

# PromISS 4 Familiarization

## Scientific background

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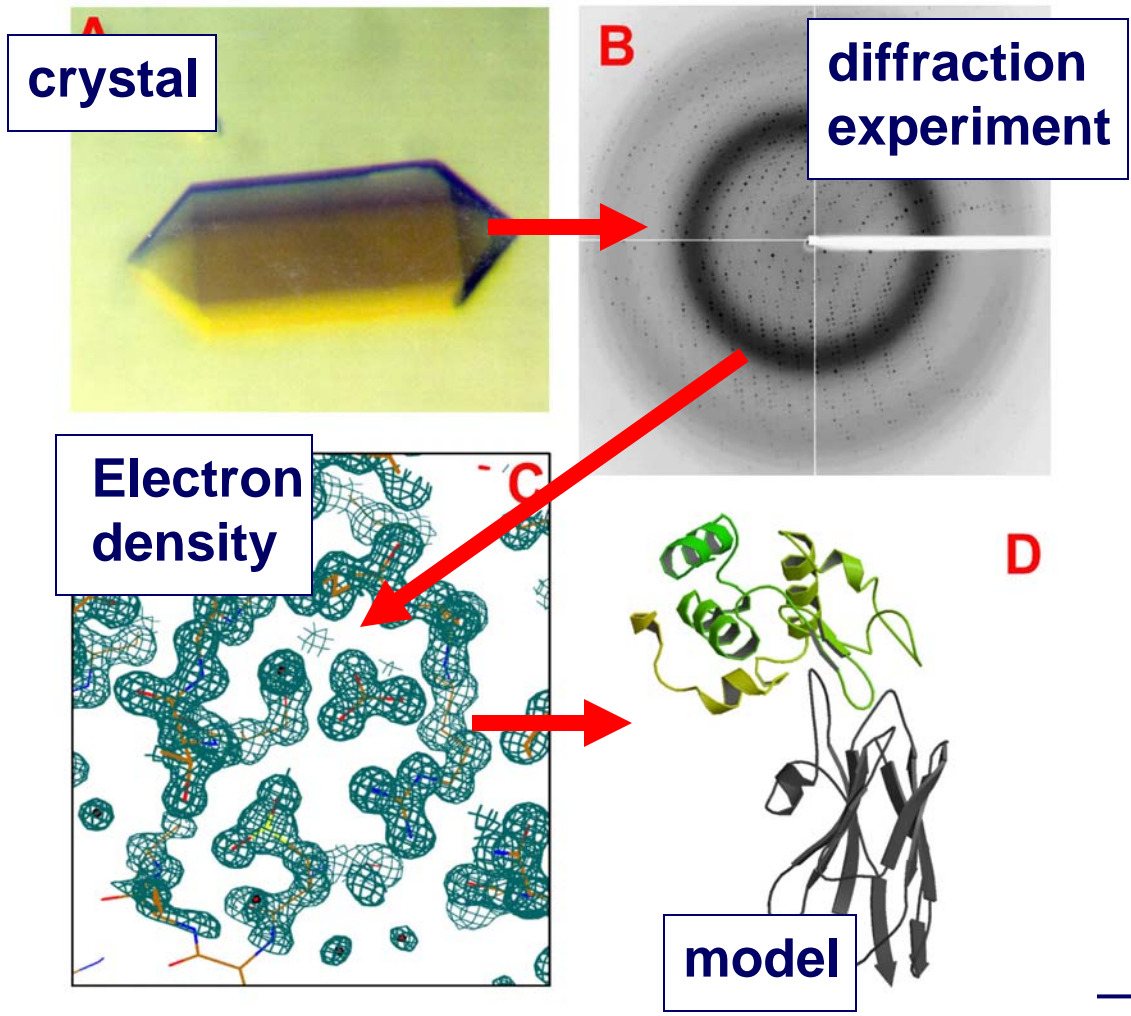


## Lesson Objectives

- 1 State the scientific background of the experiments
- 2 Describe results from previous experiments
- 3 State the objectives of the PromISS 4 experiments



# Scientific background 1: Why do we need protein crystals?



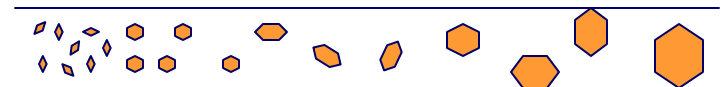
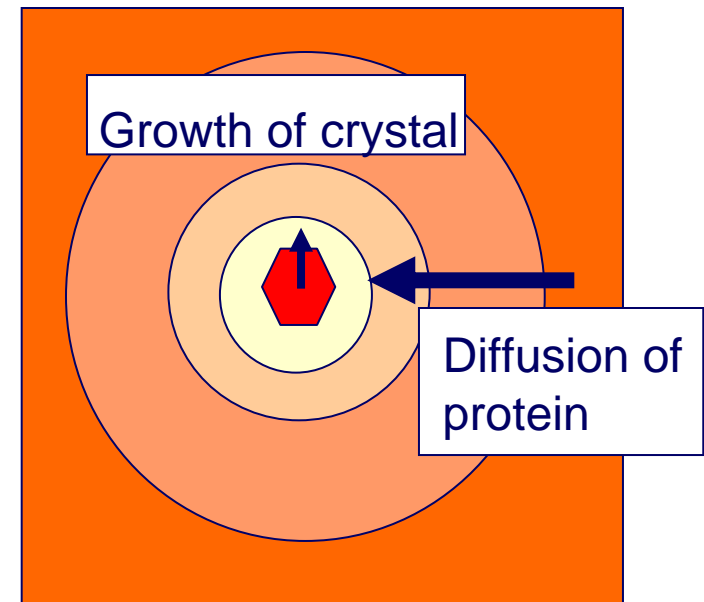
**Mycoplasma:  $\approx$  500 genes**  
**Fruit fly:  $\approx$  15.000 genes**  
**Human:  $\approx$  30.000 genes**  
**Lungfish:  $\approx$  50,000 genes**

**Hundreds of thousands of proteins need to be crystallised in the 'structural genomics' context**

## Scientific background 2: why microgravity?

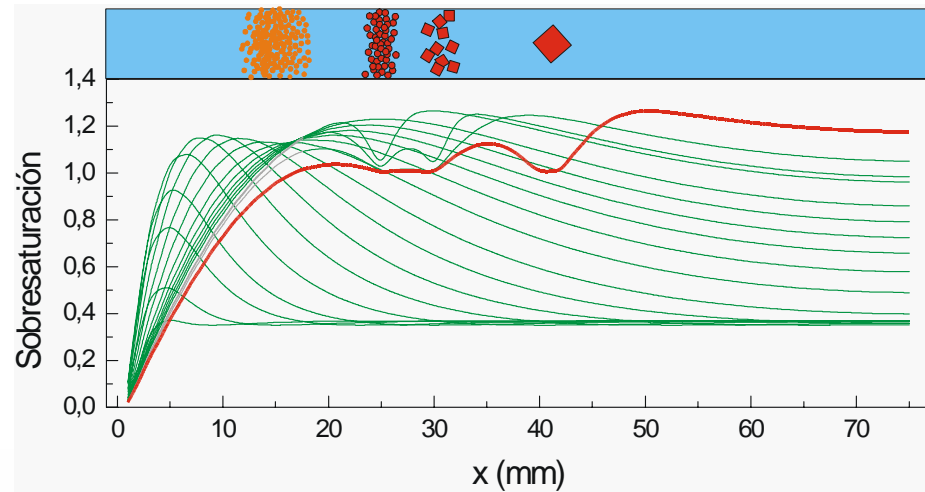
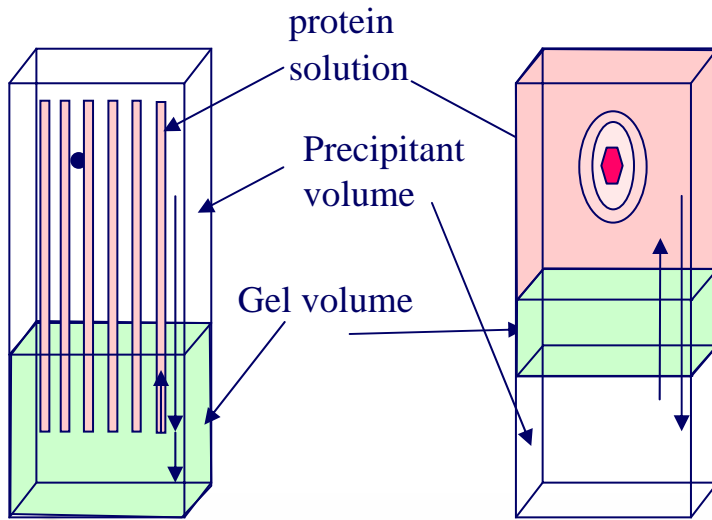
Protein crystal growth is a very complex, and fundamental processes underlying protein crystal growth are not yet well understood. With Promiss we aim to:

- Collect a large quantity of data on the **microgravity effect** (about 20 % of proteins produce better crystals in microgravity)
- Understand ‘microgravity effect’  
**verify hypotheses:**
  - Depletion zone model **Promiss I and II**
  - Impurity depletion zone **Promiss 3**
- Verify models and simulations of crystal growth processes





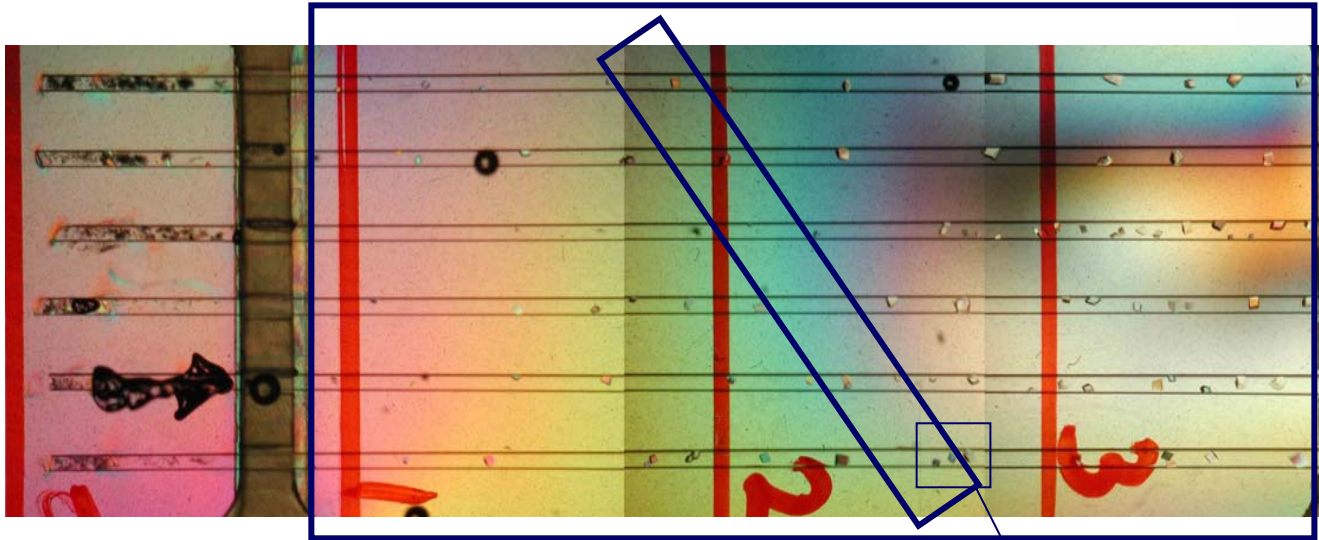
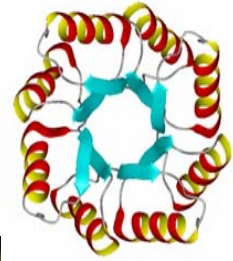
## Scientific background 3: Crystallisation in a diffusive environment



**Counterdiffusion crystallisation requires a diffusive environment**

- Gels
- Microgravity

## Previous experiments 1: TIM in PromISS I

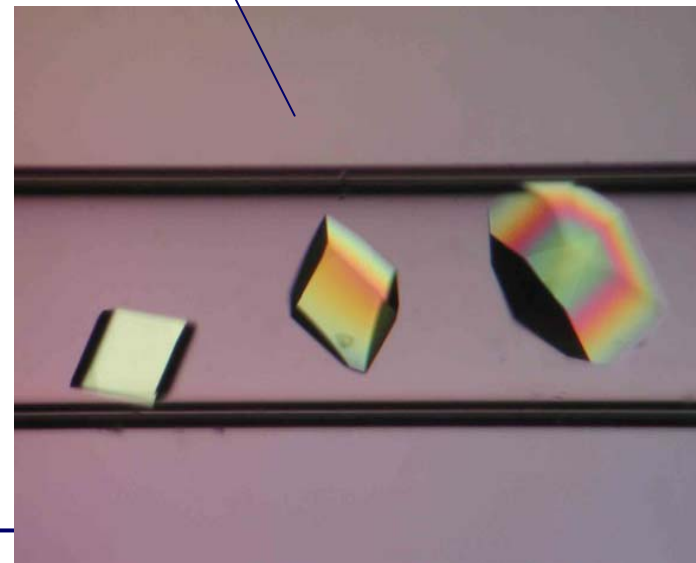


Crystallisation within timeline **OK**

Airbubbles **±OK**

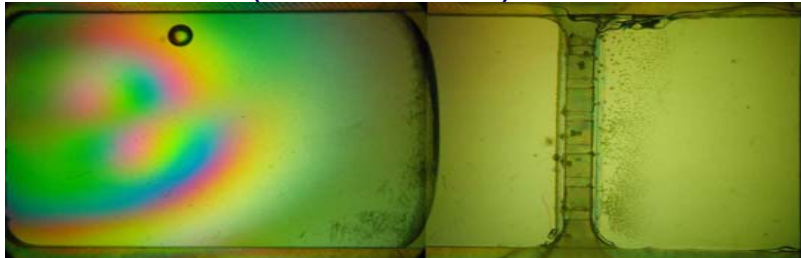
Apparent crystal quality **OK**

Range of crystals observed **OK**

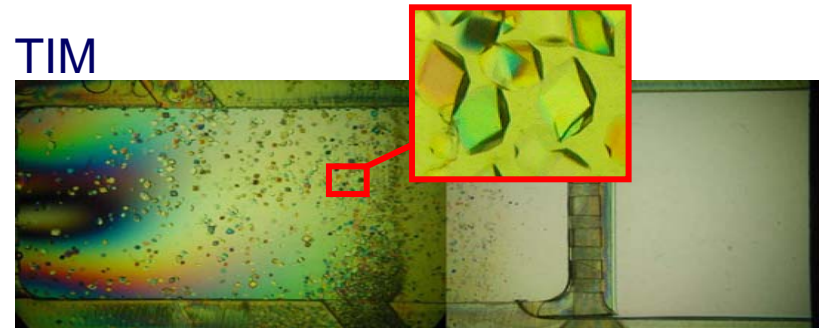


## Previous experiments 2: PromISS 3

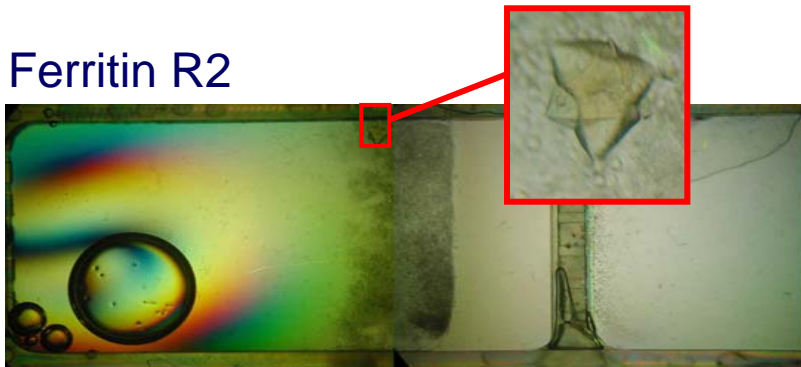
Ferritin R1 (A. Chernov)



TIM



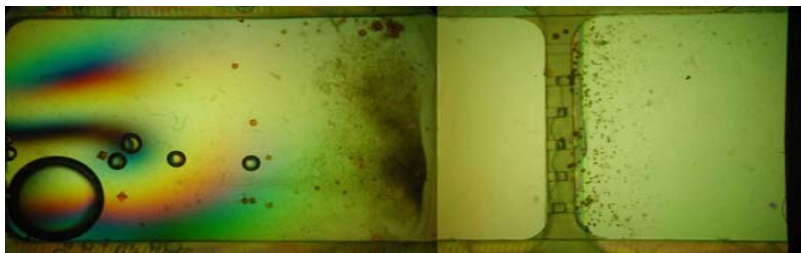
Ferritin R2



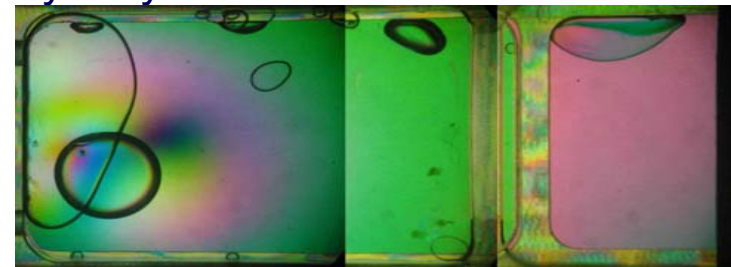
Lumazine synthase



Ferritin R3



Lysozyme seeds





## Previous experiments 3: Summary of previous observations:

1. The diffusive environment clearly affects crystal **nucleation**
2. Experiments of the ISS show there is non-negligible **crystal movement** (  $< 70 \mu\text{m/hr}$  )
3. (Comparison of results between ISS and Foton could provide more information)
4. For at least two model protein there is a **clear microgravity benefit** that can not be explained by the depletion zone model alone.
5. Crystal **Polymorphism**: in a diffusive environment the crystallising system moves through the phase diagram in a different manner, leading to different polymorphic growth.
6. crystal quality studies show gel can provide similar benefits as microgravity on the ISS, although **mosaicities are higher in gel**, and variation of quality of crystals is larger.



## Objectives: aims for PromISS 4

Verification of the depletion zone model

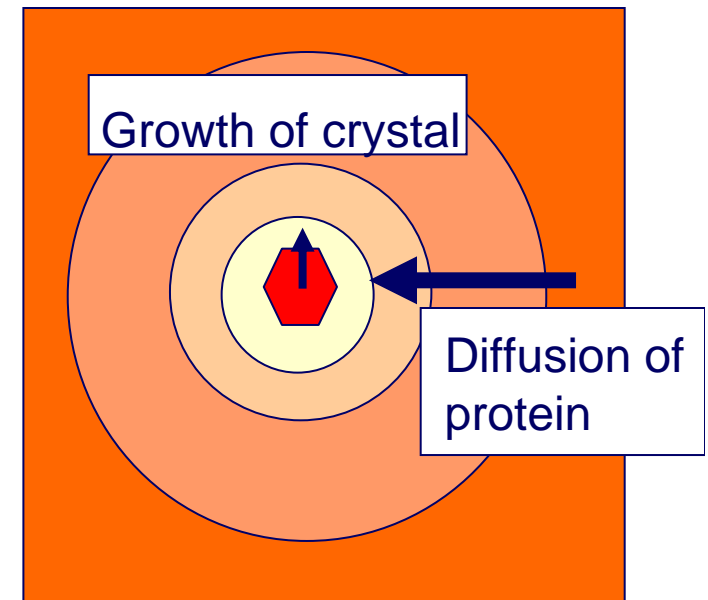
-Use proteins that crystallise in the diffusive regime, so that they can build up large depletion zones

- Catalase
- Ferritin
- Lysozyme

- Quantify the depletion zones by interferometry

- Measure the quality of crystals produced in diffusive environment (microgravity, gels) and in convective environments (ground)

Do microgravity experiments to understand protein crystallisation, and develop better crystallisation strategies on ground





## Scientific background: Participants

Promiss experiments are only possible because of many people from different fields working and discussing together

### Protein crystallisation

#### APCF

Alex Chernov (MSFC)

Lode Wyns/Ingrid Zegers (VUB)

Sevil Weinkauf (Munchen)

Jean-Paul Declercq (UCL)

Joseph Martial (Ulg)

JuanMa Garcia-Ruiz (LEC)

Adriana Zagari (Una)

### GCB Granada

#### Crystallisation box

JuanMa Garcia-Ruiz (LEC, Granada)

### Optics Interferometry

Frank Dubois/ Cedric

Schockaert/ Christophe

Minetti/ Patrick

Queeckers

ULB-MRC

### Modelling and simulation

Jean-Pierre

Boon, Gregoire

nicolis (ULB)

Luigi Carotenuto (MARS, Naples)

### PromISSdevelopment/ construction

ULB

Pedeo

Verhaert

Astrium

ESA, RSC Energia, NASA

Many thanks are due to the astronauts that actually do the experiments !