Microbial Pathogenesis

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Pre-Discussion Assignments

Discussion Session 1 **Bacterial Virulence**

Kim SI, Kim S, Kim E, Hwang SY, Yoon H. 2018. Front Microbiol. 9: 2810.

1. Read the Introduction
2. Identify the central question/claim
3. Outline an experimental design for each of the following claims from the paper. Focus on which techniques you would use to address the claim and what controls you would need to interpret your results. Feel free to use bullet points and lists.
    a. T3SS1 effectors are secreted via OMVs in *Salmonella*
    b. T3SS1 effectors are located on the outer surface of OMVs
    c. OMV-associated SipA, SipC, and SopE2 are translocated into the cytoplasm of the host cell
    d. OMVs harboring T3SS1 effectors induce host actin cytoskeletal rearrangements
    e. OMVs harboring T3SS1 effectors promote the invasive ability of *Salmonella*

Discussion Session 2 **Microbiome**

Jemielita M, Wingreen NS, Bassler BL. 2018. eLife. 7:e42057.

1. Read the Introduction
    a. Diagram the basic quorum-sensing pathway in *Vibrio cholerae* in low cell density and high cell density environments
    b. Outline the basic *Vibrio cholerae* lifecycle, indicating where quorum-sensing plays a role
2. Identify the central question/claim
3. Outline an experimental design for each of the following claims from the paper. Focus on which techniques you would use to address the claim and what controls you would need to interpret your results. Feel free to use bullet points and lists.
    a. V. cholerae forms multicellular aggregates in the HCD QS-state
    b. Aggregation dynamics are rapid
    c. Aggregation is HapR dependent
    d. A genetic screen to identify components required for V. cholerae aggregation (outline how you might design such a screen)
4. Additional claims from the paper (not required, but you can think through experiments if you're feeling ambitious)
    a. HCD-QS aggregates are VpsL-independent
    b. Exogenous supplementation of QS autoinducers induces aggregation
    c. eDNA contributes to aggregation

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Discussion Session 3 **Plant-Microbe Interactions**

Asai, *et al.* 2018. Nature Communications. 9:5192.

1. Read the introduction, skim background papers
2. Identify the central question/claim
3. Outline an experimental design for each of the following claims from the paper. Focus on which techniques you would use to address the claim and what controls you would need to interpret your results. Feel free to use bullet points and lists.
    a. HaRxL103^{Emoy2} is an *Hpa* effector recognized by *RPP4*
        i. HaRxL103 induces RPP4-dependent cell death
        ii. RPP4 recognizes HaRxL103 in an EDS1-dependent manner
    b. HaRxL103^{Hind2} evades recognition by changing localization
        i. In planta subcellular localization and RPP4-mediated HR cell death of HaRxL103 alleles
        ii. Lack of nuclear localization allows Hind2 allele to evade detection
4. Additional claims from the paper (not required, but you can think through experiments if you're feeling ambitious)
    a. Identification of *RPP4*-recognized effectors
    b. HaRxL103^{Emoy2} recognition requires RPP4 nuclear localization
    c. Localization of HaRxL103-RPP4 interaction
    d. Virulence co-segregates with lack of HaRxL103 expression

Discussion Session 4 **Fungal Virulence**

Chang AL, Kang Y, Doering TL. 2019. mBio. 10(1): 10:e02818-18. https://doi.org/10.1128/mBio.02818-18.

1. Read the introduction, skim background papers
2. Identify the central question/claim
3. Outline an experimental design for each of the following claims from the paper. Focus on which techniques you would use to address the claim and what controls you would need to interpret your results. Feel free to use bullet points and lists.
    a. Deletion of CDK8 or SSN801 in *Cryptococcus neoformans* results in abnormal, fragmented mitochondrial morphology
    b. Cells lacking CDK8 and SSN801 have increased sensitivity to nitrosative and oxidative stress
    c. Defects in intracellular survival are reversed by ROS scavengers
    d. Mice infected with CDK8 mutant strain survive longer and have altered organ burden compared to WT-infected mice
    e. The CDK8 mutant is impaired in blood-brain barrier crossing but grows normally in artificial cerebrospinal fluid

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4. Additional claims from the paper (not required, but you can think through experiments if you're feeling ambitious)
    a. Cells lacking CDK8 show reduced filamentation and resistance to cell wall stress
    b. SSN801-HA localizes to the nucleus
    c. CDK8 and SSN801 regulate specific, not global, transcriptional programs
    d. Cells lacking CDK8 and SSN801 are impaired in growth on acetate

Post-Discussion Assignments

Discussion Session 1	**Bacterial Virulence**
Kim SI, Kim S, Kim E, Hwang SY, Yoon H. 2018. Front Microbiol. 9: 2810.	Interpret the remaining figures and read the discussion. Upload your response to one of the following: 1. A question you have 2. Something you learned 3. An experiment you would like to do next
Discussion Session 2	**Microbiome**
Jemielita M, Wingreen NS, Bassler BL. 2018. eLife. 7:e42057.	Interpret the remaining figures and read the discussion. Upload your response to one of the following: 1. A question you have 2. Something you learned 3. An experiment you would like to do next
Discussion Session 3	**Plant-Microbe Interactions**
Asai, *et al.* 2018. Nature Communications. 9:5192.	Interpret the remaining figures and read the discussion. Upload your response to one of the following: 1. A question you have 2. Something you learned 3. An experiment you would like to do next
Discussion Session 4	**Fungal Virulence**
Chang AL, Kang Y, Doering TL. 2019. mBio. 10(1): 10:e02818-18. https://doi.org/10.1128/mBio.02818-18.	Interpret the remaining figures and read the discussion. Outline the proposed model from the discussion and annotate with any questions you may have or inconsistencies you notice. As we discovered in class, this paper has a surprising twist, so pay particular attention to how their model accounts for the seemingly contradictory data they collected. Responses may be uploaded as a picture if it is easier to draw the model by hand.