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### **Systemic Mycorrhiza-Induced Resistance in Blackeyed Pea**

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# Systemic Mycorrhiza-Induced Resistance in Blackeyed Pea

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## Background and Introduction

- A large portion of global crop yields are lost to insect damage every year
- Widespread use of synthetic pesticides isn't enough, but also causes other challenges
  - These endanger agricultural workers
  - These endanger non-pest animals like pollinators and natural enemies of pests
  - These diminish in usefulness as pest populations become resistant
- Alternative pest-management strategies exist, but are not as widely-implemented
  - Breeding and genetically-modifying pest-resistance traits in premium seed
  - Exploitation of existing beneficial organisms and plant pest-resistances
- Plants coevolved with their pests and developed effective innate defenses
  - These include plant-wide concentrations of insecticidal and deterrent compounds
  - Phenolic derivatives of quinic acid impair caterpillar digestion and are toxic
- Another product of plant-pest coevolution is the ability to form symbioses
  - Symbioses occur between 85% of land plants with arbuscular mycorrhizal (AM) fungi
  - These enhance plant nutrition and plants' existing defenses against many insects
- A mycorrhiza-induced resistance (MIR) hypothesis** states that symbiotic plants have faster and stronger "primed" responses against subsequent challenges
  - Host plants adjust their immune systems to accommodate the AM symbiosis
- However**, this hypothesis has been criticized
  - There is a taxonomic bias in previous research
  - Changes in plant-wide genetics and signaling are difficult to isolate
  - Pests have coevolved with their hosts and many have tools to bypass plant defenses
- Therefore**, reintroducing these microbes may one day help
- However**, this requires better far more evidence in a broad sample of plant diversity and better knowledge of the underlying biology

## Novel Model for Plant-Pest-Symbiote Interactions

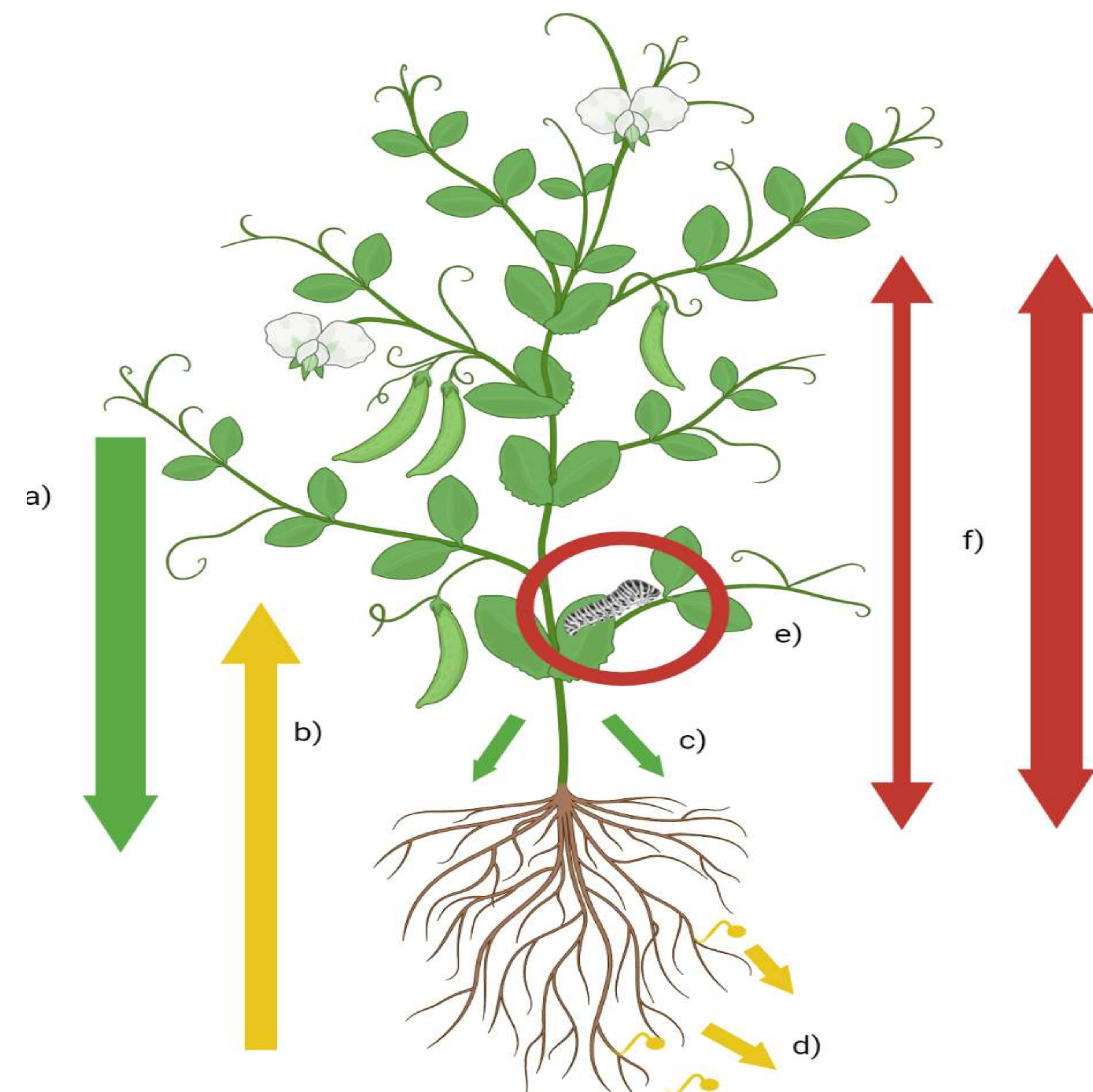


Figure 1: The proposed novel model includes Blackeyed pea, Beet Armyworm (BAW), and an AM fungus. These naturally co-occurring organisms change each other in complex mutual interactions. a) Plants share photosynthates with AM fungi. b) AM fungi share insoluble soil nutrients with plants. Plants c) and AM fungi d) signal other soil organisms. e) Caterpillars arouse the plant immune system by damaging tissues. This signal travels throughout the plant and also to AM fungi. According to the MIR hypothesis, these signals are stronger and faster in plants primed by AM fungi and previous insect attacks.

## Aims:

- Develop and evaluate a novel model system including blackeyed pea, beet armyworm (BAW), and AM fungi
- Characterize priming responses in mycorrhizal plants and caterpillars after two insect challenges

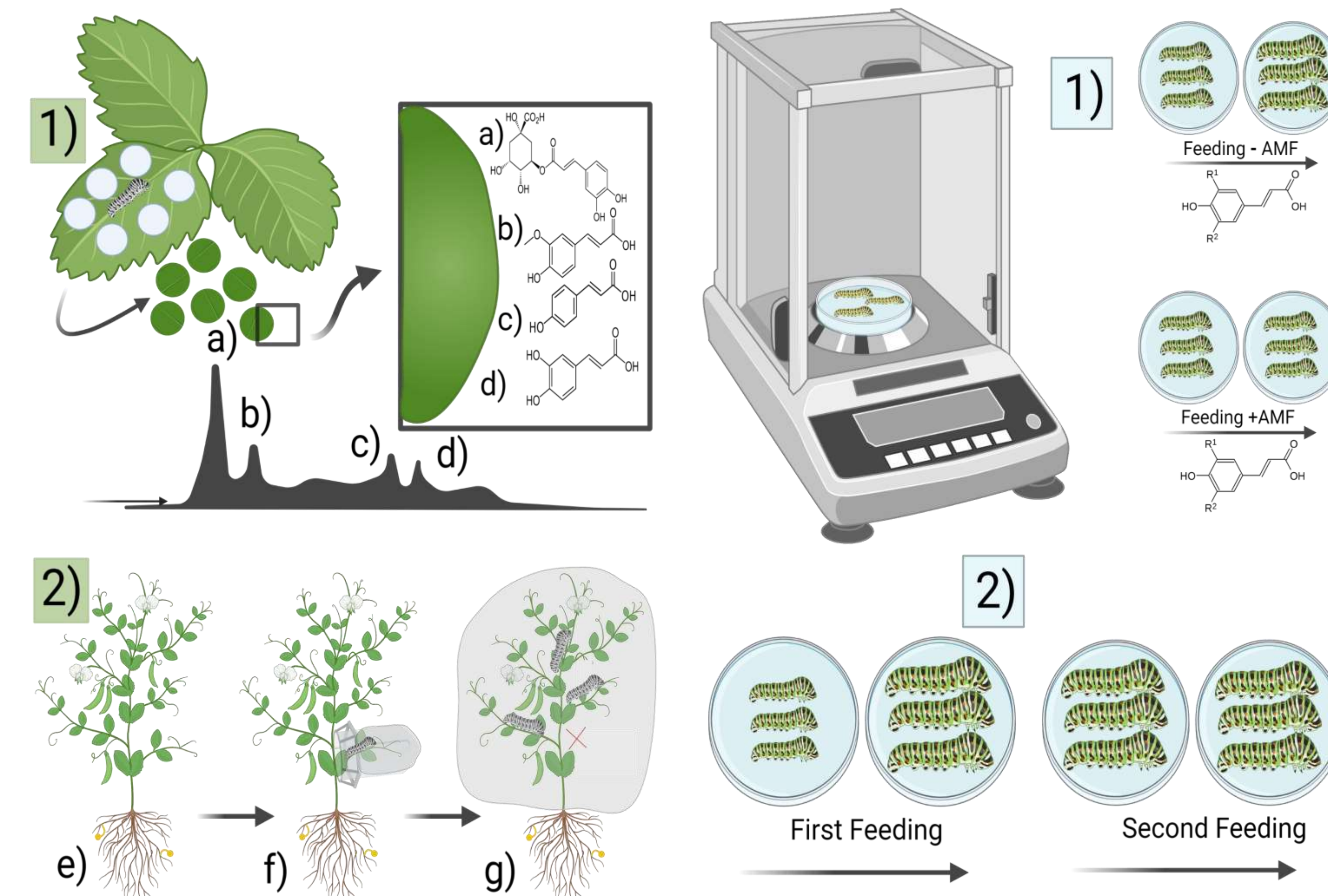


Figure 2: The second aim of the proposed research is to characterize priming responses in mycorrhizal plants and caterpillars as evidence of the MIR hypothesis. In plants (left), the priming response will be measured as plant-wide concentrations of phenolic compounds. 1) These compounds, a) chlorogenic acid, b) caffeic acid, c) p-coumaric acid, and d) ferulic acid will be quantified in leaf tissue. 2) Concentrations of these compounds will be compared between mycorrhizal and nonmycorrhizal plants, and in the same plants e) before a first feeding, g) after a first feeding, and g) after a second feeding. In caterpillars, (right) the priming response will be measured as changes in caterpillar fitness. 1) Changes in weight after feeding will be compared between caterpillars on mycorrhizal and those on nonmycorrhizal plants. 2) Changes in weight after feeding will be compared between a first and a second feeding.

## Methods

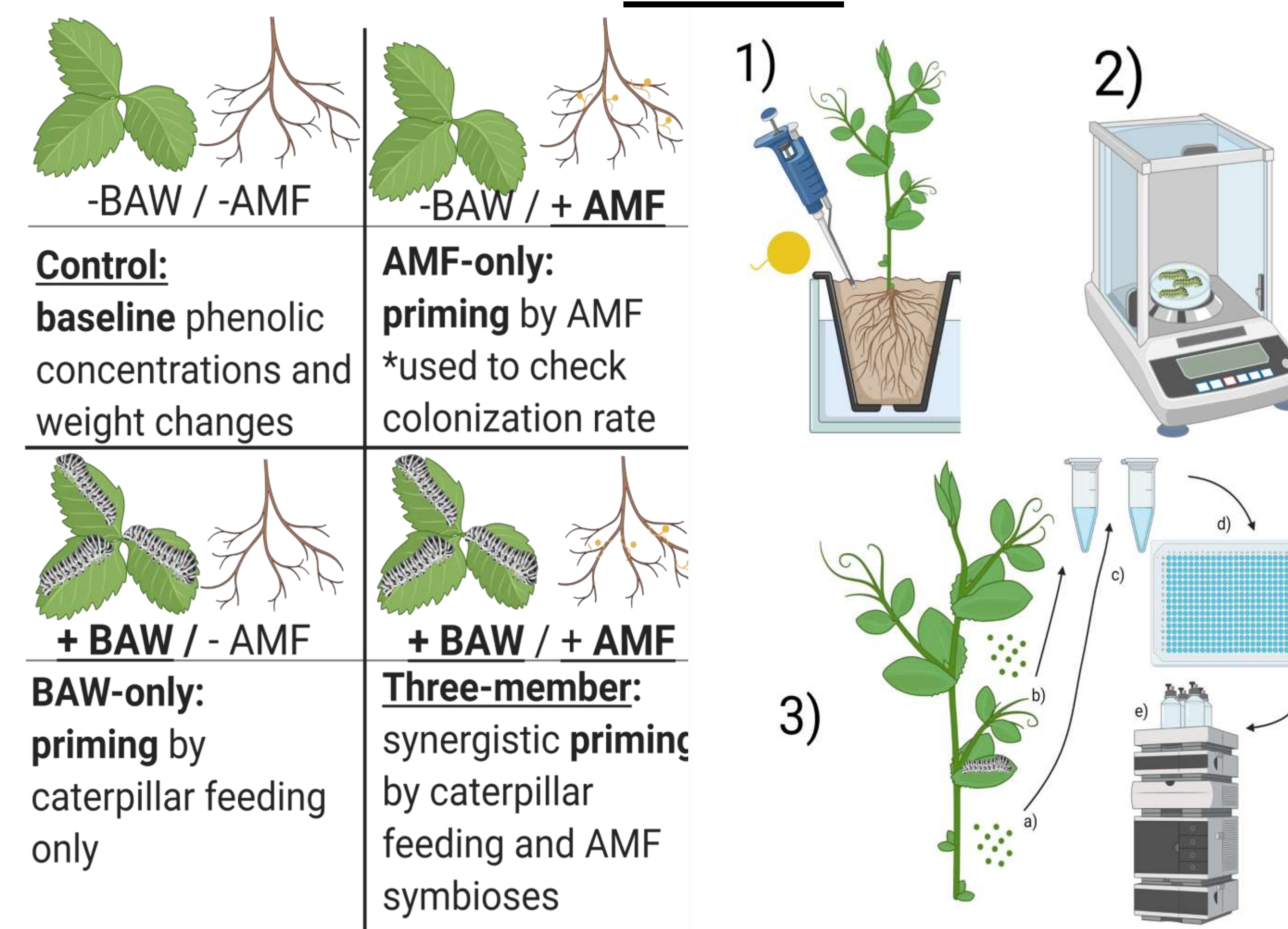


Figure 3: Data collection requires induction of priming. Data includes relative concentrations of phenolic compounds in leaf tissue and relative changes in plant weight. Left: The proposed research includes four treatments. Data from each will demonstrate the effect of priming. Right: The proposed experiment proceeds as follows. 1) Peas will be germinated, transplanted, and inoculated with AM fungi. 2) When AMF colonization is optimal, caterpillar feeding will begin. Caterpillars are weighed before and after feeding to determine weight change. 3) After each round of feeding, compounds of interest will be quantified a) After the first feeding, injured tissue will be sampled. b) After the second feeding, all leaf tissue will be sampled. c) Sampled tissue will be macerated. d) Compounds of interest will be extracted. e) Compounds will be quantified.

## Background and Introduction

- Safe and sustainable food production becomes increasingly urgent as the world population grows
- Existing pest-management methods are insufficient and, at worst, harmful
- Precious soil and water resources demand efficient use and stewardship
- Blackeyed pea is recommended as a drought-resistant food, forage, cover, and rotation crop for use across the southern US
  - It remains a principal stress-resistant crop in semi-arid regions of the world
- The AMF symbiosis enhances stress-resistant crops' most desirable qualities
  - Resistances to drought, salinity, and pests
  - Increased uptake of insoluble soil nutrients, reducing reliance on fertilizers
  - Increased protein and antioxidant content of fruit
- In collaboration with Dr. Maria Munoz-Amatriain (CSU), this research contributes to investigation of drought resistance traits in cowpea germplasm
- Evaluation of a novel model increases the taxonomic diversity of research into these three-member interactions
- The proposed experimental design compares priming by AMF with priming by multiple rounds of feeding
- The proposed experimental design better models the complexity of these interactions in natural environments

## Expected Results

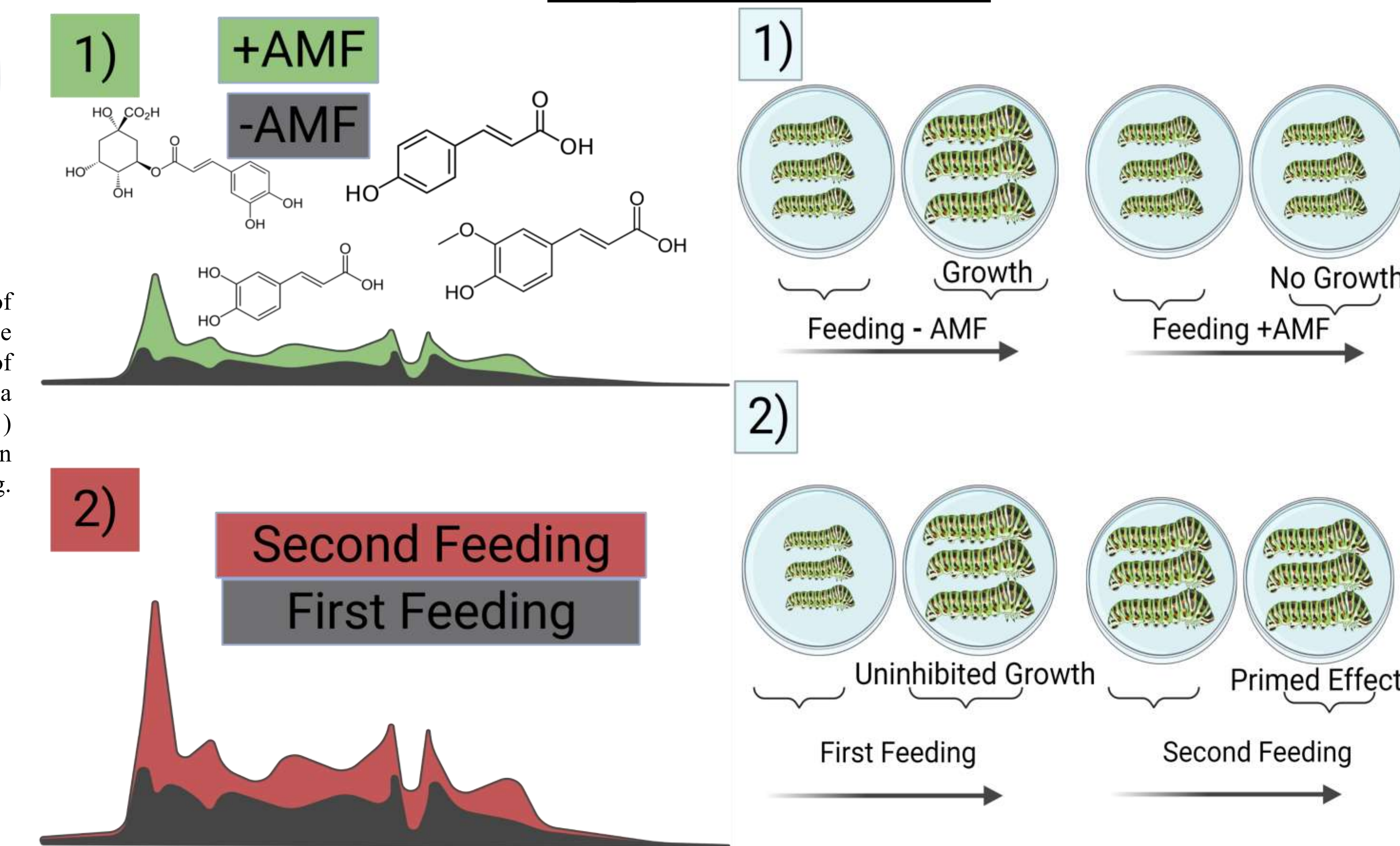


Figure 4: Predictions have been made for the second aim. Left: According to the MIR hypothesis, signals regulating plant-wide quantity of defense compounds will be stronger and faster after inoculation and then moreso after subsequent insect attacks. 1) HPLC curves showing expected relative concentrations of four phenolic compounds. These are expected to be relatively higher in mycorrhizal plant tissue for all samples. 2) HPLC curves showing higher relative concentrations after a second feeding. Right: Mycorrhization is known to deter feeding and impair digestion. 1) Caterpillars will be less fit on mycorrhizal plants. 2) The effect of priming will be subtle or negligible during the first feeding. However, this first feeding further primes the plant. During the second feeding, primed defenses will strongly deter feeding.

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