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Bone Targeting

Bisphosphonate based Osteotropic Drug Delivery System for metastatic bone treatment by oral route

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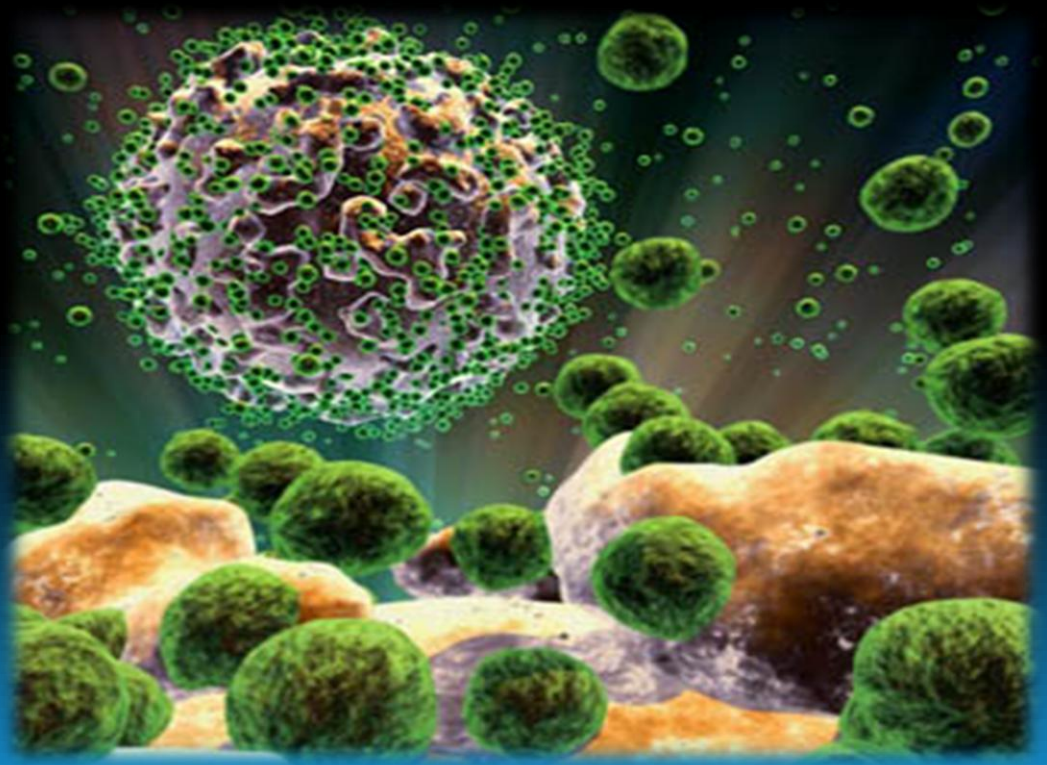

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I INTRODUCTION

Bisphosphonate **based Osteotropic Drug Delivery System for** ***metastatic bone treatment by oral route***

Many oncology patients with solid tumors get secondary metastases to bone.

Current metastatic bone treatments are *bisphosphonates by intravenous route*

(WorldWide sales > $ 1 billion)

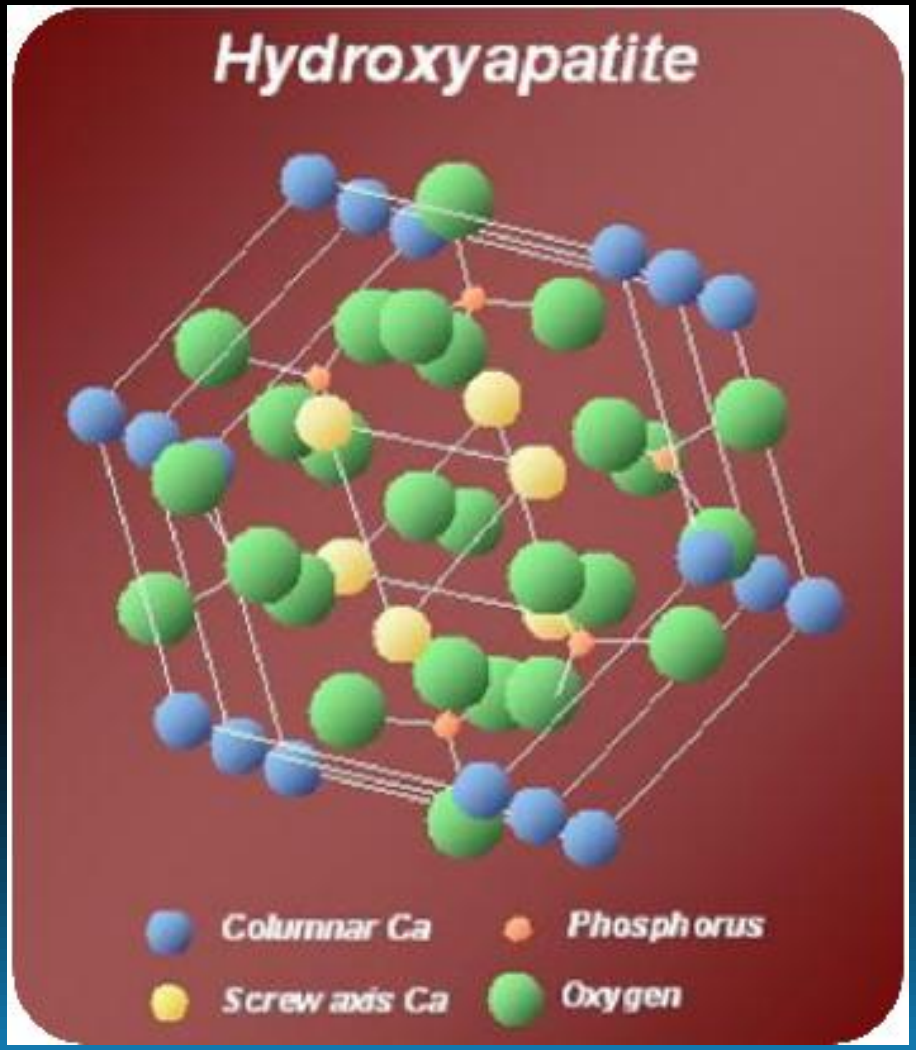
→ ***Unmet medical need*** **for patients treated by regular infusions at hospitals / clinics**

as no oral formulation is available

Bone : a complex organ responsible for structure, calcium storage and hematopoïesis

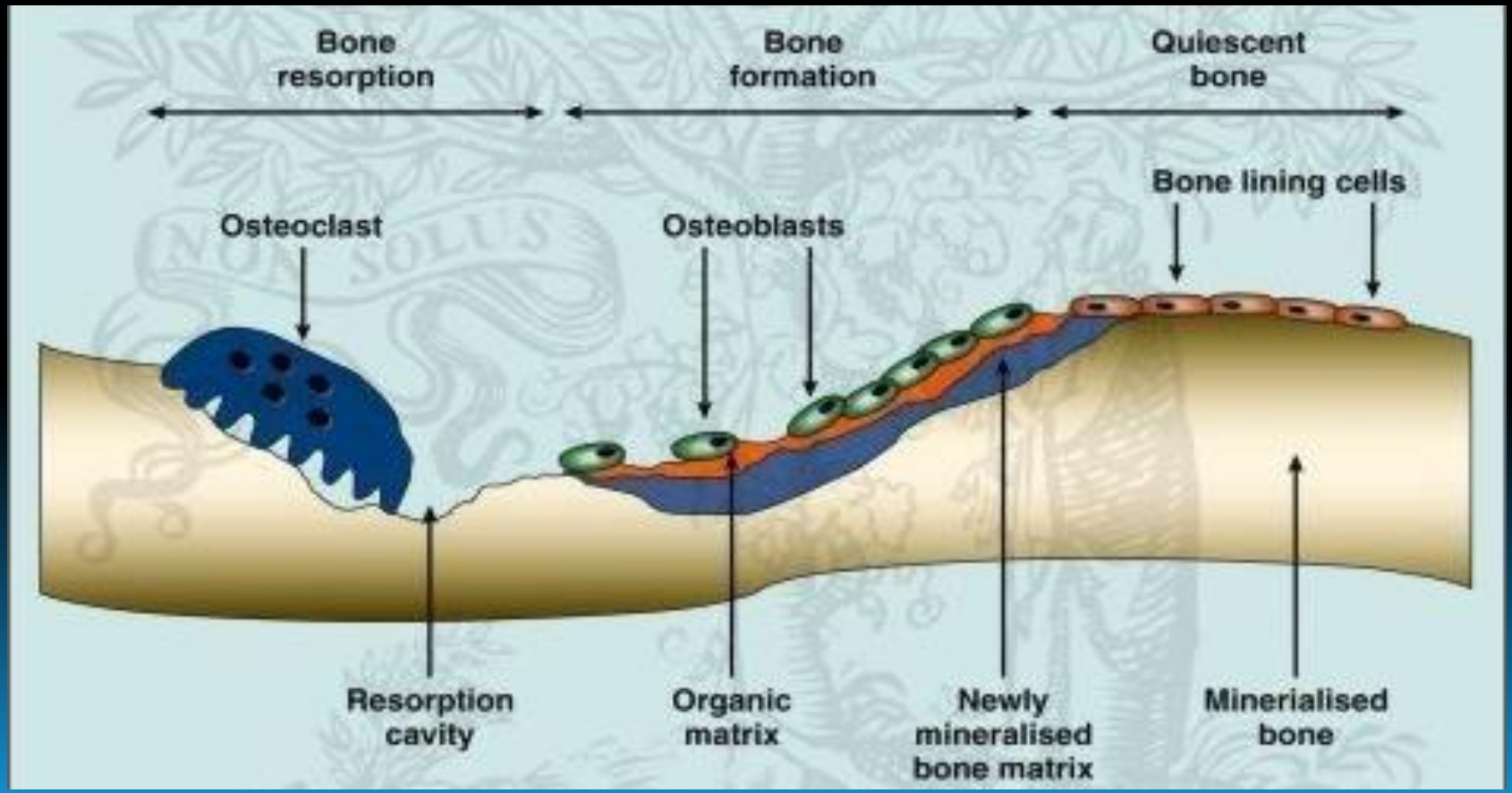
Structure:
50-70% mineral
20-40 % organic matrix
5-10% water
1-5% lipids

Main component : hydroxyapatite crystals (HAP)



Bone turn-over : a bone remodeling cycle :

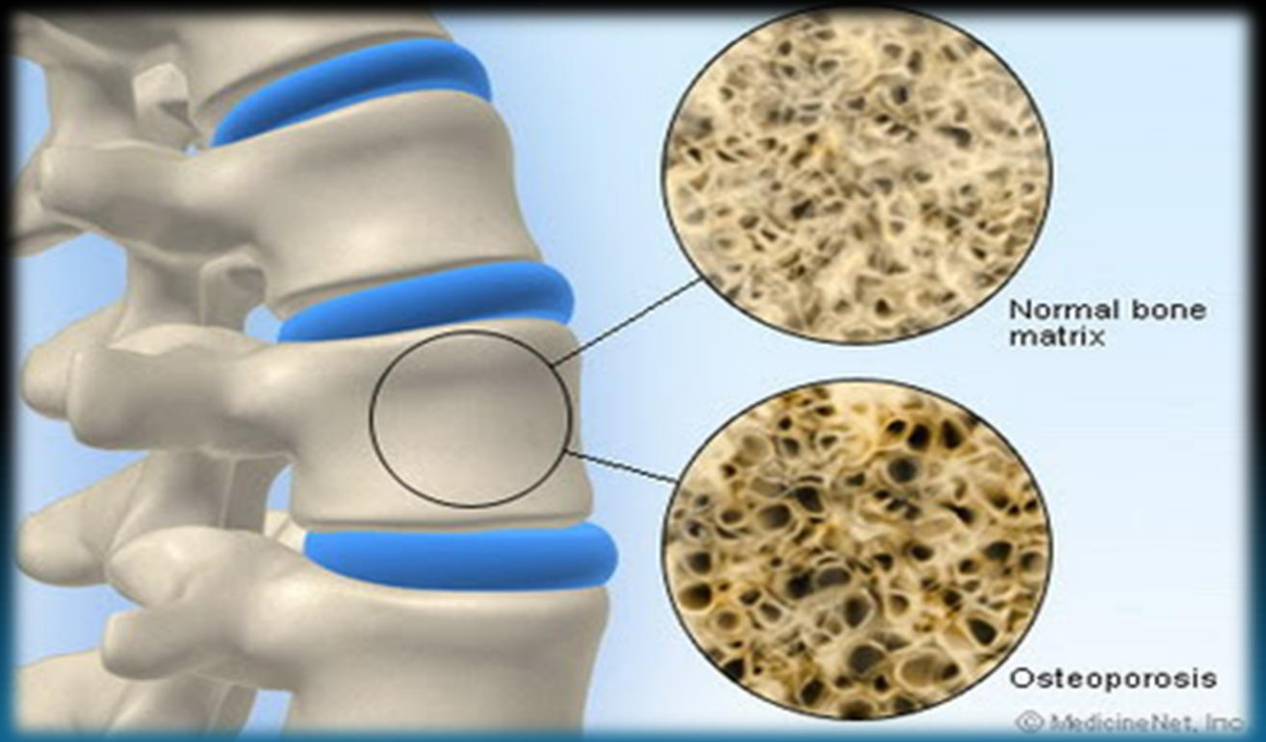
Osteoblastic bone formation
Osteoclastic bone resorption
Mineralization



Alteration of bone's catabolism and/or anabolism of bone diseases

Osteoporosis :

bone resorption
> bone formation



Other pathologies:

osteosarcoma,

bone metastasis,

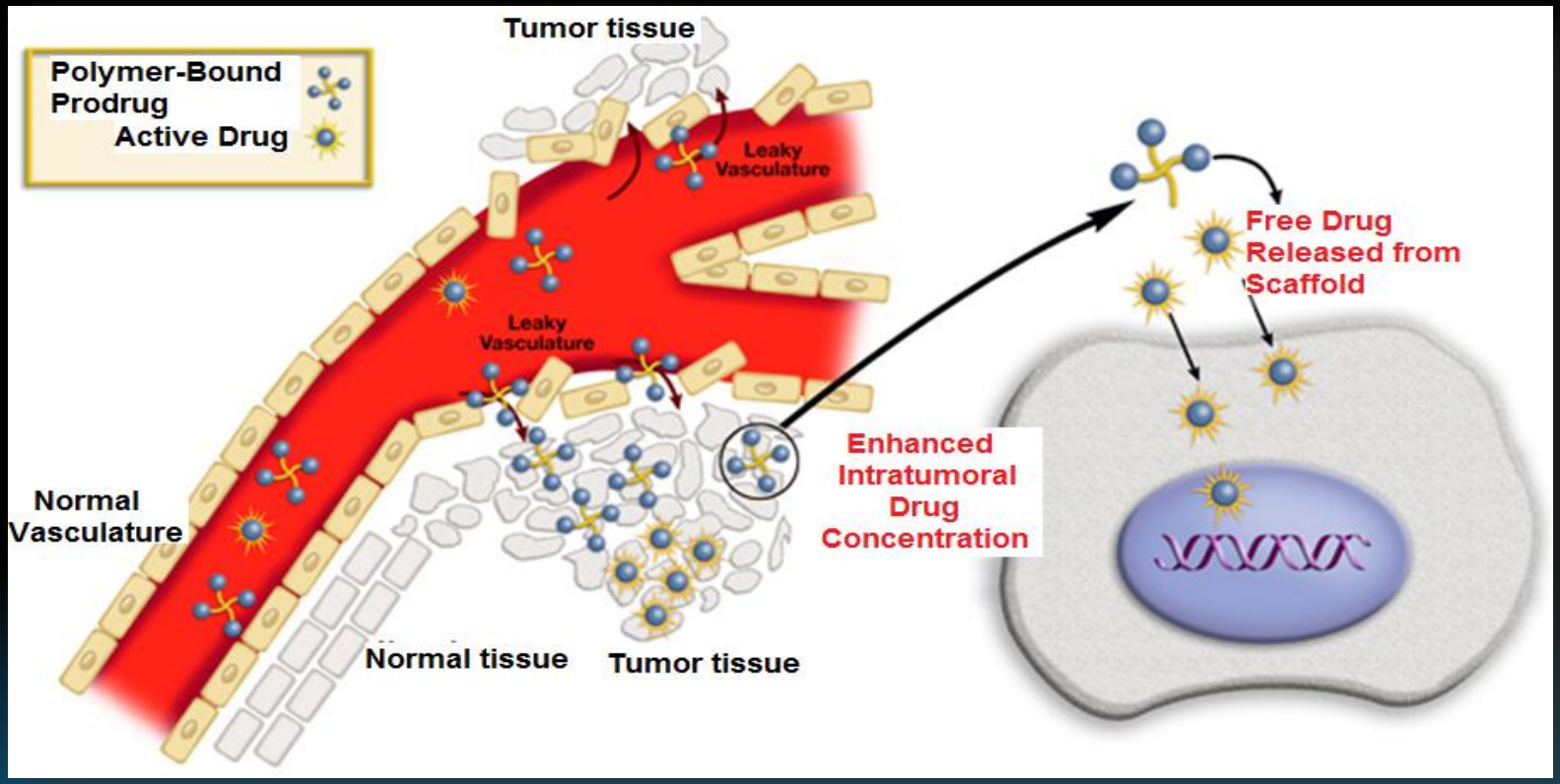
osteoarthritis,

osteomyelitis…

II Bone Targeting

Characteristics of Bone disease state : INFLAMATION

Enhanced Permeability & Retention (EPR)

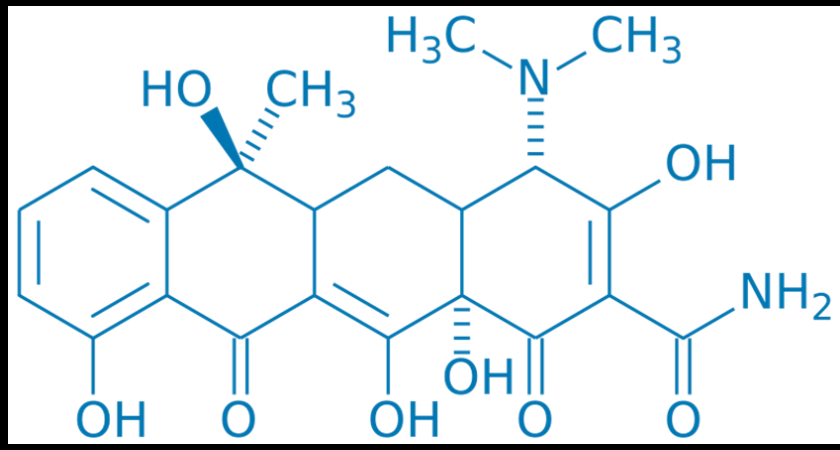


Exposure of HAP to blood used to deliver drugs to diseased tissue

Bone seeking agents

Tetracyclines

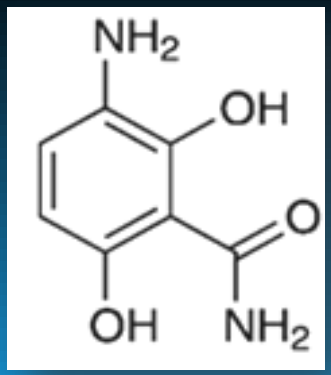
Mode of action :
Stop Protein elongation
via inhibition of aminoacyl-tRNA



Correct orientation of Tetracycline required to bind to HAP

Research of a minimalized tetracycline structure to reduce side effects and retain capacity to bind to HAP

3-amino-2,6 dihydroxybenzamide
(50 % ability to bind to HAP vs tetracycline)

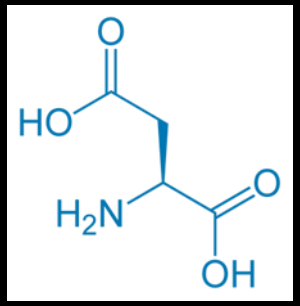


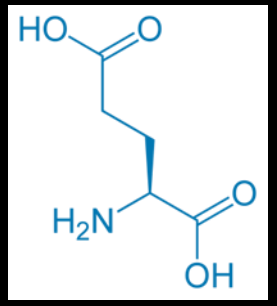
Bone seeking agents

Acidic Oligopeptides

Bone SialoProtein (BSP) : Bone protein with high affinity to HAP

From BSP Acidic, Oligopeptides
with 4 to 10 AminoAcids
have been designed for
enhanced Biocompatibility





Based on Glutamic and Aspartic acids

***In vivo* trials :**

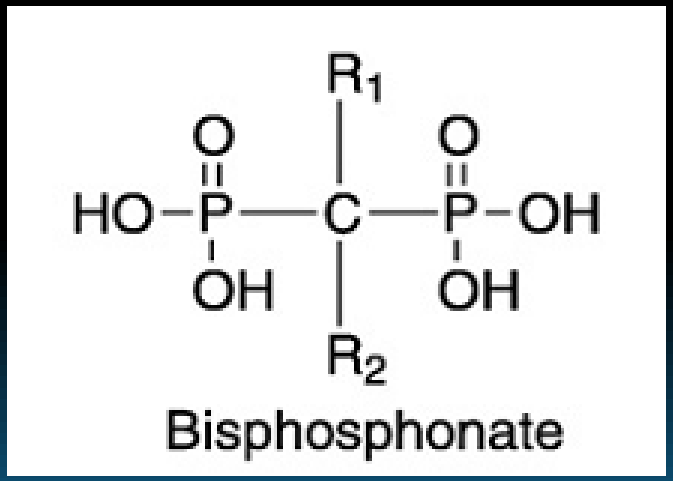
i.v injection into mice of estradiol-17β-succinate-$(L\text{-}Asp)_6$

→ ***affinities of AO depend only the number of AA, not on their L or D characters or their species***

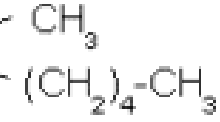
Bone seeking agents :

BISPHOSPHONATES

The most studied bone targeting molecules



Bisphosphonate

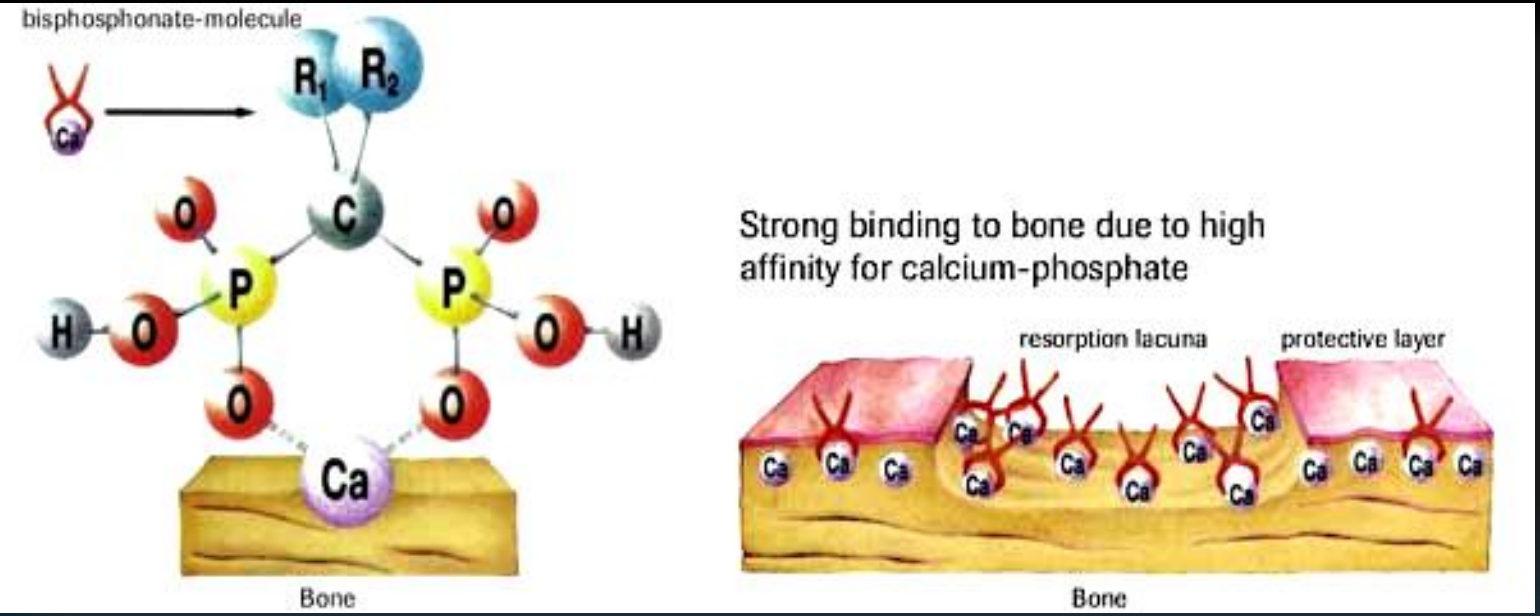
Molécule	R_1	R_2
Etidronate	-OH	$-CH_3$
Clodronate	-Cl	-Cl
Tiludronate	-H	-S-  -Cl
Pamidronate	-OH	$-CH_2-CH_2-NH_2$
Neridronate	-OH	$-(CH_2)_5-NH_2$
Olpadronate	-OH	$-(CH_2)_2N(CH_3)_2$
Alendronate	-OH	$-(CH_2)_3-NH_2$
Ibandronate	-OH	$-CH_2-CH_2N$ 
Risedronate	-OH	
Zoledronate	-OH	

<u>Mode of action :</u> binding to the inorganic part of HAP

BISPHOSPHONATES

First biological activity discovered in 1968

⇨ BPs' strong affinity to HAP :



- Retained even **conjugated** to other molecules
- Relevant in **bone scintigraphie**

Use of Tc99 labeled methylene diphosphate (MDP)
or hydroxy methylene diphosphate (HDMP)

Advantages of using Bisphosphonates as polymer/nanoparticle targeting moieties

- **Amino BP** :
  - primary amine
  - Can be conjugated to carboxylic acids

- **Conjugaison to nanomedecines** via a **degradable linker**
  - Product of synergic effects when coupled with appropriate drugs

+ Soon expiration of BPs' patent protection

→ economic option

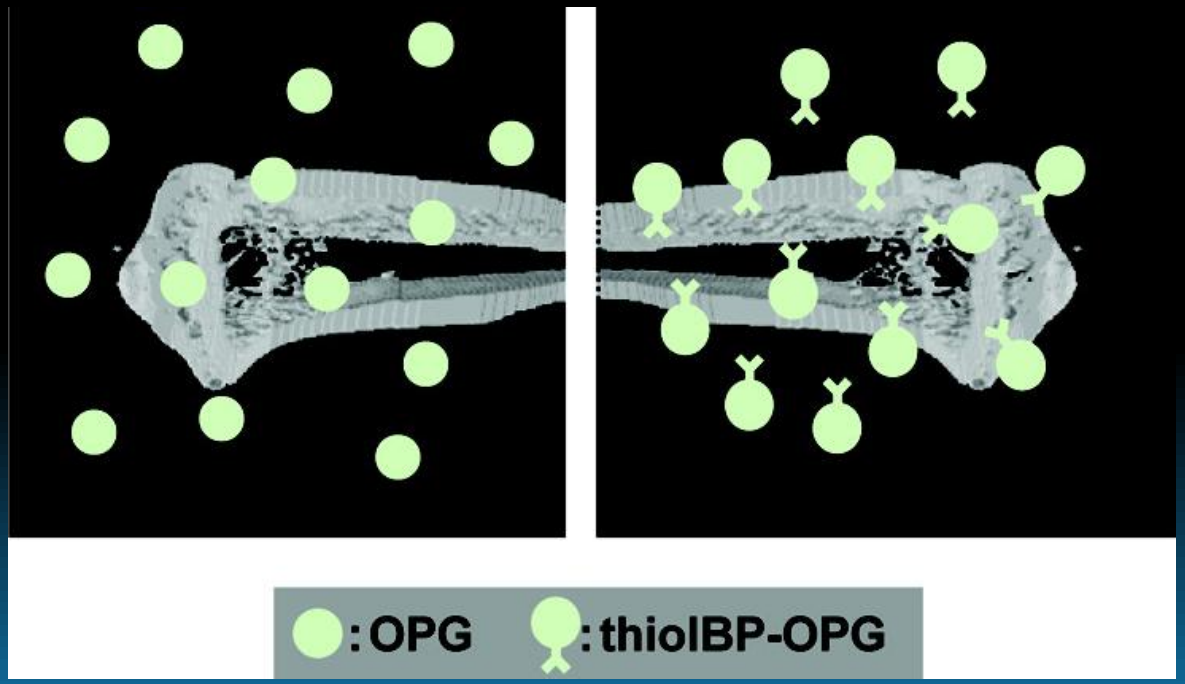
Bone Targeting using Bisphosphonates

Efficiency demonstrated *in vivo* with targeted osteoprotegerin (OPG) as a model therapeutic protein

(M.R. Doshak, Mol. Pharmaceutics 6, (2009) 634-640)

Method :

Conjugaison of OPG with a « Bone seeking » Thiol Bisphosphonate



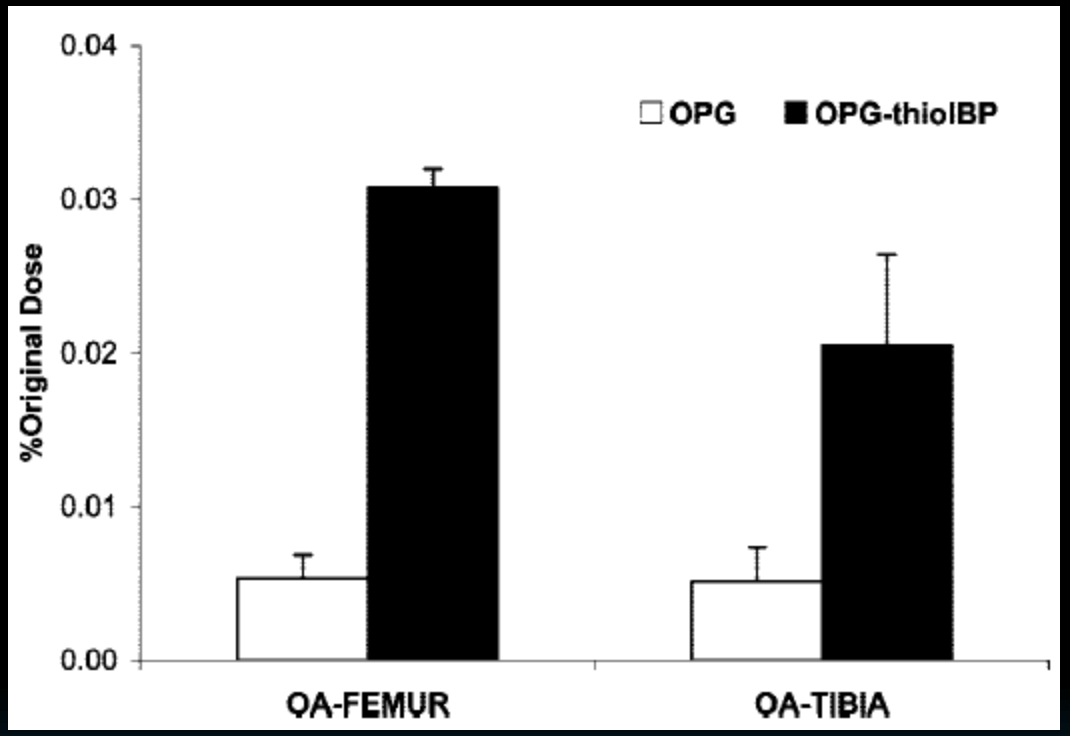
Intravenous administration in a rat model of osteoarthritis

Bone Targeting using Bisphosphonates

Results:

Delivery of OPG-thiol-BP to bone :

4 fold / control OPG in osteoarthritis rats



Targeting of control OPG and OPG-thiolBP conjugates to femur and tibia in rats with osteoarthritis

Significant **advantage of BP conjugation** as **strategy** in **osteopenic bone diseases**

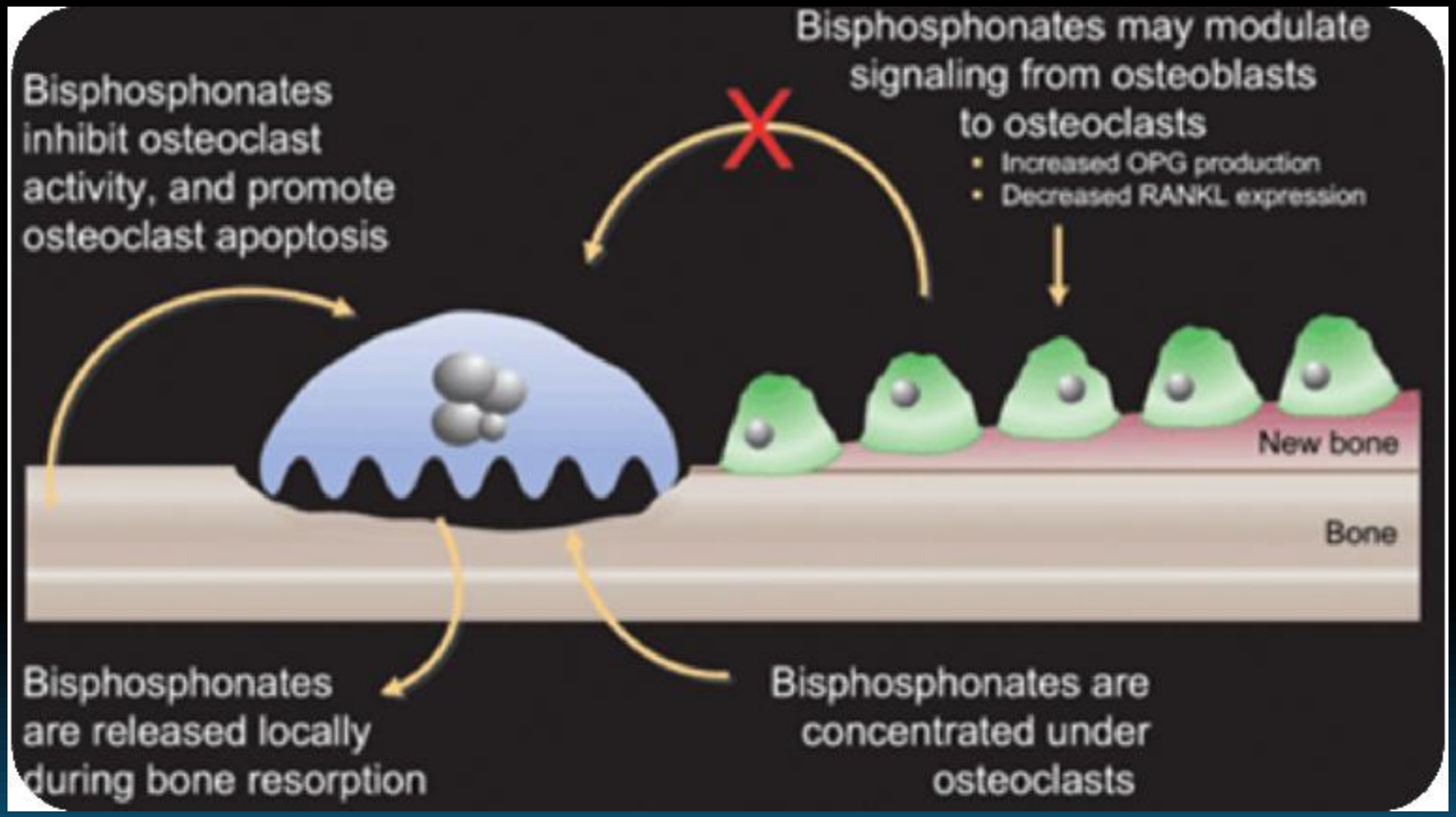
Bisphosphonates anti-angiogenic properties

most bone metastases are characterized by excess osteoclast number and activity.

Some bisphosphonates, potent inhibitors of osteoclast activity, are widely used for prevention of bone metastases.

Bisphosphonates are also used to treat hypercalcemia of malignancy, osteosarcoma and multiple myeloma.

Bisphosphonates anti-angiogenic properties



Effects: apoptosis, inhibition of migration, reduction of angiogenic sprouts of endothelial tissue

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OSTEOTROPIC DRUG DELIVERY SYSTEMS

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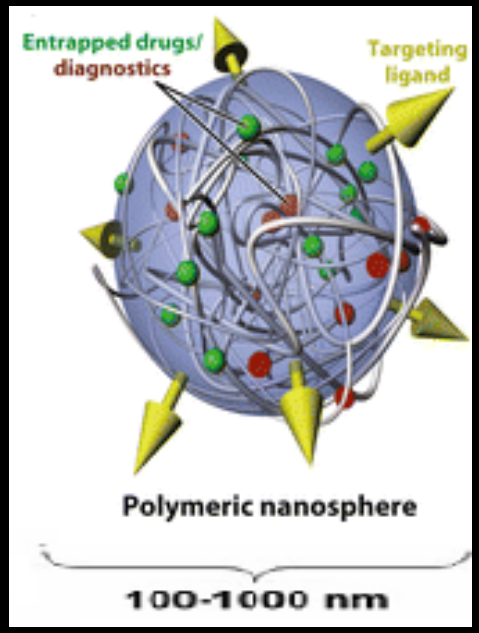
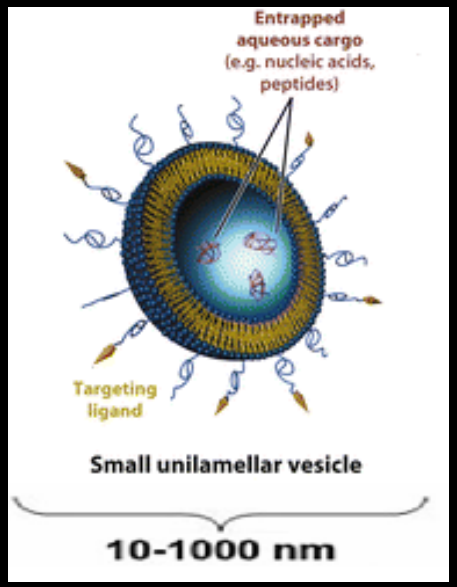
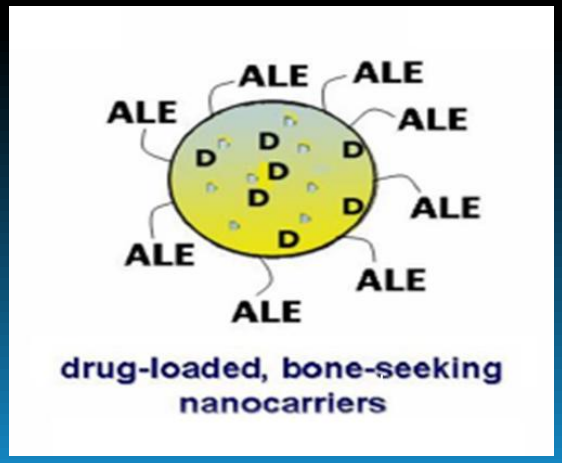
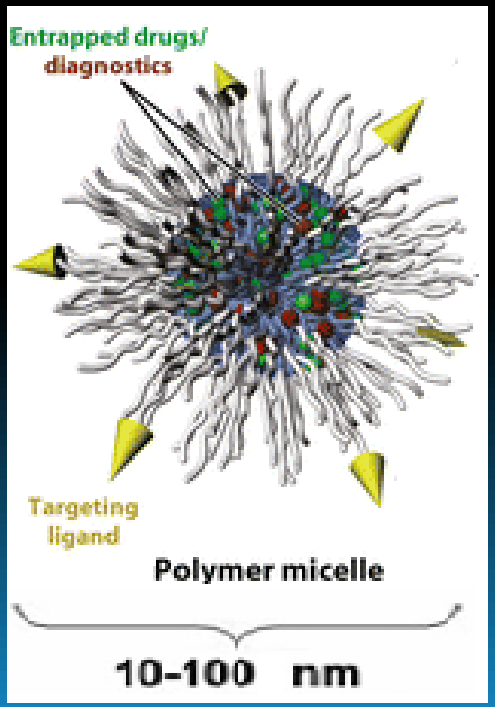
OSTEOTROPIC DRUG DELIVERY SYSTEMS

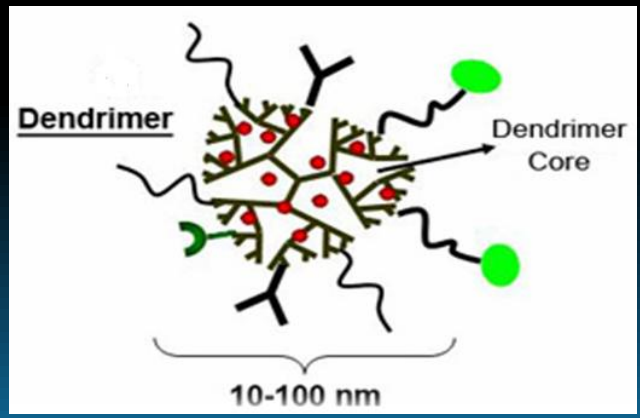
A lot have been studied !











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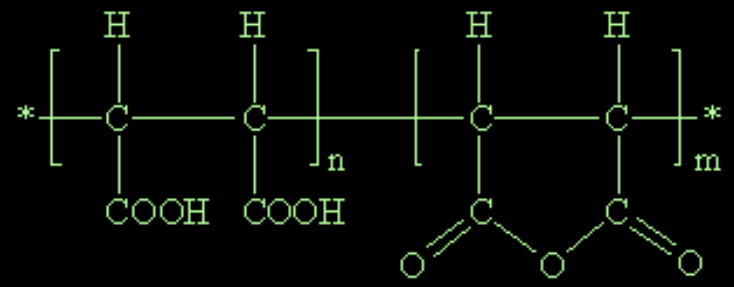
OSTEOTROPIC DRUG DELIVERY SYSTEMS

Poly[N-(2-hydroxypropyl)methacrylamide]: HPMA

The most studied polymer therapeutics to bone
Used in the design of micelles and dendrimers

Properties :
Biodistribution to bone
Bone targeting abilities
Low toxicity profile



In vivo data

HPMA copolymer – D-Asp8 conjugate: Administration i.v of bone-targeted and non-targeted HPMA copolymers into mice

HPMA copolymer−alendronate (ALN) conjugate
I.V injection of conjugates with different Mw and ALN content into mice

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All Osteotropic Drug Delivery Systems Evaluated _in vivo_ to date have been administered _intravenously_

This is why a novel Bisphosphonate Osteotropic Drug Delivery System ***(BP-ODDS)*** has been developed by INSCB ***for ORAL ROUTE***

to improve

Bisphosphonates oral bioavailability

…thereby enabling the development of oral dosage forms for oncology indications.

Bisphophonates for cancer therapies are administered *intravenously by slow infusion*.

Infusion of bisphosphonates is however associated with dose and infusion rate dependent effects on renal function.

Oral administration, is complicated by poor bioavailability and poor gastrointestinal tolerability.

This limits their use in oncology to intravenous infusion to achieve the doses required for efficacy.

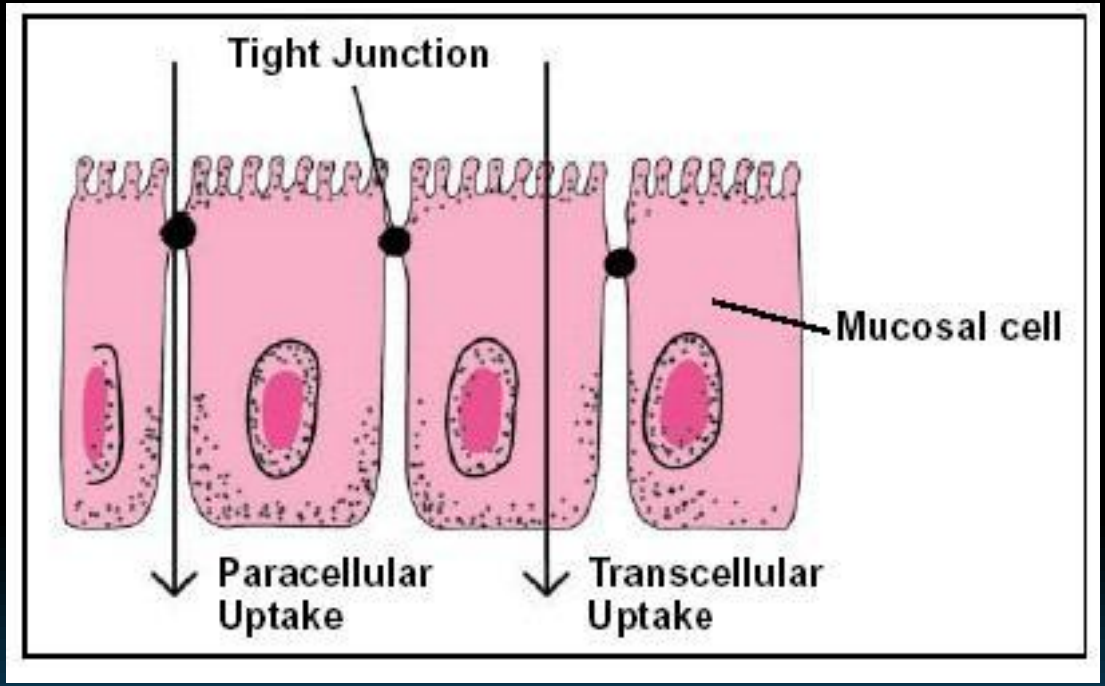
Bisphosphonates have very poor oral bioavailabilities :

0.6% in average !

Design of drug delivery systems enhancing oral absorption is required for treatment efficiency !

INSCB proprietary technology combines two factors affecting Bisphosphonates physico-chemical and pharmacological behaviour *in vivo*

Their specific transport through intestinal membrane by paracellular pathway



&… Their ability to form insoluble calcium complexes *in vivo* in the GI tract

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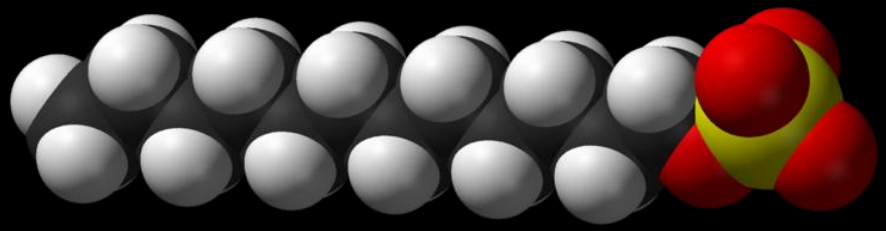
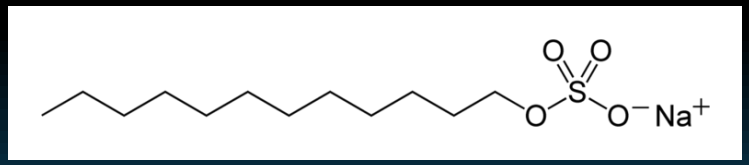
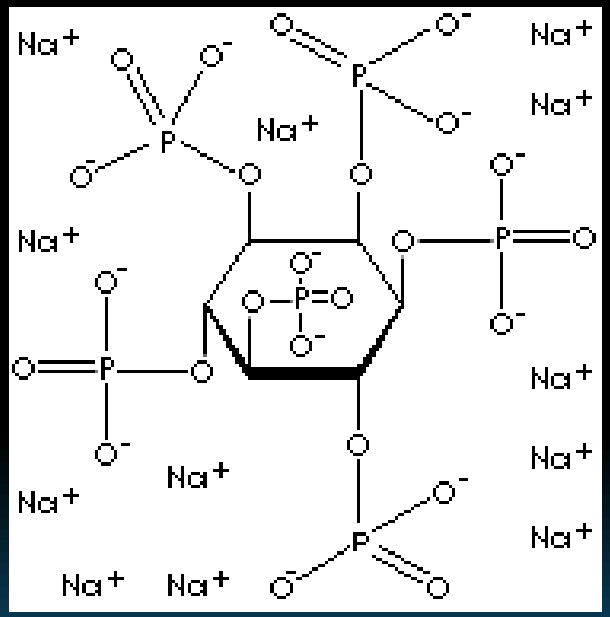
INSCB Bisphosphonate Osteotropic Drug Delivery System : (BP-ODDS)

Two excipients, both registered at pharmacopeias and authorized for oral administration were used to modulate these bisphosphonates properties





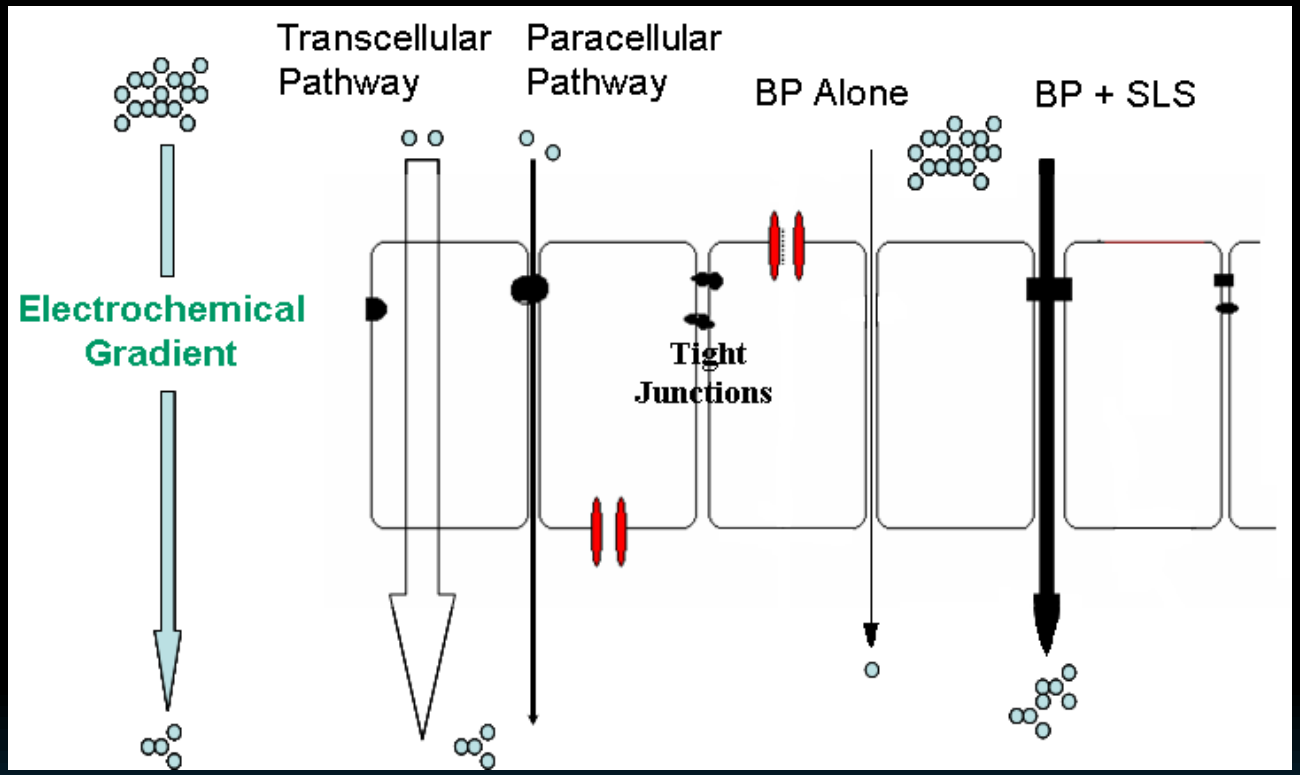


one intestinal penetration enhancer :
Sodium Dodecyl Sulfate

one calcium chelatant agent :
Myo-Inositol hexakis dihydrogen phosphate dodecasodium

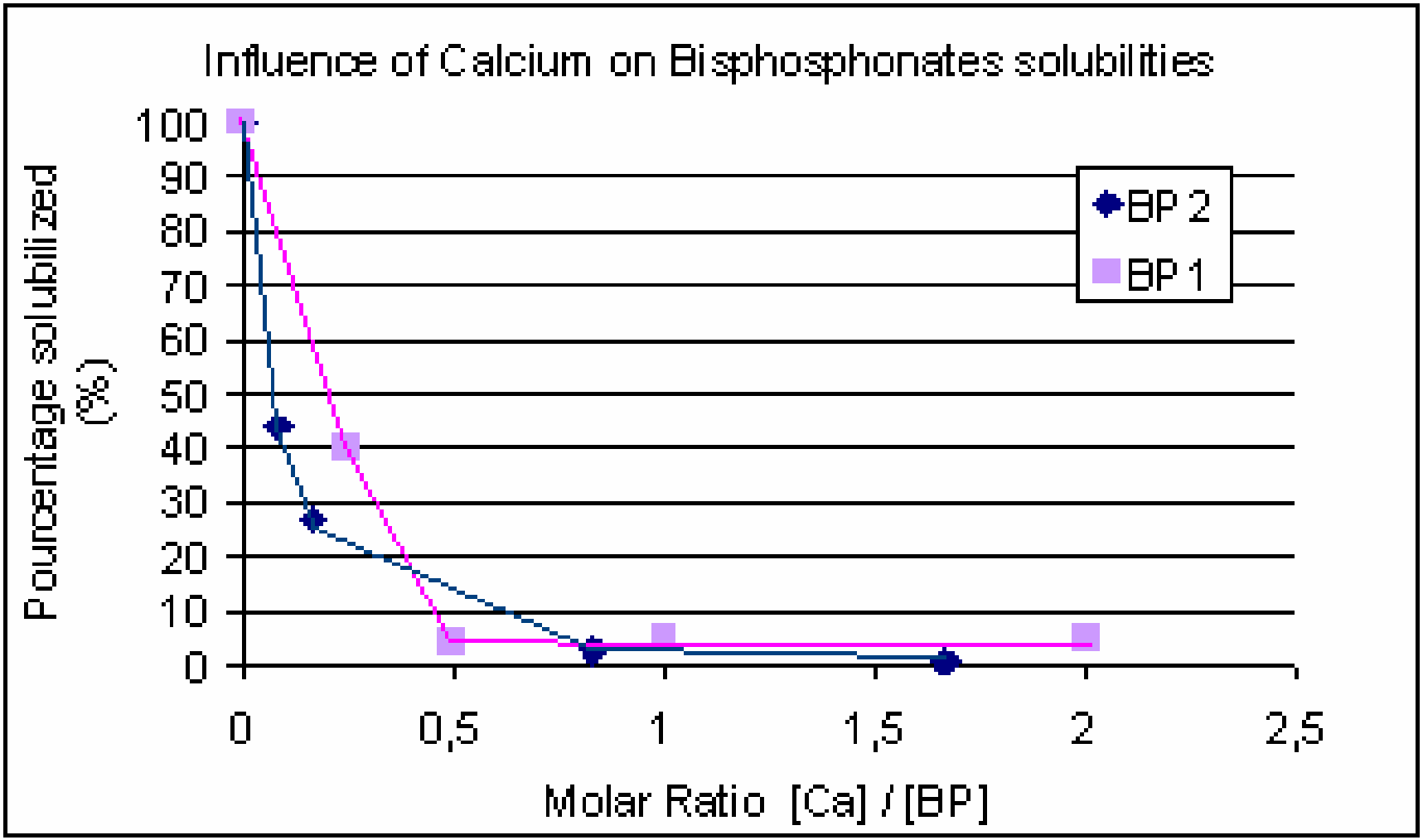
Intestinal penetration enhancement :

Bisphosphonates are highly hydrophilic, they can cross the intestinal membrane only by using the paracellular pathway of intestinal tight junctions



Sodium Dodecyl sulfate increases intestinal permeability by opening tight junctions, thus enhancing paracellular absorption of Bisphosphonates

Physiological Calcium chelation



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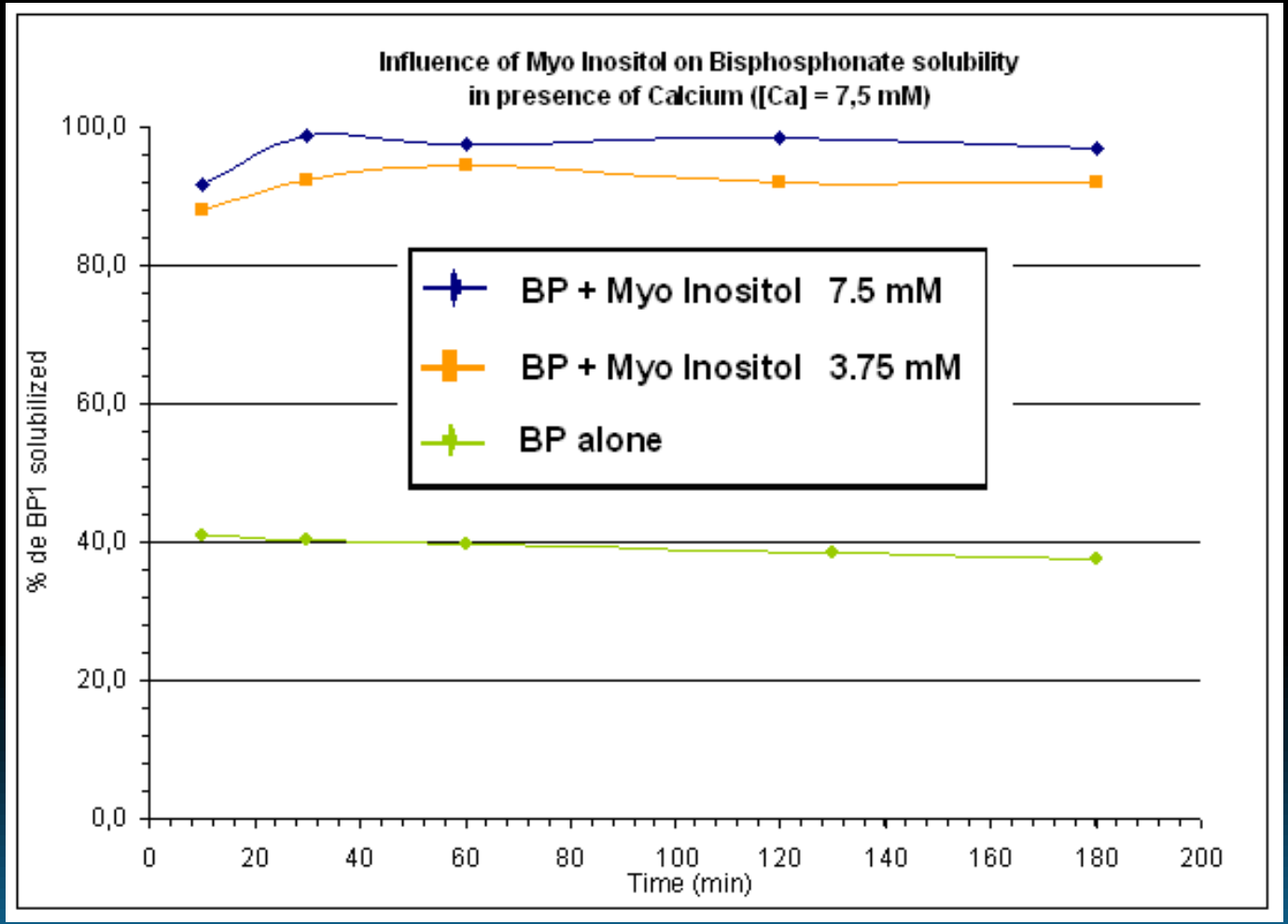
Physiological Calcium chelation

Myo-Inositol is a stronger calcium chelatant than bisphosphonates

Its presence ***increases Bisphosphonate solubility in GI Tract***



→ ***Bisphosphonates intestinal absorption Is enhanced***

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INSCB BP-ODDS

Manufacturing process

Classical physical dry mixture of excipients and drug substance

Can be processed in a film coated tablet or in hard gelatine capsules.

Technology does not increase manufacturing costs compared to classical tablet or capsule production.

The new formulation shows no degradation of the active drug after one year stability, either in film coated tablet or hard gelatine capsule form

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Comparative Bioavailability study

Purpose : To assess relative bioavailability of BP-ODDS, vs reference formulation, a marketed film coated tablet of a bisphosphonate (Actonel 35 mg ®),

Dose : 35 mg single dose, administered *per os,* one film-coated tablet of 35 mg with 200 ml of low carbonated water.

Subjects : 12 healthy volunteers in fasting conditions

Selection criteria : Male & female, aged 18-45, body mass index : 19 - 27.5

Methodology: *Two period, two sequence, cross-over, block randomized*

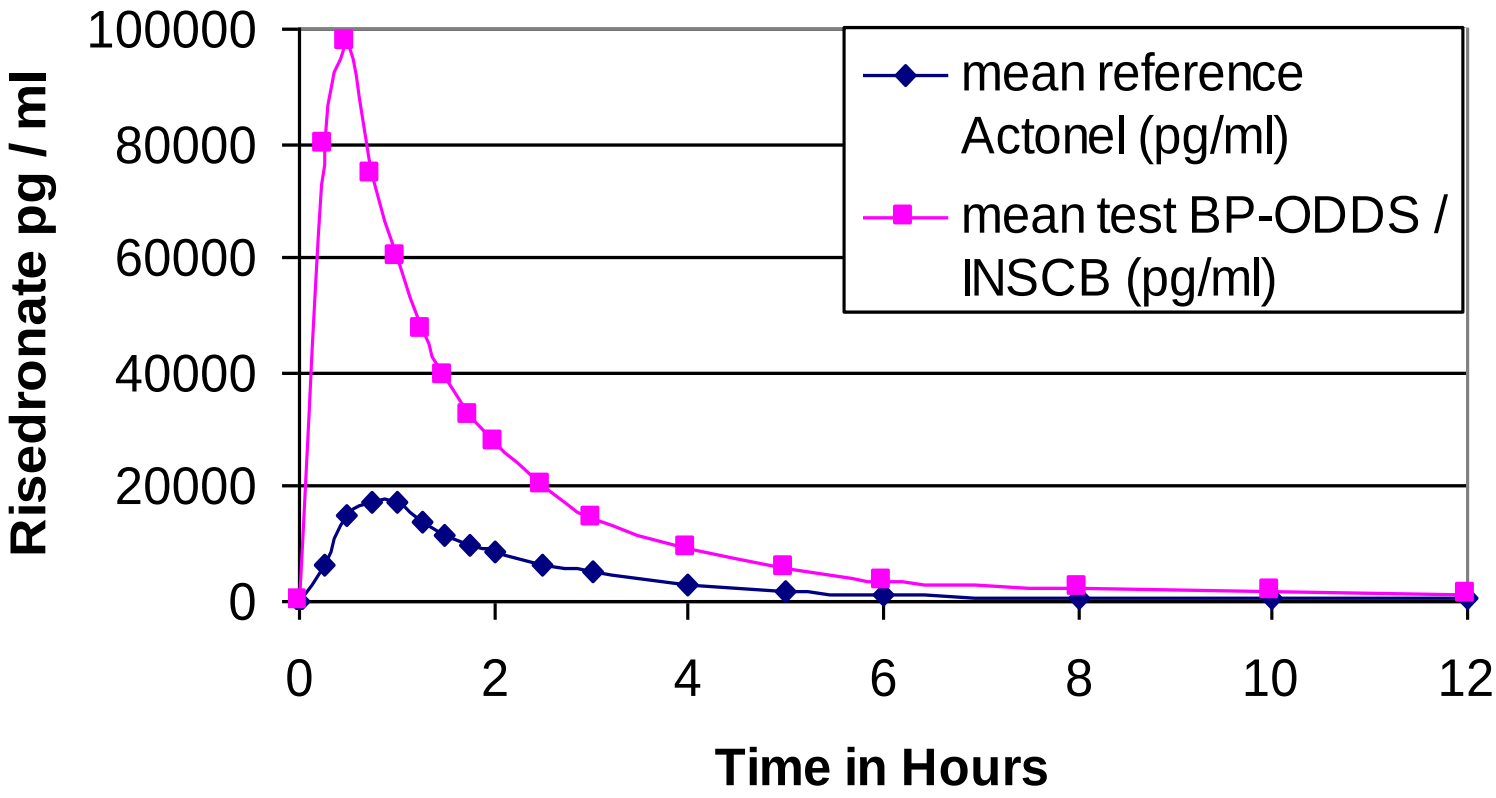
Duration of treatment : One day per period

Pharmacokinetic criteria for evaluation :

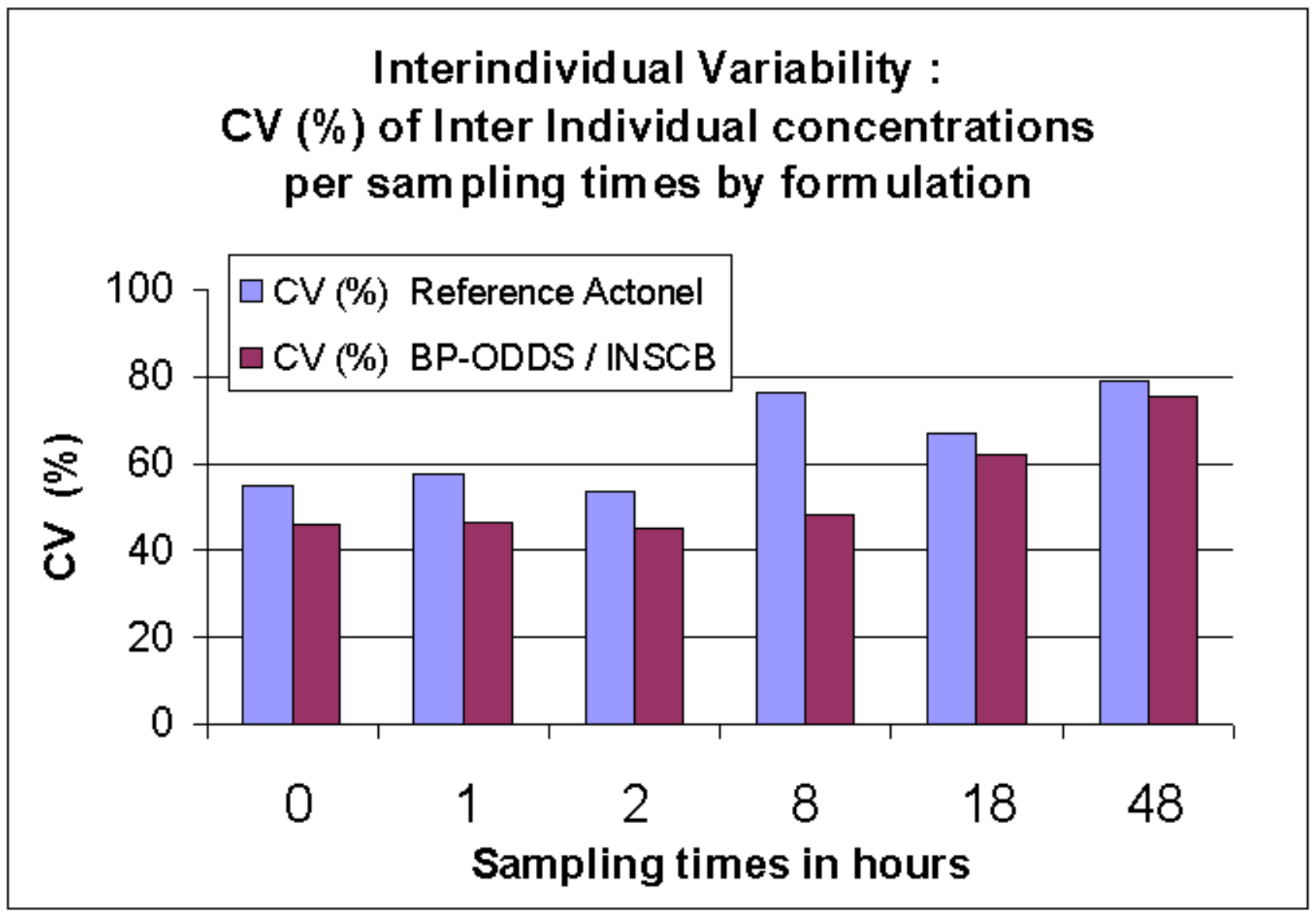
Risedronate AUC0-t , Cmax, AUC O-inf , Tmax + (T1/2 , MRT).

Comparative Bioavailability study

Comparative Bioavailability of BP-ODDS / INSCB risedronate 35 mg vs marketed Actonel



Comparative Bioavailability study



CONCLUSIONS

Statistical analysis of pharmacokinetic data obtained proves that :

Test product : BP-ODDS / INSCB Sodium Risedronate 35 mg, is relevantly better absorbed than reference ACTONEL® 35 mg

AUC of BP-ODDS is almost 300 % higher than reference Actonel®

Cmax of BP-ODDS is almost 500 % higher than reference Actonel®.

The Bisphosphonate Osteotropic Drug Delivery System (BP-ODDS) developed, is supra-bioavailable when compared to its reference marketed product

CONCLUSIONS

T_{max} of BP-ODDS is half of T_{max} of reference Actonel®

→ ***twice quicker efficiency for BP-ODDS*.**

Variability of primary pharmacokinetic parameters of BP-ODDS are lower than variability of reference Actonel®

(20 % lower for C max and 60% lower for AUC 0-inf)

+ Safety :

BP-ODDS administered in single dose, orally, was very well tolerated by the participant subjects.

BP-ODDS tablets or capsules offer a new paradigm for

metastatic bone cancer treatment
through the *oral route*

Their ability to better target growing metastatic tissues, cover an unmet market need for oral bisphosphonates in oncology.

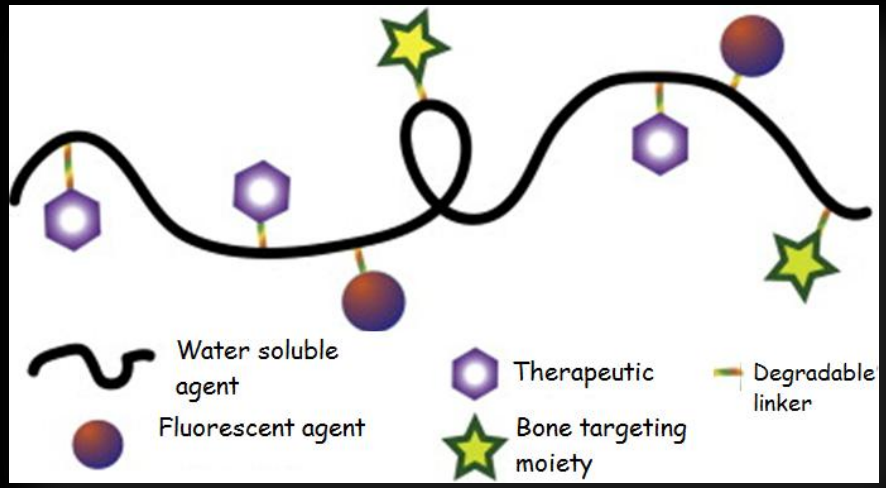
BP-ODDS is an effective and potentially safer alternative
to bisphosphonate intravenous infusions
which could offer the following advantages :

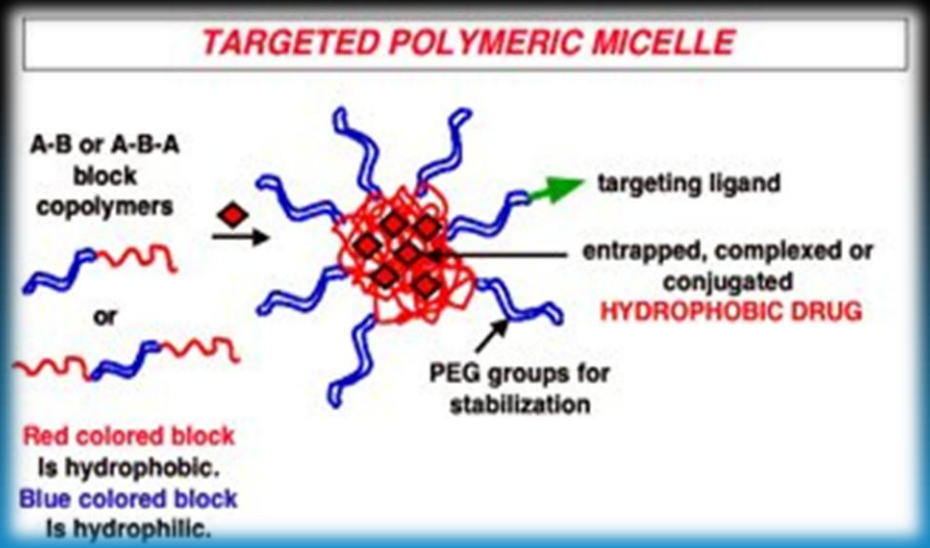
Improved quality of life for the patient
Cheaper treatment
Flexibility in the dosing regimen

BP-ODDS could also be used as a

carrier for drugs needing vectorization to bone

Either using the **Micelle forming ability of some bisphosphonates** to carry the drug candidate





Or by use of a **biodegradable linker to be conjugated to a bisphosphonate** or a bisphosphonic group

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This Osteotropic Drug Delivery System has been patented by INSCB in 2011 and is available for license

Contact : contact@inscb.org

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Thanks for your Attention !

APPENDICES

Bone seeking agents

Estradiol analogs

Localisation in bone tissue + lack of estrogenic properties

Methods : Attaching calcium chelators to an estradiol moiety via succinoyl or carboxyethyl linkers

Improvement of targeting potential

2 sorts of Bisphosphonates

Amino BP: disturbance of mevalonate pathway

More potent

inhibition of protein prenylation

and osteoclasts' loss of function

Non amino BP: creation of a modified ATP

inability to be hydrolysed

Osteoclasts' apoptosis and reduced bone turn-over

Comparative bone seeking properties of Bisphosphonates (BP) Acidic Oligopeptides (AO) and Tetracycline

Rate of binding to HAP : *Faster for AO than BP due to a larger binding area of AO*

Binding strength : *Greater for BP than AO due to a higher specificity of BP for HAP*

Binding sites :

- *All bone for BP*
- *Higher cristalline HAP for AO*
- *Growing surfaces with low cristallinity for Tetracycline*

Bone targeting principles

Osteoclast targeting :

Use of BP and AO because of their favored binding onto absorbing surfaces

Osteoblast targeting :

Use of Tetracyclines or analogs because of their favored binding on bone's growing surfaces

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Comparative Bioavailability study

Methodology:

Two period, two sequence, cross-over, block randomized

Hospitalization of subjects : until 24 hours post administration.

Washout period : 46 days.

Blood samplings collected : Before dose (0.0) & 20 sampling points
Post dose from 0.25, to 168.0 hours.

Analytical method : Determination of Risedronate in plasma
by HPLC- MS/MS.

Comparative bioavailability assessment based on plasma drug levels of test bisphosphonate used, (sodium risedronate).

+ Safety : Laboratorv data / Vital signs / Adverse events

RESULTS: Pharmacokinetics parameters :

Mean pharmacokinetics characteristics of risedronic acid after treatment with REFERENCE : Actonel ® and TEST : BP-ODDS / INSCB products

Comparative bioavailability of the primary parameters (test name : Classic 90% CI) :

Parameter	AUC 0-t	AUC 0-inf	C max	T max
Test Value (Test / Reference)	295.854	266.363	480.004	0.530

Comparative coefficients of variation (CV) of the primary pharmacokinetic parameters

Parameter	AUC 0-t	AUC 0-inf	C max
Test Value (CV Test / CV Reference)	0.85	0.38	0.79

Future of the field

Development of bone targeted anti-neoplastic agents
Because bone metastases = unsolved problem in oncology

Research about bone targeted therapeutics focusing on:

EPR effect , Delivery of **siRNA** to solid tumors
Combination therapies: (i.e : Paclitaxel / BP; Statins : BP…)
bone targeting of relevant drugs in order to reduce pain

+ improve quality of life

Continous increase in **prevalence**
of **bone diseases** with **aging population**

V.1 Gaps in current research

- Many novel drug applications: yet to be explored
- Better development of disease specific targeting
- Research of bone conditions present in each disease + targeting mechanisms
- Discovery of specific biochemical pathways => disease states