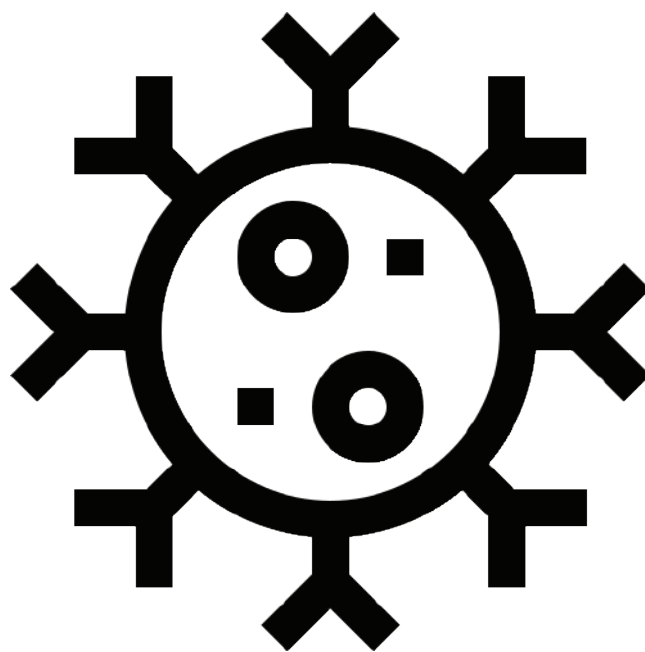


CELL COMMUNICATION AND CELL SIGNALING



Dr. KAILASH CHOUDHARY
DR. RAM PRAKASH SARAN

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Preface

Cell communication, immunology and cancer biology are demanding subjects to learn — not because any single idea in them is hard, but because the field is vast and moves quickly, and every year brings a fresh explosion of information. A student who tries to keep pace by memorising the facts as they stand today will be left behind tomorrow. Our aim in writing this book has therefore been a different one: to give the reader the underlying principles of how cells talk to one another, how the immune system turns that conversation into a defence, and what happens when the conversation breaks down and a cell becomes malignant — so that new advances can be understood and appreciated as they arrive, rather than merely recorded.

Cell Communication and Cell Signaling offers a thorough but approachable introduction to these principles. It assumes an introductory background in cell biology and biochemistry, and builds from there through a concept-based approach rather than a catalogue of facts. The fifteen chapters move deliberately from the general to the particular. The opening chapters establish the logic of signalling itself — signal molecules, receptors, second messengers, G-protein-coupled and enzyme-linked receptors and the pathways they command — before turning to signalling in bacteria and in plants. The middle chapters deal with how a cell physically engages its neighbours and its surroundings: cell junctions and adhesion molecules, the extracellular matrix, and the specialised communication of neurons and of the haematopoietic system. The chapters that follow apply all of this to the immune system — innate defences and the cells and organs that house them, antigens and antibodies, the MHC and antigen processing, lymphocyte activation, the effector mechanisms of humoral immunity, hypersensitivity and immunodeficiency, and the strategies pathogens use against the host. The last chapter turns to cancer, where every principle in the book reappears in the form of a growth signal that will not switch off.

Particular care has been taken with the topics students most often struggle with: the interaction and signalling between immune cells, the genes that cause cancer, and the adhesion proteins that tie a cell to its matrix. These are presented in a structured, diagrammatic form wherever a diagram will do the work of a paragraph. The language is deliberately simple, the page is uncluttered, and the outline drawings are meant to be clear rather than ornamental. To help the reader stay oriented, every chapter opens with a set of learning objectives; KEY POINT boxes mark the ideas that must be carried forward; DO YOU KNOW? boxes supply the historical and clinical context that makes a mechanism memorable; and a Rapid Revision summary closes the chapter for last-minute recall.

The book is written above all for students preparing for graduate and postgraduate examinations, and its purpose is as much to build confidence as to convey content. A reader who finishes it should be able to meet an unfamiliar pathway, an unfamiliar receptor or an unfamiliar tumour and reason about it from first principles.

Every effort has been made to keep this book free of error, but we are under no illusion that it is. Comments, criticism and suggestions from students and teachers alike will be received with gratitude.

Authors

Acknowledgement

Writing *Cell Communication and Cell Signaling* has been an incredible journey. Every chapter has taken its final shape only after endless inputs of time and effort, and although many colleagues assisted me in compiling this book, I would first of all like to thank all the students at the Institute for Advanced Studies (IFAS), who have helped me learn to present the concepts of cell signalling, immunology and cancer biology in their simplest form. Their questions decided what this book had to explain, and their difficulties decided how it had to be explained.

We would like to make special mention of the effort made by Er. Radheshyam Choudhary, CEO, IFAS Publications, who extended immense support in myriad ways and was an incessant force in bringing the book out in its present form.

Our special thanks to Dadasaheb Sondge and Vishal Bhujbal for reviewing the manuscript and helping me prepare its systematic layout; their comments were invaluable in improving the text, and I could not have completed it without their help. Further thanks to Digambar Jagtap for their work on the layout of the book, and to Lalji Kannoujiya, Pranshu Dwivedi, Rakesh Kumawat, and Lokesh for their creative suggestions and for reviewing the contents.

This book is a team effort, and producing it would have been impossible without the outstanding people of IFAS Publications. We are grateful to our life science teacher's suggestions on improving the quality of the content and for the regular reminders that kept the project to its schedule. Thanks are due to Deepak Nambani for turning our ideas into clear and beautiful diagrams — no small task in a book that depends so heavily on them. Special thanks to Vikendra Metha for the design of the cover page, Kuldeep Singh for the typesetting, and to all of them for the care they took during copy-editing and production.

We are also thankful to my family for their patience and encouragement through the long months this book occupied, and to the teachers whose own books first taught me this subject.

And finally, our humble greetings to all those who put in significant effort and remain unmentioned.

Dr. Kailash Choudhary

Dr. Ram Prakash Saran

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CHAPTER

1

Principles of Cell Signaling

LEARNING OBJECTIVES

- 1.1 Appreciate why every cell must receive and interpret signals from its surroundings.
- 1.2 Distinguish the modes of cell–cell signaling: contact-dependent, endocrine, paracrine and autocrine.
- 1.3 Classify the major signaling molecules – steroids, gases, neurotransmitters, and plant hormones.
- 1.4 Relate the chemical nature of a ligand (hydrophilic vs hydrophobic) to the location of its receptor.
- 1.5 Outline the three stages of signaling: reception, transduction and response.
- 1.6 Identify the core components of a signal-transduction pathway and the role of second messengers.
- 1.7 Explain how signal amplification converts a few ligand molecules into a large cellular response.

1.1 Why Cells Signal

All cells, from the simplest bacterium to the most specialised neuron, must continuously receive and respond to signals from their surroundings. Even free-living bacteria sense and swim towards high concentrations of nutrients such as glucose or amino acids. Many unicellular eukaryotes likewise respond to signaling molecules secreted by other cells, allowing rudimentary cell-to-cell communication — the mating of yeast cells, for instance, is triggered by peptides secreted by one cell that bind to receptors on the surface of another.

In multicellular organisms this conversation between cells reaches extraordinary sophistication. A vast array of signaling molecules is secreted by, or displayed on the surface of, one cell and recognised by receptors on other cells, thereby integrating and coordinating the activities of the billions of cells that make up a complex animal. The binding of a signaling molecule to its receptor initiates a chain of intracellular reactions that ultimately regulate virtually every aspect of cell behaviour — metabolism, movement, proliferation, survival and differentiation.

KEY POINT

A cell can only respond to a signal if it expresses the matching receptor. The same signaling molecule may therefore produce entirely different responses in different target cells, depending on the receptors and downstream machinery each cell possesses.

1.2 Modes of Cell–Cell Signaling

Cell signaling may arise either from the direct interaction of a cell with its neighbour (or with the extracellular matrix) or from the action of secreted signaling molecules that travel some distance before reaching their target.

1.2.1 Signaling by direct cell–cell and cell–matrix contact

Contact-dependent signaling plays a critical role in regulating the behaviour of cells in animal tissues. Adhesion proteins such as integrins and cadherins act not only as mechanical anchors but also as signaling molecules that regulate cell proliferation and survival in response to cell–cell and cell–matrix contacts. In addition, cells display a variety of surface receptors that interact with membrane-bound signaling molecules on adjacent cells (Fig 1.1a). Such direct signaling is central to the many interactions between different cell types during embryonic development and to the maintenance of adult tissues.

1.2.2 Signaling by secreted molecules

Signaling by secreted molecules is conventionally divided into three categories, based on the distance over which the signal travels (Fig 1.1).

- **Endocrine signaling:** the signaling molecules — hormones — are secreted by specialised endocrine cells and carried through the circulation to act on target cells at distant body sites. The steroid hormone estrogen, produced by the ovary and acting throughout the female reproductive system, is a classic example. More than fifty hormones are produced by endocrine glands such as the pituitary, thyroid, parathyroid, pancreas, adrenal glands and gonads.
- **Paracrine signaling:** a molecule released by one cell acts on neighbouring target cells nearby. The action of neurotransmitters carrying a signal across the synaptic cleft between two nerve cells is a familiar example.
- **Synaptic signaling:** It is a specialized form of cell communication in which a neuron transmits information to a target cell across a synapse. When an electrical impulse (action potential) reaches the axon terminal of a neuron, it triggers the release of neurotransmitters into the synaptic cleft. These chemical messengers diffuse across the small gap and bind to specific receptors on the postsynaptic (target) cell membrane. This binding generates a response in the target cell, which may be excitatory or inhibitory. Synaptic signalling is highly rapid, precise, and localized, allowing neurons to communicate efficiently within the nervous system.
- **Autocrine signaling:** a cell responds to a signaling molecule that it itself produces. Activated T lymphocytes, for instance, secrete a growth factor that drives their own proliferation, amplifying the immune response. Significantly, abnormal autocrine signaling frequently contributes to the uncontrolled growth of cancer cells, in which a tumour cell makes a growth factor to which it also responds, continuously driving its own unregulated division.

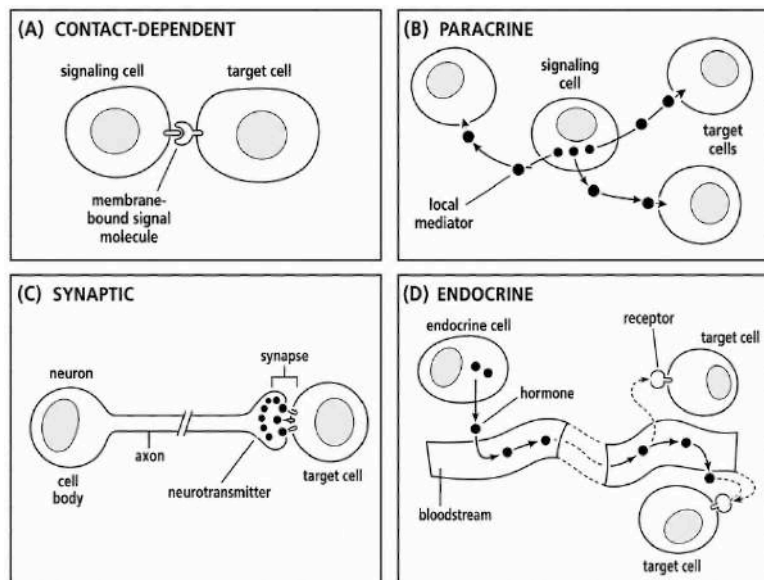


Figure 1.1 *Cell to Cell Signaling.* (A) *Contact-dependent: Direct cell–cell contact where membrane-bound ligands bind receptors on adjacent target cell.* (B) *Paracrine: Local signaling in which mediators diffuse short distances to act on nearby target cells.* (C) *Synaptic: Neuronal signaling where neurotransmitters are released across synapse to specific target cell.* (D) *Endocrine: Hormonal signaling where hormones are released into bloodstream to act on distant target cells.*

DO YOU KNOW?

A single endocrine cell may influence target cells located more than a metre away, while a synaptic signal such as a neurotransmitter acts across a synaptic gap of only ~20 nanometres — a difference in working range of nearly 50 million-fold, achieved with the same basic ligand–receptor logic.

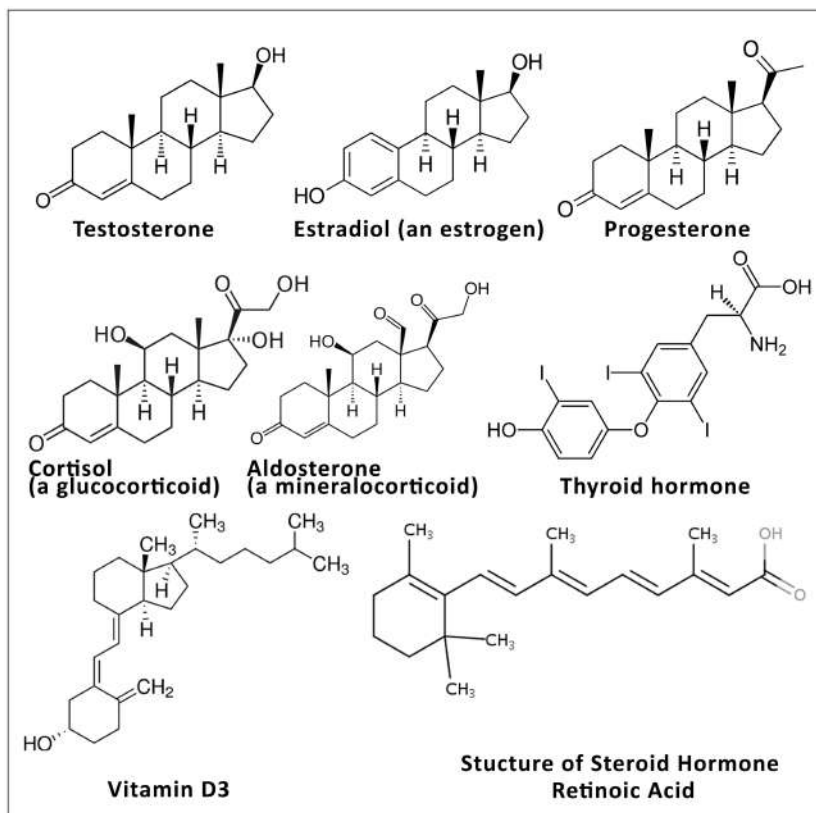
1.3 The Major Classes of Signaling Molecules

Signaling molecules are chemically diverse. Their solubility — whether they dissolve in water or in lipid — determines whether they act at the cell surface or can enter the cell, and therefore dictates the kind of receptor they use.

1.3.1 Steroid hormones

The steroid hormones — testosterone, estrogen, progesterone, the corticosteroids and the insect hormone ecdysone — are all synthesised from cholesterol (Figure 1.2). Testosterone, estrogen and progesterone are the sex steroids, produced by the gonads. The corticosteroids are made by the adrenal gland and include the glucocorticoids, which stimulate glucose production, and the mineralocorticoids, which act on the kidney to regulate salt and water balance. Ecdysone triggers the metamorphosis of insect larvae into adults.

Figure 1.2 Structures of representative steroid and steroid-related hormones synthesised from cholesterol — the sex steroids, glucocorticoids and mineralocorticoids, together with thyroid hormone, vitamin D₃ and retinoic acid.



Steroid hormone (glucocorticoids), Thyroid hormone, vitamin D₃ and retinoic acid are structurally and functionally distinct from the steroids, yet they share a common mechanism of action with them inside the target cell (Figure 1.3). Thyroid hormone, synthesised from tyrosine in the thyroid gland, governs development and metabolism; vitamin D₃ regulates Ca²⁺ metabolism and bone growth; and retinoic acid and related retinoids, derived from vitamin A, play key roles in vertebrate development. Because all of these molecules are lipid-soluble, they cross the plasma membrane and act on intracellular receptors that regulate gene transcription.

KEY POINT

Lipid-soluble signals (steroids, thyroid hormone, vitamin D₃, retinoic acid) diffuse across the membrane and bind intracellular receptors that act as transcription factors. Water-soluble signals cannot cross the membrane and must act through cell-surface receptors.

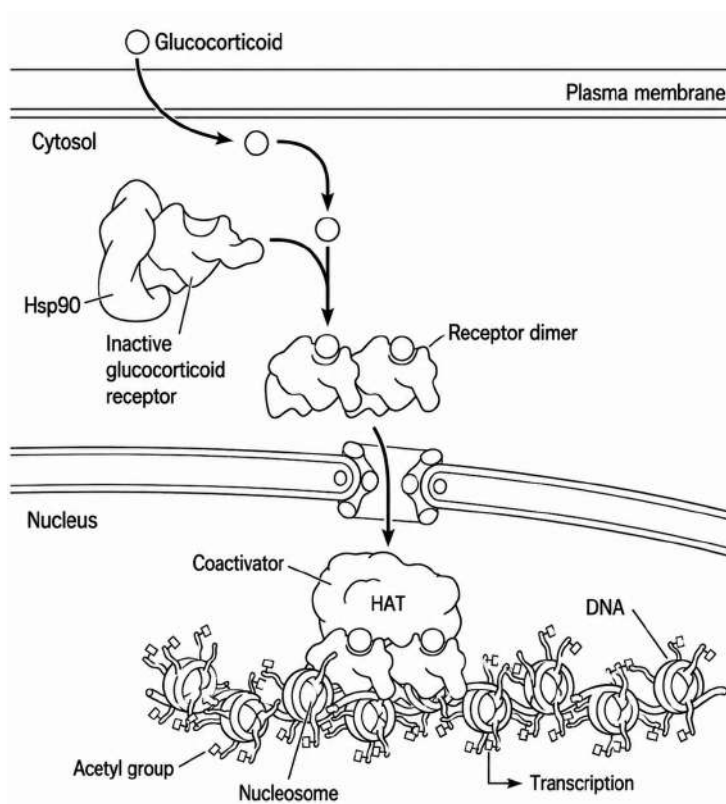


Figure 1.3 Steroid (glucocorticoid) hormone diffuses into the cell and binds its receptor, causing receptor activation and dimerization. The complex enters the nucleus and regulates gene transcription by modifying chromatin structure via coactivators like HAT.

1.3.2 Nitric oxide and carbon monoxide

The simple gas nitric oxide (NO) is a major paracrine signaling molecule in the nervous, immune and circulatory systems. Like the steroid hormones, NO diffuses directly across the plasma membrane of its target cells — but its mechanism is quite different: rather than regulating transcription, it alters the activity of intracellular target enzymes. NO is synthesised from the amino acid arginine by the enzyme nitric oxide synthase (Figure 1.4).

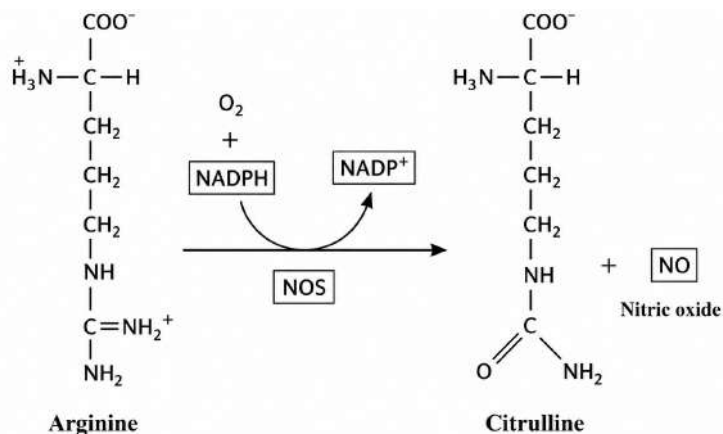


Figure 1.4 NO is synthesized from the amino acid arginine by the enzyme nitric oxide synthase

Once formed, NO diffuses out of the cell and acts only locally, because it is extremely unstable, with a half-life of just a few seconds. A well-characterised example is the dilation of blood vessels (Figure 1.5). Neurotransmitters such as acetylcholine, released from nerve terminals in the vessel wall, stimulate endothelial cells to synthesise NO. The NO then diffuses to neighbouring smooth-muscle cells, where it reacts with iron at the active site of guanylyl cyclase, increasing the enzyme's activity and raising the level of the second messenger cyclic GMP. The rise in cGMP causes the muscle to relax and the vessel to dilate. The same pathway underlies the action of nitroglycerin in heart disease — the drug is converted to NO, which dilates the coronary vessels and increases blood flow to the heart.

A second gas, carbon monoxide (CO), acts similarly in the nervous system as a neurotransmitter and mediator of vasodilation. Its synthesis in brain cells, like that of NO, is stimulated by neurotransmitters, and it too can activate guanylyl cyclase, which appears to be its principal physiological target.

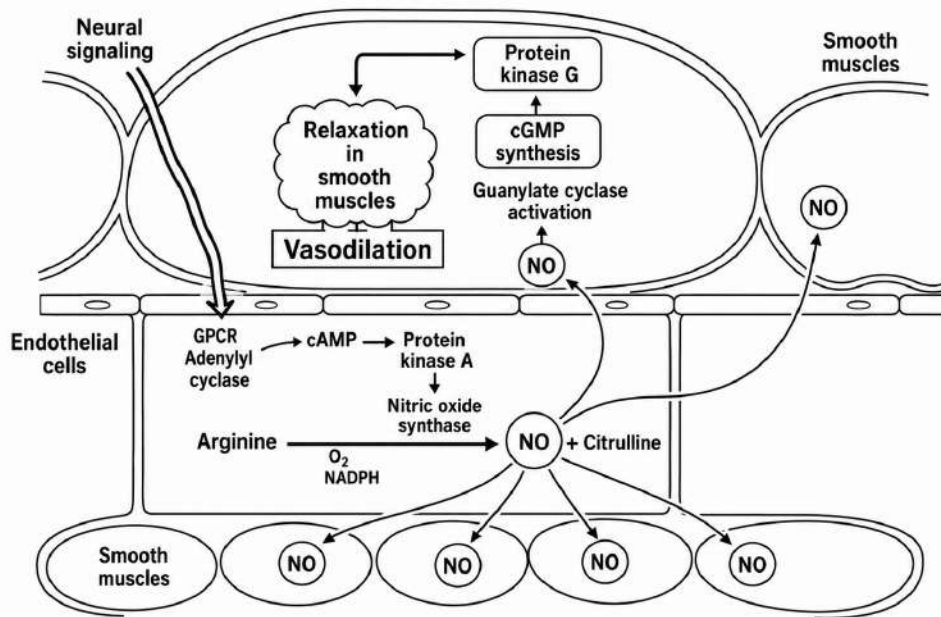


Figure 1.5 Nitric oxide signaling and smooth-muscle relaxation. Acetylcholine triggers NO synthesis in endothelial cells; NO diffuses to smooth-muscle cells, activates guanylyl cyclase, raises cGMP and causes vasodilation.

DO YOU KNOW?

The 1998 Nobel Prize in Physiology or Medicine was awarded for the discovery that nitric oxide — a toxic, unstable gas and a component of car exhaust — serves as a signaling molecule in the cardiovascular system. This insight also explains how a century-old angina remedy, nitroglycerin, actually works.

1.3.3 Neurotransmitters

Neurotransmitters carry signals between neurons, or from neurons to other target cells such as muscle. They are a diverse group of small, hydrophilic molecules including acetylcholine, dopamine, epinephrine (adrenaline), serotonin, histamine, glutamate, glycine and γ -aminobutyric acid (GABA) (Figure 1.6). Their release is triggered by the arrival of an action potential at a nerve terminal, after which they diffuse across the synaptic cleft and bind receptors on the target-cell surface. Some neurotransmitters double as hormones — epinephrine, for example, acts both as a neurotransmitter and as an adrenal hormone that signals glycogen breakdown in muscle.

Because they are hydrophilic, neurotransmitters cannot cross the membrane and therefore act exclusively through cell-surface receptors. Many of these are ligand-gated ion channels, such as the acetylcholine receptor, in which neurotransmitter binding opens the channel and directly changes ion flux. Others are coupled to G proteins, which in turn regulate ion-channel activity indirectly.

CHAPTER

4

Enzyme-Linked Receptors & Pathways

LEARNING OBJECTIVES

- 4.1 Describe RTK structure, dimerization and autophosphorylation.
- 4.2 Trace the Ras–Raf–MEK–ERK MAP-kinase pathway.
- 4.3 Explain the PI3K–Akt survival pathway and insulin signaling.
- 4.4 Relate RTK/Ras defects to cancer and to achondroplasia.
- 4.5 Outline cytokine receptors and JAK–STAT signaling.
- 4.6 Survey TGF- β /Smad, phosphatase and guanylyl-cyclase receptors.
- 4.7 Describe Wnt, Notch and Hedgehog developmental pathways.

4.1 Receptor Tyrosine Kinases (RTKs)

Unlike the G-protein-coupled receptors, many cell-surface receptors are directly linked to intracellular enzymes. The largest family of such enzyme-linked receptors are the receptor protein-tyrosine kinases (RTKs), which phosphorylate their substrate proteins on tyrosine residues. This family includes the receptors for most polypeptide growth factors — EGF, NGF, PDGF, insulin and many others — so tyrosine phosphorylation has been intensively studied as a mechanism controlling animal cell growth and differentiation. More than fifty RTKs are known, all sharing a common organisation: an N-terminal extracellular ligand-binding domain, a single transmembrane α -helix, and a cytosolic C-terminal domain with protein-tyrosine kinase activity. Most are single polypeptides, although the insulin receptor and its relatives are dimers of two pairs of chains (Figure 4.1).

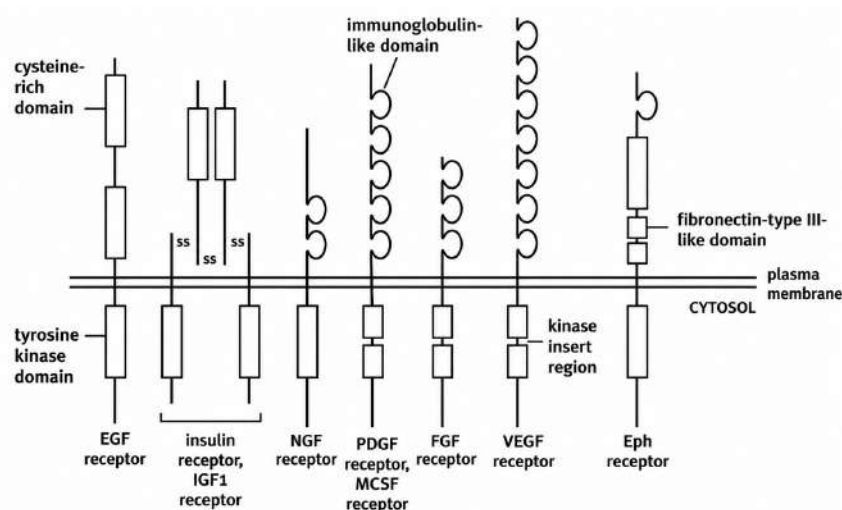


Figure 4.1 Architecture of representative single-pass receptor tyrosine kinases — the EGF, insulin and PDGF receptors.

4.1.1 Dimerization and autophosphorylation

The first step in signaling from most RTKs is ligand-induced receptor dimerization. Some growth factors (PDGF, NGF) are themselves dimers and directly cross-link two receptors; others (EGF) are monomers with two receptor-binding sites that bridge two receptors. Dimerization brings the two cytosolic kinase domains together so they cross-phosphorylate one another (autophosphorylation). This serves two roles: phosphorylation of tyrosines within the catalytic domain increases kinase activity, while phosphorylation of tyrosines outside the catalytic domain creates specific docking sites for downstream signaling proteins (Figure 4.2).

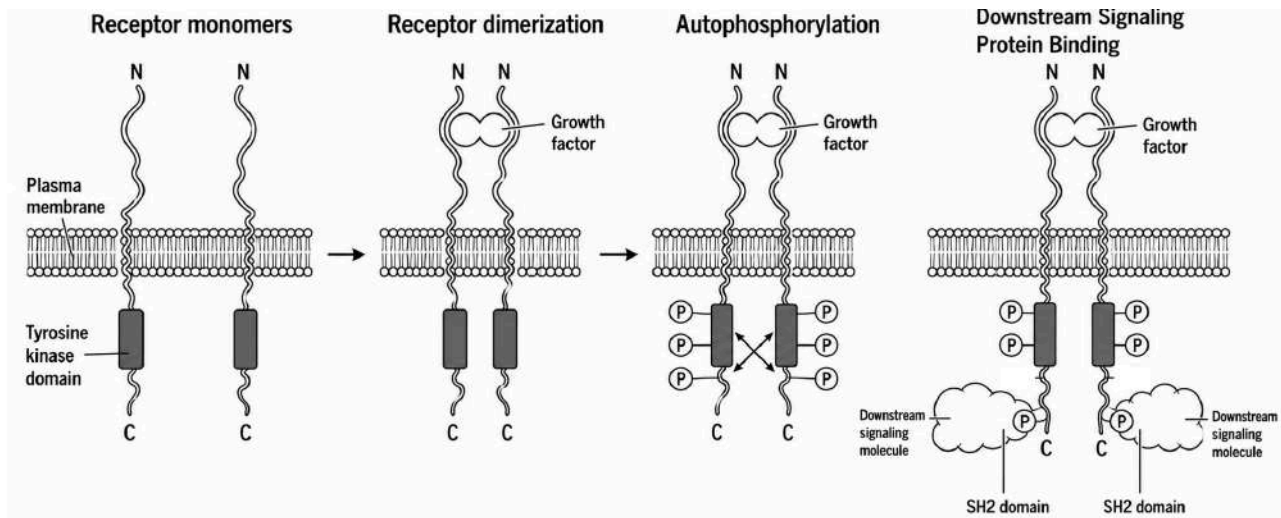


Figure 4.2 Ligand-induced dimerization and autophosphorylation of a receptor tyrosine kinase.

4.1.2 SH2 and PTB docking domains

Downstream proteins associate with the activated RTK through domains that recognise phosphotyrosine-containing peptides. The best-characterised are the SH2 domains (Src-homology 2), first recognised in kinases related to Src, the oncogenic protein of Rous sarcoma virus. An SH2 domain of about a hundred amino acids binds a short phosphotyrosine peptide; this association localises the SH2 protein to the membrane, brings it together with other proteins, promotes its phosphorylation and stimulates its activity. Other proteins dock through PTB (phosphotyrosine-binding) domains. The binding of SH2- or PTB-containing proteins to autophosphorylated receptors is the first step in transmitting the growth-factor signal into the cell (Figure 4.3). RTKs can activate the Ras–MAP kinase pathway, the DAG/IP₃ (Ca²⁺) pathway and the PIP₃ pathway (Figure 4.4).

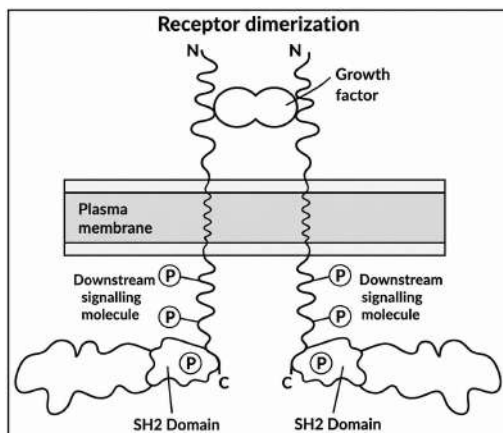


Figure 4.3 SH2-domain proteins docking onto the phosphotyrosines of an activated, dimerised receptor.

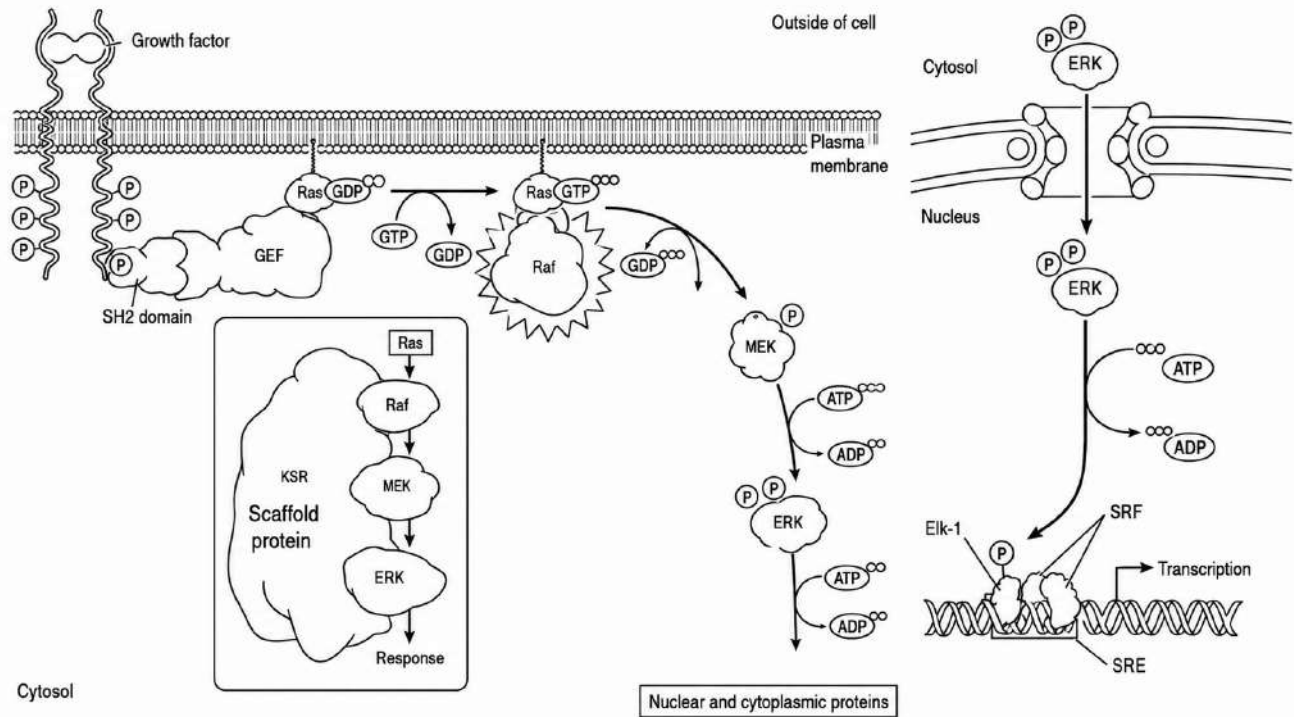


Figure 4.4 Association of downstream signaling molecules with the autophosphorylated receptor

4.2 Ras, Raf and the MAP Kinase Pathway

The MAP kinase pathway is an evolutionarily conserved cascade of protein kinases central to signal transduction in all eukaryotes, from yeast to humans. Its core elements are the MAP kinases (mitogen-activated protein kinases), serine/threonine kinases activated by growth factors and other signals. In yeasts they control mating, cell shape and sporulation; in higher eukaryotes they are ubiquitous regulators of growth and differentiation. The best-characterised mammalian MAP kinases are the ERKs (extracellular signal-regulated kinases), central to growth-factor-induced proliferation through both RTKs and GPCRs; protein kinase C, and the Ca^{2+} and cAMP pathways, also intersect with ERK signaling.

ERK is activated by two upstream kinases coupled to growth-factor receptors through the GTP-binding protein Ras. Active Ras activates the Raf serine/threonine kinase, which phosphorylates and activates MEK (MAP-kinase/ERK kinase); MEK is a dual-specificity kinase that activates ERK by phosphorylating both a threonine and a tyrosine (e.g. Thr-183 and Tyr-185 of ERK2). Activated ERK then phosphorylates many targets, including other kinases and transcription factors.

4.2.1 The Ras switch

Ras proteins are guanine-nucleotide-binding proteins that work like the α subunits of G proteins, cycling between inactive GDP-bound and active GTP-bound forms — but Ras acts as a monomer. Activation is driven by guanine-nucleotide-exchange factors that release GDP for GTP, and is terminated by GTP hydrolysis, accelerated by GTPase-activating proteins (GAPs) (Figure 4.5). Crucially, the ras mutations found in human cancers inhibit GTP hydrolysis, so the mutant Ras stays locked in the active GTP-bound form and drives unregulated proliferation even without growth factor. Ras was first identified as the oncogene of rat sarcoma viruses, and in 1982 ras mutations were directly implicated in human cancer; microinjecting active Ras induces proliferation of normal cells, while blocking Ras prevents growth-factor-induced proliferation.

7.7 Dystrophin/Dystroglycan and the Glycocalyx

A third type of ECM attachment matters in human disease. The costamere is an attachment structure at the surface of striated muscle, aligned with the Z-discs, that physically links myofibrils to the sarcolemma. Costameres contain many focal-contact proteins (β 1-integrin, vinculin, talin, α -actinin) and the intermediate-filament protein desmin, plus a specialised complex built around the large protein dystrophin.

Mutations in dystrophin cause the commonest muscular dystrophy, Duchenne muscular dystrophy (DMD) — a severe, recessive, X-linked disease affecting 1 in 3500 males, with progressive muscle degeneration leading to loss of walking and death.

Dystrophin links to a complex including the membrane protein dystroglycan and the sarcoglycan–sarcolemma complex, plus cytosolic dystrobrevins and syntrophins; dystroglycan binds laminin through the laminin G domain, anchoring the complex to the ECM. Loss of other components of the complex also causes muscular dystrophy (Figure 7.12).

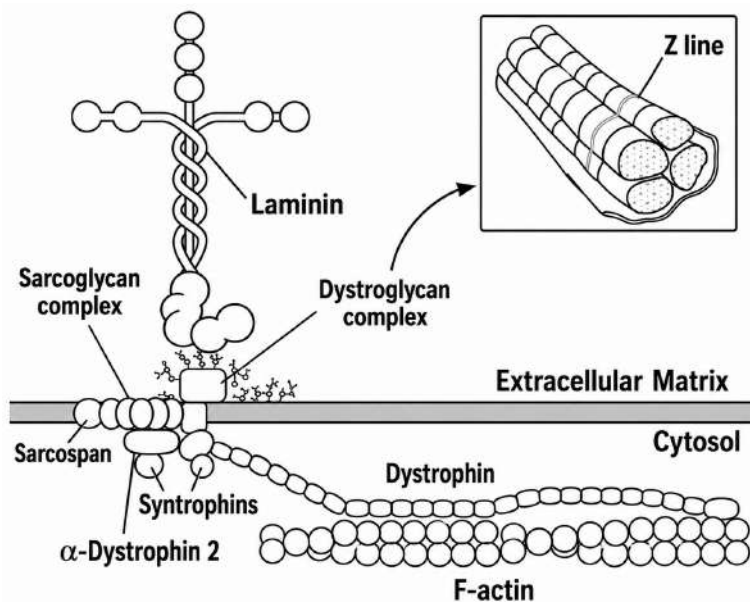


Figure 7.12 The dystrophin/dystroglycan complex linking the muscle-cell cytoskeleton across the membrane to laminin in the extracellular matrix.

7.7.1 The glycocalyx

Although animal cells lack a rigid cell wall, a carbohydrate-rich zone called the glycocalyx often surrounds the plasma membrane. Its roles include cell recognition and adhesion, protection of the cell surface and the creation of permeability barriers; the outer coat of amoebae and the jelly coats around animal eggs are specialized examples.

DO YOU KNOW?

Duchenne muscular dystrophy strikes about 1 in 3500 boys because the dystrophin gene sits on the X chromosome — the largest known human gene. Dystrophin works like a molecular shock-absorber, tethering the muscle cytoskeleton through dystroglycan to laminin in the matrix; without it, repeated contraction tears the membrane.

CHAPTER

8

Neurotransmission & Haematopoiesis

LEARNING OBJECTIVES

- 8.1 Describe chemical synaptic transmission.
- 8.2 Explain how neurotransmission is regulated and terminated.
- 8.3 Outline haematopoiesis, its sites and stem cells.
- 8.4 Describe the growth factors and signaling that regulate haematopoiesis.
- 8.5 Explain erythropoiesis and its stages.
- 8.6 Describe leucopoiesis and B-cell development.

8.1 Neurotransmission

A neuron communicates with its target — another neuron, a muscle fibre or a gland — at a specialised contact called the synapse. At a chemical synapse the two cells are separated by a narrow synaptic cleft (~20 nm), and the signal is carried across it by a chemical messenger, the neurotransmitter. Neurotransmission is therefore a specialised, very fast form of paracrine signaling, and it follows the same reception–transduction–response logic seen throughout cell signaling.

8.1.1 The sequence of synaptic transmission

An action potential arriving at the presynaptic terminal depolarises the membrane and opens voltage-gated Ca^{2+} channels. The resulting influx of Ca^{2+} triggers the fusion of neurotransmitter-filled synaptic vesicles with the presynaptic membrane, releasing transmitter into the cleft by exocytosis.

The transmitter diffuses across the cleft and binds receptors on the postsynaptic membrane. Where the receptor is a ligand-gated ion channel (ionotropic), binding opens the channel directly and changes the postsynaptic membrane potential within milliseconds; where it is a G-protein-coupled (metabotropic) receptor, the response is slower and mediated by second messengers (Figure 8.1).

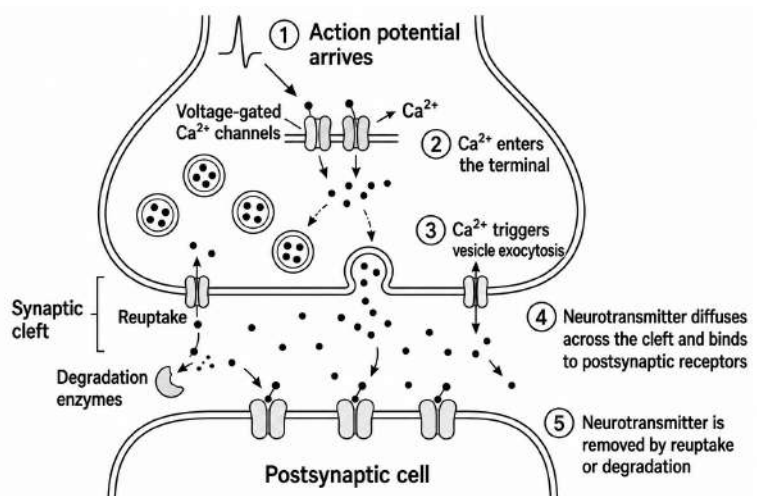


Figure 8.1 Chemical synaptic transmission: an action potential opens Ca^{2+} channels, Ca^{2+} triggers vesicle exocytosis, neurotransmitter crosses the cleft to postsynaptic receptors, and is then removed by reuptake or degradation

Excitatory transmitters produce a depolarising excitatory postsynaptic potential (EPSP), while inhibitory transmitters (such as GABA and glycine) produce a hyperpolarising inhibitory postsynaptic potential (IPSP); the neuron integrates these inputs to decide whether to fire.

The neuromuscular junction is a classic example: the motor neuron releases acetylcholine, which binds nicotinic acetylcholine receptors (ligand-gated channels) on the muscle fibre, depolarising it and triggering contraction.

KEY POINT

The trigger for neurotransmitter release is Ca^{2+} influx through voltage-gated channels at the presynaptic terminal. No Ca^{2+} entry, no vesicle fusion — this is why Ca^{2+} is the universal trigger for regulated exocytosis.

8.1.2 Regulation and termination

Because signalling must be brief and tunable, neurotransmission is tightly regulated. The transmitter is rapidly cleared from the cleft so that each impulse produces a discrete signal: acetylcholine is hydrolysed by acetylcholinesterase, while transmitters such as dopamine, serotonin, noradrenaline, glutamate and GABA are removed by specific re-uptake transporters on the presynaptic terminal and surrounding glia, after which they are recycled or degraded (for example by monoamine oxidase).

Presynaptic autoreceptors sense the transmitter the neuron has released and feed back to dampen further release, and the number and sensitivity of postsynaptic receptors can be up- or down-regulated over time. Many drugs and toxins act on these steps — acetylcholinesterase inhibitors prolong cholinergic signalling, re-uptake inhibitors raise synaptic monoamine levels, and botulinum toxin blocks acetylcholine release by cleaving the vesicle-fusion (SNARE) machinery.

DO YOU KNOW?

Acetylcholinesterase is one of the fastest enzymes known, hydrolysing acetylcholine almost as quickly as it diffuses to the enzyme. This near-instant clean-up of the cleft is what lets a neuromuscular synapse fire and reset hundreds of times a second.

8.2 Haematopoiesis: Sites and Process

Haematopoiesis is the formation of the blood's cellular components. In an adult human about 10^{11} – 10^{12} new blood cells are made every day to maintain steady-state numbers in the circulation. The cells from which all blood cells derive are the haematopoietic stem cells. In the embryo the main site is the yolk sac; in humans the principal sites are the bone marrow (femur and tibia in infants; pelvis, cranium, vertebrae and sternum in adults), liver, spleen and lymph nodes. In other vertebrates haematopoiesis occurs in the loose connective-tissue stroma of the gut, spleen, kidney or ovaries.

Table 8.1 Sites of haematopoiesis in humans

Stage	Sites
Fetus	0–2 months: yolk sac; 2–7 months: liver, spleen; 5–9 months: bone marrow
Infants	Bone marrow (most bones)
Adults	Vertebrae, ribs, sternum, skull, sacrum, pelvis, proximal femur

Pluripotent haematopoietic stem cells in the bone marrow can self-renew and give rise to all the separate blood-cell lineages. The stem cell is rare — perhaps 1 in 20 million nucleated marrow cells — and one stem cell can produce about 10^6 mature blood cells after roughly 20 divisions.

12.5.2 Maternal antibody and the neonate

Neonatal immunity: newborns are protected by maternal antibody — an example of passive immunity — acquired through the neonatal Fc receptor (FcRn). During pregnancy maternal IgG is actively transported across the placenta into the fetal circulation, and after birth ingested antibody in milk is carried across the gut epithelium by FcRn, giving the neonate the mother's IgG profile (Figure 12.6).

The same transport system can also do harm. If a Rhesus D-negative mother carries a RhD-positive fetus, fetal red cells entering her circulation at delivery can immunise her; in a later pregnancy the anti-D IgG she now makes crosses the placenta by exactly the FcRn route described above and destroys the fetal red cells, causing haemolytic disease of the newborn.

The preventive treatment is a neat piece of applied immunology: giving the mother preformed anti-D immunoglobulin at delivery clears the fetal cells before her own B cells can respond, and also engages the inhibitory FcγRIIB feedback described in Section 12.2.3.

Maternal IgG has one further consequence worth knowing: because it persists for several months, it can neutralise a live vaccine given too early, which is why measles vaccination is delayed until nine months of age or later.

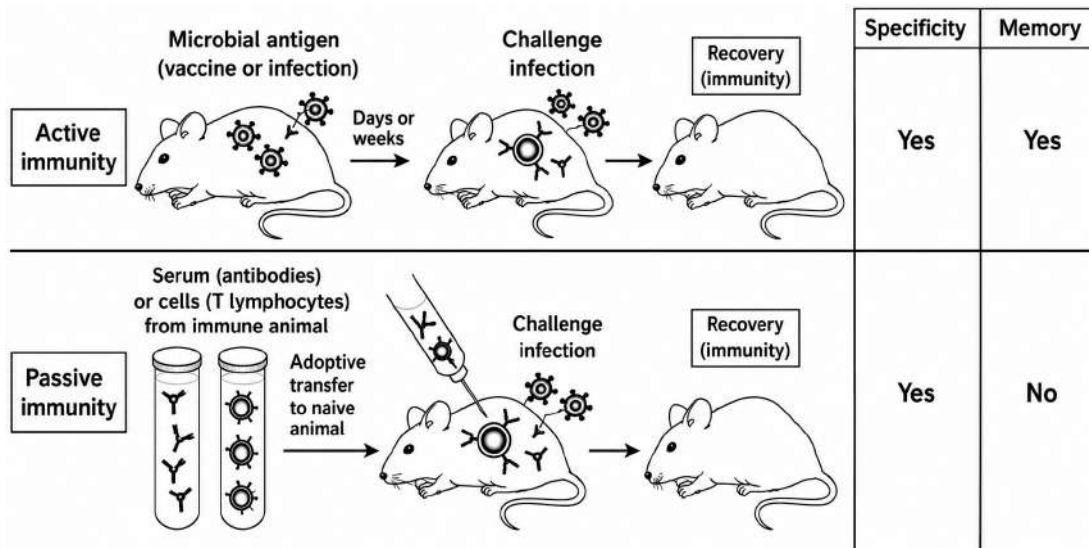


Figure 12.6 Active versus passive immunity. Active immunity is generated by the individual's own response to infection or vaccination: it takes days to weeks but leaves memory. Passive immunity is the transfer of preformed antibody or cells — maternal IgG, antiserum, or a therapeutic monoclonal — and is immediate, specific, but short-lived and leaves no memory.

KEY POINT

Passive immunity = transfer of preformed antibody (maternal IgG across the placenta via FcRn, secretory IgA in milk, or antiserum/anti-venom) — immediate but short-lived, no memory. Active immunity (infection or vaccine) is slower but lasting, with memory.

12.5.3 Microbial evasion of humoral immunity

Microbes evade antibody by antigenic variation — influenza, HIV (variable gp120) and rhinovirus mutate surface antigens, *E. coli* varies pilus antigens, and trypanosomes sequentially switch their surface glycoprotein, producing successive waves of parasitaemia each escaping the previous antibodies. Others inhibit complement or resist phagocytosis.

CHAPTER

13

Hypersensitivity, Immunodeficiency & Immunity to Microbes

LEARNING OBJECTIVES

- 13.1 Describe graft types, allorecognition and the tempo of rejection.
- 13.2 Explain how self-tolerance fails and what triggers autoimmune disease.
- 13.3 Classify hypersensitivity reactions I–IV and relate mechanism to clinical picture.
- 13.4 Describe primary and acquired immunodeficiencies, including HIV and AIDS.
- 13.5 Outline immunity and immune evasion in bacterial, fungal, viral and parasitic infection.
- 13.6 Use the pattern of infection to reason back to the defective component.

13.1 Transplant Immunology

Transplantation treats a failing organ or tissue with a healthy one: the donated tissue is the graft, the source the donor, and the host the recipient (orthotopic if placed in the normal anatomical site, heterotopic otherwise). Grafts are classified by genetic relationship: an autologous graft (autograft) moves tissue within one individual; a syngeneic graft is between genetically identical individuals; an allogeneic graft (allograft) is between genetically different members of a species; and a xenogeneic graft (xenograft) is between species. Foreign molecules recognised in an allograft are alloantigens (the responding cells and antibodies are alloreactive); in a xenograft they are xenoantigens (Figure 13.1).

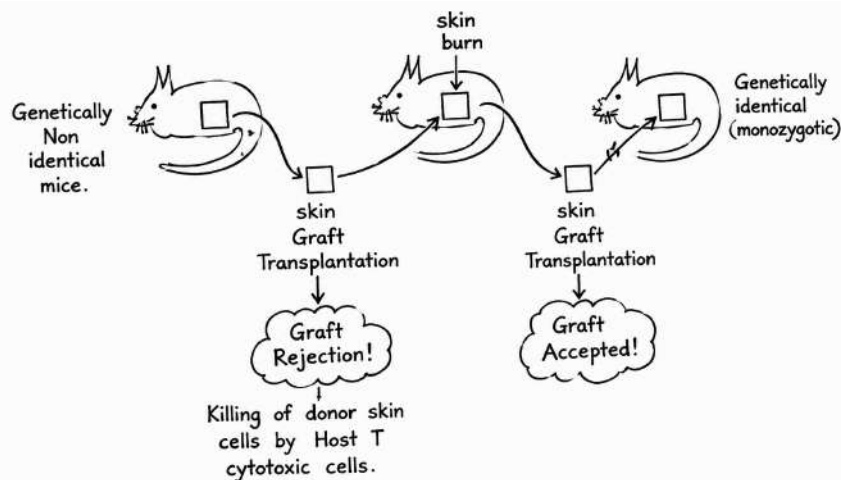


Figure 13.1 The rules of transplantation. A skin graft exchanged between genetically non-identical mice is destroyed by the host's cytotoxic T cells, while the same graft between genetically identical (syngeneic) animals is accepted. Rejection is an immunological reaction to foreign MHC, not a failure of surgery.

The molecules responsible for rejection are the MHC, and allogeneic MHC is one of the strongest immunogens known — as many as one to ten per cent of a person's T cells will respond to a single foreign MHC molecule, compared with roughly one in a hundred thousand for a conventional peptide antigen.

The reason lies in how T cells are selected. Because they are positively selected on self-MHC (Section 11.4.2), an allogeneic MHC molecule carrying its own peptides looks to a T cell very much like self-MHC carrying a foreign peptide, so allorecognition is in effect an immunological cross-reaction on a massive scale.

Alloantigen is presented in two ways: directly, when donor MHC on donor dendritic cells carried in with the graft engages host T cells without any host processing, and indirectly, when host antigen-presenting cells process graft alloantigen and present it conventionally. Direct presentation dominates early acute rejection; indirect presentation dominates chronic rejection. Class I alloantigen activates CD8⁺ cells and class II activates CD4⁺ cells, and memory T cells and B7 co-stimulation amplify both.

The classical rules of transplantation follow directly from this. Genetically identical individuals do not reject each other's tissue; genetically different individuals do. An F1 offspring accepts a graft from either inbred parent, because every MHC molecule the parent carries is also present in the offspring and is therefore seen as self — but each parent rejects a graft from the F1, because the offspring also carries the other parent's MHC, which is foreign to it. MHC genes are codominantly expressed, and this asymmetry is one of the neatest demonstrations of that fact.

13.1.1 The tempo of rejection

Rejection is classified by how quickly it happens, and the timing identifies the mechanism. Hyperacute rejection begins within minutes of the vascular anastomosis being opened and is caused by antibody that was already present before the transplant — anti-ABO isohaemagglutinins, or anti-HLA antibody generated by previous transfusion, pregnancy or a previous graft. The antibody binds donor endothelium, fixes complement, and thromboses the vessels so that the organ never perfuses. Careful ABO matching and the pre-transplant cross-match, in which recipient serum is tested against donor cells, have made it rare.

Acute rejection develops over days to weeks and is the classical T-cell-mediated response, with CD8⁺ cells killing graft parenchyma and CD4⁺ cells driving a delayed-type reaction in the vessel wall; antibody made after the transplant contributes an acute humoral component. This is the form immunosuppression is designed to prevent, and it is usually reversible if caught early. Chronic rejection appears over months to years as a slow fibrosis and concentric narrowing of graft arteries — graft arteriosclerosis — driven mainly by indirect alloantigen presentation and by repeated subclinical injury. It remains the principal reason transplanted organs are eventually lost, and no treatment reverses it.

13.1.2 Preventing rejection, xenotransplantation, transfusion and GVHD

Rejection in an immunocompetent recipient is otherwise inevitable, so immunosuppression is used: calcineurin inhibitors such as cyclosporine and tacrolimus, which block transcription of the IL-2 gene (Section 11.3.1); rapamycin (sirolimus), which blocks mTOR and so the growth-factor response rather than the activation signal; antimetabolites such as mycophenolate and azathioprine, which kill dividing lymphocytes; co-stimulation blockade with the CTLA-4-Ig fusion protein belatacept; depleting antibodies against CD3 or CD52; and corticosteroids. The price is uniform — every one of these agents is non-specific, so the patient is left susceptible to opportunistic infection and, over years, to virus-associated malignancy. Matching donor and recipient at HLA-A, -B and -DR reduces how much immunosuppression is needed, which is why sibling donors are sought first.

Xenotransplantation would solve the shortage of organs if it could be made to work, and the pig is the preferred donor on grounds of size and physiology. The first barrier is hyperacute rejection by natural antibody against the galactose- α -1,3-galactose sugar that pigs make and humans do not; pigs engineered to lack the relevant glycosyltransferase, and more recently multi-gene-edited animals, have largely overcome it, and experimental pig-to-human heart and kidney transplants have been performed. Cellular rejection and the risk of transmitting porcine endogenous retroviruses remain unsolved.

In uncoating, the nucleic acid is separated from the capsid, by host lysosomal or cytoplasmic enzymes or, in poxviruses, by a virus-encoded enzyme. Biosynthesis then produces new genomes and proteins, and maturation and release complete the cycle: capsids assemble, usually spontaneously; enveloped viruses acquire their envelope by budding, so the host cell often survives and keeps producing virus (Figure 14.8); and non-enveloped viruses are released by lysis, which kills the cell (Figure 14.6).

Figure 14.6 *Bacteriophage and animal-virus multiplication compared, stage by stage. The two follow the same overall plan — attachment, entry, uncoating, biosynthesis, release — but differ at almost every step in mechanism, and only animal viruses show latency, slow infections and transformation.*

Stage	Bacteriophages	Animal Viruses
Attachment	Tail fibers attach to cell wall proteins.	Attachment sites are plasma membrane proteins and glycoproteins.
Entry	Viral DNA is injected into host cell.	Capsid enters by receptor-mediated endocytosis or fusion
Uncoating	Not required	Enzymatic removal of capsid proteins
Biosynthesis	In cytoplasm	In nucleus (DNA viruses) or cytoplasm (RNA viruses)
Chronic infection	Lysogeny	Latency; slow viral infections; cancer
Release	Host cell is lysed.	Enveloped viruses bud out; nondeveloped viruses rupture plasma membrane.

14.5.1 Where and how the genome is copied

The single most useful way to organise the viruses is by what they must do to make mRNA, because everything else follows from it — this is the logic behind the Baltimore classification, for which David Baltimore shared the 1975 Nobel Prize. DNA viruses generally replicate their DNA in the nucleus using host or viral polymerases and make their capsid proteins in the cytoplasm; poxviruses are the famous exception, carrying their own machinery and replicating entirely in the cytoplasm. RNA viruses replicate in the cytoplasm, and their strategies differ according to the sense of the genome (Figure 14.7 and Table 14.5).

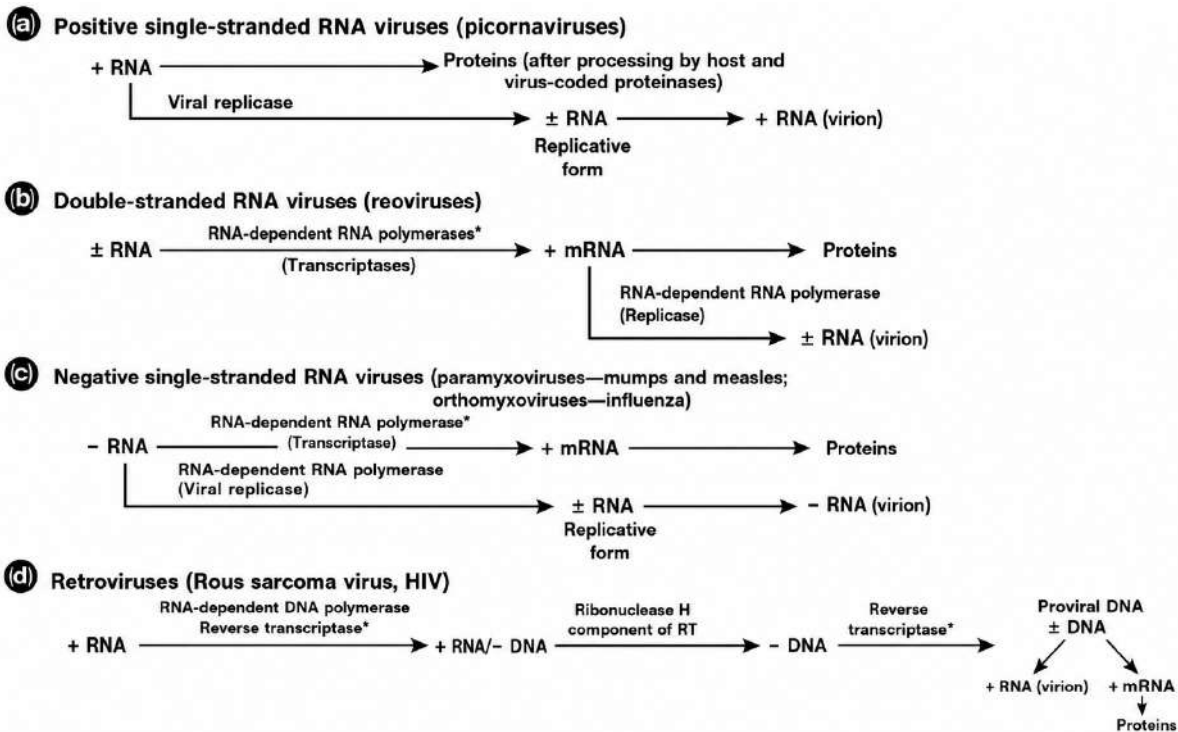


Figure 14.7 *Replication strategies of the RNA viruses.*