Algae Metabolites and Nutrient Uptake: TrueSolum® Enhances Iron Availability and Uptake

1. Introduction

TrueAlgae is pioneering metabolite research to bring innovative solutions to agriculture. A critical area of focus is enhancing the availability and uptake of essential nutrients, particularly **iron**, which is vital for plant health and productivity. Iron deficiency is a common challenge across various crops and soil types, often limiting growth and yield.

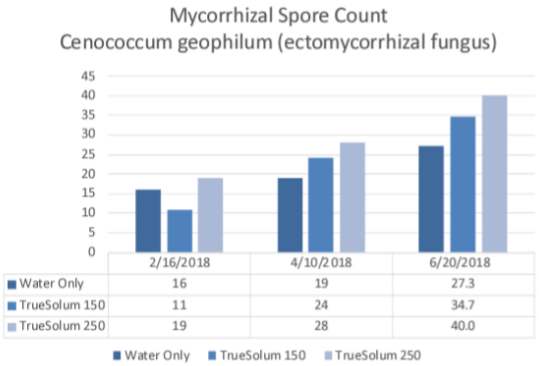
TrueSolum®, our metabolite-based solution, addresses this issue by stimulating natural plant and soil processes that improve the release and mobilization of iron, making it more accessible to plants. Through extensive research and field trials, we have discovered that TrueSolum signals and proliferates microbes that produce siderophores - molecules that chelate iron, enhancing its uptake by plants. This innovative approach provides a powerful tool for improving nutrient availability and plant health across a wide range of soil and crop conditions.

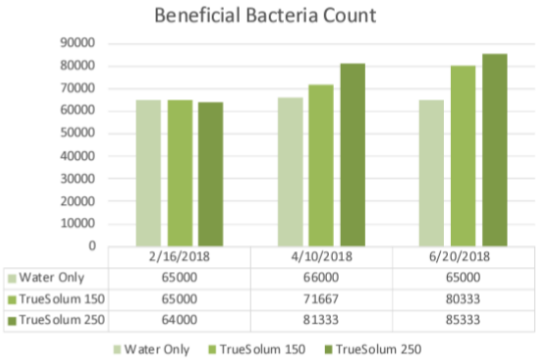
2. Discovery of the Active Metabolites in TrueSolum

Metabolite analysis was conducted by Virginia Polytechnic Institute and State University (VPI), using both GC-MS and LC-MS. Homology searches for compounds with specific peaks were conducted to identify similar structures, probable molecular classes, and/or potential metabolites in various databases. A complex mixture of compounds in the following chemical groups was discovered: 9 small peptides, 4 eicosanoids, 6 octadecanoids, fatty acids, phenolic acids, and 2-3 coumarins. In addition, the metabolites that were discovered were consistent across multiple production batches and observed in a consistent ratio between each other. Many of these compounds are known chemical signalers which activate key microbes in the soil microbiome. The following research demonstrates these activities and the resulting improvement in iron availability in the soil and uptake by crops.

3. Microbial Research

Rhizosphere microorganisms are critical for crop iron uptake. Early research conducted at AgMetrics in Dover, Florida, indicated an impact on soil microbial diversity. The soil treated with TrueSolum showed greater biological diversity and higher counts of beneficial microorganisms. Increases in mycorrhizal and other microbial populations in the soil are known to improve fertility and porosity, enhance seed germination and promote primary nutrients in the host plants. In this research trial, enhanced levels of key microbes resulted in a significant improvement in root vigor of the watermelons planted in the TrueSolum treated field.





Research conducted at the Mexican Research Center on Food and Development (CIDA) compared the presence and population of phytobenefic microorganisms per gram of soil treated with TrueSolum versus control. The analyses were conducted in two fields, one growing tomatoes and one growing table grapes. Soil samples were analyzed by means of dilution and subsequent plating on selective and semi-selective culture media for fungi and bacteria. Take note of the increases in *Pseudomonas* sp. as these bacteria are critical for iron availability and uptake.

SOIL ANALYSIS I: Tomato Farm – Sinaloa, Mexico

Soil Sample	Aerobic Bacteria	Anaerobic Bacteria	Bacillus sp	Pseudomonas Fluorescens	K-Solubilizing Bacteria	Diversity Index
TrueSolum	4,400,000	11,000,000	12,800,000	890,000	40,000	7.27
Control	3,118,000	21,733,000	6,400,000	121,000	6,666	5.94

SOIL ANALYSIS II: Table Grape Farm - Sinaloa, Mexico

Soil Sample	Aerobic Bacteria	Anaerobic Bacteria	Bacillus sp	Pseudomonas Fluorescens	K-Solubilizing Bacteria	Diversity Index
TrueSolum	8,200,000	220,000	3,633,333	560,000	1,533,333	2.24
Control	2,900,000	133,333	2,166,667	0	1,033,333	1.17

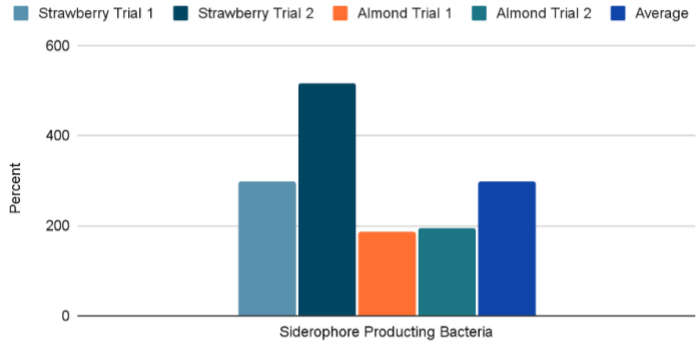
4. Siderophore Discovery

Our investigation into TrueSolum's effect on nutrient availability began with advanced genetic sequencing studies conducted in collaboration with Biome Makers. These studies were designed to analyze how TrueSolum interacts with the soil microbiome, with a focus on specific bacteria that help in the release and uptake of key nutrients, particularly iron.

Research demonstrated that TrueSolum significantly increases the population of *Pseudomonads*, siderophore-producing bacteria. Siderophores are iron-chelating molecules that help plants access iron more effectively, enhancing plant growth and development. Unlike other products that indiscriminately feed microbes, TrueSolum's metabolites are targeted signalers, providing

the specific dynamics in the soil to address the crop's needs. Field trials across strawberries, almonds, and soybeans showed that within 30-35 days of TrueSolum application, there was an average 298% increase in these beneficial bacteria. This surge in microbial activity not only improves iron availability but also enhances the uptake of iron, leading to healthier plant growth and better overall crop outcomes.

In the chart below, we observe the comparative increase in *Pseudomonas* sp. populations in plants treated with TrueSolum versus those in the control group.



Percent increase in the relative abundance of *Pseudomonas* sp. in the TrueSolum treated soil vs control

The increase in siderophore production plays a key role in mitigating Iron Deficiency Chlorosis (IDC), a widespread problem that affects many crops, particularly in soils where iron is present but not readily available to plants, such as soils with high pH. IDC occurs when plants cannot access sufficient iron, leading to symptoms like yellowing leaves and stunted growth, which can severely reduce crop yields. By enhancing the soil's microbial activity and boosting iron availability, TrueSolum offers a natural and effective solution for addressing IDC in a wide variety of crops.

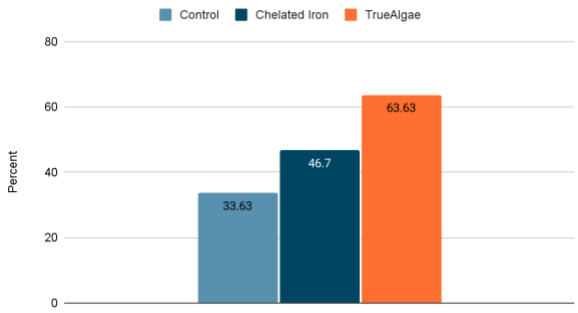
5. IDC Symptomology Testing

To better understand iron uptake challenges and address IDC, we conducted trials on strawberries, almonds, and soybeans - crops that are particularly vulnerable to iron deficiency. IDC is often marked by chlorosis, a condition where leaves turn yellow due to insufficient chlorophyll, affecting the plant's ability to photosynthesize and thrive. These trials allowed us to evaluate how TrueSolum improves iron availability and uptake, helping to alleviate the symptoms of IDC and promote healthier, greener plants.

a. Strawberries

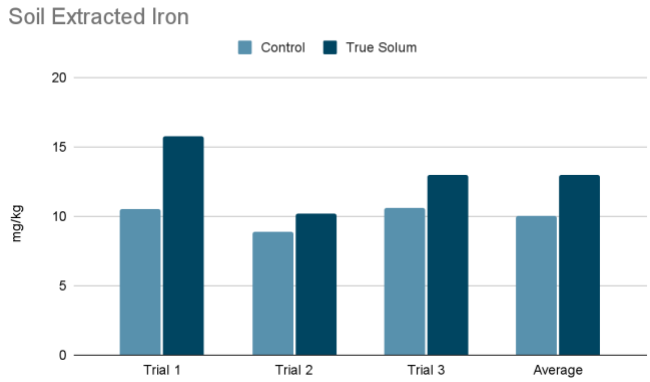
In a trial conducted in Yuma, AZ - an area with high pH soils and iron uptake challenges - TrueSolum significantly improved iron availability and plant health when compared to the standard practice of adding a chelated iron product. After 97 days, the percentage of healthy green canopy (showing no signs of IDC) was highest in the TrueSolum-treated plants, with

63.63%, compared to 46.7% for chelated iron and 33.63% for the control. The graph below shows the percentage of plant canopy classified as healthy green.



b. Almonds

In trials across three almond orchards in California's Central Valley - an area known for iron uptake challenges due to high pH soils - TrueSolum treatments resulted in an average of 30% more available iron in the soil compared to the control. The 4-5-year-old Independence variety trees benefited from this increased iron availability, driven by microbial activity stimulated by TrueSolum, which helps release iron from soil.

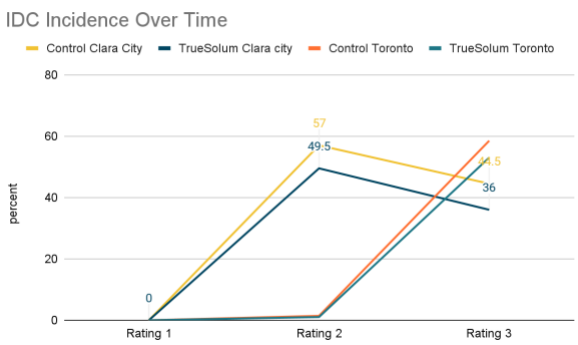


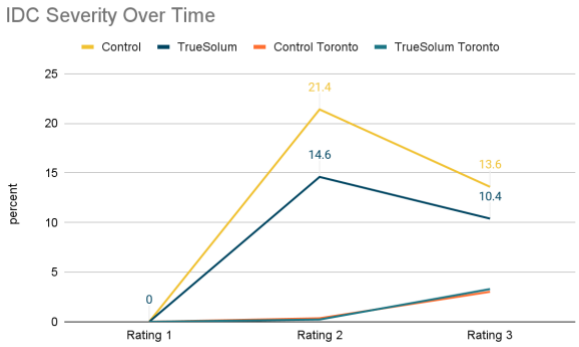
c. Soybeans

In 2024, trials in Clara City, MN, and Toronto, SD, were conducted to evaluate the effect of TrueSolum on IDC in soybeans. While IDC pressure was low at the Toronto site, the Clara City trial clearly demonstrated TrueSolum's performance.

The plots treated with TrueSolum had significantly less IDC incidence and severity compared to the control. IDC incidence was reduced by 13% during the second rating, while severity was

lowered by 32% compared to the untreated control. Although IDC incidence was low at Toronto, the trends still favored less IDC in TrueSolum-treated plots.





6. Sap Analysis

Sap samples were collected across several trials conducted on crops such as corn, soybeans, and peppers to assess nutrient uptake in plants treated with TrueSolum. Sap analysis measures the nutrients actively available to plants, providing insights into treatment efficacy.

In these trials, plants treated with TrueSolum exhibited an average of 21.5% more iron in the sap compared to the control, as shown in the table below.

Treatment	Fe
Control	1.65
TrueSolum	2.10
% Diff.	21.50%

7. Outcomes

Across multiple trials, TrueSolum consistently demonstrated a positive impact on plant health, reducing symptoms of IDC. In soybean trials, TrueSolum-treated plots exhibited lower incidence and severity of IDC compared to the controls, particularly in high-risk areas like Clara City. This reduction in IDC symptoms reflects TrueSolum's potential to improve iron availability and overall resilience in iron-deficient soils.

Similarly, in strawberry and almond trials, plants treated with TrueSolum showed higher levels of iron, supporting better growth and vigor. The sap analysis from 2022–2024 trials confirmed elevated iron levels in TrueSolum-treated plants, indicating improved nutrient uptake and likely contributing to the observed reduction in deficiency symptoms. Overall, TrueSolum enhanced plant resilience and health across various crops and conditions.

8. Conclusion

TrueSolum causes beneficial changes in the soil microbiome that influence the availability and movement of iron. Iron is widely known to be a critical component of successful crop production with many crops requiring supplemental applications of it throughout the growing season. TrueSolum provides a natural way of working with the existing ecosystem to make iron present in the soil more available to the crop by signaling *Pseudomonads* to produce iron chelating siderophores, thus making iron easy to be taken up and used by the crop. With adequate iron available, crops flourished, showing reduced symptoms of iron deficiency and increased iron uptake through sap analysis.