BS318-12 Protein Targeting

21/22

Department
Life Sciences
Level
Undergraduate Level 3
Module leader
Lorenzo Frigerio
Credit value
12
Module duration
10 weeks
Assessment
100% exam
Study location
University of Warwick main campus, Coventry

Description

Introductory description

This module allows the final year students, who have a substantial background in molecular and cell biology from a number of first and second year modules, to apply this knowledge to a research area (protein targeting) which is a field of fundamental importance in cell biology.

Module web page

Module aims

During the module, the students should gain an appreciation of the molecular nature of the targeting signals and the appropriate transport apparatus, an appreciation of the specific protein-protein interactions required at each step of a given transport pathway, and of the mechanisms by which large globular proteins are translocated across membrane bilayers which, in several cases, are impermeable even to protons. Specifically, they should become familiar with the evidence and up to date models for protein transport into the ER lumen, mitochondria and chloroplasts and for transport from the plasma membrane to lysosomes (by endocytosis) and from the ER to the cell surface via the Golgi (by exocytosis). They should also become aware of the experimental approaches used to study protein targeting and translocation across membranes.

Outline syllabus

This is an indicative module outline only to give an indication of the sort of topics that may be covered. Actual sessions held may differ.

LECTURES 1-8

Secretion

Movement of proteins through cellular membranes

What is protein targeting? The topological relationships of membrane-bound orgenelles and the proposed evolutionary pathway for these orgenelles. Introduction to the concept of sorting signals, their receptors and types of transport across membranes. Approaches to studying targeting signals, mechanisms of membrane translocation and protein sorting.

The secretory pathway

(a) Import of proteins into the ER lumen

Discovery of signal sequences and co-translational protein transport. The signal hypothesis. Discovery of the mammalian signal recognition particle, its receptor and homologues in bacteria. Segregation of membrane proteins. The components of the translocation apparatus and the nature and mechanism of membrane transfer. Methods used to study translocation of proteins into the ER lumen. Glycosylation of proteins.

(b) Protein transport through the secretory pathway

ER quality control: the calnexin cycle and retrotranslocation as aspects of ER–associated protein degradation (ERAD). The concept of a default pathway for secretion. Retrieval of ER resident proteins via retrieval signals and circulating membrane receptors. Cargo selection. Cell-free reconstituted transport systems and the identification of non-clathrin coated vesicles involved in protein trafficking. COPs, ARF and other proteins in cargo selection and vesicle budding; NSF, SNAPs and SNAREs in vesicle targeting and membrane fusion events. The SNARE hypothesis and involvement of regulatory Rab proteins in vesicle targeting. The controversy of Golgi structure and function. Sorting at the TGN, constitutive and regulated secretion.

LECTURES 9-14

Protein transport into mitochondria and chloroplasts

(a) Basics of chloroplasts and mitochondrial protein import

Structures of the organelles. Evidence for post-translational modes of protein import. Techniques used for the in vitro analysis of protein import into chloroplasts and mitochondria. Relationship to bacterial protein export.

(b) Chloroplast protein import

Structures of chloroplast targeting signals. Translocation across the envelope membranes; structure of the translocation apparatus. Energetics of translocation. Two-step import pathway for thylakoid lumen proteins. Structures of thylakoid transfer signals. Evidence for distinct translocation pathways for thylakoid lumen proteins. Insertion of membrane proteins. Evolution of targeting pathways: relation to bacterial protein transport systems.

(c) Mitochondrial protein import

Mitochondrial targeting signals. The default pathway for mitochondrial protein targeting. Energetics and mechanism of translocation. Sorting of mitochondrial proteins: outer membrane proteins, the controversy over the import of intermembrane space proteins, and the assembly of proteins in the inner membrane.

LECTURES 15-20

Receptor-mediated endocytosis

Receptor-mediated endocytosis (RME). Reasons for endocytosis. General description of RME including definition of receptors and ligands. Biosynthesis of receptors and transport to cell surface. Recycling of receptors and fate of ligands. Clathrin, coated pits and coated vesicles. Retrograde transport and protein retrieval.

Specific examples of RME will be covered:

1. RME responsible for delivering nutrients into cells, typified by low density lipoprotein and transferrin.
2. RME involved in effector function, typified by insulin and epidermal growth factor.
3. Role of clathrin-coated vesicles.
4. Transcytosis - the transfer of specific substances through polarized cells e.g. polymeric IgA or IgM molecules.
   RME involved in clearance of unwanted material from intracellular space e.g. asialoglycoproteins.
5. How unwelcome opportunists take advantage of the RME system to enter cells.

   Viruses e.g. VSF, influenza virus.
   Toxins e.g. diphtheria toxin, cholera toxin, ricin.
6. Retrograde transport from the trans-Golgi network.
7. Clathrin-independent mechanisms of endocytosis.

Learning outcomes

By the end of the module, students should be able to:

- LO1 Understand the control and mechanisms that control targeting to the ER and secretion from the ER through the Golgi apparatus
- LO2 Understand the control and mechanisms that control targeting to mitochondria and chloroplast
- LO3 Understand the control and mechanisms that control receptor mediated endocytosis
- LO4 Critical understanding of core research techniques used in the study of protein targeting and sorting

Indicative reading list

Lodish, H. et al. Molecular Cell Biology (any edition).

Alberts, B. et al. Molecular Biology of the Cell, 5th edn.
(Garland Science, 2007). Chapters 12 and 13.

Dalby, R. E. and von Heijne, G. Protein Targeting, Transport and Translocation. (Academic Press, 2002). This book should not be purchased as it is expensive. Copies are available in the Central Campus Library.

Reference material will be provided during the module.

Subject specific skills

You will gain an appreciation of the molecular nature of the targeting signals and the appropriate transport apparatus, an appreciation of the specific protein-protein interactions required at each step of a given transport pathway, and of the mechanisms by which large globular proteins are translocated across membrane bilayers which, in several cases, are impermeable even to protons. You will also learn, in a journey through the key experiments, about how this field of research developed from a simple hypothesis to a major area of cell biology.

Transferable skills

1. Critical appraisal of research papers
2. Self directed learning
3. Adult learning

Study

Study time

Type	Required
Lectures	20 sessions of 1 hour (17%)
Private study	100 hours (83%)
Total	120 hours

Private study description

Independent learning, self directed learning and revision for final year exams.

Costs

No further costs have been identified for this module.

Assessment

You must pass all assessment components to pass the module.

Students can register for this module without taking any assessment.

Assessment group B1

	Weighting	Study time
Written Examination	100%	

1.5 hour examination (April): 2 essay- style questions from a choice of 4.

Feedback on assessment

Pastoral meetings with personal tutor

Past exam papers for BS318

Availability

Courses

This module is Core for:

- Year 3 of UBSA-3 Undergraduate Biological Sciences

This module is Option list A for:

- UBSA-3 Undergraduate Biological Sciences
  - Year 3 of C100 Biological Sciences
  - Year 3 of C100 Biological Sciences
  - Year 3 of C105 Biological Sciences with Molecular Genetics
  - Year 3 of C107 Biological Sciences with Virology
- Year 3 of ULFA-C1A1 Undergraduate Biological Sciences (MBio)
- UBSA-C1B9 Undergraduate Biomedical Science
  - Year 3 of C1B9 Biomedical Science
  - Year 3 of C1B9 Biomedical Science
  - Year 3 of C1B9 Biomedical Science
- ULFA-C1A3 Undergraduate Biomedical Science (MBio)
  - Year 3 of C1A3 Biomedical Science
  - Year 3 of C1B9 Biomedical Science

This module is Option list B for:

- UBSA-3 Undergraduate Biological Sciences
    - Year 3 of C103 Biological Sciences with Environmental Resources
    - Year 3 of C104 Biological Sciences with Microbiology