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Nutrient Disorder Symptomology and Refining Leaf Tissue Nutrient Standards of Basil (*Ocimum basilicum* L.)

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Abstract. **Basil (*Ocimum basilicum* L.) is a popular annual herb grown for culinary, medicinal, and ornamental purposes. Existing foliar nutrient sufficiency ranges for basil are based on field-grown plants, which can vary from the nutritional requirements of crops grown in controlled environments. This prompts the need for refined nutrient fertilizer concentration recommendations and foliar nutrient interpretation ranges that are specific to greenhouse-grown basil. The objectives of this study were to determine critical leaf tissue concentrations when disorders were observed, to evaluate the effects of varying macronutrient fertilizer concentrations on basil growth and yield, and to develop foliar nutrient interpretation ranges for greenhouse-grown basil. Basil 'Prospera Compact DMR (PL4)' plants were grown in an automatic recirculating hydroponic system and supplied with a modified Hoagland's solution. To evaluate varying macronutrient applications, eight different concentrations (0, 8, 16, 32, 64, 100, 200, and 300%) of nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), and sulfur (S) were applied, with all other elements held constant. Similarly, micronutrient deficiencies were induced by individually omitting one element from the nutrient solution per treatment. Deficiency symptoms were photographed for all treatments except copper, manganese, and molybdenum, which did not develop visual deficiency symptoms after 57 days, when the experiment was terminated. Plant tissue was collected to measure the plant dry mass and leaf tissue nutrient concentrations. The effects of varying macronutrient fertilizer concentrations were evaluated by comparing three regression models (linear, quadratic, and quadratic plateau) to determine the foliar concentration of each element corresponding with optimal growth. To develop foliar mineral nutrient interpretation ranges for greenhouse-grown basil, additional foliar tissue analysis data (n = 1938) from greenhouse-grown basil were obtained and compiled. By expanding upon the sufficiency range approach, foliar nutrient interpretation ranges for deficient, low, sufficient, high, and excessive values were established for the 12 essential elements. For each element, Normal, Gamma, and Weibull distributions were fitted to the data, and the optimal distribution was chosen according to the lowest Bayesian Information Criterion value. The resulting macronutrient concentration recommendations and foliar interpretation ranges are valuable resources that can aid in troubleshooting nutrient disorders and optimizing growth of greenhouse-grown basil.**

Basil (*Ocimum basilicum* L.), also known as sweet basil, is an herb originating from Asia, Africa, and Central and South America (Paton 1992) grown for many uses, including its unique aroma compounds and oils that make it especially popular for culinary applications (Simon et al. 1999). Its fresh-cut leaves are used for salads, seasoning, and garnishes, while its extracts can be used for many medicinal purposes, including anti-microbial and anti-inflammatory applications (Shahrajabian et al. 2020). Basil foliage and flowers are also used for ornamental purposes, often seen as a bedding plant or in floral arrangements. Although basil is an annual, its popularity requires continuous, year-round production to meet culinary demand. As a result, it is typically grown hydroponically in a controlled environment (Walters and Currey 2015), where growers can control the temperature, nutrition, and light to optimize production.

Previous research has investigated hydroponic basil production, specifically comparing production systems including nutrient film technique and deep flow culture (Walters and Currey 2015), as well as the effects of daily light integral (DLI) on basil growth and tissue nutrient concentrations (Walters and Currey 2018) and the effects of light quality and DLIs on essential oil quality and yield (Chu et al. 2022). Further, research performed by Saha et al. (2016) reported that basil grown in crayfish-aquaponic systems, in comparison with hydroponic systems, exhibited greater growth and yield due to excess nitrogen (N) and phosphorus (P) supplied by fish excreta and unconsumed feed. Similar results were reported by Modarelli et al. (2023), who evaluated hydroponic and aquaponic basil over multiple harvests and found basil grown in the aquaponic system yielded greater dry biomass. Lastly, Rodgers et al. (2022) reported greater biomass and shoot height in basil plants grown in a conventional hydroponic solution and a nutrient-complemented aquaponic solution compared with unsupplemented aquaponic solutions due to higher nutrient applications. These findings suggest that fertilizing with higher N concentrations in hydroponic production systems can promote the growth and yield of basil.

Another important growth parameter to consider for hydroponically grown crops is the nutrient concentration of the fertilizer solution, often measured by electrical conductivity (EC). Currently, the recommended EC for growing hydroponic basil is between 1.2 to 2.2 $mS{\cdot}cm^{-1}$; however, this may depend on the production system (Hosseini et al. 2021; Wortman 2015). A lower EC, and thus a lower nutrient concentration, resulted in lower leaf mass and reduced marketable yield (Wortman 2015). Furthermore, Ren et al. (2022) evaluated the effects of varying EC levels on hydroponic basil and reported greater shoot and leaf fresh weight at 2.0 and 3.0 $mS{\cdot}cm^{-1}$, while an EC of 5.0 $mS{\cdot}cm^{-1}$ resulted in lower fresh weights. This is consistent with previous field and container studies that have reported greater basil biomass with increased N fertilization (Biesiada and Kuś 2010;

Sifola and Barbieri 2006). Conversely, Walters and Currey (2018) reported no effect on growth of hydroponically grown basil as the nutrient solution EC increased from 0.5 to 4.0 $mS \cdot cm^{-1}$.

According to a pictorial guide to nutrient deficiencies in bedding plants (Gibson et al. 2007), greenhouse-grown basil is most commonly deficient in N, magnesium (Mg), and iron (Fe), with N deficiency typically occurring during the first half of the crop cycle. The guide also includes symptomology and corresponding photos of deficiencies in P, potassium (K), calcium (Ca), sulfur (S), boron (B), copper (Cu), manganese (Mn), and zinc (Zn). Macronutrient and micronutrient deficiencies of young basil seedlings have also been studied, with similar symptomology (Song et al. 2024). Borges et al. (2016) evaluated the individual omission of macronutrients from Hoagland's solution, providing deficiency symptomology and the corresponding aerial and root tissue values. Further micronutrient deficiency studies conducted by Paparozzi et al. (2022) evaluated Zn, Fe, and Mn deficiencies in hydroponically grown purple leaf basil (*O. basilicum* L. 'Red Rubin') Another guide to nutrient deficiencies in hydroponic basil created by Mattson and Merrill (2016) also provides visual deficiency symptoms. Two nutrient-specific trade articles focused on Mg deficiency also outline visual symptoms and symptomology progression (Dickson 2019; Mattson 2018). Although these guides are useful for diagnostic purposes, they either do not provide recommended foliar nutrient concentration ranges or do not include all the major macronutrients and micronutrients.

While recommended foliar nutrient sufficiency ranges for basil exist (Bryson and Mills 2015), these guidelines are based on field-grown basil in mineral soils, not hydroponically grown plants. Although these ranges provide baseline target values, nutrients in hydroponic systems greatly differ from field production. The absence of established foliar nutrient standards for hydroponically or greenhouse-grown basil necessitates that current research compares results against the existing guidelines for field production. Providing foliar nutrient recommendations specific to basil grown in a controlled environment would provide a useful guideline for both future research and commercial growers, allowing for more precise interpretations.

As the most popular culinary herb grown in controlled environment agriculture, the economic importance of basil means it is critical for growers to have proper foliar nutrient concentrations, as well as common deficiency symptomology information that can be used to diagnose nutritional disorders. To develop a more robust interpretation model for basil that includes deficient, low, sufficient, high, and excessive ranges, this study was conducted to examine plant nutrient distribution curves for a larger leaf tissue sample set. These ranges were determined for lettuce (*Lactuca sativa* L.) (Veazie et al. 2024a) and pentas (*Pentas lanceolata* Forssk.) (Veazie et al. 2024b). Most leaf tissue nutrient concentration distributions tend to be skewed, and Normal distribution curves are less suitable for developing interpretations. Two models that account for possible skewness are the Gamma and Weibull distribution curves (Cera et al. 2022; Mhango et al. 2021; Slaton et al. 2021; Weibull 1951). Evaluating multiple models allows for more accurate fitting of the data. The objectives of this study were to develop foliar nutrient interpretation ranges for greenhouse-grown basil, determine critical leaf tissue values when disorders are observed, and evaluate the effects of varying macronutrient fertilizer concentrations on hydroponic basil growth.

Materials and Methods

Expt. 1 macronutrient concentration. Organic pelleted F1 basil [*O. basilicum* L. 'Prospera Compact DMR (PL4)'] seeds (Johnny's Selected Seeds, Winslow, ME, USA) were sown on 7 Sep 2023 into premoistened 104-count cell sheets, where each cell measured 3.6 cm tall × 3.4 cm long × 2.3 cm wide (Oasis Rootcubes; Oasis Grower Solutions, Kent, OH, USA), with six seeds per cell. Each sheet was placed in a plastic tray (Landmark Plastic, Akron, OH, USA) atop heated mats set to 22 °C (Ken-Bar, Rochester, NY, USA). Each tray was covered with a humidity dome (Super Sprouter Standard Vented Humidity Dome 7 inches; Hawthorne Gardening Company, Vancouver, WA, USA) and hand-misted with tap water to maintain moisture for 14 d until cotyledon emergence. Seedlings were grown under fluorescent lights (AgroBrite T5 Full Spectrum; Hydrofarm, Petaluma, CA, USA), which provided 17.3 $mol \cdot m^{-2} \cdot d^{-1}$ based on a 24-h photoperiod, and subirrigated with tap water for 12 d. On 2 and 3 Oct 2023, 25 and 26 d after seeds were sown, the seedlings were transplanted into 11.5-cm-diameter (0.8 L) plastic pots (BFG, Burton, OH, USA) containing silica sand [Millersville #2 (0.8 to 1.2 mm diameter); Southern Products and Silica Co., Hoffman, NC, USA]. The plants were grown in a glass-glazed greenhouse at North Carolina State University Horticulture Field Laboratory in Raleigh, NC, USA (35°N latitude) under ambient light with air temperature setpoints of 22.8 °C (day) and 22.0 °C (night). Nutrient treatments began immediately, using an automated recirculating irrigation system constructed out of 10.2-cm-diameter polyvinylchloride pipe (Charlotte Plastics, Charlotte, NC, USA). Automatic irrigation ran once per hour between 7:00 AM and 6:00 PM. Additional details regarding nutrient treatments, formulations, fertilizer salts, and the irrigation system are outlined by Barnes et al. (2012) and Veazie et al. (2022).

Expt. 2 micronutrient deficiencies. Organic pelleted F1 basil 'Prospera Compact DMR (PL4)' seeds (Johnny's Selected Seeds) were sown on 10 Nov 2023 and germinated as outlined in Expt. 1. On 4 Dec 2023, 24 d after seeds were sown, the seedlings were transplanted into 11.5-cm-diameter (0.8 L) plastic pots containing silica sand as previously described, and nutrient treatments began immediately.

Nutrient treatments. Macronutrient treatments for Expt. 1 were subdivided into eight different concentrations of each element (0%, 8%, 16%, 32%, 64%, 100%, 200%, and 300%) of a modified Hoagland's solution (Hoagland and Arnon 1950) (Table 1 lists the corresponding concentrations in $mg \cdot L^{-1}$ for each element). Sodium was avoided when creating the stock solutions and, if used, was held at <15 $mg \cdot L^{-1}$ Na. Chloride concentrations were highest in the N treatments, with the 0% N solution containing 168 $mg \cdot L^{-1}$ Cl. Each macronutrient (N, P, K, Ca, Mg, and S) was evaluated individually. Micronutrient treatments for Expt. 2 consisted of a modified Hoagland's solution adjusted to individually omit one micronutrient [B, Cu, Fe, Mn, molybdenum (Mo), or Zn] per treatment while holding all others constant. All fertilizer solutions were formulated with deionized (DI) water and custom blends of the following individual technical grade salts (Fisher Scientific, Pittsburgh, PA, USA): calcium nitrate tetrahydrate [$Ca(NO_3)_2 \cdot 4H_2O$], potassium nitrate (KNO_3), monopotassium phosphate (KH_2PO_4), magnesium sulfate heptahydrate ($MgSO_4 \cdot 7H_2O$), potassium chloride (KCl), calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$), sodium nitrate ($NaNO_3$), magnesium chloride hexahydrate ($MgCl_2 \cdot 6H_2O$), sodium phosphate monohydrate ($NaH_2PO_4 \cdot H_2O$), sodium sulfate (Na_2SO_4), iron chelated with diethylenetriaminepentaacetic acid (Fe-DTPA), manganese chloride tetrahydrate ($MnCl_2 \cdot 4H_2O$), zinc chloride heptahydrate ($ZnCl_2 \cdot 7H_2O$), copper chloride dihydrate ($CuCl_2 \cdot 2H_2O$), boric acid (H_3BO_3), and sodium molybdate dihydrate ($Na_2MoO_4 \cdot 2H_2O$).

Data collection. Expt. 1 was terminated 28 d after transplant, on 31 Oct 2023, and Expt. 2 was terminated 57 d after transplant, on 30 Jan 2024. For each nutrient treatment, four representative plants were selected for sampling. The most recently matured leaves were sampled for tissue analysis as recommended by Bryson and Mills (2015). The remainder of the shoots were also collected to evaluate the total dry mass for each sample.

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Table 1. Calculations for the modified Hoagland's solution used to determine the impacts of varying macronutrient fertilizer concentrations on the growth of greenhouse-grown basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'].

Element	Fertilizer concn (%)							
	0	8	16	32	64	100	200	300
	Macronutrient fertilizer concentration ($mg·L^{-1}$)							
Nitrogen	0.0	12.0	24.0	48.0	96.0	150.0	300.0	450.0
Phosphorus	0.0	1.63	3.25	6.5	13.0	20.0	40.0	60.0
Potassium	0.0	12.0	24.0	48.0	96.0	150.0	300.0	450.0
Calcium	0.0	4.7	9.8	18.75	37.5	75.0	150.0	225.0
Magnesium	0.0	3.2	6.4	12.8	25.6	40.0	80.0	120.0
Sulfur	0.0	2.0	4.0	8.0	16.0	25.0	50.0	76.0
	Micronutrient fertilizer concentration ($mg·L^{-1}$)							
	Iron	Manganese	Copper	Zinc	Boron	Molybdenum		
All fertilizer treatments	4.02	0.99	0.19	0.20	0.49	0.01		

The values are expressed as percentages of the standard Hoagland's solution. Nutrient concentrations are based on a modified Hoagland's solution in which all elements were held constant except the macronutrient being studied.

The collected leaves and shoots were rinsed in DI water, washed in a 0.5 M HCl solution, and then rinsed in DI water a final time. Immediately after sampling and washing, the leaf tissue and shoots were individually bagged and dried at 70 °C for 48 h in a forced-air oven, and the dry mass was weighed and recorded. Total dry mass for each sample was calculated by adding the dry mass of the recently matured leaves to the dry mass of the remaining shoot. After weighing, leaf tissue was milled through a ≤0.5-mm sieve using a Foss Tecator Cyclotec™ 1093 sample mill (Analytical Instruments, LLC, Golden Valley, MN, USA), placed in vials, and shipped to the US Department of Agriculture–Agricultural Research Service Application Technology Research Unit (Toledo, OH, USA) for analysis as outlined by Boldt and Altland (2021). To determine foliar N concentration, ≈2.5 mg of leaf tissue was placed into tin capsules (EA Consumables, Marlton, NJ, USA) and analyzed with a CHN analyzer (vario MICRO cube; Elementar, Hanau, Germany). To determine all other foliar nutrient concentrations, ≈0.25 g of leaf tissue was combined with 5 mL of nitric acid, heated to 200 °C over 20 min in a programmable microwave (MARS6; CEM Corp., Matthews, NC, USA), and held at 200 °C for an additional 20 min. Once cooled, 1.5 mL of hydrogen peroxide was added, and the samples were reheated to 200°C and held for another 20 min. Once cooled a final time, 12 mL of ultra-purified water (18 MΩ·cm) was added, and the solutions were filtered (Whatman #2). Finally, a 1.3-mL aliquot of solution was diluted with 8.7 mL of 18 MΩ·cm water and analyzed using inductively coupled plasma–optical emission spectroscopy (iCAP 6300 Duo; Thermo Electron Corp., Waltham, MA, USA).

Data analysis. This study was a randomized block design, and plant dry mass and leaf nutrient concentration data obtained from all experiments were analyzed using SAS (version 9.4; SAS Institute, Cary, NC, USA). Data from Expt. 1 were subjected to the general linear model (PROC GLM) and analyzed for differences among treatments for each element, and means were separated by least significant differences at $P \leq 0.05$. During data analysis, certain treatments (150 $mg·L^{-1}$ K, 4 $mg·L^{-1}$ S, and 16 $mg·L^{-1}$ S) had unexpectedly low mean values for plant dry mass due to faulty irrigation pumps. These values were inconsistent with the overall trends of the data set, were determined to be outliers, and consequently were omitted from the statistical analysis. The regression procedure (PROC REG) and nonlinear regression procedure (PROC NLIN) were used to determine whether a linear, quadratic, or quadratic plateau model was the best fit for each element, based on evaluating the statistical significance and R^2 of each model. When a quadratic plateau was the best fit, X_0 values were obtained to indicate the value at which growth and leaf tissue concentration plateaued, above which an increased nutrient fertilizer concentration did not result in higher biomass or foliar concentration. Data obtained from Expt. 2 were subjected to analysis of variance (ANOVA) using PROC ANOVA.

Expt. 3 nutrient distribution curves. A third aspect of the research was conducted after the conclusion of Expts. 1 and 2. Because a larger sample size better represents the population and provides more accurate results, a more robust data set was needed. Further, the incorporation of data from varying sources reduces the potential bias that can occur when establishing a distribution based on data from a single research study. Consequently, a larger data set was established by incorporating additional foliar tissue analysis data from multiple cultivars of greenhouse-grown basil, obtained from university studies and public analytical laboratories, with data obtained from Expts. 1 and 2 for a total of 1938 samples (Table 2). Distribution analyses were modeled for each element using RStudio (version 2024.04.2; RStudio Team 2024). Excessive outliers with values greater than what is biologically reasonable were removed before further analysis. Normal, Gamma, and Weibull distributions were fitted to the data (Cera et al. 2022; Mhango et al. 2021; Slaton et al. 2021; Weibull 1951). *P* values were

Table 2. Sources of greenhouse-grown basil (*Ocimum basilicum* L.) leaf tissue nutrient data ($n = 1938$) used to develop tissue nutrient interpretation ranges.

Source	Sample size	Sample type	Citation
North Carolina Department of Agriculture Laboratory	474	Diagnostic	Unpublished diagnostic and predictive grower samples
North Carolina State University	496	Research	Present macronutrient fertilizer concentration and micronutrient deficiency studies
North Carolina State University	135	Research	Substrate blend evaluation studies, unpublished
North Carolina State University	50	Research	Evaluating the effect of EC on plant growth, unpublished
USDA-ARS	14	Research	Boldt and Altland (2022)
USDA-ARS	130	Research	Evaluation of placement of semitransparent photovoltaic panels on a greenhouse roof on plant growth, unpublished
USDA-ARS	216	Research	Evaluation of Si supplementation during chronic cold stress, unpublished
Michigan State University	21	Research	Evaluating the effect of increasing N rates on basil growth, unpublished
University of Nebraska	32	Research	Paparozzi et al. (2022)
Iowa State University	30	Research	Unpublished
Iowa State University	20	Research	Currey et al. (2020)
Iowa State University	40	Research	Evaluating the effect of controlled release and water-soluble fertilizer on plant growth, unpublished
J. R. Peters Laboratory	280	Diagnostic	Diagnostic grower samples, unpublished

USDA-ARS = US Department of Agriculture–Agricultural Research Service.

calculated based on the Shapiro–Wilk test for normality for Normal and Gamma distributions and the Kolmogorov–Smirnov test for the Weibull distribution. The Bayesian Information Criterion (BIC) was calculated for each distribution, and the optimal distribution was selected based on the lowest BIC (Table 3). The results were illustrated using ggplot2 (Wickham 2011) in R. Scott's rule (Scott 1979) was used to determine bin width for optimal data visualization of macronutrients. Bin width for micronutrient distributions was determined using the Freedman–Diaconis rule (Freedman and Diaconis 1981). For macronutrients (N, P, K, Ca, Mg, and S), the sufficiency range was defined as the range between the 0.25 and 0.75 quantiles within a 95% confidence interval. The deficient and low ranges were based on the left tail (the lowest 2.5% of samples) and the area between the 0.025 and the 0.25 quantiles, respectively. The high range was determined by the area between 0.75 and the 0.975 quantiles, and the excessive range was classified by the right tail (the highest 2.5% of samples). For micronutrients (B, Cu, Fe, Mn, Mo, and Zn), the threshold between the deficient and low ranges was defined by the lowest 5% of samples of a 90% confidence interval. Likewise, the threshold between the high and excessive ranges was defined by the top 5% of observations within a 90% confidence interval. The work of Veazie et al. (2024a, 2024b) contains additional details about the development of the nutrient distribution curves.

Results and Discussion

Nitrogen

Nitrogen deficiency symptoms. In Expt. 1, initial N deficiency symptoms were stunted growth and light-green foliage (Figs. 1A and 2). As symptoms progressed, foliage continued to lighten in color, and the entire plant from the lowest nutrient treatments became chlorotic. These symptoms were first observed in plants that received the lowest two N concentrations (0 and 12 mg·L^{-1} N). Upon termination of the experiment, plants that received 0 mg·L^{-1} N had developed necrotic patches on the lower and middle foliage, and plants that received 24 and 48 mg·L^{-1} N had also developed deficiency symptoms. Plants supplied with ≥96 mg·L^{-1} N did not develop any deficiency symptoms after 28 d. The highest N fertilizer concentrations, 300 and 450 mg·L^{-1} N, did not develop any toxicity symptoms.

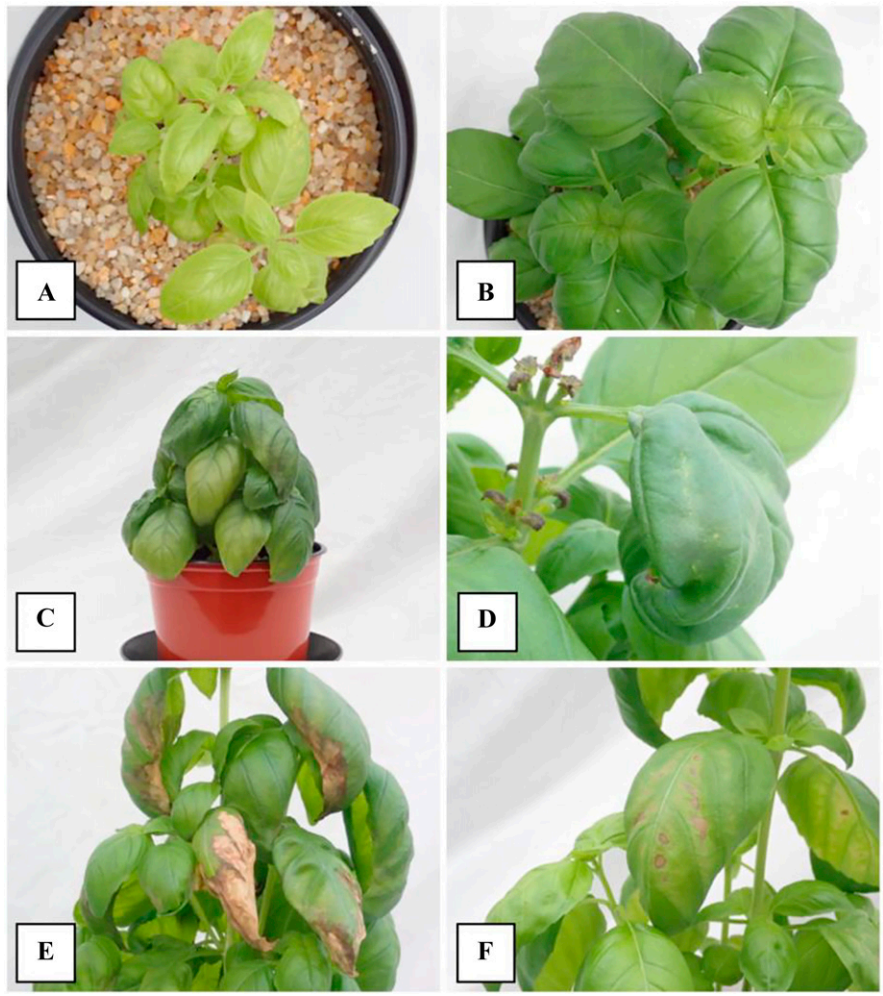

Fig. 1. Basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'] visual macronutrient deficiency symptoms. Nitrogen-deficient plants (**A**) exhibited stunted growth and light green foliage. Phosphorus-deficient plants (**B**) primarily exhibited stunted growth and faint chlorosis of the foliage. Potassium-deficient plants (**C**) displayed stunted growth and interveinal chlorosis of the lower leaves, beginning at the margins; leaves also grew in a downward and slightly cupped orientation. Calcium-deficient plants (**D**) exhibited death of the growing tips, cupped leaves, and marginal necrosis due to the leaves being unable to fully expand. Magnesium-deficient plants (**E**) exhibited interveinal chlorosis of the lower foliage, which quickly progressed into largely necrotic leaves, starting near the leaf margin and progressing inward. Sulfur-deficient plants (**F**) were stunted and developed interveinal chlorosis of the middle foliage, which developed necrotic spotting.

Table 3. Bayesian Information Criterion (BIC) values for Normal, Gamma, and Weibull distribution models used to develop sufficiency range approach distribution models for each of the 12 essential elements in greenhouse-grown basil (*Ocimum basilicum* L.).

	BIC Value		
Element	Normal	Gamma	Weibull
Nitrogen	5,801.98	6,361.72	**5,720.45**
Phosphorous	1,930.38	**1,601.85**	1,672.12
Potassium	**7,200.96**	7,631.77	7,225.35
Calcium	**2,957.67**	2,981.23	3,080.55
Magnesium	749.81	**303.79**	633.41
Sulfur	−2,921.11	**−3,042.60**	−2,894.38
Boron	16,381.00	**15,743.70**	16,129.88
Copper	14,968.45	**12,596.46**	12,720.57
Iron	20,266.09	**19,590.82**	20,045.87
Manganese	21,698.66	**20,837.24**	21,005.27
Molybdenum	3,254.21	1,132.89	**1,120.30**
Zinc	19,603.79	**18,727.71**	18,891.47

The lowest BIC value is in bold type, indicating the selected model.

Nitrogen leaf tissue biomass and accumulation. After 28 d, plants from the control group, which received 150 mg·L^{-1} N, had a dry mass 17.4× greater than the plants that received 0 mg·L^{-1} N (Fig. 3A). Overall, the relationship between plant dry mass and N fertilizer concentration was best modeled by a quadratic plateau, with a plateau observed at 53.2 mg·L^{-1} N (Fig. 3A). This suggests that N fertilizer concentrations greater than 53.2 mg·L^{-1} N do not result in significantly greater dry mass. On the contrary, N accumulation in leaf tissue was best modeled by a quadratic equation, with no plateau in N foliar concentration observed. Nitrogen concentration in the leaf tissue increased from 0.97% to 6.25% N as fertilizer concentrations increased from 0 to 450 mg·L^{-1} N (Fig. 3B). The most rapid increase in N foliar concentration occurred at the intermediate fertilizer concentrations, while relatively smaller increases were observed at the lower and higher fertilizer concentrations. These findings demonstrate that although plants continued to accumulate N at fertilizer concentrations above 53.2 mg·L^{-1} N, the accumulation was luxury consumption and did not result in greater biomass (Marschner 1995). Walters and Currey (2018) reported a similar trend, with N foliar concentrations increasing with an increasing fertilizer solution EC, while no impact on growth was observed.

Nitrogen nutrient distribution curve. Of the three models evaluated, the Weibull distribution had the lowest BIC value and was determined

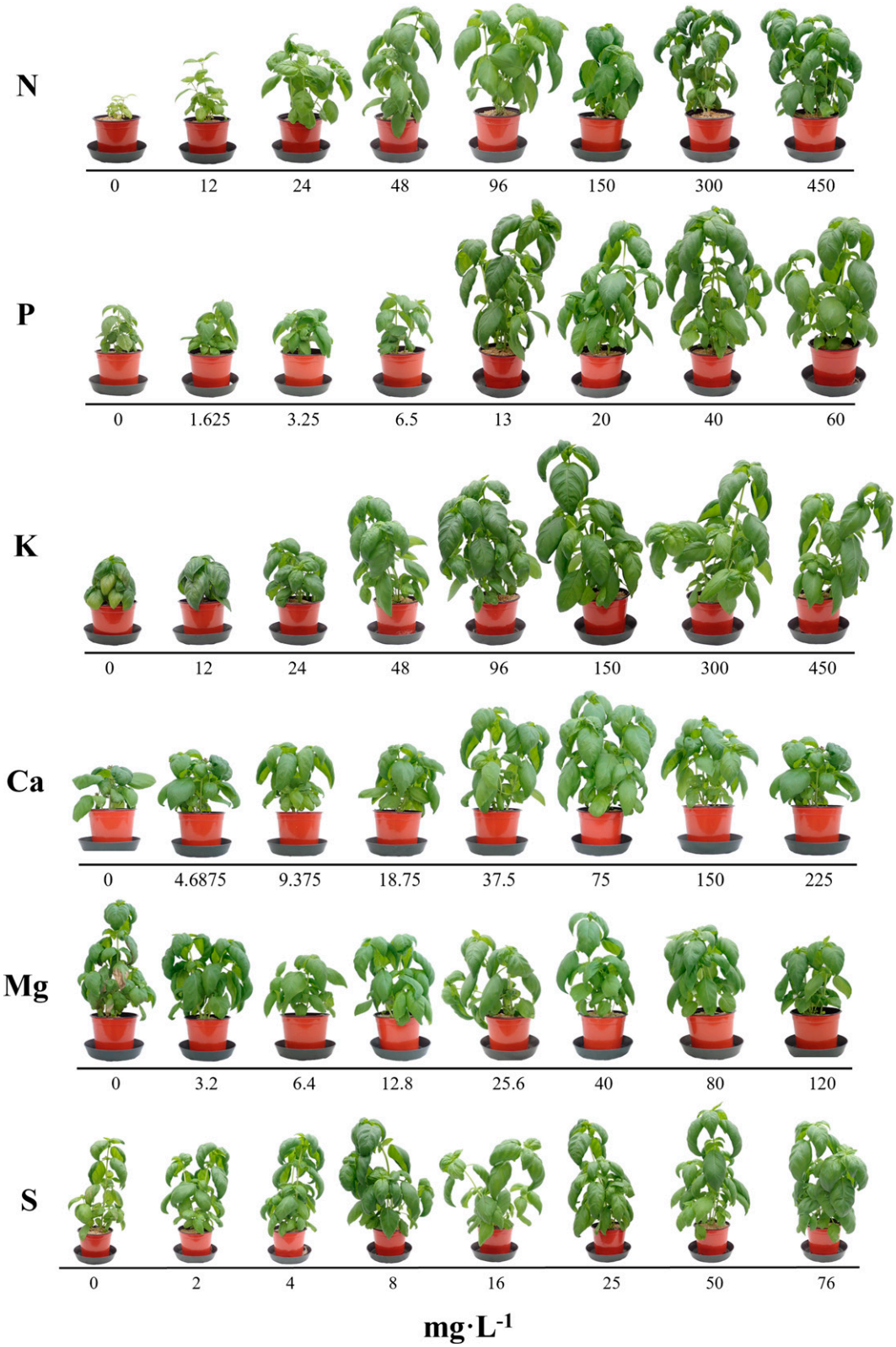

Fig. 2. Visual impact of nitrogen (**N**), phosphorus (**P**), potassium (**K**), calcium (**Ca**), magnesium (**Mg**), and sulfur (**S**) fertilizer concentrations on hydroponically grown basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)']. Eight concentrations of each nutrient based on a Modified Hoagland's solution were evaluated. All photos were taken after 28 d of growth.

to be the best fit for N foliar concentrations (Table 3; Fig. 4A). The recommended sufficiency range of 4.13% to 5.54% N (Table 4) narrows the current range of 4% to 6% N defined by Bryson and Mills (2015). The deficiency value threshold of 2.63% N is supported by the N foliar concentrations (≤1.92% N; Fig. 3B) of symptomatic plants that received the four lowest N fertilizer concentrations. The distribution also established the excessive foliar concentration range as >6.65% N. While N toxicity is uncommon, excessive N application can result in luxury consumption and may inhibit the uptake of other essential elements, including B, Cu, and K (Marschner 1995).

Although a plateau in total basil dry mass occurred at 53.2 mg·L^{-1} N, plants supplied

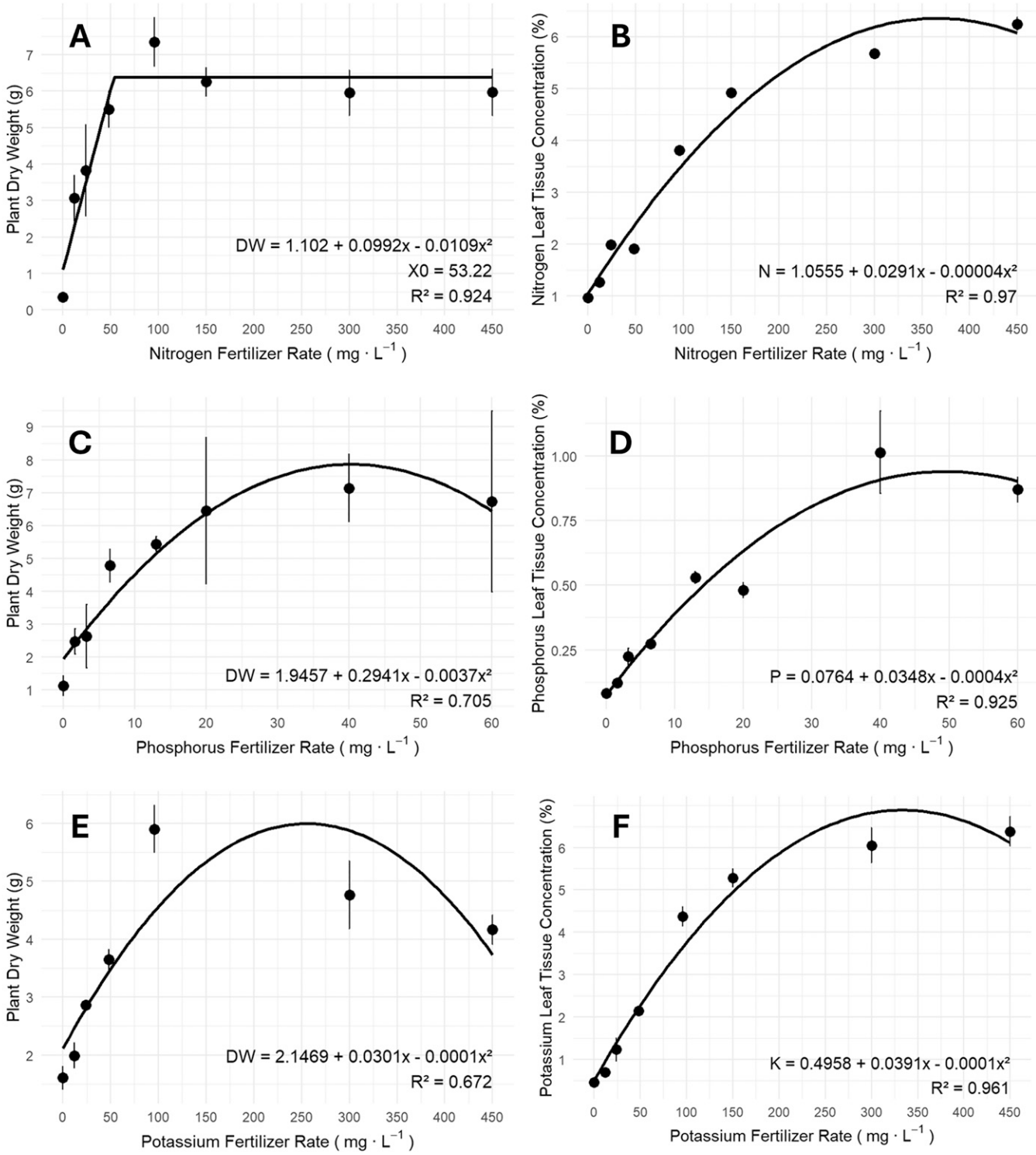

Fig. 3. Impact of nitrogen (**A–B**), phosphorus (**C–D**), and potassium (**E–F**) fertilizer concentrations on plant dry mass and elemental leaf tissue concentration of greenhouse-grown basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'] (n = 1938). Eight concentrations of each nutrient based on a modified Hoagland's solution were evaluated.

with less than 150 mg·L^{-1} N had N foliar concentrations below the recommended sufficiency range of 4.13% to 5.54% N (Table 4). While the dry mass of basil plants was not affected by a N fertilizer concentration as low as 48 mg·L^{-1} N, deficiency symptoms were still evident. Considering both the dry mass and foliar concentration, 150 mg·L^{-1} N is recommended to ensure visually healthy basil plants and to maximize yield without overapplication of N. Nitrogen applications as low as 53.2 mg·L^{-1} N may not negatively affect yield, but N foliar concentrations are likely to fall below the recommended sufficiency range. While N fertilizer concentrations as high as 450 mg·L^{-1} N would still increase N foliar concentration, excessive application did not promote greater yield and may inhibit the uptake of other essential nutrients, particularly K and Ca (Bryson and Mills 2015).

Phosphorus

Phosphorus deficiency symptoms. Symptoms of P deficiency were stunted growth and light-green foliage, which occurred on plants that received the two lowest P concentrations, 0 and 1.63 mg·L^{-1} P (Figs. 1B and 2), within 14 d. After 28 d, plants supplied with 3.25 mg·L^{-1} P also exhibited deficiency symptoms. Plants that received 6.5 mg·L^{-1} P were noticeably stunted but did not exhibit

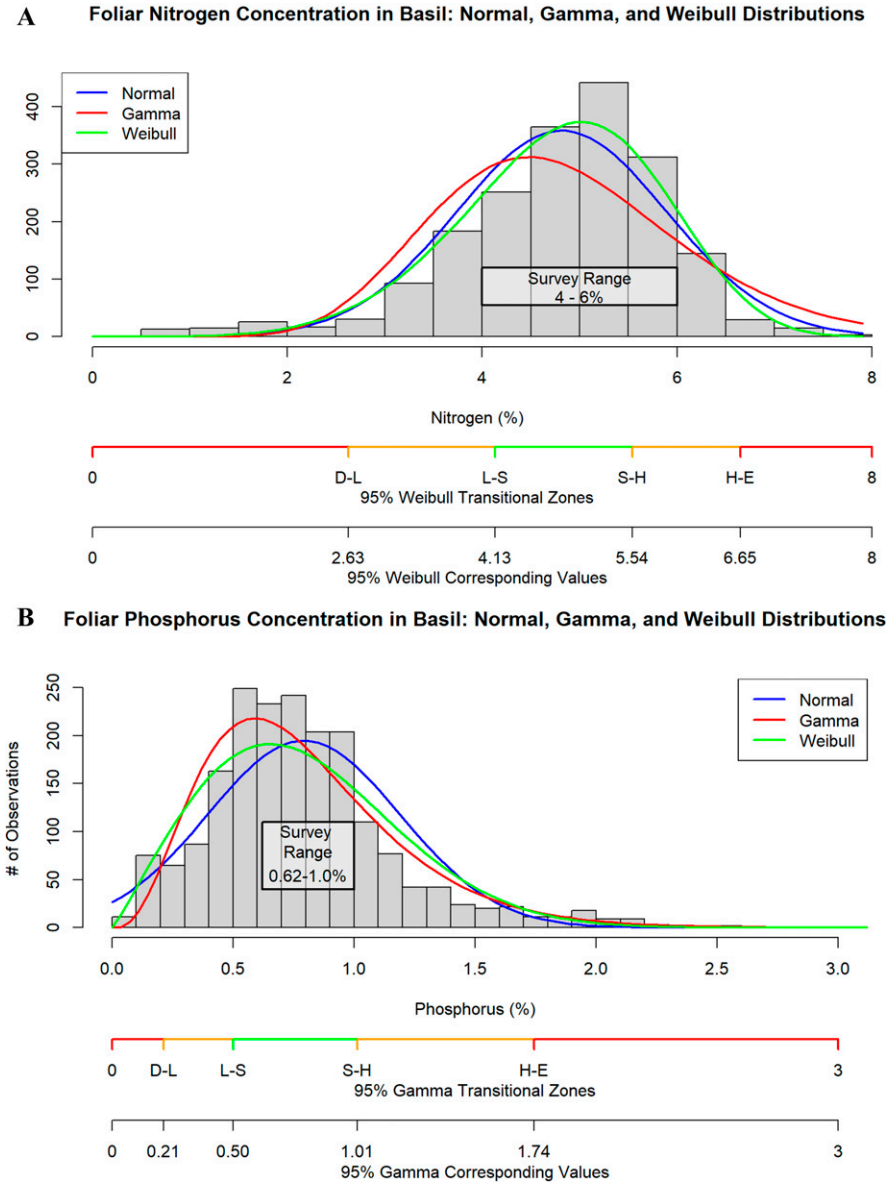

Fig. 4. Nitrogen (**A**) and phosphorus (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 2.5%, 25%, 75%, and 97.5% of sample observations (n = 1938), respectively. Survey ranges from Bryson and Mills (2015) are overlayed for reference.

any other deficiency symptoms. The plants that received 13, 20, 40, or 60 mg·L^{-1} P did not develop visible deficiency or toxicity symptoms at any time during the experiment.

Phosphorus leaf tissue biomass and accumulation. After 28 d of growth, plant dry mass increased as P concentration increased. The relationship between P fertilizer concentration and plant dry mass was best modeled by a quadratic equation, with the highest plant dry mass observed at 40 mg·L^{-1} P (Fig. 3C). The relationship between P fertilizer concentration and P foliar concentration was also best represented by a quadratic model, with the greatest P foliar concentration observed at 40 mg·L^{-1} P (Fig. 3D). Lower plant dry mass and P foliar concentrations occurred at 60 mg·L^{-1} P. With yield being the top priority in basil production, these findings suggest an optimal fertilizer concentration of 40 mg·L^{-1} P to maximize yield without overapplication.

Phosphorus nutrient distribution curve. Based off the BIC values (Table 3), a Gamma distribution best represented the P leaf tissue data (Fig. 4B). The recommended sufficiency range, 0.50% to 1.01% P (Table 4), expands the previously suggested range of 0.62% to 1.00% P (Bryson and Mills 2015). The deficiency threshold was 0.21% P, and plants that received 0 and 1.63 mg·L^{-1} P (Fig. 3D), which exhibited deficiency symptoms, had P foliar concentrations below this threshold value. The plants supplied with 3.25 mg·L^{-1} P, which also exhibited deficiency symptoms, had a P foliar concentration that fell in the low range. The model determined the value between high and excessive P foliar concentrations to be 1.74% P. While excessively high P foliar concentrations may inhibit plant growth, the P concentration of plants that received 40 and 60 mg·L^{-1} P did not exceed the sufficiency range.

Although limited information exists on the effects of P concentrations on hydroponically grown basil, research on container-grown basil has yielded similar results. Currey et al. (2020) reported the greatest shoot dry mass in basil supplied with 20 or 40 mg·L^{-1} P, compared with those that received less. While basil plants in our study supplied with 40 mg·L^{-1} P were

Table 4. Revised nutrient distribution interpretation ranges for greenhouse-grown basil (*Ocimum basilicum* L.) defined from the distribution models, denoting deficient, low, sufficient, high, and excessive foliar elemental concentrations based on the sufficiency range approach using 1938 samples.

		Nutrient ranges				
Element	Unit	Deficient	Low	Sufficient	High	Excessive
Nitrogen	%	<2.63	2.63–4.13	4.13–5.54	5.54–6.65	>6.65
Phosphorous	%	<0.21	0.21–0.50	0.50–1.01	1.01–1.74	>1.74
Potassium	%	<1.39	1.39–3.81	3.81–5.80	5.80–7.47	>7.47
Calcium	%	<0.79	0.79–1.53	1.53–2.11	2.11–2.92	>2.92
Magnesium	%	<0.29	0.29–0.52	0.52–0.88	0.88–1.35	>1.35
Sulfur	%	<0.16	0.16–0.26	0.26–0.42	0.42–0.60	>0.60
Boron	mg·kg^{-1}	<16.2	16.2–26.3	26.3–46.6	46.6–66.2	>66.2
Copper	mg·kg^{-1}	<1.1	1.1–4.3	4.3–16.6	16.6–33.2	>33.2
Iron	mg·kg^{-1}	<51.0	51.0–83.1	83.1–147.6	147.6–210.0	>210.0
Manganese	mg·kg^{-1}	<27.6	27.6–58.4	58.4–132.6	132.6–213.0	>213.0
Molybdenum	mg·kg^{-1}	—[i]	<0.1	0.1–0.8	0.8–2.0	>2.0
Zinc	mg·kg^{-1}	<14.8	14.8–32.8	32.8–77.6	77.6–127.0	>127.0

[i] Concentration was too low to report.

11% bigger by dry mass than plants supplied with 20 mg·L^{-1} P, the P foliar concentration was within the suggested sufficiency range for both treatments. Because P is a finite, irreplaceable natural resource (Van Vuuren et al. 2010), growers may consider applying 20 mg·L^{-1} P to prevent overapplication while still maintaining healthy plant growth and conserving it as a resource.

Potassium

Potassium deficiency symptoms. The primary symptom of K deficiency was stunted growth and interveinal chlorosis of the lower leaves, beginning at the margins (Fig. 1C). Severe deficiency symptoms included downward cupping of the leaves and necrotic spotting near the margins of lower leaves. Plants that received 0 or 12 mg·L^{-1} K were the first to exhibit symptoms within 14 d, with both the new and old leaves becoming chlorotic. After 28 d of growth, plants that received 24 mg·L^{-1} K had significantly less dry mass than plants supplied with higher K concentrations (Figs. 2 and 3E), although no other symptoms were present. Plants that received 48, 96, 150, 300, or 450 mg·L^{-1} K did not develop visual deficiency symptoms during the experiment.

Potassium leaf tissue biomass and accumulation. After 28 d of growth, basil plants that received 96 mg·L^{-1} K had the greatest dry mass (Fig. 3E). Due to the omission of the control treatment (150 mg·L^{-1} K), plants that received 96 mg·L^{-1} K were used for comparison. The relationship between K fertilizer concentration and basil dry mass was best modeled by a quadratic equation. Plants supplied with 300 or 450 mg·L^{-1} K had 19% and 29% less dry mass, respectively, than plants that received 96 mg·L^{-1} K. Likewise, the relationship between K fertilizer concentration and K foliar concentration was also best modeled by a quadratic equation. The highest K foliar concentration was observed at 450 mg·L^{-1} K (Fig. 3F), despite lower plant dry mass. Plants that received 300 or 450 mg·L^{-1} K both had a 52% lower Mg foliar concentration than plants supplied with 96 mg·L^{-1} K. These treatments also had 32% and 59% lower Ca foliar concentrations, respectively. This is likely due to the antagonistic effect that K has on and Mg and Ca uptake (Bryson and Mills 2015). As a result, excessively high K foliar concentrations inhibited the uptake of Mg and Ca, which likely inhibited plant growth.

Potassium nutrient distribution curve. A Normal distribution best fit the K foliar concentration data based on the BIC values and visual fitness (Table 3; Fig. 5A). The recommended sufficiency range of 3.81% to 5.80% K (Table 4) suggests a much higher K foliar concentration than the previously recommended range of 1.55% to 2.05% K (Bryson and Mills 2015). While our range is much higher, it is corroborated by the K foliar concentration of the control plants, which received 150 mg·L^{-1} K (Fig. 3F). The deficiency range was defined as <1.39% K, which aligns with the leaf tissue values of plants that received 0, 12, or 24 mg·L^{-1} K (Fig. 3F). The excessive range was defined as >7.47% K.

To optimize yield and avoid overapplication, a fertilizer concentration of 96 mg·L^{-1} K is recommended for hydroponically grown basil plants. Plants grown at this fertilizer concentration had the highest total dry mass and had a K foliar concentration falling within the sufficiency range. While plants given 48 mg·L^{-1} K did not exhibit deficiency symptoms and also had a K foliar concentration within the sufficiency range, the dry mass was 38% lower than that of plants given 96 mg·L^{-1} K. Fertilizer concentrations greater than 96 mg·L^{-1} K would not be recommended due to the antagonistic effects on uptake of other elements, especially Mg. Avoiding Mg antagonism is especially important due to basil's higher Mg requirement than other leafy greens (Bryson and Mills 2015; Mattson 2018).

Calcium

Calcium deficiency symptoms. Visual symptoms of Ca deficiency were stunted growth, death of the growing tips, and cupped leaves (Fig. 1D). Symptoms were most prominent in plants that received the two lowest Ca fertilizer concentrations, 0 and 4.7 mg·L^{-1} Ca, with plants in both treatments exhibited death of the apical meristem and pronounced leaf cupping. Plants that received 9.38 mg·L^{-1} Ca also exhibited stunted growth and cupped leaves, although death of the growing tip was not observed. Although the plants supplied with 0, 4.7, 9.8, or 18.75 mg·L^{-1} Ca were visibly smaller than plants that received higher Ca fertilizer concentrations (Fig. 2), there were no

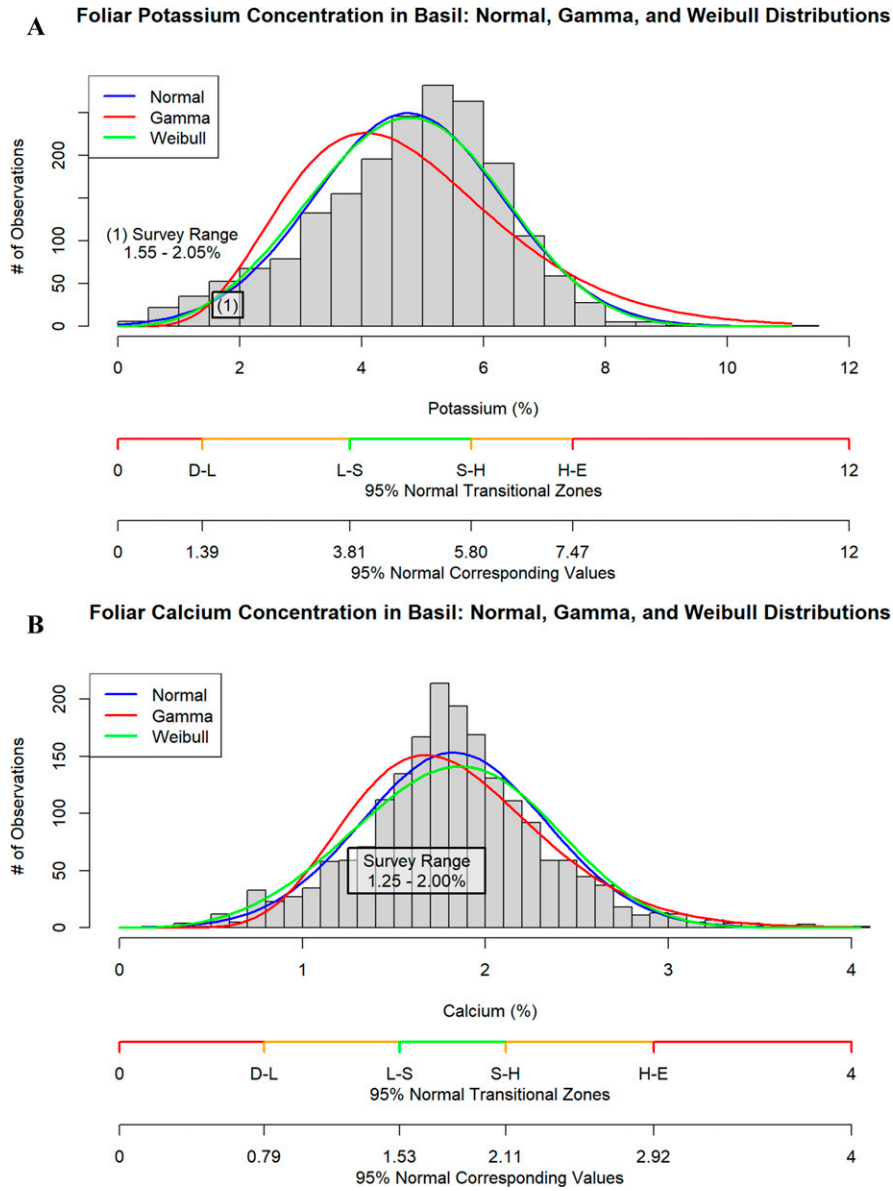

Fig. 5. Potassium (**A**) and calcium (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 2.5%, 25%, 75%, and 97.5% of sample observations (n = 1938), respectively. Survey ranges from Bryson and Mills (2015) are overlayed for reference.

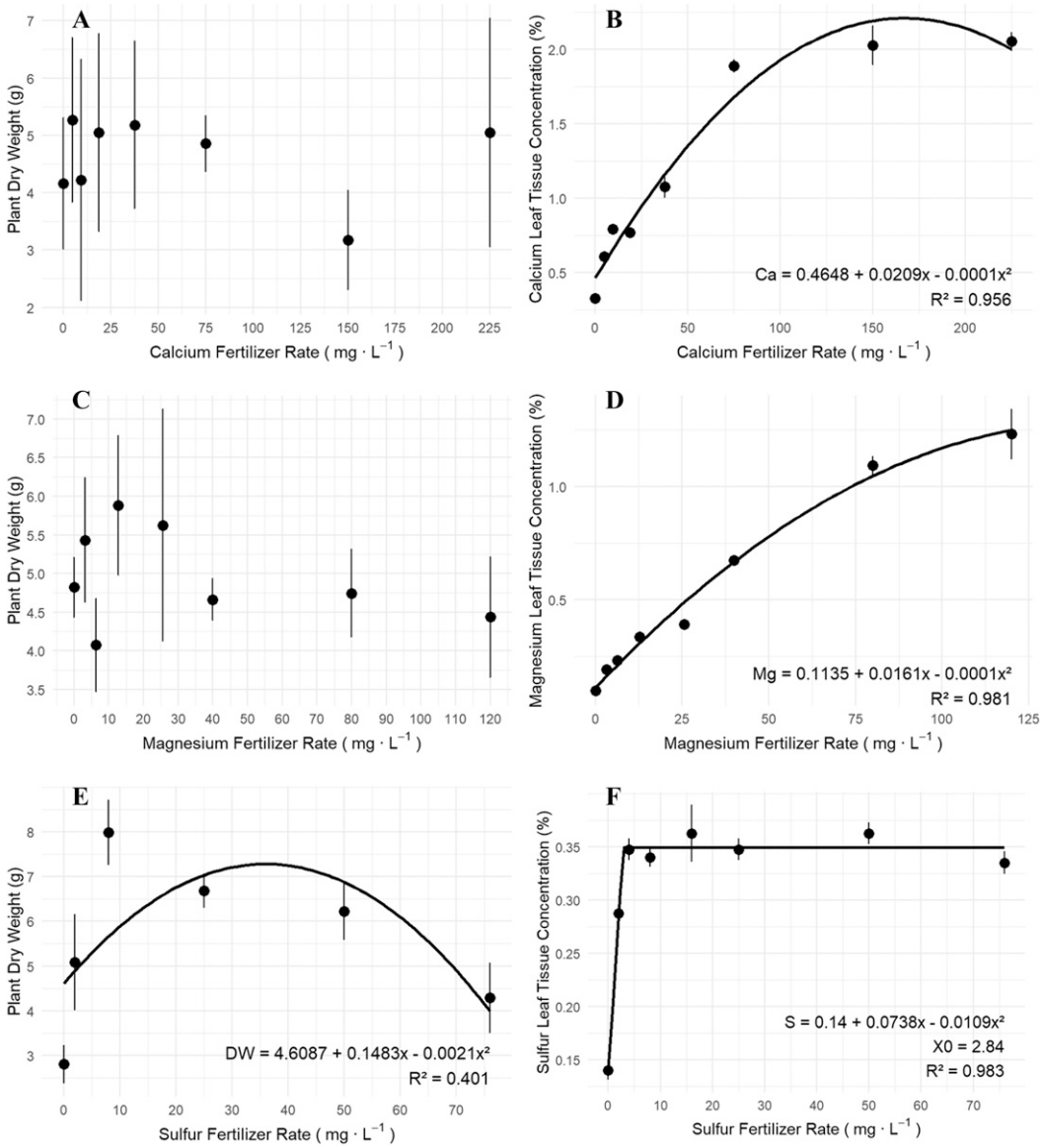

Fig. 6. Impact of calcium (**A–B**), magnesium (**C–D**), and sulfur (**E–F**) fertilizer concentrations on plant dry mass and elemental leaf tissue concentration of greenhouse-grown basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'] (n = 1938). Eight concentrations of each nutrient based on a modified Hoagland's solution were evaluated.

significant differences in dry mass among treatments (Fig. 6A). While plants supplied with 9.8 mg·L^{-1} Ca appeared visually stunted, no other symptoms were observed during the experiment. Plants from the remaining treatments, 37.5, 75, 150, and 225 mg·L^{-1}, Ca did not develop symptoms during the experiment.

Calcium leaf tissue biomass and accumulation. While plants grown at low Ca concentrations exhibited clear deficiency symptoms, no relationship between plant dry mass and Ca fertilizer concentration was observed among the treatments. This could be attributed to a combination of sampling time and thickened plant tissue due to Ca deficiency. While a longer growing period may lead to differences in dry mass among the treatments, commercial crops are often grown for a similar period of time. Research on grapevine (*Vitis vinifera* L.) and bush bean (*Phaseolus vulgaris* L.) reports that Ca deficiency can increase lignification and thickening of leaf tissue (Duan et al. 2022; Wallace et al. 1980), which may have contributed to the similar dry masses of Ca-deficient basil plants compared with the control treatment. The relationship between Ca fertilizer concentration and Ca foliar concentration was best modeled by a quadratic equation, with the maximum foliar concentration seen between 150 and 225 mg·L^{-1} Ca (Fig. 6D). Notably, plants grown with 150 or 225 mg·L^{-1} Ca had low Mg foliar concentrations due to the antagonistic relationship between Ca and Mg (Bryson and Mills 2015). While plants from the 150 mg·L^{-1} Ca treatment still had Mg foliar concentrations within the sufficient range, plants from the 225 mg·L^{-1} Ca treatment had Mg foliar concentrations that fell within the low range.

Calcium nutrient distribution curve. Of the three distributions, a Normal distribution had the lowest BIC value and best represented Ca foliar concentration (Table 3; Fig. 5B). The defined Ca sufficiency range was 1.53% to 2.11% Ca (Table 4), which suggests a slightly higher Ca foliar concentration than the previously recommended range of 1.25% to 2.00% Ca (Bryson and Mills 2015). Plants from the control treatment, which received 75 mg·L^{-1} Ca, had a Ca foliar concentration of 1.89%, supporting this sufficiency range. The deficiency range, <0.79% Ca, includes the Ca foliar concentration of plants that received the two lowest Ca concentrations, 0 and 4.7 mg·L^{-1} Ca (Fig. 6B), and exhibited severe deficiency symptoms. The excessive range was defined as Ca foliar concentrations >2.92%.

Despite minimal differences in dry mass among the treatments, all plants that received a Ca fertilizer concentration <75 mg·L^{-1} had a Ca foliar concentration below the recommended sufficiency range. Although plants from the 18.75 and 37.5 mg·L^{-1} Ca treatments

did not exhibit clear deficiency symptoms, symptoms may have arisen given a longer growing period. Plants that received the two highest Ca concentrations, 150 and 225 mg·L^{-1}, had Ca foliar concentrations within the sufficiency range but contained low Mg foliar concentrations. As a result, it is recommended that basil plants be supplied with a maximum of 75 mg·L^{-1} Ca to ensure sufficient Ca foliar concentrations without inducing Mg deficiency. Further research is necessary to determine whether a lower fertilizer concentration, such as 18.75 or 37.5 mg·L^{-1} Ca, may be sufficient for growing healthy basil plants without negatively affecting yield.

Magnesium

Magnesium deficiency symptoms. The primary symptom of Mg deficiency was interveinal chlorosis of the lower foliage, with severely deficient plants exhibiting interveinal necrosis toward the margins of the leaves (Figs. 1E and 2). Plants that received 0 mg·L^{-1} Mg were the only plants to develop symptoms during the experiment, beginning with interveinal chlorosis of the older leaves after 21 d. Upon completion of the experiment, 28 d after transplant, the older leaves had become mostly necrotic, starting at the margins. With the exception of plants that received 12.8 mg·L^{-1} Mg, there was no significant difference in dry mass among the treatments (Fig. 6C). Plants from the 12.8 mg·L^{-1} Mg treatment had a dry mass that was significantly greater than the other treatments.

Magnesium leaf tissue biomass and accumulation. Despite deficiency symptoms in plants that received 0 mg·L^{-1} Mg, no significant relationship between Mg fertilizer concentration and plant dry mass occurred. Although there were minimal differences among the dry mass, the relationship between Mg fertilizer concentration and Mg foliar concentration was best modeled by a quadratic regression (Fig. 6D). While Mg foliar concentrations increased with higher Mg fertilizer concentrations, a lack of concurrent increase in dry mass suggests that high Mg fertilizer concentrations led to luxury consumption, where the plant accumulates more Mg than is necessary for optimizing yield. Due to the antagonistic effect of Mg on K and Ca uptake, there were slightly lower K and Ca foliar concentrations in plants that received 80 or 120 mg·L^{-1} Mg. While the K foliar concentrations still fell within the sufficiency range, plants supplied with 120 mg·L^{-1} Mg had Ca concentrations that fell below the sufficiency range.

Magnesium nutrient distribution curve. A Gamma distribution had the lowest BIC value and best represented Mg foliar concentration (Table 3; Fig. 7A). The defined sufficiency range, 0.52% to 0.88% Mg (Table 4), lowers the range presented by Bryson and Mills (2015). This range is also corroborated by the Mg foliar concentration (0.68% Mg) of the control plants, which received 40 mg·L^{-1} Mg (Fig. 6D). The deficiency range was defined as Mg foliar concentrations <0.29%. The Mg foliar concentrations of plants that received 0, 3.2, or 6.4 mg·L^{-1} Mg fell within this range. Although plants supplied with 3.2 or 6.4 mg·L^{-1} Mg did not exhibit visual deficiency symptoms during the experiment, symptoms may have arisen given more time. While an excessive range of >1.35% Mg was established, plants that received 80 or 120 mg·L^{-1} Mg did not have excessive Mg foliar concentrations.

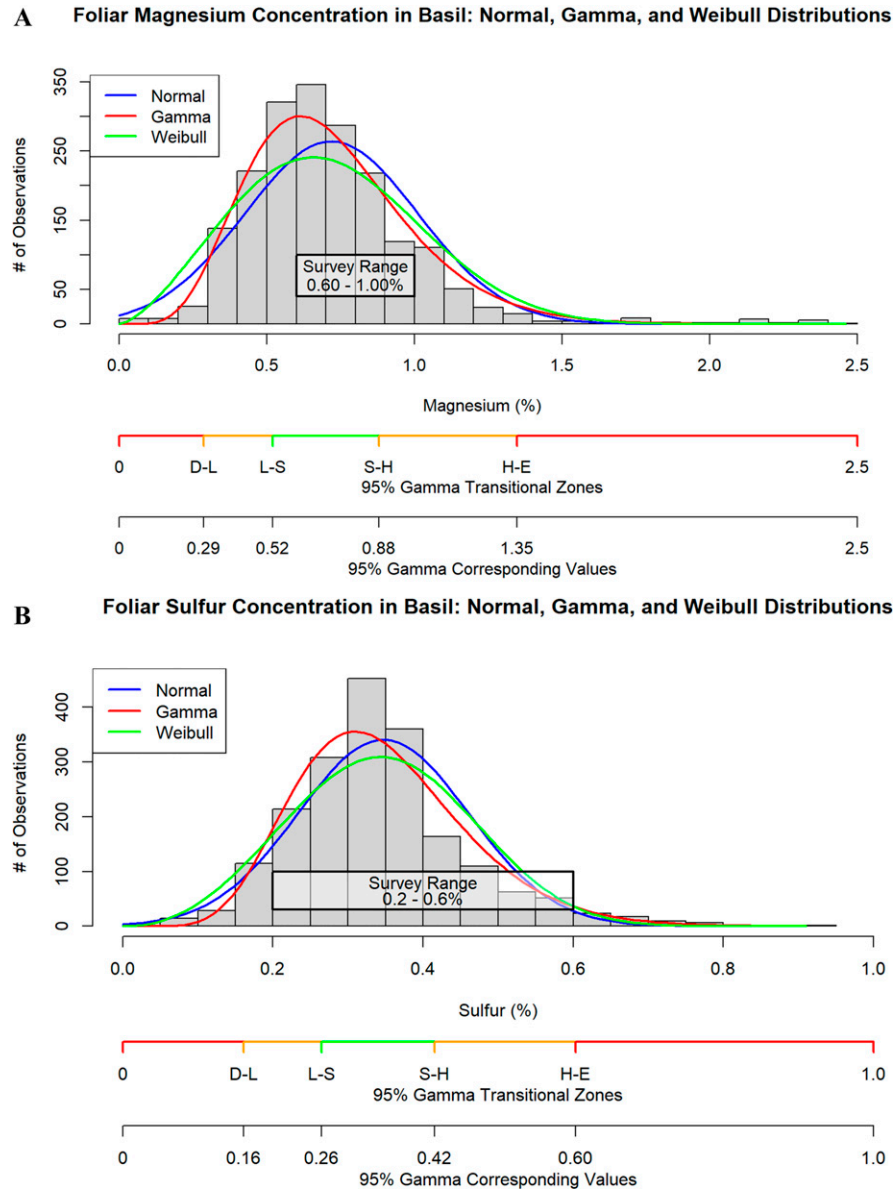

Fig. 7. Magnesium (**A**) and sulfur (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 2.5%, 25%, 75%, and 97.5% of sample observations (n = 1938), respectively. Survey ranges from Bryson and Mills (2015) are overlayed for reference.

These findings contradict basil's reportedly high Mg requirement and instead suggest basil is not as sensitive to Mg deficiency as previously suggested (Dickson 2019; Mattson 2018). While the updated sufficiency range for Mg foliar concentrations in basil are still higher than what is recommended for other leafy greens (Bryson and Mills 2015), the absence of deficiency symptoms at low Mg fertilizer concentrations does not suggest a sensitivity to Mg deficiency. The occurrence of Mg deficiencies in commercial production may be due to high Ca concentrations in the fertilizer solutions rather than a high Mg requirement in basil. Hydroponic fertilizer solutions are often formulated for the most economically important crops, such as tomato and lettuce, which are most susceptible to Ca deficiency disorders (Bangerth 1979). Excessive Ca can impede Mg uptake, and due to basil having a higher foliar Mg concentration requirement, Mg deficiencies may be more commonly induced in hydroponically grown basil crops. Currently, based on the Mg foliar concentrations, a minimum of 40 mg·L^{-1} Mg should be applied to maintain sufficient Mg concentrations. Plants that received the two highest Mg fertilizer concentrations, 80 and

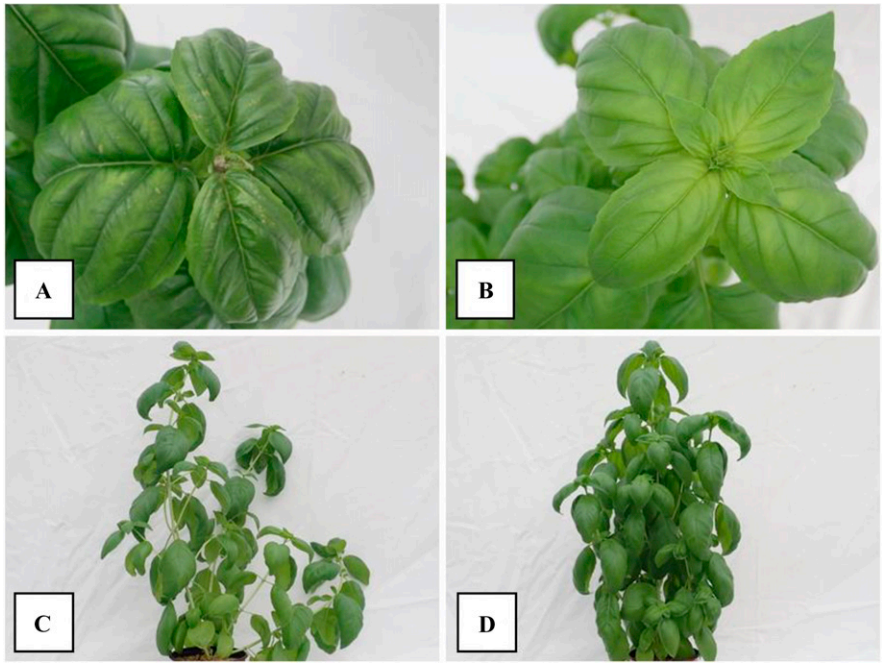

Fig. 8. Basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'] visual micronutrient deficiency symptoms. Boron-deficient plants (**A**) developed interveinal chlorosis of the newest leaves, and the thick, brittle upper foliage that exhibited a downward cupped appearance. Iron-deficient plants (**B**) exhibited subtle interveinal chlorosis of the upper foliage. Zinc-deficient plants (**C**) had spindly growth, with long internodes and weak stems. (**D**) Control plant which received sufficient concentrations of all nutrients and exhibited healthy growth.

120 mg·L^{-1}, had Mg foliar concentrations that fell in the high range but did not have greater yields. While 40 mg·L^{-1} Mg is still currently recommended, the results of this study prompt the need for further research investigating Mg deficiency in basil and to determine whether lower Mg applications may be sufficient for growing visually healthy basil plants while still maximizing yield.

Sulfur

Sulfur deficiency symptoms. Symptoms of S deficiency were only observed in plants that received 0 mg·L^{-1} S. Plants from this treatment exhibited stunted growth and interveinal chlorosis of the middle foliage (Figs. 1F and 2). Advanced deficiency was observed in plants that received 0 mg·L^{-1} S after 35 d of growth, with the foliage developing a pale-green coloration of the entire canopy. While plants from the 2 mg·L^{-1} S treatment did not exhibit visual deficiency symptoms, they had 24% less dry mass than plants supplied with 8, 25, or 50 mg·L^{-1} S (Fig. 6E). Although no clear toxicity symptoms were observed, plants supplied with the highest S fertilizer concentration, 76 mg·L^{-1}, also exhibited lower dry mass than the control plants (Fig. 6E).

Sulfur leaf tissue biomass and accumulation. The relationship between S fertilizer concentration and plant dry mass was best represented by a quadratic model. The highest dry mass was observed in plants supplied with 8 mg·L^{-1} S, followed by an incrementally lower dry mass in plants that received 25, 50, or 76 mg·L^{-1} S. The relationship between S fertilizer concentration and S foliar concentration was best modeled by a quadratic plateau, in which S foliar concentration plateaued at a S fertilizer concentration of 2.8 mg·L^{-1} S (Fig. 6F). This low plateau, in conjunction with a lack of deficiency symptoms, suggests that a lower S fertilizer concentration may be sufficient for hydroponically grown basil crops.

Sulfur nutrient distribution curve. A Gamma distribution best represented the S foliar concentrations (Table 3; Fig. 7B). The model defines the sufficiency range as 0.26% to 0.42% S (Table 4), which narrows the survey range of 0.2% to 0.6% S presented by Bryson and Mills (2015). Control plants, which received 25 mg·L^{-1} S, had a foliar concentration of 0.35% S and fell within the suggested sufficiency range. The deficiency range was defined as <0.16% S, which is corroborated by the foliar concentration of plants that received 0 mg·L^{-1} S and exhibited deficiency symptoms (Fig. 6F). The excessive range was defined as >0.60% S. Plants that received the highest S fertilizer concentrations (50 and 76 mg·L^{-1} S) did not have high or excessive S foliar concentrations.

While basil plants supplied with 0 mg·L^{-1} S had a S foliar concentration that fell within the deficiency range, all remaining treatments had a S foliar concentration within the established sufficiency range. Plants that received 2 mg·L^{-1} S had a S foliar concentrations within the sufficiency range, and while they visually appeared healthy, they yielded lower dry mass than plants supplied with higher S fertilizer concentrations. This suggests that basil can be supplied with a S fertilizer concentration as low as 8 mg·L^{-1} S without negatively affecting plant health or dry mass. A S concentration as low as 2 mg·L^{-1} S may be supplied to produce visually healthy plants while managing plant growth. While plants that received 25 mg·L^{-1} S had 16% less dry mass than plants supplied with 8 mg·L^{-1} S, it is unlikely due to S toxicity. Given a longer growing period, plants that received a higher S fertilizer concentration may have had a higher dry mass. Due to the lower dry masses observed in plants supplied with 50 or 76 mg·L^{-1} S, fertilizer concentrations exceeding 25 mg·L^{-1} S are not recommended.

Boron

Boron deficiency symptoms. Boron-deficient plants exhibited interveinal chlorosis of the newest leaves, and the upper foliage became thick and brittle. The newest growth also exhibited a downward, cupped appearance (Fig. 8A). As symptoms advanced, the interveinal chlorosis

Table 5. Greenhouse-grown basil [*Ocimum basilicum* L. 'Prospera Compact DMR (PL4)'] plant dry mass and tissue nutrient concentration as affected by deficient micronutrient treatments.

Treatment	−B	−Cu	−Fe	−Mn	−Mo	−Zn
	Dry mass (g)					
Complete control	15.59	15.59	15.59	15.59	15.59	15.59
Disorder	8.67	6.83	12.52	10.28	13.40	12.04
P value[i]	**	***	NS	*	NS	NS
	Tissue nutrient concentration (mg·kg^{-1})					
Complete control	54.50	8.49	89.97	80.41	0.74	27.19
Disorder	3.98	0.82	87.76	29.73	0.05	12.05
P value[i]	***	***	NS	**	**	***
Survey range[ii]	25–60	5–10	75–200	30–150	25–60	30–70

[i] *, **, and *** denote statistically significant differences between the sample means based on *F* test at $P \leq 0.05$, ≤ 0.01, and ≤ 0.001, respectively. NS indicates that the *F* test differences between sample means was not significant ($P > 0.05$).
[ii] Bryson and Mills (2015).

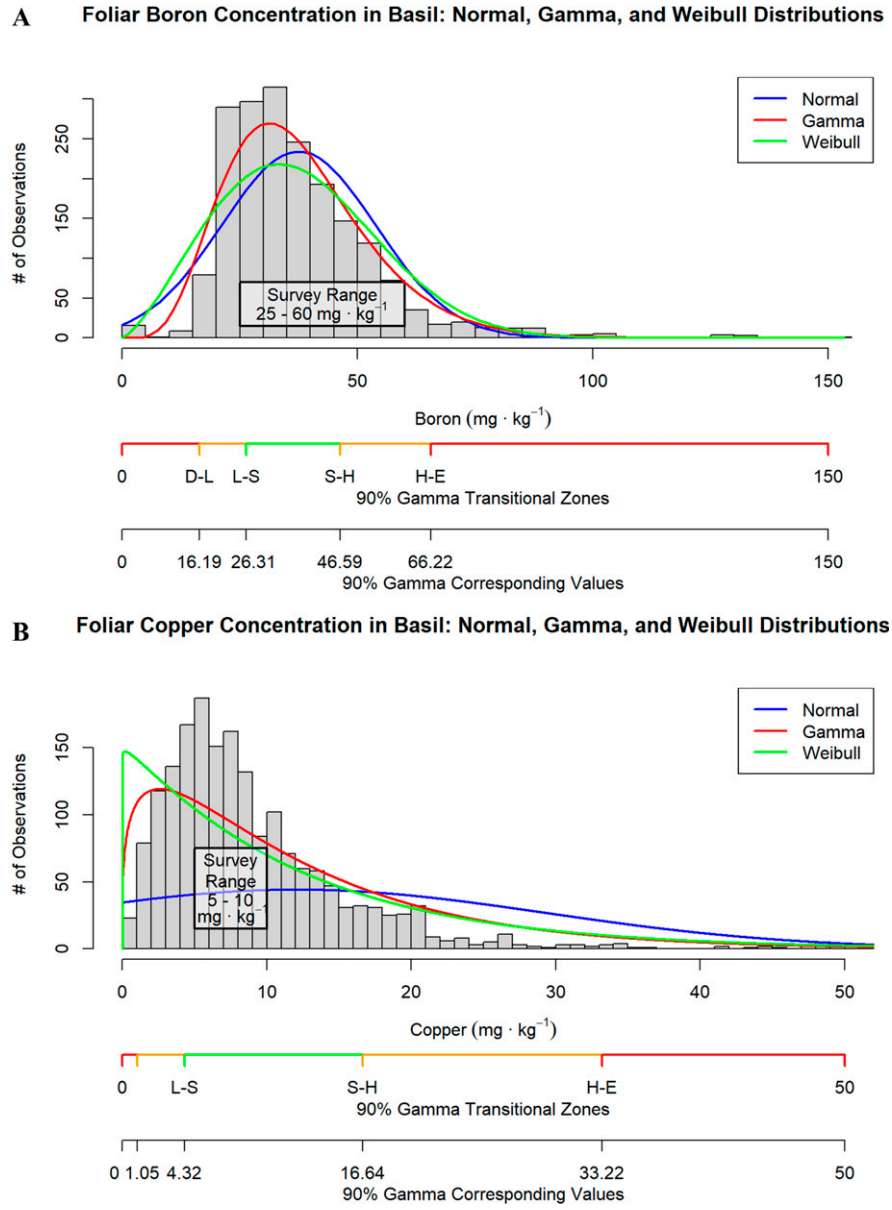

Fig. 9. Boron (**A**) and copper (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 5%, 25%, 75%, and 95% of sample observations (n = 1938), respectively. Survey ranges from Bryson and Mills (2015) are overlayed for reference.

became more pronounced, and necrotic patches developed on the margins of the most recently matured leaves. Boron-deficient plants had significantly less dry mass than the control plants, with means of 8.7 and 15.6 g, respectively. The B foliar concentration of the B-deficient plants was also lower than the control plants, 4.0 mg·kg^{-1} B compared with 54.5 mg·kg^{-1} B, respectively (Table 5).

Boron nutrient distribution curve. A Gamma distribution best represented the B leaf tissue data (Table 3; Fig. 9A). The recommended sufficiency range of 26.3 to 46.6 mg·kg^{-1} B (Table 4) narrows the previously recommended range of 25 to 60 mg·kg^{-1} B (Bryson and Mills 2015). The recommended deficiency range, <16.2 mg·kg^{-1} B, is supported by the B foliar concentration of the symptomatic B-deficient plants. Boron foliar concentrations exceeding 66.2 mg·kg^{-1} were determined to be excessive, after which toxicity symptoms may occur.

Copper

Copper deficiency symptoms. After 57 d, Cu-deficient plants did not exhibit deficiency symptoms other than stunted growth. The Cu-deficient plants had 56.2% less dry mass than the control plants (Table 5). Likewise, Cu-deficient plants had a lower foliar concentration (0.8 mg·kg^{-1} Cu) than the control plants, which had a Cu concentration of 8.5 mg·kg^{-1}.

Copper nutrient distribution curve. Of the three examined models, a Gamma distribution best represented the Cu foliar concentrations (Table 3; Fig. 9B). The recommended sufficiency range, 4.3 to 16.6 mg·kg^{-1} Cu (Table 4) expands the previous range of 5 to 10 mg·kg^{-1} Cu, provided by Bryson and Mills (2015). The defined deficiency range was <1.1 mg·kg^{-1} Cu. These ranges are both supported by the Cu foliar concentrations of the control and Cu-deficient plants. Excessive Cu foliar concentrations were defined as >33.2 mg·kg^{-1} Cu.

Iron

Iron deficiency symptoms. After 57 d, Fe-deficient plants developed subtle interveinal chlorosis of the upper foliage (Fig. 8B). The dry mass of Fe-deficient plants was 20% lower than the control plants, but the difference was not statistically significant (P > 0.05) (Table 5). Similarly, despite signs of Fe deficiency, there was no significant difference between the Fe foliar concentration of the control and Fe-deficient treatments.

Iron nutrient distribution curve. A Gamma distribution had the lowest BIC value and best represented the Fe foliar concentrations (Table 3; Fig. 10A). The sufficiency range was defined as 83.1 to 147.6 mg·kg^{-1} Fe (Table 4), substantially narrowing the range of 75 to 200 mg·kg^{-1} Fe previously recommended by Bryson and Mills (2015). This adjustment suggests lower Fe foliar concentrations are adequate for greenhouse-grown basil. The control plants, which had an Fe foliar concentration of 90.0 mg·kg^{-1} Fe, corroborate this sufficiency range. The model also defined the deficiency range as <51.0 mg·kg^{-1} Fe. While our Fe-deficient plants did not have low Fe foliar concentrations to corroborate this deficiency value, it can still provide a baseline for diagnostic purposes. Lastly, the excessive range was defined as >210.0 mg·kg^{-1} Fe, above which Fe toxicity may occur.

Manganese

Manganese deficiency symptoms. Manganese-deficient plants did not exhibit deficiency symptoms within 57 d. Despite a lack of visual deficiency symptoms, the Mn-deficient plants had 34% less dry mass than the control plants. Additionally, the Mn foliar concentration of the control and Mn-deficient plants differed and were 80.4 and 29.7 mg·kg^{-1} Mn, respectively (Table 5).

Manganese nutrient distribution curve. A Gamma distribution best represented the Mn foliar concentrations (Table 3; Fig. 10B). The sufficiency range of 58.4 to 132.6 mg·kg^{-1} Mn (Table 4) narrows the 30 to 150 mg·kg^{-1} Mn range presented by Bryson and Mills (2015). The deficiency range was defined as <27.6 mg·kg^{-1} Mn, which is slightly lower than the foliar concentration of the Mn-deficient plants. The excessive range was defined as >213.0 mg·kg^{-1} Mn.

Molybdenum

Molybdenum deficiency symptoms. The Mo-deficient plants did not develop any visual deficiency symptoms within 57 d. Further, there was no significant difference (P > 0.05) between the dry masses of the control plants and plants from the Mo-deficient treatment.

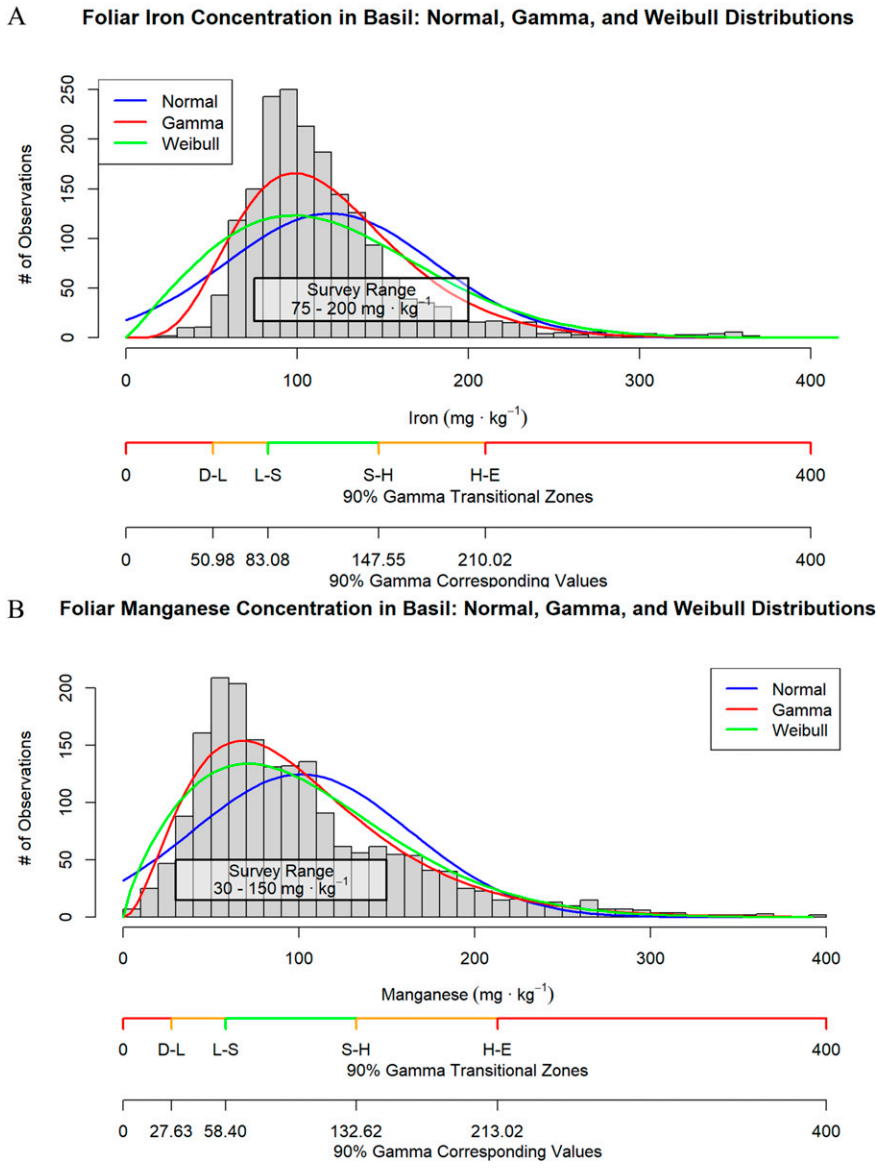

Fig. 10. Iron (**A**) and manganese (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 5%, 25%, 75%, and 95% of sample observations (n = 1938), respectively. Survey ranges from Bryson and Mills (2015) are overlayed for reference.

Despite this, the Mo-deficient plants had a foliar concentration of 0.05 mg·kg^{-1} Mo, while the control plants had a foliar concentration of 0.7 mg·kg^{-1} Mo. (Table 5).

Molybdenum nutrient distribution curve. Some samples used to define the foliar interpretation ranges did not include Mo leaf tissue concentrations, so those samples were excluded, and the interpretation ranges for foliar Mo were determined using a smaller sample size (n = 1265). Additionally, Mo foliar concentrations below the detection limit were assigned a foliar concentration of 0.01 mg·kg^{-1} Mo for analysis purposes. Of the three examined models, a Weibull model had the lowest BIC and best represented the Mo foliar concentrations (Table 3; Fig. 11A). The resulting sufficiency range was 0.1 to 0.8 mg·kg^{-1} Mo (Table 4), which expands the previously recommended range of 0.1 to 0.5 mg·kg^{-1} Mo recommended by Bryson and Mills (2015). Due to the highly skewed distribution of Mo foliar concentrations, a left tail was not established, and a deficiency range could not be confidently identified. The low and deficient ranges were combined, and the value for the deficient and low range was defined as <0.1 mg·kg^{-1} Mo. While the Mo-deficient plants did have a Mo foliar concentration within this range, no deficiency symptoms were present. This suggests that basil is not significantly affected by low Mo concentrations. The distribution also defined the excessive range as >2.0 mg·kg^{-1} Mo. While this range provides a useful reference, Mo toxicity is relatively rare and has not been observed in basil. Other crops, such as French marigold (*Tagetes patula* L.), do not exhibit symptoms of Mo toxicity until foliar concentrations as high as 5010 mg·kg^{-1} (Choi et al. 1996). In terms of Mo toxicity in basil, further research is needed to determine whether the upper limit is higher than currently reported. While the Weibull distribution was deemed the best fit, the high number of samples with low Mo foliar concentrations suggests that further refinement would be advantageous.

Zinc

Zinc deficiency symptoms. After 57 d of growth, plants from the Zn deficiency treatment exhibited spindly growth, with long internodes and weak stems (Fig. 8C). However, there was no significant difference (P > 0.05) between the dry masses of the control and Zn-deficient plants. The Zn foliar concentration of the Zn-deficient plants was 12.1 mg·kg^{-1}, in contrast to the 27.2 mg·kg^{-1} Zn of the control plants (Table 5).

Zinc nutrient distribution curve. A Gamma distribution had the lowest BIC value and best represented the Zn foliar concentrations (Table 3; Fig. 11B). The defined sufficiency range of 32.8 to 77.6 mg·kg^{-1} Zn (Table 4) slightly shifts upward the range presented Bryson and Mills (2015), which suggested 30 to 70 mg·kg^{-1} Zn. The Zn foliar concentration of the control plants fell slightly below this sufficiency range, although no deficiency symptoms were present. The deficiency range was defined as <14.8 mg·kg^{-1} Zn, which includes the foliar concentration of our Zn-deficient plants. The model defined the excessive range as >127.0 mg·kg^{-1} Zn.

Conclusions

Visual symptoms of all macronutrient deficiencies (N, P, K, Ca, Mg, and S) manifested within 28 d of growth. Symptoms of micronutrient deficiencies were only seen for the B-, Fe-, and Zn-deficient treatments within 57 d of growth. All observed nutrient deficiency symptoms were consistent with previously reported deficiency symptoms in basil (Gibson et al. 2007; Mattson and Merrill 2016), although Fe deficiency symptoms were not as prominent as those reported by Mattson and Merrill (2016). Lastly, despite a lack of expected deficiency symptoms, plants from the Cu- and Mn-deficient treatments had significantly less dry mass. For plants from the Mo-deficient treatment, despite having Mo foliar concentrations that fell within the deficient range, biomass was not negatively affected, and deficiency symptoms were not observed. These results suggest that basil is not sensitive to low Mo concentrations.

While optimal macronutrient fertilizer concentrations varied by element, low N, P, and K had the greatest impact on plant dry mass. For some elements, such as P, the relationship between plant dry mass and fertilizer concentration was best modeled by a quadratic equation in which plant dry mass increased with fertilizer concentration until a maximum point, above which dry mass decreased. The relationship between N fertilizer concentration and dry mass was best modeled by a quadratic plateau in which dry mass

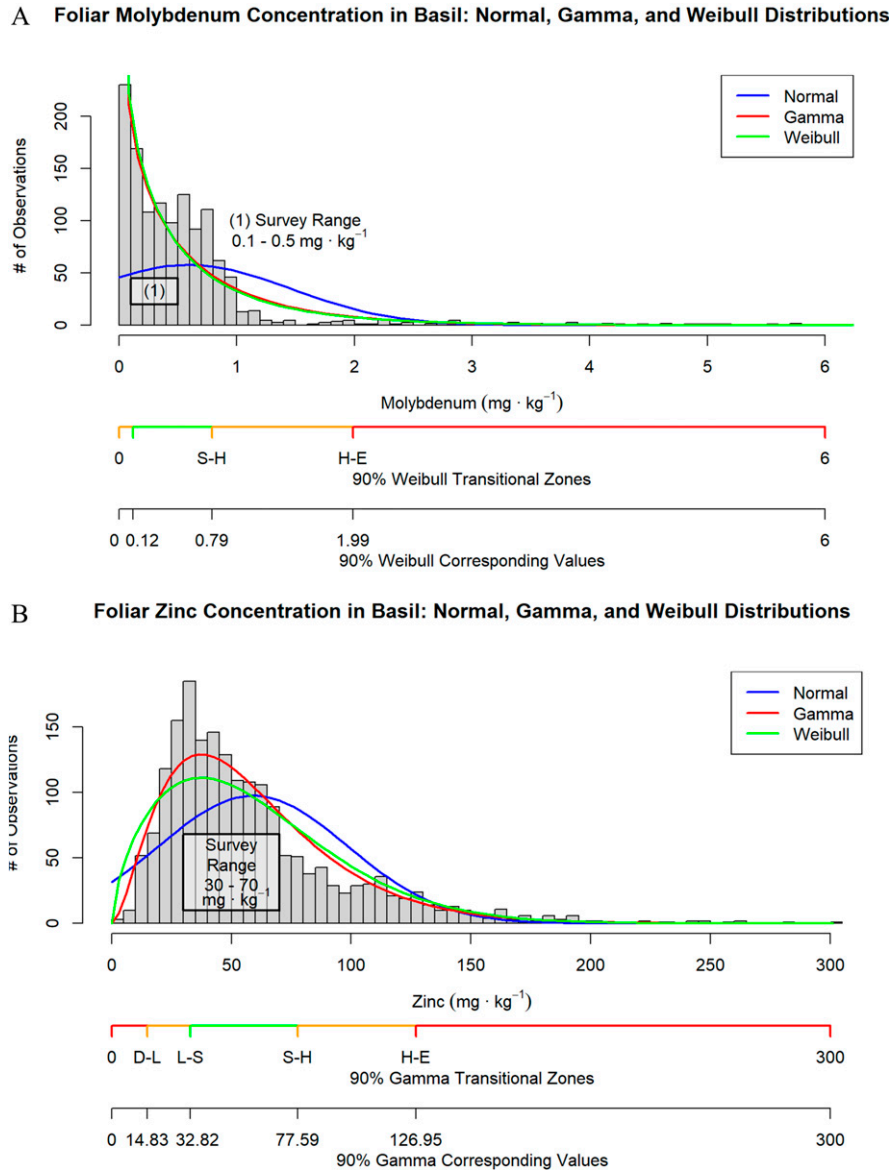

Fig. 11. Molybdenum (**A**) and zinc (**B**) foliar concentrations of greenhouse-grown basil (*Ocimum basilicum* L.) modeled using Normal, Gamma, and Weibull distributions. Interpretation ranges are based on the denoted distribution with four transitional points of deficient to low (D–L), low to sufficient (L–S), sufficient to high (S–H), and high to excessive (H–E), which correspond to 5%, 25%, 75%, and 95% of sample observations (n = 1938), respectively. For Mo, the deficient and low ranges were combined, creating one low range corresponding to the lowest 30% of sample observations (n = 1265). Survey ranges from Bryson and Mills (2015) are overlayed for reference.

increased with increasing N fertilizer concentration before reaching a plateau. While Ca and Mg concentrations did not have a significant effect on plant dry mass, deficiency symptoms were still evident at low fertilizer concentrations.

In addition to evaluating macronutrient fertilizer concentrations, refined foliar nutrient standards were also established for each essential element. By expanding upon the sufficiency range approach method, deficient, low, and excessive ranges for each element were established. These ranges will provide greenhouse-specific reference points for both growers and researchers, enabling more accurate diagnoses of nutrient disorders in basil.

The collected dry mass and foliar nutrient concentration data, in conjunction with the established sufficiency ranges study, suggest that optimal basil growth can be achieved using a nutrient solution containing (in mg·L^{-1}) 150 N, 20 P, 96 K, 75 Ca, 40 Mg, and 8 S. While a 150 mg·L^{-1} is recommended to maintain N foliar concentrations within the sufficiency range, a fertilizer concentration as low as 53.2 mg·L^{-1} may be applied without negatively affecting yield. These findings provide more comprehensive recommendations than what is currently available, providing optimal fertilizer concentrations for each macronutrient. These recommendations will aid producers in preventing fertilizer overapplication to save costs and resources without sacrificing yield.

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