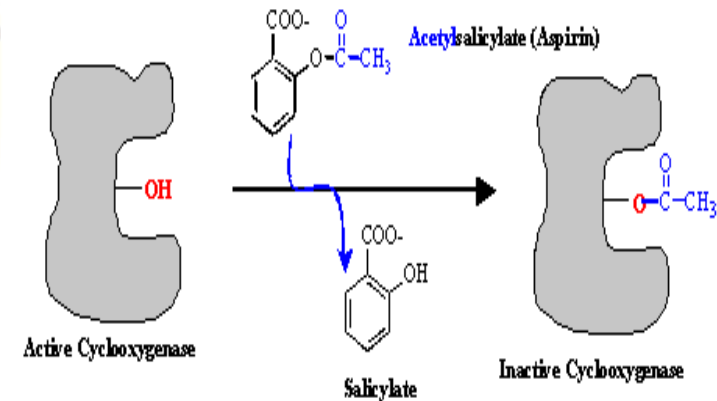
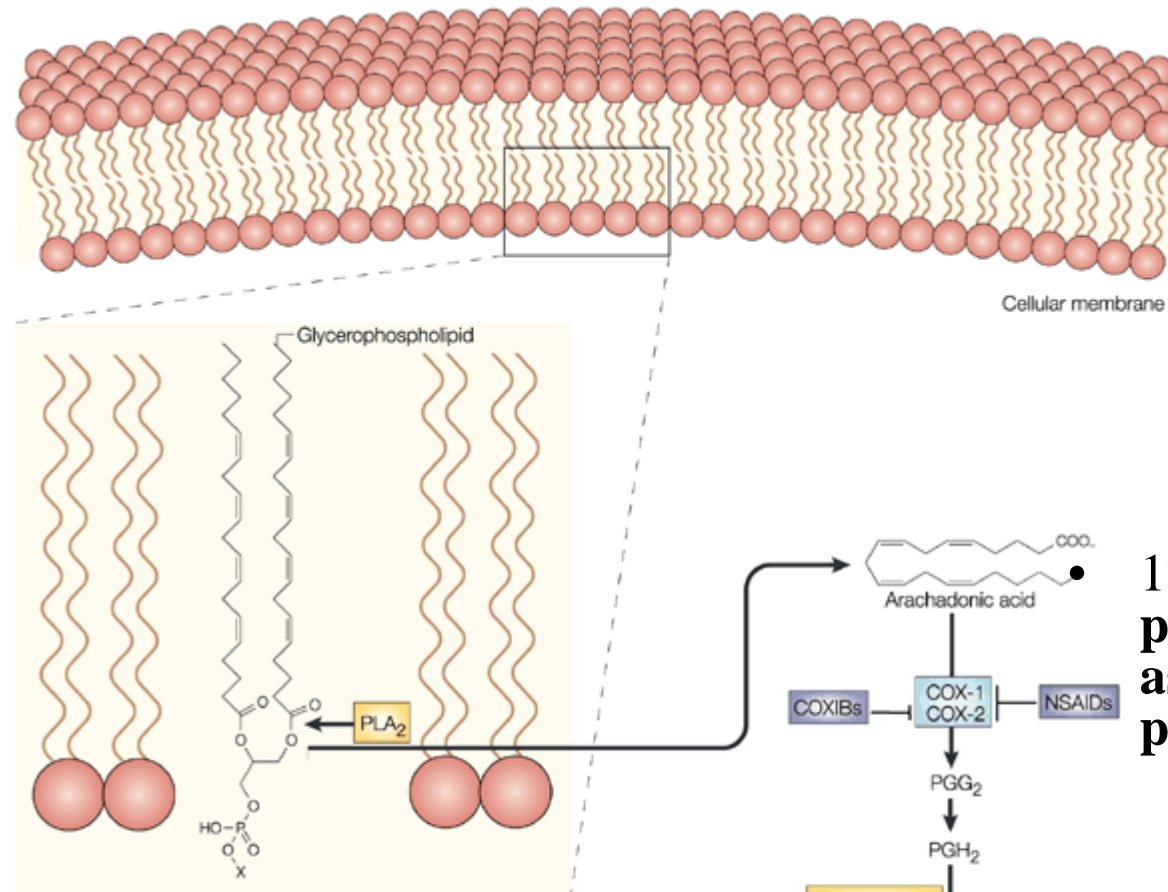
Cafeína	127
---------	-----

Cocaína	18
---------	----

Heroína	1.4
---------	-----

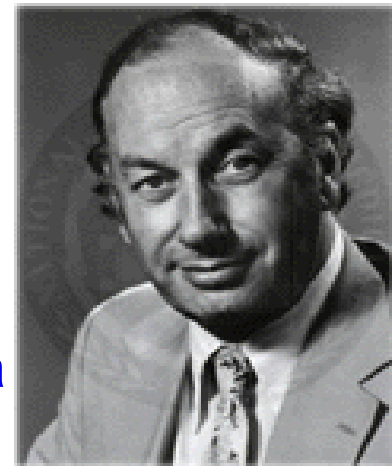
<u>LD<sub>50</sub></u>	<u>Provável Dose letal</u>
< 5	< 0.3 g
5 - 50	0.3 - 3 g
50 - 500	3 - 30 g
500 - 5000	30 - 300 g

# Mecanismo de Ação do AAS



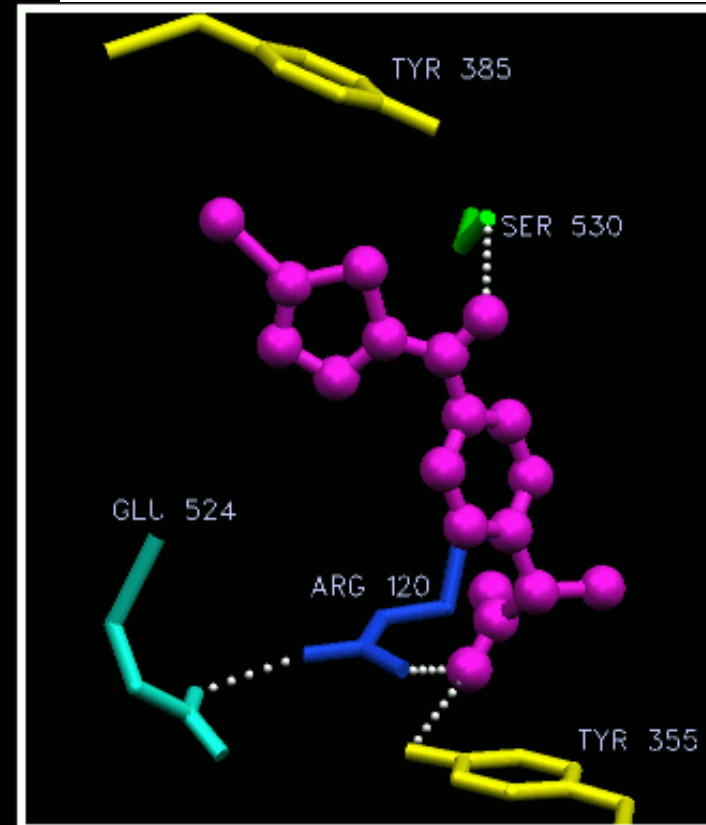
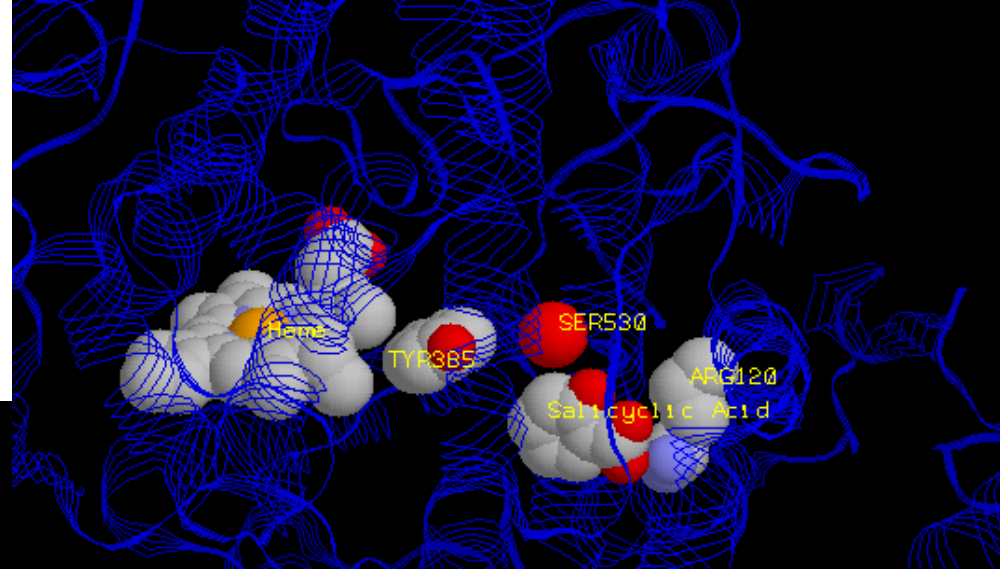
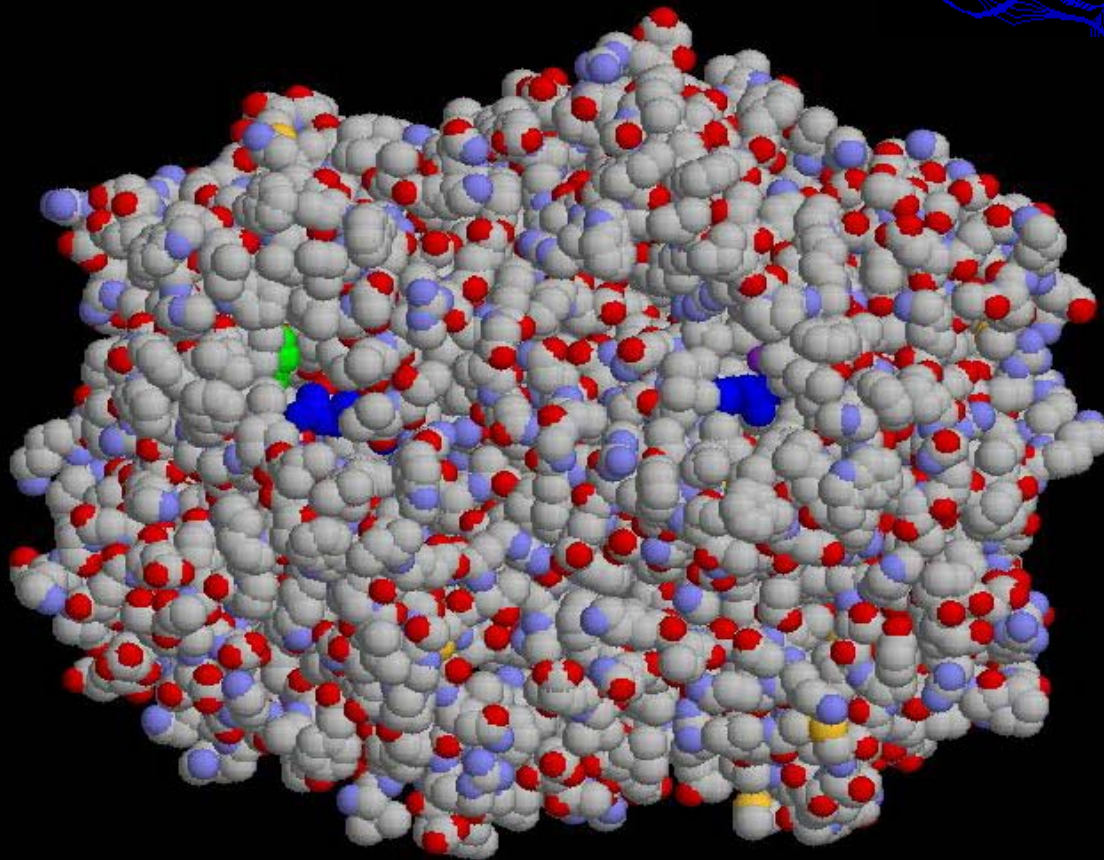
1971: John Vane, British pharmacologist, discovers that aspirin works] by inhibiting the production of prostaglandins.

**Nobel  
Medicina  
1982**



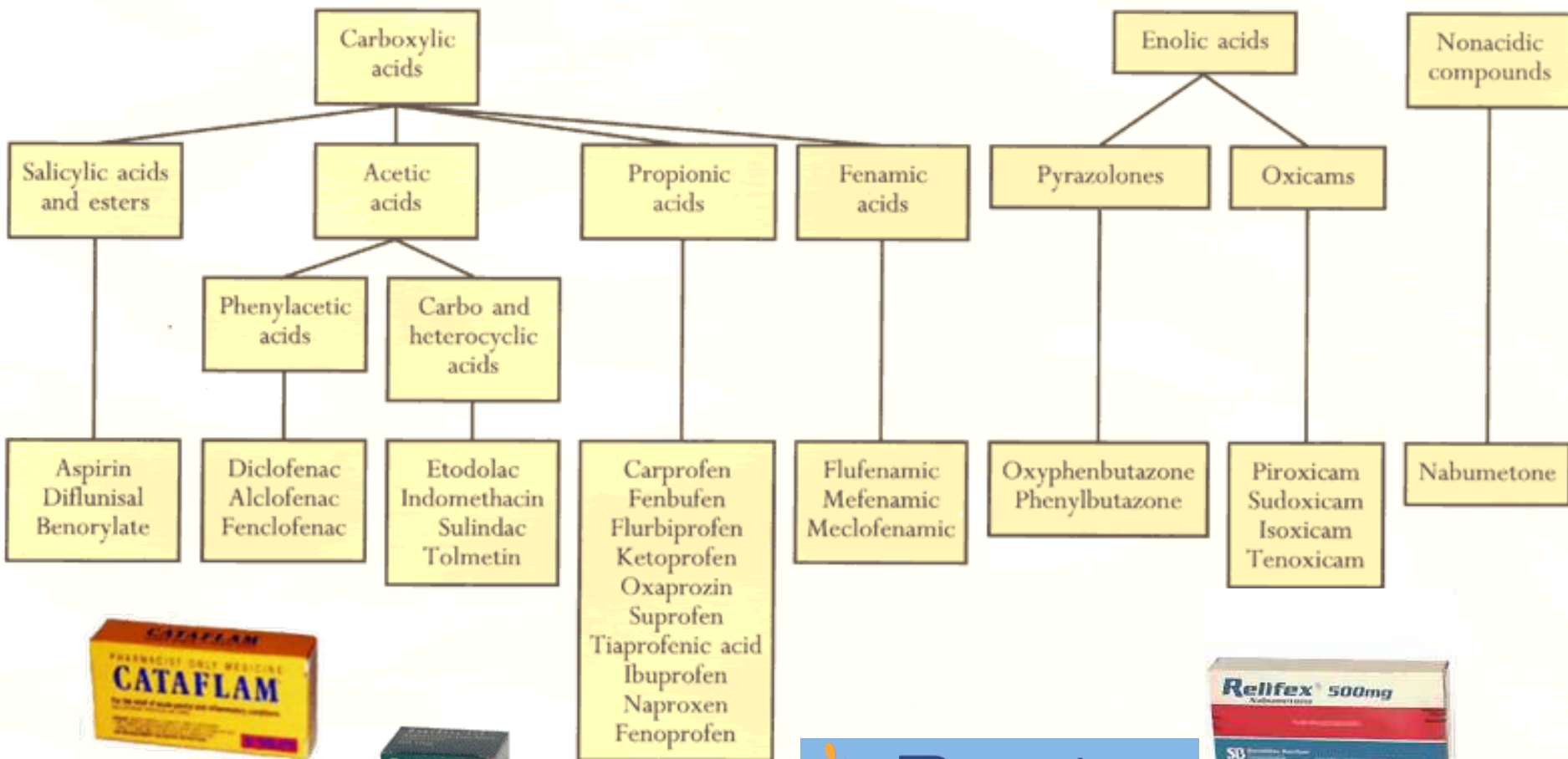
# Modo de Interação com a COX

COX-1

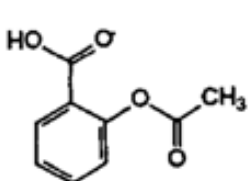




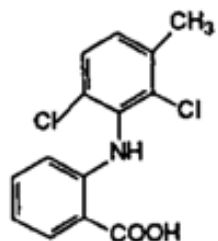
# Classificação dos AINE's



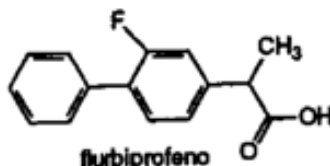
# A Descoberta do Piroxicam



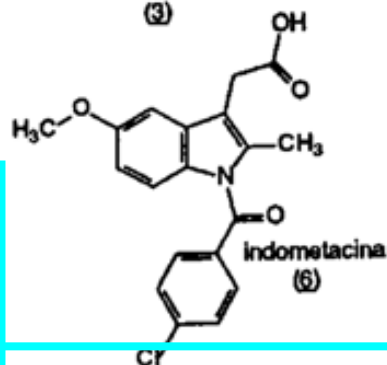
ácido acetil-salicílico (1)



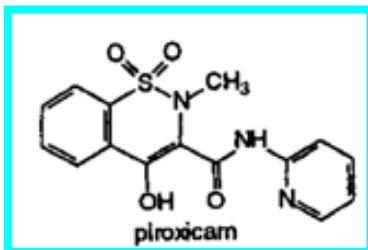
ácido meclofenâmico (2)



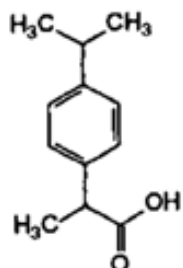
flurbiprofeno (3)



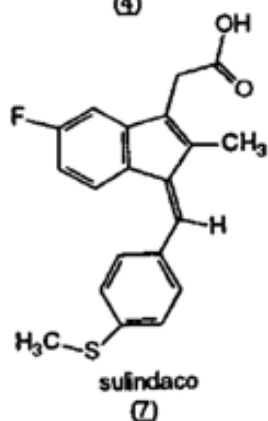
indometacina (6)



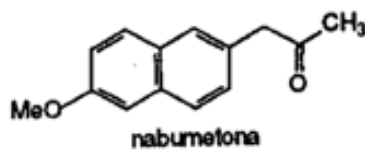
piroxicam (5)



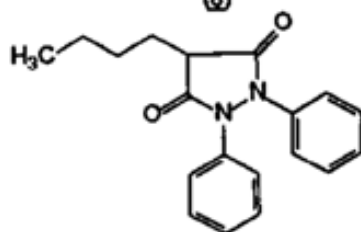
ibuprofeno (4)



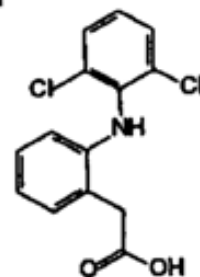
sulindaco (7)



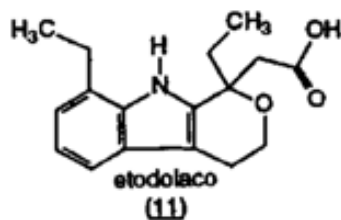
nabumetona (8)



fenilbutazona (10)



diclofenaco (9)



etodolaco (11)



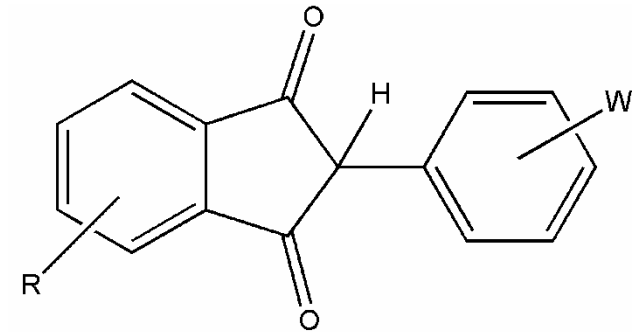
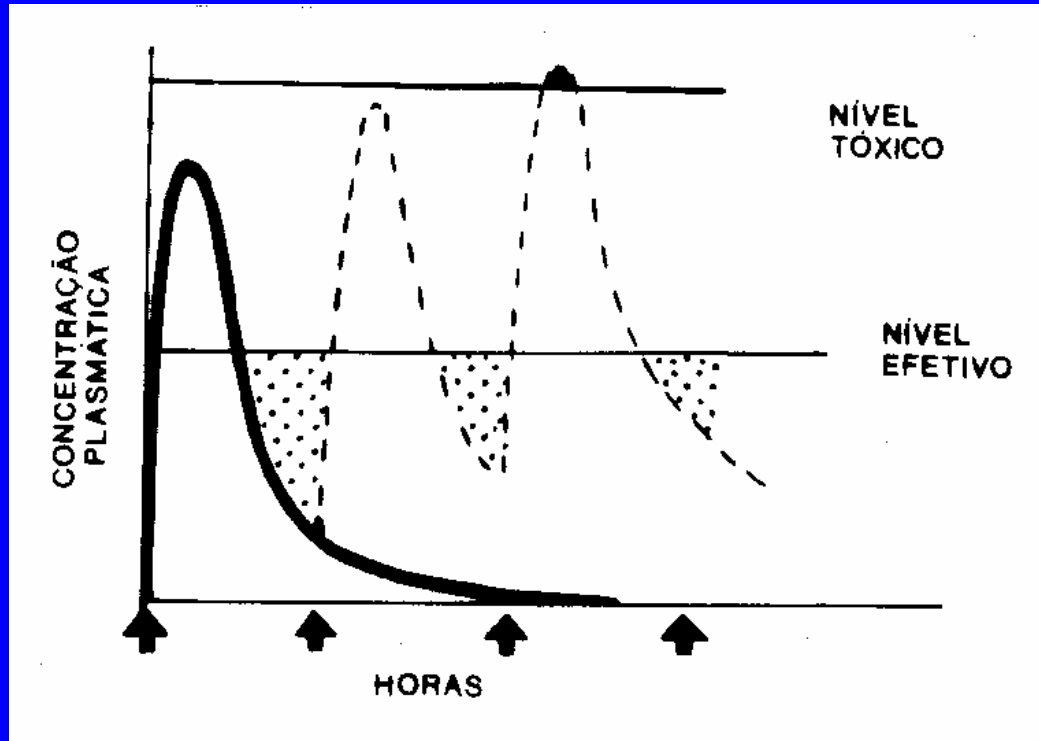
Joseph G. Lombardino

1971



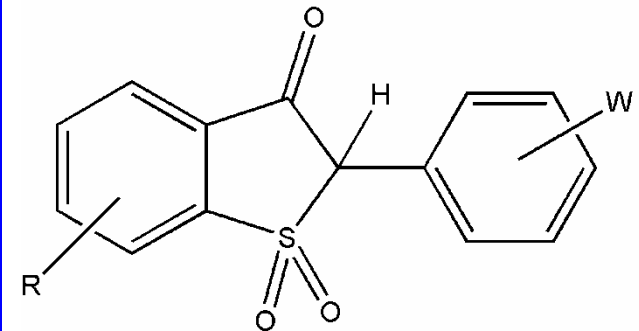


# O Desafio: Obter um AINE ácido, porém de natureza não-carboxílica



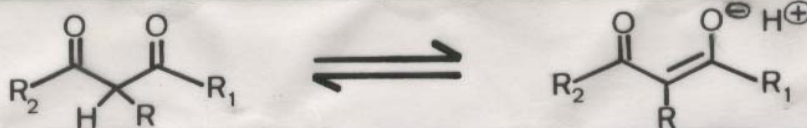
## 1,3-Indanodionas

AI + Anticoagulante (Inibem a síntese hepática de Protrombina)

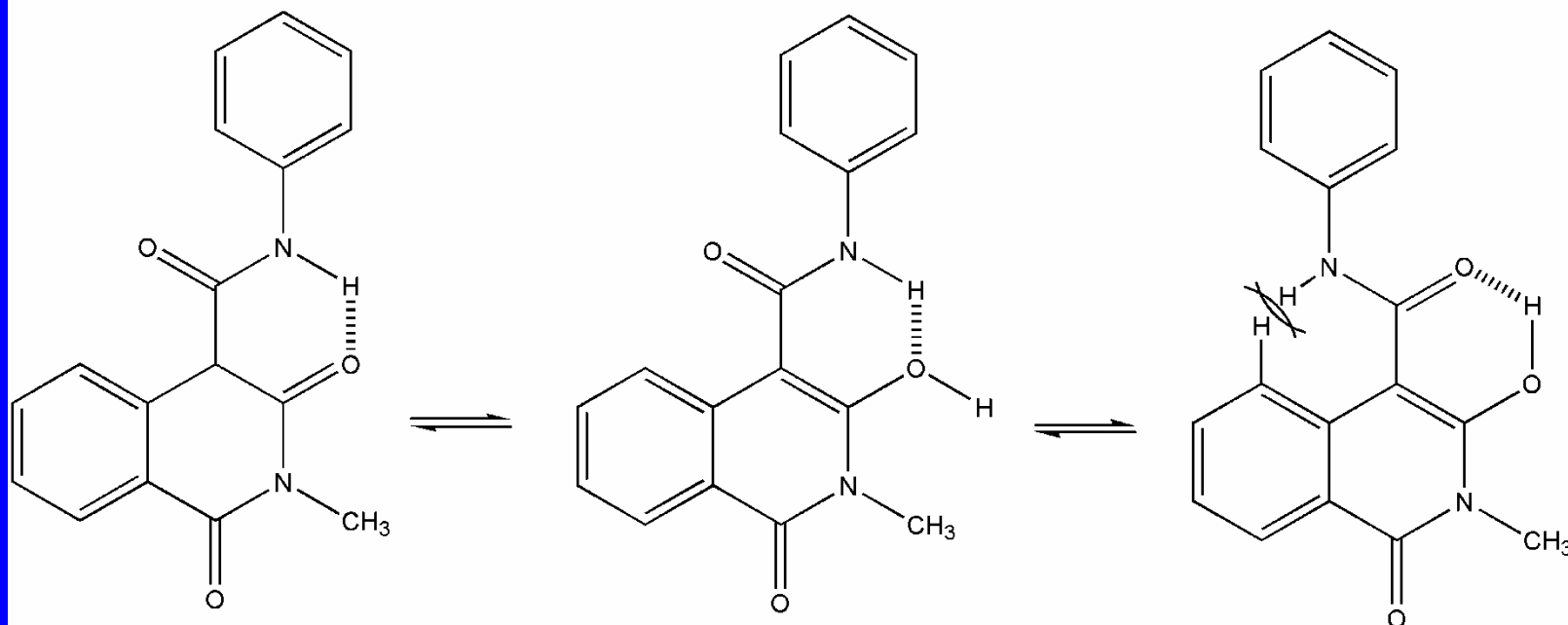
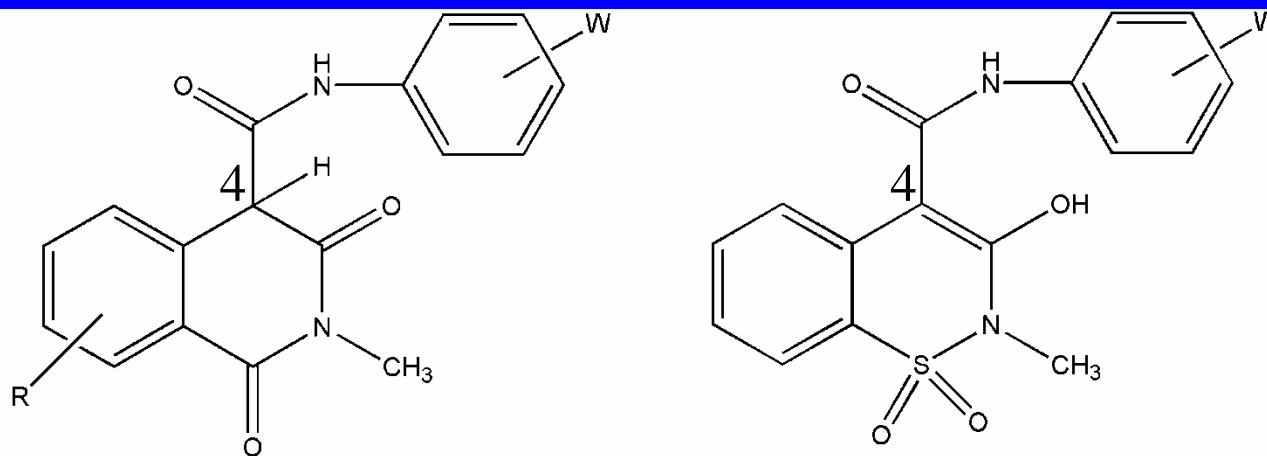


## Benzotiofen-3-onas

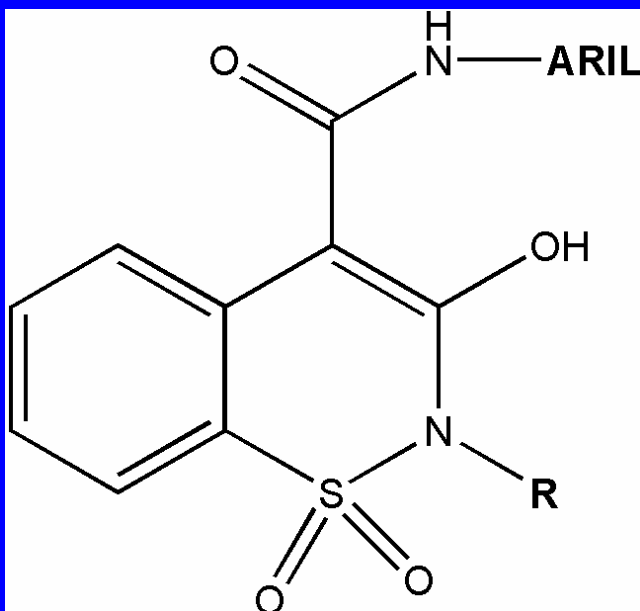
Atividade Antiinflamatória  
Reduzida Ação Anticoagulante  
**Nefrotoxicidade em cães e ratos**



# Isoquinolinas e 1,2-Benzotiazinas-4-carboxamidas

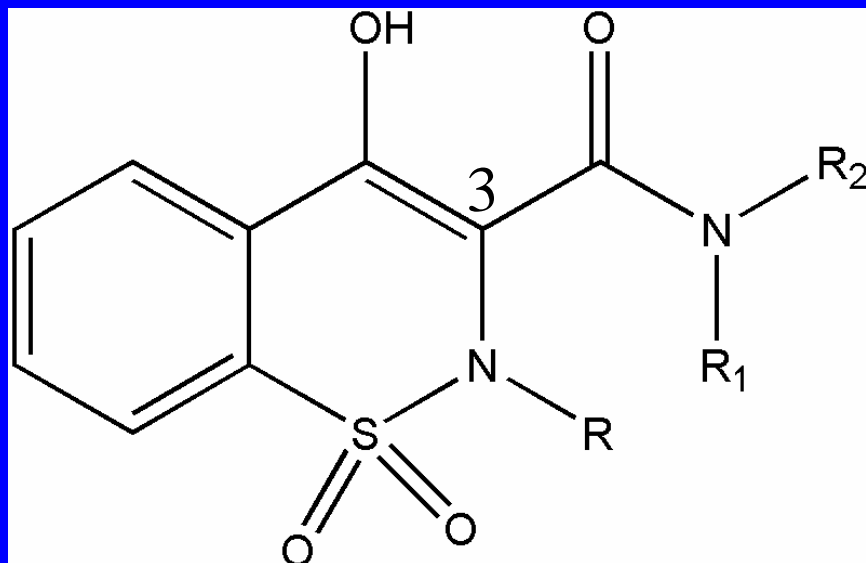


# SAR das 1,2-Benzotiazinas-4-carboxamidas



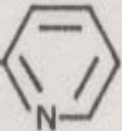


COMPOSTO N°	R	Aril	$\sigma_p$ HAMMET	ATIVIDADE A.I. RELATIVA
25 a	CH <sub>3</sub>		0.0	++
25 b	CH <sub>3</sub>		0.23	+++++
25 c	CH <sub>3</sub>		0.78	+++++
25 d	CH <sub>3</sub>		0,5	-
25 e	CH <sub>3</sub>		0,72	+
25 f	H		0,0	-
25 g	CH <sub>3</sub>		(-)	-

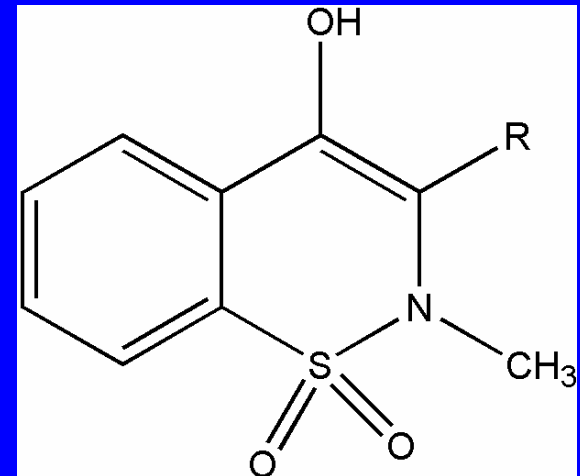
# Derivados 1,2-Benzotiazínicos 3-Carboxamidas





COMPOSTO Nº	R	R <sup>1</sup>	R <sup>2</sup>	ATIVIDADE AI RELATIVA
26	CH <sub>3</sub>		H	+++++
27	H		H	++
28	CH <sub>2</sub> CH <sub>3</sub>		H	+
29	CH <sub>3</sub>	-CH <sub>2</sub> CH <sub>2</sub> φ	H	-
30	CH <sub>3</sub>		H	+
31	CH <sub>3</sub>		H	++++
32	CH <sub>3</sub>		H	++
33	CH <sub>3</sub>		CH <sub>3</sub>	-

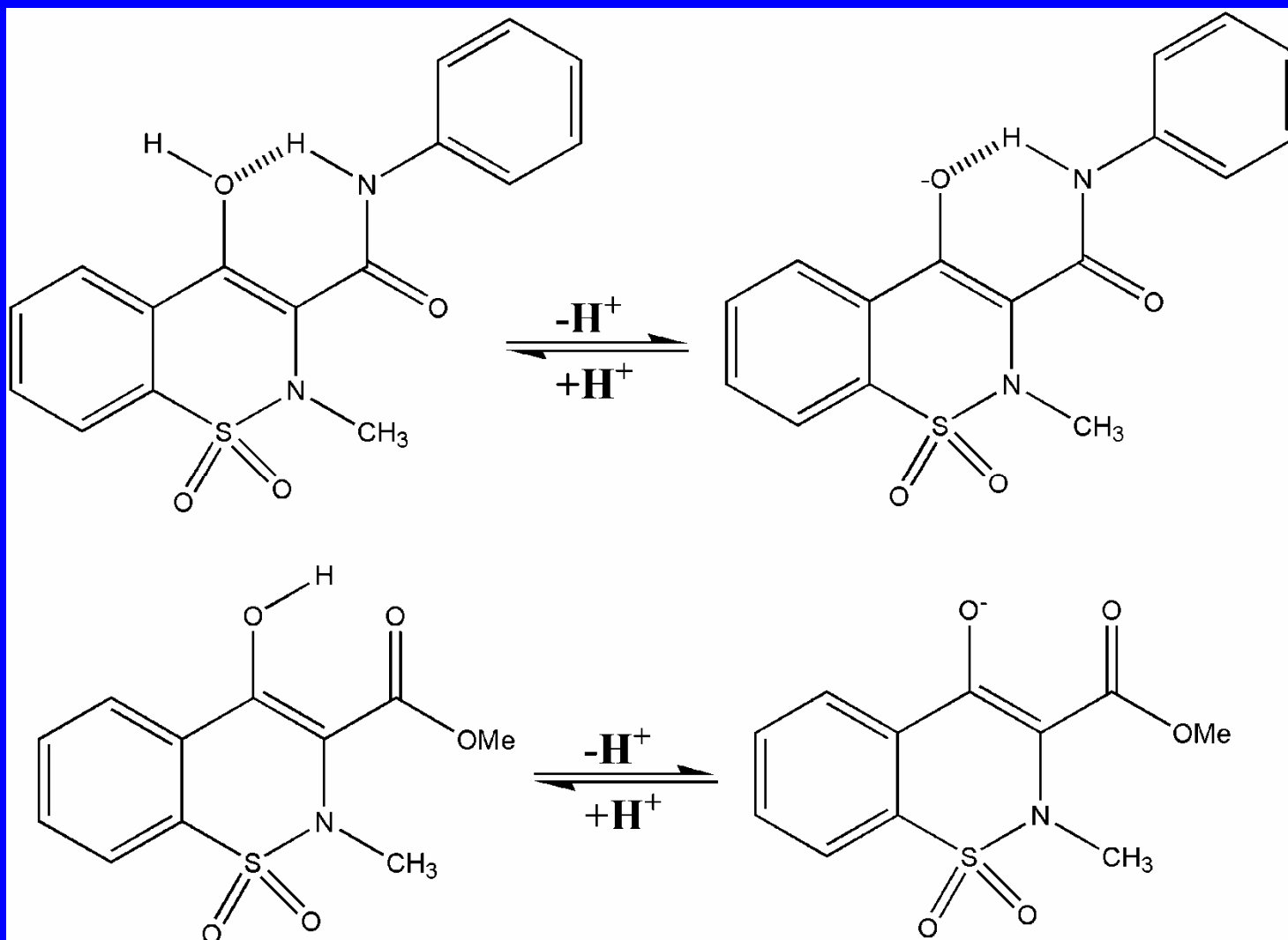
# Acidez X Atividade Antiinflamatória de Derivados 1,2-Benzotiazínicos 3-Carboxamidas

COMPOSTO N°	R	INIBIÇÃO DA SÍNTESE DE PG's IC <sub>50</sub> (μM)
3	CONH- 	0,76
26	CONH- 	3,6
34	CONH- 	22
19	COOCH <sub>3</sub>	>50
35	COOH	>100
36	H	>100



Composto n°	R	pKa
26	CONH- 	7,3
19	COOMe	8,4
33	CONCH <sub>3</sub> - 	9,8

# Ionização de de Derivados 1,2-Benzotiazínicos 3-Carboxamidas





# Propriedades Fisico-químicas dos Oxicams

Coeficiente de partição  
(Lipofilicidade)

1,8  
(octanol/tampão pH 1,4)

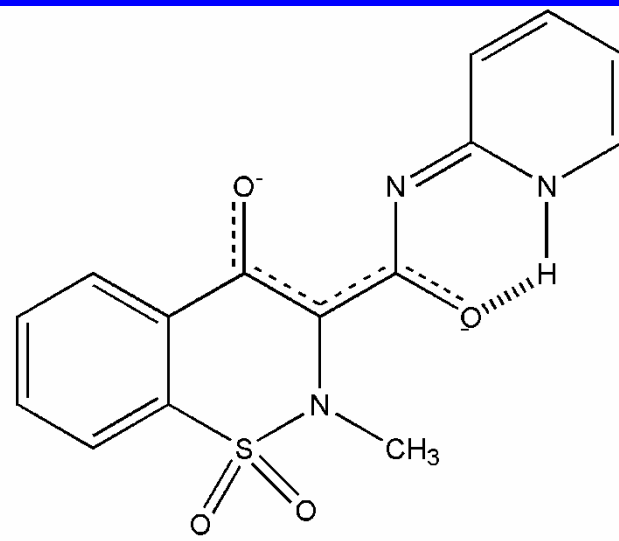
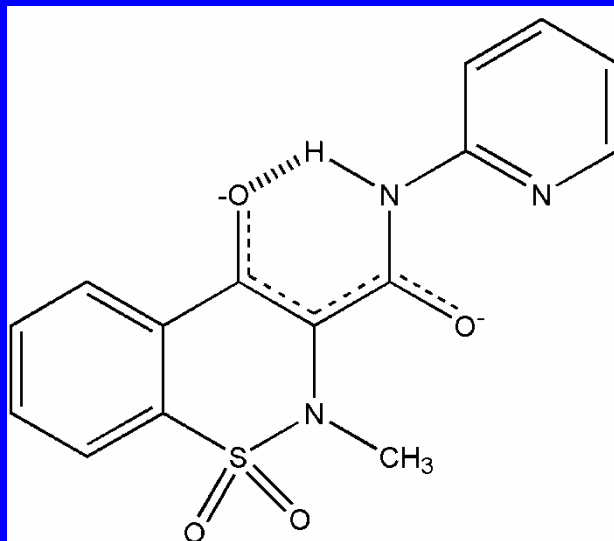
pKa  
(Acidez)

6,3  
(dioxana/água (2:1))

Ligação à proteínas  
plasmáticas humanas

99%  
(em conc. de 5µg/ml)

Tautomeria  
2-aminopiridina  
/2-iminopiridona



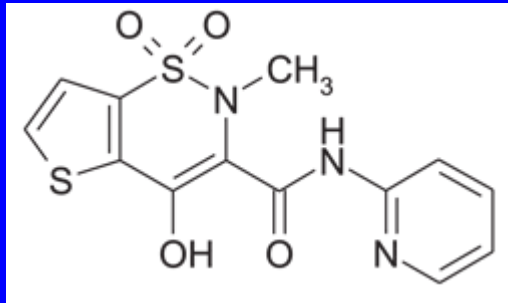
## Tempo de Meia-Vida dos Oxicams

AINE	Tempo de Meia-Vida (h)			
	Ratos	Cães	Macacos	Homem
Fenilbutazona	6	6	7	72
Indometacina	4	0,3	0,3	2
Ibuprofeno	1	2,5	---	3
Naproxeno	5	0,3	1,9	13,9
Piroxicam	16	45	5	45

# Potência Relativa no Modelo de CIRPE

AINE	Potência Anti-Edema Relativa (Ratos)
Fenilbutazona	2
Indometacina	10
Ibuprofeno	2
Naproxeno	3
Piroxicam	20

# Índice de Ulcerogenicidade



Tenoxicam - Roche



AINE	UD <sub>50</sub> /ED <sub>50</sub>
Aspirina	1,6
Fenilbutazona	1,8
Diclofenaco	3,1
Indometacina	9,2
Piroxicam	10,9
Tenoxicam	50,0

# Comparação das Doses Clínicas de Alguns AINES

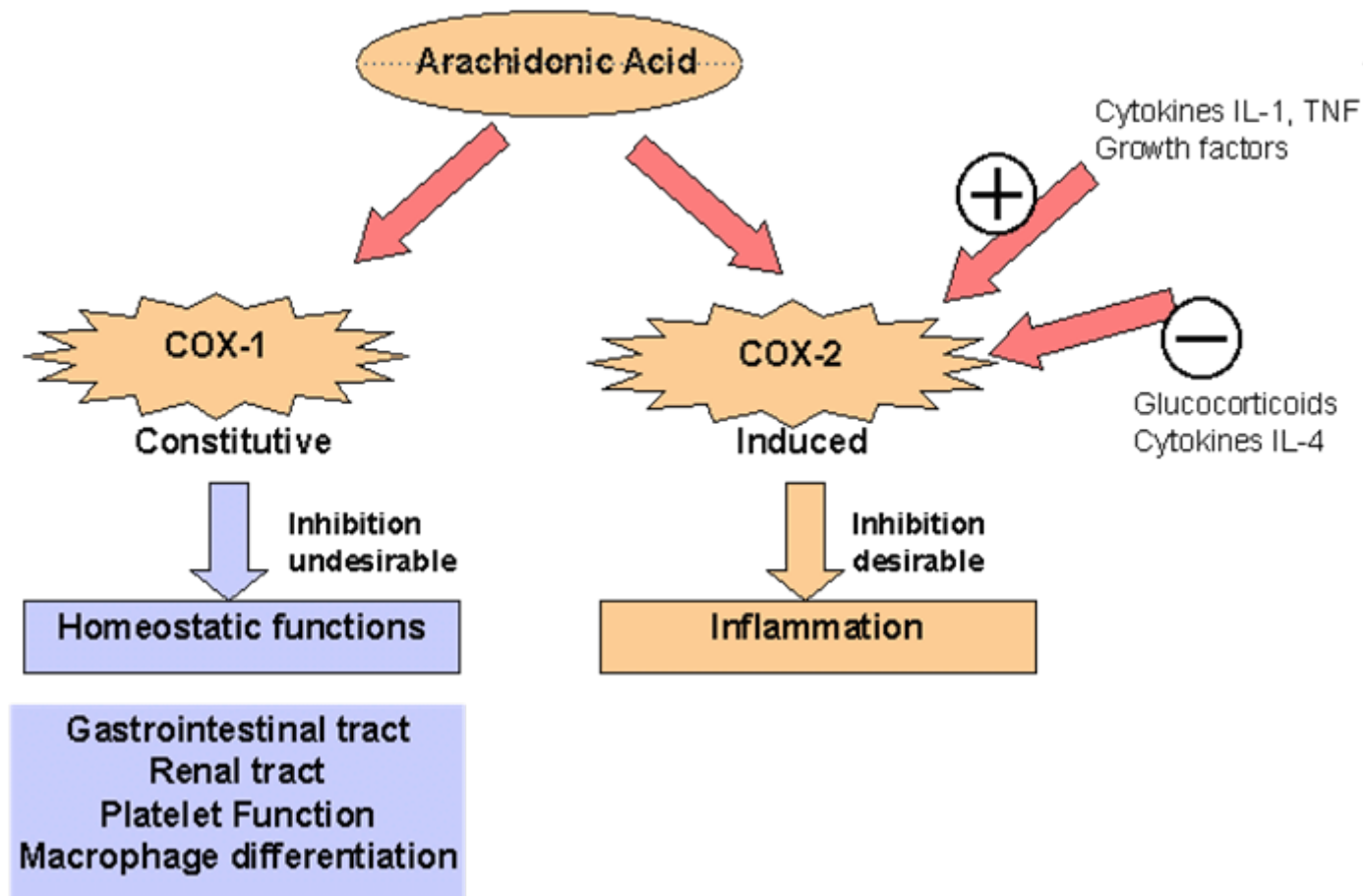
Fármaco (ano da patente)	Acidez (pKa)	DOSE CLÍNICA DIÁRIA		VENDAS <sup>3</sup> (a)	
		Número	Quant. total (mg)	1989	1993
ASPIRINA (-----)	3,5	4	3000-4800	----	----
INDOMETACINA (1964)	4,6	3	75-150	69	39
IBUPROFEN (1968)	5,3	3-4	1600-2400	106	77
FENILBUTAZONA (1951)	4,6	3	300-400	----	----
PIROXICAM (1971)	6,3	1	20	248	280

(a) Consumo mundial em 1989 e projeção de vendas em 1993, em milhões de dólares americanos.

# O conceito atual: Inibição Seletiva da COX-2

◆ 1990: Masferrer hipotetizou a existência de duas diferentes isoformas de ciclooxigenase, baseado na evidência que os AI esteróides inibem o aumento da atividade COX induzida por LPS de bactérias em macrófagos, sem afetar a produção basal de PG's e LT's.

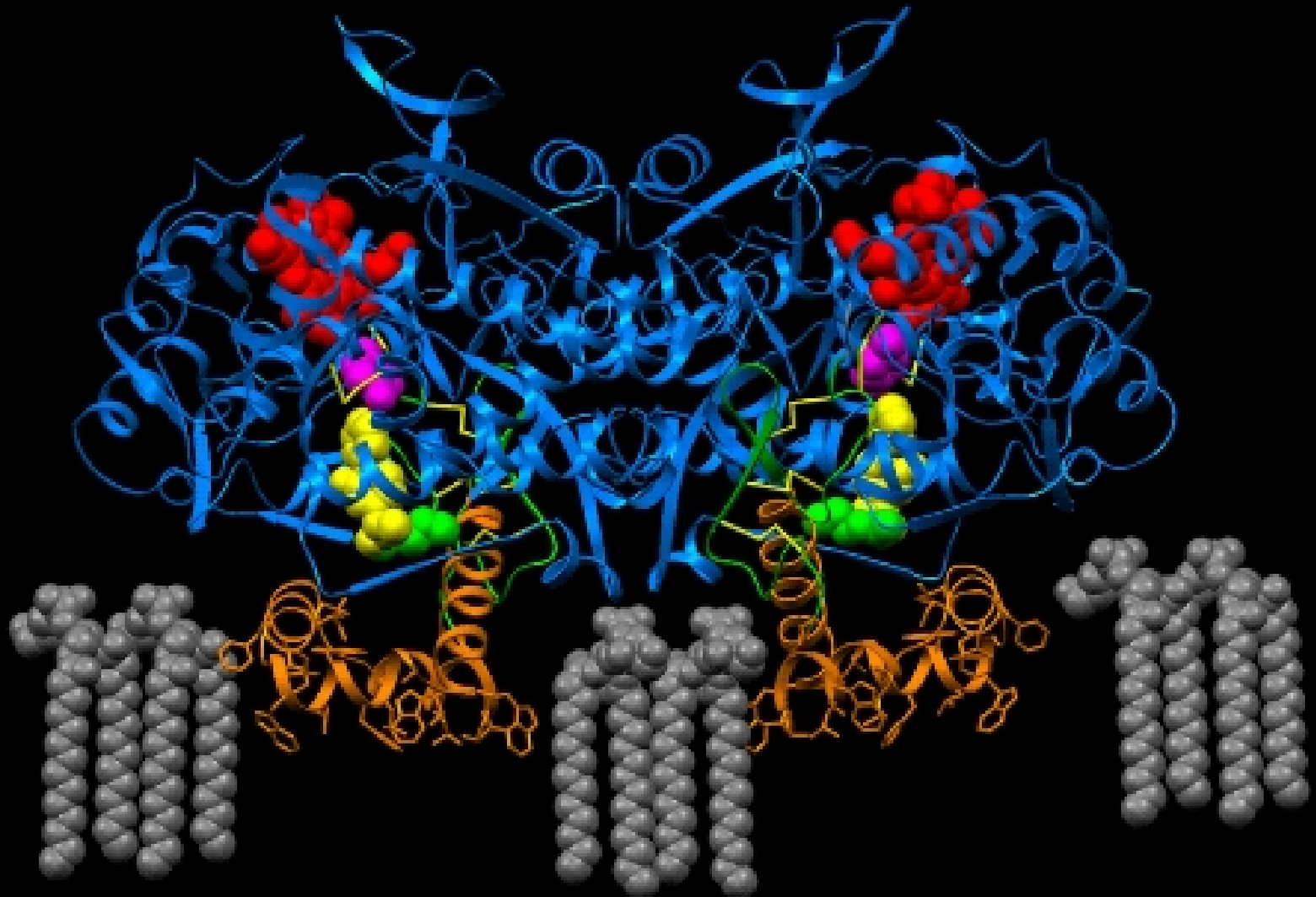
◆ Esta evidência levou ao isolamento da enzima COX-2 humana em 1991.



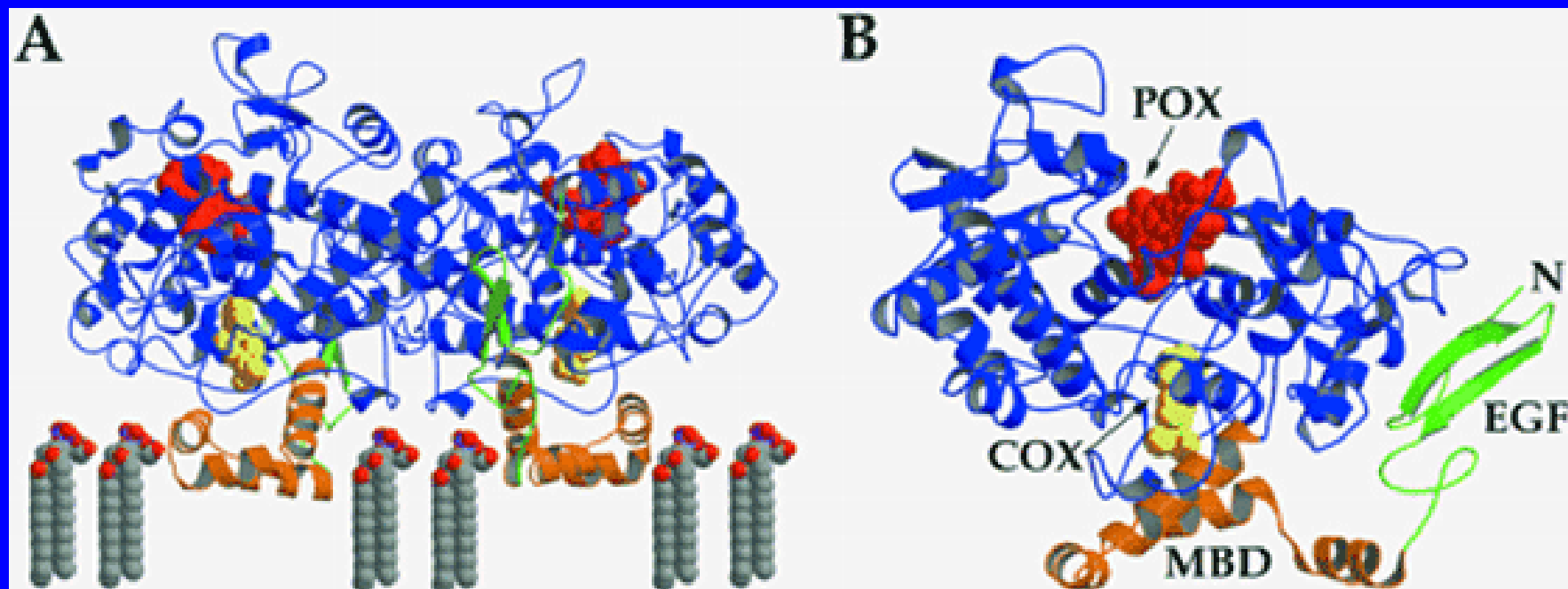


# PGHS-1 (COX-1)

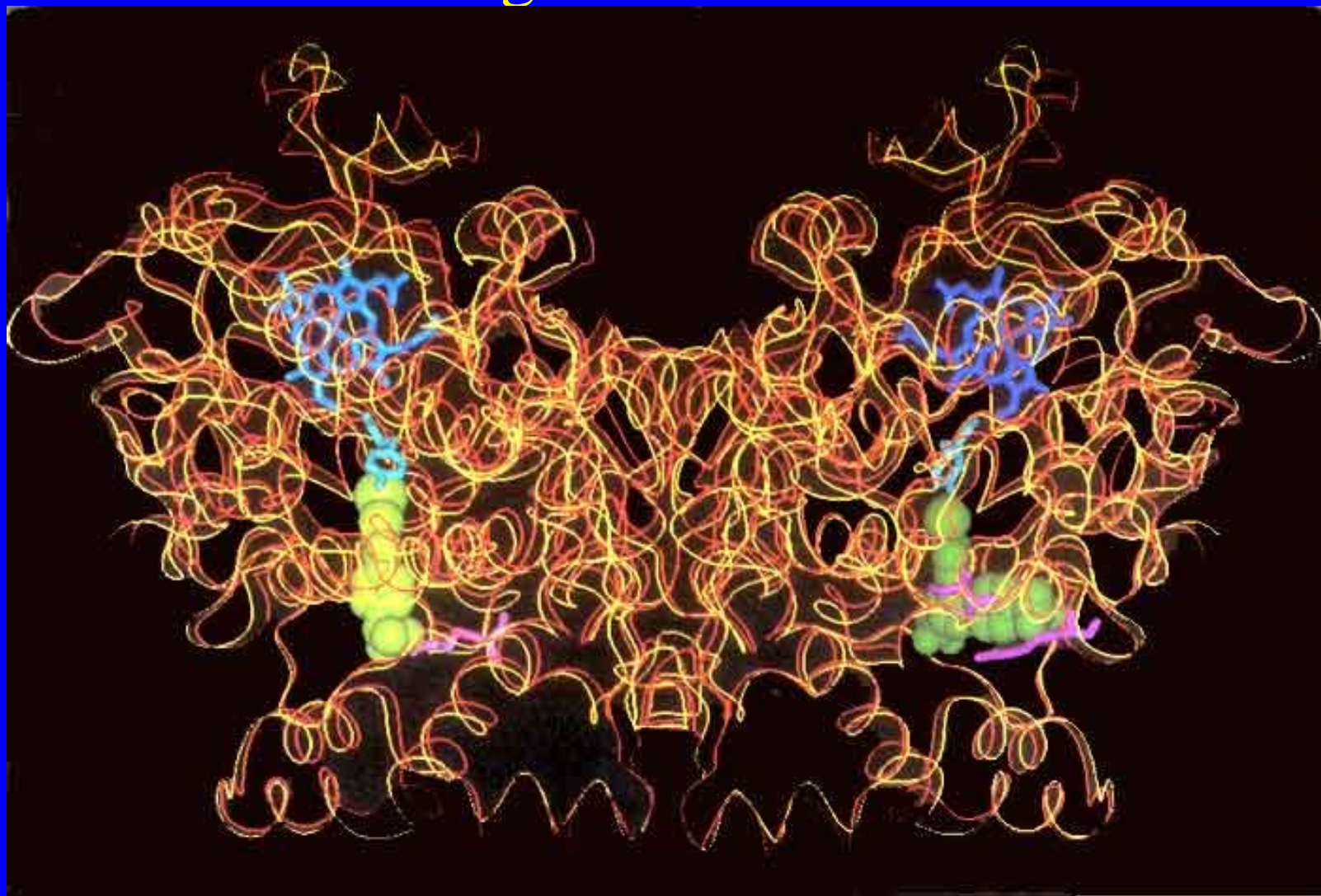
ER lumen



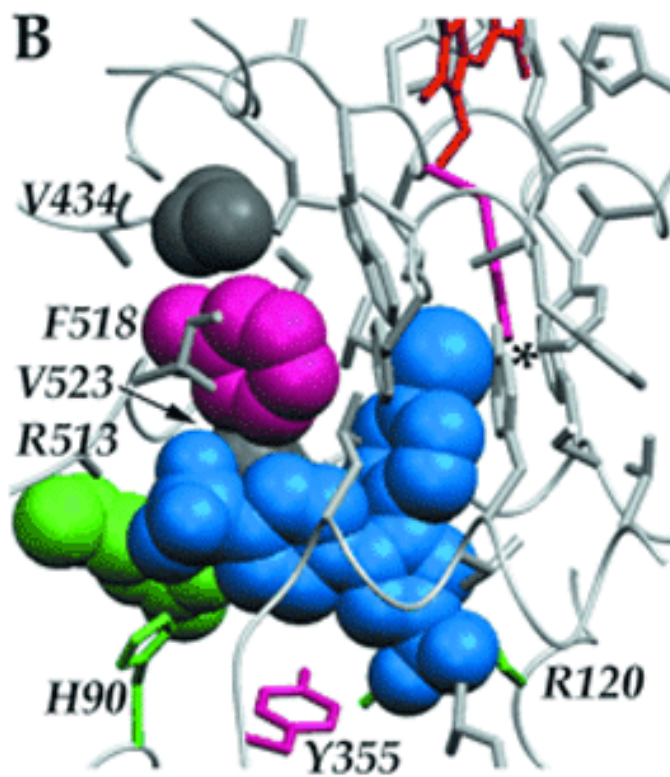
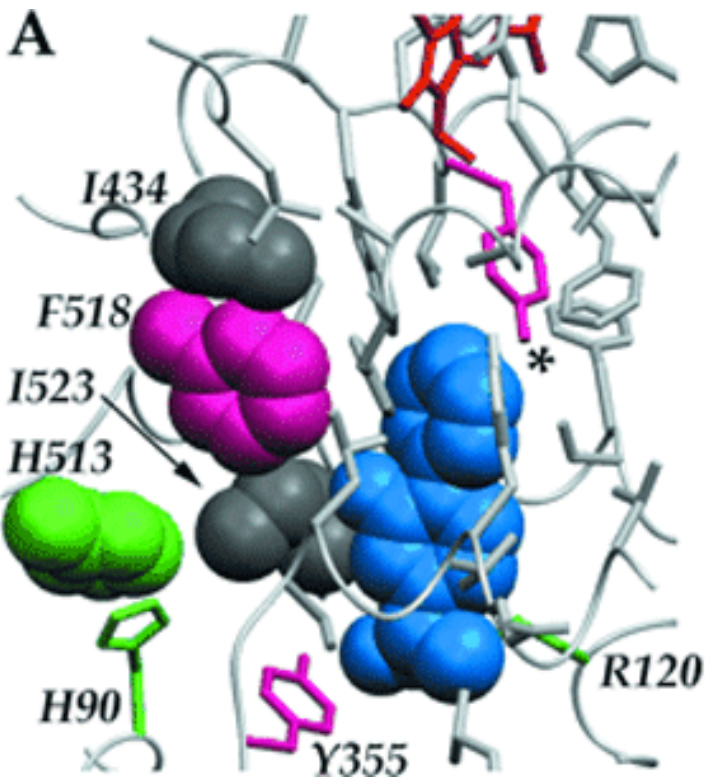
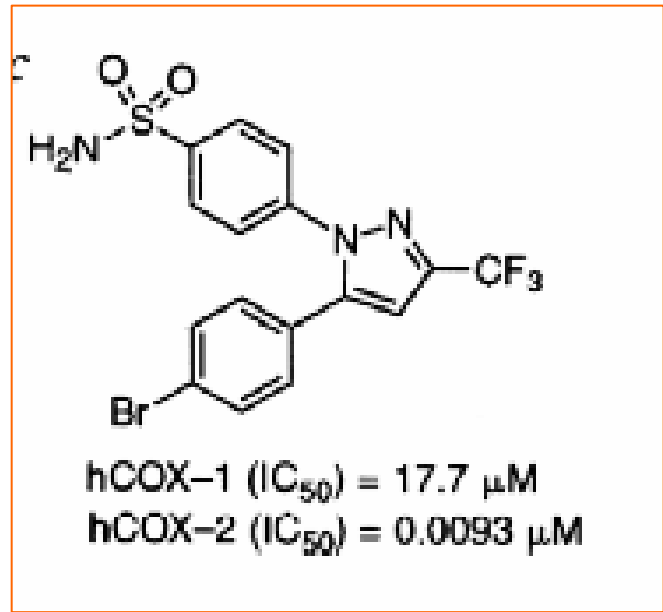
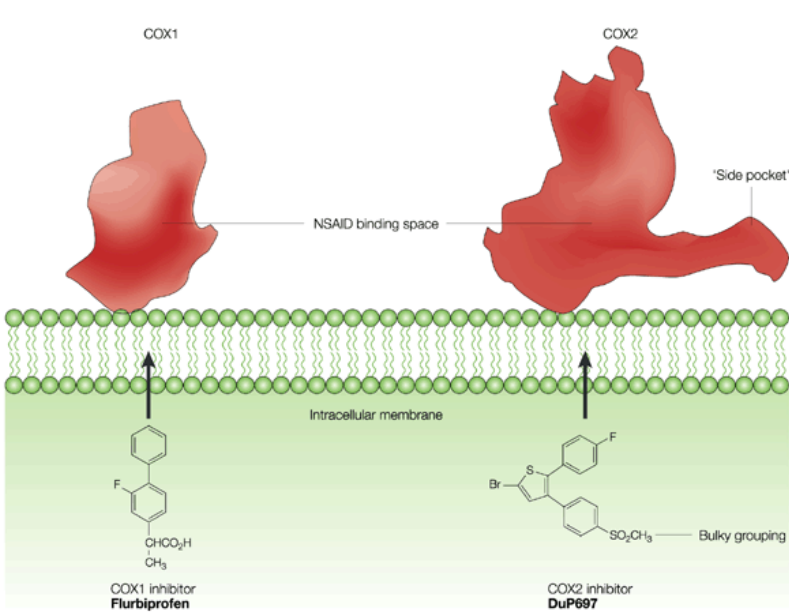
# PGHS-2 (COX-2)



# Homologia COX-1/COX-2



COX-1 ovina (dímero); COX-2 murina (dímero); Flurbiprofeno;  
Heme e Tir-385; Arg-120; SC-558; Val-509 e Arg-499.



Razões  
Estruturais  
da  
Seletividade  
COX-1  
/COX-2

# Inibidores Seletivos de Ciclooxigenase 2: Sulidos



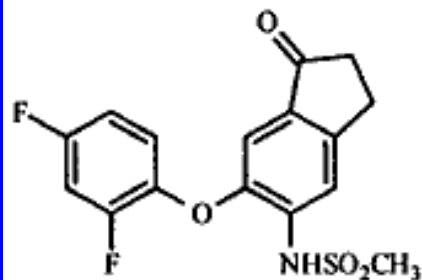
(49) **Nimesulida**

$\text{IC}_{50}$  PGHS-1 > 100  $\mu\text{M}$   
 $\text{IC}_{50}$  PGHS-2 = 0.07  $\mu\text{M}$   
 SI = 1,400



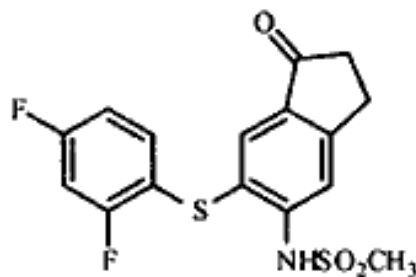
NS-398 (50)

$\text{IC}_{50}$  PGHS-1 > 100  $\mu\text{M}$   
 $\text{IC}_{50}$  PGHS-2 > 0.05  $\mu\text{M}$   
 SI > 2,000



Flosulide (51)

$\text{IC}_{50}$  PGHS-1 = 72.3  $\mu\text{M}$   
 $\text{IC}_{50}$  PGHS-2 = 0.015  $\mu\text{M}$   
 SI = 5,000



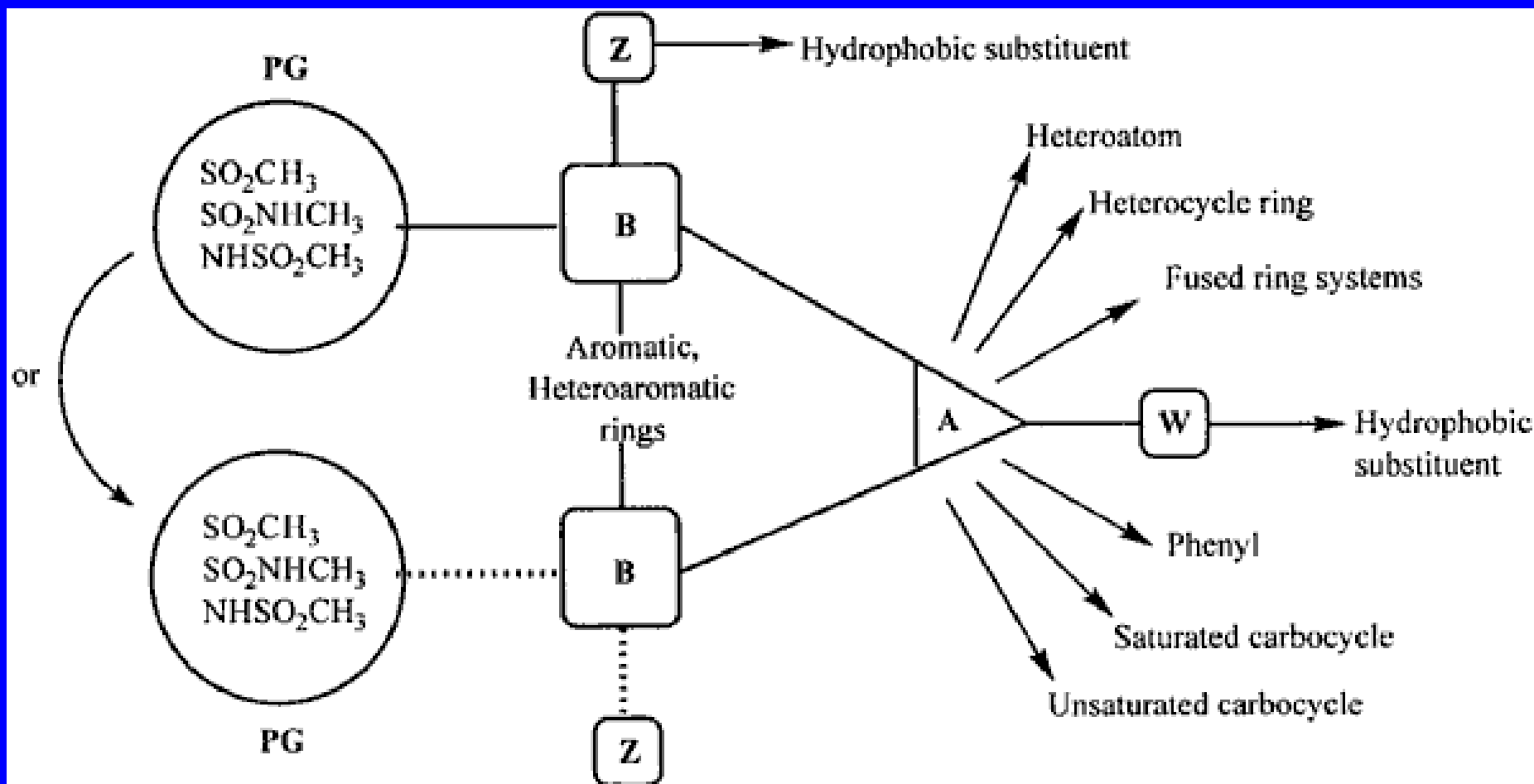
L-745,337 (52)

$\text{IC}_{50}$  PGHS-1 > 50  $\mu\text{M}$   
 $\text{IC}_{50}$  PGHS-2 = 0.023  $\mu\text{M}$   
 SI = 2,100



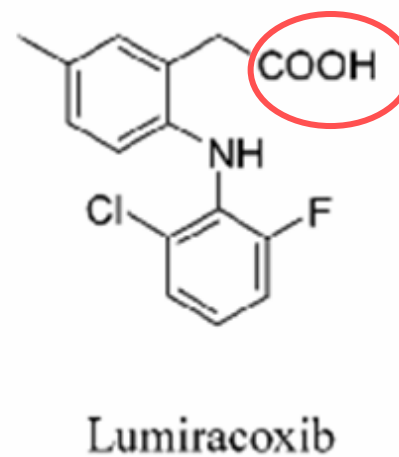
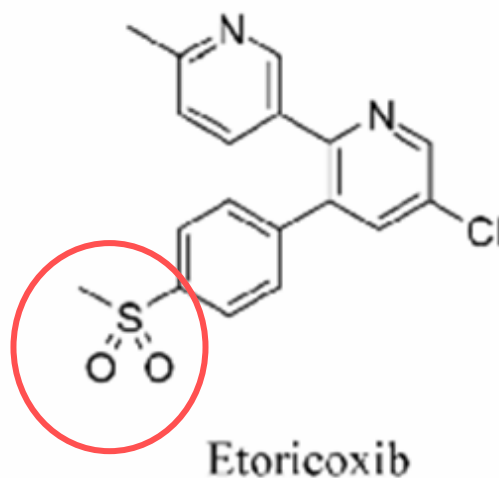
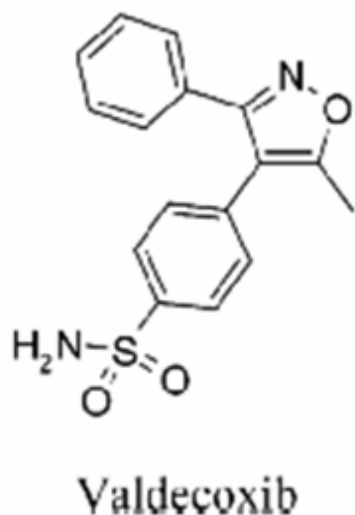
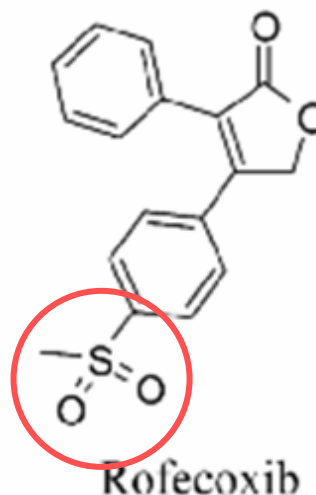
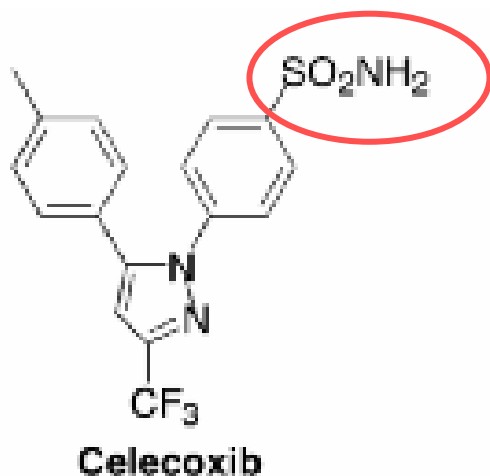


# Derivados Diaril-Heterocíclicos: SAR

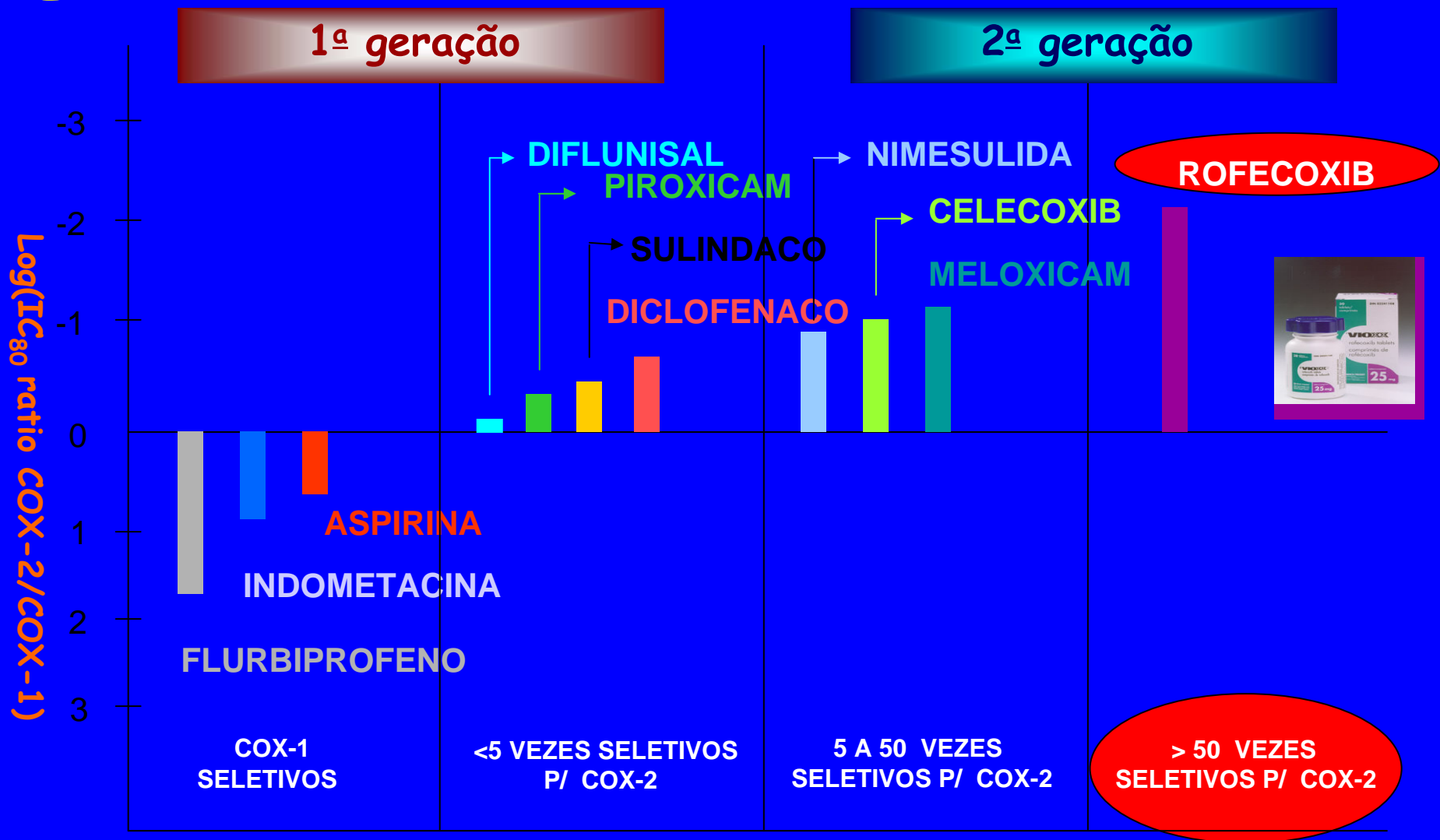




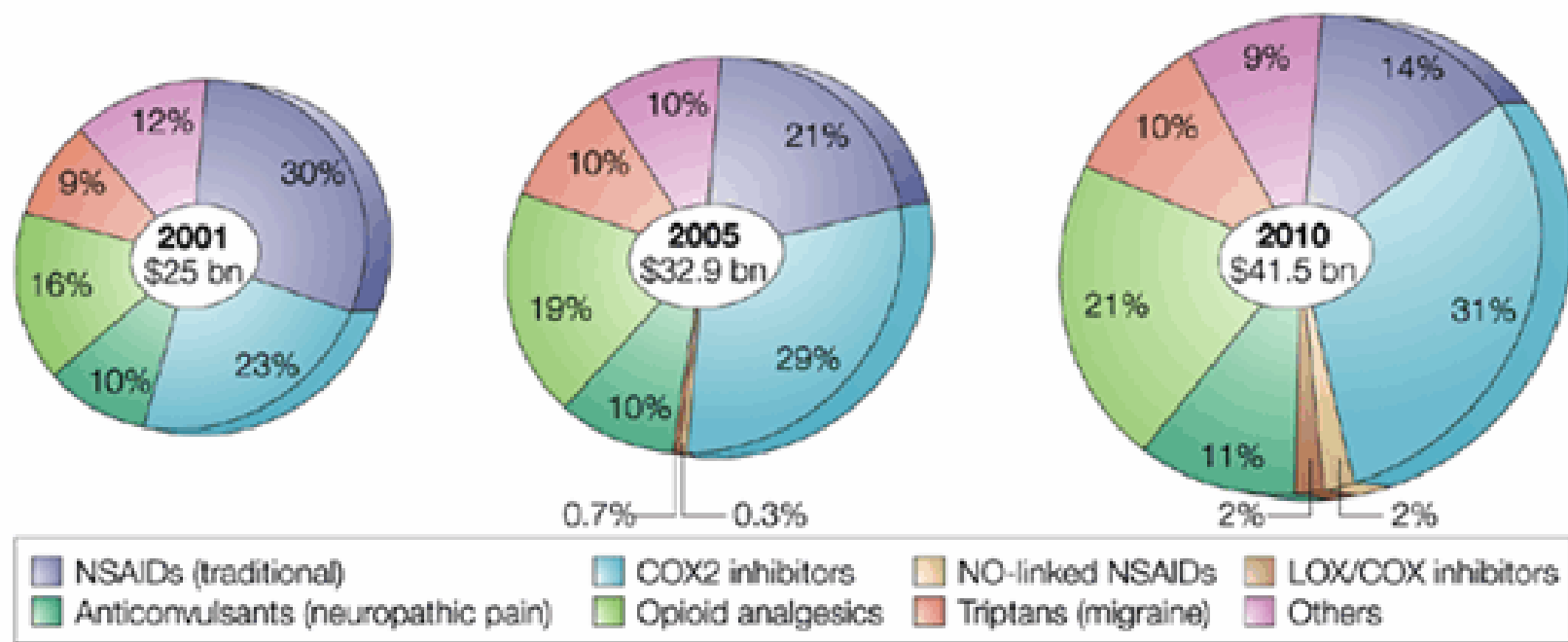
# Principais Inibidores de COX2 Diaril-Heterocíclicos



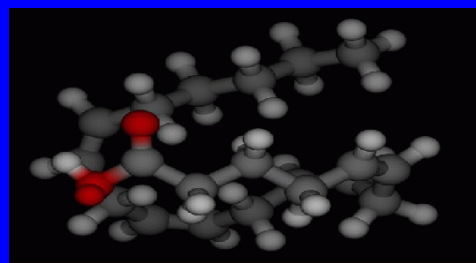
# Seletividade dos Agentes Antiinflamatórios não Esteroidais



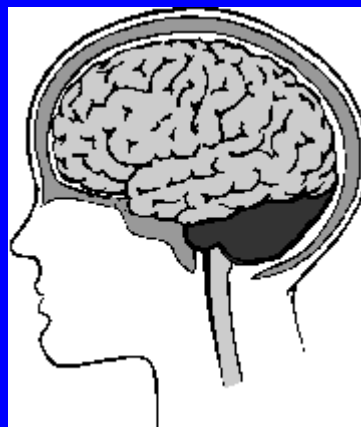
# O Mercado dos Inibidores Seletivos da COX-2



# Cascata do AA e o SNC



Gene  
COX-1



Gene  
COX-2



COX-1  
Constitutiva

COX-3  
Constitutiva

COX-2b?  
Induzida

COX-2  
Constitutiva

COX-2  
Induzida

AINEs clássicos

Acetaminofeno

Seletivos COX-2

Paracetamol

Trato gastrointestinal ( $\text{PGE}_2$ ,  $\text{PGI}_2$  – proteção da mucosa)

Rins ( $\text{PGE}_2/\text{I}_2$  – fluxo sangüíneo)

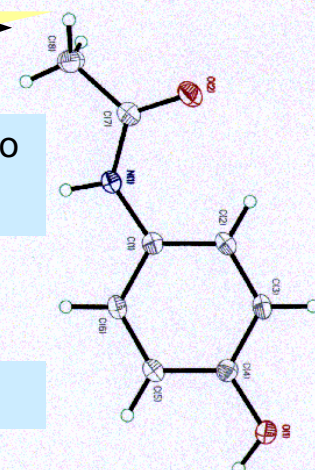
Rins ( $\text{PGE}_2$  – balanço hídrico)

Circulação ( $\text{PGI}_2$ )

Plaquetas ( $\text{TXA}_2$  – agregação)

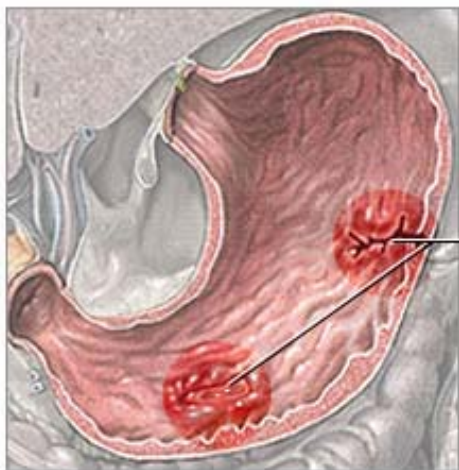
SNC ( $\text{PGE}_2$  – dor, febre)

Tecido inflamado  
(e.g.  $\text{PGE}_2$ )



# Definindo uma Fisiopatologia:

## Úlcera Péptica

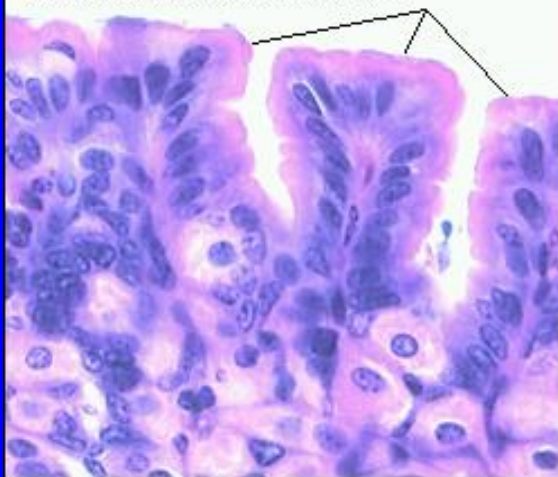


Úlceras  
estomacales



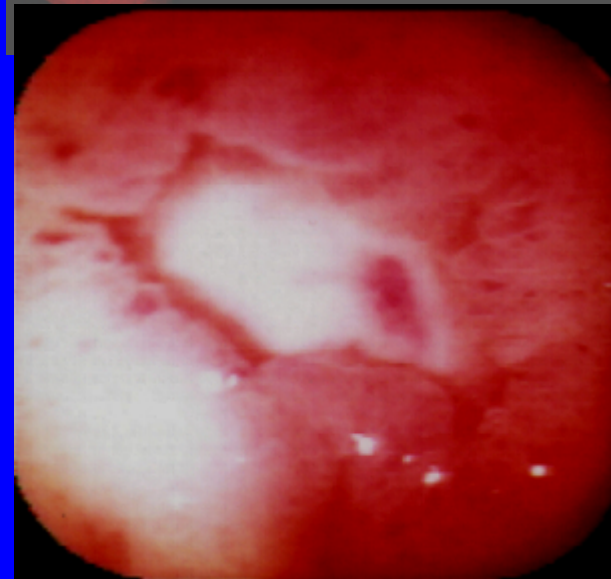
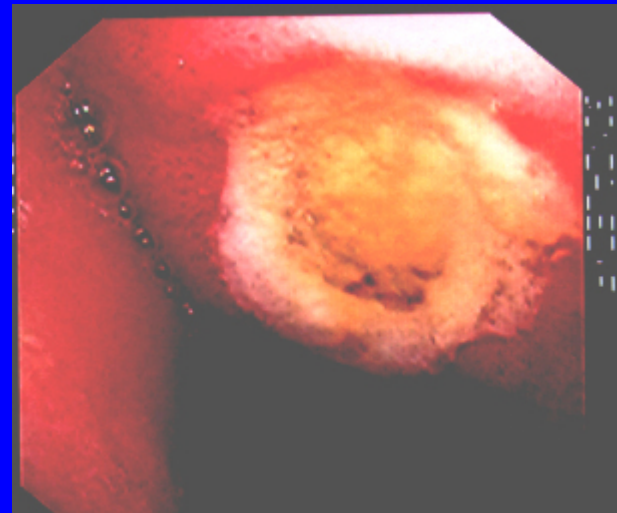
10-20% da população mundial

Mucus-secreting cells cover the surface of the stomach and extend into gastric pits



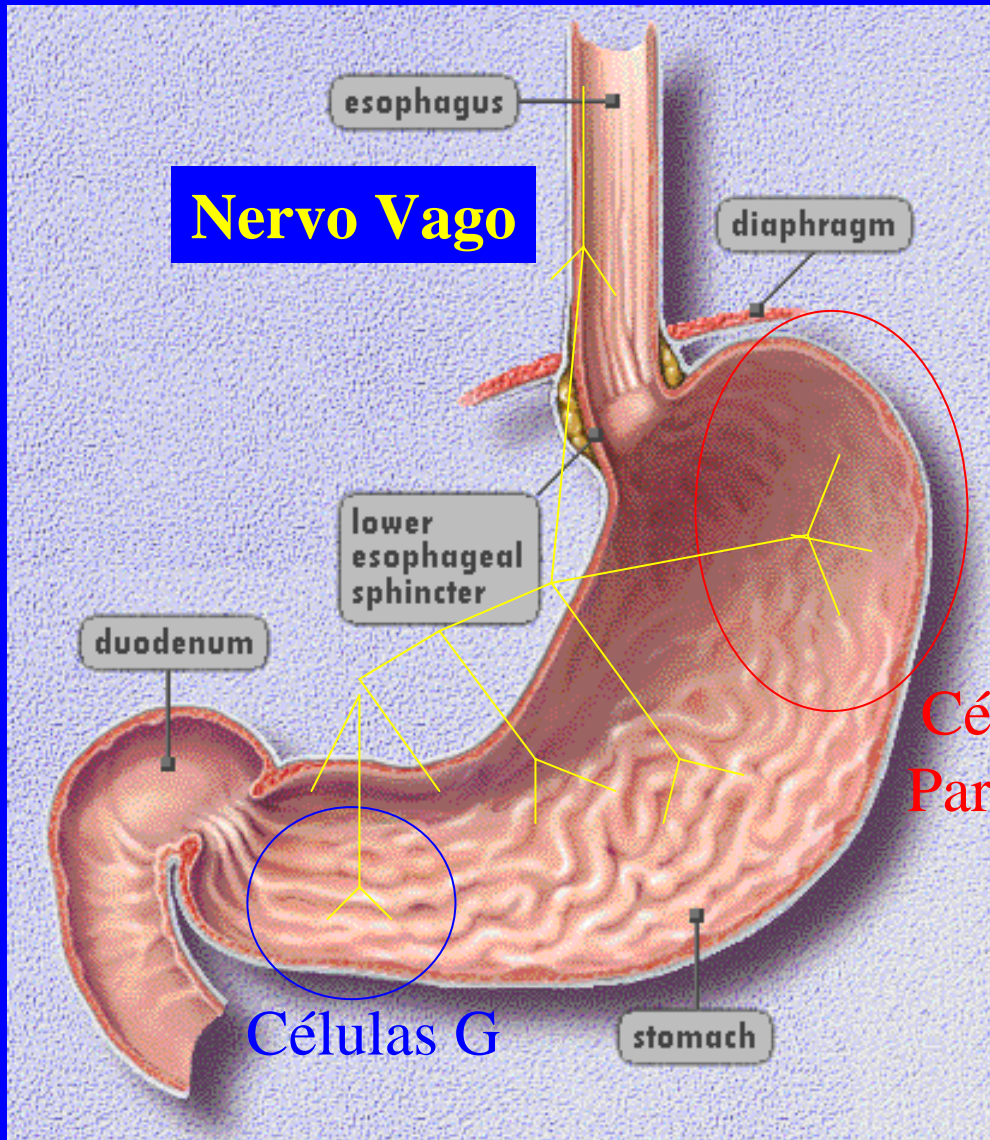
### Causas:

- Produção Excessiva de Ác. Clorídrico;
- Comidas picantes e/ou condimentads;
- Tensão Emocional;
- Infecção

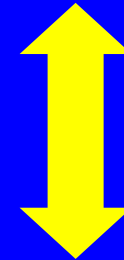




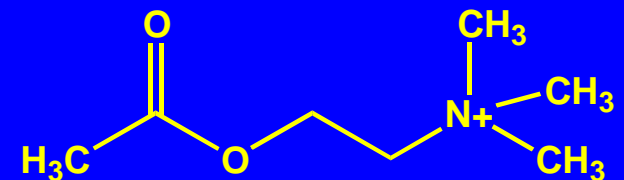
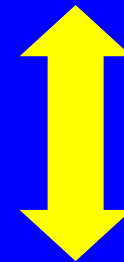
# Aspectos da Fisiologia Gátrica



Pensamento, Visão, Cheiro e  
Gosto de Comida

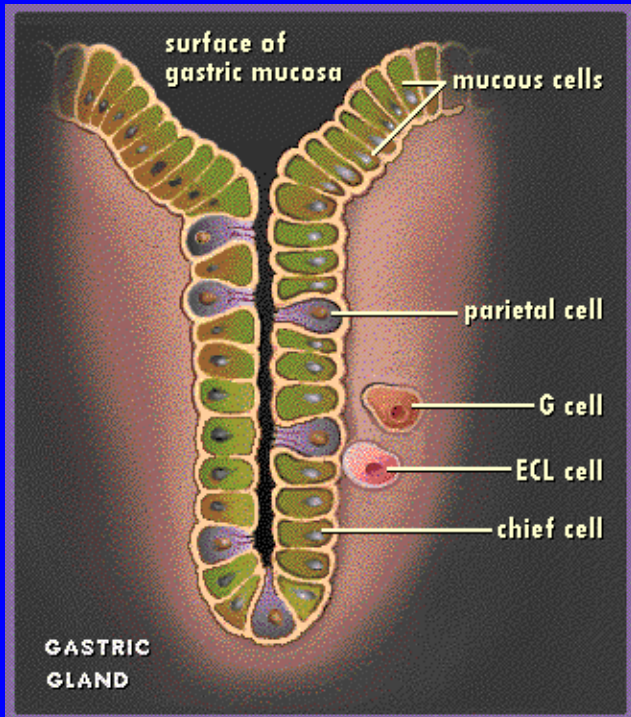


Estimulação Vagal





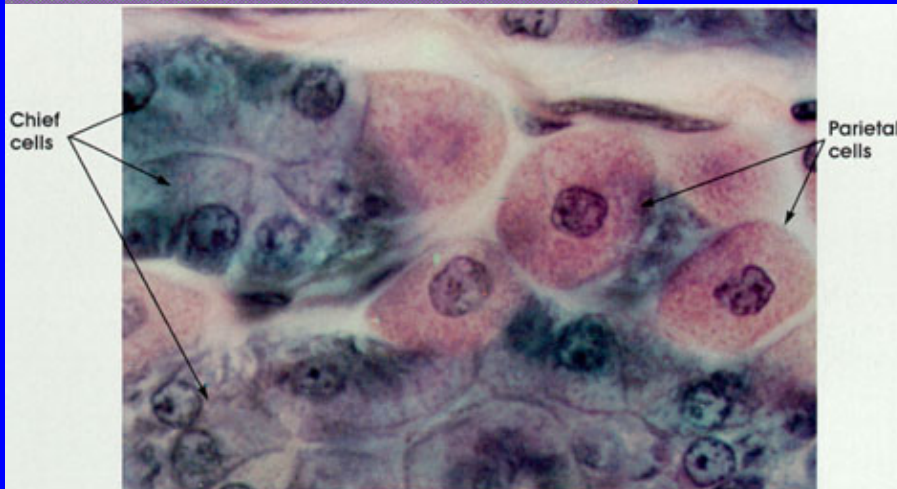
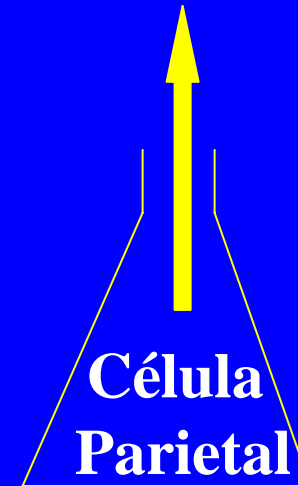
# Célula Parietal



pH ~ 1-2

HCl

Pepsina  
Muco



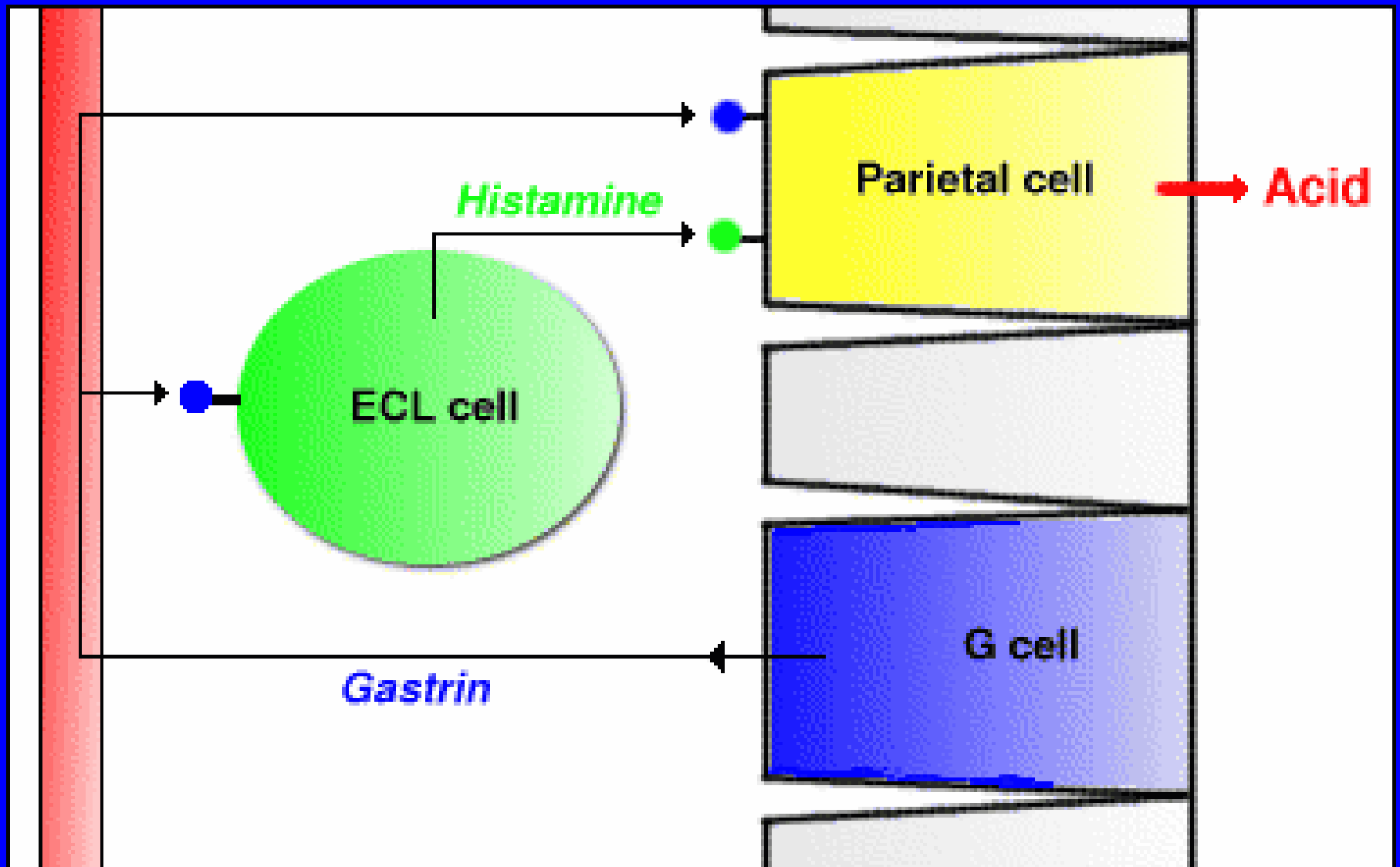
Acetilcolina

Gastrina

Histamina

Glu-Gly-Pro-Trp-Leu-  
[Glu]<sub>5</sub>-Ala-Tyr-Gly-  
Trp-Met-Asp-Phe-NH<sub>2</sub>





# Aspectos Históricos

- (1905) Edkins inferiu a presença de um hormônio que estimulava a secreção gástrica de ácido, a ele deu o nome de **gastrina**;
- (1920) Popielski descobre que a **histamina** é um potente estimulante da secreção gástrica de ácido, e concluiu que a **gastrina** de Edkins e a **histamina** eram a mesma substância;
- (1938) Até que Komarov, descobriu a natureza peptídica da **gastrina** e explorou esta característica química para separá-la da **histamina**;
- Papel dos secretagogos?
- (1948-50) Foi caracterizado que fármacos antihistamínicos não reduziam a secreção gástrica de ácido, colocando em questão o papel da **histamina**;
- (1964) Gastrina foi isolada e sequenciada na Universidade de Liverpool, convencendo a maior parte dos gastro-fisiologistas de sua importância no controle da secreção gástrica de ácido; os quais passaram a considerar que o papel da histamina era muito pouco importante

# Tratamentos Disponíveis na Época

- Antiácidos: neutralização química do HCl liberado; trissilicato de magnésio; hidróxido de alumínio; bicarbonato de sódio; carbonato de cálcio; **Grandes quantidades / Efeito Rebote**
- Fármacos Anticolinérgicos; Ativos apenas em altas doses, levando ao bloqueio de outros receptores de acetilcolina (**Efeito tóxico**)
- Vagotomia

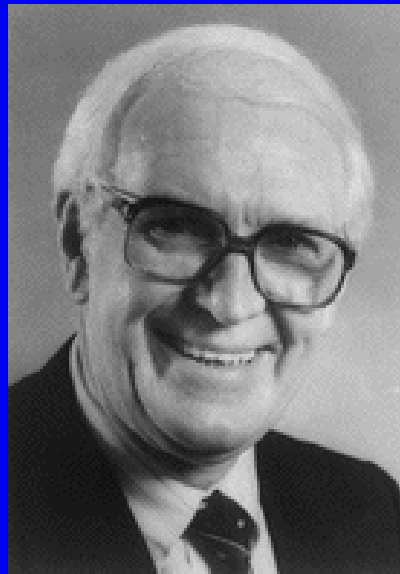


# A Pesquisa de Novos Fármacos Anti-Úlceras

- Anos 60: Compreensão da fisiologia da secreção gástrica de ácido;
- Programas para a descoberta de Inibidores da ação de mensageiros químicos;
- Reino Unido: ICI Pharmaceuticals, Pfizer, Smith Kline French:  
**Antagonistas de Receptores Histaminérgicos**



R. Ganellin  
G. Durant  
J. Emmett

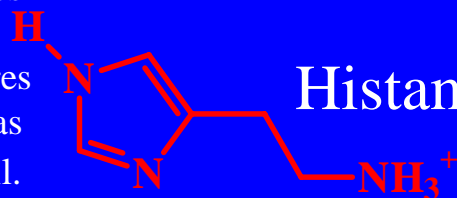


Sir James Black  
Prêmio Nobel  
de Medicina, 1988

# Programa Antagonistas-H<sub>2</sub>

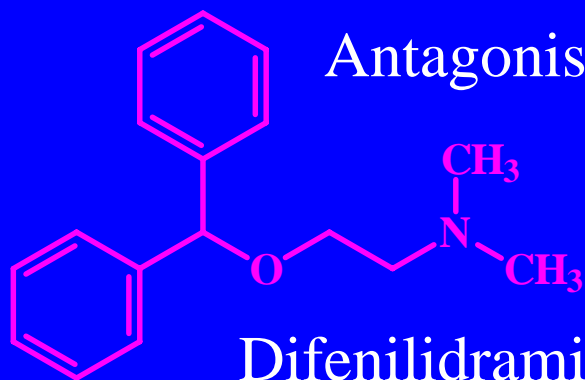
(1948) Folkow, Haeger e Kahlson: Reduzem, mas não abolem as propriedades Vasodilatadora da histamina, sugerindo a existência de 2 tipos de receptores sensíveis à histamina, dos quais apenas um podia ser bloqueado pelo Benadril.

(1966) Ash e Schild: Receptor H<sub>1</sub>



Histamina (Monocátion)

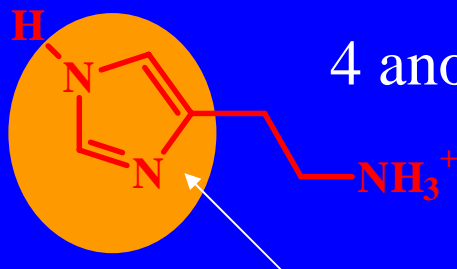
## Antagonistas de Receptores Histaminérgicos



Difenilidramina  
(Benadril, Parke Davis)

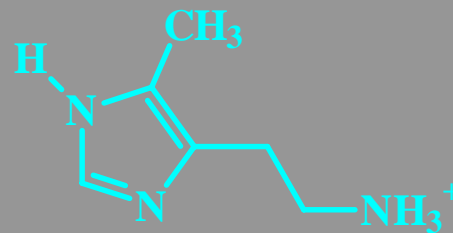


Mepiramina



4 anos, >200 compostos

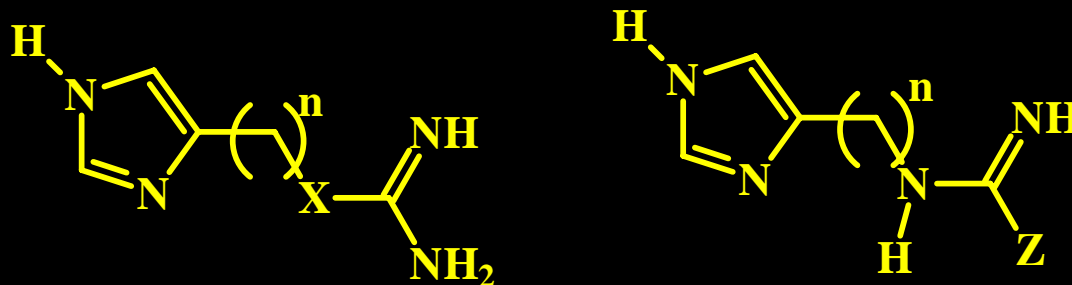
Idéia Original de Black: Alquilação  
Lipofilicidade X Hidrofilicidade



4-Metilhistamina: Agonista Seletivo  
(Induz a secreção gástrica de ácido)



# REA de algumas imidazolilisotiouréias, guanidinas e amidinas

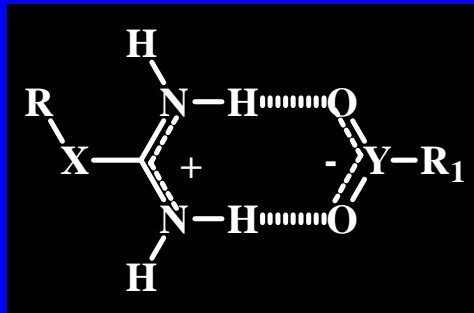


Composto	n	Substituinte	Atividade
1	2	X = NH	+
2	2	X = S	++
3	3	X = NH	+++
4	3	X = S	±
5	2	Z = SMe	±
6	2	Z = Me	±
7	3	Z = SMe	+++
8	3	Z = Me	+++

Teste de inibição da secreção gástrica de ácido, induzida por histamina; ± (detectável);  
 + (ID<sub>50</sub> > 500 µmol/Kg); ++ (ID<sub>50</sub> ~ 200 µmol/Kg); +++ (ID<sub>50</sub> = 100-50 µmol/Kg).  
 ID<sub>50</sub> = Dose intravenosa capaz de reduzir a secreção de ácido em 50%.

# Imidazolilisotiouréias, Amidinas e Guanidinas (Monocátions)

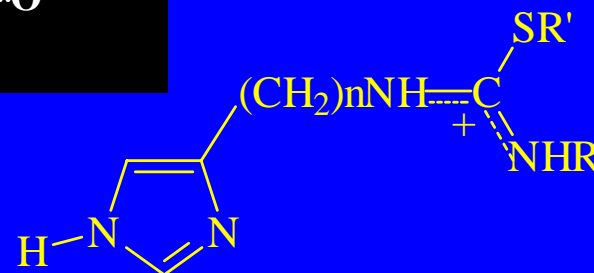
Agonismo X Antagonismo



Isotiouréias

**n = 2-4**

**R = H ou alquila**



Isotiouréias  
Invertidas

**n = 2-5**

**R = H ou alquila**

**R' = alquila**

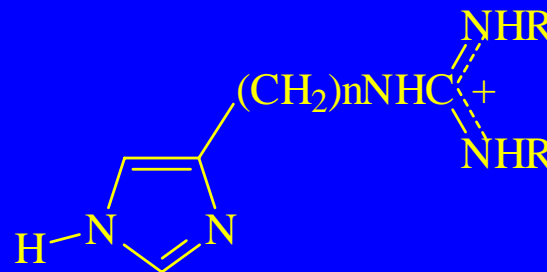


Amidinas

**n = 2-5**

**R = H ou alquila**

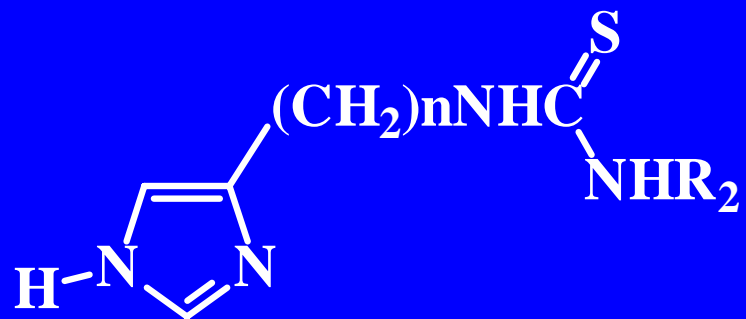
**R'' = alquila, arila, etc.**



Guanidinas

# Descoberta da Burimanida

Introdução de Grupos Polares,  
Não Básicos

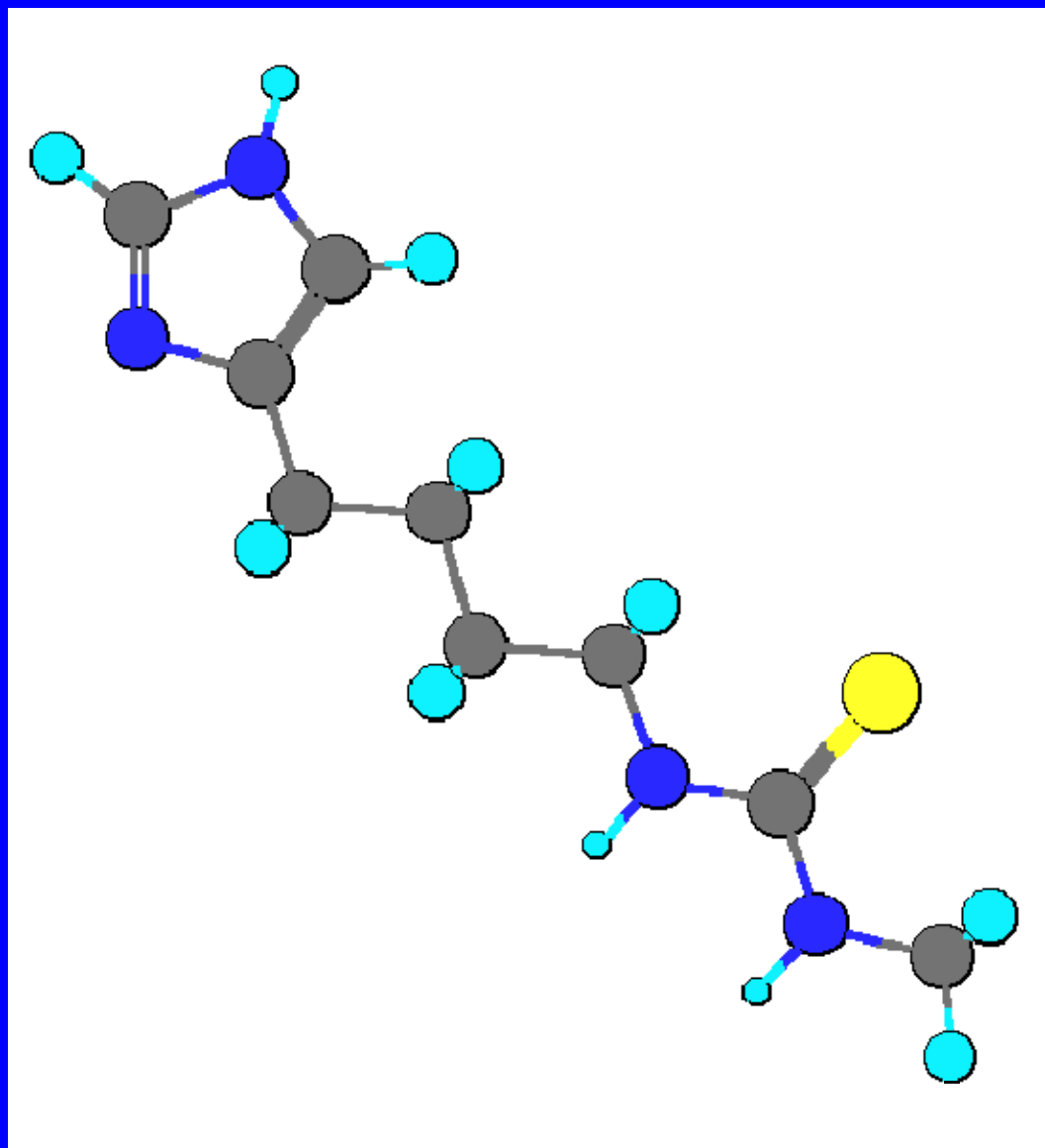


## Tiouréias

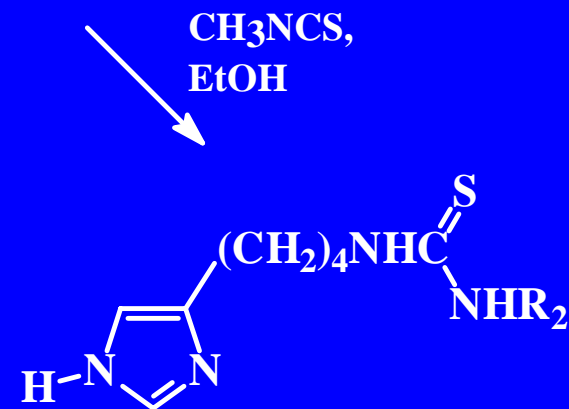
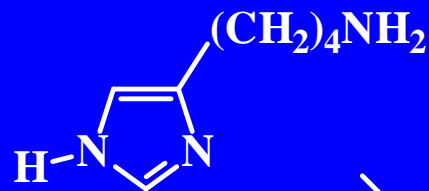
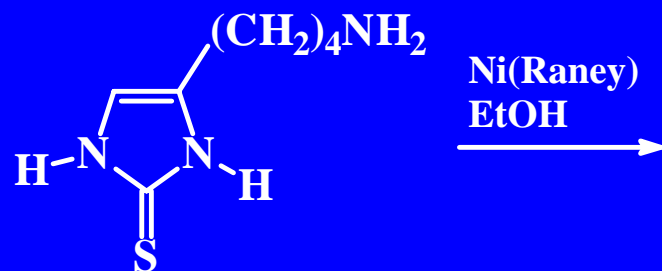
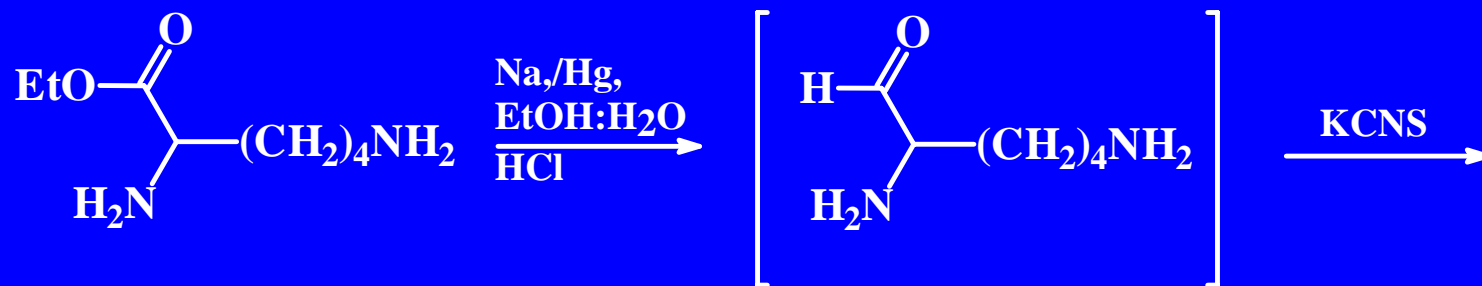
(9) SK&F 91581  $n = 3$ ,  $\text{R}_2 = \text{H}$

(10) SK&F 91863  $n = 4$ ,  $\text{R}_2 = \text{H}$

(11) Burimamida,  $n = 4$ ,  $\text{R}_2 = \text{CH}_3$

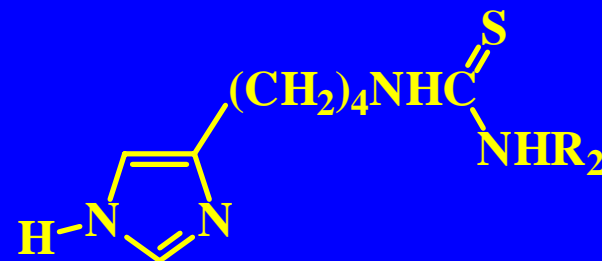
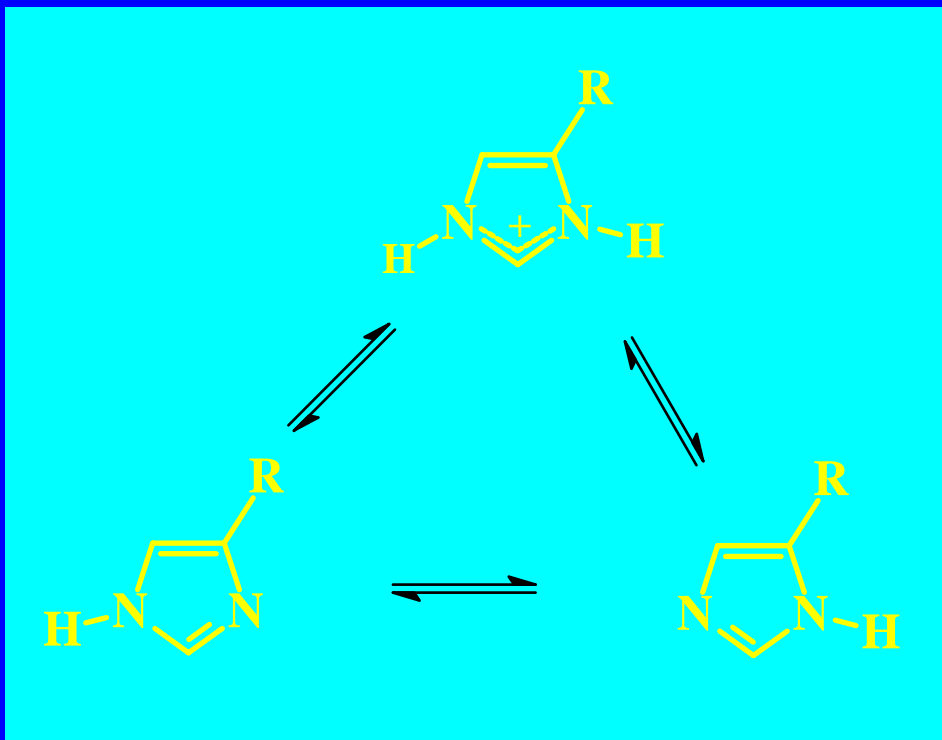


# Síntese da Burimamida

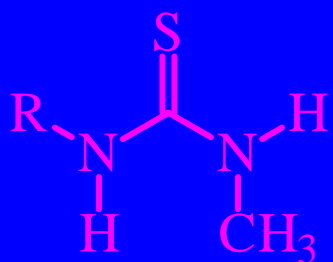
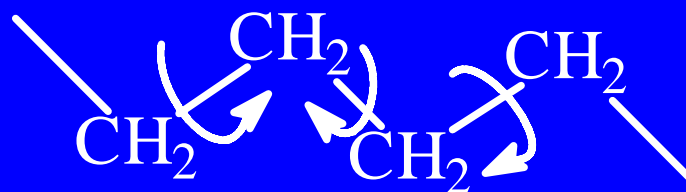


**Burimamida**

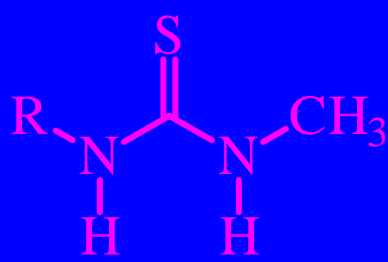
# Características Estruturais da Burimamida



**Burimamida**



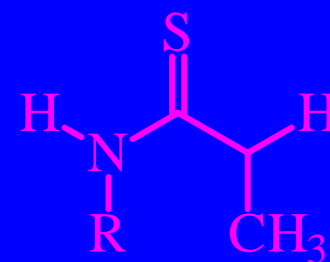
(s-cis,s-trans)



(s-cis,s-cis)

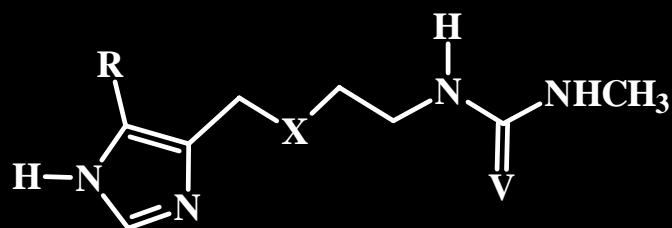


(s-trans,s-cis)



(s-trans,s-trans)

# Planejamento Estrutural de Isósteros da Burimamida

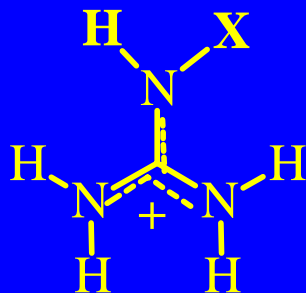


Atividade em Receptores  $H_2$

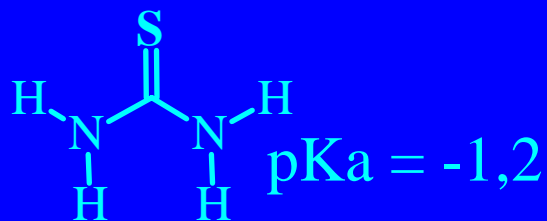
Composto	R	Estrutura		<i>In Vitro</i>		<i>In Vivo</i> Secreção de HCl ID <sub>50</sub> (µg/mol)
		X	V	Átrio ( $K_B \times 10^{-6} M$ )	Útero ( $K_B \times 10^{-6} M$ )	
Burimamida	H	CH <sub>2</sub>	S	7,8	6,6	6,1
Tiaborimamida	H	S	S	3,2	3,2	5
Oxaburimamida	H	O	S	28	6,6	ND
Metiamida	CH <sub>3</sub>	S	S	0,92	0,75	1,6
Isósteros Ureídicos	CH <sub>3</sub>	S	S	22	7,1	27
Isósteros	CH <sub>3</sub>	S	NH	16	5,5	12
Gunidínico						
Isósteros	CH <sub>3</sub>	S	N-NO <sub>2</sub>	1,4	1,4	2,1
Nitroguanidínico						
Isósteros	CH <sub>3</sub>	S	N-CN	0,79	0,81	1,4
Cianoguanidínico						
Isósteros	CH <sub>3</sub>	S	N-CONH <sub>2</sub>	7,1	6,9	7,7
Guaniluréia						



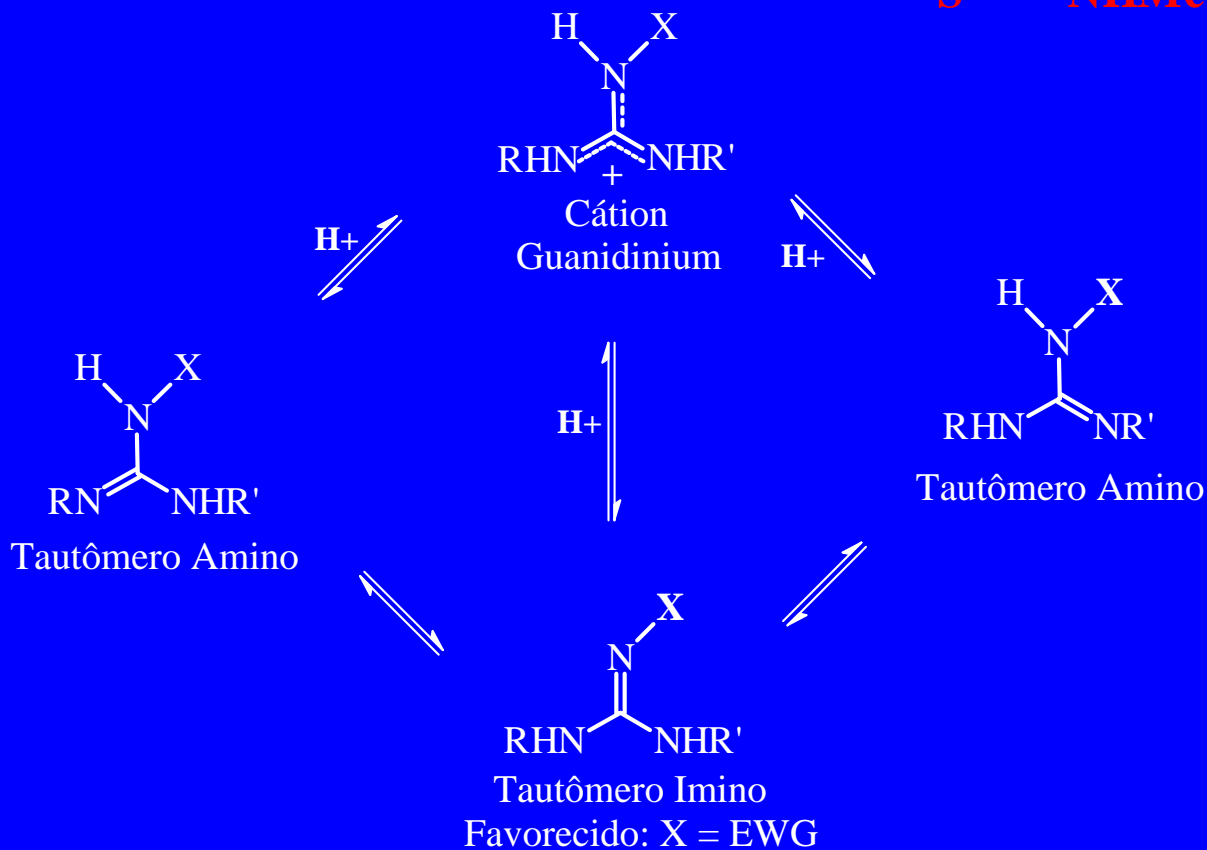
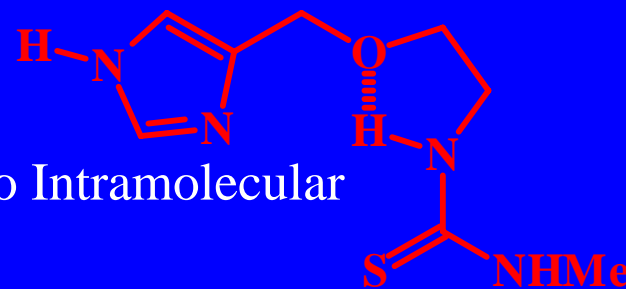
# Aspectos Estruturais de Guanidinas Substituídas e Derivados



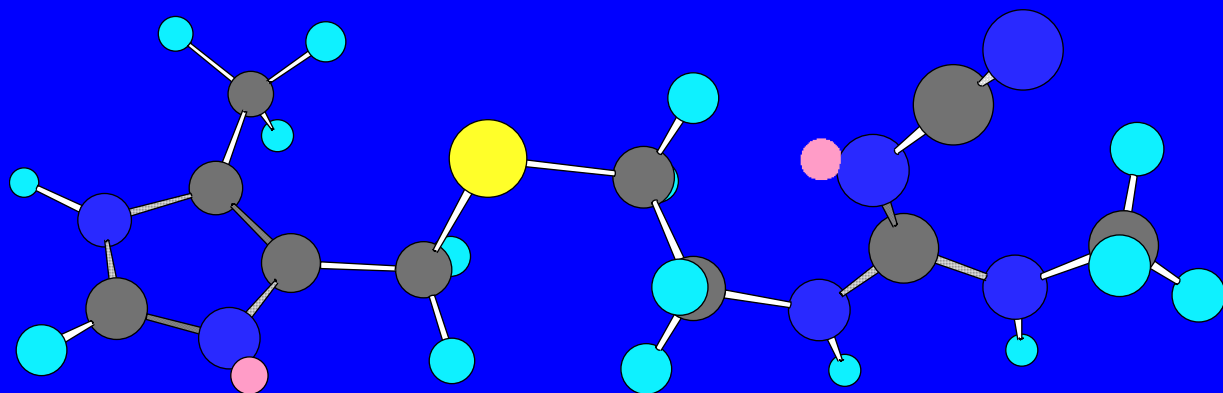
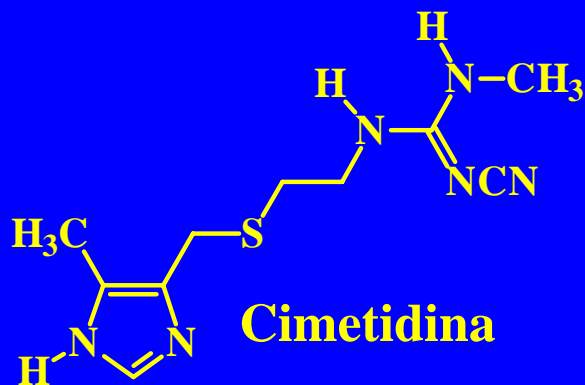
X	$\sigma$	pKa'
H	0	14,0
Ph	0,11	10,6
Ac	0,32	8,0
CONH <sub>2</sub>	0,30	7,3
OMe	0,32	6,3
CN	0,59	0,4
NO <sub>2</sub>	0,62	0,9



Ligação de Hidrogênio Intramolecular



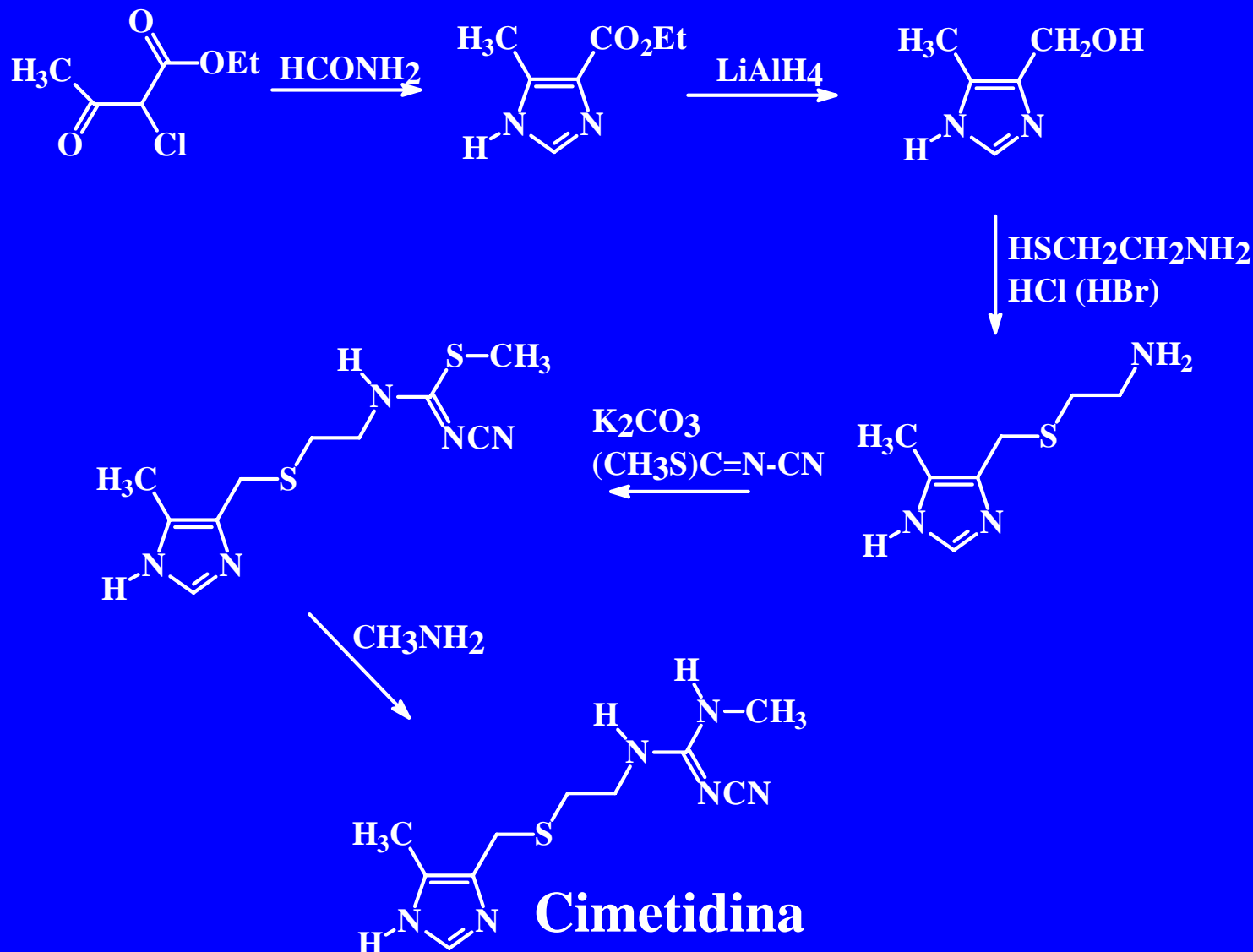
# O Novo Fármaco: Cimetidina



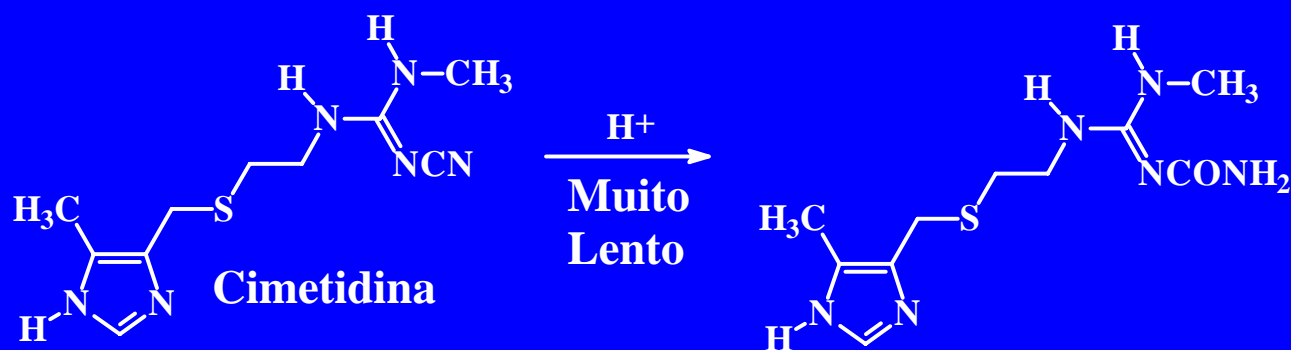
Animal	Preparação	Estimulante	ID <sub>50</sub> (μmol/Kg) intravenoso
Ratos	Lúmen Estomacal Perfundido	Histamina	1,37
		Pentagastrina	1,40
Gatos	Lúmen Estomacal Perfundido	Histamina	0,85
		Pentagastrina	1,45
Cães	Lúmen Estomacal Perfundido	Histamina	1,70
		Pentagastrina	2,00

“Reduz a secreção gástrica de ácido estimulada por comida”

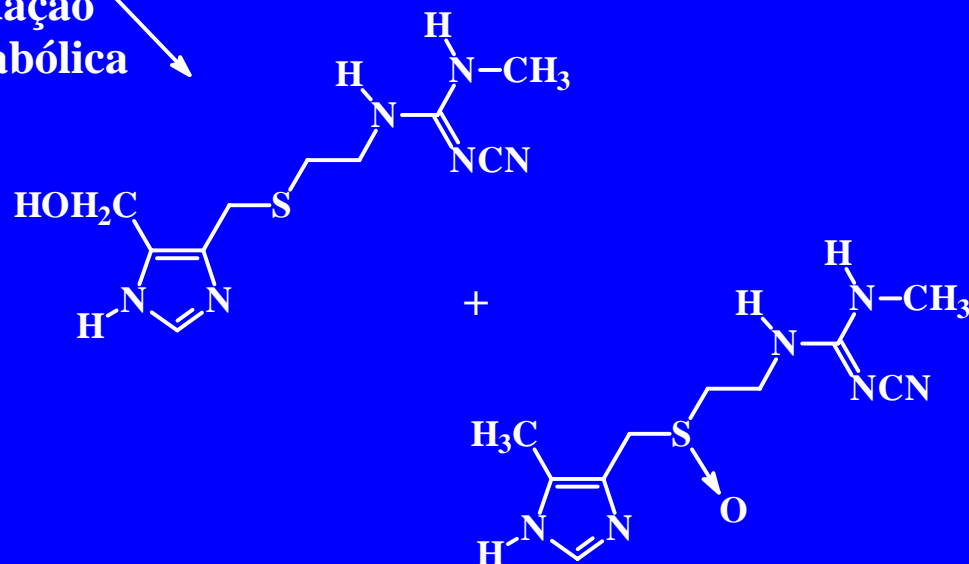
# Síntese da Cimetidina



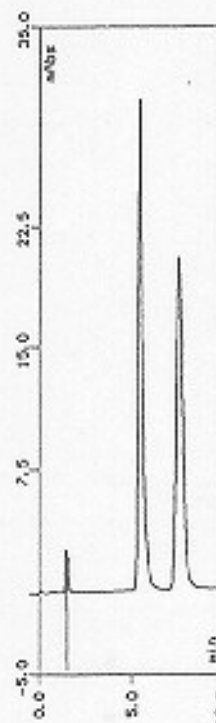
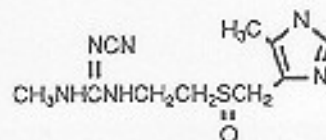
# Metabolismo da Cimetidina



Oxidação  
Metabólica



CIMETIDINE SULPHOXIDE

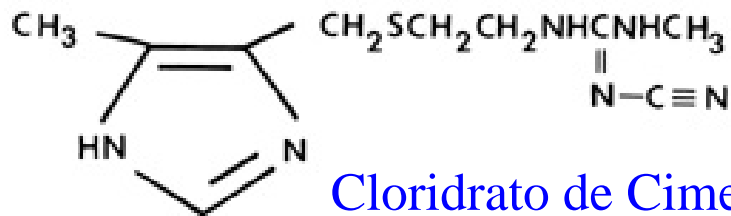


Column:  
CHIRAL-CEH  
150 x 4.0 mm

Mobile phase:  
10 mM sod.ph.b,  
pH 6.0 + 50  $\mu$ M di-  
sodium EDTA

Sample conc.:  
0.03 mg/ml

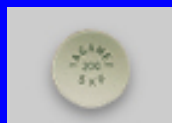
# Formulações Farmacêuticas



# Cloridrato de Cimetidina



# Tagamet (SK&F)



100 mg HB

200 mg

300 mg

400 mg

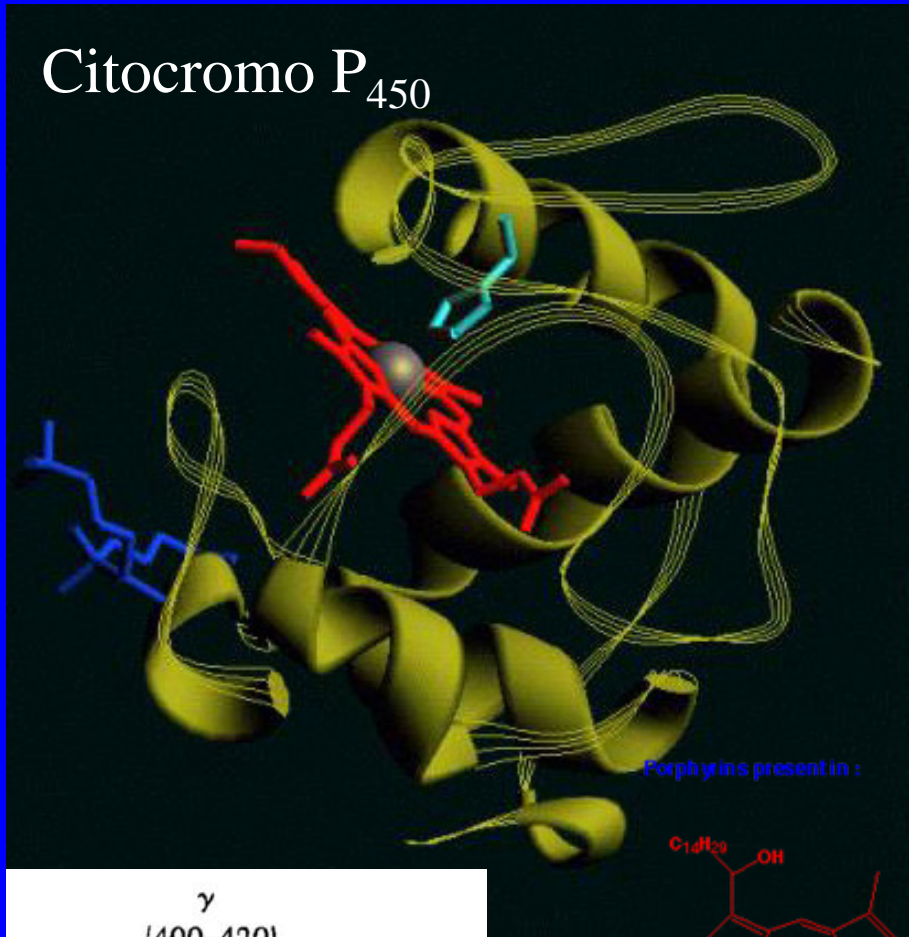
800 mg

Este medicamento é também disponível como genérico

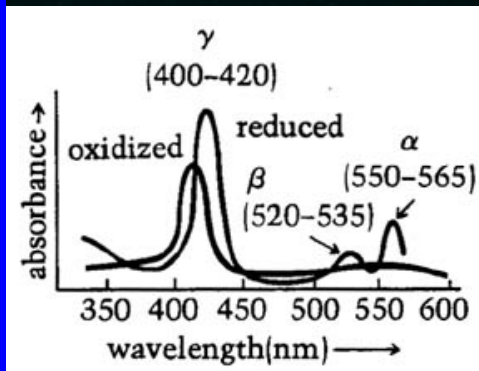


# Efeitos Colaterais da Cimetidina

## Citocromo P<sub>450</sub>



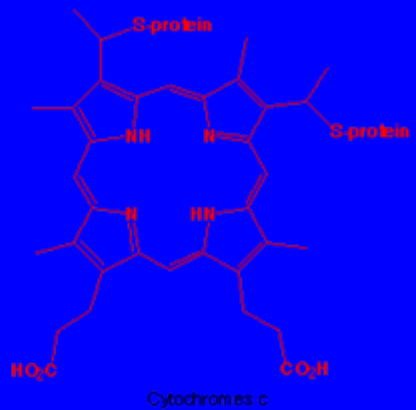
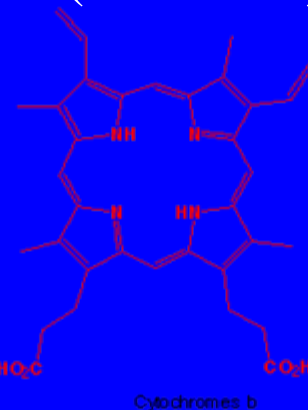
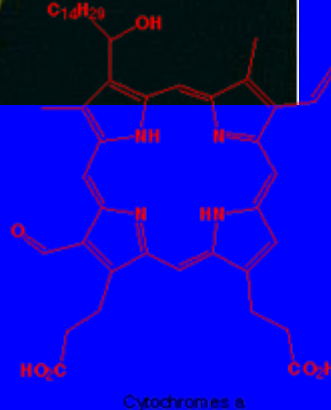
Porphyrins present in :



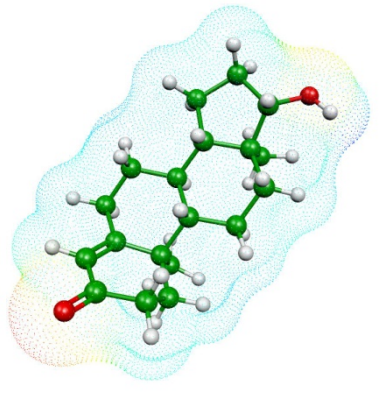
## Principais Efeitos Colaterais Em Homens:

- 1) Crescimento de Seios (Ginecomastia)
- 1) Perda da Libido
- 2) Impotência

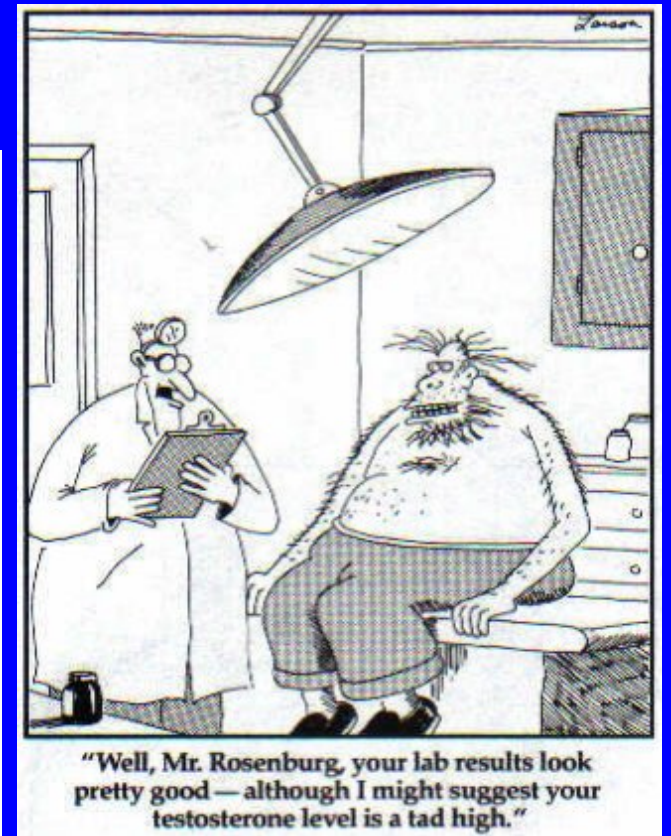
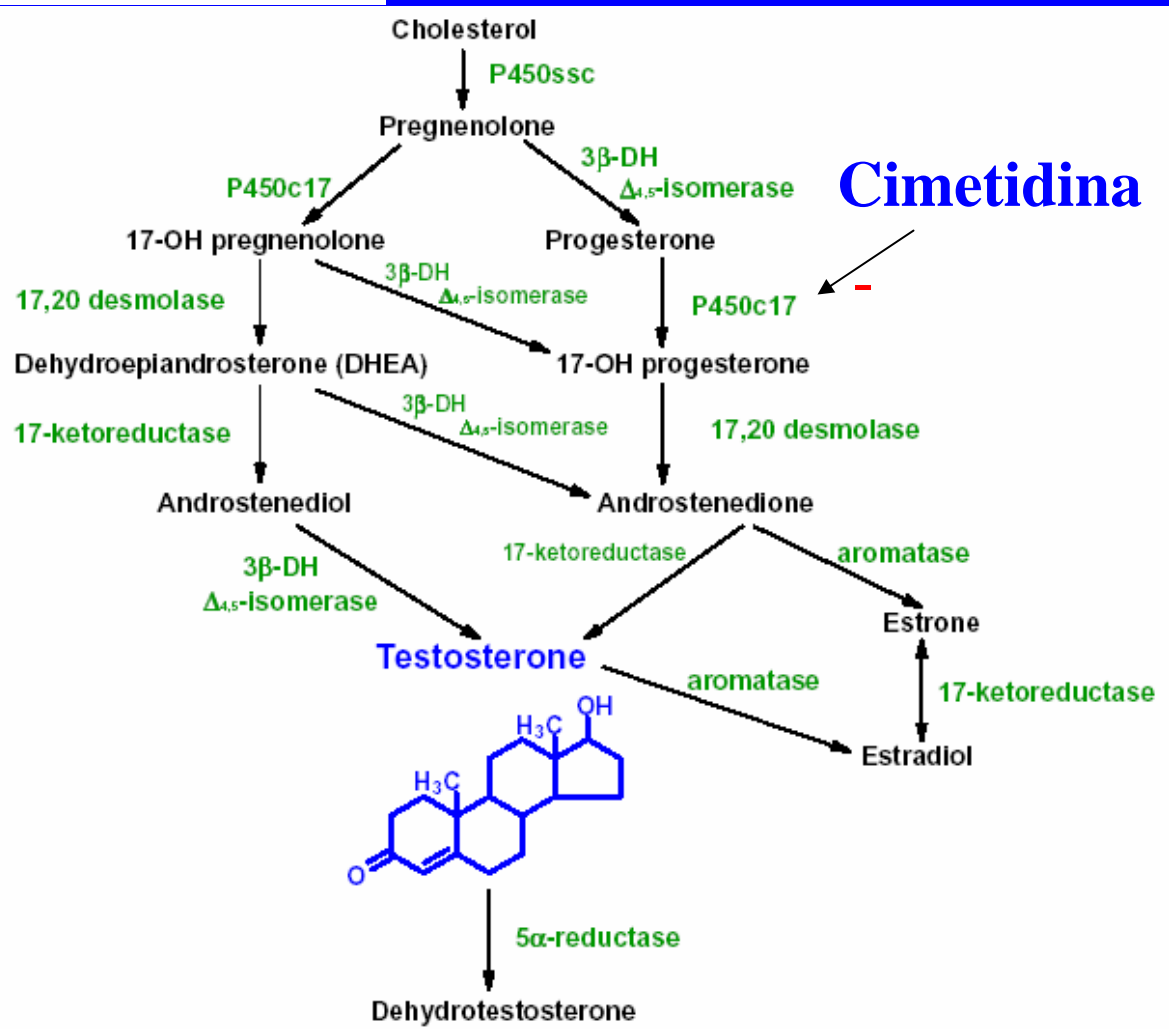
## Interações Medicamentosas com vários outros fármacos metabolizados por oxidação hepática (CYP450)





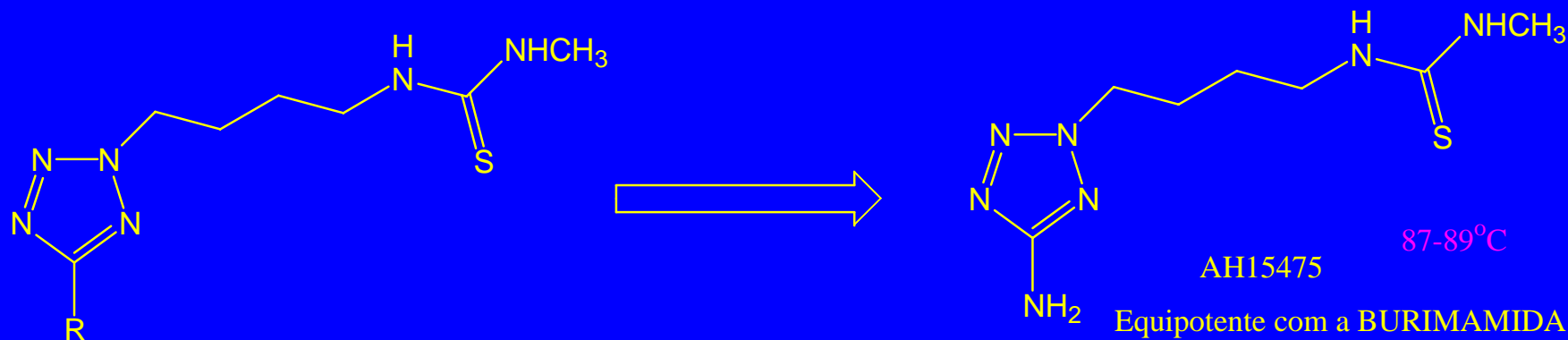
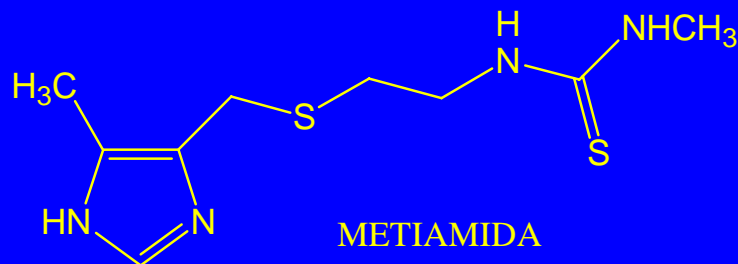


# Ação da cimetidina na biossíntese de testosterona



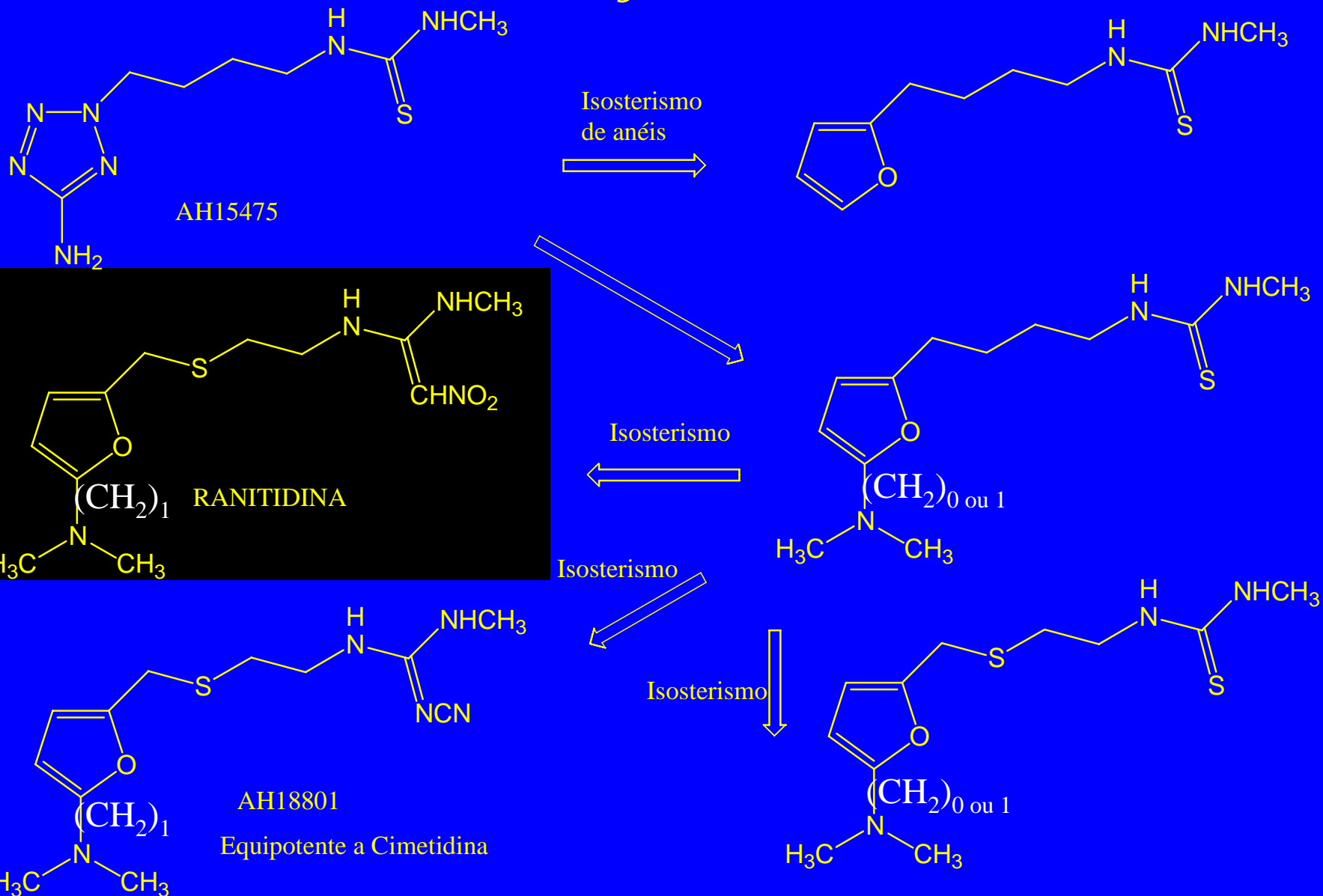
"Testosterone increases secondary sex characteristics and also increases bone growth, muscle development and sex drive. Examples of secondary sex characteristics are an increase in facial hair, muscle development, deepening of voice, production of sperm."

# Novo Programa de Pesquisas para Antagonista de Receptores H<sub>2</sub>: Glaxo Group Research Ltd.

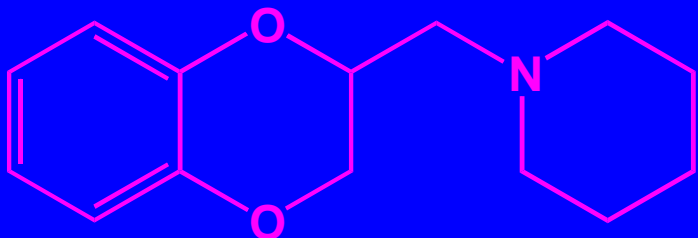


Não há incremento significativo na atividade sobre a secreção gástrica de HCl

# Isosterismo de Anéis com Estratégia de Modificação Molecular

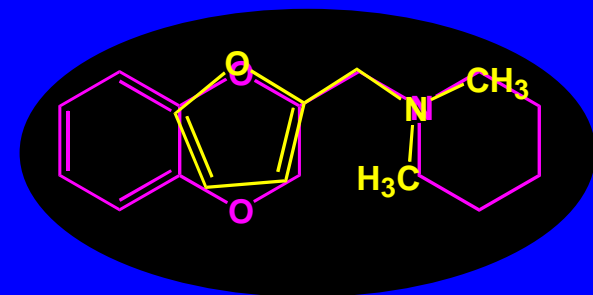
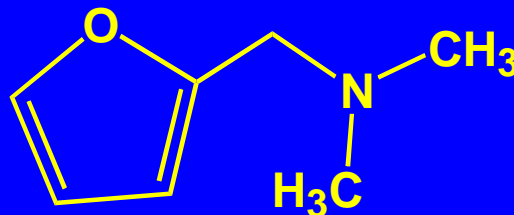


# Bioisosterismo de Anéis

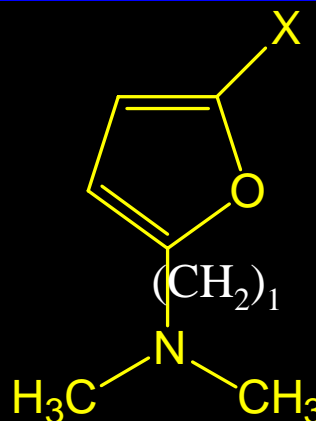
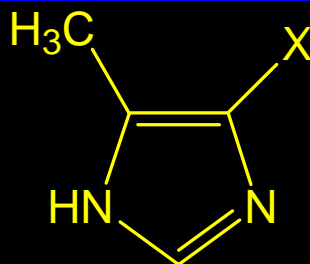


Piperoxano

Antagonista de Receptores Histaminérgicos



**X**



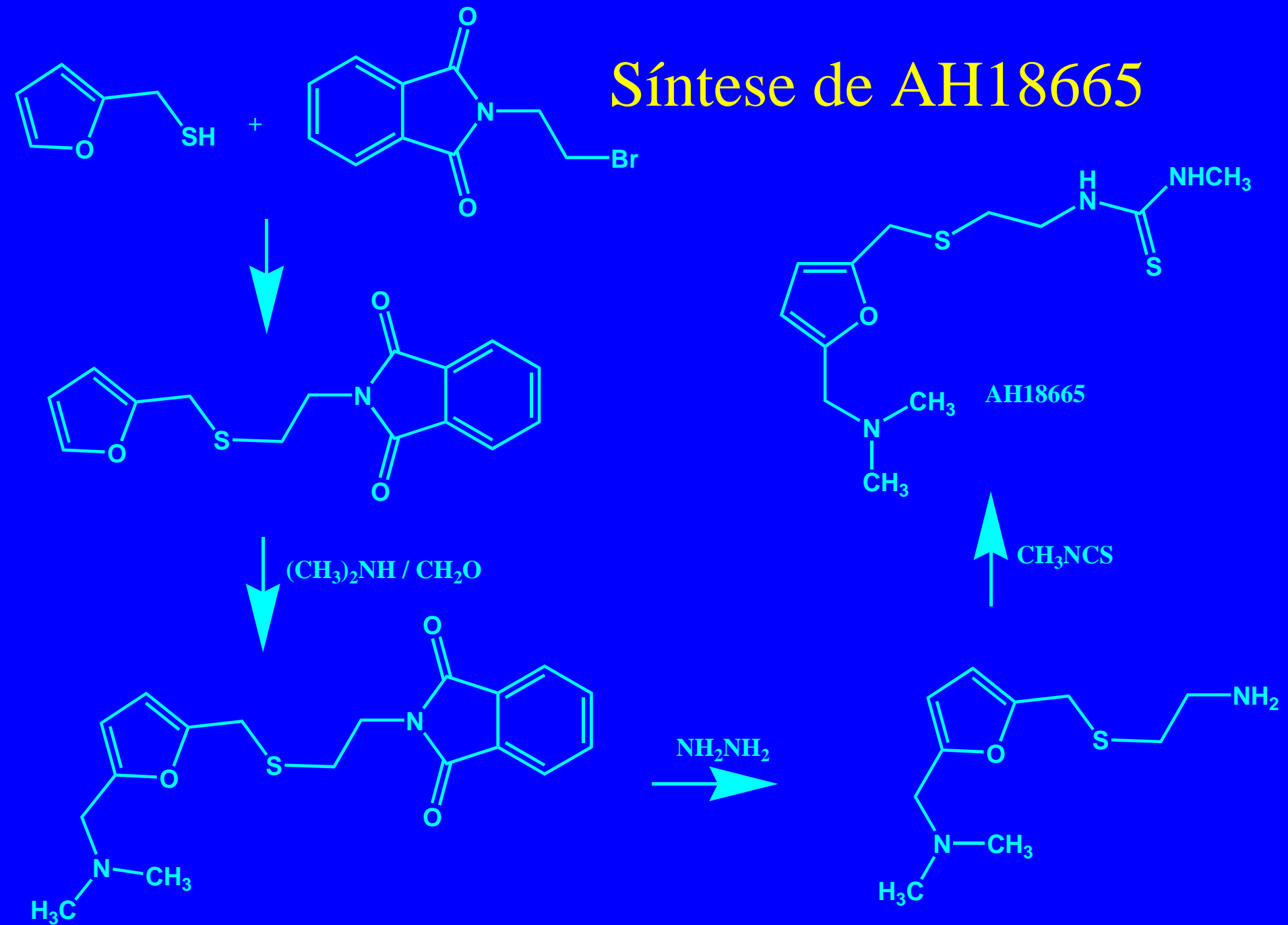
-NHC(=S)NHCH <sub>3</sub>	0,52	2,32
-NHC(=NCN)NHCH <sub>3</sub>	1,12	1,39
-NHC(=CHNO <sub>2</sub> )NHCH <sub>3</sub>	1,75	0,18



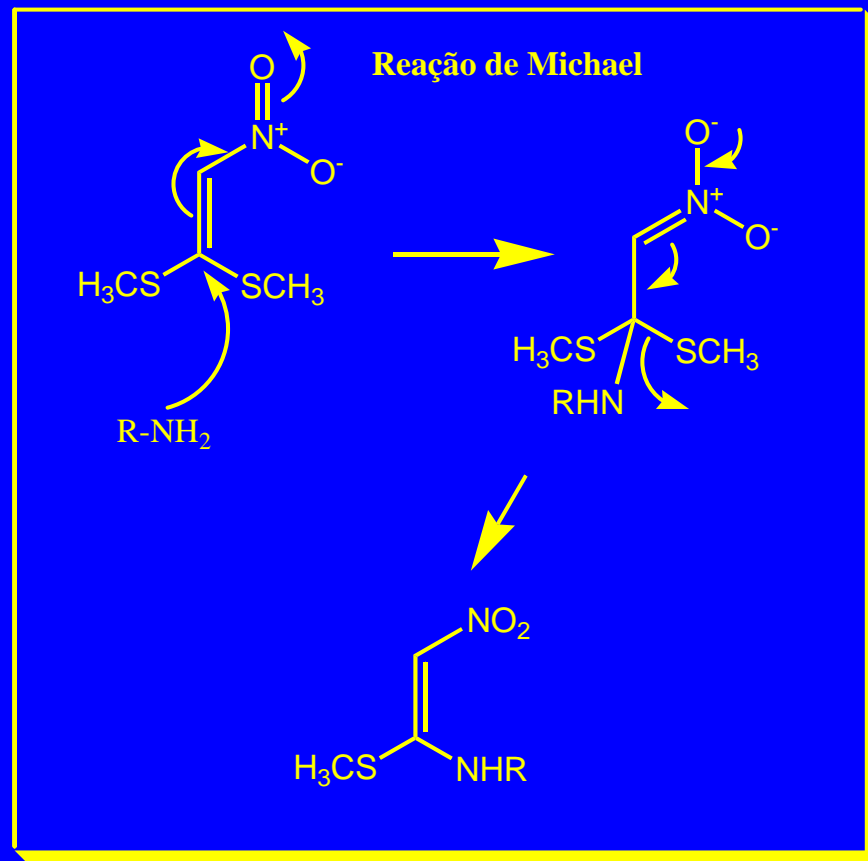
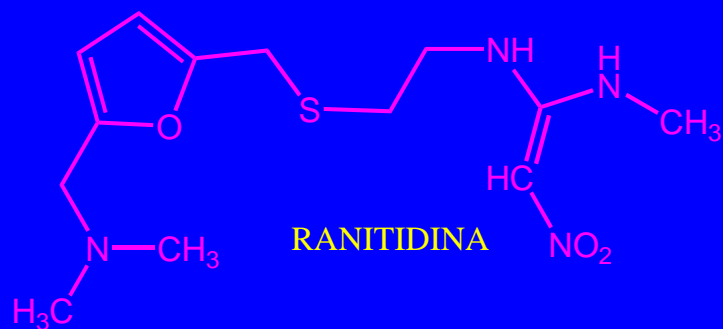
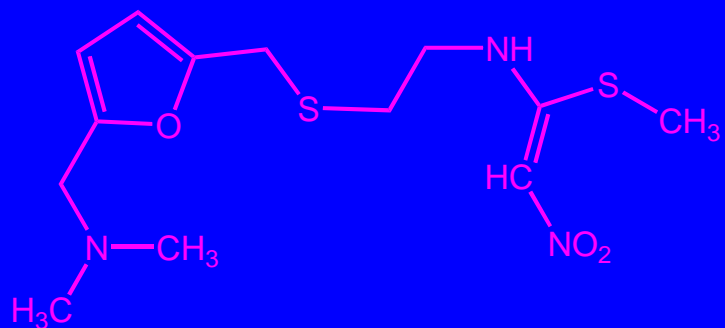
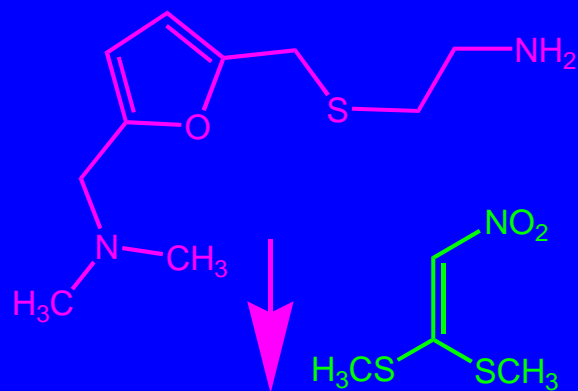
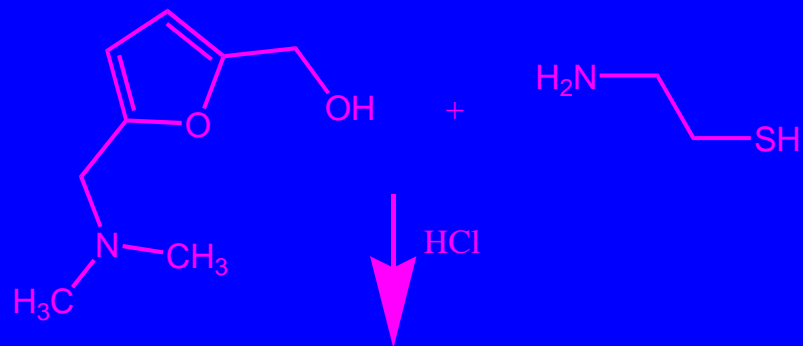
Principais Variações  
Estruturais no Esqueleto  
Furânico

Atividade (mg/Kg) na preparação de estômago de ratos perfundidos

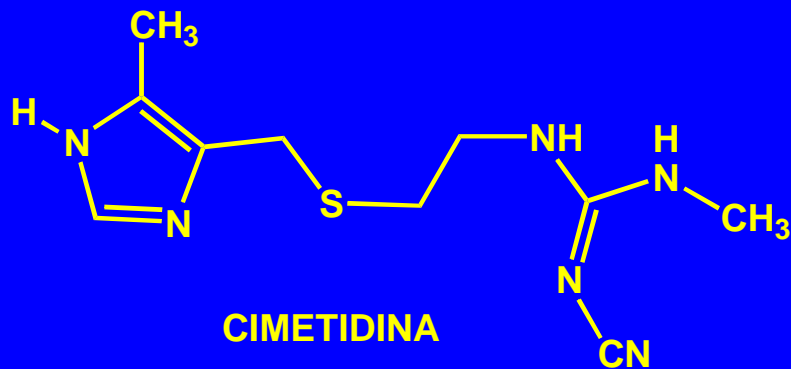
# Síntese de AH18665



# Síntese da Ranitidina







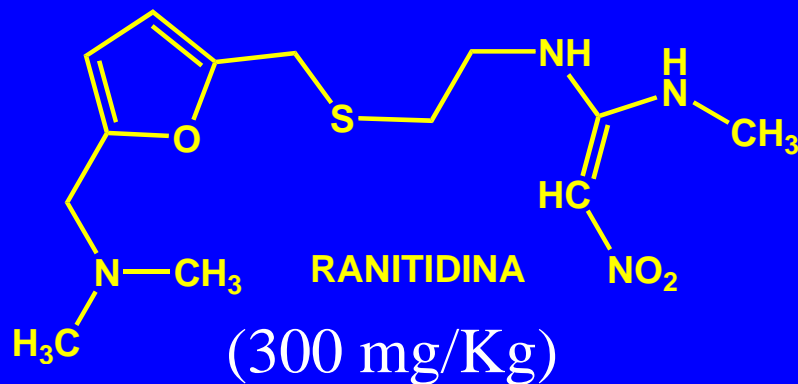
**TAGAMET**

SK&F: Total em

vendas em 1976: U\$ 386 milhões

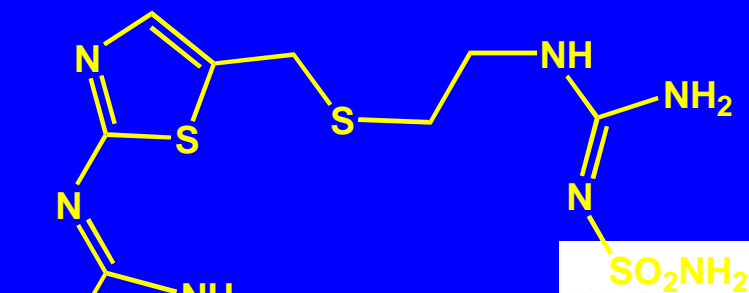
Vendas de Cimetidina em 1980: U\$ 580 milhões

(800 mg/Kg) 1983: Top (U\$ 1 milhão)



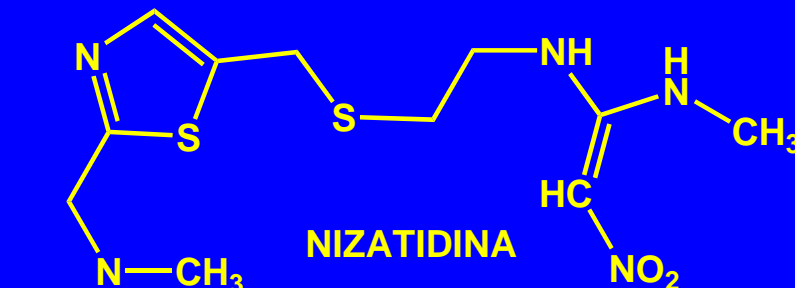
Glaxo

1987: Top

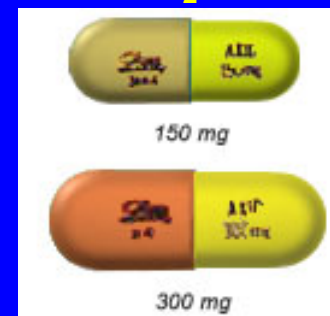


ICI

(40 mg/Kg)



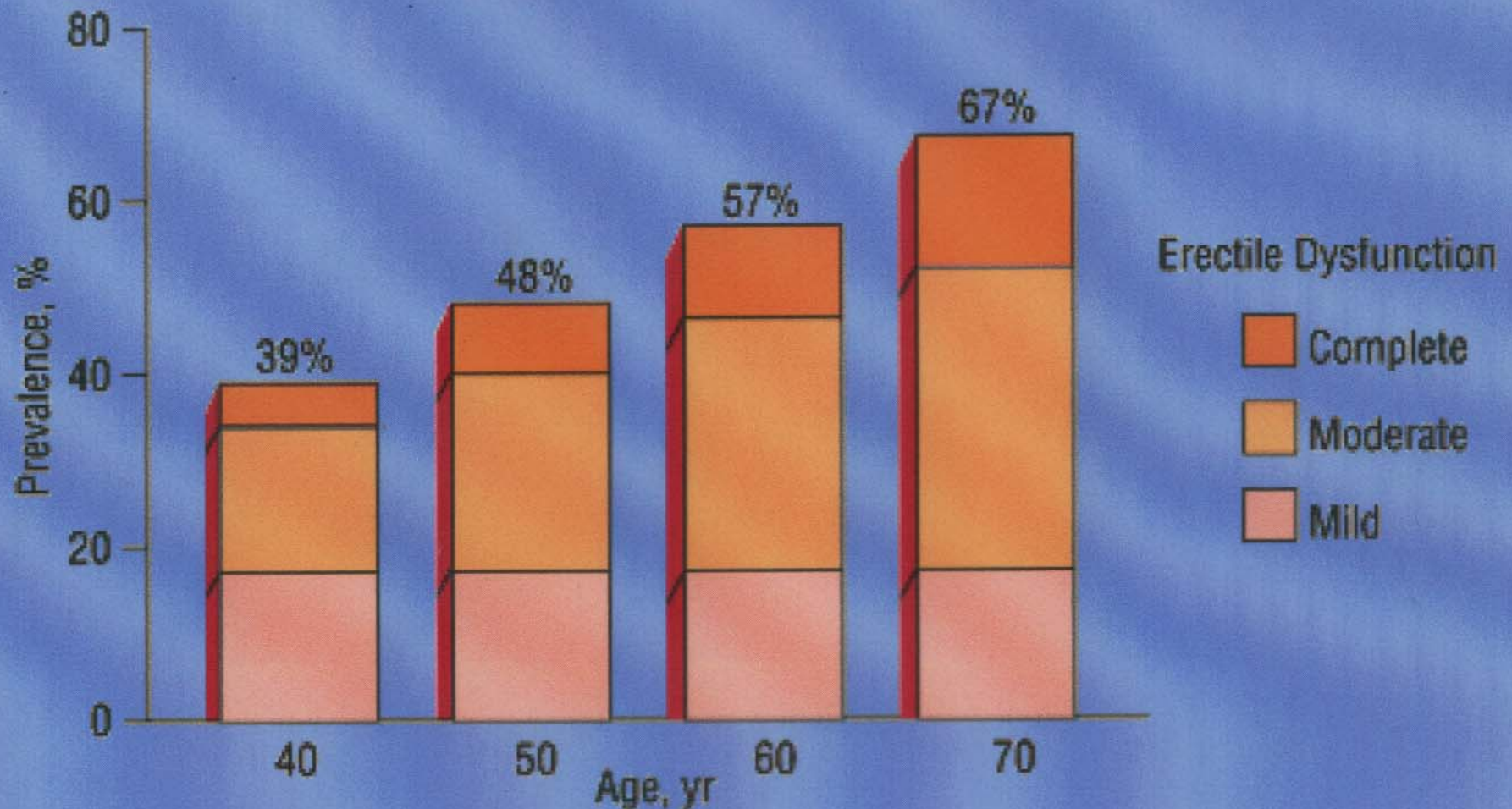
Lilly



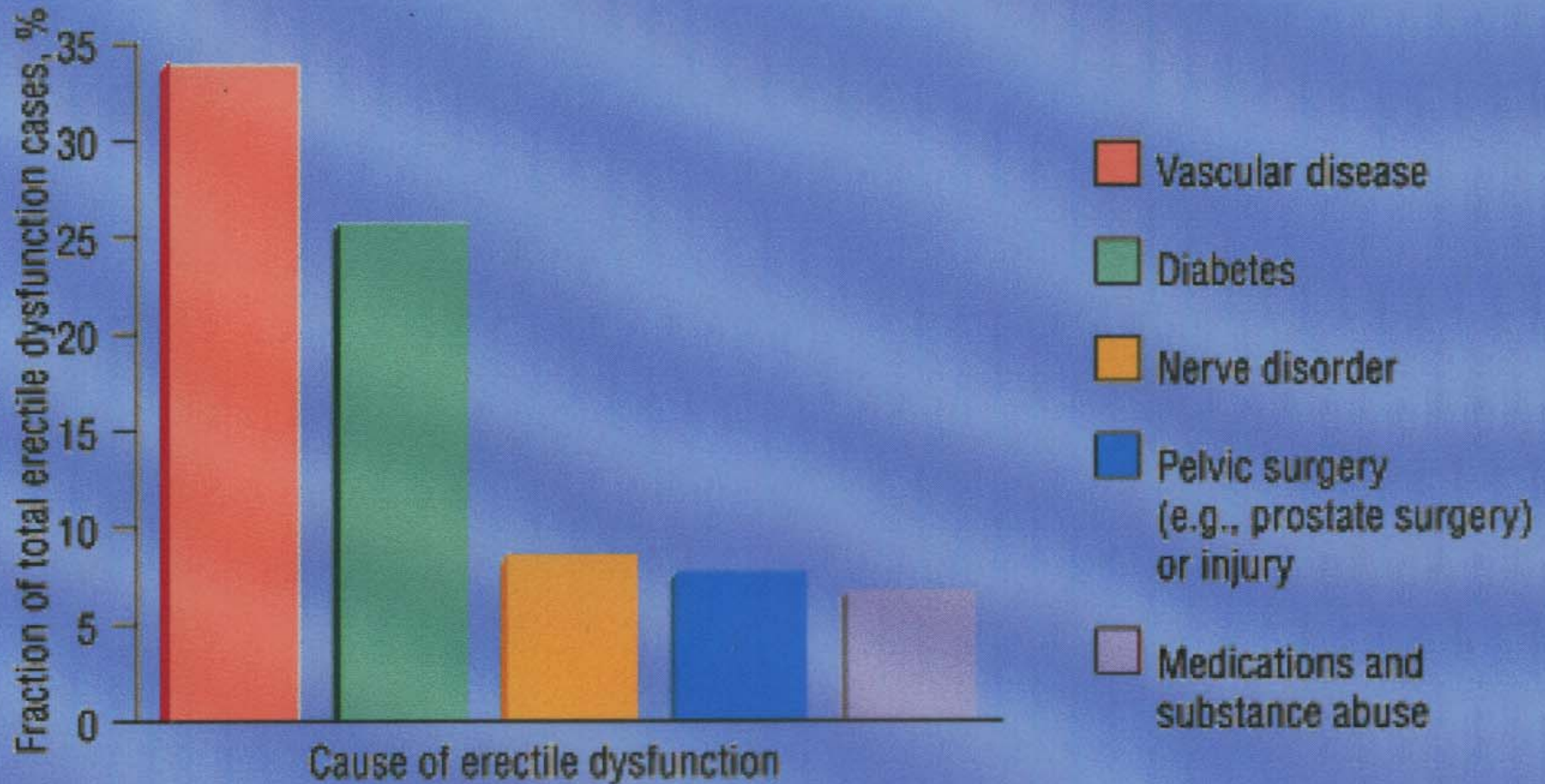
# A História do Sildenafil (Simon Campbell - Pfizer)

- **1985** Programa de Descoberta de Fármacos iniciado nos laboratórios Pfizer para desenvolver uma tratamento para a Angina. O ponto de partida foi a modificação química do conhecido vasodilatador ZAPRINAST, o qual tinha sido desenvolvido por May and Baker (agora na Rhone Poulenc, a qual se fundir com a Hoechst para se tornar a Aventis) mas que nunca alcançou o mercado.
- **1989** Aproximadamente 1600 compostos foram sintetizados no âmbito deste programa, incluindo o Sildenafil.
- **1990** Sildenafil entrou em Fase I de triagem clínica em pacientes saudáveis. Entretanto, ele não preencheu as expectativas para o tratamento de doenças coronárias. Efeito colateral em homens durante estes ensaios indicaram seu potencial para o tratamento da disfunção erétil (DE).
- **1994** Fase 2 de Triagem Clínica para o tratamento da DE.
- **1998** O FDA aprova o uso do Sildenafil (Viagra) para o tratamento da DE.

# Disfunção Erétil x Idade



# Principais Causas da DE



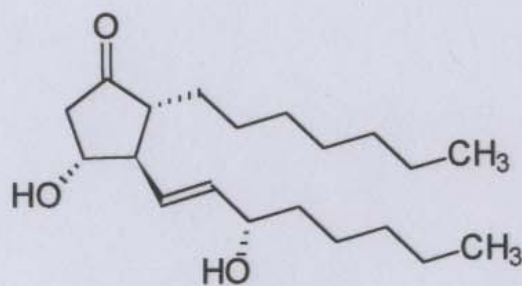


# Fármacos Indutores de DE

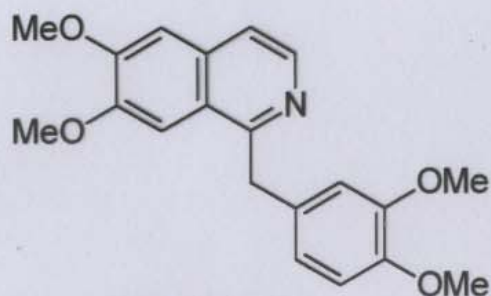
Drug Class	Chemical class or generic drug
Stimulants	amphetamines, cocaine
Anticancer drugs	leuprolide
Anticonvulsants	carbamazepine
Antidepressants	tricyclics, monoamine oxidase inhibitors
Antifungals	ketoconazole
Antihistamines	cimetidine
Antihypertensives	beta-blockers, clonidine, guanethidine, methyldopa, spironolactone, thiazides
Antipsychotics	butyrophenones, phenothiazines
Alcohol	ethanol
Cannabinoids	marijuana (delta-9-THC)
Narcotics	methadone
Sedatives	barbiturates

# Estratégias Terapêuticas usualmente para a correção da DE, antes do Viagra

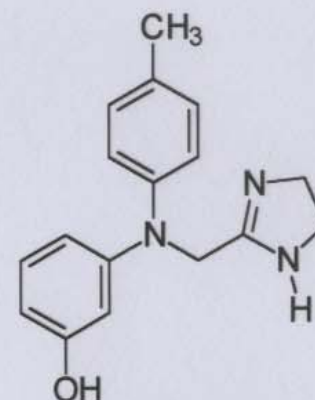
Therapeutic Option	Pros	Cons
1) Vacuum constriction device (for venous disorders)	Reliable, safe, cost-effective	Interruption of sexual act
2) Penile prosthetic implants (rigid, malleable, hinged, or inflatable)	Cosmetically satisfactory (especially inflatable implants)	Multiple surgical procedures usually necessary
3) Surgery (arterial balloon dilation, arterial bypass, or venous ligation)	If ED mostly due to vascular problems, is a semi-permanent solution	Risks of surgical procedure



ALPROSTADIL

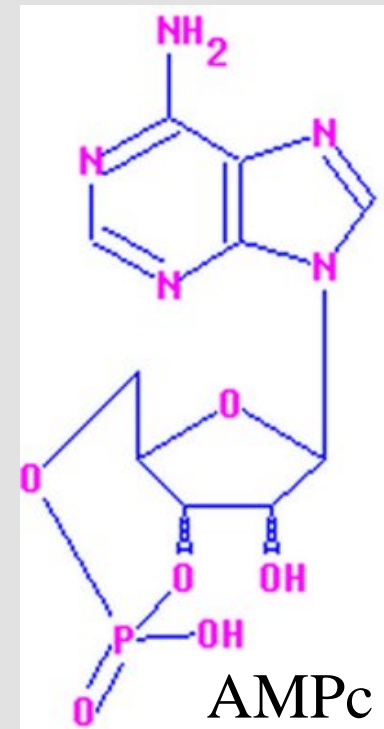
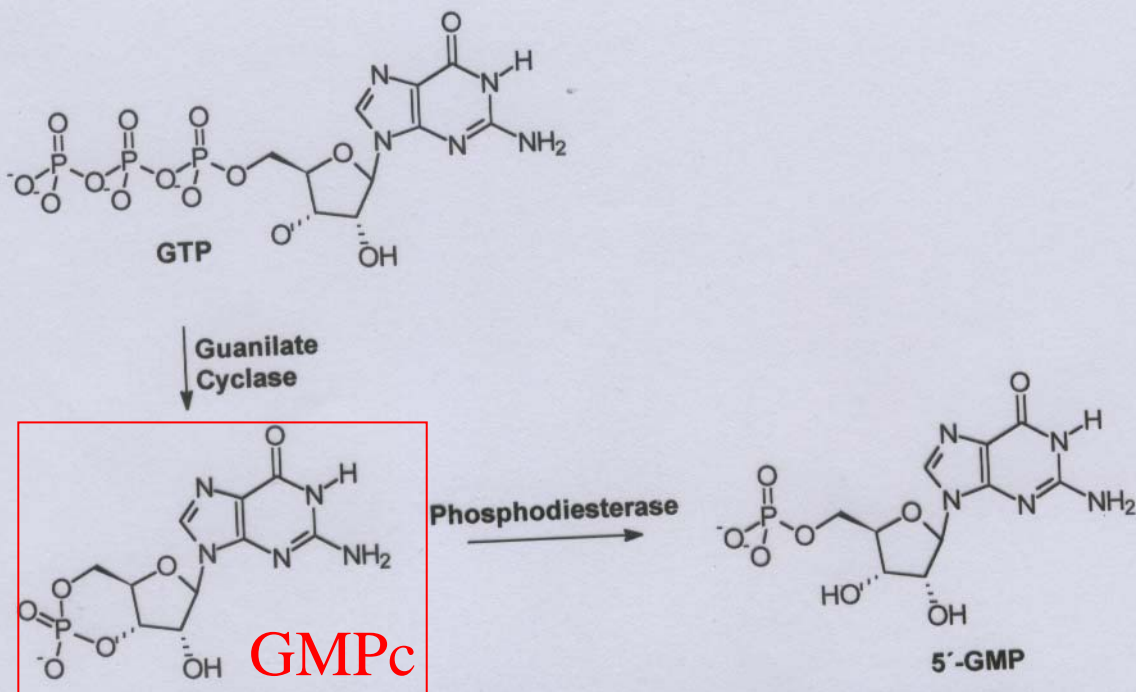
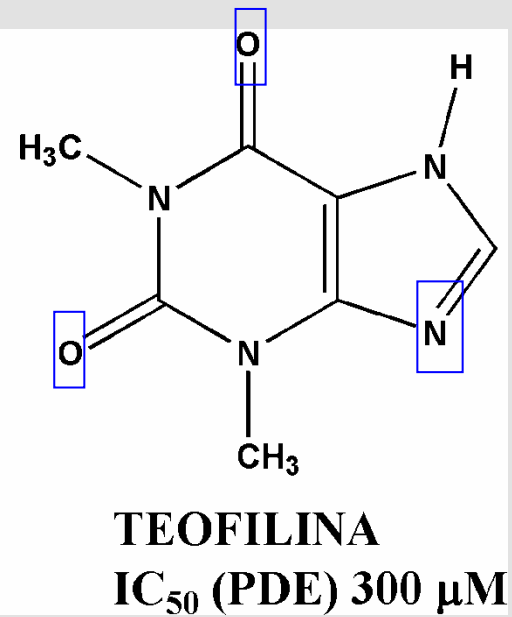
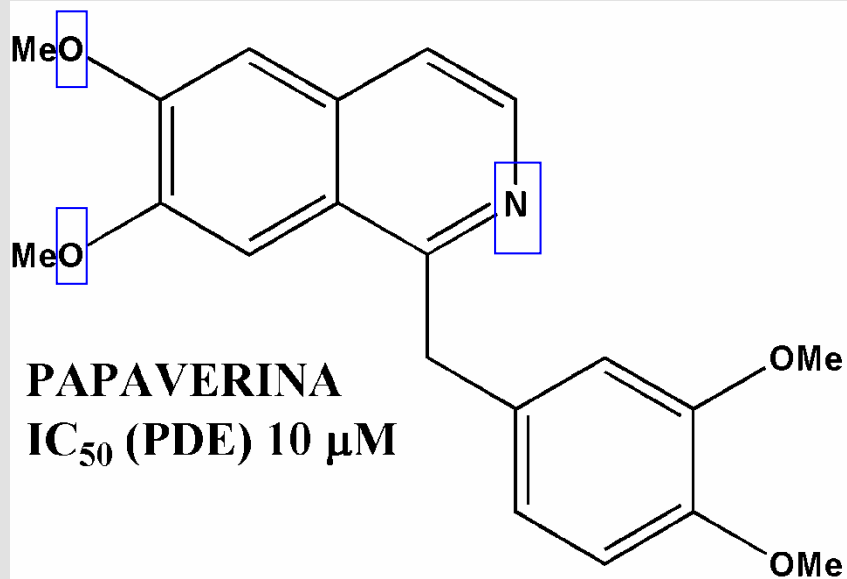


PAPAVERINA



FENTOLAMINA





# Fosfodiesterases

Table 2  
Phosphodiesterase (PDE) Types and Associated Organ Systems

PDE Family	Tissue Localization (Where Well Characterized)	
PDE-1	Brain, heart, kidney, liver, skeletal muscle, vascular and visceral smooth muscle	(Calmodulina Dependente)
PDE-2	Adrenal cortex, brain, corpus cavernosum, heart, kidney, liver, skeletal muscle, visceral smooth muscle	(Estimulada por GMPc)
PDE-3	Corpus cavernosum, heart, platelets, vascular and visceral smooth muscle, liver, kidney	(Inibida por GMPc)
PDE-4	Kidney, lung, mast cells, heart, skeletal muscle, vascular and visceral smooth muscle	(Específica para AMPc)
PDE-5	Corpus cavernosum, platelets, skeletal muscle, vascular and visceral smooth muscle	(Específica para GMPc)
PDE-6	Retina	(Específica para Fotoreceptores)

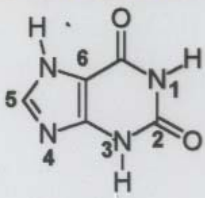
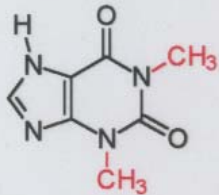
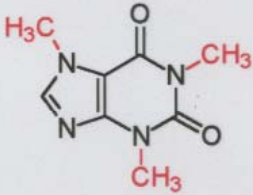
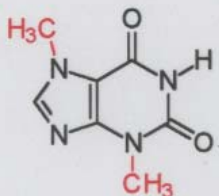
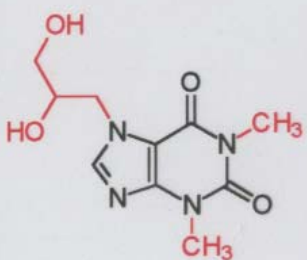
Adapted from Wallis RM et al. *Am J Cardiol.* 1999;83(5A):3C-12C."

# Isoformas de PDE

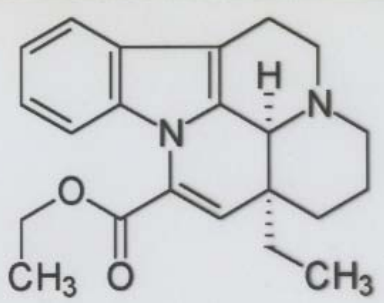
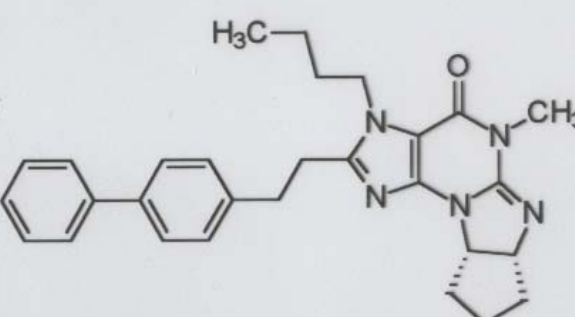
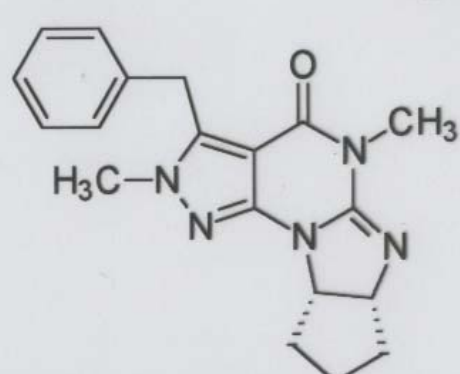
Table 1  
Phosphodiesterase Family Tree  
(11 Families, 21 Subfamilies, 53 Isoforms)

	Phosphodiesterase Family	Subfamily	Splice Variants
All discovered pre-sildenafil	1	A	4
	1	B	1
	1	C	5
	2	A	3
	3	A	1
	3	B	1
	4	A	8
	4	B	3
	4	C	4
	4	D	5
	5	A	3
	6	A	1
	6	B	1
	6	C	1
All discovered post-sildenafil	7	A	3
	7	B	1
	8	A	1
	8	B	1
	9	A	4
	10	A	2
	11	A	4

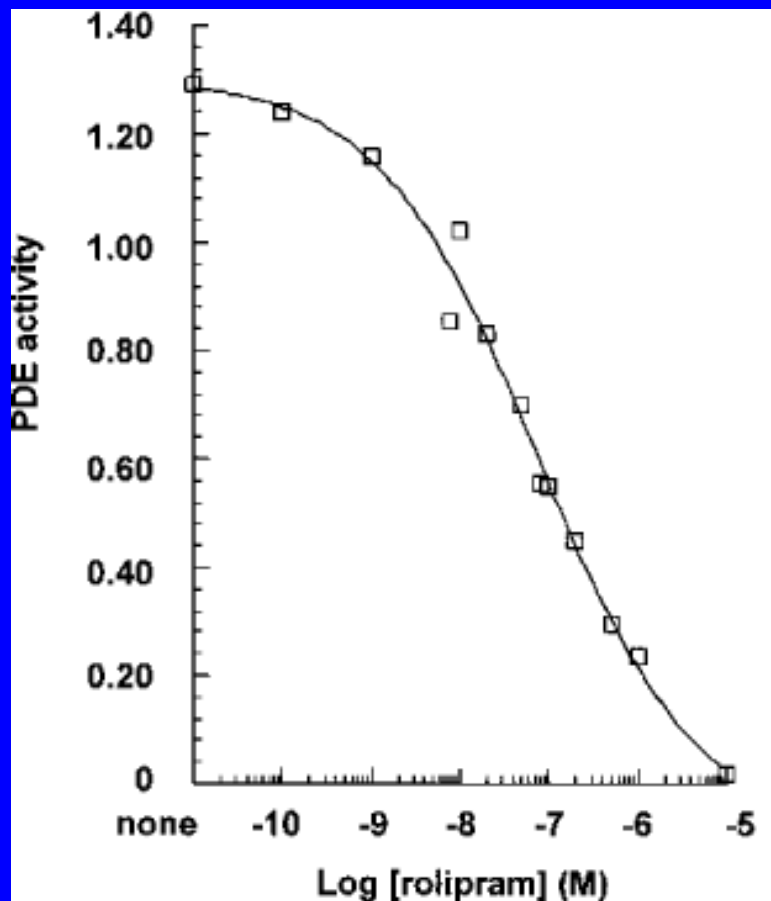
# Inibidores de PDE não-seletivos

Drug name	Structure	Clinical utility	Side effects/misc.
xanthine (prototype)		renal vasodilator (hypertension), relaxation of bronchial smooth muscle (asthma), increase in coronary blood flow	CNS (nervousness, restlessness, irritability, insomnia, nausea, vomiting, seizures), increase in heart rate
theophylline		used since 1930's as a bronchodilator. Inhibits the late-phase inflammation of asthma	orally active
caffeine		causes cerebral vasoconstriction (included in many headache medicines)	
theobromine		low potency	as name suggests, found in tea
dyphylline		bronchodilator (asthma) though less potent than theophylline	discovered in 1946. Less nausea than theophylline

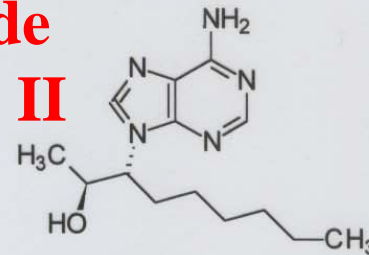
# Inibidores de PDE I

Compound name	Structure	IC50 (PDE-I) (nM)	IC50 (PDE-III) (nM)	IC50 (PDE-V) (nM)
vinpocetine		19,000	> 300,000	> 300,000
# 31		0.07	3500	305
4b		55	80,000	100

# Inibidores de PDE seletivos

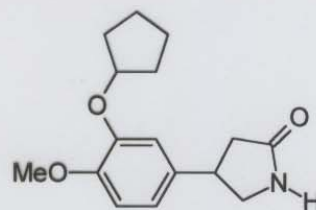


## Inibidores de Fosfodiesterase II

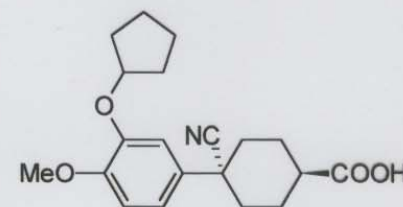


erythro-9-(2-hydroxyl-3-nonyl) adenine  
(EHNA)

## Inibidores de Fosfodiesterase IV

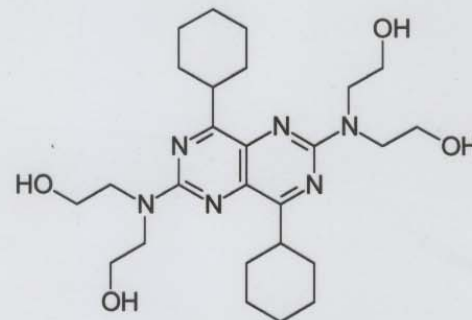


ROLIPRAM



SB207499

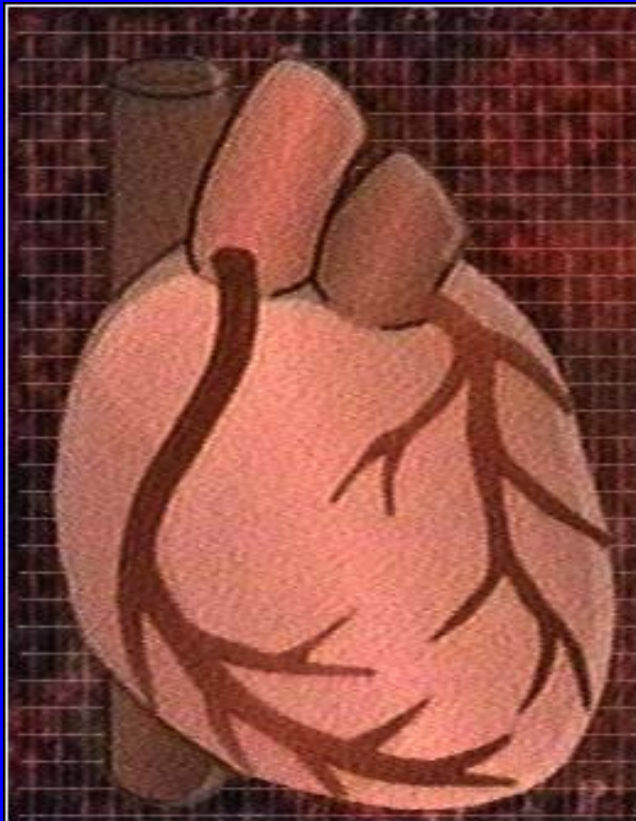
## Inibidores de Fosfodiesterase V



DIPYRIDAMOLE



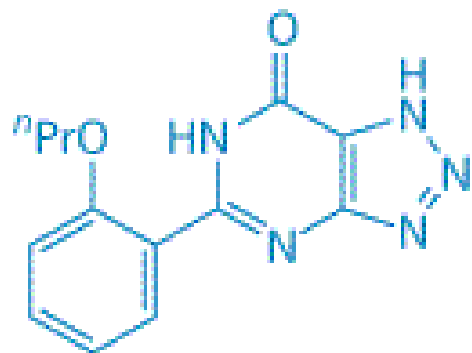
# Inibidores Seletivos de PDE III



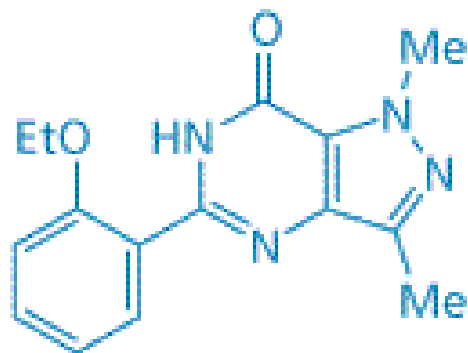
(CNN)

Chemical class	Example	Structure
Bipyridines	amrinone	
Imidazolones	enoximone	
Imidazolines	CI930	
Dihydropyridazinones	indolidan	
Dihydroquinilones	cilostamide	

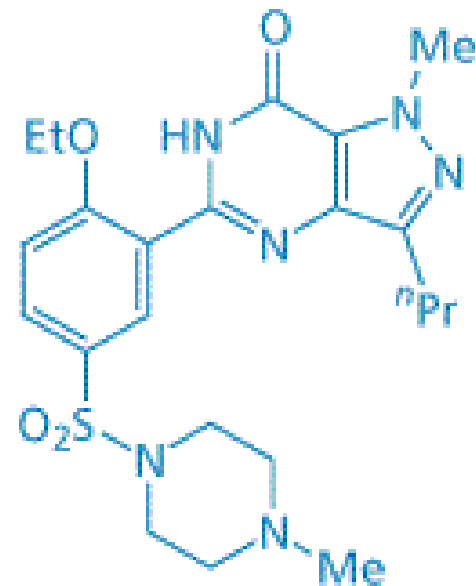
# Zaprinast X Sildenafil



(1) Zaprinast



(2) Pyrazolopyrimidinone

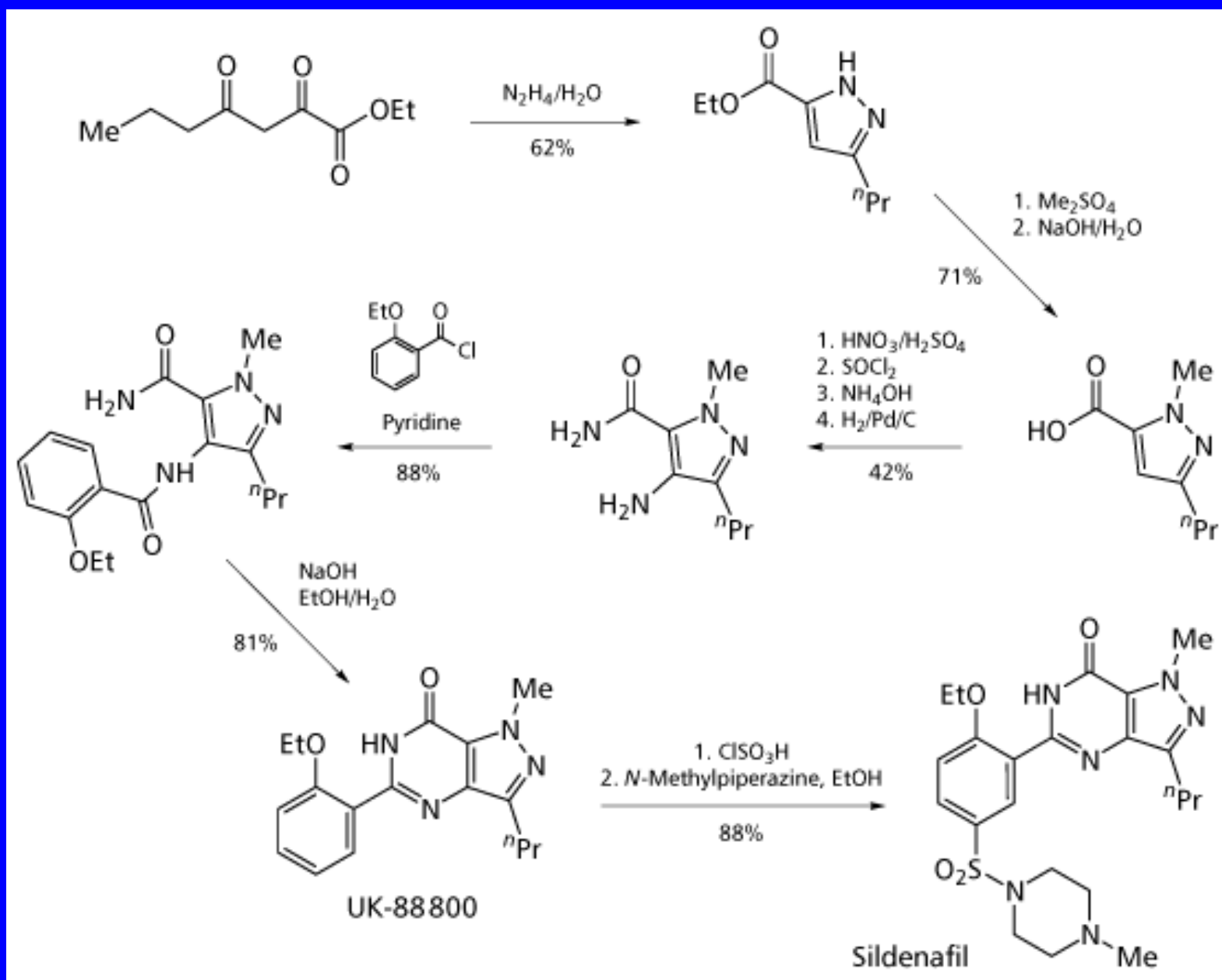


(3) Sildenafil

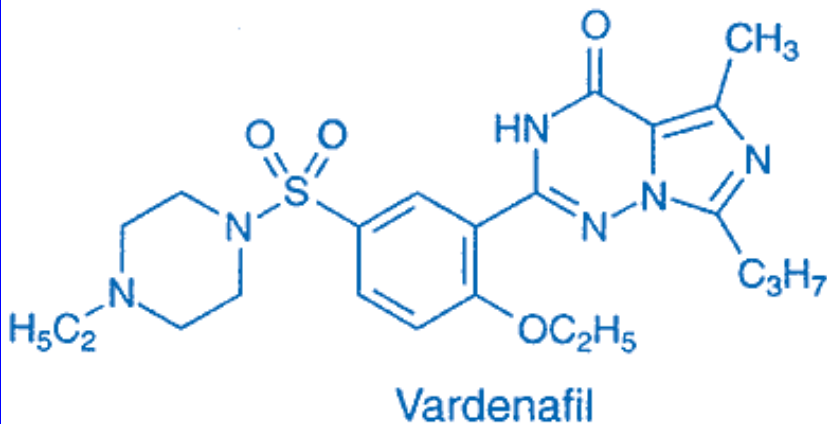
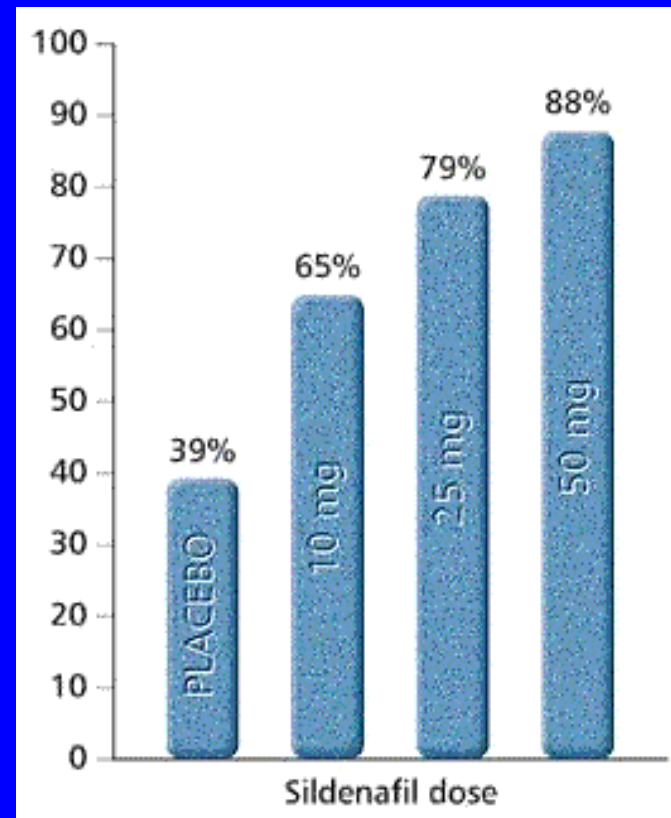
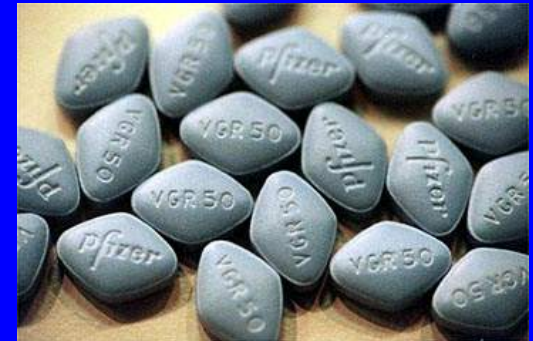
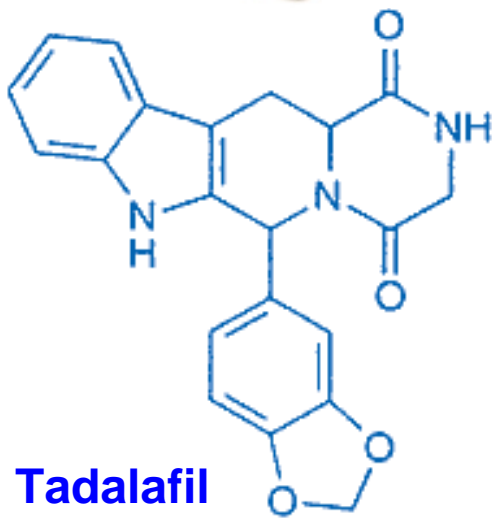
Isozyme	$\text{IC}_{50}$ (nM)
PDE I	9400
PDE III	> 100.000
PDE V	2000

Isozyme	$\text{IC}_{50}$ (nM)
PDE I	260
PDE III	65.000
PDE V	3

# Síntese do Sildenafil

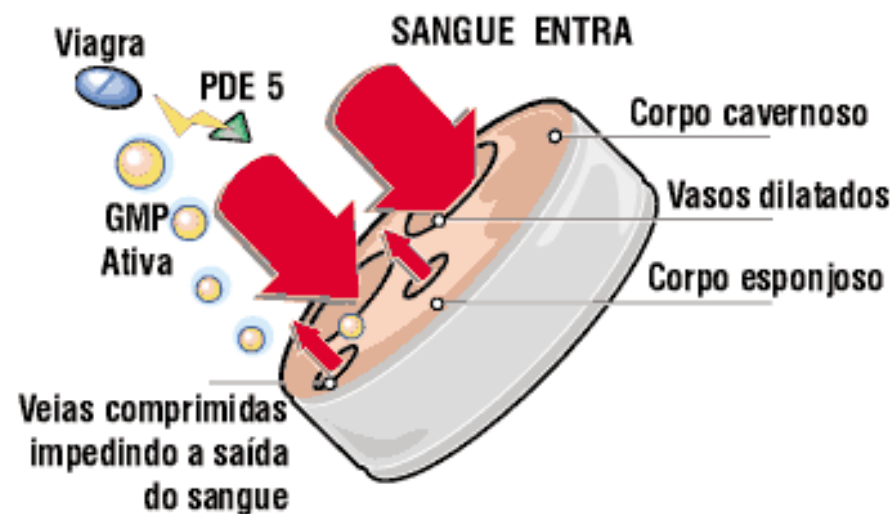
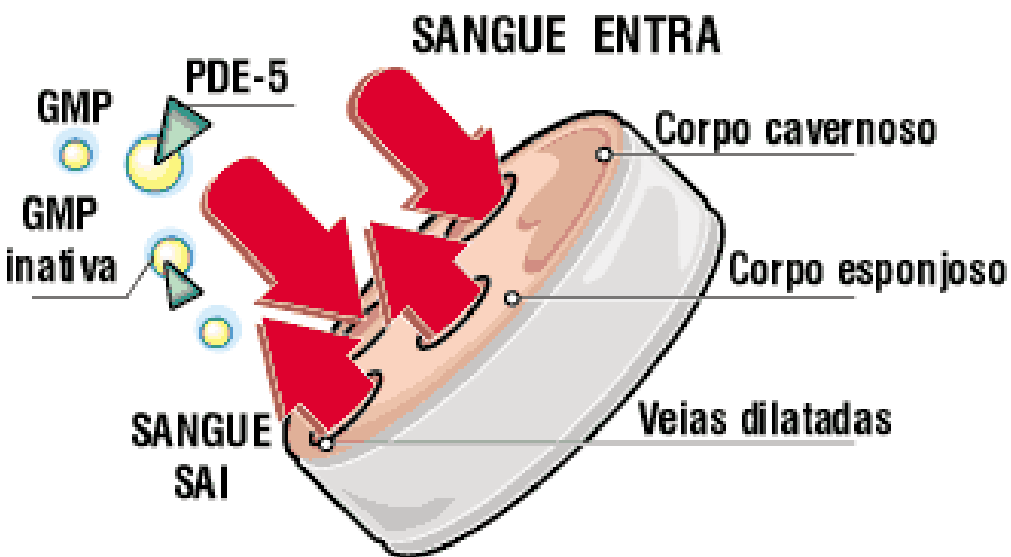
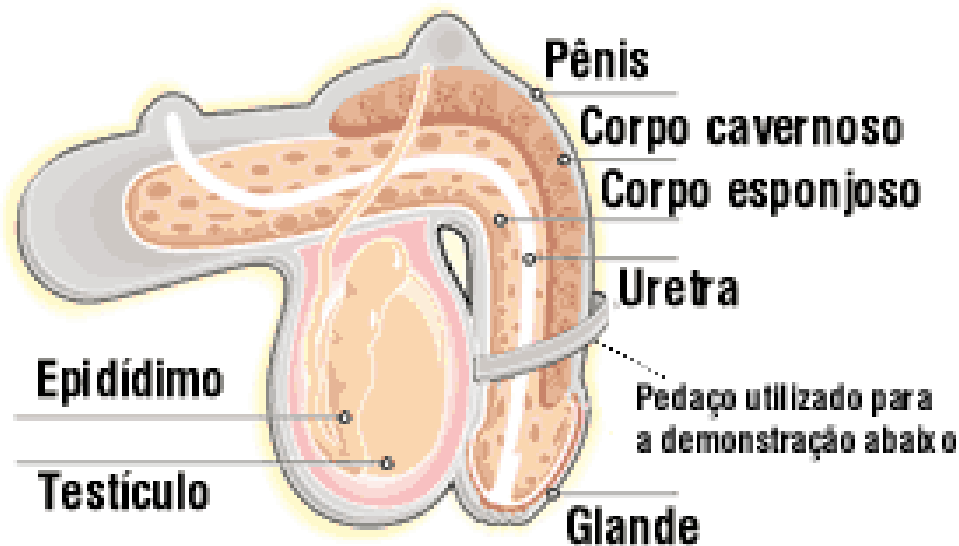


# Sildenafil e Outros Inibidores de PDE 5 seletivos anti-DE



**Levitra<sup>R</sup>**

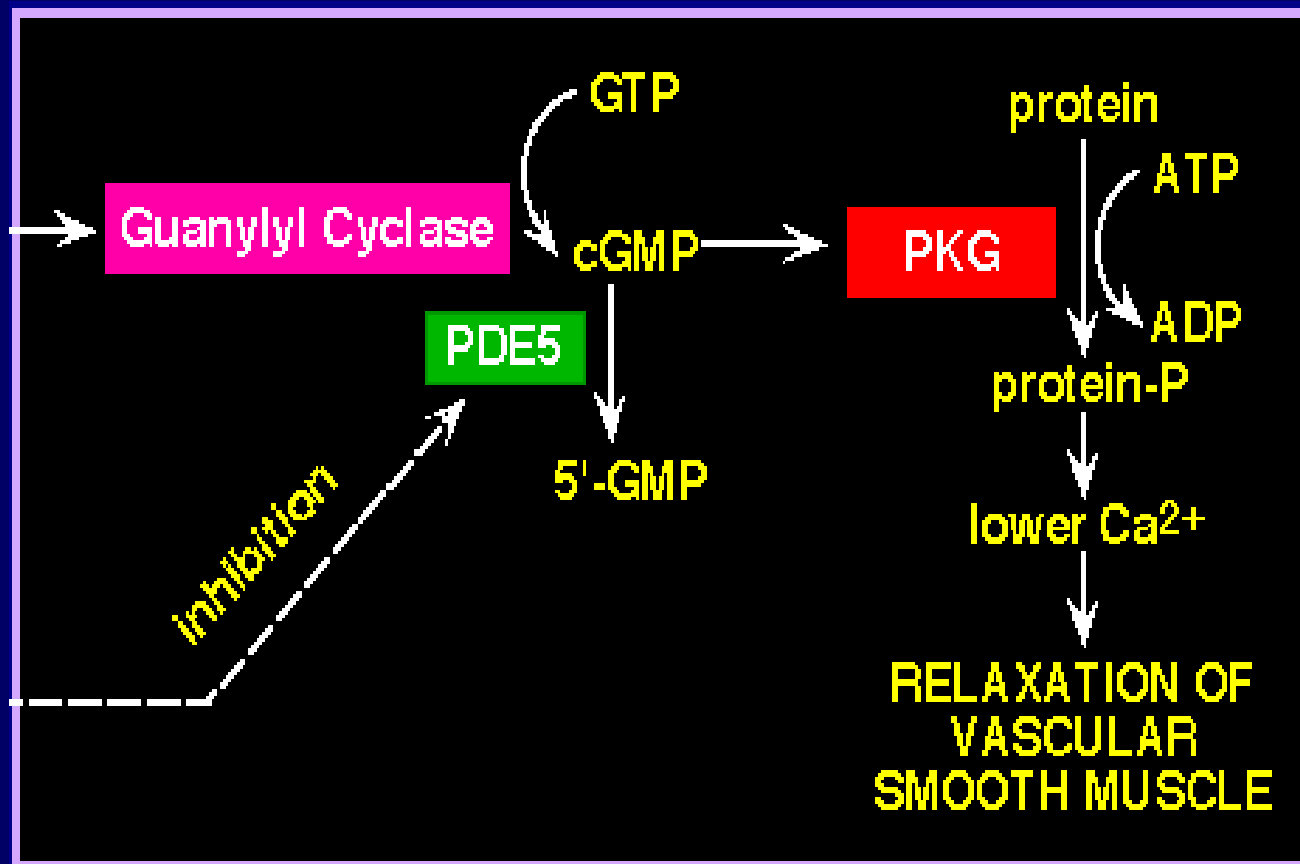
# COMO O VIAGRA ATUA



# Sildenafil e a Regulação do Relaxamento da Musculatura Lisa

Neurons or Endothelium

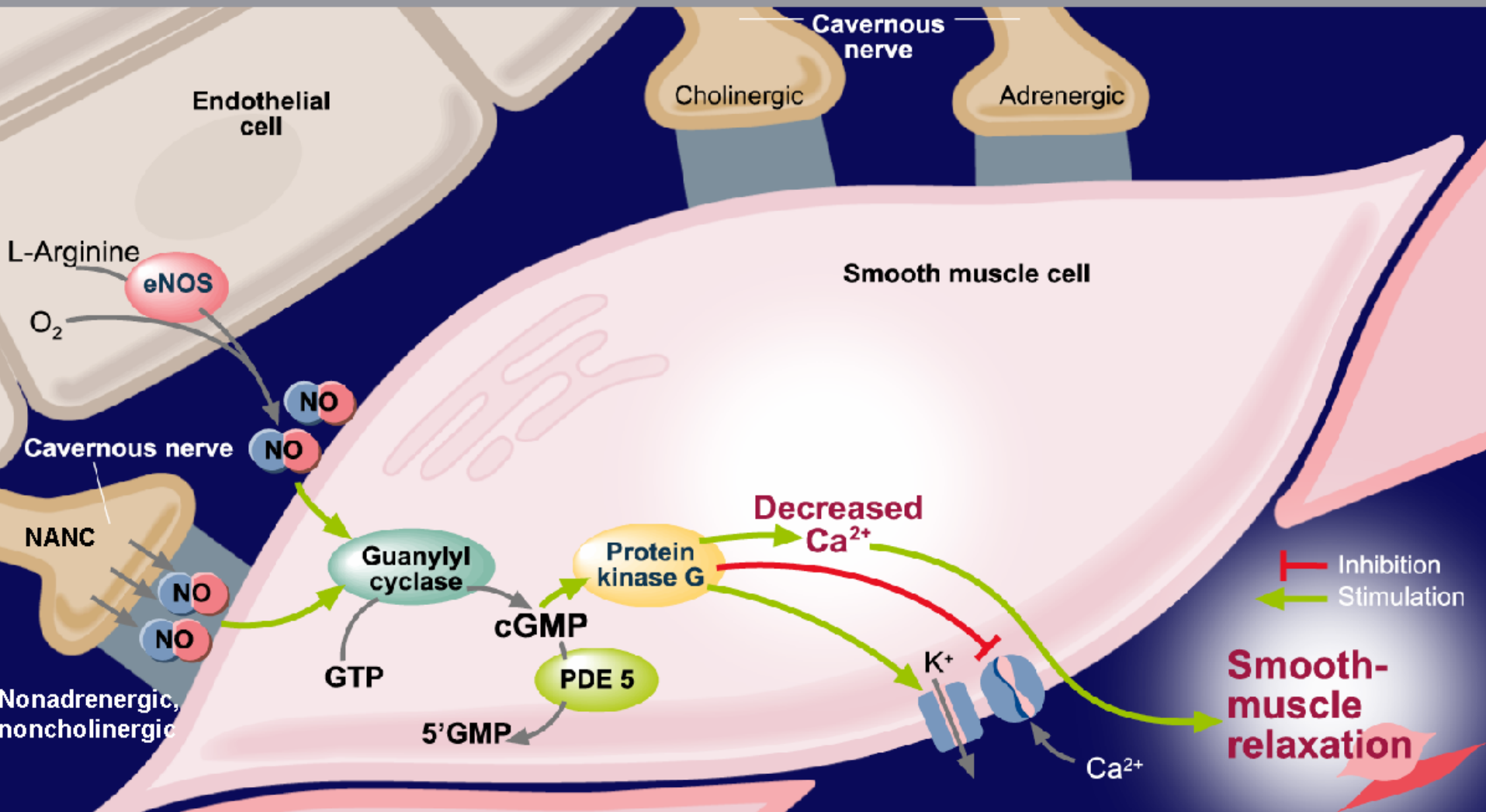
Nitric Oxide



Sildenafil



# Mecanismo de Ereção Peniana dependente de NO/cGMP





**Pharmacology/  
Biochemistry**  
**BIOLOGICAL  
PROPERTIES**

**Evaluation of the Bioactivity  
Profile:  
SAR, SAFIR  
Lead-Compound  
Identification**



**Organic Chemistry  
(Computational)**  
**STRUCTURAL  
PROPERTIES**

**Medicinal  
Chemistry**

**Evaluation of the Bioactivity  
Profile:  
Lead-Compound  
Structural Optimization**



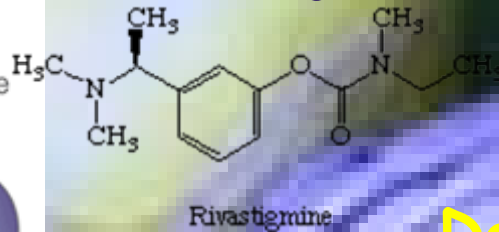
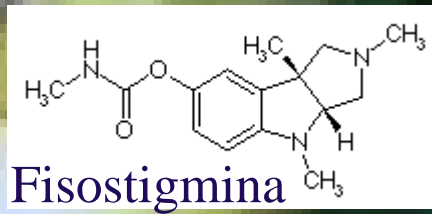
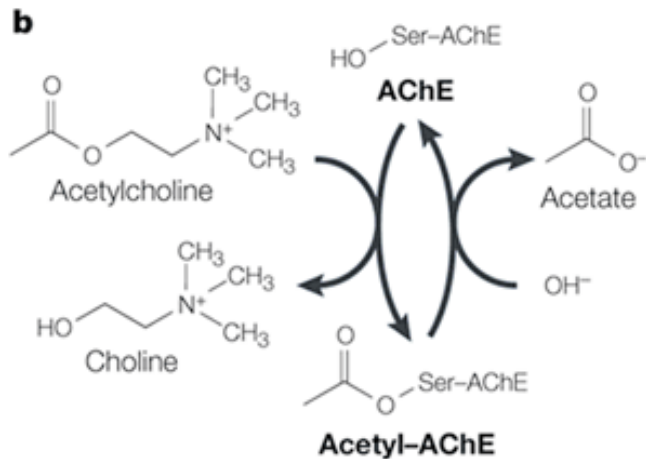
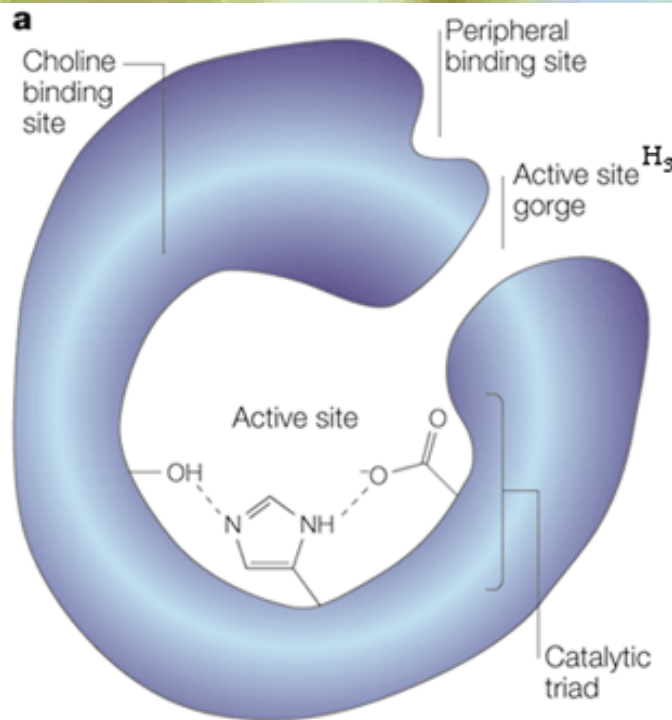
**Organic Chemistry  
(Synthetic)**  
**CHEMICAL REACTIVITY**

**Structural Design:  
Bioisosterism,  
Molecular Hybridization,  
Simbiosis**

**New Chemical Entities**

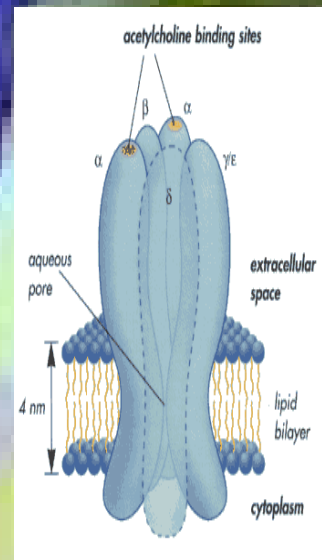


# Novos Inibidores de Acetilcolinesterase



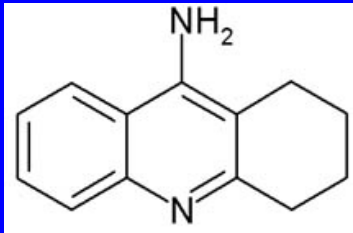
Doenças  
Neuro-Degenerativas

Mal de Alzheimer



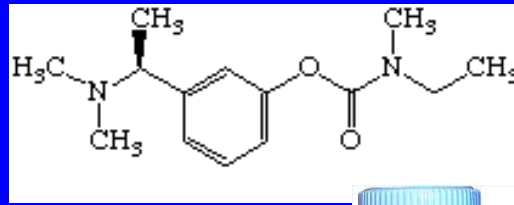


# Inibidores de AChE

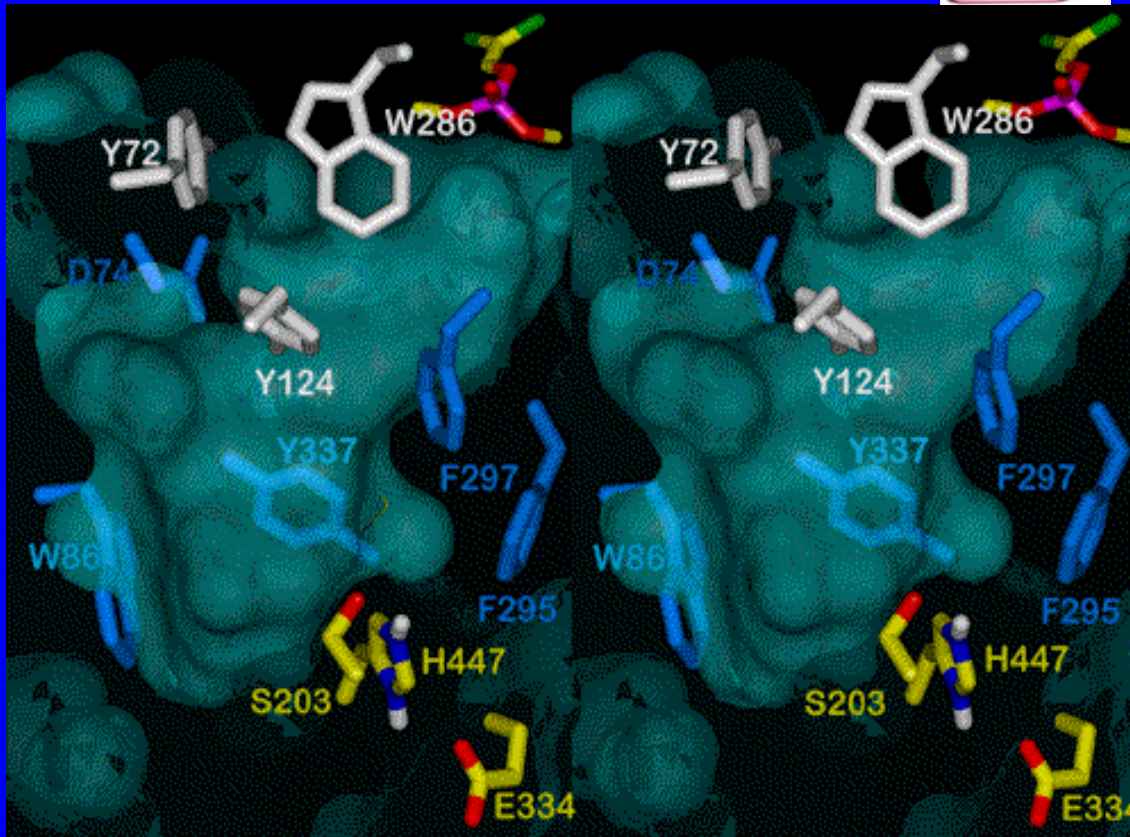
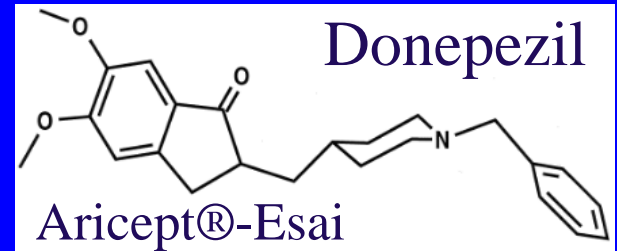


Tacrine

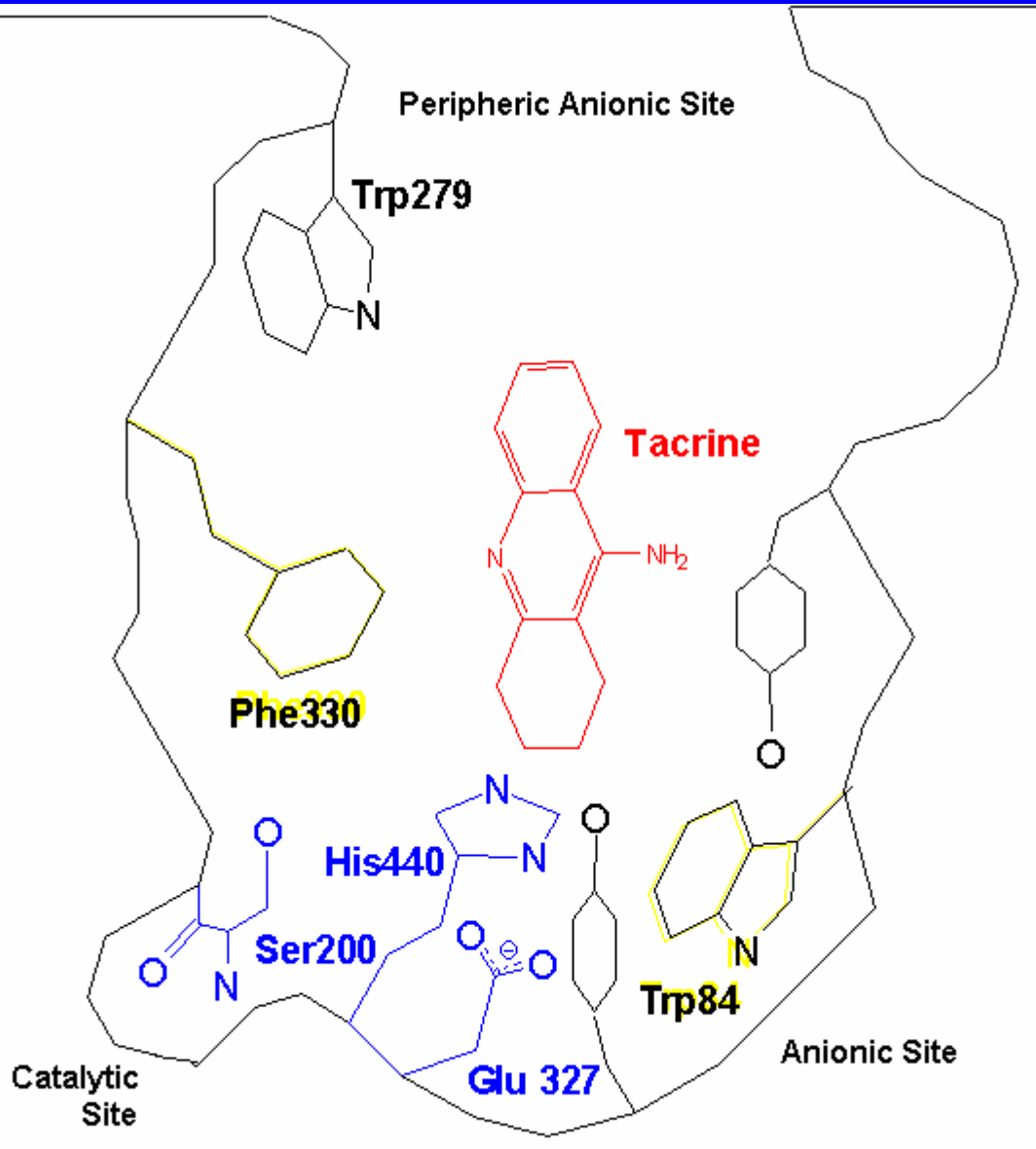
Cognex®-Parke Davis



Rivastigmina



# ACETYLCHOLINESTERASE (AChE)



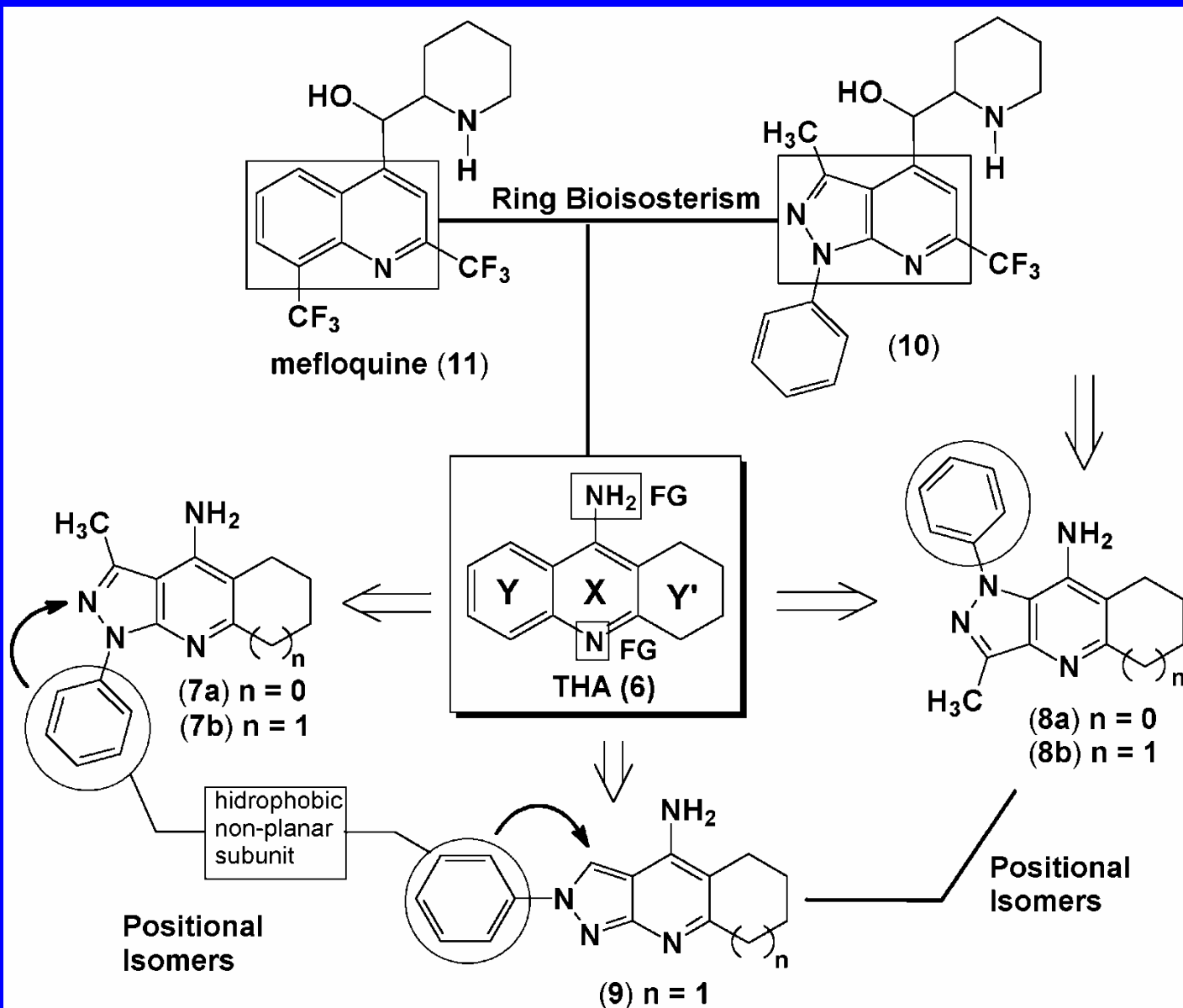
## 4 Domains:

- **Estearatic Site - Catalytic Triad:**
- **Ser200, His440 e Glu327.**
- **Anionic Site:  $\geq 4,7\text{\AA}$  of Ser200**
- **Hydrophobic Domains**
- **Peripheral Anionic Site:**
- **$\pm 20\text{\AA}$  of Estearatic Site**

Quinn, D. M. (1987) Acetylcholinesterase: Enzyme, structure, reaction, and virtual transition states, *Chem. Rev.* **87**, 955-979.

# Novos Derivados Pirazolopiridínicos



## Inibidores de AChE



Câmara, C. A. (2000)  
Tese de Doutorado,  
IQ-UFRJ / LASSBio

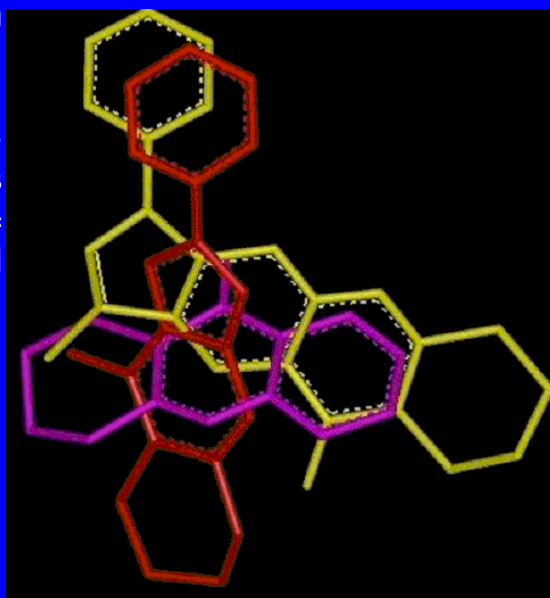


# Atividade e Seletividade Anticolinesterásica

Compound	Total Cholinesterase <sup>b</sup>		Butyrylcholinesterase		SI <sup>c</sup>
Tacrine (6)	0.16 ± 0.03	(4)	0.24 ± 0.10	(2)	1.5
LASSBio-504	31.19 ± 4.23	(2)	 LASSBio-501	 LASSBio-560	
LASSBio-505	26.71 ± 3.13	(3)			
LASSBio-503	28.49 ± 0.31	(3)			
LASSBio-506	32.17 ± 4.39	(2)			
LASSBio-502	23.24 ± 5.30	(3)			
<b>LASSBio-501</b>	<b>6.39 ± 0.86</b>	<b>(3)</b>	<b>133.80 ± 14.51</b>	<b>(2)</b>	<b>20.9</b>
LASSBio-560	6.01 ± 1.22	(3)	31.90 ± 10.09	(2)	5.3
Gаланthamine (1)	3.10 ± 0.18	(2)			

<sup>a</sup>Values are means ± S.E.M. of the IC<sub>50</sub> from the indicated number of animals in parenthesis; <sup>b</sup>Total cholinesterase (ChE) and butyrylcholinesterase (BuChE) activities were determined by Ellman's method,<sup>26</sup> as described in the Experimental section; <sup>c</sup>SI = Selectivity Index = IC<sub>50</sub> BuChE / IC<sub>50</sub> Total ChE ratio.

Barreiro, E. J. *et al.* (2003)  
*J. Med. Chem.* **46**, 1144.



Dinâmica  
Molecular  
Ligante-AChE

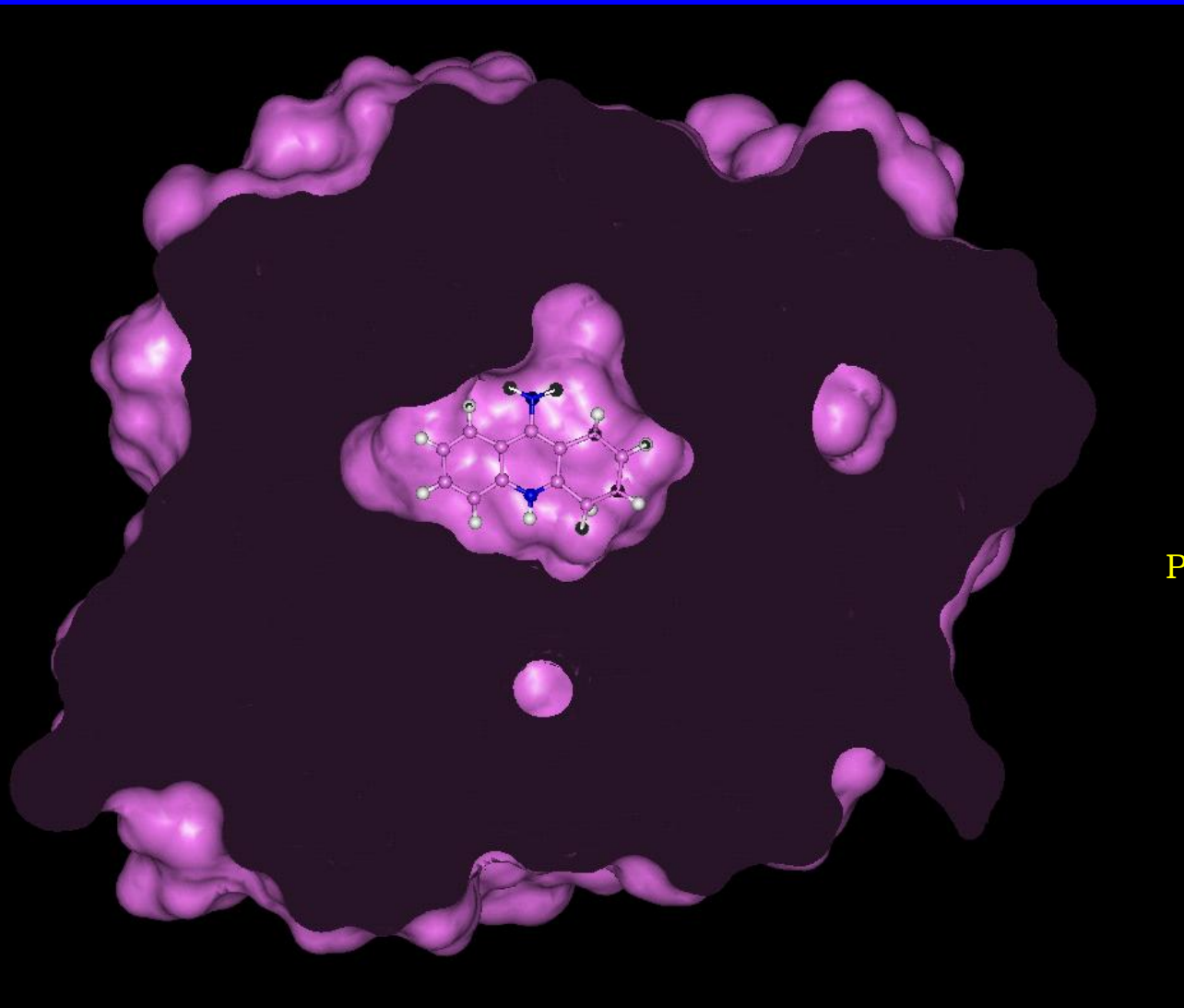
# Molecular Dynamics of Acetylcholinesterase-THA Complex



*Torpedo californica*  
Acetylcholinesterase  
(PDB-1ACJ, 2.8 Å)

Harel. M. *et al.* (1993)  
Proc. Natl. Acad. USA 90, 9031.

**Cut-off 16Å**  
**600 ps Molecular**  
**Dynamics Simulation**



# Molecular Dynamics of Acetylcholinesterase-THA Complex

RMS = 1,55Å

Phe330

(-8,22\*)

His440

(-3,15\*)

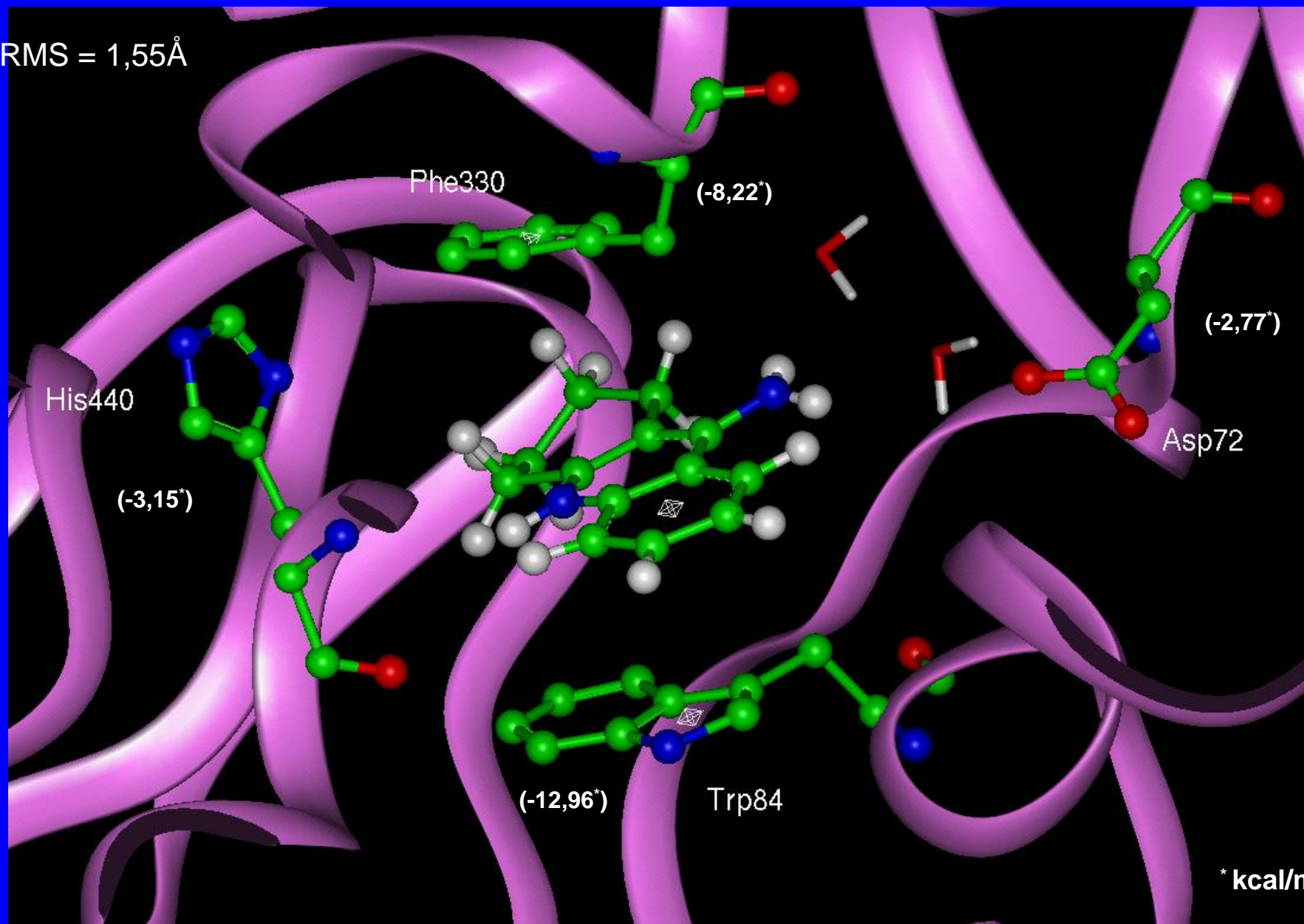
(-2,77\*)

Asp72

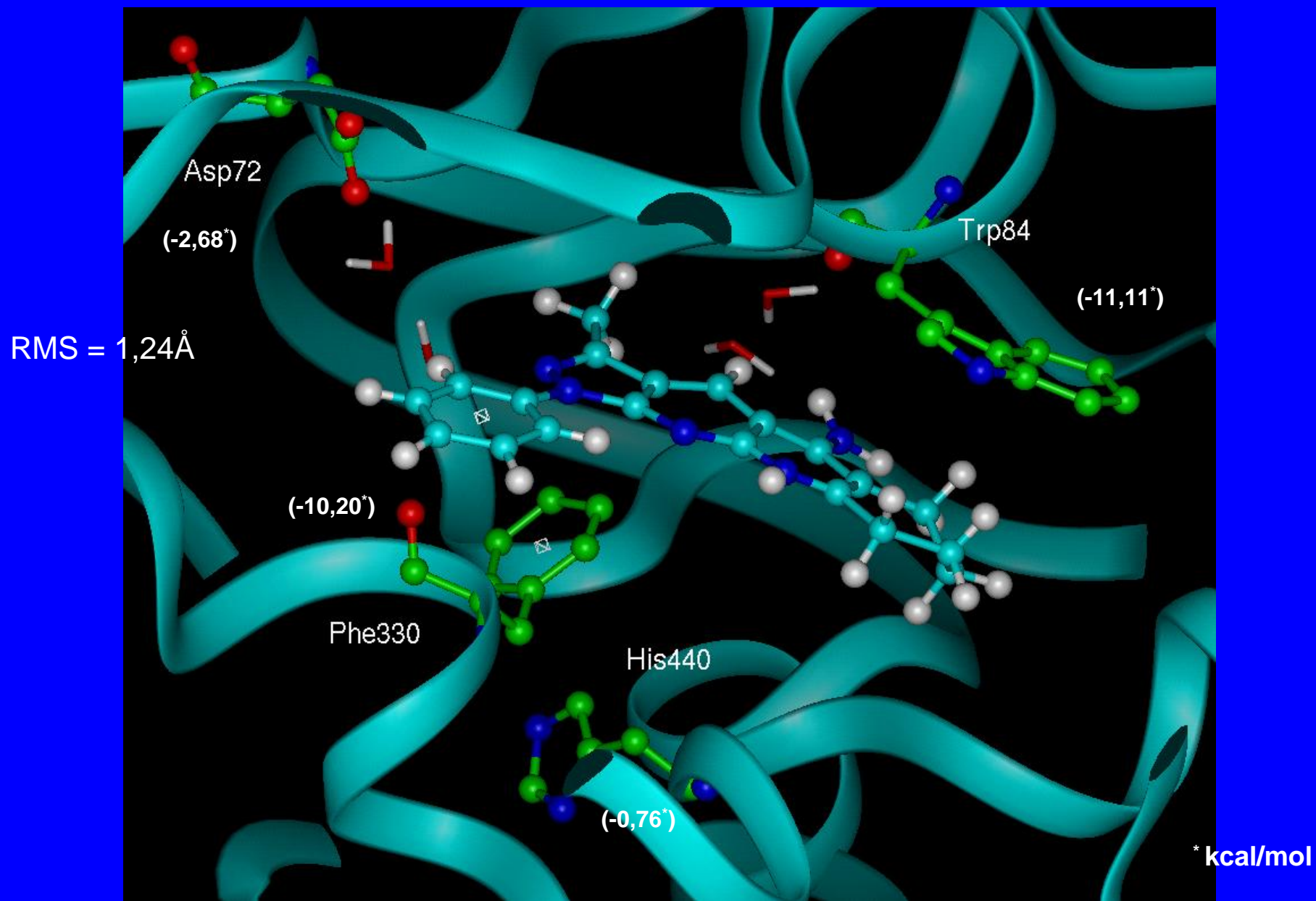
(-12,96\*)

Trp84

\* kcal/mol



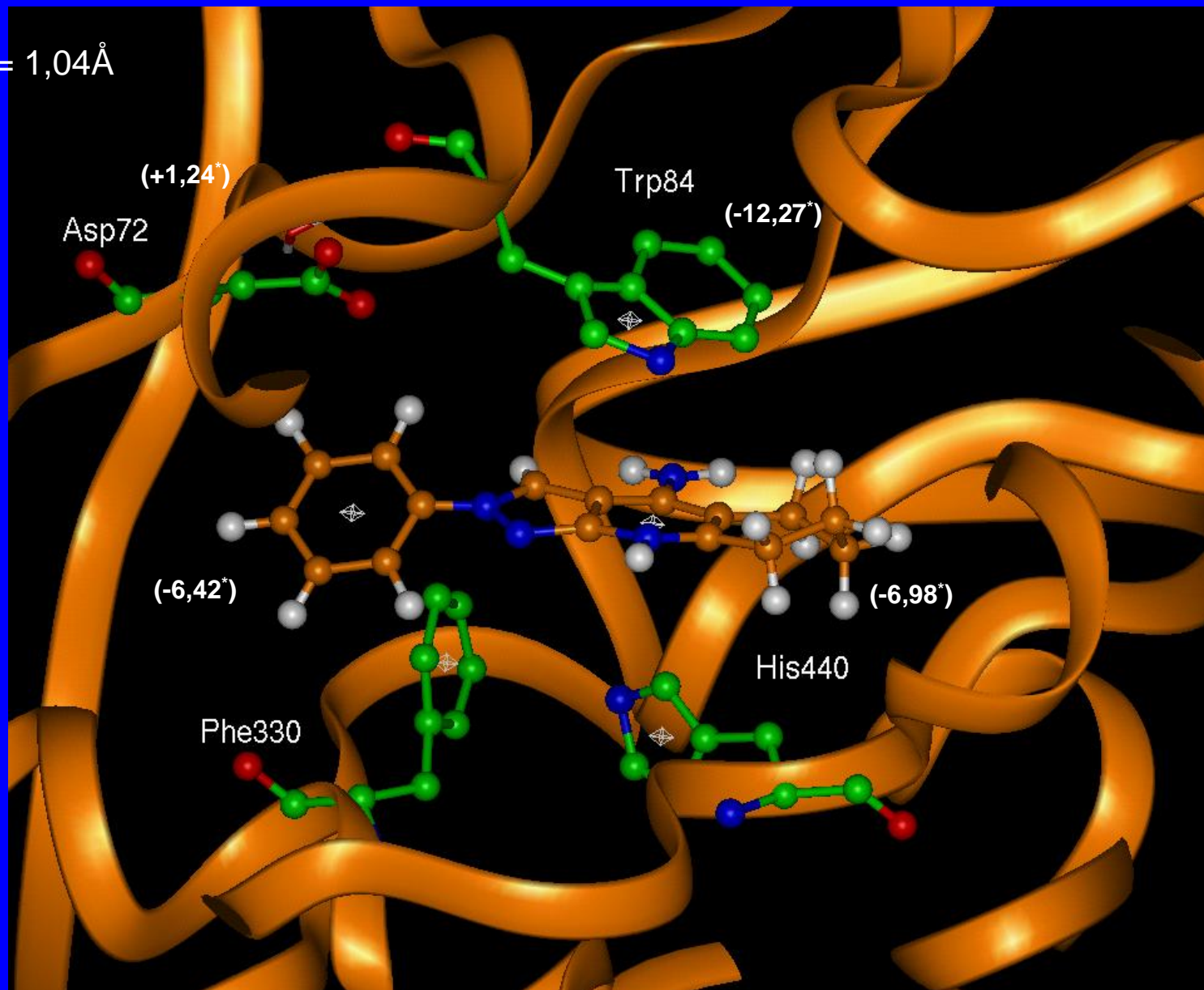
# Molecular Dynamics of Acetylcholinesterase-LASSBio501 Complex





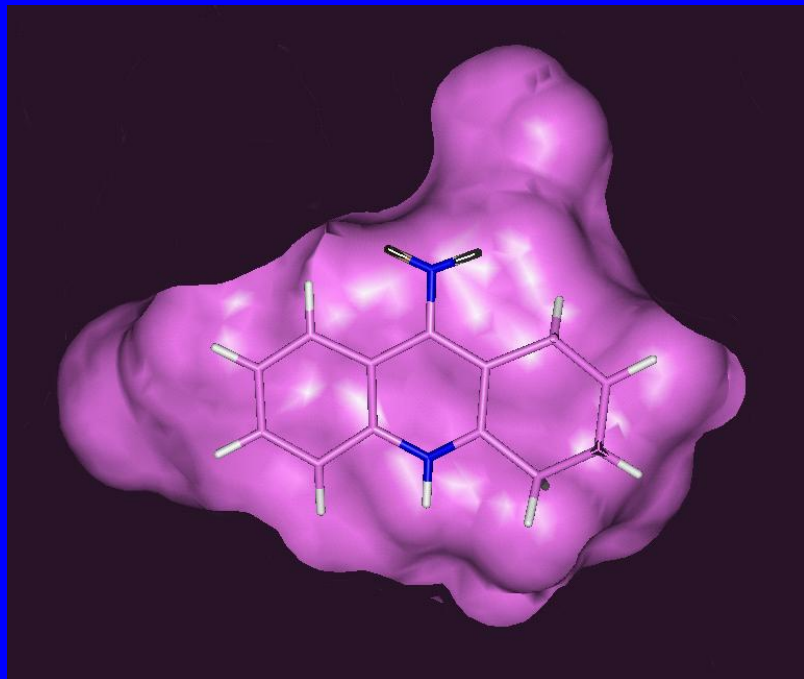
# Molecular Dynamics of Acetylcholinesterase-LASSBio560 Complex

RMS = 1,04Å

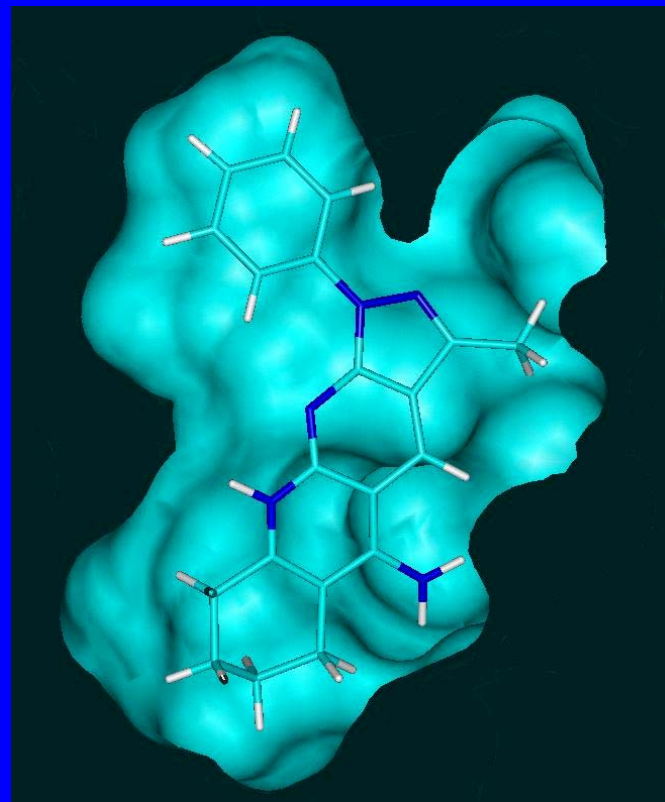


\* kcal/mol

# Comparison of Active Site Shape of AchE Complexed with THA and LASSBio-501



**Tacrine**

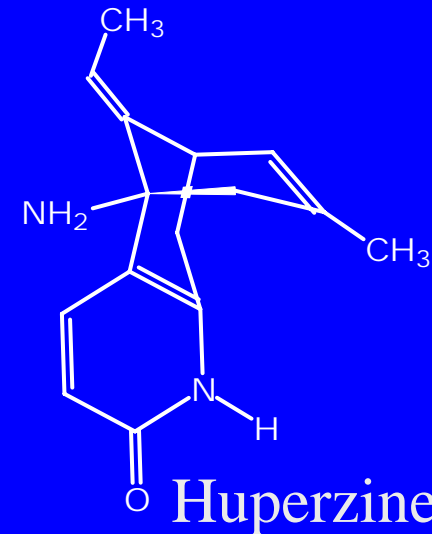
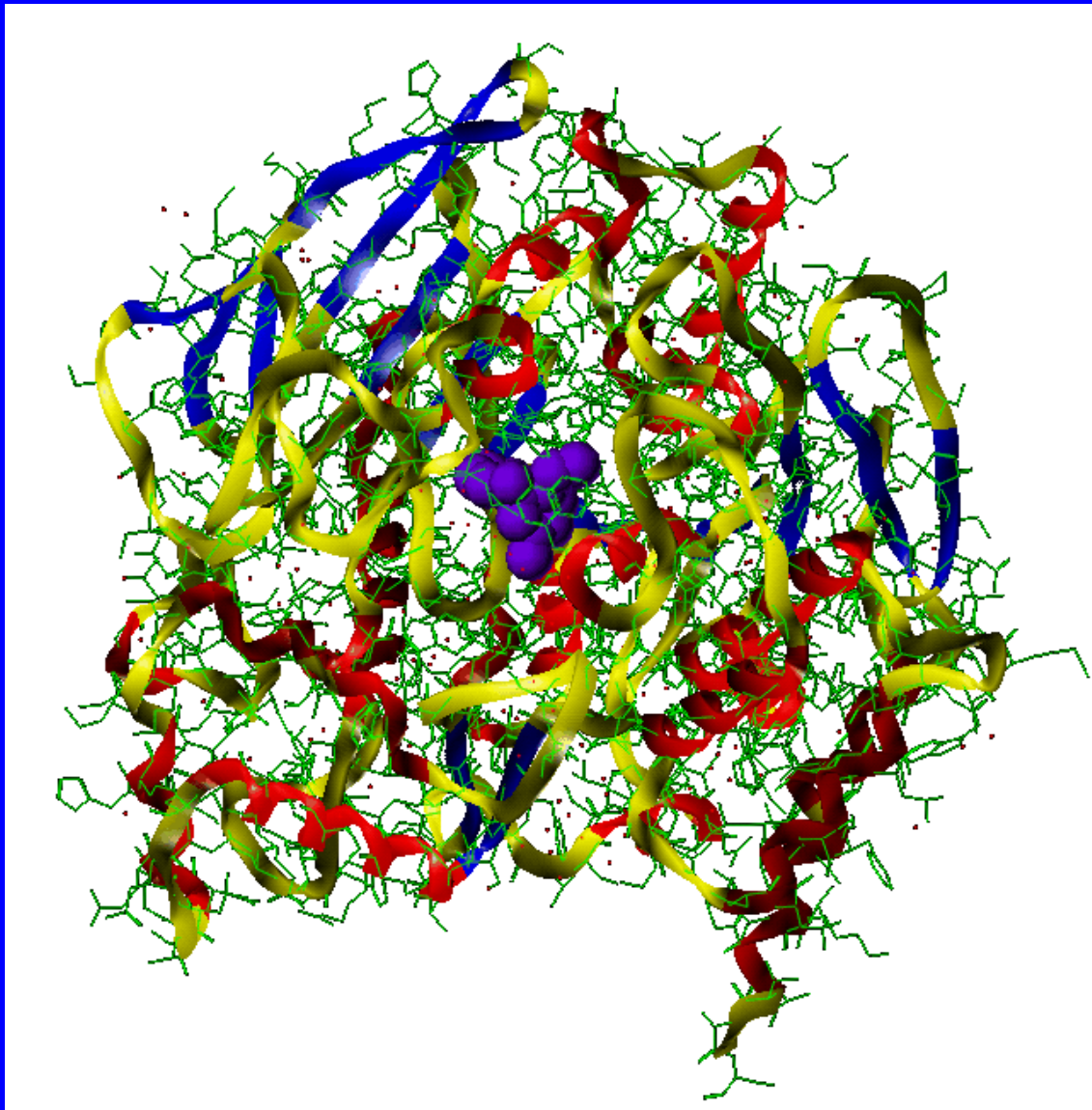


**LASSBio-501**

Eliezer J. Barreiro, Celso A. Camara, Hugo Verli, Leonora Brazil-Más, Newton G. Castro, Wagner M. Cintra, Yasco Aracava, Carlos R. Rodrigues and Carlos A. M. Fraga, DESIGN, SYNTHESIS AND PHARMACOLOGICAL PROFILE OF NOVEL FUSED PYRAZOLO[4,3-d]PYRIDINE AND PYRAZOLO[3,4-b][1,8]NAPHTHYRIDINE ISOSTERES: A NEW CLASS OF POTENT AND SELECTIVE ACETYLCHOLINESTERASE INHIBITORS, *J. Med. Chem.* 46 (7), 1144-1152 (2003) .



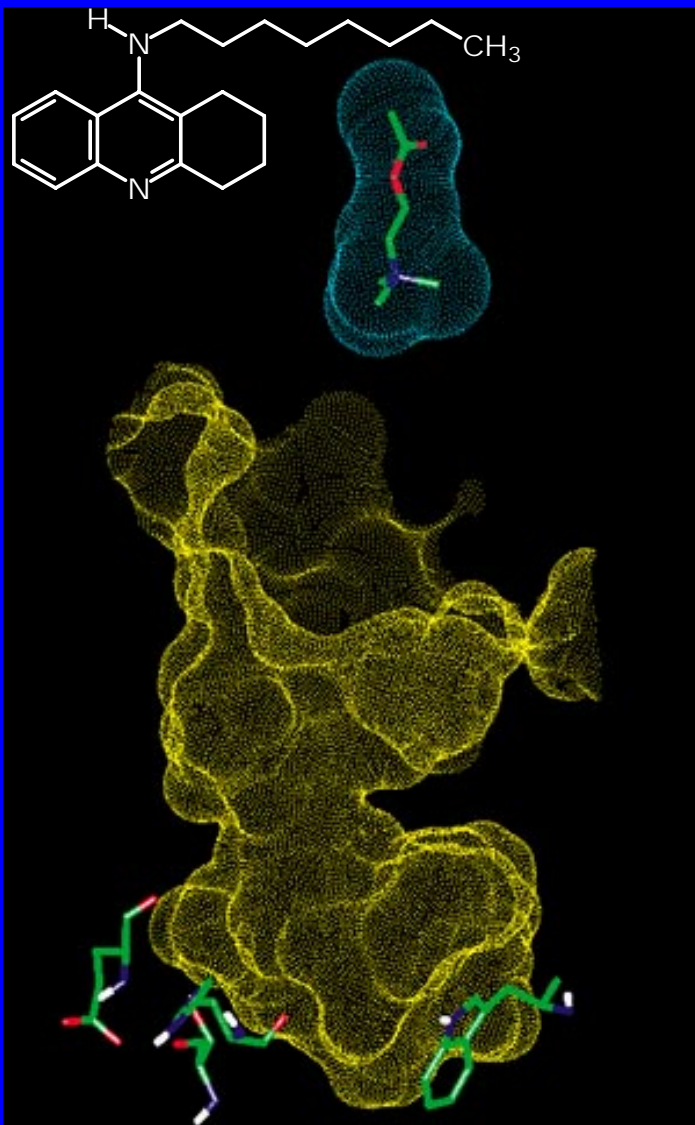
# Otimização Estrutural de Protótipos de Inibidores de AChE Naturais



Huperzine A  
(*Huperzia serrata*)



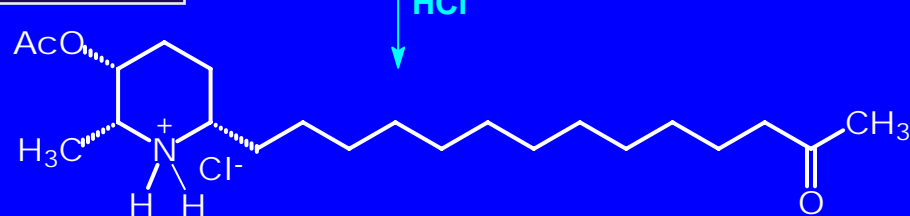
# Otimização Estrutural de Protótipos de Inibidores de AChE Naturais



*Cassia leptophylla*

Analgesia  
Capsaicina ???

Modificação  
Estrutural  
(SAR)



LASSBio-767

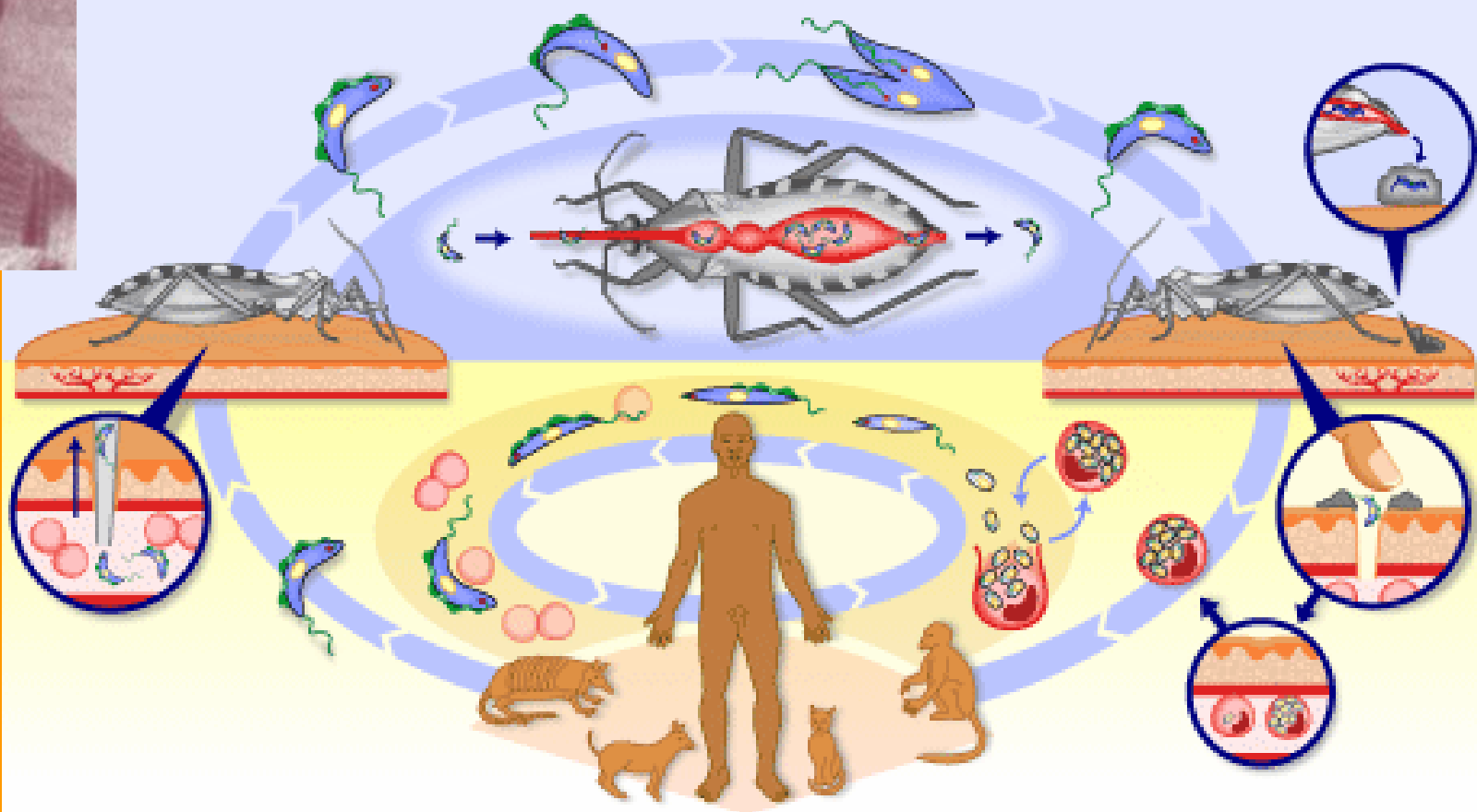
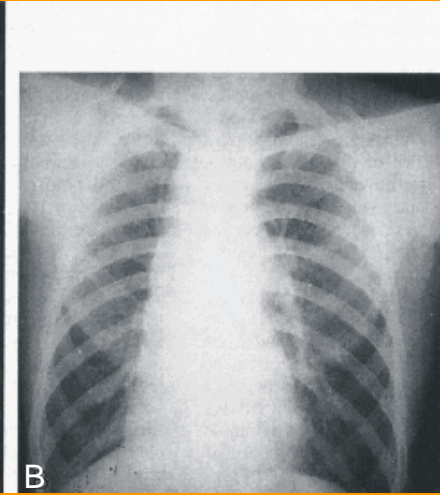
IC<sub>50</sub> = 4 μM (AChE)

# Doença de Chagas

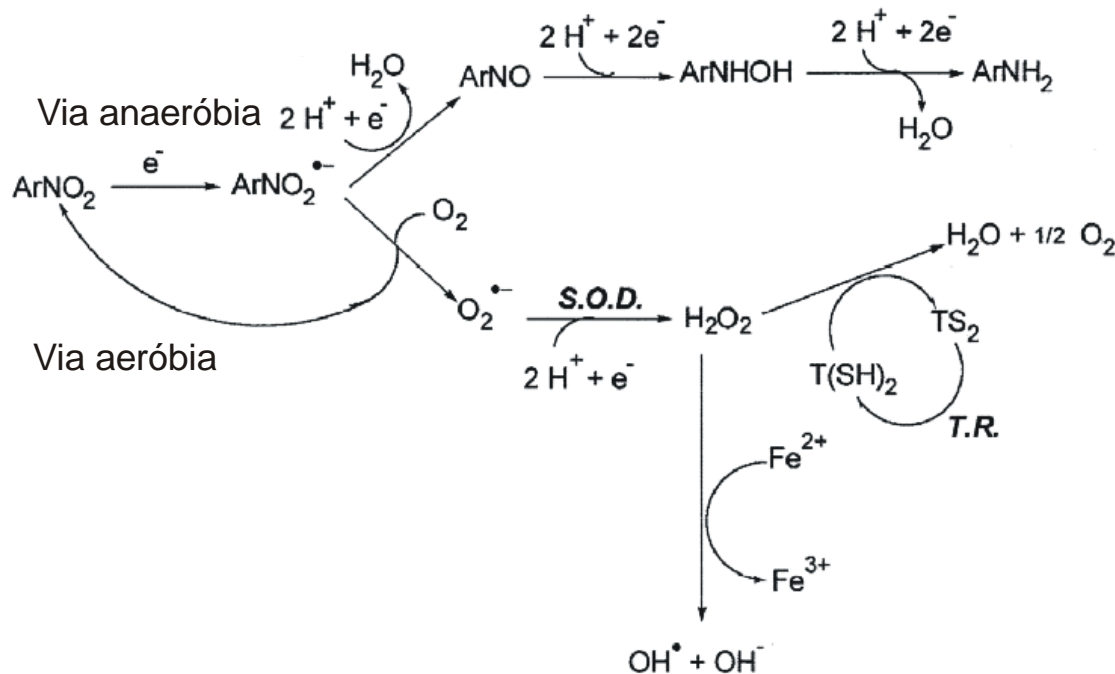
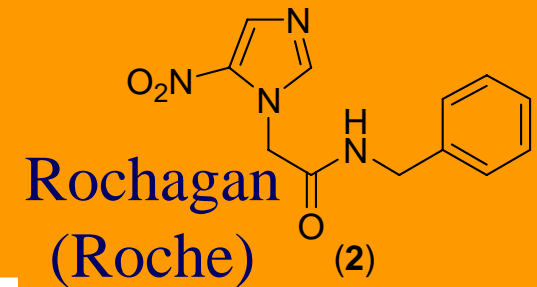
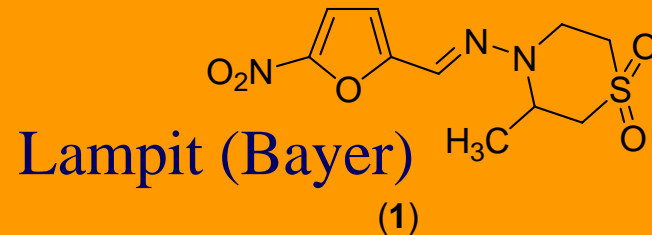
Carlos Chagas,  
1909



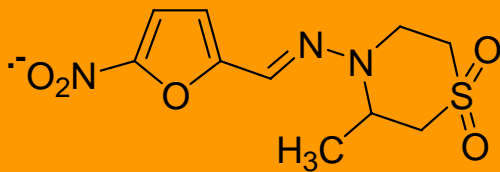
Mortalidade produzida por  
enfermidades endêmicas no Brasil  
1980-1990



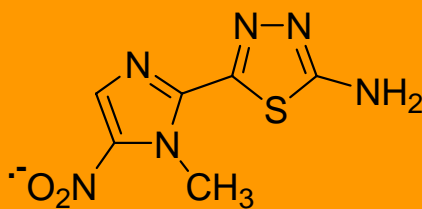
Desde o final de 1960 início dos anos 70 dois fármacos têm sido usados no tratamento da doença de Chagas: nifurtimox (1) e benzonidazol (2).





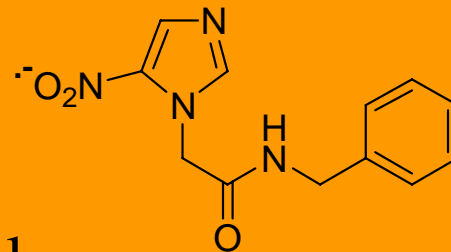


(A)



(B)

Megazol

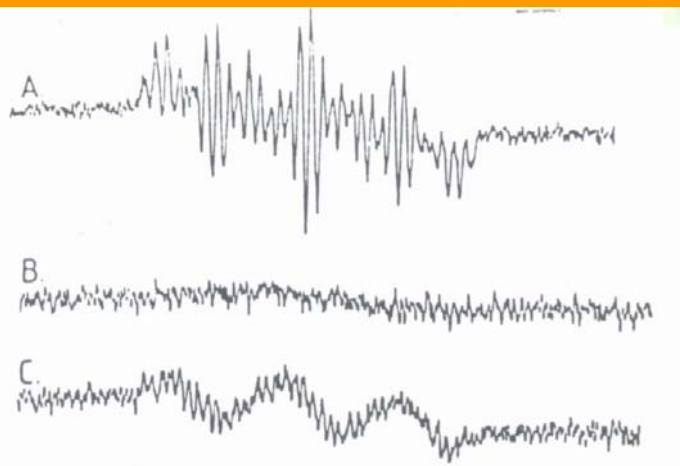


(C)

**Tabela 1**—Percentagem de cura em grupos de ratos inoculados com cepas de *T. cruzi*  $\gamma$  e *colombiana* tratados com o megazol (6), benzonidazol (2), nitrofurazona (7) e nifurtimox, por via oral.

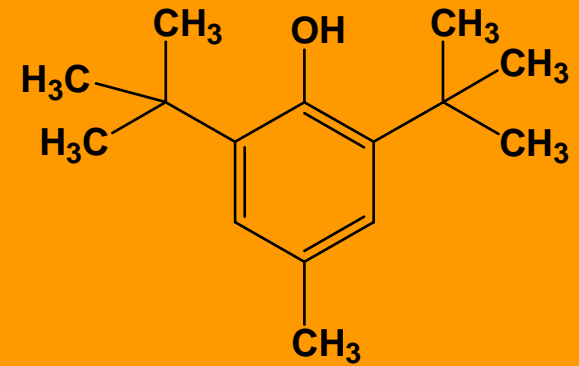
Cepa de <i>T. cruzi</i>	Fármaco	Dose (mg.kg <sup>-1</sup> )	Nº. Doses	No.cured/ No.tratado s	% cura
$\gamma$	Megazol	25	20	9/18	50,0
$\gamma$	Megazol	50	20	19/19	100,0
$\gamma$	Megazol	100	20	17/17	100,0
$\gamma$	Megazol	50 <sup>*</sup>	20	17/20	85,0
$\gamma$	Megazol	500 <sup>T</sup>	1	8/9	88,8
$\gamma$	Nitrofurazona (8)	100	20	5/18	27,7
$\gamma$	Benzonidazol	100	20	5/17	29,4
$\gamma$	Nifurtimox	25	20	1/17	5,8
$\gamma$	Nifurtimox	500 <sup>T</sup>	1	0/10	0,0
Colombiana	Megazol	50	20	13/15	85,6
Colombiana	Megazol	100	20	14/14	100,0
Colombiana	Nifurtimox	50	20	0/15	0,0

# Qual é o Mecanismo De Ação do Megazol?

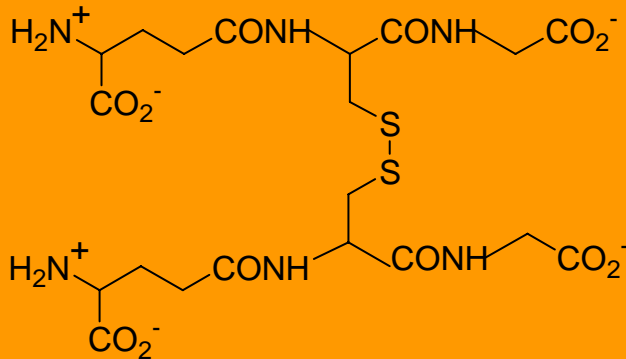


Espectro de **ERS** obtido durante incubação anaeróbica de microssomas de fígado de rato com compostos nitro-heterocíclicos. As misturas reacionais em tampão fosfato (20 mM), pH 7.4, contêm microssomas de fígado de rato (2mg de proteína/mL), NADPH (5 mM), DTPA (1 mM), KCl (150 mM), DMSO (10%, v/v) e: (A) nifurtimox (5 mM); (B) megazol (10 mM); e (C) benzonidazol (10 mM).

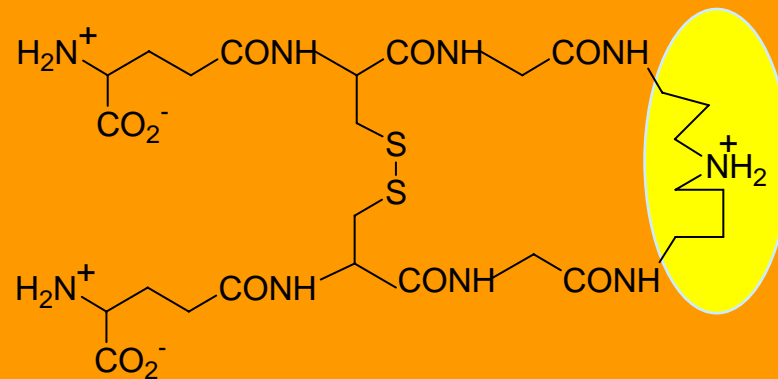
# Moduladores do Sistema Tripanotiona Redutase



**BHT**



1 Glutathione Dissulfeto **GSSG**

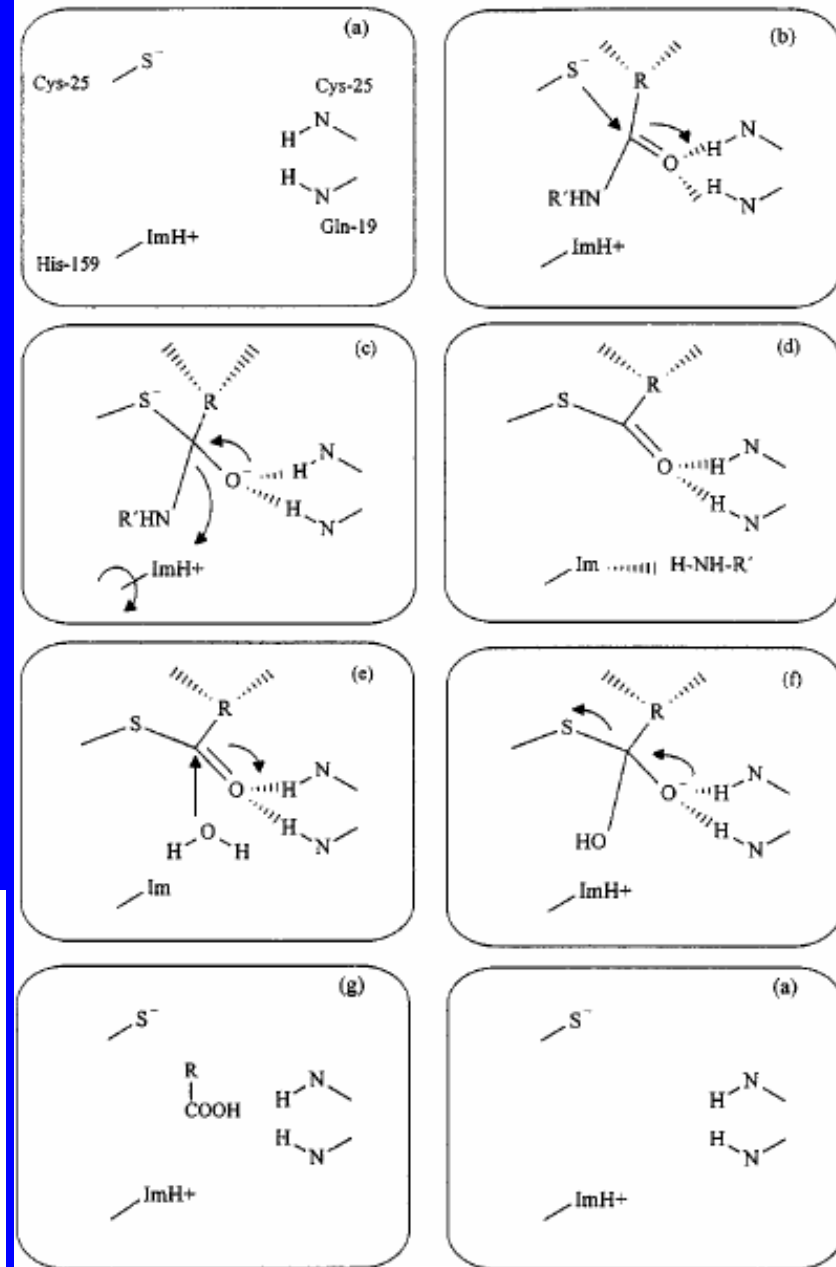
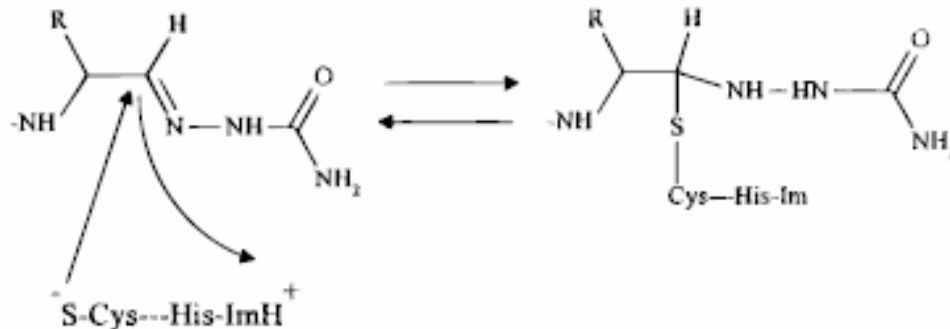
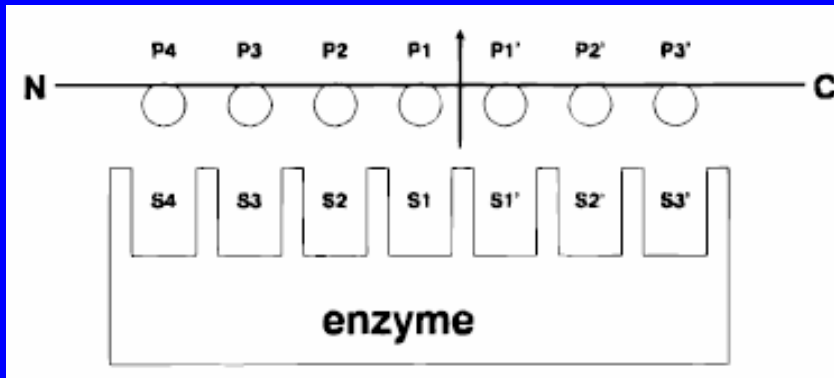
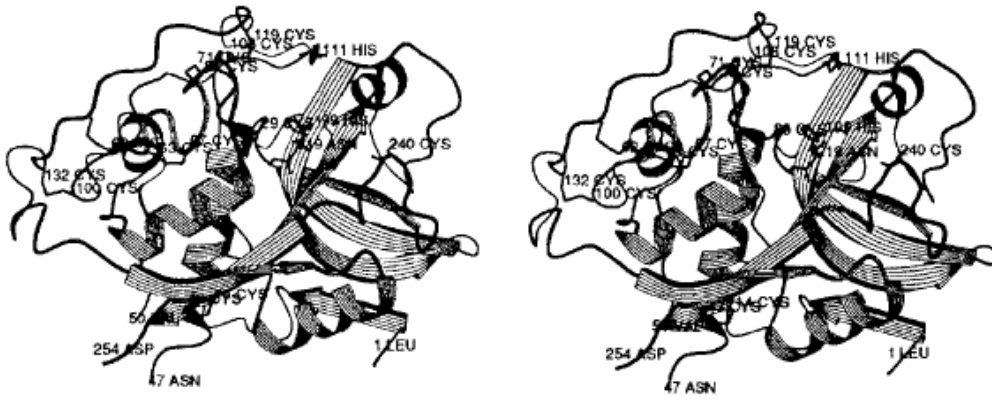


2 Tripanotiona Dissulfeto **T[S]<sub>2</sub>**

**spermidina**

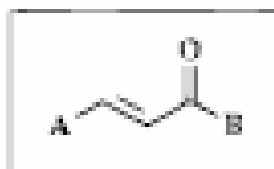


# Cisteína-Proteases (Cruzipaína)



# Inibidores de Cruzipaina

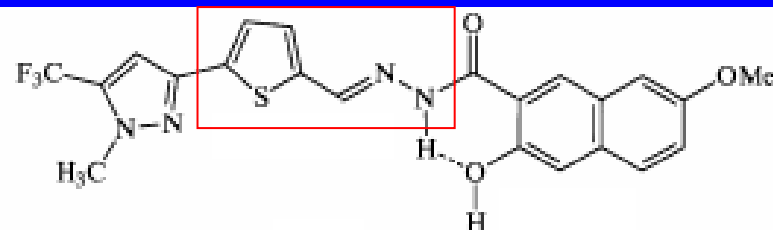
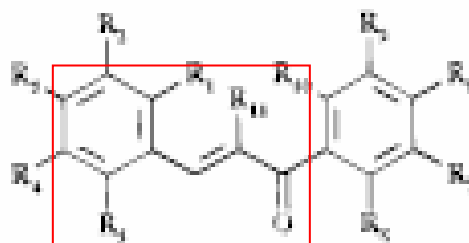
## a) Chalcone derivatives <sup>532</sup>



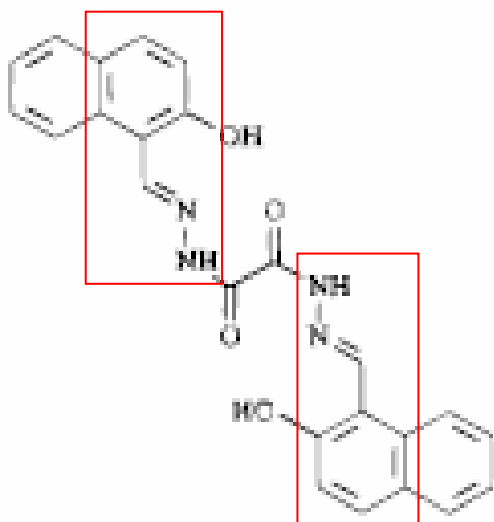
A, B = aryl, heteroaryl e.g.:

$R_1 - R_{10} = \text{H, F, Cl, Br, NO}_2, \text{CF}_3$   
 $\text{OH, OCH}_3, \text{N(CH}_3)_2$

$R_{11} = \text{H, CH}_3$

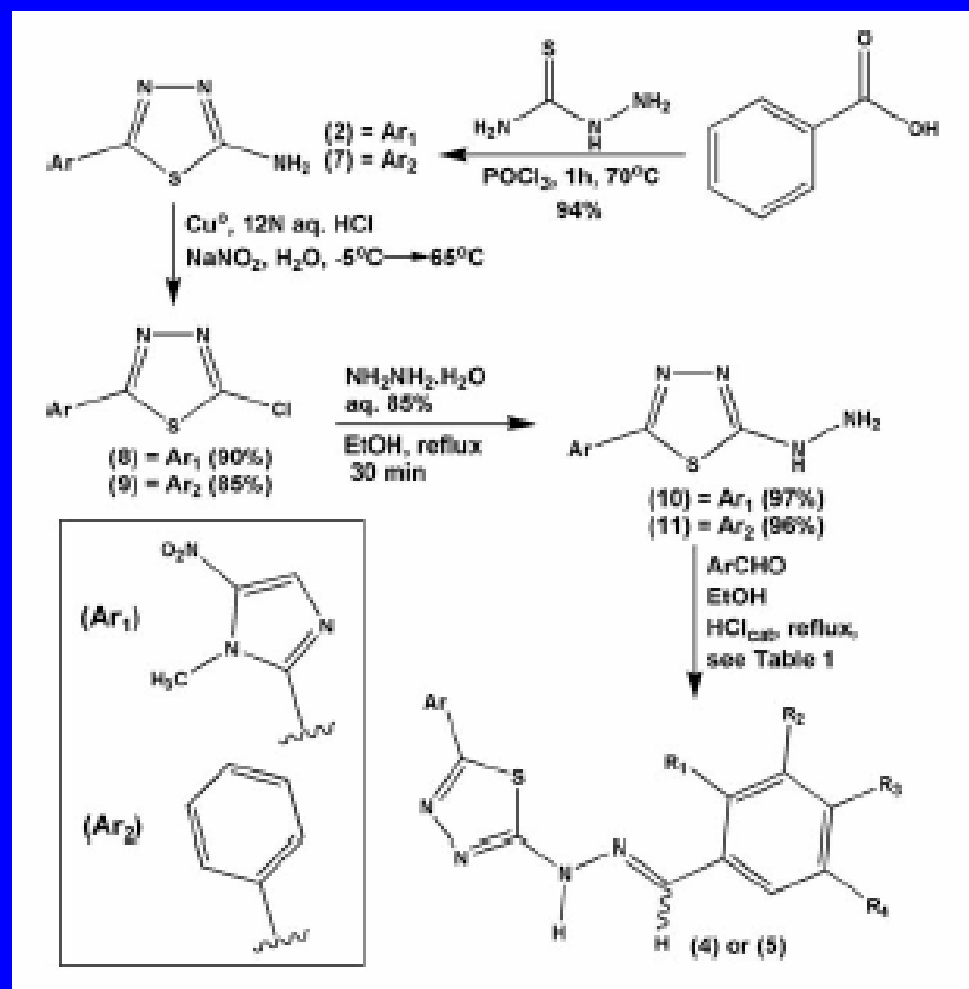
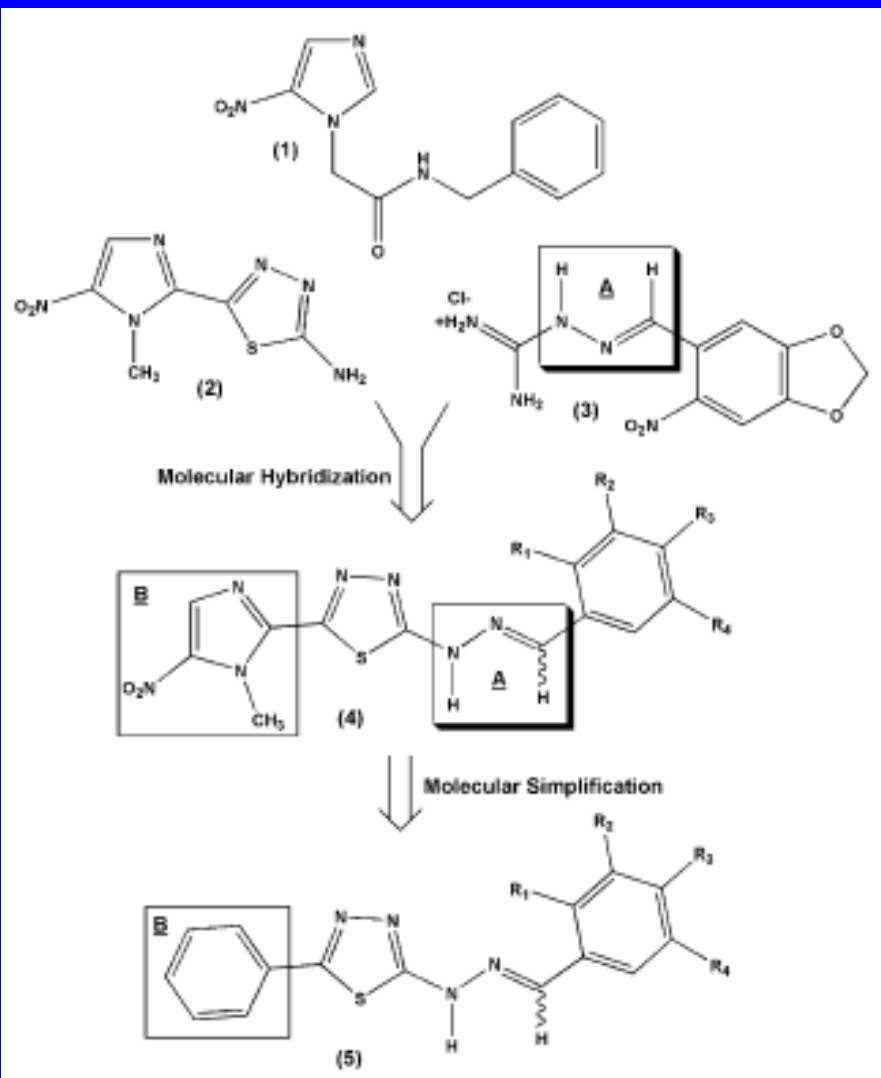


## b) Oxalic bis[(2-hydroxy-1-naphthylmethylene)hydrazide] <sup>531</sup>



Otto, H-H *et al.*  
 (1997)  
 Chem Rev.  
 97, 133-171

# Synthesis and antitrypanosomal profile of new functionalized 1,3,4-thiadiazole-2-arylhydrazone derivatives, designed as non-mutagenic megazol analogues

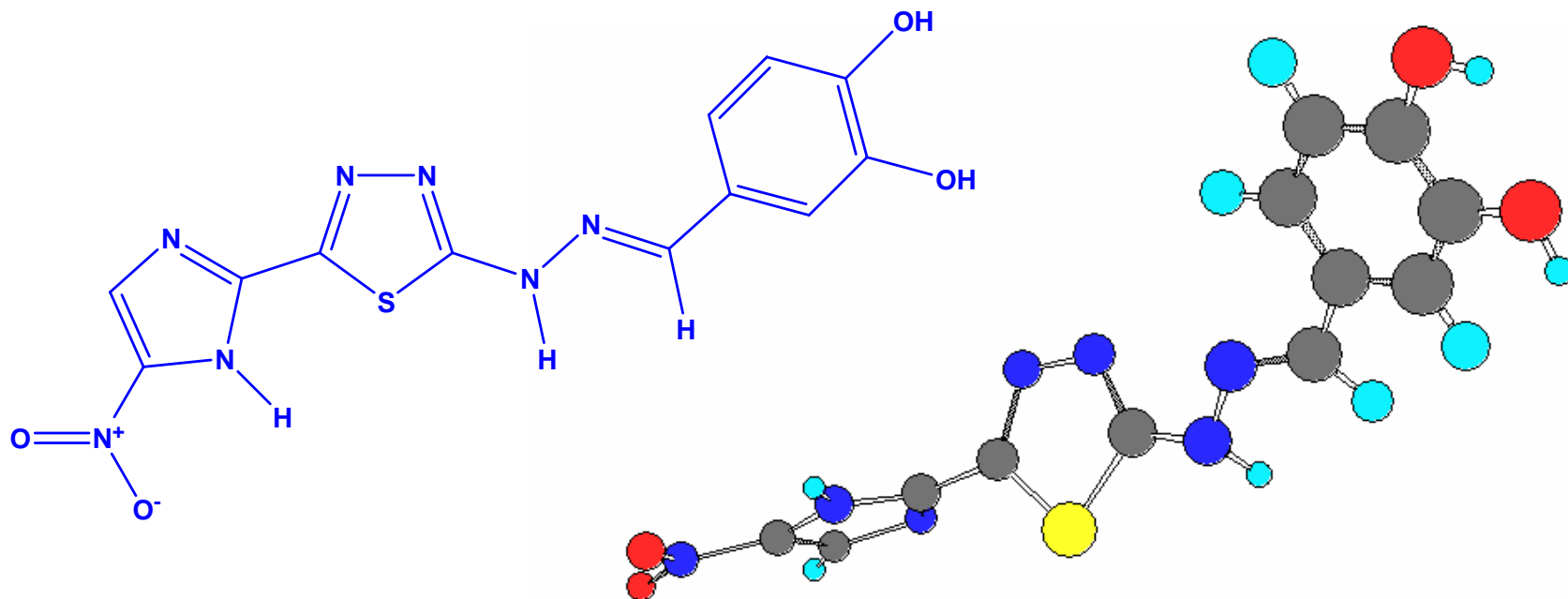


Carvalho, S. A.; Silva, E. F.; Santa-Rita, R. M.; Castro, S. L.; Fraga, C. A. M.

(2004) Bioorganic Medicinal Chemistry Letters, *in press*



# Descoberta da Brazilizone A



“Brazilizone A was able to inhibit in 91% the UV absorption of DPPH radical while megazol used as standard inhibited 32%, at the same molar concentration (0.1mM)”

Carvalho, S. A.; Silva, E. F.; Santa-Rita, R. M.; Castro, S. L.; Fraga, C. A. M. (2004)  
Bioorganic Medicinal Chemistry Letters, *in press*



**“The most fruitful basis for discovery of a new drug is to start with an old drug.”**

**James Black, vencedor do prêmio Nobel de 1988 em fisiologia e medicina.**



# Agradecimentos

*À Comissão Organizadora da  
XIII Escola de Verão em QF&M*

**Obrigado!**

A nighttime aerial photograph of Rio de Janeiro, Brazil. The city's lights are visible, reflecting on the water of the bay. In the background, the iconic Sugarloaf Mountain (Pão de Açúcar) is illuminated. The word "Obrigado!" is written in large, bold, 3D orange letters across the center of the image.