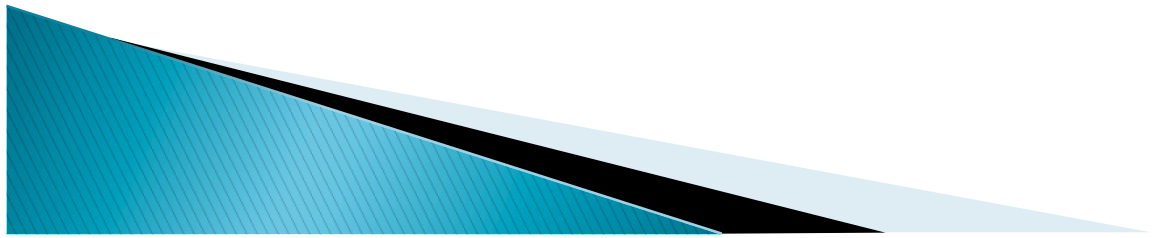
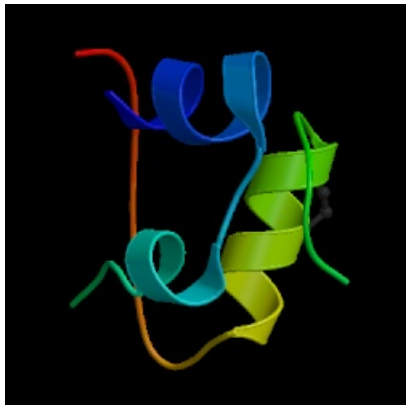
Pharmaceutical based strategies for proteins and peptides delivery: Special emphasis on interferon

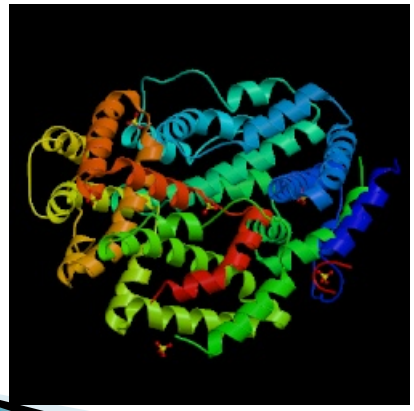
Omaima N. El Gazayerly, Ph.D.
Professor of Pharmaceutics and Industrial Pharmacy,
Faculty of Pharmacy, Cairo University



Protein Pharmaceuticals



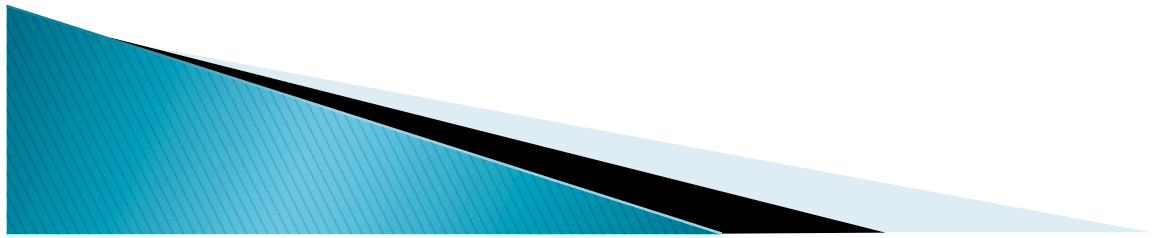
Insulin (diabetes)



Interferon α (HCV)

Protein Pharmaceuticals

- The number of approved protein drugs is increasing rapidly.
- Protein pharmaceutical sales currently approach $46 billion/yr
- By 2011 they are expected to reach $52 billion/yr



Challenges with Proteins

In vivo

- Elimination
- Proteolysis by peptidases
- Small proteins, filtered out by the kidneys very quickly
- Unwanted allergic reactions may develop (even toxicity)
- Loss due to insolubility/ adsorption

In vitro

Denaturation

Aggregation

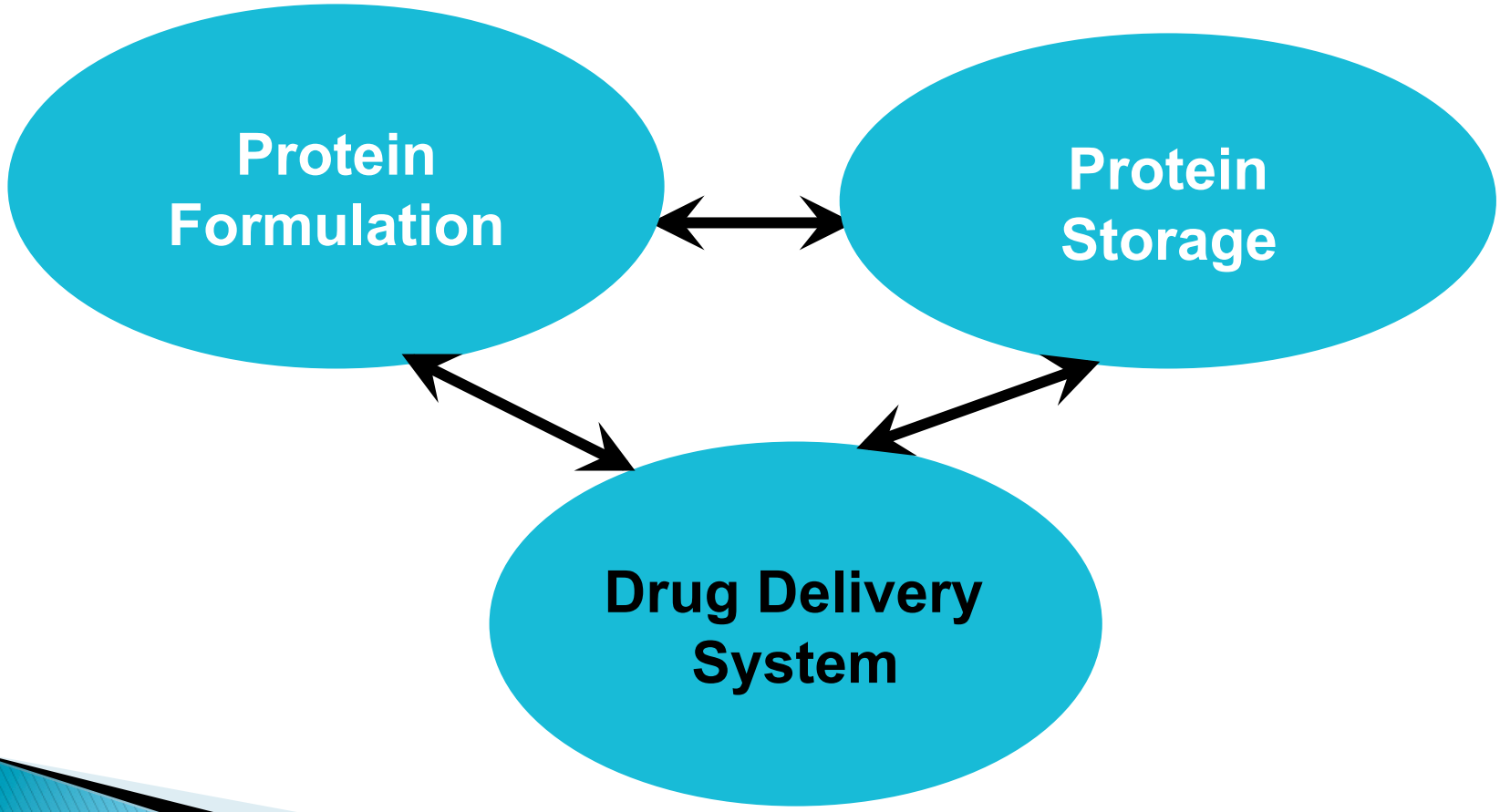
Precipitation

Adsorption

Deamidation

Oxidation

How to Deal with These Problems

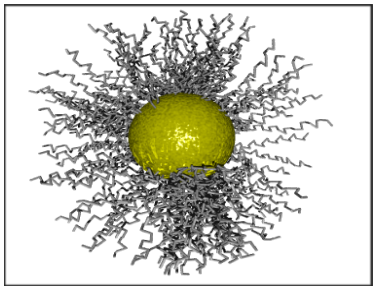


Protein Formulation

- PEGylation (Increases half life and protease resistance

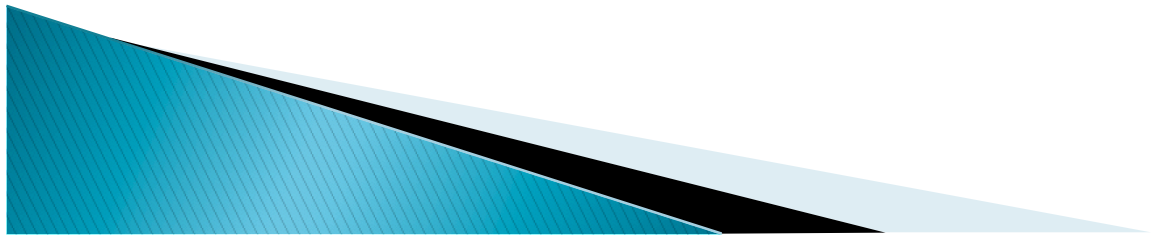
Increases solubility & stability, Reduces depot loss at injection sites)

- Proteinylation (attachment of additional protein or cross linking with serum albumin to increase half life)
- Peptide Micelles
- Formulating with permeabilizers (salicylates, fatty acids, EDTA)



Storage

- Refrigeration
- Packaging (under vacuum)
- Additives (stabilizing salts, Zn for insulin, surfactants to decrease adsorption and aggregation)
- Freeze-Drying



Drug Delivery Systems

Oral

- Ease of administration
- Patient Compliance

- Exposure to extremely acidic pH
- Poor absorption of larger drugs
- Degradation by enzymes

Parenterals

- Fast action
- No absorption issues

- Lesser patient compliance
- Fast clearance of drugs

Parenteral Delivery of Proteins

Route of delivery for 95% of proteins

Allows rapid and complete absorption

Allows smaller dose size (less waste)

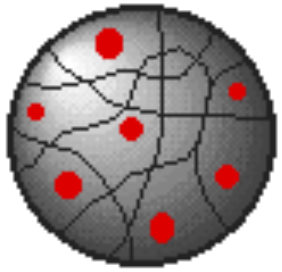
Avoids first pass metabolism

Problems with overdosing, necrosis

Local tissue reactions/hypersensitivity

Poor patient compliance

Polymeric Drug Delivery

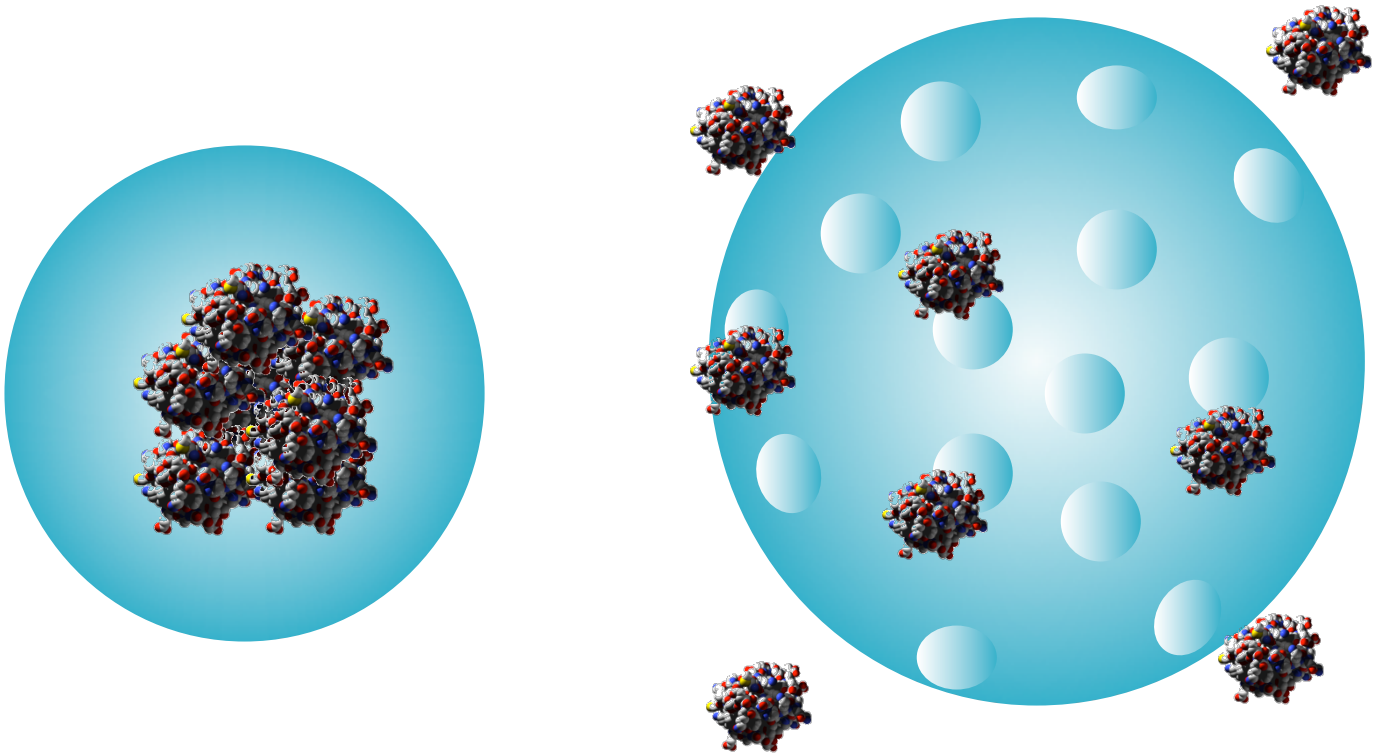


- Frequency of doses reduced
- Drug utilized more effectively
- Drug stabilized inside the polymer matrix
- Reduced side effects

- Possibility of dose-dumping
- De-activation of drug inside polymer

- Polymers should be:
  - Biodegradable
  - Biocompatible
  - Nontoxic
- Examples:
  - Polylactides/glycolides
  - Polyanhydrides
  - Polyphosphoesters

Oral Delivery by Microsphere

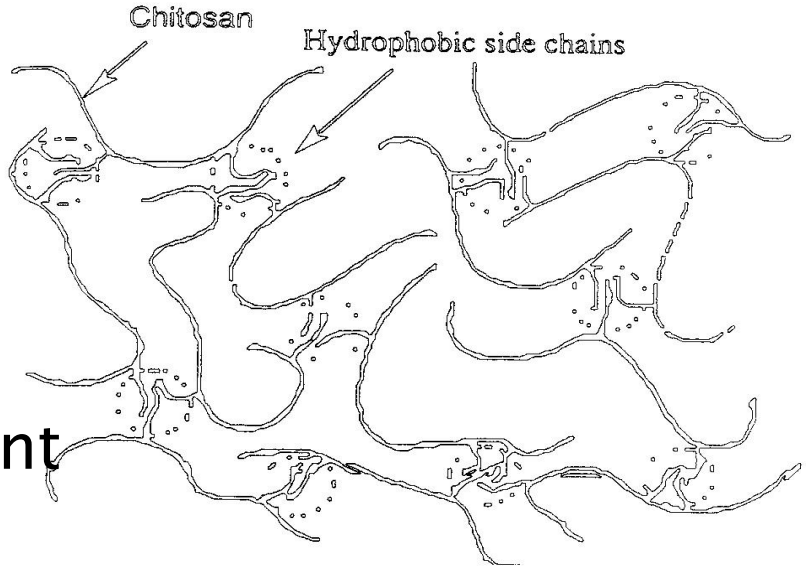


pH 2

pH 7

Hydrogel Based Drug Delivery

Hydrogels are three dimensional networks of hydrophilic polymers that are insoluble

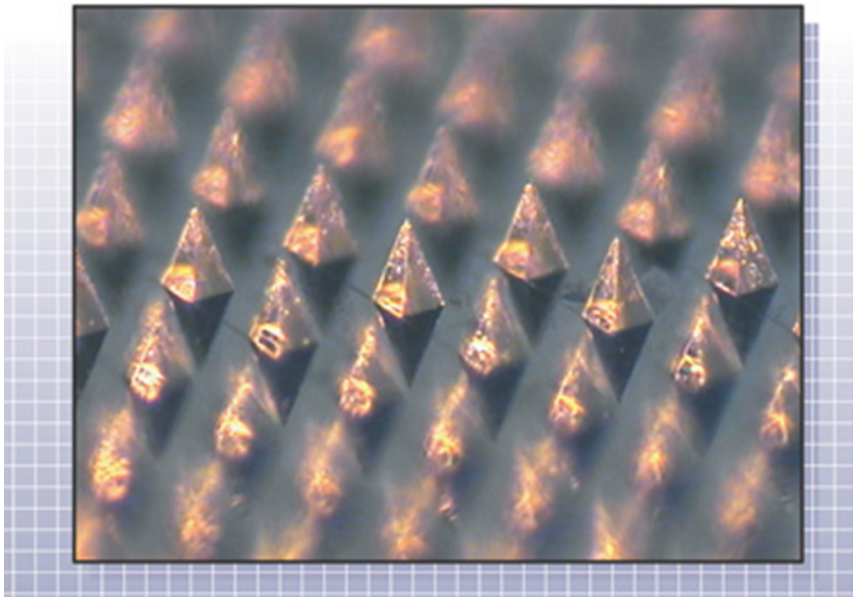


Hydrogels can swell as a result of changes in pH, temp., ionic strength, solvent composition, pressure and the application of electric fields.

Insulin has been one drug that was incorporated in hydrogels and investigated by researchers extensively

Transdermal Patches

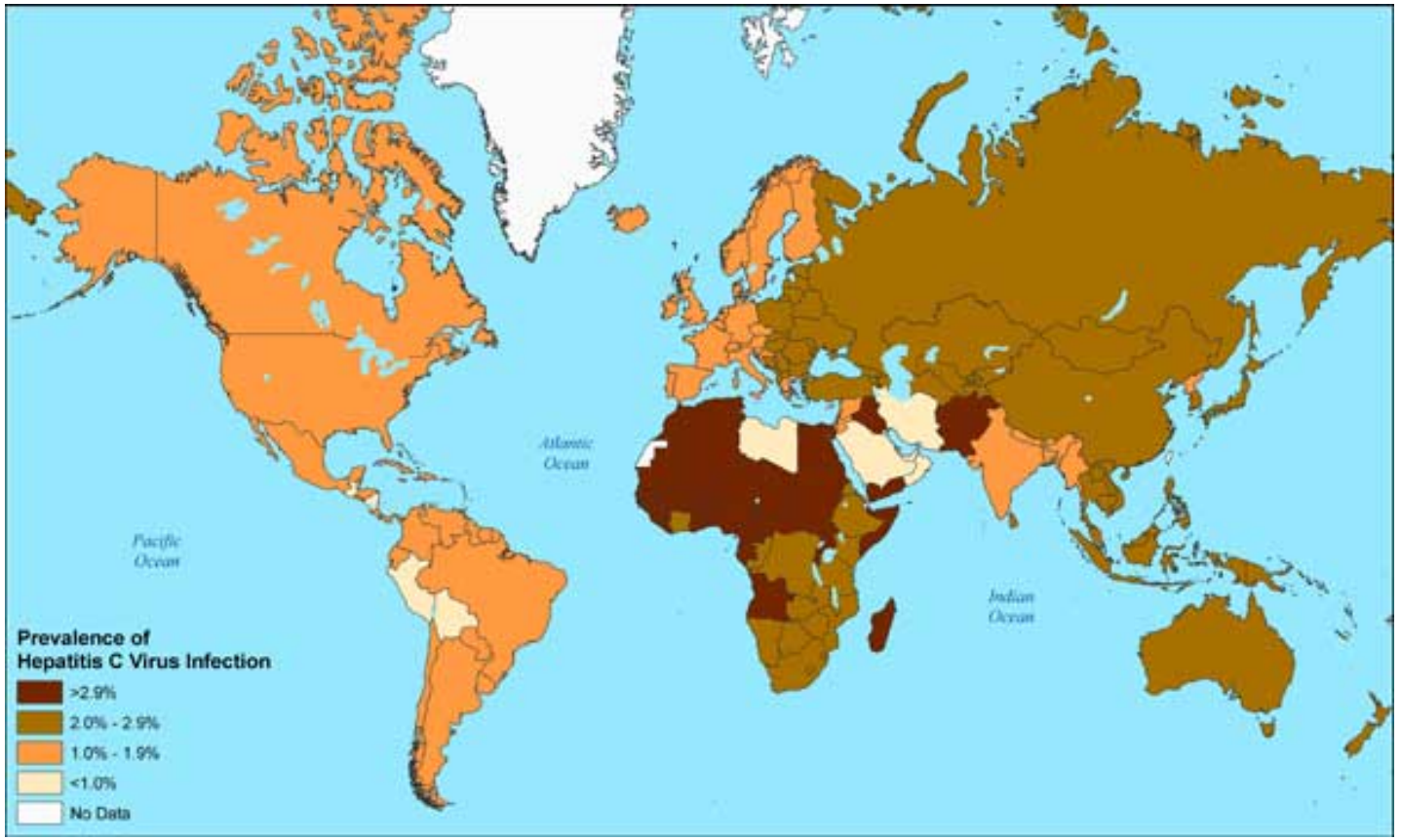
- Proteins imbedded in a simple matrix with appropriate additives
- Patch is coated with small needles that penetrate the dermal layer
- Proteins diffuse directly into the blood stream via capillaries
- Less painful form of parenteral drug delivery



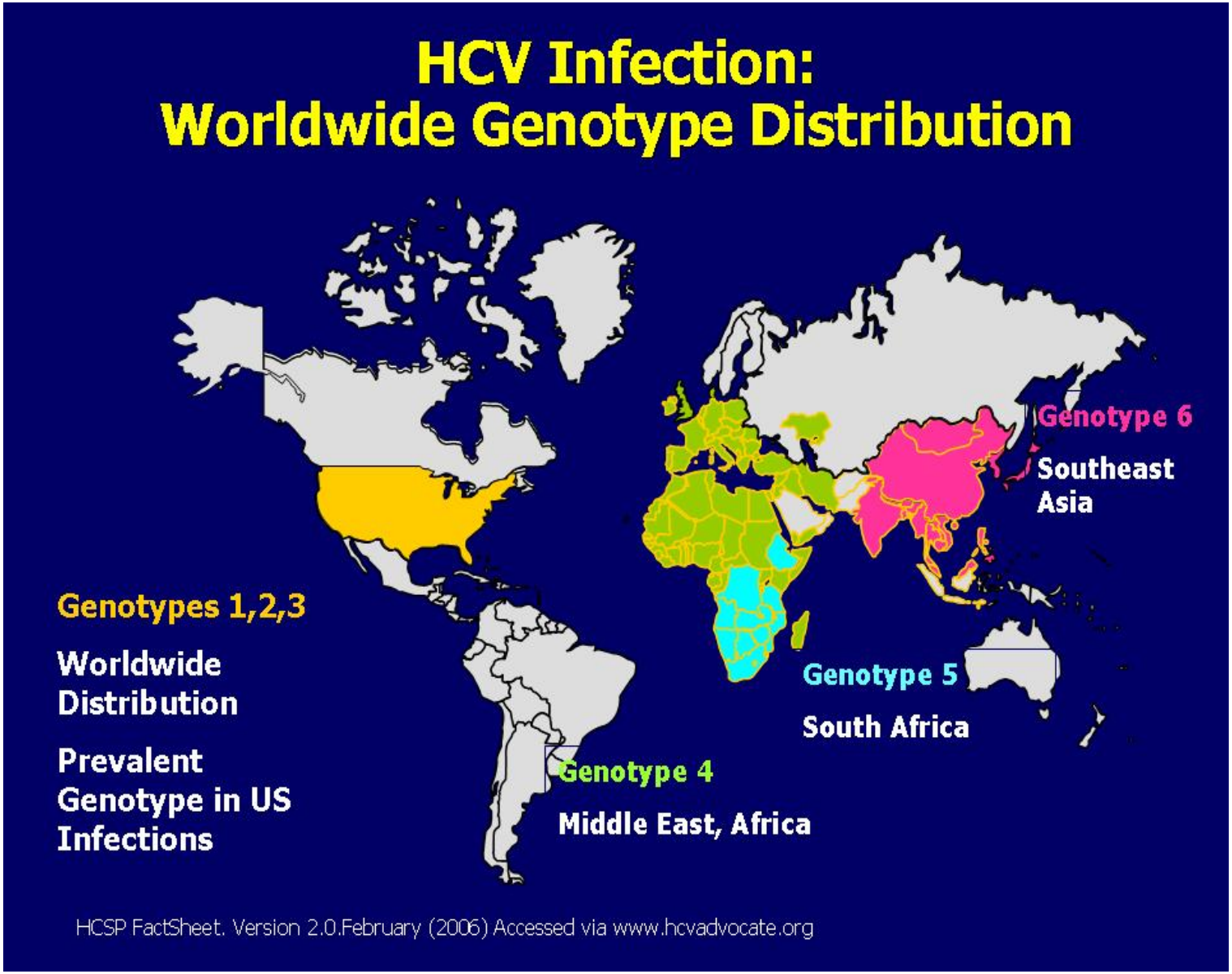
Micro fabricated needles to facilitate permeation of peptide drugs

Hepatitis C virus (HCV) is a common infectious agent, affecting about 170million individuals worldwide

Incidence in Japan is about 1.2%. Alarming rates were reported reaching as high as 14.5% in Egypt



HCV Infection: Worldwide Genotype Distribution



HCSP FactSheet. Version 2.0.February (2006) Accessed via www.hcvadvocate.org

Current therapies for hepatitis C

The main goal of treatment of chronic hepatitis C is to eliminate detectable viral RNA from the blood

Lack of detectable HCV RNA from blood 6 months after completing therapy is known as a sustained virological response (SVR)= **CURE**

Monotherapy

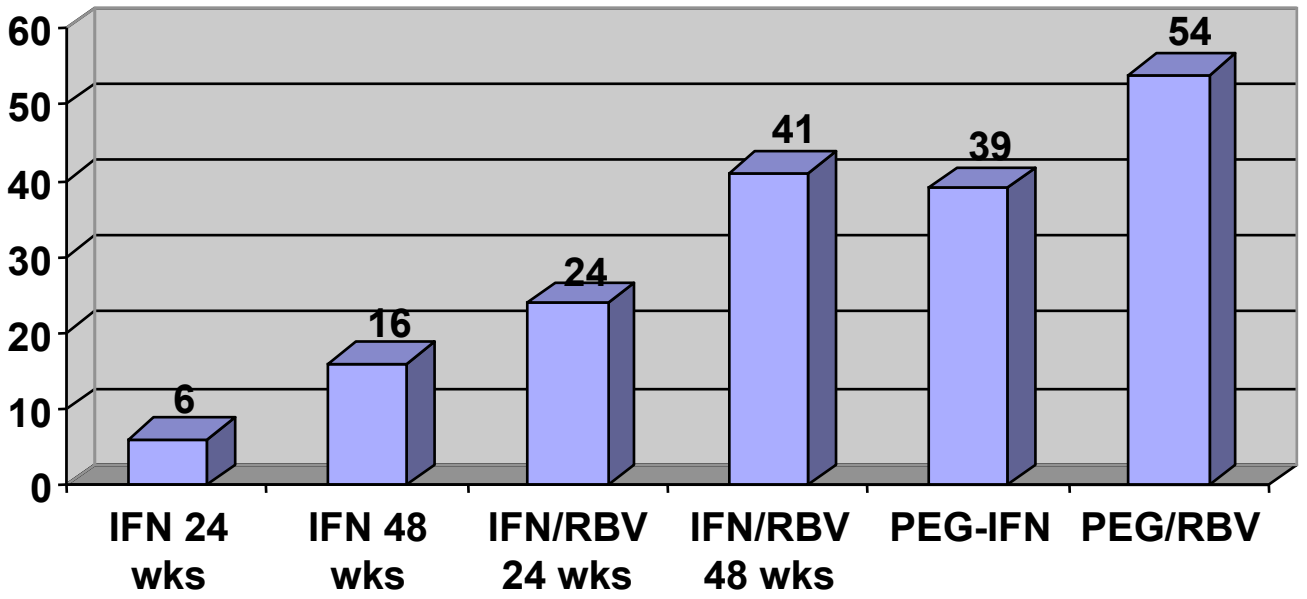
Interferon α-2a and inteferon α-2b are approved for the treatment of adults with chronic hepatitis C as single agent. IFNα-2a and IFNα-2b are administered by IM or SC.

Combination therapy

IFNα-2a or IFNα-2b plus ribavirin for 48 weeks led to a marked improvement of hepatitis C therapy compared to monotherapy with IFN.

HCV – Treatment

Sustained Virologic Response Rates



Multiple randomized controlled trails

Properties of Interferon

Physicochemical

IFN is a family of highly homologous, species-specific proteins, water-soluble, antiviral effect, leads to an enhancement of immune response in hepatitis C virus treatment

Pharmacological

IFN limits amplification and spread of viruses during infections by direct and indirect mechanisms leading to the activation of the adaptive immune response

Pharmacokinetic properties

IFN is degraded by gastric acid and proteolytic enzymes in GIT

Administered by IM or SC injection for systemic effects (80%)

IFN is widely and rapidly distributed into body tissues after parenteral administration with the highest concentrations occurring in spleen, kidney, liver, and lung

Elimination of IFN is rapid from plasma following IV injection

Modified IFN

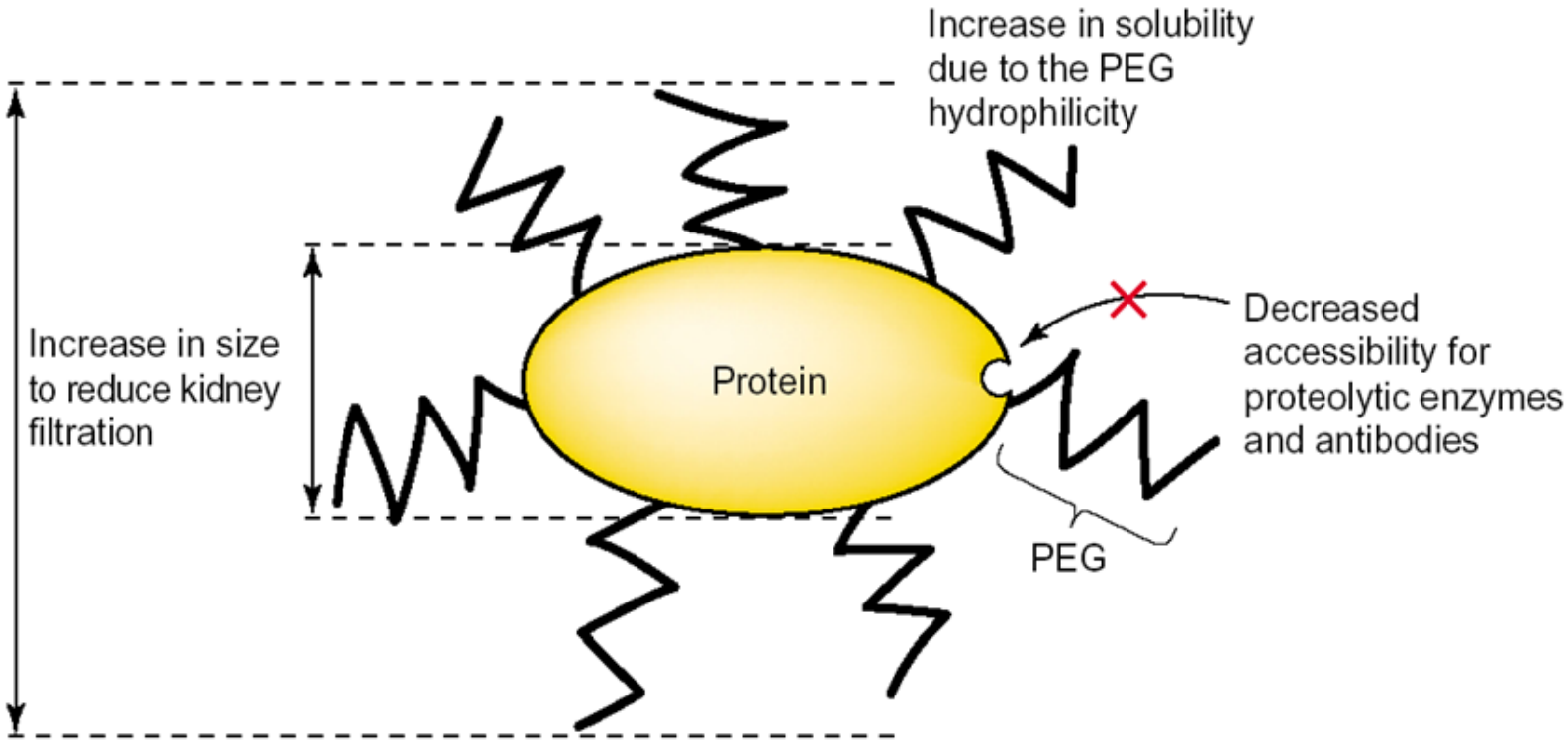
Pegylated interferon
For long-circulation
The objective of PEGylation is to shield the IFN molecule from enzymatic degradation, thereby reducing systemic clearance to provide long-circulating IFN in the patient's serum.

Albuferon (albumin–IFNα-2b). Albuferon is a protein consisting of the antiviral properties of IFN*α* -2b genetically fused to human serum albumin (HSA). Decrease dosing frequency to 1 SC every two weeks

Attaching polymers to proteins is called “PEGylation”

PEG is FDA approved, Biocompatible

- Decreases immunogenicity
- Reduces depot loss at injection sites



PEG–IFN ↑ size of the IFN molecule. ↑

Prolonged biological half-life (4-400X)

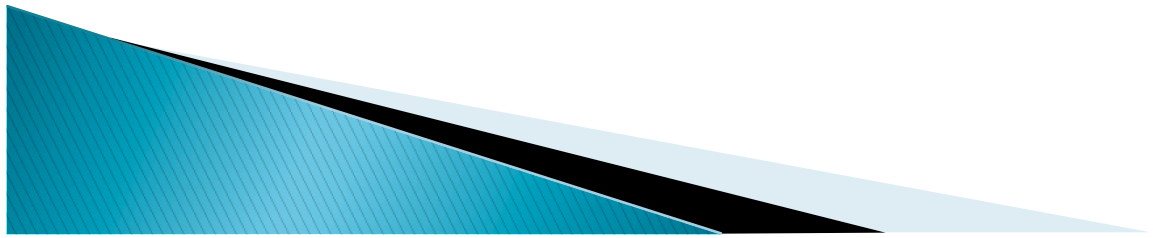
As a result the PEG–IFN has allowed the weekly dosing frequency from 3 to 1. ↓

A Pilot study on the **efficacy and safety of Pegylated Interferon in Children and Adolescents Infected With Chronic Hepatitis C in Egypt** was conducted.

Study Population

Children whose age between 10 and 18 years, meeting the preset inclusion and exclusion criteria and who were infected with HCV were eligible for enrollment into the study.

Thirty patients were enrolled to receive pegIFN once a week with ribavirin twice daily; viral load and experienced adverse effects were then assessed.



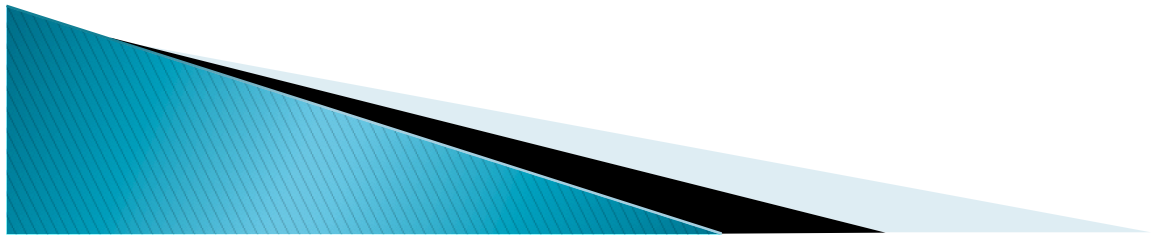
Efficacy reporting 24 and 48 weeks after therapy commencing was assessed.

Out of the thirty patients who completed 48 weeks of therapy:

1. **Fifteen** patients are considered non-responders to the therapy regimen.

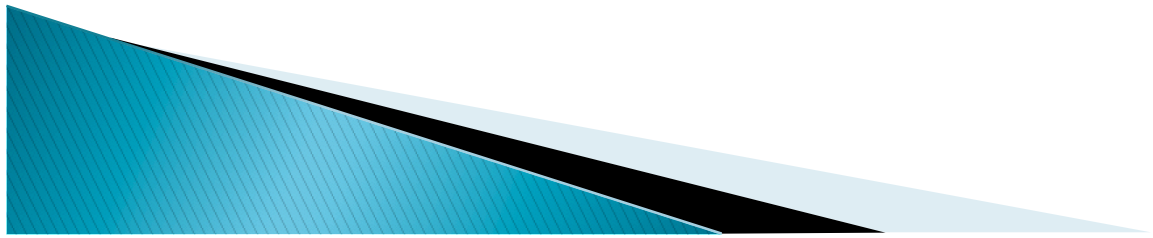
2. **Fifteen** patients have been cleared from the virus according to the results of qualitative PCR obtained at week 48.

The percentage cure or SVR is 50%



Side effects recording showed expected side effects including pyrexia, headache, anorexia

The recorded side effects subsided by week 12 and all children attended their schools without difficulties.



Study limitations:

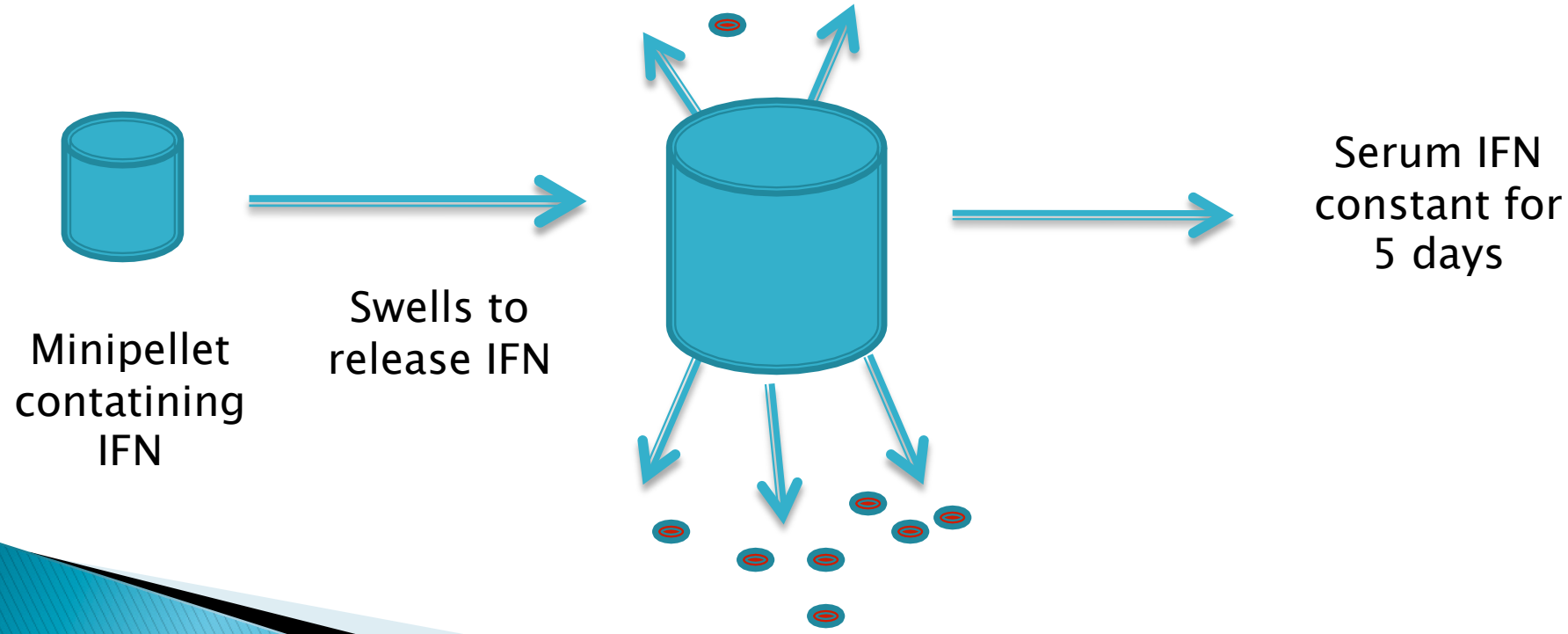
1- Sample size

2- Control group

3- The age range lying in the upper range above 10 years.

Novel sustained release injectable drug delivery systems

IFN minipellet is a matrix-type long-acting drug delivery system of IFN incorporated in collagen as a biodegradable carrier



IFN MLV

provide controlled release and stabilize IFN

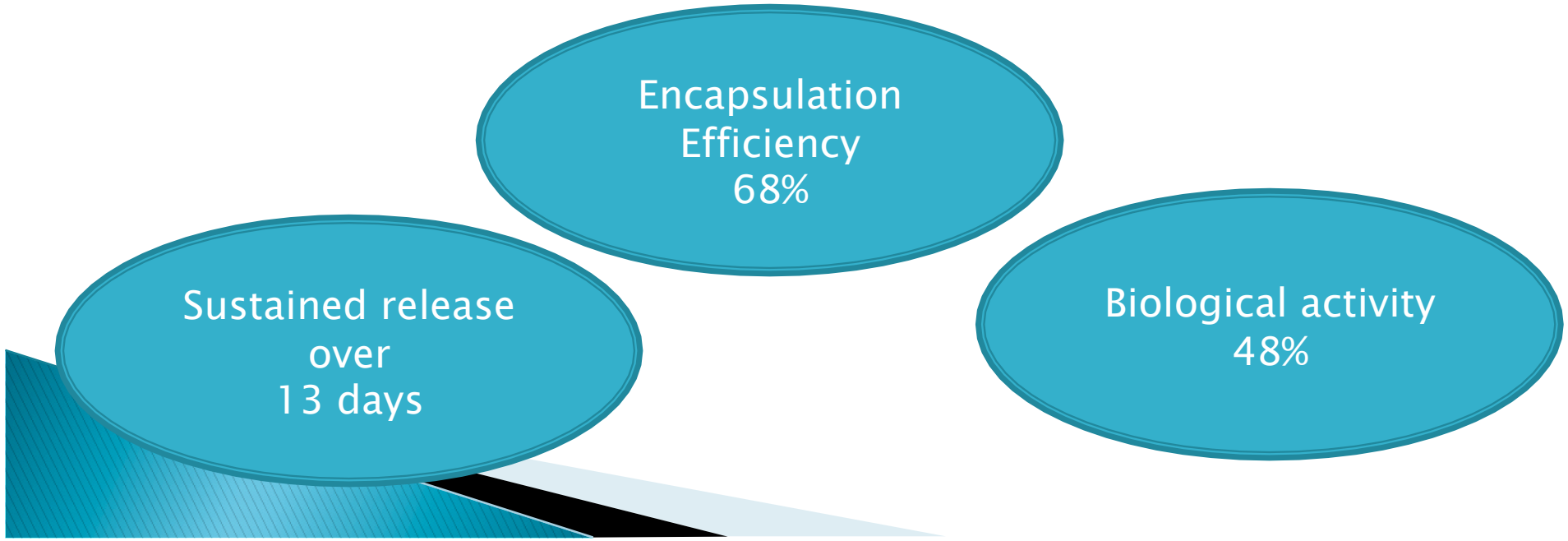
alter the pharmacokinetics, tissue distribution and uptake of IFN

Good encapsulation for IFN

Activity of IFN and size of liposome are stable for 4 months

IFN Microspheres

IFNα-2a was loaded within alginate microcores and then microencapsulated into poly-dl-lactide-poly(ethylene glycol) (PELA) copolymer using a w/o/w solvent extraction method.



Transdermal Route

Transdermal delivery systems present an attractive, noninvasive delivery method to achieve steady blood levels of IFN

Iontophoresis did not have any effect on the bioactivity of the IFNα-2b

When iontophoresis was applied to microporated skin, the amount delivered

No peak and valley pharmacokinetic profile

Implants

An implant device containing lyophilized IFNα-2a and PEG 6000 for continuous delivery of IFNα-2a provided sustained release

Buccal Route

Provides constant level of IFN, decreases side effect, convenient and safe transport method
Requires an absorption promoter for the delivery of IFN across the buccal mucosa

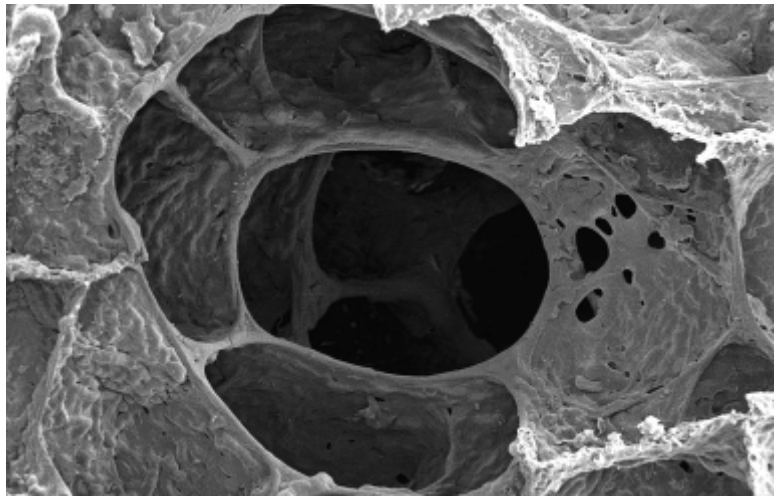
The Nasal Route has been actively investigated in the last few years for IFN

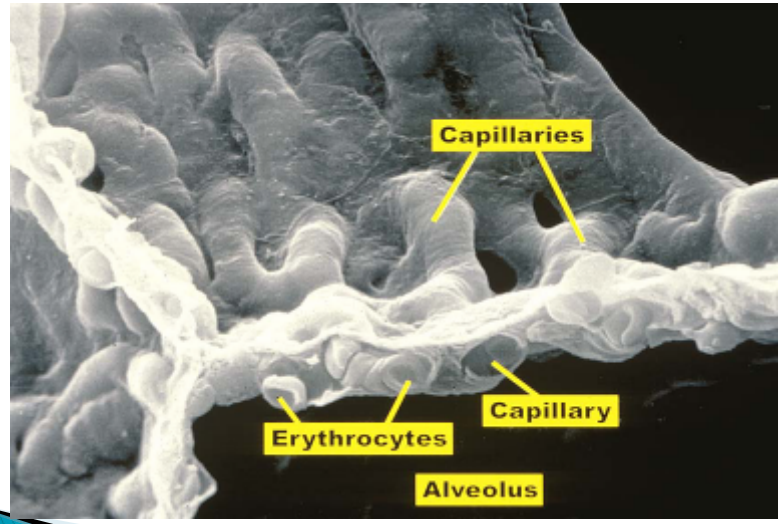
- The nasal administration has attracted much interest now a days due to
  - Relatively rapid absorption of drug
  - Low metabolic degradation
  - Relative ease of administration
  - Selective to peptide structure and size

The use of absorption enhancers seems necessary for significant absorption of IFN

Nasal bioavailability of IFN was increased up to 32.3% by the addition of 1% sodium taurocholate.

Pulmonary Route





Promising alternative for delivering IFN:

1- large surface area of the alveolar epithelium 140 m^2 (500 million alveoli)

2- short distance of the air to blood pathway from the lung. (1 µm)

Some absorption enhancers are needed to enhance the pulmonary absorption of IFN from the lung

ORAL ROUTE

Development of oral formulation of IFN would greatly expand its clinical application. However, bioavailability of oral IFN has been known to be low

Only about 40% of IFNα absorbed from the GIT can reach the systemic circulation due to proteolysis.

Enteric coated tablets for oral administration of Belerofon were prepared (a single point aminoacid mutation of native IFN, designed for higher resistance to proteolysis)

Oral bioavailability of Belerofon is based on its low sensitivity to protease degradation in the intestine which renders the molecule available for absorption into the bloodstream.
In animal studies, appropriate oral doses of Belerofon can reach blood levels comparable to those obtained from SC IFN. Furthermore, antiviral activity of Belerofon in rats after oral administration is significantly superior to that of native IFN. Consequently, a clinical phase 1 study is performed on oral Belerofon in healthy adult subjects for safety, tolerability and pharmacokinetics.

New research focuses on the development of novel modified interferon molecules which demonstrate reduced side effects and extended systemic circulation time, to ultimately provide greater efficacy. Alternative routes for IFNα delivery, such as oral delivery, demonstrate challenging but promising areas of research for improving patient compliance.

Thank you

دومو أريجاتو

جوزايماس