

Differential Effects of Potassium Chloride on Vascular Tissues, Morphological Traits and Germination of Tomato with Sperm Swarm-based Nutrient Optimization

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Abstract

Potassium chloride (KCl)-induced stress has been predominantly tested on higher crops such as cotton, rice, and wheat. What open agriculture needs now is the extended understanding of possible contributions and detrimental effects of KCl dynamics to tomato as this is one of the major crops consumed worldwide. In this study, the impacts of KCl on the three Philippine tomato (*Solanum lycopersicum*) genotypes namely, Perlas, Diamante Max F1, and Mica, in separate mesocosms were visualized and measured based on morphological, vascular tissues, and germination variability responses. Three KCl treatments were deployed, which are control (Tc, 0.05 mM), deficit (Td, 0.025 mM), and excess (Te, 0.5 mM), in 3 replicates. KCl-deficit treatment showed improvement in germination rate and vigor index and deteriorated Timson germination index for tomato seedlings. Excess KCl treatment distinctively promotes a higher root (R) to shoot (S) length ratio for Perlas variant with rusty red-orange root color. R/S length increases from control, deficit to excess KCl and R/S fresh and dry weights quadratically increase from control, excess to deficit KCl. The inconsistency of Td and Te treatments to provide equally productive outcomes across the 3 genotypes on the first 37 days after sowing led to the use of 3 bio-inspired algorithms namely, moth-flame, sperm-swarm, and jellyfish swarm optimizers, in determining the most suitable KCl concentration for growth promotion. Multigene symbolic genetic programming was used in constructing the fitness models of five phytomorphological phenotypes namely, root and shoot lengths and weights and leaf count, as functions of KCl concentration and cultivation period. Light microscopy showed that sperm swarm-optimized KCl (0.038 μ M) widens the diameter of xylem and phloem vessels in the vegetative stage which is important as they are responsible for transporting nutrients, water, and photosynthesis by-products. Hence, KCl is an essential micronutrient that could alter the growth of crops.

Keywords: Bio-inspired optimization, Nutrient toxicity, Plant physiology, Plant-soil interaction, Plant nutrient stress, Precision agriculture, Sperm swarm optimization

Introduction

Nutrient and water-enriched soil naturally promote crop growth. Issues such as nutrient deficiency and toxicity happen due to agricultural malpractices, particularly, by not measuring the initial nutrient concentration in the soil system before sowing. The plant-soil interaction where the nutrient is one of the prime movers is very important in enabling effective production [1]. The elemental micronutrients including boron, chlorine, copper, iron, molybdenum, manganese, and zinc are required in the growth cycle of plants. They support different mechanisms inside the plant system particularly in the development of new cells, tissues, and organs. Severely drained and drought soil is characterized by the deficiency of chloride while the toxicity of chloride is more common worldwide due to improper soil amendments [2]. Chloride toxicity has been recognized as a major challenge in the open fields of Syria, Pakistan, and Turkey [3]. In the Middle East and Asian countries, tomato (*Lycopersicon esculentum*) has

been observed to be sensitive to micronutrient deficiencies and toxicity [3]. Thus, chloride stress should be taken into consideration for comprehensive experiments and analyses to help farmers improve their production with given challenging soil conditions.

Chlorine in the form of chloride ion (Cl^-) is mainly used by plants for photosynthesis reactions, gas exchange, resisting diseases, and water balance in turgor regulations [4]. Unlike iron, manganese, and zinc that are not sufficiently needed during the maturity and senescence stage, and boron being more effective only during vegetative and flowering stages, chloride is essential throughout the life cycle [5,6]. Chloride cannot be obtained from igneous and sedimentary rocks unlike iron, manganese, zinc, copper, molybdenum, and boron [7]. Hence, the importance of enriching the soil system with the right concentration of chloride is necessary to manifest its beneficial impacts on crops. This demand should be consistently employed to promote agricultural production as micronutrients are minimally present in soil systems. Agrochemicals are injected into the growth medium to suffice the required concentration [8]. Chloride salts that are commercially available are sodium chloride (NaCl), magnesium chloride (MgCl_2), calcium chloride (CaCl_2), and potassium chloride (KCl) [9]. However, MgCl_2 and CaCl_2 are used only as supplementary salts when sulfur and nitrogen are deficient, respectively [10]. Also, CaCl_2 was proven effective in extending the shelf-life quality of tomatoes [11]. NaCl has the initial fallback of dehydrating plant tissues, especially the root systems [12,13]. Among these four chloride salts, KCl is the most suitable source of chlorine for plant consumption [14]. In general, there is chloride deficiency for 100 mg/kg and toxicity happens within 500 - 100 mg/kg [3]. If fertilizer salts are added with the irrigation system, initial tempering of nutrient concentration must be automatically performed by the system so no excess nutrients may alter the natural growth rate of crops. Agricultural abiotic intervention is also different when crops are cultivated in soil-based and soilless approaches [15-17] and with consideration of the cultivation site such as near coastal area, highland, or domestic water dumping areas [18,19].

Potassium chloride has been utilized in stabilizing the impact of hydroxyapatite and cadmium contamination in soil-based cultivation of chili, rape, and basil with biochar [20]. Polymer-coated potassium chloride (PCPC) salts were applied to the cotton field with potassium sulfate (K_2SO_4) that resulted in delayed leaf senescence even if there is high salinity stress [21]. KCl -conditioned tomato plants appeared to be more standing during drought stress than those with added antioxidant enzymes [22]. Another promising usage of KCl is its inhibitory effects in the consumption of radioactive nanoparticles in rice cultivated in Japan [23]. On the other hand, induction of stress due to potassium chloride has a direct threat to seed germination and stomatal operations [24]. Hence, the injection of potassium chloride into the soil system should be properly characterized as crops require different concentrations for their growth.

Tomato is considered of the main crops cultivated in a greenhouse and is demanding in nutrients and water [25]. Leaf chlorosis and wilting of mature leaves are the most common symptoms of potassium chloride in tomatoes on top of blossom-end rot disease [26,27]. Due to an imbalance in water distribution and cell turgidity, root morphology, and leaf chlorophyll fluorescence are affected [28]. This negatively enhances nutrient uptakes. It was recommended that applying controlled-release potassium chloride (CRK) helped in maintaining the consistent nutrient absorption of maize roots in high calcium-concentrated agricultural regions [29]. Blending of 50 % CRK and 50 % soluble KCl resulted in even better improvements to the root morphology of wheat [30]. Interactive effects of PCPC with irrigation level confirmed the decrease in the utilization of fertilizers for soilless tomato cultivation [25]. The potential of KCl in improving growth quality can be seen not only through the architecture of the crop but also in its anatomy. The dimensions and geometry of parenchyma cells and xylem and phloem vessels may characterize the stress brought by KCl as this micronutrient is specialized in cell turgidity. In the perspective of computational intelligence, particularly the bio-inspired techniques, moth-flame [31], sperm swarm [32], and jellyfish swarm [33] have been used as the basis for optimizing gear train, smart grid, and structural tower design issues respectively. Exploring the usefulness and effectiveness of these bio-inspired models aside from engineering problems like agricultural challenges would be beneficial.

Despite the thorough investigations of the impacts of macronutrient (nitrogen, phosphorus, and potassium) dynamics on tomato [34], direct differential effects of potassium chloride have not experimented comprehensively. As of this writing, there is no published study that focuses on KCl assay in tomato vascular tissues even though vacuole cells are immediately affected by changes in nutrient consumption. Also, KCl -induced stress has been prominently tested on other higher crops including cotton, rice, and wheat. For cotton, KCl deficiency was tolerated using coronatine that resulted in increased root diameter and lateral root counts [35]. On the other hand, both KCl deficiency and toxicity were tolerated by using *Trichoderma asperellum* in cultivating rice [36]. Combining controlled-release KCl (CRK) and soluble KCl enhanced root morphology of wheat [30]. As of this writing, there is no

published study that have used computational intelligence in related to KCl-induced stress with cotton, rice, and wheat. Tomato was utilized mostly in drought stress and PCPC-based experiments based on the above-mentioned literature. What open agriculture needs now is the extended understanding of the possible contributions and detrimental effects of KCl dynamics to crops. Moreover, no computational intelligence tools have been used previously concerning KCl concentration optimization in spite of the strong potentials it poses in quantifying crop phenotypes [37].

To address the challenges in chloride impacts to tomatoes, this study focuses on cultivating 3 Philippine tomato genotypes namely, Diamante Max F1, Perlas, and Mica, in separate mesocosms with 3 KCl treatments (control, deficient, and toxic) to measure and visualize morphological, vascular tissues, and germination variability. Multigene genetic programming was used to characterize the differential impacts of KCl. Three bio-inspired algorithms namely, moth-flame (MFO), sperm swarm (SSO), and jellyfish swarm optimization (JSO), were used to determine the most suitable KCl concentration that will induce plant growth. There is a need to test using different bio-inspired algorithms in enhancing KCl concentration as computational techniques are prone to premature convergence that implies no optimization was performed. MFO, SSO, and JSO have unique mathematical basis [31-33]. This study explores the suitability of MFO, SSO, and JSO to ensure on generating a feasible and realistic KCl concentration for ideal fertigation in tomato. The output of this study will contribute to the: 1) confirmation of the suitability of selected bio-inspired algorithms for non-linear agricultural and plant science data and 2) visualization of the transverse cross-section of stem and morphological structure of tomato as affected by KCl-induced stress. The choice of tomato is because of its high demand worldwide and its sensitivity to micronutrients especially in seedling and flowering stages. This study does not include the measurement of absorbed chloride inside the plant systems.

Materials and methods

This study involves 4 major phases which are 1) initial seed quality test, 2) preparation of experimental design using three different KCl-induced stress treatments and greenhouse setup, 3) tomato architectural phenotype measurement, stem's vascular tissue dissection and microscopy, and 4) the application of computational intelligence in characterizing KCl impacts to phytophysiology and determination of optimum KCl concentration for enhanced tomato seedling growth quality (**Figure 1**). MATLAB R2021b is the only computational intelligence software used in the study.

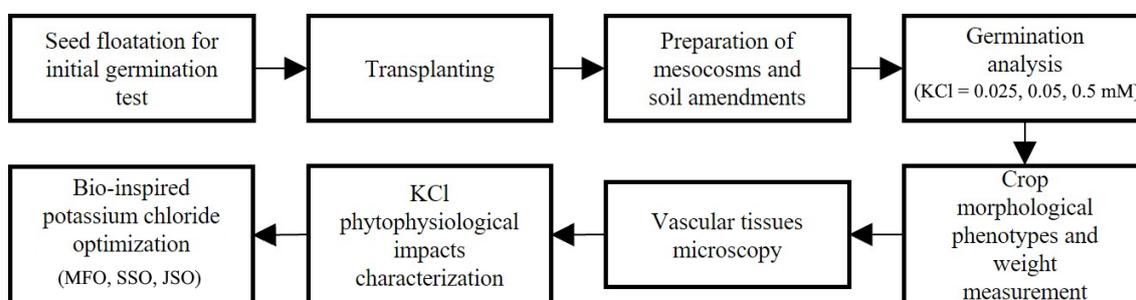


Figure 1 Developmental architecture in determining the differential effects of potassium chloride on the morphological phenotypes and vascular tissues of tomatoes with bio-inspired nutrient optimization.

Plant material and experimental design

Three tomato (*Solanum lycopersicum*) genotypes namely, Diamante Max F1, Perlas, and Mica (East-West Seed, Philippines) were used as cultivars. The experiment was performed in a greenhouse at Bacoor, Cavite, Philippines (14° 27' 26.676" N, 120° 55' 42.492" E) from September 14 to October 19, 2021. Seed floatation test was performed to initially filter out those non-viable seeds, then, they were transferred to an enclosed storage container with tissue paper at the bottom for the germination to start. Distilled water was poured inside the container up to 2 mm level from the bottom and the lid was sealed using a plastic wrap. It is then placed in a controlled environment with 27 °C air temperature and light intensity of 3800 lumens for 1 week until the emergence of a 5 mm radicle. Thirty germinated tomato seeds from each variety were transferred to the mesocosm for 30 days where each 6 in × 6 in × 12 in pot holds one seed only. The mesocosm consists of 20 % loam, 30 % sand, 40 % perlite, and 10 %

vermiculite. Three potassium chloride nutrient treatments were employed, namely: Control (Tc), deficient (Td), and excess (Te). Tc, Td, and Te has potassium chloride concentration of 0.05, 0.025 and 0.5 mM, respectively. All treatments were supplied with the following salts: 16 mM potassium nitrate, 6 mM, calcium nitrate, 14 mM magnesium sulfate, 25 μ M boric acid, 2 μ M manganese sulfate, 2 μ M zinc sulfate, and 0.5 μ M copper sulfate. Nutrients are injected daily at 8:00 a.m. into the individualized mesocosms with a volume of 0.75 L. Three replicates of the whole experiment have been performed utilizing a total of 810 seedlings (30 seeds/genotype \times 3 genotypes \times 3 treatments \times 3 replicates). The greenhouse is maintained with 24 °C air temperature and 3800 lumens artificial photosynthetic light through full-spectrum LED for 16 h photoperiod and 8 h dark period. Tomato seedlings are the crop stage of interest in this study.

Time-series germination analysis

Germination analysis was based on three computed parameters which are germination rate (GR, %), Timson germination index (TGI, seeds/day), and vigor index (VI, seeds/g) based on Eqs. (1) to (3) [38]. For this analysis, germinate seeds are those seeds inside mesocosm with at least 5 mm of radicle emergence. G is defined as the number of germinated tomato seeds at the end of a certain period (DAS 14 for GR computation and DAS 14 and DAS 37 for TGI), n is the total number of cultivated seeds, p is the germination period which is 37 days, and W is the fresh weight of the seedlings at day 37. This time-series germination analysis was employed for each tomato genotype.

$$GR = \frac{G}{n} \times 100 \quad (1)$$

$$TGI = \sum \frac{G}{p} \quad (2)$$

$$VI = GW \quad (3)$$

Crop morphological phenotypes and weight measurement

Tomato genotypes are expected to exhibit different phenotypes as affected by potassium chloride stress from day 8 to 37 after sowing. Morphological phenotypes include root length (mm), shoot length (mm), and the number of leaves. The first 2 traits are measured using a vernier caliper and the number of leaves is counted manually. Five random tomato seedlings were sampled weekly for weight measurement using a digital weighing scale with a sensitivity of 1 mg. At the end of DAS 37, 5 random tomato seedlings undergo fresh and dry weight measurements. Dry weight measurement was performed through oven dehydration of fresh leaf tissues under 80 °C for 16 h.

Vascular tissues microscopy

The impact of KCl-stress on the morphoanatomy of tomato seedlings was investigated and visualized using a trinocular compound microscope (Howell, Philippines) on a 400 \times magnification (40 \times objective, 10 \times ocular). Tomato seedling stems were dissected in transverse orientation using a scalpel blade. Note that collected plant stem tissues are those within the 1 cm region from the base node of the shoot system to make more on-point visualizations and analyses on where on the main stem region KCl may materialize its impacts. Tissue samples were histologically stained using toluidine blue O (C₁₅H₁₆N₃S⁺, TBO) within 10 seconds, then washed consecutively using ethanoic acid and distilled water. The average major axis lengths or diameters (μ m) of xylem and phloem vessels in tomato stem were measured in ImageJ software.

Potassium chloride phytophysiological impacts characterization and bio-inspired nutrient optimization

Genetic programming, a computational intelligence technique that is based on Charles Darwin's evolutionary concept [39], through MATLAB R2021b GPTIPS v2 tool was employed to develop the fitness model for the five phytomorphological phenotypes as characterized by the potassium chloride concentration ($[KCl]$, mM) and cultivation period (t , DAS) Eq. (4) [40,41]. The multigene symbolic regression genetic programming (MSRGP) hyperparameters were configured as 50 population size, 100 maximum generations, tournament size of 50, 0.05 elite fraction with enabled lexicographic selection pressure and 0.1 Pareto tournament probability, 10 maximum genes, 5 maximum tree depth, 0.1 ephemeral random constant, crossover rate of 0.85, the mutation rate of 0.14 and mathematical function set of {times, minus, plus, sqrt, square, sin, cos, log, cube, neg, abs}. By using MSRGP, the phenotype

expressions are characterized by $[KCl]$ and t . Five contour plots showing the dynamic map of KCl concentration and cultivation period to phytomorphological phenotypes were constructed in Minitab 20.4 through the distance interpolation method with a distance power of 2.

$$\{RL, SL, RFW, SFW, NOL\} = f([KCl], t) \quad (4)$$

Bio-inspired optimization techniques namely, moth-flame, sperm swarm, and jellyfish swarm optimization, were modeled for maximization optimization as adapted from [31-33] and employed as a 2-dimensional problem based on the developed MSRGP-based models (**Table 1**). The moth-flame optimization (MFO) model is based on the navigational transverse orientation of the moth flying in a straight path while maintaining a specific angle with the moon which is considered as the flame [31]. MFO was configured with an initial moth population of 100 and 30 maximum number of flames. The sperm swarm optimization (SSO) model is based on the motility of the swarm of sperm from the cervix to reaching the egg in the ovary [32]. SSO was configured with 100 initial sperms that are divided into 3 parts employed with non-uniform mutation, uniform mutation, and no mutation operators respectively, random sperm initial velocity in the cervix region between 0 and 1, vaginal pH value of 7 to 14 for chemotactic, and cervix to the fallopian tube temperature range of 35.1 to 38.5 °C for thermotaxis. The jellyfish swarm optimization (JSO) model is based on the movement of jellyfish in the ocean as mainly affected by ocean current caused by solar heating and wind surface velocity in search for food [33]. JSO was configured with an initial jellyfish population of 100 using a logistic chaotic map presenting the tossing nature of ocean waves and a time control mechanism value of 0.5 for changing between active and passive jellyfish motions in a jellyfish bloom. For MFO, SSO, and JSO, the upper and lower bounds were set to [0, 1] for KCl concentration and [0, 37] for the cultivation period, respectively, and the iteration of 100 was employed as the convergence criteria. The outputs of these three bio-inspired metaheuristic algorithms are the global best solutions of the values of KCl concentration and cultivation day required to induce the best tomato growth.

Table 1 Developed fitness model for tomato seedling phytomorphological phenotypes: Root length, shoot length, root fresh weight, shoot fresh weight, and number of leaves, as a function of potassium chloride concentration ($[KCl]$, mM) and cultivation period (t , DAS) using multigene genetic programming.

Phytomorphological phenotype	Developed fitness model
Root length (RL, mm)	$RL = 1.21t - 402 [KCl] - 1290 \cos([KCl]) - 0.327 [KCl]t + 6.6 [KCl]^2t + 1300$
Shoot length (SL, mm)	$SL = 8.57t - 2.82e^4 [KCl] + 2.95e^4 \sin([KCl]) + 2530 [KCl]t - 2650 t \sin([KCl]) - 93.8$
Root fresh weight (RFW, g)	$RFW = 25.9 [KCl] - 0.0533 t - 6.25 \log([KCl]) + 1.17 [KCl]t - 1.77 [KCl]^2t - 17.5$
Shoot fresh weight (SFW, g)	$SFW = 29 [KCl] - 0.00354 t - 6.8 \log([KCl]) + 2.1 [KCl]t - 3.65 [KCl]^2t - 20$
Number of leaves (NOL)	$NOL = 0.275 t - 129 [KCl] - 0.0475 [KCl]t^2 + 5.22 [KCl]^2t + 170 [KCl]^2 + 6.46$

Statistical analysis

Data were analyzed using Pearson's correlation coefficient ($p < 0.05$), all conducted in Minitab 20.4 (Minitab LLC). Both data processing and plots generation was done using Microsoft Excel 2018.

Results and discussion

Time-series analysis of germination of tomato

Germination is the emergence of radicle from tomato seed and is a sign of a live and adapted embryo. Based on DAS 14 and 37 germination tests, tomato seedlings treated with KCl deficiency of 0.025 mM concentration exhibited an average germination rate of 82.222 % and excess KCl leads the seedlings to advanced senescence (GR is 0 % at DAS 32) (**Figure 2(a)**). KCl-deficit treatment also performed consistently as a suitable concentration in terms of Timson germination index with an average value of 2.667 seeds/day (**Figure 2(b)**) and vigor index with an average score of 309.238 seeds/g (**Figure 2(c)**). Interestingly, the Mica seedling variant in the control treatment resulted in a very low vigor index

that is 13.236 % only of that of its counterpart with KCl deficiency (**Figure 2(c)**). This is primarily affected by the number of sprouted leaves in the shoot system that are included in the weight component of VI. The TGI of the three tomato variants scored around 0.5 seeds/day in excess treatment because of the low germination in DAS 14 which is around 6 to 8 seedlings only. Increasing the nutrient concentration applied for each tomato seedling does not guarantee the improvement of GR, TGI, and VI. Too low and high concentration does not enhance the emergence of radicle and may not sustain its early seedling development. On the other hand, it is hypothetically expected that control treatment (0.05 mM of KCl) will result in a higher seedling count at the end of the experiment [3] but it appears that deficient KCl promoted it instead. Hence, it gives rise to the need to reevaluate the ideal KCl concentration that is suitable for these specific Philippine tomato variants, and it was done in this study through bio-inspired optimization. Moreover, germination due to varying phosphorus mediation suggests that phosphorus deficiency in maize allows the expansive primary growth of lateral and seminal roots and the significant increase in the secondary growth of nodal roots [9]. Because germination is based on the emergence of at least 5 mm of the radicle, strong evidence that KCl is an essential micronutrient during this growth stage of tomato especially in terms of vigor index (**Figure 2**).

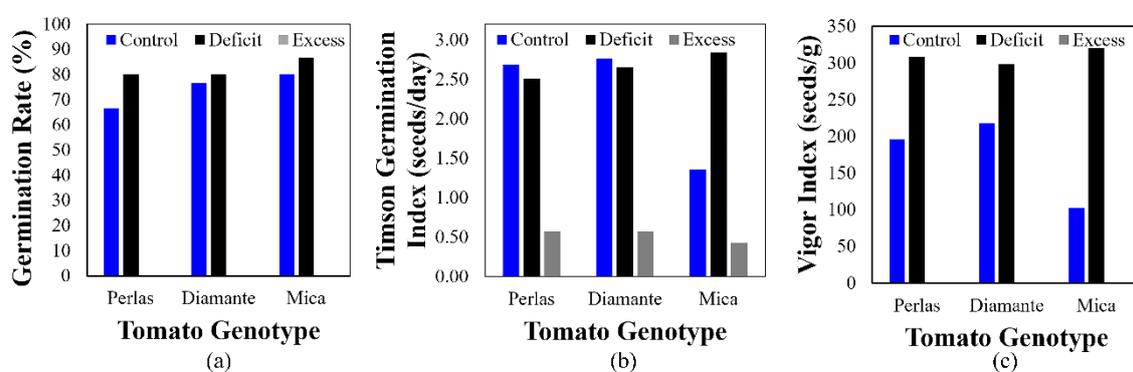


Figure 2 Germination dynamics of Perlas, Diamante, and Mica tomato genotypes as affected by potassium chloride concentration in soil system characterized by (a) germination rate, (b) Timson germination index, and (c) vigor index. These germination measures characterize the 37-day old tomato seedlings.

Differential effects of KCl concentration on root and shoot systems of tomato

After a total of 37 days of cultivation with 30 days in actual KCl treatment, strong evidence was found that increasing KCl to 0.5 mM, shoot length, root and fresh weights, and leaf count diminish which results in poorer cultivation (**Table 2**). On the average, root length of tomato seedlings has a very weak positive relationship with KCl concentration (**Table 2**). This minimal increase in root length as the soil becomes highly concentrated with KCl is mainly affected by Perlas seedlings as this tomato genotype was observed to grow root with an average of 84.7 mm long during the toxic condition (**Figure 3(a)**). This length is about 188.852 and 237.589 % longer than the Diamante and Mica seedlings have. It is apparent that the included phytomorphological traits increase in measure concerning the cultivation period with root and shoot length as the most significant ones (**Table 2**). This finding adds up to [26,29-31,41], that nitrogen, phosphorus, and chloride which is the form of KCl in this study, are essential to root and shoot primary growth. It has been a known usage of phosphorus is to make root girth thicker [9] but this study proves that a specific concentration of KCl increases root length (**Figure 3(a)**). In a more detailed view, excess in KCl during the DAS 8 to 37 resulted in longer root length for Perlas and Diamante seedlings (**Figure 3(a)**) and KCl-deficient soil resulted in longer shoot length for the three seedling variants (**Figure 3(b)**). Concerning leaf count, the KCl-deficit treatment resulted in the highest scores ranging from 12 to 14 (**Figure 3(c)**). It was observed in this treatment that Perlas and Mica exhibited the lowest and highest leaf counts, respectively, which has an inverse relation with the measured shoot length. In control and KCl-excess treatments, the leaf count and shoot length have the same trend for each genotype. This suggests that KCl-deficit seedlings have the highest tendency to promote synthesis because denser stomates may be present in a larger number of leaves. For both shoot and root, the KCl-deficit treated tissues resulted in the heaviest fresh and dry weights (**Figures 3(d)** and **3(e)**) which are profoundly opposite of the findings in the dry tissue of tomato seedlings which decreases in weight as NaCl and

MgCl₂ concentrations increase [10-12]. In the view of tomato genotypes, Perlas is characterized by the heaviest fresh and dry weights, and Diamante and Mica are characterized by the lightest fresh and dry weights, respectively. Also, Diamante seedlings retained the heaviest dry tissues after oven drying which is quantified to 42.66 %, followed by Mica and Perlas with 39.977 and 39 %, respectively. This means that the Diamante tomato variant can withstand KCl stress and still induces cell growth and tissue development. Another important finding in this study is that the ratio of root and shoot (R/S) lengths is increasing from control, deficit, to excess KCl (**Figure 3(f)**), and the R/S fresh and dry weights are quadratically increasing from control, excess, to deficit KCl (**Figure 3(g)**). This means that reducing the amount of KCl in the soil system promotes heavier root weight and longer root in general. Moreover, there is a clear resemblance to the trend in **Figure 3(g)** and findings in **Figure 3(a)** that excess treatment also provides an advantage in impressive root primary growth for the three tomato variants. These expressions of dynamic KCl are a manifestation that chloride has an essential function in the primary growth of root and shoot systems.

Table 2 Pearson's correlation values with 95 % confidence level in describing the strength of linear relationships of tomato phytomorphological phenotypes as dynamically affected by the combination of KCl concentration and cultivation period.

Growth factors	Root length	Shoot length	Root fresh weight	Shoot fresh weight	Leaf count
KCl concentration	0.125	-0.308	-0.437	-0.519	-0.160
Cultivation period	0.809	0.812	0.023	0.418	0.745

This study emphasizes that chloride in the form of KCl introduces new usage during the vegetative stage of tomatoes. Based on the measured tomato seedling phenotypes, KCl concentration from 0.1 mM to toxic 0.5 mM applied before DAS 30 may impede root primary growth (**Figure 4(a)**). In actual observations, the toxic KCl level dissolves the delicate radicles of each tomato seedling. Conservatively increasing the amount of KCl (0.005 mM per day) with the cultivation day may help improve the root primary growth. On the other hand, shoot length requires at least 0.03 mM of KCl at DAS 25 to help improve its quality (**Figure 4(b)**). For root and shoot fresh weights, it requires at least 0.035 mM of KCl throughout the first 37 DAS to maintain the proper internal distribution of liquids and formation of plant tissues (**Figures 4(c)** and **4(d)**). A KCl concentration of 0.045 mM may result in more than 6 leaves starting at DAS 25 (**Figure 4(e)**). These morphological phenotypes are observable in seedling samples presented in **Figure 5(a)**. These entail the differential impacts of KCl on tomato seedling morphology. Chloride is present in the soil system in the form of NaCl, MgCl₂, CaCl₂, and KCl which has a specialized role in the growth and quality of plant systems [9]. The first three salts have been considered useful for crop protection [4,9,12]. The current study partially agrees with the trend of chloride in the form of NaCl that its low concentration treatment corresponds to longer and heavier root and shoot lengths and weight of tomato seedling, respectively [4,5,12]. This is because chloride through excess KCl treatment was observed to significantly induce primary root length. The unique impact of KCl other than the common effects of NaCl might be attributed to the potassium content that clearly reacts with plant roots. Potassium is known to make sturdy stems [14,21,24,25,47] and it was observed in this study.

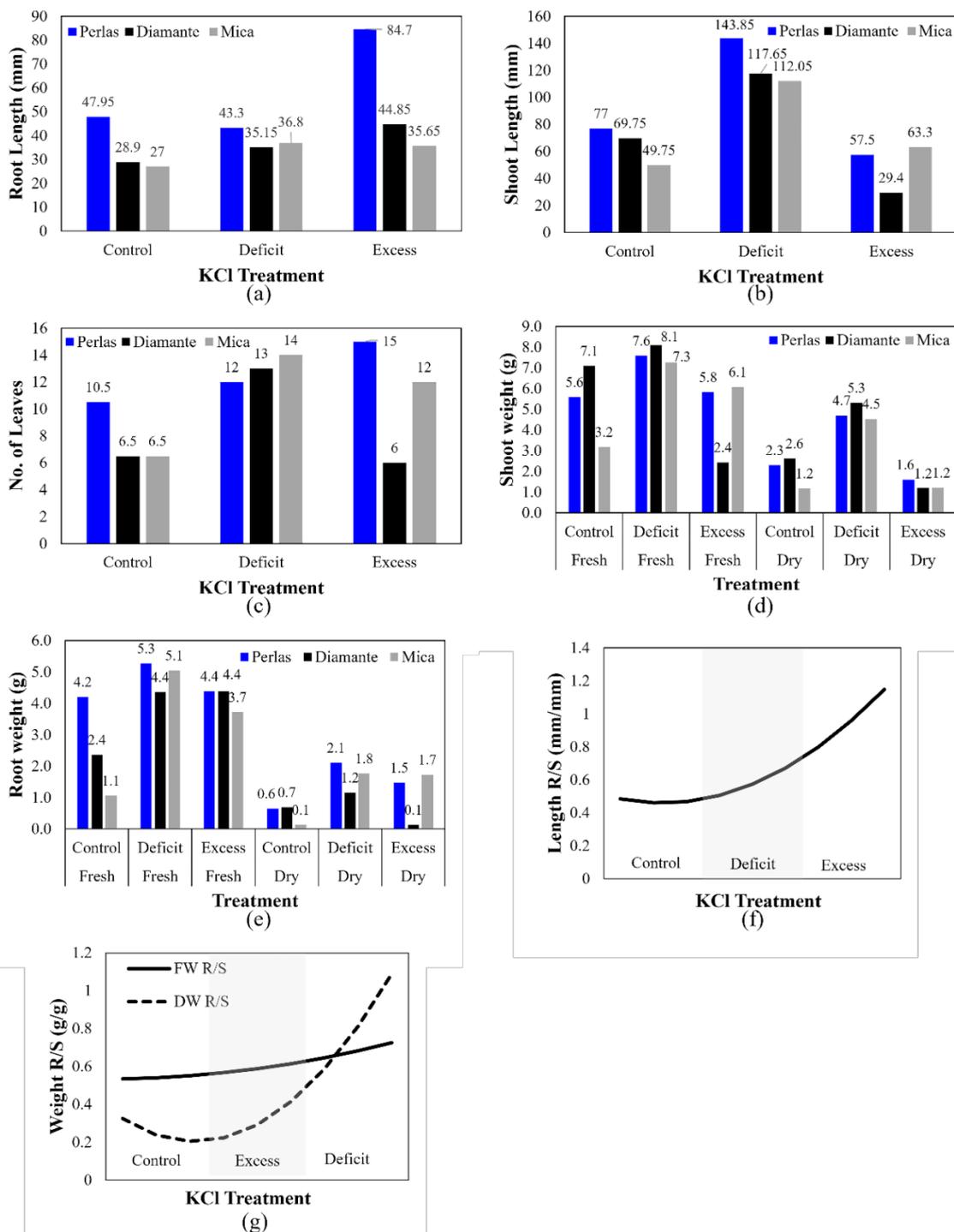


Figure 3 Impacts of KCl stress on (a) root length, (b) shoot length, (c) leaf count, (d) shoot weight, and (e) root length. Trends representing the fresh (f) root/shoot (R/S) length and (g) R/S weight in relation to control, deficit, and excess KCl treatments.

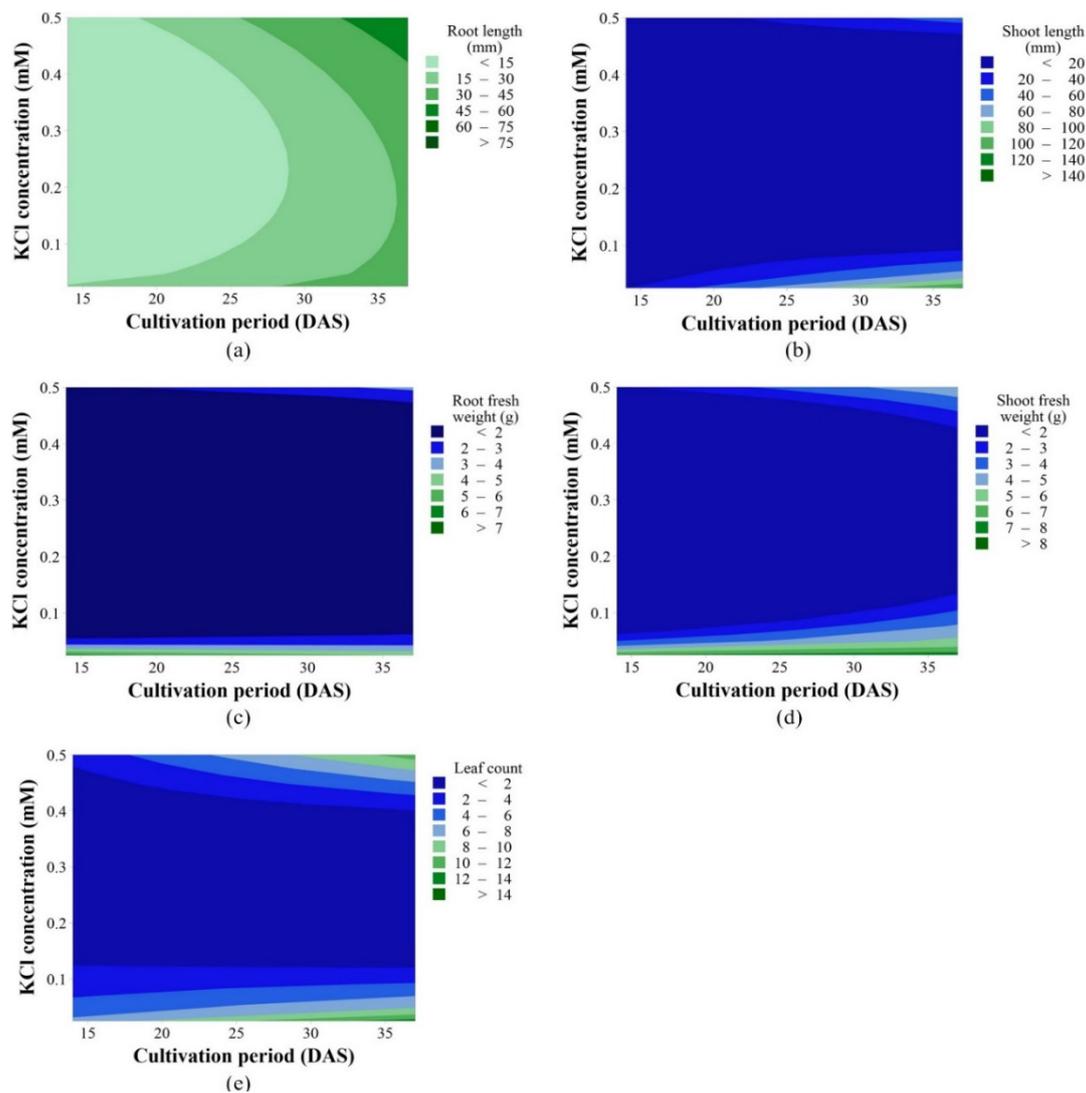


Figure 4 Perlas, Diamante Max F1, and Mica tomato varieties’ combined morphological phenotype dynamic maps as affected by potassium chloride concentration (0.05, 0.025, 0.5 mM) and the cultivation period based on genetic programming symbolic expression results.

It was suggested that the variability of resistance to chloride stress depends on the species of crops exposed to it [24,25,47]. The visual appearance of tomato seedling as a direct indicator of stressors under different chloride concentrations can also be seen through its anatomy, particularly, through the vascular tissues that are responsible for transporting the nutrients, minerals, and by-products of photosynthesis all around the plant systems from the apical tip of the root to shoot and fruits. For the current study, xylem and phloem vessels are the tissue of interest as they are responsible for the moving of essential fluids. Based on visual traits of harvested tomato seedling samples (**Figure 5(a)**), KCl-deficit-treated Perlas, Diamante, and Mica (Td-P, Td-D, Td-M) have the thickest stems and whiter roots as compared to the control (Tc) and excess treatments (Te). Also, the first true leaves (dicotyledon leaves) of KCl-deficit seedlings are still present at DAS 37 while for the control and excess treatments, they are already wilted. Seedlings exposed with toxic concentration have exhibited a shorter shoot system and longer and thicker root system with significantly visible rusty orange-brown color (**Figure 5(a)**). Orangy root might be an indicator of overwatering [4,20] but this current finding lies in the fact that a higher concentration of KCl drives the root to mature faster. This is probably caused by reinforcing each cell to enhance osmotic pressure resulting in faster root elongation which agrees with [45]. All roots are not exposed to air that may dry them out of the water as they have completely emerged underground. No root rot has been observed in all treatments. To better analyze the impacts of KCl on tomato seedlings, the micrographs of

stem stained with TBO are presented (**Figure 5(b)**). Another interesting finding here is that trichomes are evident only to seedlings cultivated above 0.05 mM of KCl. As one of the responsibilities of trichomes is to aid plants in acquiring nutrients, excess KCl-treated seedlings might have its advantage causing them to have a longer root system. Xylem tissues are responsible for transporting water and dissolving nutrients from roots to leaves in a unidirectional flow. In this study, xylem vessels in KCl-deficit treatment are constantly exhibited by wider maximum major axis length or diameter with 184.146 and 193.736 % higher than the control and excess treatments (**Figures 5(b)** and **5(c)**). Also, it is observable that the presence of a large density of tracheids or small xylem vessels is because the tomato seedling is still growing and not fully mature yet. The lignified walls of the xylem are responsible for holding internal system fluids and maintaining the osmotic pressure. On the other hand, the average diameter of phloem vessels is observed to be wider also in KCl-deficit treatment with 157.215 and 170.136 % higher than the control and excess treatments (**Figures 5(b)** and **5(d)**). This suggests 2 realizations: First, xylem and phloem diameters are directly proportional as affected by KCl concentration; and second, it proves that as plant uptakes more water and nutrients allowed by wider xylem vessels, the distribution of glucose from plant leaves is also active as supported by wider phloem vessels. Unlike NaCl, KCl exhibits a water softening property that allows the plant cells to directly consume the macronutrients essential for their growth like potassium [4,11,20,45]. Moreover, Tc-M and Tc-D seedlings have resulted in diminished growth (**Figure 5(a)**) which has been resolved by employing bio-inspired algorithms in determining the exact KCl concentration suitable for these tomato variants.

After several iterations in bioinspired combined exploration and exploitations phases, MFO, SSO, and JSO converged on their global optimum value for KCl concentration at day 37 of cultivation for the selected Philippine tomato genotype (**Figure 6** and **Table 3**). For the bases of root and shoot lengths, root and shoot fresh weight, and leaf count, the SSO exhibited the highest fitness function value that is within the range of deficient (0.025 mM) and control (0.05 mM) (**Figure 6**). The fitness curves exhibited by JSO in optimizing KCl concentration for root length and fresh weight are characterized by sigmoidal property which is smooth suggests poor enhancement (**Figures 6(a)** and **6(c)**). It is also manifested by MFO in optimizing KCl concentration for leaf count (**Figure 6(e)**). Interestingly, SSO reveals to be the most suitable bio-inspired algorithm for this specific KCl concentration and tomato traits data and application. This claim is also supported by averaging the generated KCl concentration in relation to the tomato traits (**Table 3**). MFO and JSO has recommended 0.016 mM and 0.019 mM KCl to be fertigated on tomato crops from DAS 1 to 37. These nutrient concentrations are far weaker than the deficient concentration performed in the experiment. On the other hand, SSO-recommended KCl concentration is 0.038 mM which lies between the deficient and control concentrations. Another testing using 0.038 mM KCl was conducted to verify if the SSO result is suitable for actual fertigation. There is significant improvement in average xylem diameter which increased by 103.501, 112.395, and 105.759 % for Perlas, Diamante, and Mica genotypes, respectively, in using SSO-recommended KCl concentration (**Figure 7**). Phloem average diameter also increased by 114.226, 135.432, and 117.737 % for Perlas, Diamante, and Mica genotypes, respectively (**Figure 7**). These findings proved that SSO is a suitable bioinspired algorithm in improving tomato seedling traits and 0.038 mM of KCl fits the chloride requirements of Perlas, Diamante, and Mica seedlings. By employing 0.038 mM of KCl instead of the conventional 0.05 mM [3,9] also contributes to lessening the volume of inorganic fertilizers injected to the soil system. This approach of bioinspired SSO indirectly contributes to lessening the emission to the environment that might be caused by excess fertigation. Meanwhile, previous studies have shown that applying 80 % irrigation and 80 % of controlled-release KCl-20 % resulted in higher tomato fruit yield in China [48], experimental output-based selection of KCl concentration from 112 to 336 kg/ha for almond cultivation resulted in stronger tree and lesser nut cracks [49], and manual selection from 0, 1, 6 and 40 mM KCl based on maximum net photosynthetic rate of *Prunella vulgaris* L. [50]. Hence, there is no prior comprehensive computational technique used to optimized KCl concentration for crops aside from this current study. It is either by having biological intervention like microorganisms [35] and other interactive crops [36,50] or adjusting in the irrigation-fertigation system by manual selection based on the resulting crop trait [48,49] to compensate the nutrient stress caused by KCl. Here, the potential of bioinspired algorithms was highlighted through the sperm swarm optimization which resulted in more acceptable tomato seedling phenotypes by fertigating with exact KCl nutrient requirement. The developed technique using computational intelligence principle can be extended on improving other nutrients that have specific impacts on crops.

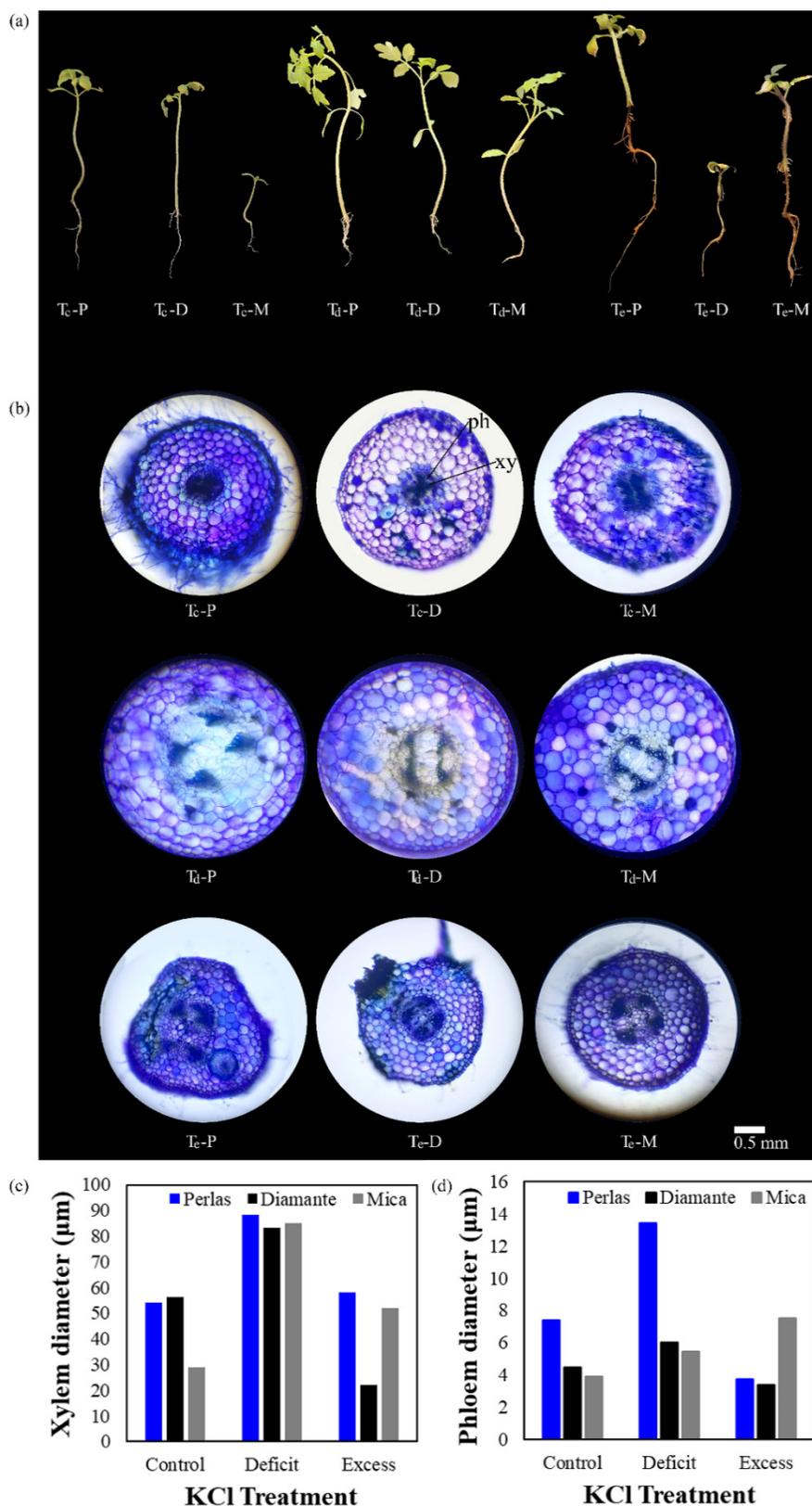


Figure 5 Impacts of KCl stress as observed in the (a) actual harvested tomato seedlings at the end of DAS 37 and (b) transverse cross-section regions of seedling stem showing the vascular tissues that are stained with TBO. Measured (c) xylem (xy) and (d) phloem (ph) mean diameters. Note that Tc, Td, and Te are the control, deficit, and excess KCl treatments; P, D, and M are Perlas, Diamante Max F1, and Mica tomato genotypes.

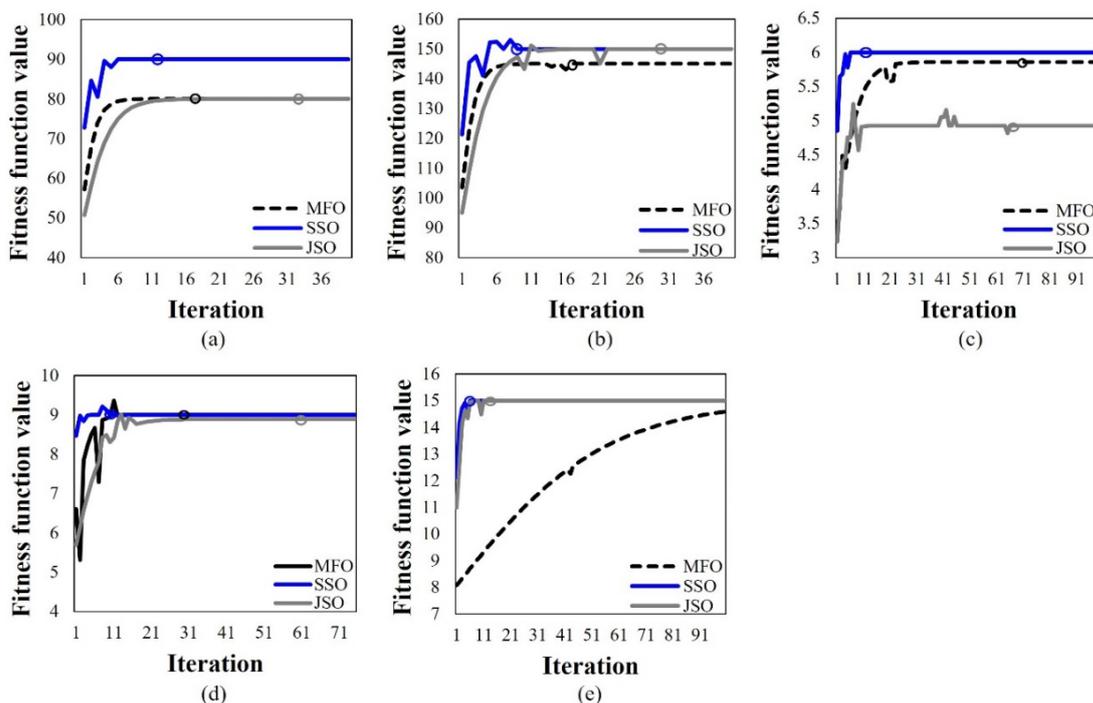


Figure 6 Convergence curve of each phytomorphological phenotype namely (a) root length, (b) shoot length, (c) root fresh weight, (d) shoot fresh weight, and (e) leaf count as optimized by using the configured moth-flame (MFO), sperm swarm (SSO) and jellyfish swarm (JSO) optimizers. Plotted circles are the start of the iteration of convergence.

Table 3 Global best results of potassium chloride concentration as generated by moth-flame, sperm swarm, and jellyfish swarm optimizers for tomato seedling until DAS 37.

Bio-inspired optimizer	KCl concentration (mM)					
	Root length	Shoot length	Root weight	Shoot weight	Leaf count	Average (recommended)
Moth-flame (MFO)	0.012	0.111	0.021	0.005	0.008	0.016
Sperm swarm (SSO)	0.039	0.040	0.037	0.037	0.037	0.038
Jellyfish swarm (JSO)	0.008	0.015	0.015	0.023	0.055	0.019

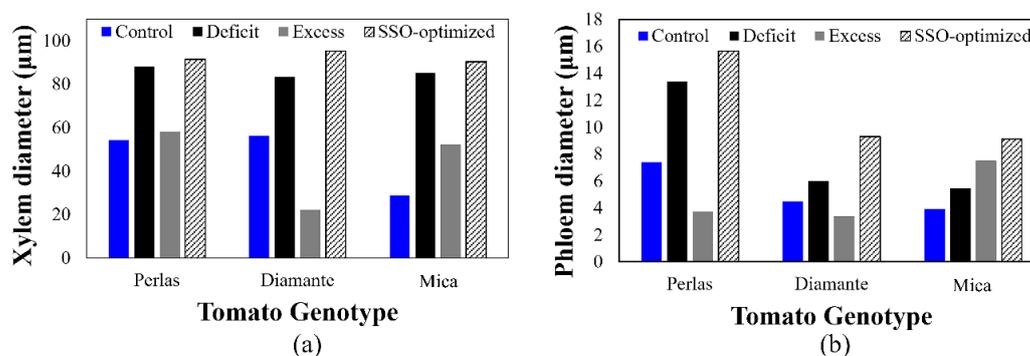


Figure 7 Comparison of measured average xylem and phloem diameters of Perlas, Diamante Max F1, and Mica at DAS 37 as affected by control, deficit, excess, and SSO-optimized KCl concentration (0.038 mM).

Conclusions

This study has shown that potassium chloride is an essential nutrient that enhances the growth productivity of tomato seedlings based on germination, vascular tissues, and phytomorphological architectures. The differential impacts of KCl (control, deficit, and toxic) have been expressed on 3 tomato genotypes namely, Perlas, Diamante Max F1, and Mica. KCl-deficit treatment was confirmed to improve germination rate and vigor index and has an adverse effect on the Timson germination index for the 3 tomato variants. It also promotes primary root growth, heavier root, and shoot length, thicker stem, and induced leaf development. The KCl-excess treatment distinctively promotes a higher root to shoot length ratio for Perlas variant and speeds up the formation of the root as manifested by the rusty orange root color. Three bio-inspired algorithms have been explored to generate the exact KCl concentration that would maximize root and shoot lengths and weights, and leaf count for the first 37 days of cultivation. By using the MSRGP-constructed fitness functions, sperm swarm optimizer generated the global best of 0.038 mM of KCl that provided acceptable growth productivity as evident by wider xylem and phloem vessels which are important vascular tissues as they are responsible for the translocation of nutrients, water, and by-products of photosynthesis inside the plant system. Further research should be done in determining the impacts of KCl on nitrogen consumption of tomato plants and the corresponding growth phenotypes and fruit quality. Also, the use of computational techniques can be applied to cotton, rice, and wheat cultivation in relation to KCl-induced stress.

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