EVALUATION OF THE BIOLOGICAL EFFECTIVENESS OF ACF-SR IN STRAWBERRY CULTIVATION UNDER GREENHOUSE CONDITIONS.

NOVEMBER, 2018

SUMMARY OF FINAL REPORT from TLC Products, Inc.:

Dr. J. C DR. J. CONCEPCIÓN RODRÍGUEZ MACIEL Investigated the application of the product ACF-SR and its effect on greenhouse strawberry plants. The investigation showed dramatic improvements in all measured parameters, including weight of fresh and dry plant, weight of fresh and dry roots,

dramatic increase in fruit numbers, weight per fruit, and fruit quality (BRIX).

- At the lowest dose of ACF-SR, yield was 38% improved compared to an untreated control
- At the highest dose of ACF-SR, yield was a dramatic 70% improved compared to an untreated control

Zero negative effects were found by the university investigator (zero phytotoxicity).

This study demonstrates the excellent results attainable with the non-toxic, non-GMO, probiotic product ACF-SR!

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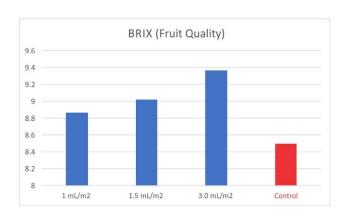
Summary of ACF-SF Test Data (see details in remainder of report)

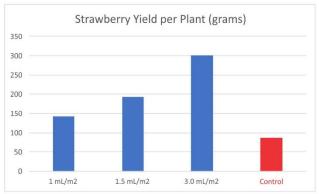
The charts below show the dramatic and statistically significant improvement in all aspects of this university based greenhouse based strawberry study.

While detail can be found in the pages that follow, note particularly that fruit quality, overall yield, and fresh and dry weight of plant and roots improved with ACF-dosing.

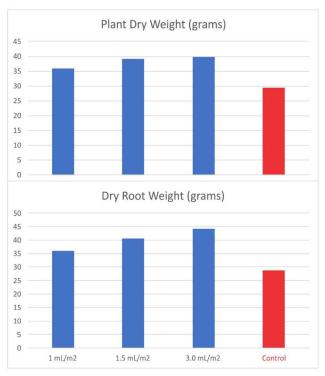
Note further that three dose rates of ACF-SF were used in this study, and that the results improved as the dose rate increased.

This study proved that ACF-SF use improved plant yield up to 70% compared to the control in this authoritative university study.









General and specific objectives:

- 1. Evaluate the biological effectiveness of ACF-SR in the cultivation of Strawberries (Fragaria x ananassa), Var. CAMAROSA under greenhouse conditions.
- 2. Secondly, to evaluate the possible phytotoxic effects of ACF-SR under these conditions.

Dates Study Was Conducted:

Begin July 06 of 2018, End September 26, 2018

Application Methods and Doses.

Four applications applications of ACF-SR were made: the first directed to the floor "In drench" and three more foliar. The first application was at the time of the transplant applied directly to the soil "In drench", the second foliar in vegetative development, a third foliar in the pre-flowering stage and the fourth in the same foliar way in the fruiting stage.

The type of soil was a clay loam with 20 to 45% silt, and between 15 and 25% clay. Actual doses were as follows:

- Low Dose (A): 1 ml / square meter
- Medium Dose (B): 1.5 ml / square meter
- Higher Dose (C): 3 ml / square meter
- Control (D): Untreated, no dose of ACF-SR

Experimental design

Experimental design: completely randomized with four treatments and four repetitions (4 plants will be taken per treatment), for 16 experimental units.

Treatment arrangement: capital letters indicate the treatment.

C	В	D	D
D	Α	В	Α
Α	C	Α	C
В	D	C	В

- Low Dose (A): 1 ml / square meter
- Medium Dose (B): 1.5 ml / square meter
- Higher Dose (C): 3 ml / square meter
- Control (D): Untreated, no dose of ACF-SR

Each of the plants is an experimental unit allocated one square meter. Then per the chart, four plants selected at random for each of A, B, C, and D.

TOTAL SURFACE: 16 m2.

NUMBER OF APPLICATIONS: Four applications (as noted previously, at transplant directly to soil, then during vegetative development, then at preflowering, and lastly during fruiting stage).

Evaluation methods

- Fresh and Dry Weight of the plant: the fresh weight of the plant was determined, then dried in an oven at 70 ° for 24 hours to give the dry weight.
- Fresh and dry weight of root (g): The fresh weight of the root was determined, then dried in an oven at 70 ° for 12 hours for dry weight.
- Root volume (ml): Performed with a graduated cylinder, the milliliters of water displaced by the root were measured.
- <u>Number and weight of fruits per plant:</u> The total number of fruits present per plant was quantified by visual observation, then the total weight taken.
- Fruits: pH and Brix degrees (° Brix): A random sample of five fruits of the four strawberry plants for each type (A, B, C, and D) was ground to determine the pH with the help of a portable potentiometer
- Fruit Yield Performance in weight / plant: With the data of weight and number of fruit, the yield per plant will be estimated.
- <u>Phytotoxicity:</u> A total of four evaluations were carried out, one before each application and one last before the harvest, using the scoring scale proposed by the EWRS (European Weed Research Society)

Statistical analysis:

The variables were subjected, without transformation, to an analysis of variance to determine if at least one treatment was different from the others (ANOVA, $\alpha = 0.05$). Subsequently, the data were subjected to a multiple comparison test to order the biological effectiveness of the treatments under study (TUKEY, $\alpha = 0.05$).

Results – Fresh and Dry Weight of the Above Ground Plant

Statistical differences were found where the best treatments were doses of 1.5 and 3.0 mL / m2 when recording an average in fresh weight of 221.32 and 226.67 g respectively, with and increase of 26.08 and 29.13% compared to the control. Similarly for the Dry Weight; show a positive effect since the treatments in which the inoculant was used show an efficacy of 21.52 to 35.29% higher than that reported by the control (Tables 4 and 5)

Fresh weight of plant

ID	Dose	Fresh Plant Weight	% Increase
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Α	1 mL/m2	209.54	19.37
В	1.5 mL/m2	221.32	26.08
С	3.0 mL/m2	226.67	29.13
D	Control	175.54	0

Dry weight of plant

ID	Dose	Plant Dry Weight	% Increase
Α	1 mL/m2	35.79	21.52
В	1.5 mL/m2	39.08	32.71
С	3.0 mL/m2	39.84	35.29
D	Control	29.45	0

Results - Fresh and Dry Weight of Roots

ACF-SR dosing to strawberry plants presented positive effects on fresh and dry root weight, statistical differences between the treatments and the control.

ID	Dose	Fresh Root Weight	% Increase
Α	1 mL/m2	76.76	22.41
В	1.5 mL/m2	86.7	38.26
С	3.0 mL/m2	89.63	42.92
D	Control	62.71	0

ID	Dose	Dry Root Weight	% Increase
Α	1 mL/m2	36.11	25.68
В	1.5 mL/m2	40.53	41.07
С	3.0 mL/m2	44.13	53.6
D	Control	28.73	0

Results – Root Volume (ml displacement by roots in graduated cylinder)

Root volume of ACF-SR treated strawberries were larger (highly significant statistical differences between the treatments evaluated with respect to the control:

ID	Dose	Root Volume (ml)	% Increase
Α	1 mL/m2	28	27.32
В	1.5 mL/m2	32	45.72
С	3.0 mL/m2	34	54.26
D	Control	22	0

Results - Number of Fruits per Plant

The number of fruits per plant increased with ACF-SR dosing, in a statistically significant way, with each higher dose (as has been the case with all results presented so far).

ID	Dose	Number Fruits per Plant	% Increase
Α	1 mL/m2	13.5	27.78
В	1.5 mL/m2	16	39.06
С	3.0 mL/m2	21.75	55.17
D	Control	9.75	0

Results - Weight of Fruits per Plant

The weight of fruits per plant increased with ACF-SR dosing, in a statistically significant way, with each higher dose (as has been the case with all results presented so far).

		Weight of Fruit per Plant	
ID	Dose	(grams)	% Increase
Α	1 mL/m2	10.56	12.78
В	1.5 mL/m2	12.01	23.31
С	3.0 mL/m2	13.8	33.26
D	Control	9.21	0

Results - Ph and Brix degrees (°Brix)

Better fruit quality was obtained when dosing the highest level of ACF-SR. As has been the case with all measurements, the statistical validity confirms improving quality as ACF-SR dose increases.

ID	Dose	pH of Fruit	Brix
Α	1 mL/m2	4.57	8.87
В	1.5 mL/m2	4.59	9.02
С	3.0 mL/m2	4.63	9.37
D	Control	4.36	8.5

Results - Fruit Yield Performance (gr / plant)

Based on the analysis of variance for the yield variable of strawberry plant there were significant statistical differences between the treatments, noting that the ACF-SR in high dose of 3.0 mL / m2 achieved the highest yield per plant.

ACF-SR at the highest dose had fruit yield of 300.26 gr / plant, which represents an increase of 70.58% with respect to the control that only achieved a production per plant of 87.53 gr. Note the increase in yield compared to the control in the table below. The yield increase at each dose was significant per Tukey's statistical analysis of differences.

ID	Dose	Yield per Plant (grams)	% Increase
Α	1 mL/m2	143.05	38.81
В	1.5 mL/m2	192.44	54.52
С	3.0 mL/m2	300.26	70.85
D	Control	87.53	0

Results – Phytotoxicity

Phytotoxicity is defined as a delay of seed germination, inhibition of plant growth or any adverse effect on plants caused by specific substances (phytotoxins) or growing conditions (WRAP, 2002). None of the treatments showed phytotoxicity, regardless of the dose.

CONCLUSIONS

Based on the results presented in this final report, ACF-SR dosing to strawberry plants showed the following statistically significant results:

- Fresh and dry weight of above ground plant increase 19% to 35% with increasing ACF-SR dose and compared to control.
- Fresh and dry weight of root increased 22% to 54% with increasing ACF-SR dose, and root volume increased 27% to 54% with ACF-SR dose
- Number of fruits per plant increased 28% to 55% with ACF-SR, and weight of fruit increased by 13% to 33% with increasing ACF-SR dose.
- Fruit quality, or BRIX, increased with increasing ACF-SR dose
- Actual yield (total weight of fruit per plant, and with superior quality) increased a dramatic 38% to 70% with increasing ACF-SR dose.
- Zero phytotoxicity was measured at any of ACF-SR dosing rates

Actual data and statistical analysis are available on request