Medicinal Chemistry: An Overview

Course Outline

Lecture	Date	Topic
1	2015/12/17	General Aspects of Medicinal Chemistry
2	2016/01/07	General Biochemistry
3	2016/01/21	Principles of Chemical Synthesis
4	2016/02/04	Chemical Synthesis of Small and Complex Molecules
5	2016/02/18	Chemical Synthesis of Peptides
6	2016/04/07	Strategies for Discovering Lead Compounds
7	2016/04/17	Structure-Activity Relationships
8	2016/04/25	Spatial Organization, Receptor Mapping and Molecular Modeling
9	2016/05/02	Pharmacokinetic Properties
10	2016/05/09	Legal and Economic Aspects of Drug Development

Pharmacophore Identification

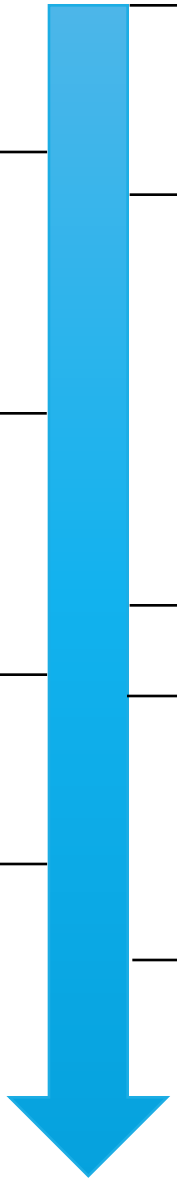
A pharmacophore is an abstraction of the essential molecular features responsible for the binding of a set of ligands to a macromolecular target.[1] It is not a discreet functional group or substance class possessing biological activity.

If the 3D structure of a receptor and the binding site is known, then ligands can be docked into the binding site to check for optimal interaction. However, in the absence of such information, analogs of active ligands can be designed to generate novel active compounds.

A pharmacophoric element or pharmacophore feature is an atom or group of atoms (a hydrogen bond donor atom or an aromatic ring system) common to biologically active compounds with respect to a target protein and critical for the activity.

Wermuth *et al. Annu. Rep. Med. Chem.* **1998**, *38*, 385–395.

History of Pharmacophore Development



1878; Langley introduced the term "receptive substances."

1894; Fisher introduced the lock- and-key concept.

1900; Ehrlich introduced the terms "pharmacophore" and "receptor."

1940s; Simple 2D models were employed.

1963; Perutz solved the X-ray structure of haemoglobin.

1969; Marshall pioneered the development of the pharmacophore concept and its application to structure-activity relationships.

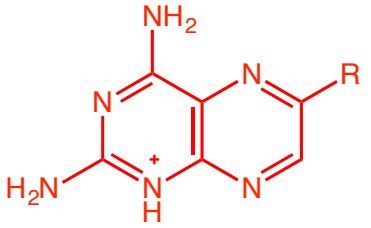
1970; Gund carried out the first *in silico* screening method.

1990; Marshall displayed carried out the first *in silico* screening method.

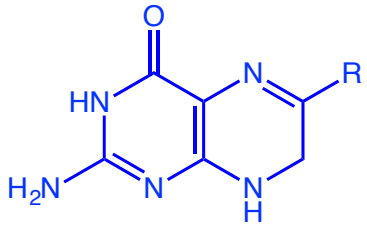
2012; an array of automated pharmacophore discovery programs have been developed.

Significance of the Pharmacophore Concept

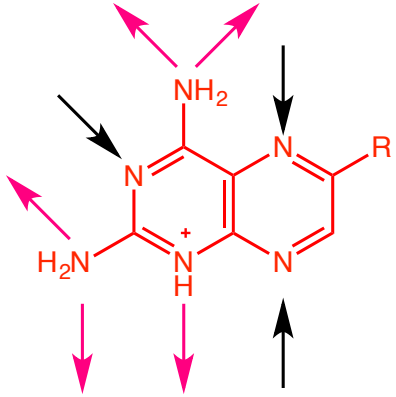
Pharmacophoric features are more useful than topological ones since the latter are often misleading in the superpositioning of two molecules in relation to their binding mode.

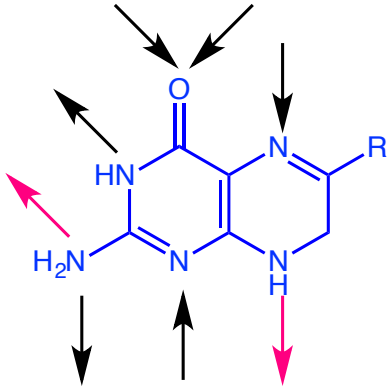


Methotrexate

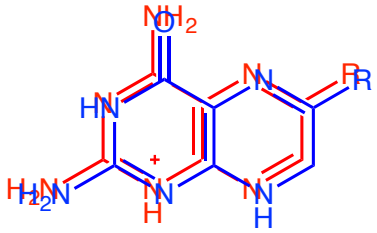


Dihydrofolate

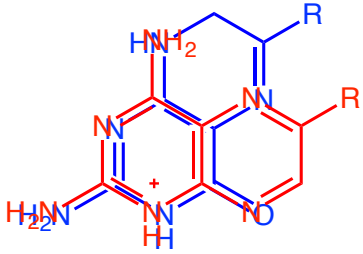




Hydrogen bond donors and acceptors



Atom-based alignment



Pharmacophore alignment

Bohm *et al.* [*Drug design: The path to drugs*]. Heidelberg, Germany: Spektrum Akademischer Verlag; **1996**.

Application of Pharmacophores

1) To define relevant pharmacophoric features.

2) Scaffold hopping: detection of molecules with novel chemotypes by virtual screening of large compound libraries.

3) Parallel pharmacophore-based screening to predict pharmacological profiles for lead structures *in silico.*

Pharmacophore Modeling

Step 1: choosing a set of active ligands that bind to identical target (binding site)

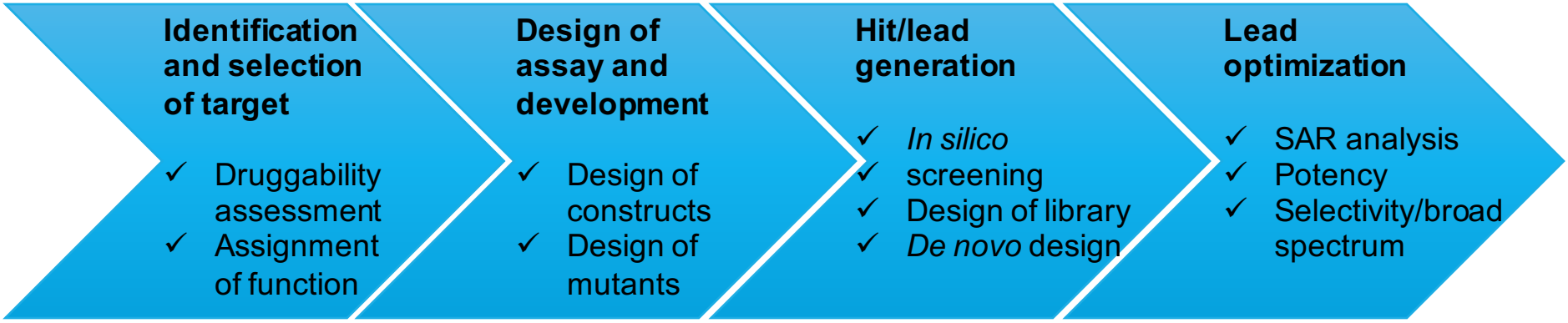
Step 2: conformational analysis of all the ligands.
The lowest energy minimum conformation is not necessarily the bioactive conformation, hence, all low-energy conformations must be calculated. Experimental techniques such as NMR can only give information on a few conformations of a molecule. The more rotatable bonds a molecule has the more demanding the calculation of all its minima.

Step 3: defining pharmacophoric elements

Step 4: determination of a common 3D-pharmacophore by molecular superposition of the ligand conformations.
Conformational flexibility is a major challenge when addressing 3D-pharmacophore–ligand superposition. A feasible but time-consuming solution is to carry out the identification of common chemical features and the conformational search concurrently (flexible alignment). Alternatively, pre-generation of conformation (rigid-body alignment) makes the overlay algorithm faster.

Protein Crystallography: Introduction

The impact of protein crystallography on the drug discovery process is immense.

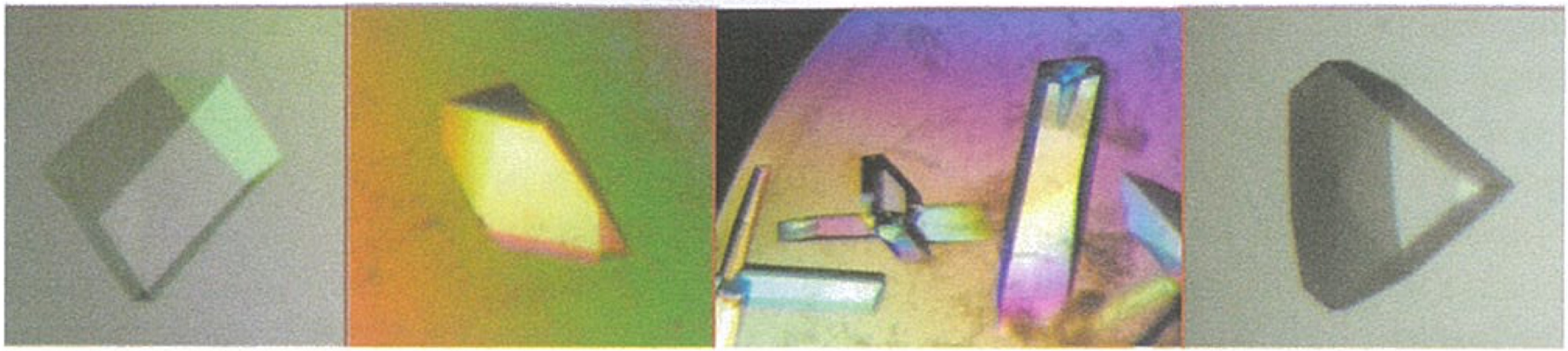


The development of protein crystallography dates back to 1840 when Hünefeld reported hemoglobin crystals. This paved the way for the determination of the molecular nature of viruses and enzymes by their isolation in crystalline form between 1926 and 1935. X-ray diffraction led to the double helical structure of DNA in 1953. In the intervening years every aspect of of the crystallographic process has been proved such that there are over 100,000 crystal structures in the Protein Data Bank.

However, the bottleneck of protein crystallography is the preparation of suitable crystals for X-ray analysis.

Protein Crystallography: Basic principles

Protein crystals are regular 3D arrays of identical molecules or molecular complexes. It is the combined diffraction of individual molecules in a crystal that produces measurable.



β-secretase inhibitor complex

human farnesyl pyrophosphatase in complex with zoledronic acid

abl kinase domain in complex with imatinib

cdk2 inhibitor complex

Crystal Packing

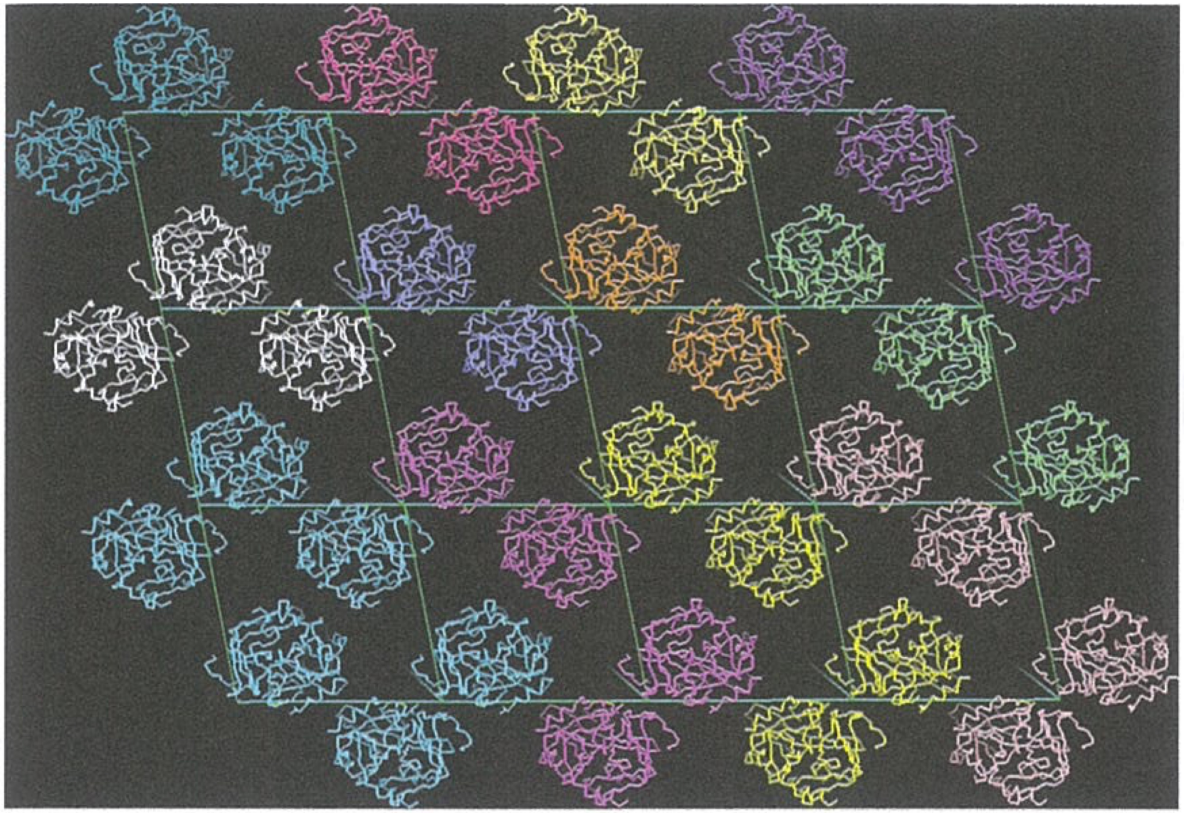
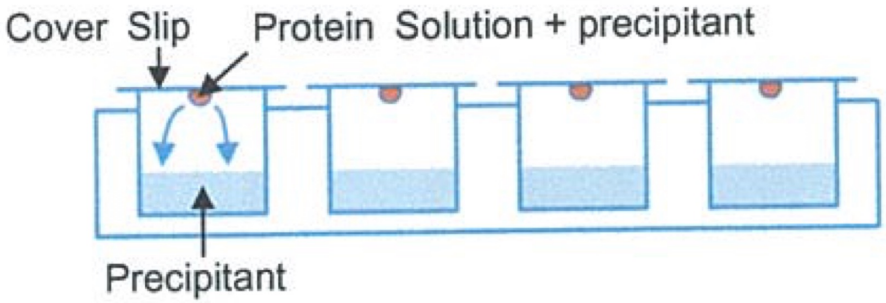


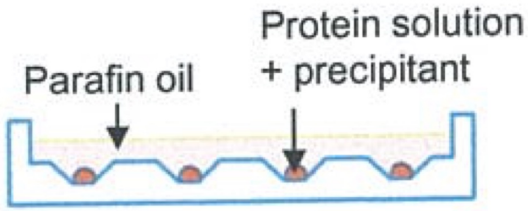
FIGURE 22.4 Crystal packing of a human thrombin complex. Twelve unit cells with one layer of molecules are shown. By looking carefully, one can see that the two molecules in each unit cell are rotated 180° with respect to each other. Protein crystals used for X-ray diffraction extend into three dimensions and consist of many layers of molecules. The next layer of thrombin molecules fits into the holes present in the layer shown.

How to Obtain Crystals

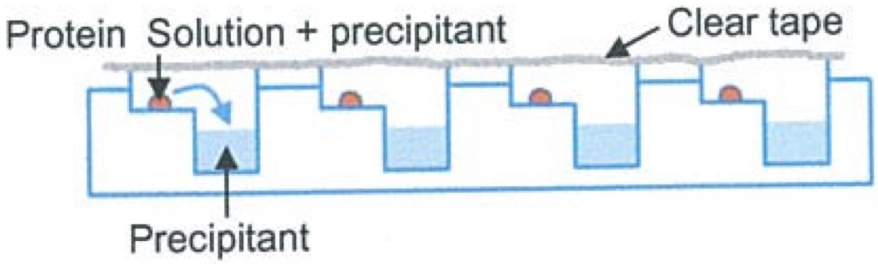
Hangingdrop



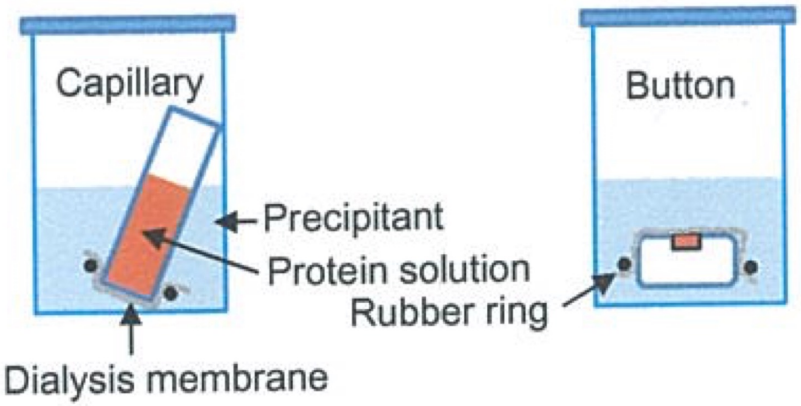
Microbatch



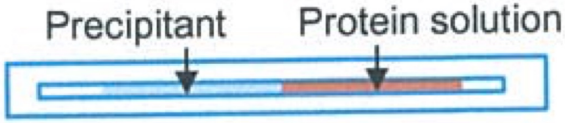
Sitting drop



Dialysis



Free interface diffusion



Obtaining Crystals: Problems and Solutions

1) Published crystallization conditions

2) *De novo* crystallization

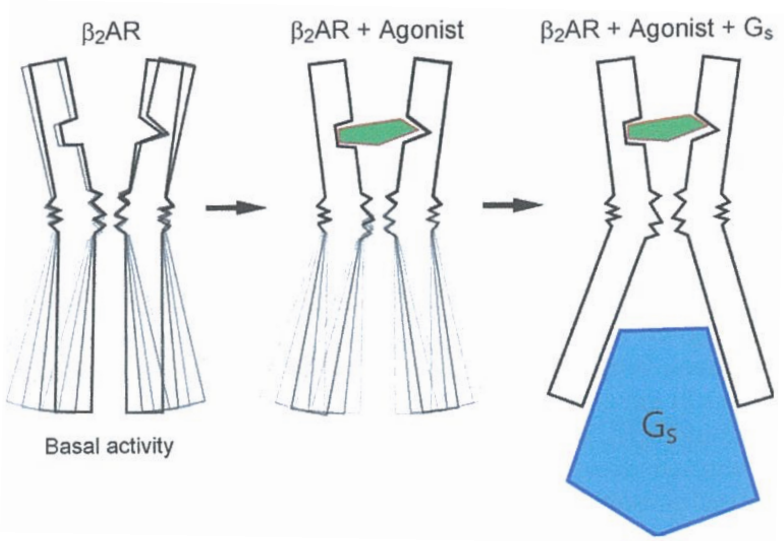
3) Conformational heterogenicity

4) Glycosylation

5) Proteolytic cleavage

6) Phosphorylation

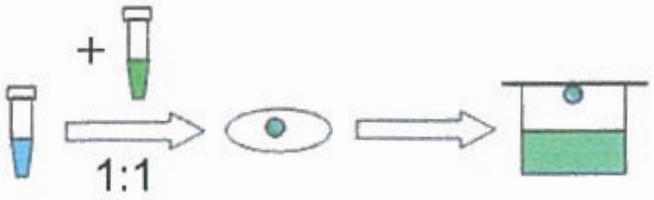
7) Membrane proteins



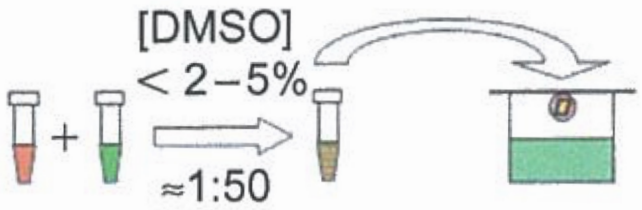
Preparation of Crystals of Protein-Ligand Complexes

- Reagents

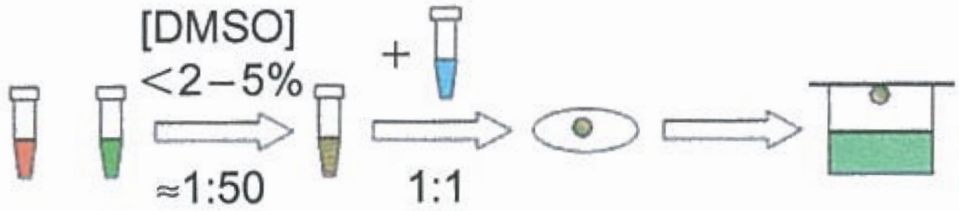
- Crystallization, standard protocol



- Soaking experiment

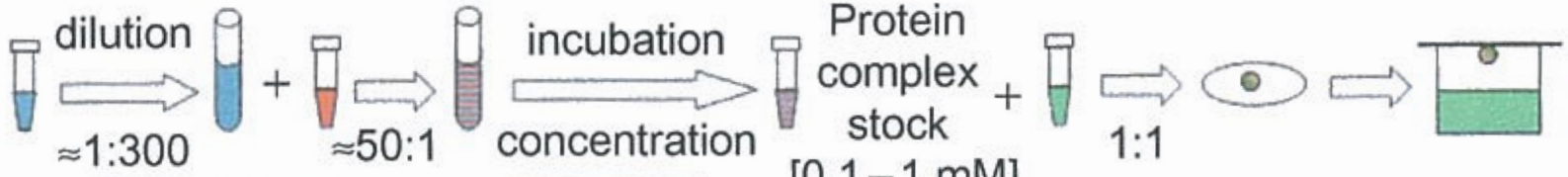


- Co-crystallization, standard protocol



Molar excess of ligand
≈ 1.5 to 5-fold

- Co-crystallization, "ligand fishing" protocol



Molar excess of ligand
≈ 450 to 1,500-fold

X-ray Diffraction Experiment

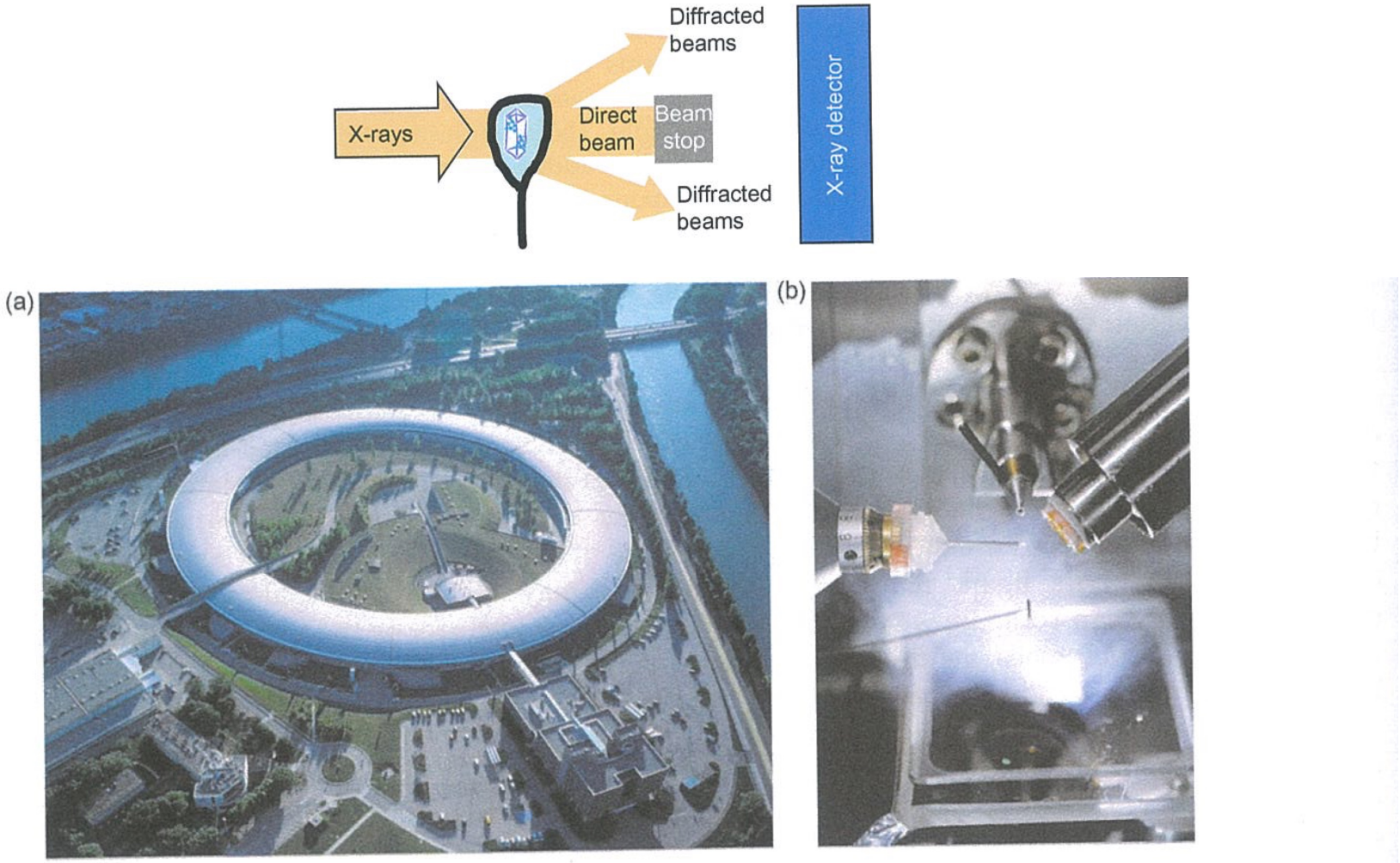
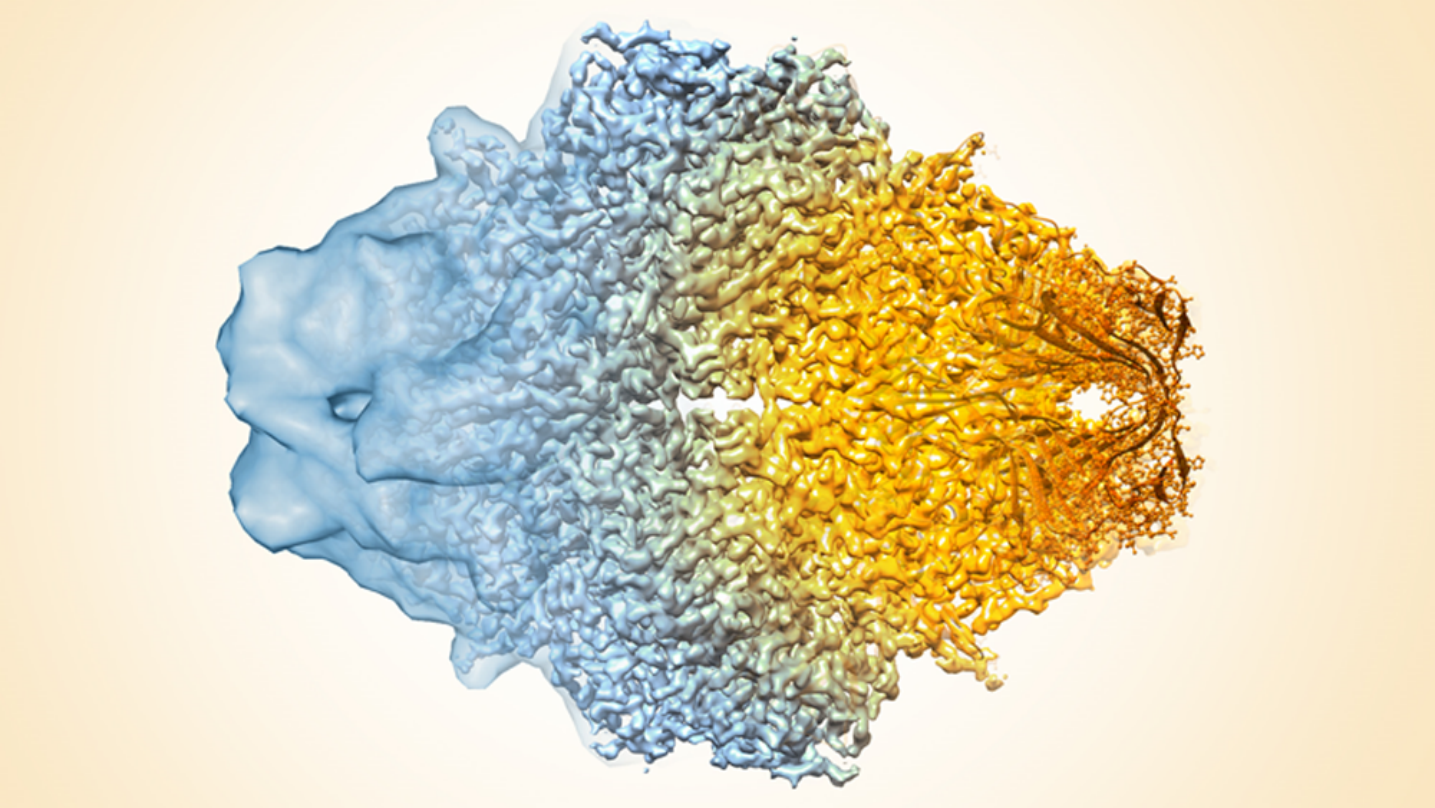


FIGURE 22.8 (a) Aerial view of the ESRF, located between the rivers Isère and Drac in Grenoble, France. Electrons circle around in a large ring inside the circular building, and when they pass bending magnets or assemblies of magnets called undulators they emit powerful X-rays. (b) Crystal being exposed, viewed from the position of the detector. X-rays emanating from the narrow steel tube in the back hit a frozen crystal in the cryo-loop in the center of the picture. The direct beam is stopped by a beam-stop, the small piece of metal just below the center of the picture. On the left is the goniometer, which is used to rotate the crystal, and from the nozzle on the right cold (100 k) nitrogen gas is blown over the crystal. *Pictures courtesy of ESRF/Morel.*

Cryo-Electron Microscopy



This composite image of the protein β-galactosidase shows how cryo-EM has progressed over the years, from the indistinct blobs once obtained with the technique (left) to the nearly 2-Å resolution structures possible today (right).

Berg E. G. *ACS Cent. Sci.* **2015**, *1*, 283–285.

Summary

The “picture” of the ligand-receptor complex is obtainable through pharmacore identification, protein crystallography, and cryo-electron microscopy. However, a technique that can capture the dynamic nature of the ligand-receptor interaction remains elusive.

Next Lecture, 2016/05/02

Pharmacokinetic Properties