Master thesis projects in plant pathology for 2022

Below are suggestions for master thesis projects in plant pathology for 2022. However, other ideas for thesis work may be discussed further with one of the following teachers:

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1 Virus	**Tobamovirus i vektshuskulturer**
	Tobamoviruses in greenhouse crops

Viruses represent one of the most important threats to agriculture and horticulture. Several viral families include very stable and highly infective pathogens. Especially viruses in the genus *Tobamovirus* can cause unpredictable loss to tomato and cucumber production in Norway. In tomato, *Tomato brown rugose fruit virus* and other resistance breaking viruses and strains are posing a threat to the tomato industry. In cucumber, *Cucumber green mottle mosaic virus* has been a growing problem for the last decade.

Tobamovirus in tomato gives uneven ripening, necrosis and chlorosis.

The viral particles of tobamoviruses are extremely stable and infectious. They can be preserved in seeds for several years. In addition, tobamoviruses can survive outside of the host on inert and biological surfaces, as well as in water and soil for months or even years, without losing their virulence. Due to import of tomato and cucumber product as fresh vegetables and other products, we could be exposed to these viruses in our daily life, and infective virus particles may spread further into sewage, drainage and environmental waters which could be an unpredictable threat for plant health and food production in Norway.

Several aspects of tobamovirus identification, testing, epidemiology and control needs further investigations.

The master thesis project needs to be discussed and defined in cooperation with the student. Please contact Dag-Ragnar Blystad (dag-ragnar.blystad@nibio.no) or Zhibo Hamborg (zhibo.hamborg@nibio.no) for more information if you are interested in this topic.

2 Virus	**Virus i jordbær – diagnostikk og kartlegging**
	Strawberry viruses – diagnosis and survey

One of the causal agents, which can reduce fruit quality and quantity are viruses. Viral diseases can reduce strawberry yield by 20-40%, sometimes up to 70-80% or the plants can even die. The use of classical molecular biological methods (grafting of indicator clones, transmission electron microscopy, PCR) suggested that the most economically important strawberry viruses are strawberry mild yellow edge virus (SMYEV), strawberry crinkle virus (SCV), strawberry mottle virus (SMoV) and strawberry vein banding virus (SVBV), especially when it occurs in mixed infections. A new study showed that infection with only SMYEV caused a reduction in the number and size of fruits ranging from 28% to 63% compared to a healthy control.

Single virus infection usually does not show any specific symptoms on these plants, but only occurs when mixed viral infections are present. Therefore, often the most effective spreaders of viruses in strawberry are growers themselves, who unknowingly distribute diseased seedlings. Detection of viral pathogens and subsequent production of recovered seedlings is a very important prevention for the cultivation of healthy cultures.

We would like to develop a more cost-efficient and sensitive method to detect strawberry viruses with q-PCR in a new KAPPA (Norway and Czech Republic collaboration) project starting from 2021. In addition, survey of strawberry viruses in Norway and identification of unknown viruses that can infect strawberry cultures will also be studied in this project.

Strawberry crickle virus showing symptoms in a sensitive indcactor host.

Please contact Dag-Ragnar Blystad (dag-ragnar.blystad@nibio.no) or Zhibo Hamborg (zhibo.hamborg@nibio.no) for more information if you want to study strawberry viruses in your master thesis.

3 Virus	**Potetbanken og genressurser**
	The potato tissue culture bank and genetic resources

Potetbanken hos NIBIO er grunnleggende for et friskt utgangsmateriale av potet i Norge.

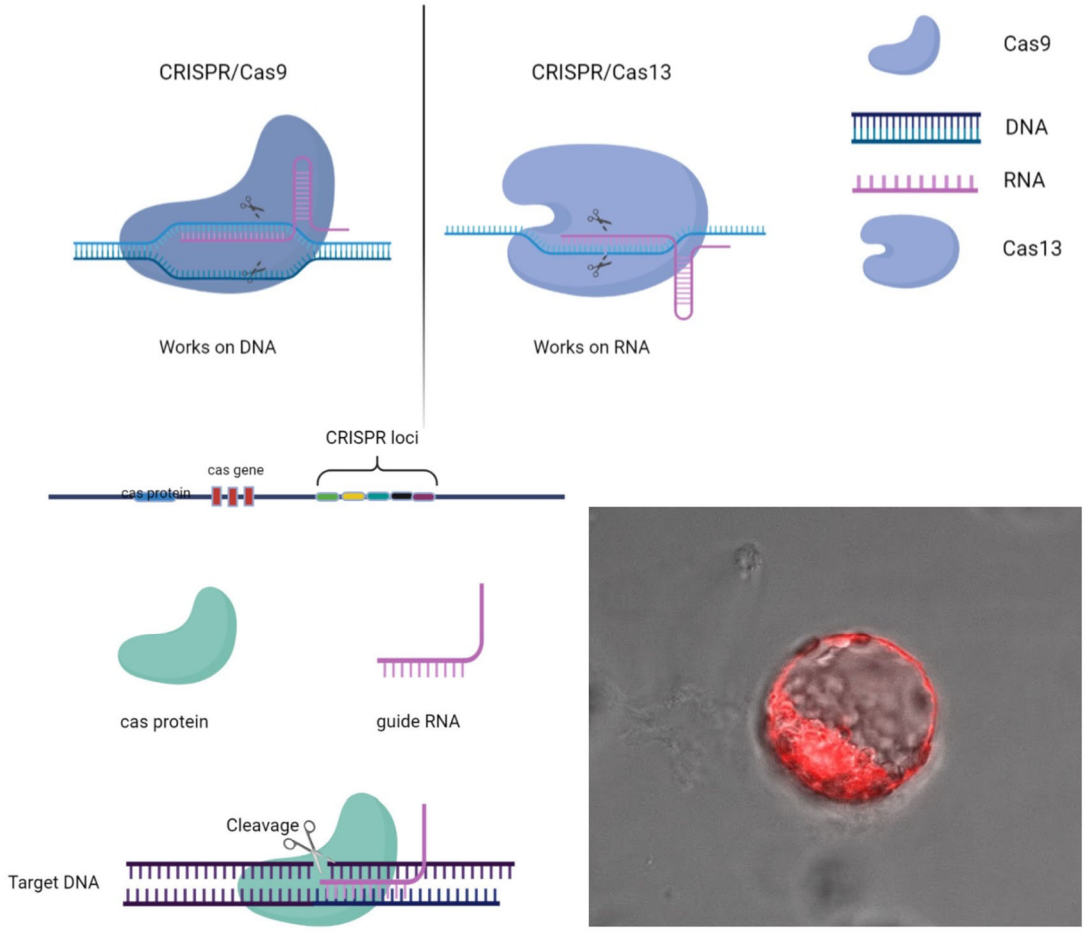
NIBIO Divisjon for bioteknologi og plantehelse har en samling av friskt, virusfritt kjernemateriale av mandatsorter av potet (Potetbanken) og vegetativt formert løk. Kompetansen til å lage og vedlikeholde et slikt friskt utgangsmateriale er av avgjørende betydning for plantehelsen til klonfomerte vekster. NIBIO har gjennom dette arbeidet viktige oppgaver knyttet til mat- og dyrkingssikkerhet, bevaring av genressurser og innovasjon.

En masteroppgave innenfor dette området vil kunne defineres i flere mulige retninger:

- Rensing av «nye», gamle sorter som bør inn i Potetbanken / klonarkivet. Dette vil være en oppgave som vil fokusere på vevskultur og virustesting med ELISA-metoden, PCR og testplanter
- Beskrive og sammenligne gamle sorter som vi har i banken for om mulig å avsløre dubletter som egentlig er samme sort, men med forskjellige navn. Dette arbeidet kan ha flere hovedretninger:
    - Genetisk analyse som utføres i samarbeid med andre kompetansemiljøer i NIBIO.
    - Dyrkingsforsøk for å studere og beskrive utvikling og utseende med beskrivelser og fotografering. Dette kan innbefatte studier av utvalgte sorters dyrkingsverdi og matkvalitet.
    - Sammenstilling av gamle data om sortene, dyrkingsverdi og bruk.

Ta kontakt med Dag-Ragnar Blystad (dag-ragnar.blystad@nibio.no) dersom du er interessert i en oppgave innen dette temaet. Zhibo Hamborg (zhibo.hamborg@nibio.no) og Morten Rasmussen (morten.rasmussen@nibio.no) vil være i veilederteamet. Oppgaven blir definert nærmere i samtale med studenten og kan involvere flere medveiledere etter hva som er naturlig ut fra vinklingen som defineres i samtale med studenten.

4 Virus	**Avansert bruk av CRISPR-Cas-systemene for RNA undertrykking**
	Advanced uses of the CRISPR-Cas-system for RNA suppression



Summary

CRISPR Cas9 has caught the attention of many scientists during the last 5 years. There are many groups doing research on CRISPR Cas9 in plant biology. The focus of these groups is mainly on introducing new trains to plants by DNA modifications and to diversity the CRISPR toolbox for enhanced gene editing. My lab is focusing on CRISPR in a very different way. In my lab we are exploring a different Cas protein (Cas13a and the orthologues Cas13b and Cas13d) which does not target DNA, but rather RNA. In addition, my lab is not focusing on introducing traits or modifying the plant genome, but rather on using Cas13 as a method to localize plant viruses within the cells and to kill them. In addition, we are also studying the ability of Cas13 to be fused with specific viral proteins to induce RNA silencing. The thesis will be focused on characterizing the activities, cellular localization, and potential uses Cas13 proteins. This type of study involves good amount of gene cloning and molecular techniques, the use of delivery vectors, plant cell transfections and cellular localization studies. The student will gain competence in the following areas and techniques: Molecular biology, virology, gene technology, recombinant DNA techniques, plant genetics and plant pathology. If you are interested drop me a line.

Subject area

Molecular Biology, Virology, Cell Biology, Plant physiology, Plant-pathogen interactions

Language thesis (Norwegian and/or English)

Bachelor or Master thesis

Credits

60 credits

Project/company

NIBIO

Please contact

Carl.spetz@nibio.no Dag-ragnar.blystad@nibio.no

Our publications on CRISPR:

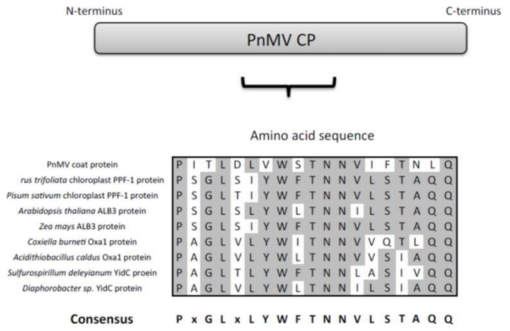
- Xu Z, Kuang Y, Ren B, Yan D, Yan F, **Spetz C**, Sun W, Wang G, Zhou X, Zhou H (2021). SpRY greatly expands the genome editing scope in rice with highly flexible PAM recognition. ***Genome Biol***. Jan 4;22(1):6. doi: 10.1186/s13059-020-02231-9. PMID: 33397431; PMCID: PMC7780387.

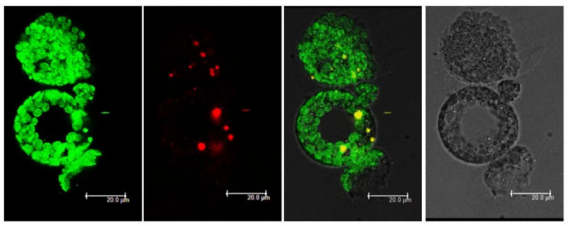
- Liu, L., Kuang, Y., Yan, F., Li, S., Ren, B., Gosavi, G., **Spetz, C**., Li, X., Wang, X., Zhou, X., & Zhou, H. (2020). Developing a novel artificial rice germplasm for dinitroaniline herbicide resistance by base editing of OsTubA2. ***Plant biotechnology journal***, 10.1111/pbi.13430. Advance online publication. https://doi.org/10.1111/pbi.13430

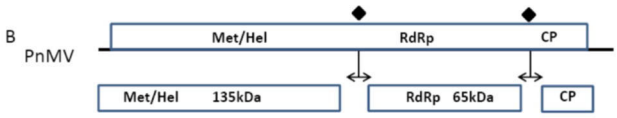
- Wang, M., Xu, Z., Gosavi, G., Ren, B., Cao, Y., Kuang, Y., Zhou, **C., Spetz**, C., Yan, F., Zhou, X., & Zhou, H. (2020). Targeted base editing in rice with CRISPR/ScCas9 system. ***Plant biotechnology journal***, 10.1111/pbi.13330. Advance online publication. https://doi.org/10.1111/pbi.13330

- Kuang, Y., Li, S., Ren, B., Yan, F., **Spetz, C**., Li, X., Zhou, X., & Zhou, H. (2020). Base-Editing-Mediated Artificial Evolution of OsALS1 In Planta to Develop Novel Herbicide-Tolerant Rice Germplasms. ***Molecular plant***, *13*(4), 565–572. https://doi.org/10.1016/j.molp.2020.01.010

5 Virus	**Dissekere virale replikasjonsmekanismer**
	Dissecting viral replication mechanisms







Summary

Replication of all positive-sense single-stranded RNA viruses occurs in specific structures in close association with cellular membranes. Targeting of the viral replication complex (RC) to the site of replication is mediated by the interaction of viral-encoded proteins and host factors. Electron microscope studies have shown that *Poinsettia mosaic virus* (PnMV, family *Tymoviridae*) infection is associated with the presence of vesicular structures in the chloroplasts, which indicates that the replication of PnMV might occur in association with chloroplast-derived membranes. Using computer assisted homology search, we have identified that the coat protein (CP) of PnMV shows similarity to membrane bound proteins and contains a conserved amino acid sequence motif found in members the Alb3/Oxa1/YidC protein family. This protein family is involved in the insertion of proteins into intracellular membranes. We hypothesize that the targeting of the PnMV RC to the chloroplast is mediated by viral-encoded CP. We plan to test this hypothesis carrying out co-localization studies using transient and stable expression of GFP-tagged viral proteins and confocal laser scanning microscopy. This type of study involves good amount of gene cloning and molecular techniques, the use of delivery vectors, plant cell transfections and cellular localization studies. The student will gain competence in the following areas and techniques: Molecular biology, virology, gene technology, recombinant DNA techniques, plant genetics and plant pathology.

Subject area

Molecular Biology, Virology, Cell Biology, Plant physiology, Plant-pathogen interactions

Language thesis

Norwegian and/or English

Bachelor or Master thesis

Credits

60 Credits

Project/company

NIBIO

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Our publications on Poinsettia mosaic virus

- Spetz C & Blystad D-R (2015) A Membrane-Binding Conserved Motif in the Coat Protein of PnMV Seems to Mediate Chloroplast Targeting. *Acta Horticulturae* 1072, 97-104.
- Spetz C, Clarke JL, Dees MW, Haugslien S, Moe R, & Blystad D-R. (2012) RNA silencing-based resistance in Nicotiana benthamiana Is transiently broken by graft inoculation of Poinsettia mosaic virus January 2012. *Acta horticulturae* 941(941):91-108
- Clarke, J.L., Spetz, C.J., Haugslien, S., Dees, M.W., Moe, R. & Blystad, D.-R. (2011). Production of transgenic Poinsettia with Resistance against Poinsettia mosaic virus (PNMV) using Agrobacterium- Mediated Transformation. Acta Horticulturae 901:87-93.
- Dees, M.W., Spetz, C.J.J. & Blystad, D.-R. (2011). First report of Tulip virus X (TVX) in Norway. Acta Horticulturae 901:215-222.
- Fløistad, E., Spetz, C. J.J. & Blystad, D.-R. (2011). Virus elimination and retention of free - branching in Poinsettia. Acta Horticulturae 901:207-213.
- Spetz, C., Moe, R. and Blystad, D-R. (2008). Symtomless infectious cDNA clone of Norwegian isolate of *Poinsettia mosaic virus*. *Archives of Virology* 153, 1347-1351
- Clarke, J.L., Spetz, C., Haugslien, S., Xing, S., Dees, M.D., Moe, R. and Blystad, D- R. (2008). Agrobacterium tumefaciens-mediated transformation of poinsettia, *Euphorbia pulcherrima*, with virus-derived hairpin RNA constructs confers resistance to Poinsettia mosaic virus. *Plant Cell Reports* 27, 1027-1038

6	Integrated management of Fusarium head blight in cereals

Fusarium head blight (FHB) is a widespread and destructive disease of cereals caused by a range of different fungal species within the genus *Fusarium* and *Microdochium*. Several *Fusarium* species can reduce grain quality due to the production of a range of toxic metabolites (mycotoxins) that have adverse effects on human and animal health. To reduce the risk of mycotoxin-contaminated grain lots to enter the food and feed chain, it is of importance to identify climatic and agricultural factors that influence Fusarium head blight infection and mycotoxin development in cereals. The aim of NIBIO's involvement and activities within this subject is to develop and disseminate knowledge about measures to reduce the risk of *Fusarium* and mycotoxins in cereals.

The effect of weather on development of *Fusarium* and mycotoxins

Weather factors such as rainfall and temperature in the critical period around flowering have a major impact on the development of *Fusarium* head blight (FHB) and mycotoxins in cereals. Forecasting models to predict development of mycotoxins in a specific field due to cultivation practise and weather are developed by NIBIO in collaboration with the Norwegian Extension Service.

The effect of agronomic practice on development of *Fusarium*

Deep tillage and crop rotation are among the agronomic practices considered to be of prime importance for combating *Fusarium* and the production of mycotoxins in cereals. NIBIO study the effect of field factors such as cultivar, tillage, preceding crop, soil humidity, and pesticide treatment on development of *Fusarium* and mycotoxins in cereals.

Resistance to *Fusarium* in Norwegian cereals:

No commercial cultivar of wheat, oat or barley displays absolute resistance to *Fusarium* infection, although cultivar differences exist. NIBIO perform field trials, greenhouse and laboratory experiments to study interactions between *Fusarium* spp. and cereal varieties.

Chemical and biological control of FHB and mycotoxins:

NIBIO carry out efficacy testing of fungicides on behalf of the Norwegian Food Safety Authority. The effects of biopesticides and chemical fungicides on development of *Microdochium* spp., *Fusarium* spp. and mycotoxins in cereals have been tested in field trials. The active ingredient prothioconazole is found to reduce DON content in oats by on average 50% compared to untreated control. However, none of the tested biopesticides has resulted in a reduction of DON, and no fungicide treatment has resulted in a reduction of *F. langsethiae* and HT2+T2-toxins in oats.

This master thesis will focus on Integrated management of Fusarium head blight in cereals. The practical work will be to characterize the impact of selected management strategies (choice of variety, tillage practice and/or fungicides) on the development of Fusarium head blight and mycotoxins in cereals. The student will learn to identify symptoms of FHB in cereals, and to identify fungal species within the genus *Fusarium* and *Microdochium*. Depending on the topic for a specific master thesis, the use of molecular methods for fungal identification, as well as chemical methods for the quantification of mycotoxins may also be feasible.

Supervisor: Ingerd Skow Hofgaard (Norwegian Institute of Bioeconomy Research, NIBIO and NMBU. In collaboration with co-supervisors from NIBIO (to be decided, depending on project plan).

7 Sopp-sjukdommer	**Lagersjukdomar på eple, inokulerings- og lagerforsøk**
	Postharvest diseases on apple, inoculation and storage experiments

Background
Apple is the most important fruit crop in Norway. There is a lack of knowledge about the biology and risk for latent infections of the pathogen in young trees.

Apple fruit can have several different diseases and decays.

Thesis
The master thesis work has several options: 1. Storage experiments of inoculated fruit to gain basic knowledge about the relationship between fruit, storage condition and pathogen. The work will start in summer/early autumn and finish in December/January. 2: Sample pathogens in field during summer and autumn, do isolations and do identification with molecular methods in autumn/winter in order to gain new pathogen biology knowledge. 3. Storage experiments with focus on physiological decay in order to gain more knowledge about the mechanism and influencing factors. Can be focused on metabolism study at Ås, storage of fruit at Ås or at NIBIO Ullensvang. NIBIO Ullensvang may host the student and offer a summer job/internship.

Supervisors:
If pathology focus: Arne Stensvand (NMBU/NIBIO) arne.stensvand@nmbu.no/ arne.stensvand@nibio.no, May Bente Brurberg (NIBIO/NMBU), may.brurberg@nmbu.no/ may.brurberg@nibio.no and Jorunn Børve (NIBIO) jorunn.borve@nibio.no

If storage/fruit quality/postharvest physiology focus: Siv Fagertun Remberg (NMBU) siv.remberg@nmbu.no and Jorunn Børve (NIBIO) jorunn.borve@nibio.no

8	**Effect of cover crops on soilborne diseases of carrot**
	Effekt av fangvekster på jordboende sykdommer av gulrot

Examples of soil-borne diseases of carrot: Sclerotinia rot and liquorice rot. Photo: Belachew Asalf

Background: Soil-borne disease are major problems of carrot, both in the field (pre-harvest) and in storage (postharvest). Survival structures of pathogens, such as sclerotia, chlamydospores and oospores, can survive in the soil for many years. Management of soil-borne diseases are difficult because they cannot easily be targeted by the conventional contact fungicides, and most systemic fungicides are not translocated into the roots. There is therefore a need for alternative measures to control soilborne pathogens in carrot.

Use of cover crops are one of the strategies for reducing soil-borne pathogens and improving soil health and productivity. Several studies have demonstrated the effect of cover crops on soil-borne pathogens. Cover crops can modify the soil microbial community and increase beneficial organisms, while specific biofumigants released from the cover cops (especially plant species within Brassicaceae) may release metabolites which directly suppress soilborne plant pathogens. Rye and white mustard used as cover crops in carrot production increased numbers of antagonistic bacteria and fungi. There is limited or lack of knowledge on the effect of cover crops on soil-borne pathogens of carrot under Norwegian growing conditions. The objective of the master thesis will be to evaluate the efficacy of cover crops on selected soil-borne diseases of carrot in the field, at harvest and after cold storage for 3-5 months.

Supervisors: Arne Stensvand (NMBU/NIBIO), arne.stensvand@nmbu.no/arne.stensvand@nibio.no, Belachew Asalf Tadesse, Belachew.asalf.tadesse@nibio.no

9	**Management of onion white rot**
	Bekjempelse av løkhvitråte

Onion infected by *S. cepivora* and sclerotia on onion and artificial growth medium. Photo: Belachew Asalf

Background: Onion white rot caused by *Stromatinia cepivora* (= *Sclerotium cepivorum*) is a major disease of all *Allium* spp. worldwide. It is a quarantine disease in Norway, and is especially challenging for the onion set production. The pathogen survives as sclerotia (0.2 - 0.5 mm in diameter) in the soil for 20-40 years in the absence of the host, and it is very difficult to control. We have limited knowledge about the biology of the pathogen under Norwegian conditions. It is also important to evaluate the effect of different Integrated disease management practices to reduce the survival of the pathogen in the soil and find means to clean contaminated onion waste so it can be used for composting.

The master thesis work has several options: i) Evaluate germination stimulants and cold treatment to induce sclerotia germination; ii) study the effect of steaming and composting on the survival of *S. cepivora* as sclerotia; iii) evaluate the effect of *S. cepivora* antagonists; iv) evaluate susceptibility of different *Allium* spp. to *S. cepivora.*

Supervisors: Arne Stensvand (NMBU/NIBIO), arne.stensvand@nmbu.no/arne.stensvand@nibio.no, Belachew Asalf Tadesse, Belachew.asalf.tadesse@nibio.no