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Lecture 11**
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- 2  **UNSW Copyright Notice**
- 3  **A Sample Signaling**
- 4  **Signaling Text References**
 - Essential Cell Biology
 - Chapter 15
 - Molecular Biology of the Cell
 - Chapter 15
 - Molecular Cell Biology
 - Chapter 20
 - Nature Signaling Gateway
 - <http://www.signaling-gateway.org/molecule/>
- 5  **Lecture Summary**
 - Messengers and Receptors
 - chemical signals
 - cellular receptors
 - signal transduction
 - intracellular pathways
 - second messengers
 - Examples of signaling
- 6  **Signaling Mechanisms**
 - Endocrine
 - Hormone
 - Paracrine
 - Locally
 - Neurotransmitter
 - Specific form of paracrine
 - Autocrine
 - Contact Dependent
 - Locally
- 7  **Cell Communication**
 - Contact Mediated
 - display molecules on cell surface
 - recognized by receptor on another cell
 - Non-Contact Mediated
 - chemical signal
 - nearby or at a distance

- 8  **Common Signals**
- 9  **Signals and Receptors**
- 10  **Signal Transduction Model**
- 11  **Signal Transduction Model**
- 12  **Signal Transduction Model**
- 13  **Signal Transduction Model**
- 14  **Signaling between Tissues**
 - Regulation of cells and tissues
 - Hormones
 - secreted by one tissue to regulate function of other cells or tissues
 - Chemical Signal Types
 - water soluble
 - lipid soluble
- 15  **Movie: Hormone Signaling**
- 16  **Extracellular Signal Steps**
 - Signaling Molecule
 - Synthesis
 - Release by signaling cell
 - Transport to target cell
 - Detection by a specific receptor protein
 - Change by receptor-signal complex (trigger)
 - Metabolism
 - Function
 - development
 - Removal of the signal
 - often terminates cellular response
- 17  **Messenger /Receptor Interaction**
 - Binding of messenger (ligand) has to lead to a change in the receptor
 - like enzyme and substrate
 - Specific recognition
 - Receptor affinity
 - activation
 - signal transduction
 - Signal cascade
 - Secondary messengers
- 18  **Chemical Signals**
 - Water Soluble
 - bind to surface receptors
 - Lipid Soluble
 - bind to cytoplasmic or nuclear receptors
 - steroid hormones
- 19  **Receptor Pathways**

- 20  **Second Messengers**
- 21  **Activation of the GTPase Rac in living motile fibroblast**
- 22  **Second Messengers**
- Cyclic nucleotides
 - cAMP, cGMP
 - Calcium Ions
 - Protein Kinase A
 - PKA, B, C
 - diacylglycerol (DAG)
 - modified lipid activates PKC
 - Kinase cascades
 - small GTP binding proteins
 - related to RAS which is G protein family
- 23  **Movie: Signaling IP3 / calcium**
- 24  **2 Signal Transductions**
- 25  **cAMP and Kinases**
- 26  **Lipids in Cell Signaling**
- Arachidonic Acid (AA) pathway
 - generates many of the lipids involved as second messengers in cell signaling pathways
- 27  **Lipid Soluble-Steroids**
- 28  **Steroid Responses**
- 29  **Intracellular Receptors**
- Steroid Hormones
 - thyroxine
 - vitamin D3
 - retinoic acid
 - Nuclear location
 - Cytosol location
 - translocates to nucleus on ligand binding
 - binds ligand and DNA
 - becomes transcription factor
- 30  **Steroid Receptors**
- steroid binding region
 - near C-terminus
 - DNA binding
 - central region
 - zinc finger motif
 - alpha helix and 2 beta sheets held in place by cysteine or histidine residues by a zinc atom
 - multiple fingers typical
 - DNA response element
 - Enhancer
- 31  **Steroid Receptor Pathway**
- 32  **Steroid Receptor Pathway**

- 33  **Steroid Receptor Pathway**
- 34  **Steroid Receptor Pathway**
- 35  **Other Transcription Factors**
- 36  **Membrane Receptors**
 - embedded in plasma membrane
 - ligand binding
 - leads to conformational change in receptor
 - activation of intracellular pathway
 - G Protein Linked receptors
- 37  **Cell Surface Receptor Types**
- 38  **G Protein Receptors**
- 39  **G Protein-Coupled Signal Pathways**
 - Transmembrane proteins transduce extracellular signals
 - common structural motif of 7 membrane spanning regions
 - Receptor binding promotes interaction
 - between receptor
 - G protein on interior surface of membrane
- 40  **G Protein-Coupled Signal Pathways**
 - induces an exchange of GDP for GTP on G protein α subunit and dissociation of the α subunit from the $\beta\gamma$ heterodimer
 - Depending on isoform, GTP- α subunit complex mediates intracellular signaling either
 - **indirectly** by acting on effector molecules
 - adenylyl cyclase (AC)
 - phospholipase C (PLC)
 - **directly** by regulating ion channel or kinase function
- 41  **G Protein Linked**
- 42  **Receptor associated with Kinase**
 - many growth factors use this pathway
 - Vascular Endothelial Growth Factor
 - Epidermal Growth Factor
 - Nerve Growth Factor
 - Bone Morphogenic Protein
 - Transforming Growth Factor-beta
 - Ligand binding
 - Receptor association
 - Phosphorylation
 - Kinase cascade
- 43  **VEGF Receptor and Ligands**
- 44  **EGF Receptor Transduction Pathway**
- 45  **Signaling Pathway of TGF- β**
- 46  **TrkA Receptor**

- Trk proto-oncogenes
 - TrkA, TrkB, TrkC, TrkE
- variably expressed in CNS and PNS
- TrkA binds to nerve growth factor (NGF) and autophosphorylates
 - leading to activation of multiple downstream effector proteins

47 Proto-oncogenes

- proto-oncogenes
 - Normal cell proteins that have potential to cause uncontrolled growth when mutated
- loss of receptor regulation
- cells grow out of control
- mutation in TK Receptor
 - receptor always activated
- mutation of activating protein
 - always active
- Oncogenes
 - Ras
 - mutants detected in 30% cervical cancers

48 Movie: Methods Receptor/Ligand

49 Movie: Receptor Internalization

- HEK-293 cells express GFP tagged Beta-2 adrenergic receptors
 - treated with noradrenaline and imaged by time-lapse confocal microscopy at 5 s intervals over 30 minutes
- movement underneath plasma membrane due to noradrenaline-evoked internalisation of receptors

50 Movie: GLUT4 Dynamics

- dynamics of glucose transporter isoform 4 (GLUT4)-containing vesicles in 3T3-L1 adipocytes microinjected with GFP-GLUT4
- After 24h adipocytes were serum-starved for 3h prior to imaging
- Insulin was added at t=0
- cell was imaged at 1 frame/s
- Two types of movement GLUT4 vesicles are evident:
 - rapid vibrational-type displacements
 - rapid movements over short distances

51 Movie:

- Expressed transiently in porcine aortic endothelial (PAE) cells
- GFP tagged 32 kDa PtIns(3,4,5)P3-binding protein (DAPP1)
- translocated from cytosol to plasma membrane in response to platelet-derived growth factor (PDGF)

52 Movie: Agonist-induced translocation of EGFP-PHPLC

- Agonist-induced translocation of EGFP-PHPLC _ in SH-SY5Y and CHO-lac-mGlu1_cells
- Single-cell imaging of graded Ins(1,4,5)P3 production following G-protein-coupled-receptor activation

53 Growth Factors

54 Online References

- ANAT3231 Lectures
 - <http://cellbiology.med.unsw.edu.au/units/science/lectures.htm>
- Molecular Biology of the Cell (Ch15)
 - <http://www.ncbi.nlm.nih.gov:80/books/bv.fcgi?call=bv.View..ShowSection&rid=cell.section.3834>
- Developmental Biology (Ch6)
 - <http://www.ncbi.nlm.nih.gov:80/books/bv.fcgi?call=bv.View..ShowSection&rid=.TWQLjiW2xLXyWUOVemh0sDWJf2YbG1QHHjC->
- Molecular Cell Biology (Ch20)
 - <http://www.ncbi.nlm.nih.gov:80/books/bv.fcgi?call=bv.View..ShowSection&rid=mcb.chapter.5687>

- The Cell- A molecular approach (Ch13)
 - <http://www.ncbi.nlm.nih.gov:80/books/bv.fcgi?tool=bookshelf&call=bv.View..ShowSection&searchterm=cell&rid=cooper.chapter.2198>
- Sigma Apoptosis Brochure
 - http://www.sigmaaldrich.com/Area_of_Interest/Life_Science/Cell_Signaling.html

55 **Signal Transduction Research Labs**

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56 **Reference: Molecular Biology of Cell**

- III. Internal Organization of the Cell
 - 15. Cell Signaling
 - Introduction
 - General Principles of Cell Signaling
 - Signaling via G-Protein-linked Cell-Surface Receptors
 - Signaling via Enzyme-linked Cell-Surface Receptors
 - Target-Cell Adaptation
 - The Logic of Intracellular Signaling: Lessons from Computer-based "Neural Networks"
 - References

57 **Reference: Molecular Cell Biology**

- 20. Cell-to-Cell Signaling: Hormones and Receptors
 - 20.1 Overview of Extracellular Signaling
 - 20.2 Identification and Purification of Cell-Surface Receptors
 - 20.3 G Protein –Coupled Receptors and Their Effectors
 - 20.4 Receptor Tyrosine Kinases and Ras
 - 20.5 MAP Kinase Pathways
 - 20.6 Second Messengers
 - 20.7 Interaction and Regulation of Signaling Pathways
 - 20.8 From Plasma Membrane to Nucleus
 - PERSPECTIVES
 - Future
 - Literature

58 **Reference: The Cell**

- IV. Cell Regulation
 - 13. Cell Signaling
 - Signaling Molecules and Their Receptors
 - Functions of Cell Surface Receptors
 - Pathways of Intracellular Signal Transduction
 - Signal Transduction and the Cytoskeleton

- Signaling in Development and Differentiation
- Regulation of Programmed Cell Death
- Summary
- Questions
- References and Further Reading

59  **Reference: Developmental Biology**

- Part 1. Principles of development in biology
 - 6. Cell-cell communication in development
 - Induction and Competence
 - Paracrine Factors
 - Cell Surface Receptors and Their Signal Transduction Pathways
 - The Cell Death Pathways
 - Juxtacrine Signaling
 - Cross-Talk between Pathways
 - Coda
 - Principles of Development:Cell-Cell Communication
 - References