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BMED 600.01: Advanced Cellular Biochemistry

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Advanced Cellular Biochemistry Bioc/Phar 600 Spring 2007

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Class 8:40 am - 10:00 am	MF	Skaggs Building 336	Jan 22, 2007 - May 4, 2007
Class 8:40 am - 10:00 am	W	Skaggs Building 114	Jan 22, 2007 - May 4, 2007

Catalog course description (4 cr.) Exploration on a molecular level the regulation of structure, function, and dynamics of eukaryotic cells. Topics include membranes, cytoskeleton, transcription, translation, signal transduction, cell motility, cell proliferation, and programmed cell death.

Overview

Cell Biology is vast and dense and encompasses biochemistry, biophysics, molecular biology, microscopy, genetics, physiology, computer science, and developmental biology. This course will use as a main text Alberts, et al., *Molecular Biology of the Cell,* 4th ed. (5th if it is out; Garland Science). This text will be a jumping off point into reading from the scientific literature. We will explore the topics listed below by reading reviews and papers from the primary literature. Papers will be chosen, where possible, that are at the interface between two fields, so a large amount of background reading will be necessary to understand the paper and put it in context. The two main learning goals are 1) to learn about a number of topics in cell biology; and 2) to gain the confidence and skills to attack any scientific paper even if it is in an unfamiliar area.

Format

Presentations will be made by instructors and students. Instructors will introduce a topic with a lecture, and a student presentation of a paper with data will follow in the next session.

The student's presentation should set the stage for the paper being presented with a brief introduction that draws on recent reviews. Keep in mind the following questions when presenting a paper. What is the key question being addressed by the experiments? What are the key experiments that address the question? Do you believe their interpretation, and did they do the proper controls? Many of the methods used to study cell biology and biochemistry are evolving, and far from perfect, so it is important to look with a critical eye at the data, the methods used to obtain the data, and how the data are interpreted.

When tackling any research paper in an unfamiliar area, the best way to start is by reading one or more textbooks (use the index and table of contents) and reviews, looking up unknown concepts mentioned in the paper's introduction (often reviews are cited there too). Then look at the data in the figures. If you don't understand the methods, look them up[1]. Then read the results and discussion, and decide whether the author's interpretation of the data is the same or different than yours.

Student's presentation papers will be assigned in advance to allow time for preparation. Students are strongly encouraged to seek instructors' help. Please ask questions by email, phone, or by stopping by any time (call first to make sure we are around). We intend a relaxed atmosphere where we all ask questions, an no questions are dumb questions. All students will be required to read all papers. It is expected that the student presenting the paper will be better informed on the topic, which will require extra work. While errors and misunderstandings are forgivable, we expect you to make an effort to understand the paper being presented, especially if you are the one presenting it!

Blackboard (http://umonline.umt.edu/) will be used to post course documents such as lecture slides, papers, and assignments. The website has excellent instructions on how to login and use Blackboard.

[1] Note: Some methods can be found on the web at: http://www.scienceboard.net/resources/protocols.asp?criteria=

Assessment

The course grade will be assigned based on oral presentations, written assignments, quizzes and exams. One assignment will be a critical review of a paper from the primary literature. The other will be a hypothesis and experimental design to answer a question in cell biology in grant proposal format. There will be two exams in which interpretation of data will be emphasized. Quizzes will be called spontaneously during one class period for the next class period. Written questions from all students (except presenting students) will be required for student presentations. Assignments, presentations, quizzes, written questions and exams will be proportionally used to determine the grade, with class participation added as bonus points. The final exam will be comprehensive.

The Provost's Official Fine Print
All students must practice academic honesty. Academic misconduct is subject to an academic penalty by the course instructor and/or a disciplinary sanction by the University.

All students need to be familiar with the Student Conduct Code. The Code is available for review online at http://www.umt.edu/SA/VPSA/index.cfm/page/1321.

Topics

(Note: the order of topics and recent papers are subject to change.)

Cell structure, lipids and membrane traffic (Alberts, Chapters 10, 12, and 13)

Reviews

van Meer, G., and Sprong, H. (2004). Membrane lipids and vesicular traffic. Curr Opin Cell Biol *16*, 373-378.

Perret, E., Lakkaraju, A., Deborde, S., Schreiner, R., and Rodriguez-Boulan, E. (2005). Evolving endosomes: how many varieties and why? Curr Opin Cell Biol 17, 423-434.

De Matteis, M.A., and Godi, A. (2004). PI-loting membrane traffic. Nat Cell Biol 6, 487-492.

Behnia, R., and Munro, S. (2005). Organelle identity and the signposts for membrane traffic. Nature 438, 597-604.

Seabra, M.C., and Wasmeier, C. (2004). Controlling the location and activation of Rab GTPases. Curr Opin Cell Biol *16*, 451-457.

Emery, G., and Knoblich, J.A. (2006). Endosome dynamics during development. Curr Opin Cell Biol *18*, 407-415.

Primary papers

Matsuo, H., Chevallier, J., Mayran, N., Le Blanc, I., Ferguson, C., Faure, J., Blanc, N.S., Matile, S., Dubochet, J., Sadoul, R., Parton, R.G., Vilbois, F., and Gruenberg, J. (2004). Role of LBPA and Alix in multivesicular liposome formation and endosome organization. Science *303*, 531-534.

Sharma, D.K., Brown, J.C., Choudhury, A., Peterson, T.E., Holicky, E., Marks, D.L., Simari, R., Parton, R.G., and Pagano, R.E. (2004). Selective stimulation of caveolar endocytosis by glycosphingolipids and cholesterol. Mol Biol Cell *15*, 3114-3122.

Emery, G., Hutterer, A., Berdnik, D., Mayer, B., Wirtz-Peitz, F., Gaitan, M.G., and Knoblich, J.A. (2005). Asymmetric Rab 11 endosomes regulate delta recycling and specify cell fate in the Drosophila nervous system. Cell *122*, 763-773.

Glebov, O.O., Bright, N.A., and Nichols, B.J. (2006). Flotillin-1 defines a clathrin-independent endocytic pathway in mammalian cells. Nat Cell Biol *8*, 46-54.

The cytoskeleton (Alberts, Chapters 16 and 18)

Reviews

Stradal, T.E., and Scita, G. (2005). Protein complexes regulating Arp2/3-mediated actin assembly. Curr Opin Cell Biol.

Gundersen, G.G., Gomes, E.R., and Wen, Y. (2004). Cortical control of microtubule stability and polarization. Curr Opin Cell Biol *16*, 106-112.

Guzik, B.W., and Goldstein, L.S. (2004). Microtubule-dependent transport in neurons: steps towards an understanding of regulation, function and dysfunction. Curr Opin Cell Biol *16*, 443-450.

Kaksonen, M., Toret, C.P., and Drubin, D.G. (2006). Harnessing actin dynamics for clathrin-mediated endocytosis. Nat Rev Mol Cell Biol *7*, 404-414.

Munro, E.M. (2005). PAR proteins and the cytoskeleton: a marriage of equals. Curr Opin Cell Biol.

Primary papers

Dujardin, D.L., Barnhart, L.E., Stehman, S.A., Gomes, E.R., Gundersen, G.G., and Vallee, R.B. (2003). A role for cytoplasmic dynein and LIS1 in directed cell movement. J Cell Biol *163*, 1205-1211.

Munro, E., Nance, J., and Priess, J.R. (2004). Cortical flows powered by asymmetrical contraction transport PAR proteins to establish and maintain anterior-posterior polarity in the early C. elegans embryo. Dev Cell 7, 413-424.

Signal transduction and intracellular localization (Alberts, Chapters 15 as well as 12, 13, and 16)

Reviews

Polo, S., Pece, S., and Di Fiore, P.P. (2004). Endocytosis and cancer. Curr Opin Cell Biol *16*, 156-161.

Di Fiore, P.P., Polo, S., and Hofmann, K. (2003). When ubiquitin meets ubiquitin receptors: a signalling connection. Nat Rev Mol Cell Biol *4*, 491-497.

Prior, I.A., and Hancock, J.F. (2001). Compartmentalization of Ras proteins. J Cell Sci 114, 1603-1608.

Miaczynska, M., Pelkmans, L., and Zerial, M. (2004). Not just a sink: endosomes in control of signal transduction. Curr Opin Cell Biol *16*, 400-406.

Lefkowitz, R.J., and Whalen, E.J. (2004). beta-arrestins: traffic cops of cell signaling. Curr Opin Cell Biol 16, 162-168.

Paratcha, G., and Ibanez, C.F. (2002). Lipid rafts and the control of neurotrophic factor signaling in the nervous system: variations on a theme. Curr Opin Neurobiol 12, 542-549.

Bucci, C., and Chiariello, M. (2006). Signal transduction gRABs attention. Cell Signal 18, 1-8.

Hoeller, D., Volarevic, S., and Dikic, I. (2005). Compartmentalization of growth factor receptor signalling. Curr Opin Cell Biol 17, 107-111.

Rocks, O., Peyker, A., and Bastiaens, P.I. (2006). Spatio-temporal segregation of Ras signals: one ship, three anchors, many harbors. Curr Opin Cell Biol *18*, 351-357.

Primary papers

Rocks, O., Peyker, A., Kahms, M., Verveer, P.J., Koerner, C., Lumbierres, M., Kuhlmann, J., Waldmann, H., Wittinghofer, A., and Bastiaens, P.I. (2005). An acylation cycle regulates localization and activity of palmitoylated Ras isoforms. Science 307, 1746-1752.

Miaczynska, M., Christoforidis, S., Giner, A., Shevchenko, A., Uttenweiler-Joseph, S., Habermann, B., Wilm, M., Parton, R.G., and Zerial, M. (2004a). APPL proteins link Rab5 to nuclear signal transduction via an endosomal compartment. Cell *116*, 445-456.

Polo, S., Sigismund, S., Faretta, M., Guidi, M., Capua, M.R., Bossi, G., Chen, H., De Camilli, P., and Di Fiore, P.P. (2002). A single motif responsible for ubiquitin recognition and monoubiquitination in endocytic proteins. Nature *416*, 451-455.

Haglund, K., Sigismund, S., Polo, S., Szymkiewicz, I., Di Fiore, P.P., and Dikic, I. (2003). Multiple monoubiquitination of RTKs is sufficient for their endocytosis and degradation. Nat Cell Biol *5*, 461-466.

Chen, W., Ren, X.R., Nelson, C.D., Barak, L.S., Chen, J.K., Beachy, P.A., de Sauvage, F., and Lefkowitz, R.J. (2004). Activity-dependent internalization of smoothened mediated by beta-arrestin 2 and GRK2. Science *306*, 2257-2260.

Chen, W., ten Berge, D., Brown, J., Ahn, S., Hu, L.A., Miller, W.E., Caron, M.G., Barak, L.S., Nusse, R., and Lefkowitz, R.J. (2003). Dishevelled 2 recruits beta-arrestin 2 to mediate Wnt5A-stimulated endocytosis of Frizzled 4. Science *301*, 1391-1394.

The Cell Cycle (Alberts Chapters 17 and 23)

Reviews

Yamasaki, L., and Pagano, M. (2004). Cell cycle, proteolysis and cancer. Curr Opin Cell Biol *16*, 623-628.

Humbert, P.O., Brumby, A.M., Quinn, L.M., and Richardson, H.E. (2004). New tricks for old dogs: unexpected roles for cell cycle regulators revealed using animal models. Curr Opin Cell Biol *16*, 614-622.

Primary papers

Bashir, T., Dorrello, N.V., Amador, V., Guardavaccaro, D., and Pagano, M. (2004). Control of the SCF(Skp2-Cks1) ubiquitin ligase by the APC/C(Cdh1) ubiquitin ligase. Nature *428*, 190-193.

Wei, W., Ayad, N.G., Wan, Y., Zhang, G.J., Kirschner, M.W., and Kaelin, W.G., Jr. (2004). Degradation of the SCF component Skp2 in cell-cycle phase G1 by the anaphase-promoting complex. Nature *428*, 194-198.

Programmed cell death (Alberts Chapter 17)

Reviews

Willis, S.N., and Adams, J.M. (2005). Life in the balance: how BH3-only proteins induce apoptosis. Curr Opin Cell Biol 17, 617-625.

Boatright, K.M., and Salvesen, G.S. (2003). Mechanisms of caspase activation. Curr Opin Cell Biol *15*, 725-731.

Bossy-Wetzel, et al., Mitochondrial Fission in apoptosis, neurodegeneration and aging. Current Opinion in Cell Biology **15:**706-716, 2003.

Kuwana, T., and Newmeyer, D.D. (2003). Bcl-2-family proteins and the role of mitochondria in apoptosis. Curr Opin Cell Biol *15*, 691-699.

Bergmann, A., Yang, A.Y., and Srivastava, M. (2003). Regulators of IAP function: coming to grips with the grim reaper. Curr Opin Cell Biol *15*, 717-724.

Breckenridge, D.G., and Xue, D. (2004). Regulation of mitochondrial membrane permeabilization by BCL-2 family proteins and caspases. Curr Opin Cell Biol *16*, 647-652.

Ekert, P.G., and Vaux, D.L. (2005). The mitochondrial death squad: hardened killers or innocent bystanders? Curr Opin Cell Biol 17, 626-630.

Primary papers

Chipuk, J.E., Kuwana, T., Bouchier-Hayes, L., Droin, N.M., Newmeyer, D.D., Schuler, M., and Green, D.R. (2004). Direct activation of Bax by p53 mediates mitochondrial membrane permeabilization and apoptosis. Science *303*, 1010-1014.

Kuwana, T., Mackey, M.R., Perkins, G., Ellisman, M.H., Latterich, M., Schneiter, R., Green, D.R., and Newmeyer, D.D. (2002). Bid, Bax, and lipids cooperate to form supramolecular openings in the outer mitochondrial membrane. Cell *111*, 331-342.

Breckenridge, D.G., Stojanovic, M., Marcellus, R.C., and Shore, G.C. (2003). Caspase cleavage product of BAP31 induces mitochondrial fission through endoplasmic reticulum calcium signals, enhancing cytochrome c release to the cytosol. J Cell Biol *160*, 1115-1127.

Adhesion and stem cells (Alberts Chapters 19, 21, 24 and 25)

Reviews

Braga, V.M., and Yap, A.S. (2005). The challenges of abundance: epithelial junctions and small GTPase signalling. Curr Opin Cell Biol 17, 466-474.

Bos, J.L. (2005). Linking Rap to cell adhesion. Curr Opin Cell Biol 17, 123-128.

Kleber, M., and Sommer, L. (2004). Wnt signaling and the regulation of stem cell function. Curr Opin Cell Biol *16*, 681-687.

Mayhall, E.A., Paffett-Lugassy, N., and Zon, L.I. (2004). The clinical potential of stem cells. Curr Opin Cell Biol *16*, 713-720.

Molofsky, A.V., Pardal, R., and Morrison, S.J. (2004). Diverse mechanisms regulate stem cell self-renewal. Curr Opin Cell Biol *16*, 700-707.

Näthke, I. (2006), "Cytoskeleton out of the cupboard: how changes in the cytoskeleton induced by loss of APC contribute to colon cancer" *Nat. Cancer Reviews*, **6**: 967-974. (Cover image)

Näthke, I. and Nelson, W.J. (2005) "Cell-to-cell contact and extracellular matrix Laying down the rules for proper cell behavior", Curr Opin Cell Biol. **17**:443-445.

Primary papers

Kroboth, K., Newton, I.P., Zumbrunn, J., Li, Z., Kita, K., Waterman-Storer, C.M., Näthke, I.S., "Lack of adenomatous polyposis coli protein correlates with a decrease in cell migration and overall changes in microtubule stability", *Mol. Biol. Cell*, in press.

Dikovskaya, D., Schiffmann, D., Newton, I.P., Oakley, A., Kroboth, K., Sansom, O., Jamieson, T.J., Meniel, V., Clarke, A., and Nathke, I.S. (2007). Loss of APC induces polyploidy as a result of a combination of defects in mitosis and apoptosis. The Journal of Cell Biology *176*, 183-195.

del Pozo, M.A., Alderson, N.B., Kiosses, W.B., Chiang, H.H., Anderson, R.G., and Schwartz, M.A. (2004). Integrins regulate Rac targeting by internalization of membrane domains. Science *303*, 839-842.

Molofsky, A.V., Pardal, R., Iwashita, T., Park, I.K., Clarke, M.F., and Morrison, S.J. (2003). Bmi-1 dependence distinguishes neural stem cell self-renewal from progenitor proliferation. Nature *425*, 962-967.

Hari, L., Brault, V., Kleber, M., Lee, H.Y., Ille, F., Leimeroth, R., Paratore, C., Suter, U., Kemler, R., and Sommer, L. (2002). Lineage-specific requirements of beta-catenin in neural crest development. J Cell Biol *159*, 867-880.

Lee, H.Y., Kleber, M., Hari, L., Brault, V., Suter, U., Taketo, M.M., Kemler, R., and Sommer, L. (2004). Instructive role of Wnt/β-catenin in sensory fate specification in neural crest stem cells. Science *303*, 1020-1023.

Transcription, gene regulation and chromatin remodelling

Alberts et al. (2004) Ch. 4, pp. 191-216; Ch. 6, pp. 299-315; and Ch. 7, pp. 375-415 & pp. 422-435.

Reviews

Boeger et al. (2005) Structural basis of eukaryotic gene transcription. FEBS Lett 579:899-903.

Ebright (2000) RNA polymerase: structural similarities between bacterial RNA polymerase and eukaryotic RNA polymerase II. J Mol Biol 304:687-98.

Jin et al. (2005) In and out: histone variant exchange in chromatin. Trends Biochem Sci 30:680-7.

Johnson et al. (2005) Chromatin remodeling complexes: ATP-dependent machines in action. Biochem Cell Biol 83:405-17.

Kornberg (2005) Mediator and the mechanism of transcriptional activation. Trends Biochem Sci 30:235-9.

Malik & Roeder (2005) Dynamic regulation of pol II transcription by the mammalian Mediator complex. Trends Biochem Sci 30:256-63.

Meinhart et al. (2005) A structural perspective of CTD function. Genes Dev 19:1401-15.

Mellor (2005) The dynamics of chromatin remodeling at promoters. Mol Cell 19:147-57.

Orphanides & Reinberg (2002) A unified theory of gene regulation. Cell 108:439-451.

Sims et al. (2004) Recent highlights of RNA-polymerase II mediated transcription. Curr Opin Cell Biol 16:263-271

Primary papers

Ahn et al. (2004) Phosphorylation of serine 2 within the RNA polymerase II C-terminal domain couples transcription and 3' end processing. Mol Cell 13:67-76.

Alper et al. (2006) Engineering yeast transcription machinery for improved ethanol tolerance and production. Science 314:1565-8.

Lorch et al. (2006) Chromatin remodeling by nucleosome disassembly in vitro. (2006) Proc Natl Acad Sci USA 103:3090-3.

Wang et al. (2005) Mediator requirement for both recruitment and postrecruitment steps in transcription initiation. Mol Cell 17:683-94.

Wang et al. (2006) Structural basis of transcription: role of the trigger loop in substrate specificity and catalysis. Cell 27:941-54

mRNA processing

Alberts et al. (2004) Ch. 6, pp. 315-335; and Ch. 7, pp. 435-451.

Reviews

Bentley (2005) Rules of engagement: co-transcriptional recruitment of pre-mRNA processing factors. Curr Opin Cell Biol 17:251-6.

Maquat (2004) Nonsense-mediated mRNA decay: splicing, translation and mRNP dynamics. Nat Rev Mol Cell Biol 5:89-99.

Matlin et al. (2005) Understanding alternative splicing: towards a cellular code. Nat Rev Mol Cell Biol 6:386-98.

Moore (2005) From birth to death: the complex lives of eukaryotic mRNAs. Science 309:1514-8.

Roy & Gilbert (2006) The evolution of spliceosomal introns: patterns, puzzles and progress. Nat Rev Genet 7:211-21.

Valadkhan (2005) snRNAs as the catalysts of pre-mRNA splicing. Curr Opin Chem Biol 9:603-8.

Primary papers

Gudikote et al. (2005) RNA splicing promotes translation and RNA surveillance. Nat Struct Mol Biol 12:801-9.

Nielsen et al. (2005) An mRNA is capped by a 2', 5' lariat catalyzed by a group I-like ribozyme.

Science 309:1584-7.

Sheth & Parker (2006) Targeting of aberrant mRNAs to cytoplasmic processing bodies. Cell 125:1095-109.

Yeo et al. (2005) Identification and analysis of alternative splicing events conserved in human and mouse. Proc Natl Acad Sci USA 102:2850-5.

Young et al. (2005) Regulation of RNA splicing by the methylation-dependent transcriptional repressor methyl-CpG binding protein 2. Proc Natl Acad Sci USA 102:17551-8.

Small RNAs, RNAi and riboswitches

Alberts et al. (2004) Ch. 7, pp. 451-452.

Reviews

Bernards (2006) Exploring the uses of RNAi--gene knockdown and the Nobel Prize. N Engl J Med 355:2391-3.

Gottesman (2004) The small RNA regulators of *Escherichia coli*: roles and mechanisms. Annu Rev Microbiol 58:303-28.

Hammond (2005) Dicing and slicing: the core machinery of the RNA interference pathway. FEBS Lett 579:5822-9.

Mandal & Breaker (2004) Gene regulation by riboswitches. Nat Rev Mol Cell Biol 5:451-63.

Nishikura (2006) Editor meets silencer: crosstalk between RNA editing and RNA interference. Nat Rev Mol Cell Biol 7:919-31.

Sontheimer (2005) Assembly and function of RNA silencing complexes. Nature Rev Mol Cell Biol 6:127-138.

Zamore & Haley (2005) Ribo-gnome: the big world of small RNAs. Science 309:1519-24.

Primary papers

Chowdhury (2006) Molecular basis for temperature sensing by an RNA thermometer. EMBO J 25:2487-97.

Macrae et al. (2006) Structural basis for double-stranded RNA processing by Dicer. Science 311:195-8.

Montange & Batey (2006) Structure of the S-adenosylmethionine riboswitch regulatory mRNA element. Nature 441:1172-5.

Rivas et al. (2005) Purified Argonaute2 and an siRNA form recombinant human RISC. Nat Struct Mol Biol 12:340-9.

Wassarman and Saecker (2006) Synthesis-mediated release of a small RNA inhibitor of RNA polymerase. Science 314:1601-3.