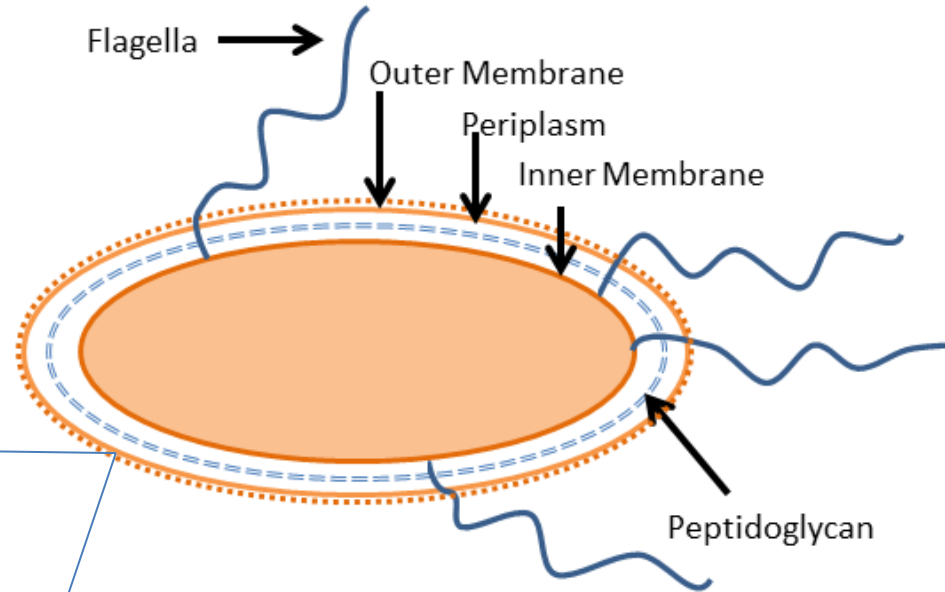
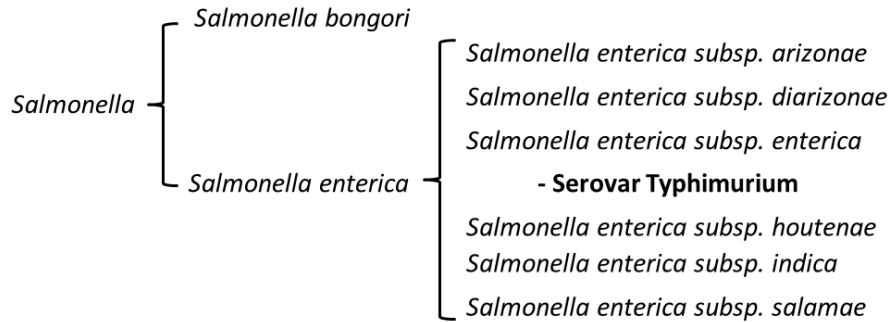


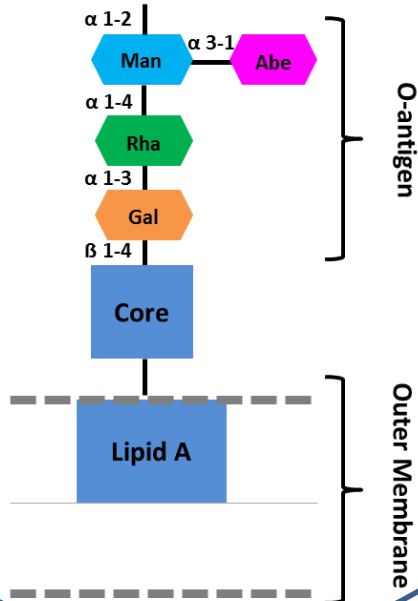
O-antigen modifying enzymes in the bacterial pathogen *Salmonella*

Caroline Pearson - September 2016

Salmonella LPS O-antigen



Lipopolysaccharide



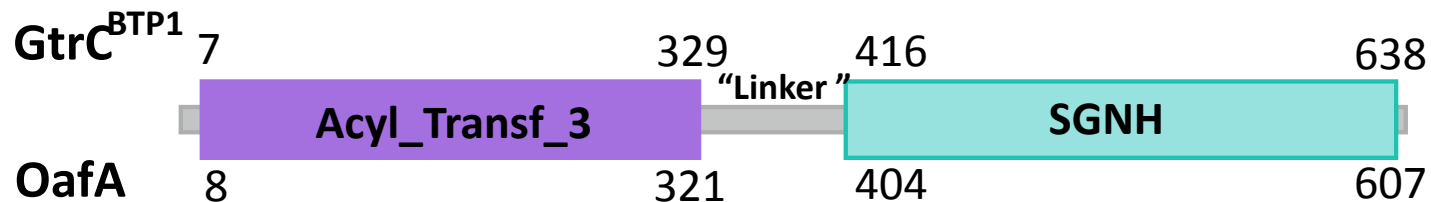
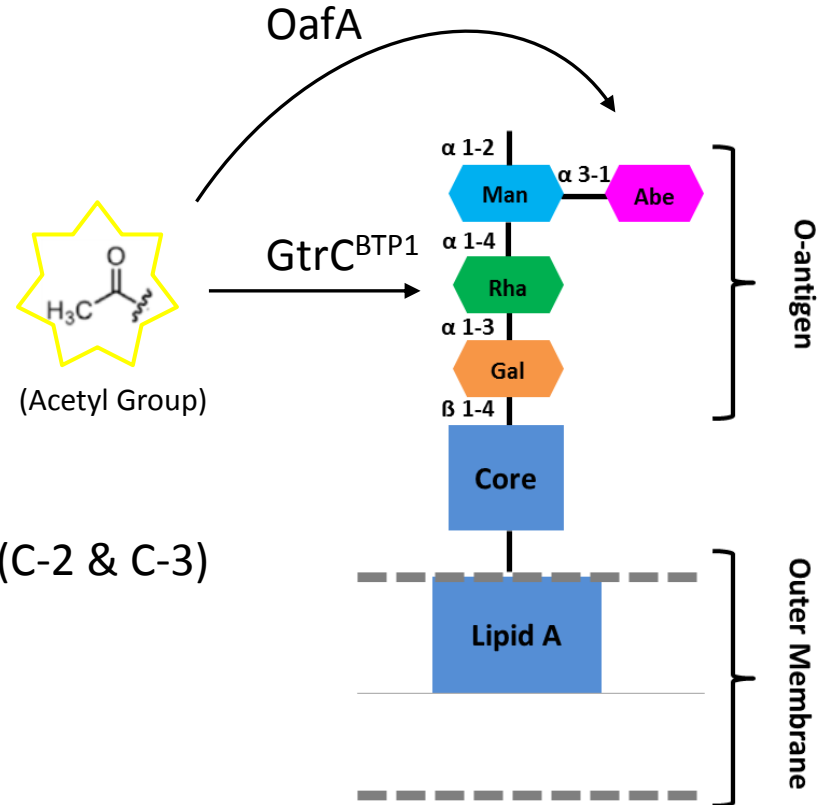
OafA and GtrC^{BTP1} Acetylate LPS O-antigen

OafA

- Found in multiple Typhimurium strains
- Acetylates the abequose residue (C-2)
- Confers O:5 serotype

GtrC^{BTP1}

- Found in BTP1 prophage of ST313 strains (C-2 & C-3)
- Acetylates rhamnose residue



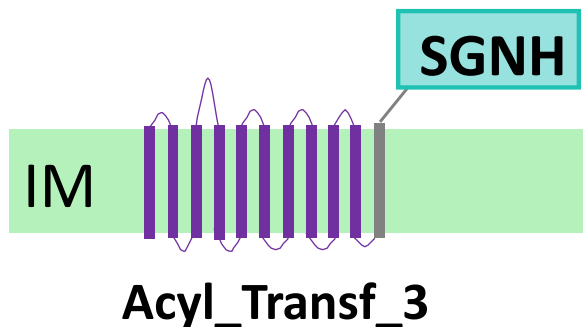
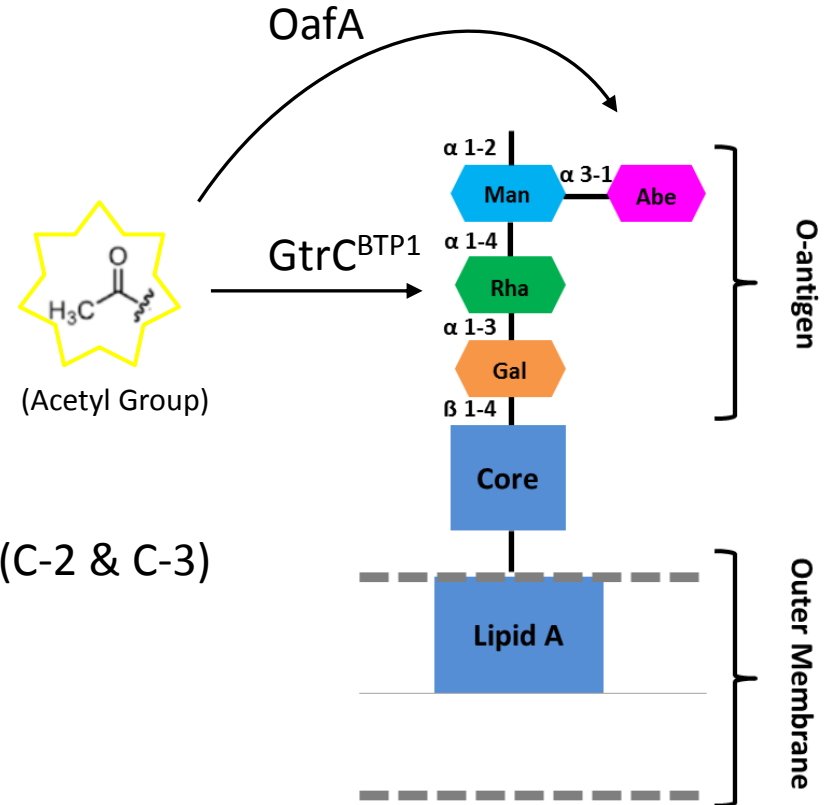
OafA and GtrC^{BTP1} Acetylate LPS O-antigen

OafA

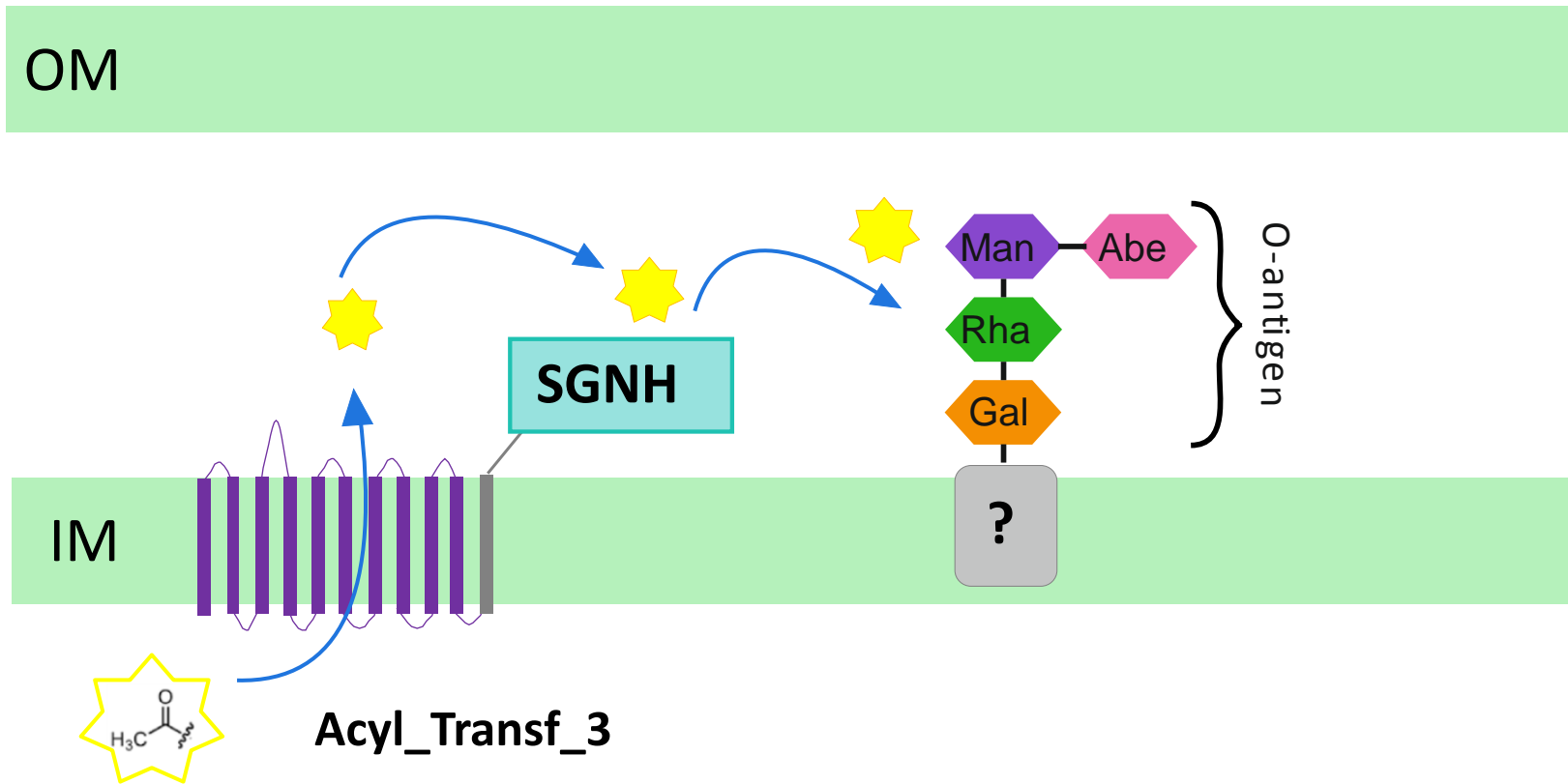
- Found in multiple Typhimurium strains
- Acetylates the abequeose residue (C-2)
- Confers O:5 serotype

GtrC^{BTP1}

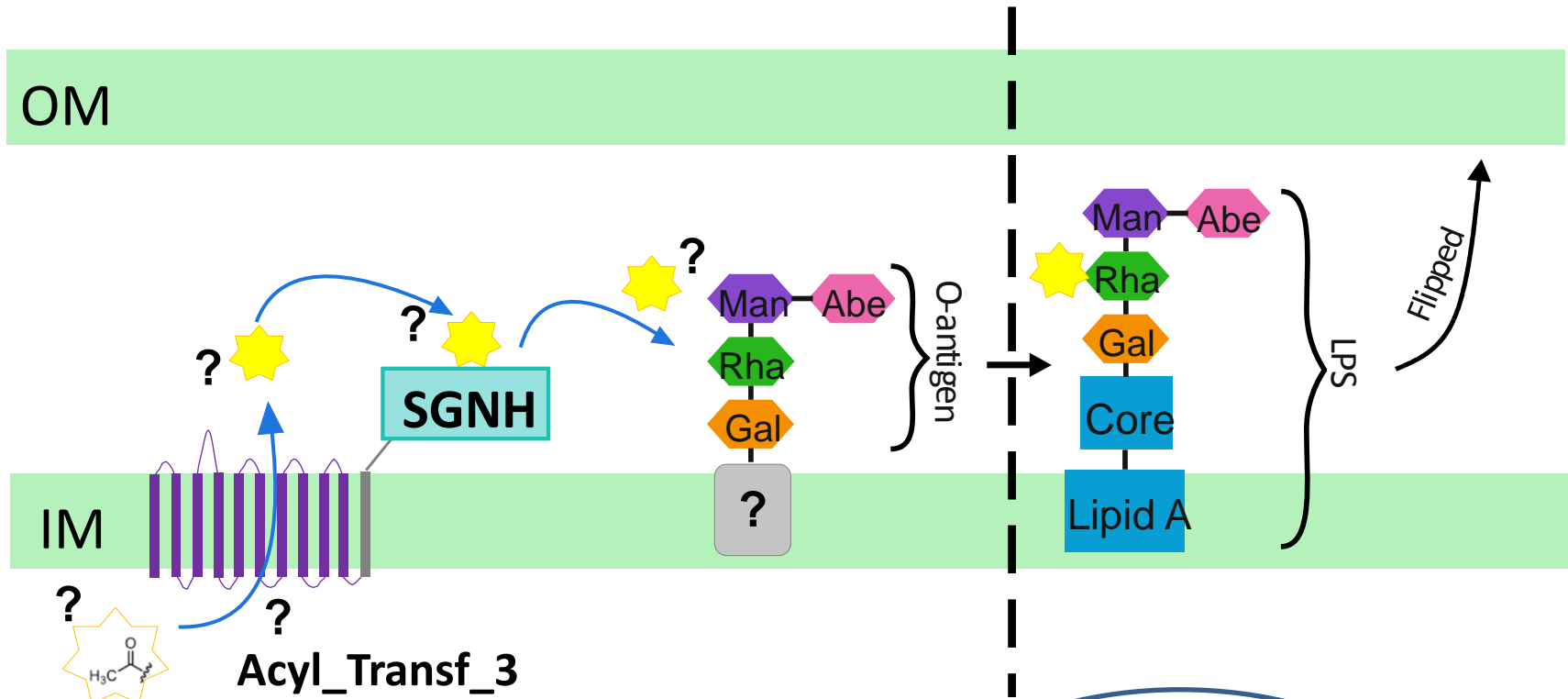
- Found in BTP1 prophage of ST313 strains (C-2 & C-3)
- Acetylates rhamnose residue



OafA and GtrC^{BTP1} Mechanism of Action



What don't we know?



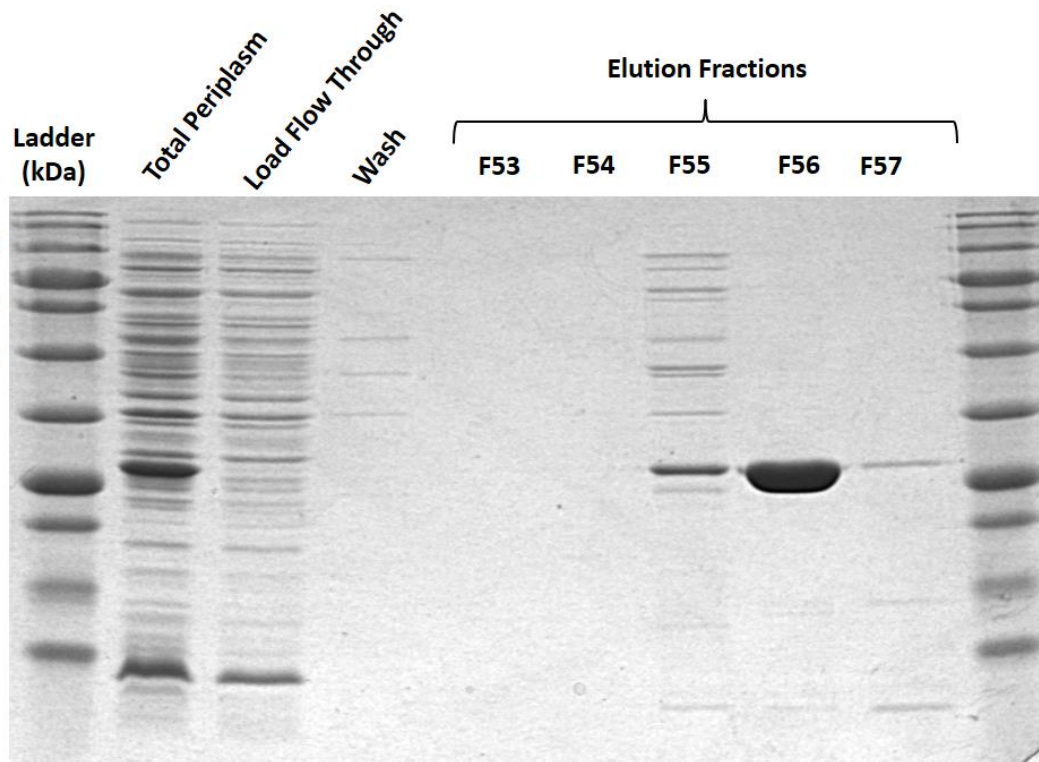
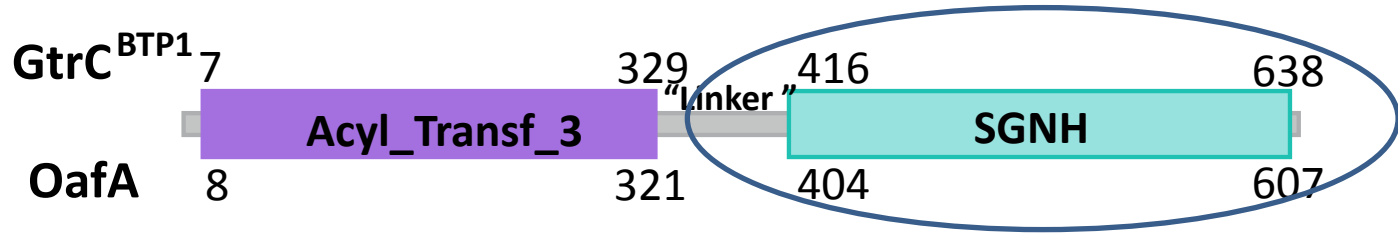
Acyltransferase 3 domain:

- Acetyl group donor
- Acetyl group binding site in Acyl_Transf_3
- Transfer of acetyl group between domains

SGNH hydrolase domain:

- Acetyl group binding site
- LPS recognition site
- Mechanism of addition

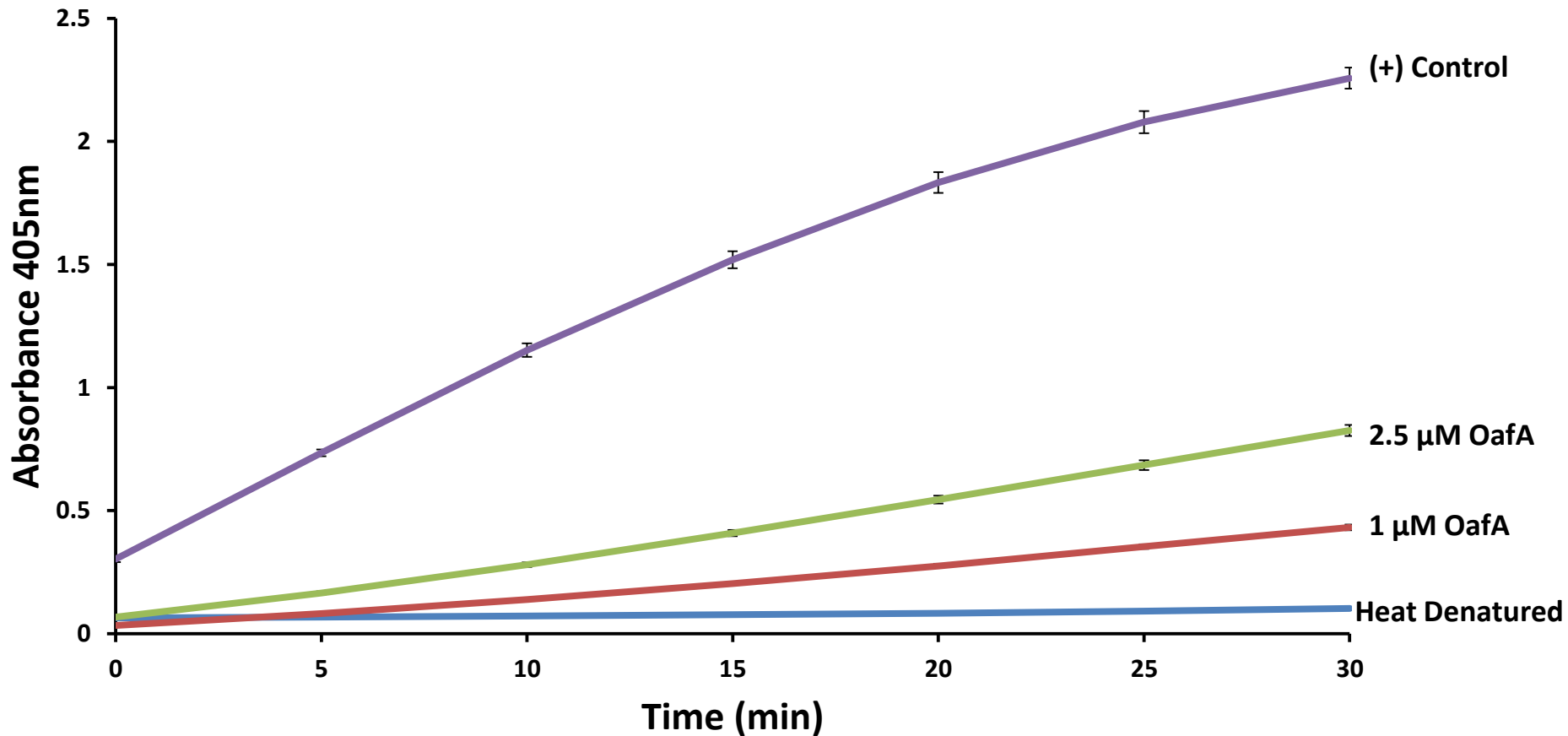
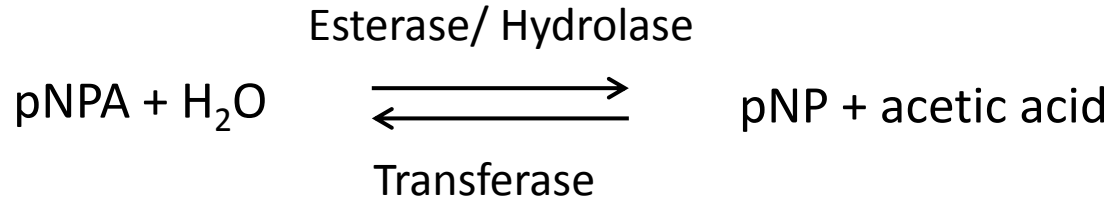
Expression and purification of C-terminal OafA



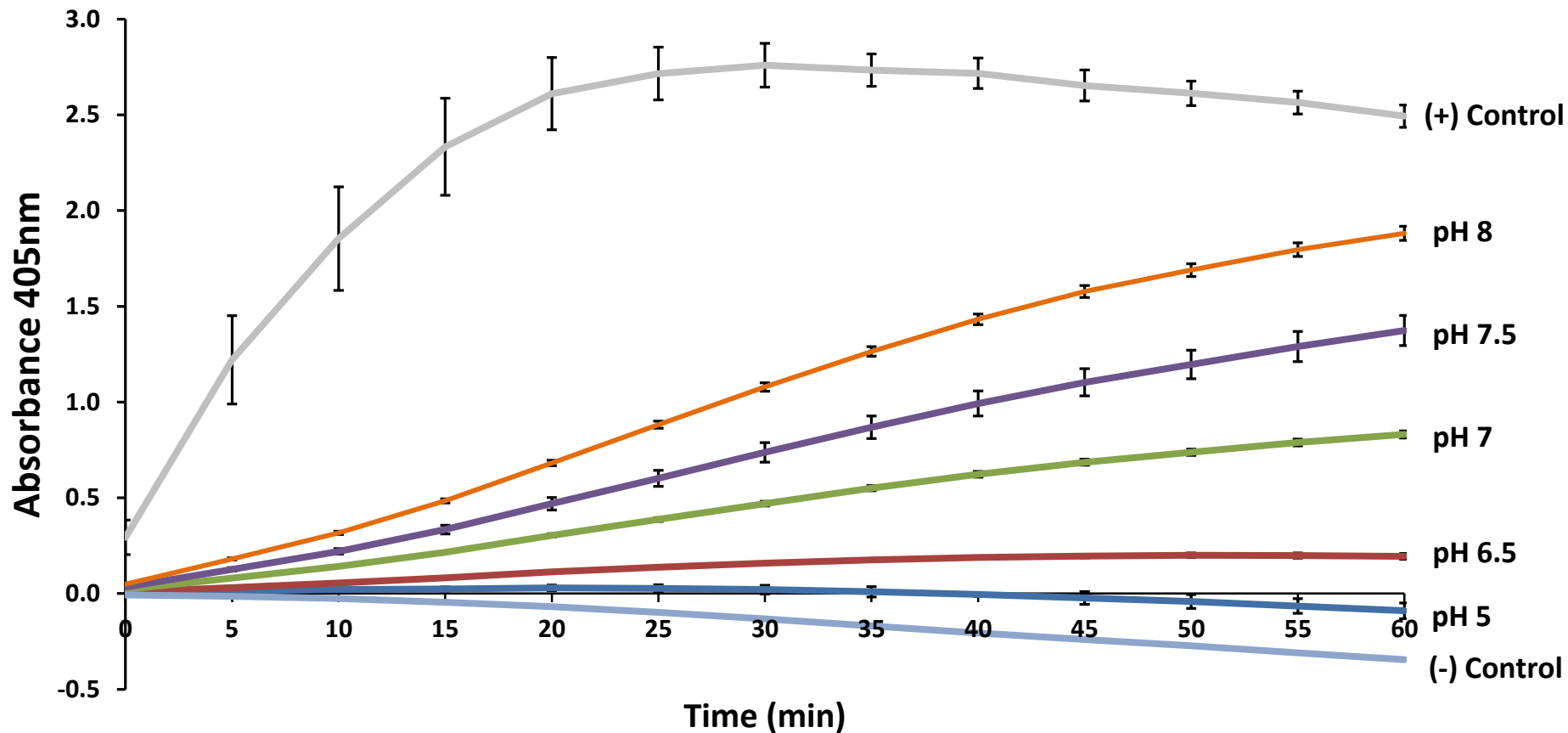
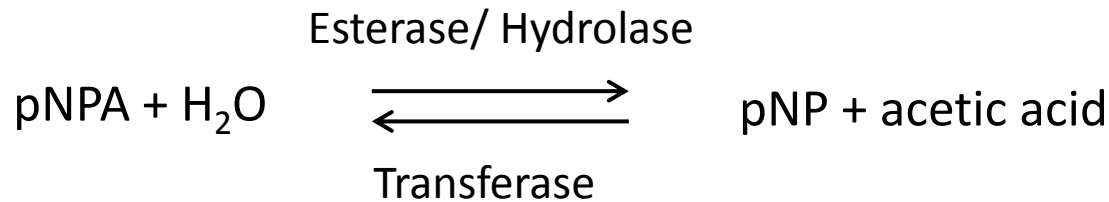
Periplasmic extraction

Nickel affinity purification

Acetyltransferase activity of OafA SGNH domain



Low pH inhibits catalytic activity of OafA SGNH domain



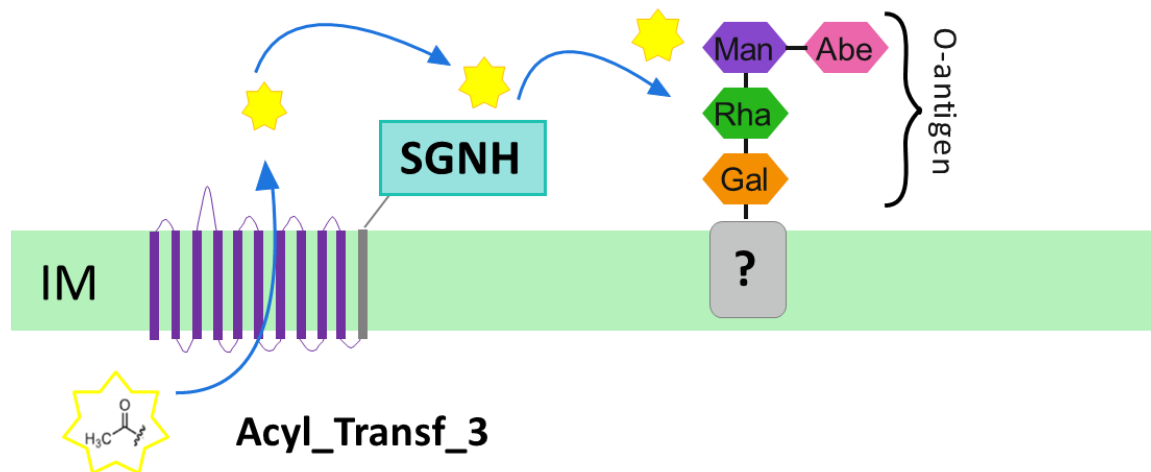
Where next?

***In vitro* analysis:**
NMR spectroscopy

***In vivo* functional analysis:**
Mutation analysis

Very long term goals:

1. Work out the mechanism of action of these proteins.
2. Apply this knowledge to antibiotics, vaccines, industrial processes.



Thank you

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Dr Marjan Van Der Woude

Thesis advisory panel:

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Dr Michael Plevin

All members of the Thomas
and Van Der Woude groups.

