PhD Diary Entry for week beginning October 8th 2018

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Contents

1 Preface

This week has been mostly ASM talks - a lot of work still labelled `TODO` will have been moved to week 42

2 Things that need doing

2.1 TODO Need to find specific and relevant modelling papers

- Preferably with some concrete data to illustrate how to handle and what it may end up looking like

2.2 TODO Would also be good to poss discuss analytical methods prior to data production?

3 Paper Synopsis'

3.1 Architecture and Dynamics of the JA Gene Regulatory Network [1]

Half finished - need to investigate if JA worth reading more on?

- In nature plants are subject to a broad range of attack from pests and pathogens
- Sophisticated immune signalling networks enable them to mount effective defences
- The phyotohormone, jasmonic acid and its derivatives are key regulators in this network
  - Usually in response to insect, infection and necrotrophic pathogen attacks
- Enhanced JA production mediates large-scale reprogramming of the plant's transcriptome
- This is largely influenced by the antagonistic or synergistic action of other hormones produced during parasitic interactions such as SA, ethylene (ET) or abscisic acid (ABA)
- The JA network coordinates the production of a braod range of defence related proteins and secondary metabolites
  - The composition of which is adapated to the environmental context and the nature of the initial JA inducing situation

4 Moved to Next week!

4.1 TODO Understanding Plant Immunity as a surveillance system to detect invasion [2]

4.1.1 Intro

- Crop production needs doubled by 2050 by some current estimates
- A lot of work has been done on conceptual models for plant pathogen systems

  - Details in the models vary
  - Though they all agree in the observation that plants mainly rely on innate immune systems that are largely controlled by encoded receptors that identify invasion
- It's important to note that models:
  1. Are generalisations and therefore incomplete
  2. Increasing the details of a model decreases its general applicability
  3. Multiple models can be used to explain the same phenomenon
  4. All models should be continually challenged via experimentation to advance scientific knowledge
- Models are full of limitations and narrowly define molecular plant-invader interactions
  - Additionally do not integrate experimental data from diverse systems
- Introduces the *Invasion Model*

4.1.2 Advances in explaining the plant immune system

- Talks about gene-for-gene hypothesis
- And how other research suggests "general elicitors" from microbes which could **not** be used to determine race specificity or were detected by multiple plant species
  - In contrast to pathogen Avrs that induced responses only on particular varieties of a host species
  - These seeminly different observations were regarded as disparae phenomena
  - The identification and characterisation of general elicitors and their receptors in vertebrate immunity helped to further refine the concept in plant immunity
- Charles Janeway postulated the concept of conserved microbial ligands for innate immunity to account for lapses in the conceptual model of vertebrate immunity
  - He reasoned that microbes possess pathogen-associated molecular patterns (PAMPs)
  - That are recognised by host patter recognition receptors (PRRs) as nonself
  - He anticipated that PRRs perceive microbe-derived conserved general structural patters that are critical for the organism and require significant changes to avoid recognition
- PAMP is noted to be a misnomer, as it often concerns molecules present in both pathogenic and nonpathogenic organisms
  - Hence why we now use the term MAMPs (microbe-associated molecular pattern)
- Talks about zigzag model (see figure in key words/phrases)

4.1.3 Limitations and incongruities in the MAMP-effector dichotomy

- Continued research into plant microbe-interactions have identified a ...

4.2 TODO A review on Jasmonic acid and its derivatives [3]

4.3 TODO Necrotrophic Pathogens Use the SA Signaling Pathway to Promote Disease Development in Tomato [4]

4.4 TODO A single fungal MAP kinase controls plant cell-to-cell invasion by the rice blast fungus [5]

5 Key words/phrases

5.1 Jasmonic Acid

- Is an organic compound found in plants
- Is biosynthesized from linolenic acid by the octadecanoid pathway
- The major function of JA is regulation responses to abiotic and biotic stresses
  - In addition to growth and development
- Regulating growth includes:
  - Senescence
  - Tendril coiling
  - Flower development
  - Leaf abscission (natural detachment, e.g. dead leaves and ripe fruit)
- JA has been considered as a seed treatment in order to stimulate natural anti-pest defences

5.1.1 Relationship with SA

- Due to the antagonistic relationship between SA and JA, in some plant species it may result in increased susceptibility to viral agents and other pathogens
- Some pests, **through unknown mechanisms**, are able to abuse the relationship with the SA pathway. This allows for increased levels of SA resulting in the depression of JA synthesis
- Though thanks to the NPR1 gene, the levels aren't significantly reduced

5.2 Phytohormone

- Fancy way of saying: Plant hormone
- Is a signalling molecule
- Occur in low concentrations
- Exert strong control over plant development
- Can either act locally or in a more distant part of the plant
- Unlike animals, each plant cell is capable of producing hormones

- **some** phytohormones also occur in microorganisms, such as unicellular fungi and bacteria
  - However, these cases do not play a hormonal role and can be better regarded as secondary metabolites

5.2.1 Phyto -

- word-forming element meaning "plant," from Greek phyton "plant," literally "that which has grown," from phyein "to grow"

5.3 Necrotrophic

- A parasitic organism that kills the cells of its host an then feeds

5.4 Metabolites

- A metabolite is the intermediate end product of metabolism
- It is usually restricted to small molecules
- They have various functions
  - Including: fuel; structure; signalling; stimulatory and inhibitory effects on enzymes; defence; interactions with other organisms
- Examples include: ethanol; isoascorbic acid; glycerol

5.4.1 The metabolome

- Refers to a the complete set of small-molecule chemicsals found within a biological sample
- The sample can be a cell, a cellular organelle, a tissue extract or an entire organism

5.5 C-Terminal

- The c-terminus (also known as the carboxyl-terminus, C-Terminal) is the end of an amino acid chain
  - Protein or polypeptide
- When a protein is translated from messenger RNA it is created fron N-terminus to C-terminus
- The convention for writing peptide sequences is to put the c-terminal end on the right and write the sequence from N-C-terminus

5.6 CPG Site

A region of DNA where cytosine and guanine appear consecutively

5.7 Gene Promoters

- https://www.addgene.org/mol-bio-reference/promoter-background/
- For transcription to take place, the enzyme that synthesises RNA, known as polymerase, must attach to the DNA near a gene
- Promoters contain specific DNA sequences such as *response elements* that provide a secure initial binding site for RNA-P, as well as proteins called transcription factors that recruit RNA-P
- These TFs have specific activator or repressor sequences of corresponding nucleotides that attach to specific promoters and regulate gene expression

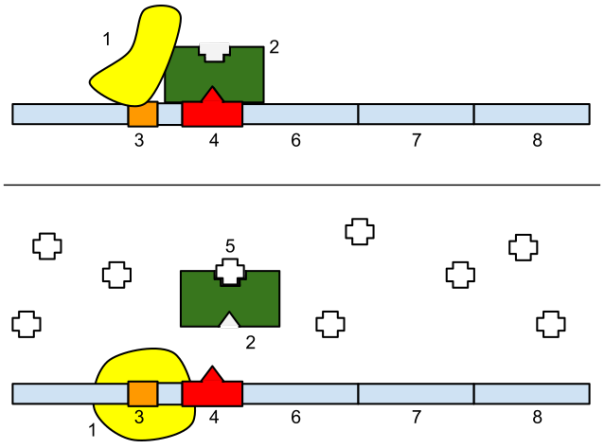

Figure 1: **1**: RNA Polymerase, **2**: Repressor, **3**: Promoter, **4**: Operator, **5**: Lactose, **6**: lacZ, **7**: lacY, **8**: lacA. **Top**: The gene is essenetially turned off. There is no lactose to inhibit the repressor, so the repressor binds to the operator, which obstructs the RNA polymerase from binding to the promoter and making lactase. **Bottom**

5.7.1 In bacteria

- The promoter is recognised by RNA-P and an associated sigma factor
- Which in turn are often brought to the promoter DNA by an activator protein's binding to its own DNA binding site, nearby

5.7.2 In eukaryotes

- The process is more complicated
- At least seven different factors are necessary for the binding of an RNA-P II to the promoter

1. RNA polymerase II
    - https://www.wikiwand.com/en/RNA_polymerase_II
    - Is a multiprotein complex.
    - Is one of the three RNAP enzymes found in the nucleus of eukaryotic cells
    - Catalyses the transcription of DNA to begin mRNA, snRNA and microRNA processes.

5.7.3 Sigma Factor

- Is a protein needed only for initiation of transcription
- It is a bacterial transcription initiation factor that enables specific binding of RNA polymerase to gene promoters
- It is homologous to archaeal transcription factor B and to eukaryotic TFIIB
- The specific sigma factor used to initiate transcription of a given gene will vary, depending on the gene
  - and the environmental signals needed to initiate transcription of that gene

1. TFIIB
   - Transcription factor II B is a general transcription factor that is involved in the formation of the RNA polymerase
   - It aids in stimulation transcription initiation
   - Recruits RNA polymerase II and other TFs

5.7.4 Response elements

- Are short sequences of DNA within a gene promoter region
- They are able to bind specific transcription factors
- Can regulate transcription of genes
- Under conditions of stress, a transcription activator binds to the response element and stimulates transcription
- If the same response element sequence is located in the control regions of different genes, then these genes will be activated by the same stimuli
- Thus producing a coordinated response

5.8 The RNA team

5.8.1 mRNA

- Messenger RNA
- Convey genetic information to the ribosome from DNA
- They specify the amino acid sequence of the protein products
- RNA polymerase transcribes primary transcript mRNA (known as Pre-mRNA) into mature mRNA
- This matured mRNA is translated into a polymer of amino acids

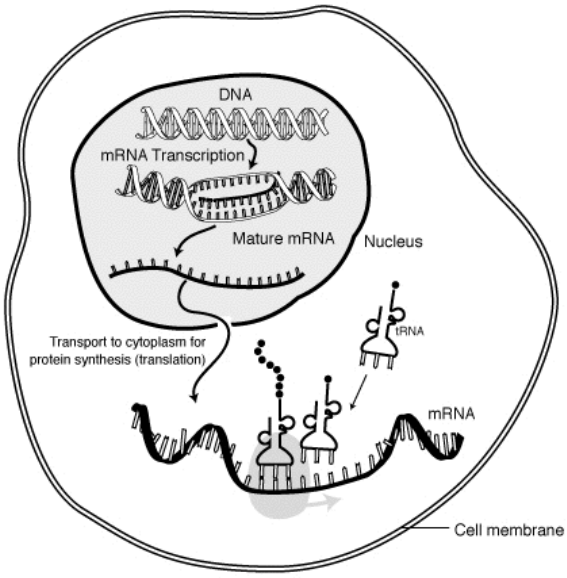

Figure 2: mRNA

5.8.2 snRNA

- http://bioscience.jbpub.com/cells/MBIO5245.aspx
- Small nuclear RNA is one of the many small RNA species confined to the nucleus
- several of the snRNAs are involved in splicing or other RNA processing reactions

5.8.3 microRNA

- Is a small non-coding RNA molecule (about 22 nucleotides) found in plants, animals and some viruses
- Functions in RNA silencing and post-transcriptional regulation of gene expression
- microRNAs function via base-pairing with complementary sequences iwhtin mRNA molecules
- As a result these mRNA molecules are silenced by one of the following processes:
  1. Cleavage of the mRNA strand into two pieces
  2. Destabilisation of the mRNA through shortening of its poly(A) tail
  3. Less efficient translation of the mRNA into proteins by ribosomes
- Often abbreviated to miRNA

5.9 Binding site

- Is a region on a protein or piece of DNA or RNA to which ligands may form a chemical bond

5.10 Gene Repressor

- Is a DNA or RNA-binding protein that inhibits the expression of one or more genes by binding to the operator or associated silencers
- A DNA-binding repressor blocks the attachment of RNA polymerase to the promoter
  - Thus preventing transcription of genes to mRNA
- The blocking of expression is called repression

5.11 Gene Operator

- An operon is a functioning unit of DNA containing a cluster of genes under the control of a single promoter
- The genes are transcribed together into an mRNA strand and either translated together in the cytoplasm or undergo splicing to create monosistronic mRNAs that are translated separately

5.12 Polymerase

- Is an enzyme that synthesises long chains of polymers or nucleic acid s
- DNA and RNA polymerase are used to assemble DNA and RNA molecules, respectively

5.13 lac operon

- Is an operon required for the transport and metabolism of lactose
- Many entric bacteria use is
- Although glucose is the preferred carbon source for most bacteria, the lac operon allows for the effective digestion of lactose when glucose is not available
- Gene regulation of the lac operon was the first genetic regulatory mechanism to be understood clearly
- So it's an example for prokaryotic gene regulation
  - *which explains why it's in my notes!*

5.13.1 Lactase

- Is an enzyme that breaks-down milk, only needed here to help explain lac operon figure!

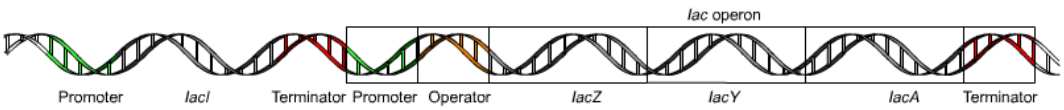

Figure 3: lac operon in more detail than previous fig

5.14 Zigzag model

The zigzag model discriminates four phases of the plant immune system that determine the plant-microbe interactions manifested today. (Following text is almost verbatim from [2])

1. In the first phase, plants recognise MAMPs by cell surface-localised PRRs (pattern recognition receptor), leading to broad spectrum resistance
   - This is termed MTI (Microbe-asscociated molecular-patterns-triggered immunity)
2. Next, microbial-produced molecules, termed effectors, enable successful pathogens to overcome MTI
   - This results in effector-triggered susceptibility (ETS)
3. Subsequently, these effectors may be recognised by intra-cellular receptors (R proteins)
   - This activates ETI (effector-triggered immunity)
4. In turn, the microbe may evade ETI and restore ETS

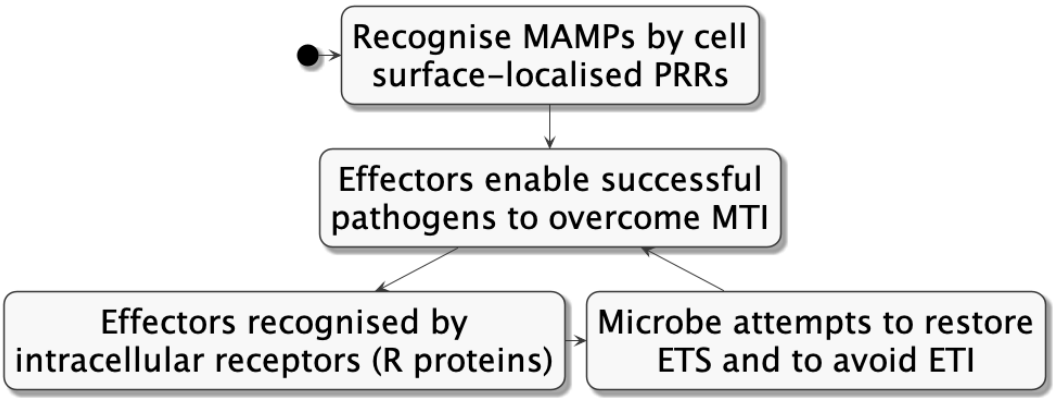

Figure 4: The zigzag model in plant immune system

6 Interesting

6.1 **IDEA** Read up on the links between jasmonic and salicylic acid

7 Last week leftover

7.1 Transcriptome

- Is the set of all RNA molecules in one cell or a population of cells
- Sometimes it is used to refer to all RNAs or just mRNA, depending on the experiment

7.2 Kinase

- A kinase is an enzyme that catalyses the transfer of phosphate groups from high-energy, phosphate-donating molecules to specific substrates.

- This process is known as phosphorylation
  - Where the substrate gains a phosphate group
  - And the high-energy ATP molecule donates a phosphate group
  - Conversely, it is referred to as dephosphorylation when the phosphorylated substrate donates a phosphate group and ADP gains a phosphate group

7.3 Phosphorylation

- In chemistry, phosphorylation of a molecule is the attachment of a phosphoryl group. Together with its counterpart, dephosphorylation, it is critical for many cellular processes in biology.

7.4 ATP

- Adenosine triphosphate, energy providing molecule used in active transport!

7.5 ADP

- Adenosine diphosphate also known as adenosine pyrophosphate (APP), is an important organic compound in metabolism and is essential to the flow of energy in living cells. ADP consists of three important structural components: a sugar backbone attached to adenine and two phosphate groups bonded to the 5 carbon atom of ribose. (verbatim from wikipedia)

7.6 DNA methylation

- Is the process by which methyl groups are added to the DNA molecule
- is an epigenetic mechanism (Says so here)
- Methylation can change the activity of a DNA segment without changing the sequence
  - Can act like a switch?
- When located in a gene promoter, DNA methylation typically acts to repress gene transcription
- It is essential for normal development and is associated with a number of key processes including:
  - Genomic imprinting; X-chromosome inactivation; Ageing
- Two of DNA's four bases C(ytosine) and A(denine) an be methylated
- Cytosine methylation is widespread in both eukaryotes and prokaryotes
  - Even though the rate of cytosine DNA methylation can differ greatly between species

Arabidopsis thaliana	8%
Drosophila	0.03%

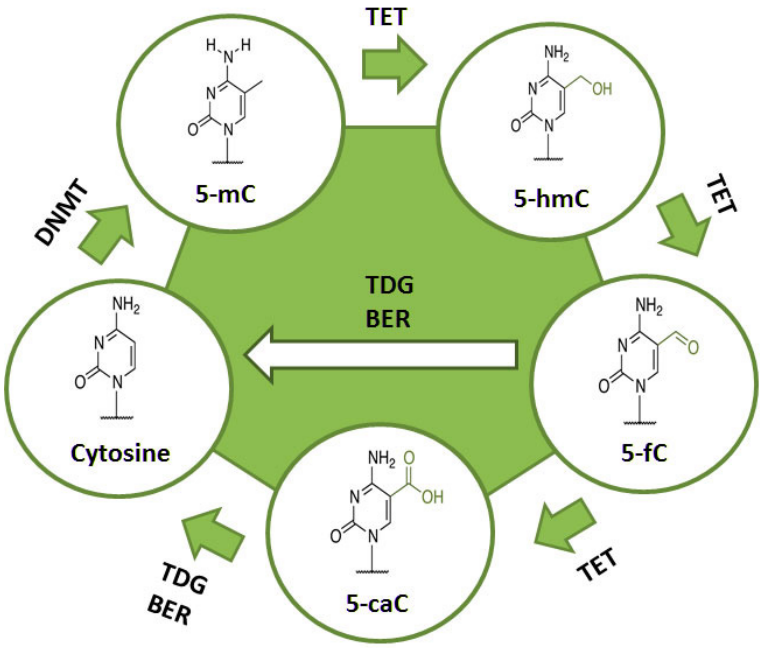

Figure 5: Cytosine's methylation and demethylation process

7.6.1 *What is epigenetics* website

- https://www.whatisepigenetics.com/dna-methylation/
- Addition of a methyl group is often (always?) covalent
- The methylcytosine is sometimes, informally, referred to as the "fifth" base of DNA

7.6.2 A Methyl group

- Is an alkyl dervied from emthane, containing one carbon atom bonded to three hydrogen atoms
- It is a very stable compound
- Usually part of a larger molecule, though can exist independently

7.6.3 DNA Aceltylation

- Is an essential part of gene regulation
- Aceltylation is the process where an acetyl functional group is transferred from one molecule to another
  - Deacetylation is the reverse, where an acetyl group is removed from a molecule

7.6.4 Pathways

- A pathway just refers to a sequence of interactions amongst molecules in a cell that leads to a product or change in a cell

- Such a pathway can trigger the assembly of new molecules such as fat or protein
- Pathways may also turn genes on and off, or make a cell move
- Common pathways are invovled in metabolism and regulation of gene expression as well as transmission of signals
- Gene regulatory network is an example of the genetics pathway

7.6.5 Induced systemic resistance (ISR)

- Is an activated resistance process that is activated by biological or abiotic factors
- Is dependant on the physical or chemical barrier of the host plant
- And its action is characterised by no direct killing or inhibition of the pathogen, but through the induction of plant disease resistance to disease prevention and control purposes

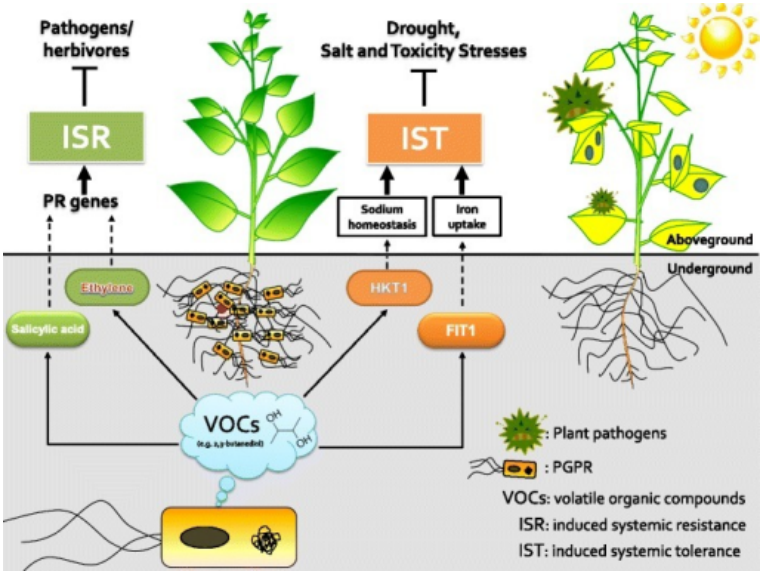

Figure 6: ISR

7.6.6 Systemic acquired resistance (SAR)

- Is a whole plant resistance response that occurs following an earlier localised exposure to a pathogen
- SAR is analogous to the innate immune system found in animals
- There is evidence that SAAR in plants and innate immunity in animals may be evolutionary conserved

8 Misc Questions

- When is a cell "dead". Or rather what makes one such
- Where is the instruction-set for methylation held?

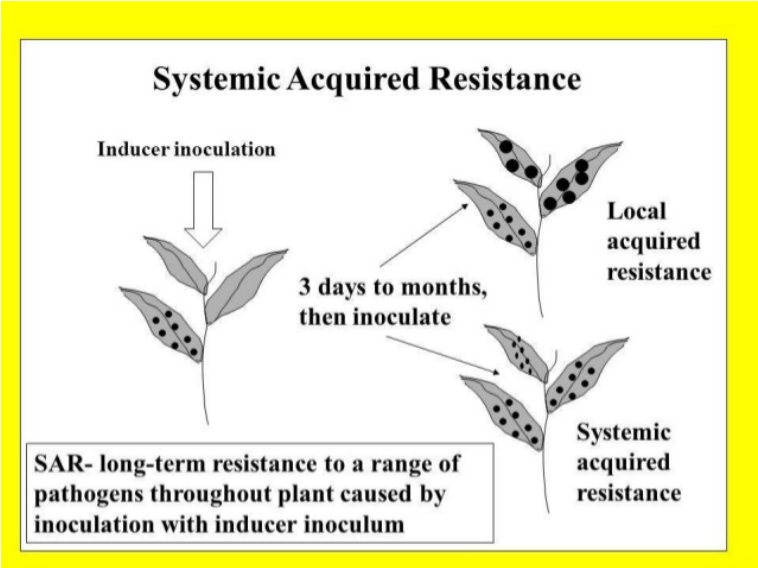

Figure 7: SAR

- PD, what happens to them when cells divide
  - Follow up, what happens when certain paths become deprecated because of connection to previous "major" paths?
- PD, is signalling directed, is there a feedback mechanism? i.e. radio towers
- Pathogens which feed on their hosts:
  1. Why don't they let the cells continue to function and just "consume a litte" in a pesudo-perpetual relationship
  2. What specifically are they "feeding" on inside a cell
  3. Do they consider the cell's function before attack, and sometimes "disappointed" with the molecules inside?
- Do PD of different cells have different behaviour?
  - if you take root PD and leaf and do same attack, same action?
    - If not why
    - Can you trick them to if not?
    - Do they change over time if isolated
  - Something something delta resilience, are older PD more conserved i.e. do they lack basic economic principals?

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