

Effects of Gibberellin and Melatonin Foliar Sprays on Basil Chemical Composition and Volatile Oil Yield under Water Stress

Hala Faisal Khazal*, and Majid Ali Hanshal

Department of Horticulture and Landscape Gardening, College of Agricultural Engineering Sciences, University of Baghdad, Baghdad, Iraq

*Corresponding author; email: hala.khazal2105p@coagri.uobaghdad.edu.iq

Abstract

This study investigated the potential of exogenous gibberellic acid (GA₃) and melatonin (MT) to mitigate the adverse effects of drought stress on basil (*Ocimum basilicum* L.). Conducted in 2022 at the University of Baghdad, Iraq, the experiment employed a split-plot design within a completely randomized block design. Three irrigation intervals (3, 5, and 7 days) were assigned to the main plots, while subplots received a factorial combination of the plant growth regulator gibberellic acid (GA₃; 0, 100, and 200 mg/L) and melatonin (0, 20, and 40 mg/L). Results indicated that moderate water stress with a 5-day irrigation interval significantly enhanced leaf nitrogen, phosphorus, and potassium nutrient content, total phenolic content, and volatile oil yield compared with the 3-day and 7-day irrigation intervals. The application of 200 mg/L GA₃ and 40 mg/L melatonin consistently yielded the highest values for all physiological and chemical parameters. Significant double- and triple-interactions suggest that the synergistic application of GA₃ and melatonin effectively improved basil secondary metabolism under water-limited conditions. These findings highlight a strategic approach for optimizing essential oil production in arid environments.

Keywords: basil, growth regulator, irrigation interval, water stress

Introduction

The use of medicinal plants has become an urgent need with the increase in population size and the increase in awareness and knowledge in treating diseases that may affect humans, as well as their use and circulation is simple and can be used as concentrates or extracts or in the form of capsules and dry pills due to the presence of the active substance in them with a physiological effect, as attention turned in the late last century in developed countries towards alternative medicine instead of taking medical drugs that humans take (Al-Shahat, 2006). Medicinal plants have been used since ancient times to treat and cure diseases, and many countries still rely on them. The World Health Organisation also indicated that a large portion of the world's population relies on medicinal herbs as a primary form of treatment for many diseases (Mamedov, 2012).

Basil belongs to the mint family, which includes a variety of aromatic plants found worldwide. Its scientific name is *Ocimum basilicum* L., and it is believed to be native to Asia and Africa (Arya & Thakur, 2012; Zolfaghari et al., 2013). Basil is widely cultivated around the world due to its nutritional and medicinal importance, as it was used in ancient China and India to treat kidney and stomach problems (Leung & Foster, 1996; Manzoor et al., 2023), its oil has antioxidant, antibacterial, antifungal, and antimicrobial properties, and has medicinal and economic value for the production of cosmetics, perfumes, and pharmaceuticals. It is also used as a food preservative and flavour enhancer

(Al-Shahat, 2006; Ru et al., 2024). Basil oil is characterised as a white-to-pale-yellow liquid with a pleasant odour (Al-Dajwi, 2004; Mounira et al., 2022). Agricultural production worldwide is affected by various stresses, both biotic and abiotic. Medicinal plants are among those directly affected by stress, including water stress, as this stress affects the development, formation, and growth stages of plant organs, and the occurrence and duration of these stages vary with accompanying environmental conditions (Jaleel et al., 2009). Water shortages or water stress are one of the biggest environmental challenges in basil-growing regions, especially in arid and semi-arid climates. Water shortages or water stress can lead to reduced relative water content of leaves and impaired physiological functions, such as poor growth and decreased total essential oil content. Water shortages reduce leaf area and crop dry weight, and plants sometimes respond by increasing glandular hair density or improving water-use efficiency (WUE), which helps maintain reasonable yields under stress. In these environments, careful irrigation management, the selection of drought-resistant varieties, and strategies such as microbial inoculation or the use of growth regulators are critical to enhance plant resilience. Integrating water stress management with crop quality is essential to ensuring sustainable basil production in a changing climate (Shahrajabian et al., 2021).

The physiological processes that occur in plants and are associated with water stress require further studies, especially the process of nutritional balance that plants perform to maintain their water content in harmony with climatic conditions and call for to seriously consider not wasting water and finding technologies that increase the efficiency of water use, such as spraying some antioxidant compounds such as melatonin, which is of great importance for growth and multiple physiological processes, including enhancing photosynthesis, regulating nitrogen metabolism, maintaining nutritional balance within plant tissues, protecting plant pigments, delaying leaf aging, and balancing oxidation and reduction processes (Ren et al., 2022). Melatonin also has essential functions in plant growth and

development. Many studies have demonstrated the regulatory effects of melatonin on plant systems under various abiotic stress conditions, as melatonin has multiple physiological effects, including stimulating cells in the thin layer of plant tissue between the epidermis and the bark and increasing the activity and growth of root hairs, resulting in the emergence of new lateral roots and adventitious roots (Al-Janabi et al., 2024; Arnao & Hernández-Ruiz, 2018).

Plant growth regulators, specifically gibberellic acids (GA_3), are fundamental endogenous regulators of cell division and elongation (Davies, 2012; Taiz & Zeiger, 2010). Beyond their primary role in promoting growth, GAs significantly enhance plant adaptability to water deficits by facilitating stem and root elongation, thereby optimizing subsoil moisture acquisition. Furthermore, GAs modulate the antagonistic crosstalk between growth hormones (auxins) and stress hormones (abscisic acid), thereby mitigating the physiological constraints of drought. Previous research suggests that exogenous GA application improves cell wall elasticity, enhances nutrient uptake efficiency, and maintains photosynthetic pigments under moisture-limited conditions (Maghsoudi et al., 2018). Recently, melatonin has emerged as a synergistic signaling molecule that further bolsters antioxidant defenses. There are still limited studies on how plant growth regulators can improve oil yield of aromatic species under water stress. This study, therefore, evaluates the combined effects of foliar-applied gibberellins and melatonin on the volatile oil profile and chemical composition of basil (*Ocimum basilicum* L.) under stage-specific water stress.

Materials and Methods

Experimental Site and Design

The study was conducted during the spring season of 2022 at the experimental field of the Faculty of Agricultural Engineering Sciences, University of Baghdad. The research evaluated the physiological and chemical responses of basil (*Ocimum basilicum* L.) to exogenous gibberellic

acid (GA_3) and melatonin applications under varying water-stress regimes. The experiment was organized in a split-split plot design within a completely randomized block design with three replications. The irrigation intervals of 3, 5, and 7 days, reflecting water stress levels, was assigned as the main plot; sub-plots with GA_3 concentrations of 0, 100, and 200 mg/L; and sub-sub-plots with melatonin concentrations of 0, 20, and 40 mg/L.

Basil seeds were initially sown in seedling trays filled with a standardized growth medium (peat moss and perlite). After 2–4 true leaves emerged, the seedlings were thinned to ensure uniformity. At 21–28 days after sowing (DAS), the seedlings were transplanted into the experimental field. For the drought stress treatment, a standard irrigation was maintained during the initial establishment period to ensure successful root development. Water stress treatments were initiated 14–21 days post-transplantation, when the plants were approximately 35–45 days old. At this phenological stage, the plants had reached a stable vegetative state, allowing for a clear assessment of how and melatonin influence stress resilience and volatile oil synthesis.

Leaf nutrient analysis and secondary metabolite quantification were performed following standardized protocols. Nitrogen (N) content was determined using the semi-micro Kjeldahl method (AOAC, 1980). Phosphorus (P) was estimated colorimetrically via spectrophotometry, while potassium (K) was quantified according to AOAC (1980) guidelines. The total phenolic content (TPC) was evaluated using the spectrophotometric method described by Mahadevan and Sridhar (1986). Volatile oil content was extracted from fresh leaf material using a Clevenger-type apparatus for hydrodistillation (AOAC, 1980), and the volatile oil yield was subsequently calculated as the product of the fresh leaf weight and the oil percentage (g per plant).

Results and Discussion

Result

Effects of Irrigation Intervals and Growth Regulators on Leaf N, P, K

The mineral composition of basil leaves, specifically nitrogen (N), phosphorus (P), and potassium (K), was significantly influenced by irrigation frequency and the application of exogenous growth regulators (Tables 1, 2, 3, 4, 5, and 6). The three-way interaction among 5-day watering interval, GA_3 at 200 mg/L, and melatonin at 40 mg/L had the greatest positive impact on nutrient uptake. The maximum values recorded were 2.920% for N (Table 1), 0.811% for P (Table 3), and 1.793% for K (Table 5). Conversely, the lowest nutrient accumulation was consistently observed in the 7-day watering interval without growth regulators, resulting in 0.974% N (Table 1), 0.270% P (Table 3), and 0.598% K (Table 5), illustrating the severe limitations on mineral absorption imposed by prolonged water deficits without hormonal mitigation.

The moderate irrigation interval of 5 days consistently yielded the highest nutrient concentrations: N (2.153%, Table 1), P (0.598%, Table 3), and K (1.322%, Table 5). In contrast, extending the irrigation interval to 7 days resulted in the lowest recorded values across all mineral traits (Tables 1, 3, 5), respectively.

Regarding hormonal treatments, the high-concentration applications of GA_3 (200 mg/L) and melatonin (40 mg/L) significantly enhanced nitrogen accumulation compared to their respective controls (Table 2). Melatonin at 40 mg/L was particularly effective for nitrogen, achieving a peak of 2.016% (Table 2), while GA_3 at 200 mg/L recorded phosphorus and potassium peaks of 0.548% (Table 4) and 1.210% (Table 6). Significant two-way interactions were observed between all studied factors (Tables 2, 4, and 6). The combinations of 5-day watering intervals and GA_3 at 200 mg/L or melatonin at 40 mg/L produced the highest leaf N (Table 2), P (Table 4), and K (Table 6) concentrations. For instance, basil treated with 5-day watering intervals and

melatonin at 40 mg/L had N and K concentrations of 2.399% (Table 2) and 1.473% (Table 6), respectively.

Total Phenolic Content (%)

Statistical analysis (Tables 7 and 8) revealed that the total phenolic content was significantly enhanced by moderate irrigation (5-day irrigation interval; 0.466%) and by the highest hormonal concentrations (200 mg/L GA₃: 0.415%; 40 mg/L melatonin; 0.428%). The synergistic effect of the three factors was most evident in the 5-day irrigation interval, 200 mg/L GA₃, and 40 mg/L melatonin combination, which achieved 0.679%, a significant increase over the 7-day irrigation interval without growth regulators (0.139%).

Leaf Oil Content

The volatile oil percentage exhibited a response pattern consistent with the leaf nutrient accumulation trends (Tables 9, 10). Statistical analysis revealed that irrigation intervals, GA₃ application, and melatonin treatments all exerted significant primary and interactive effects on these parameters (Tables 9, 10).

The moderate irrigation frequency of 5 days achieved a maximum oil yield of 7.421 g per plant, significantly outperforming the severe drought condition of 7 days, which reduced yield to 5.062 g per plant (Table 11). Among the growth regulators, the highest concentrations of GA₃ (200 mg/L) and melatonin (40 mg/L) resulted in peak individual yields of 6.509 g per plant and 6.744 g per plant, respectively (Table 12). These results confirm that exogenous GA₃ and melatonin act as potent biostimulants for secondary metabolite synthesis in basil.

Table 1

The Effects of GA₃ and Melatonin Spraying under Different Watering Intervals on the Leaf Nitrogen (%)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	1.891	1.702	1.810	1.801	1.798
	100	1.891	1.696	1.795	1.794	
	200	1.911	1.725	1.761	1.799	
5 days	0	1.998	1.900	1.945	1.948	2.153*
	100	2.043	2.178	2.333	2.185	
	200	2.025	2.039	2.920	2.328	
7 days	0	0.974	1.637	1.698	1.436	1.679
	100	1.566	1.850	2.030	1.815	
	200	1.857	1.651	1.850	1.786	
Melatonin mean		1.795	1.820	2.016		
LSD irrigation interval			0.083			
LSD GA ₃			0.083			
LSD melatonin			0.083			
LSD irrigation interval x GA ₃ x melatonin			0.250			

Table 2

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Leaf Nitrogen Content

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	1.621	1.747	1.818	1.728
100	1.833	1.908	2.052	1.931
200	1.931	1.805	2.177	1.971
Melatonin mean	1.795	1.820	2.016	
LSD GA ₃ x melatonin	0.144			
Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	1.898	1.708	1.789	1.798
5	2.022	2.039	2.399*	2.153
7	1.465	1.713	1.859	1.679
Melatonin mean	1.795	1.820	2.016	
LSD irrigation interval x melatonin	0.144			
Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	1.801	1.794	1.799	1.798
5	1.948	2.185	2.328*	2.153
7	1.436	1.815	1.786	1.679
GA ₃ mean	1.728	1.931	1.971	
LSD irrigation interval x GA ₃	0.144			

Table 3

The Effects of GA₃ and Melatonin Spraying under Different Watering Intervals on the Leaf Phosphorus (%)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	0.522	0.473	0.503	0.499	1.798
	100	0.525	0.471	0.499	0.498	
	200	0.531	0.479	0.489	0.500	
5 days	0	0.555	0.528	0.540	0.541	2.153*
	100	0.568	0.605	0.648	0.607	
	200	0.562	0.566	0.811	0.647	
7 days	0	0.270	0.455	0.472	0.399	1.679
	100	0.435	0.514	0.564	0.504	
	200	0.516	0.459	0.514	0.496	
Melatonin mean		0.498	0.505	0.560		
LSD irrigation interval		0.023				
LSD GA ₃		0.023				
LSD melatonin		0.023				
LSD irrigation interval x GA ₃ x melatonin		0.069				

Table 4

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Leaf Phosphorus

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	0.449	0.485	0.505	0.480
100	0.509	0.530	0.570	0.536
200	0.536	0.501	0.605	0.548
Melatonin mean	0.498	0.505	0.560	
LSD GA ₃ x melatonin	0.040			

Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	0.526	0.474	0.497	0.499
5	0.562	0.566	0.666	0.598
7	0.407	0.476	0.517	0.466
Melatonin mean	0.498	0.505	0.560	
LSD irrigation interval x melatonin	0.040			

Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	0.499	0.498	0.500	0.499
5	0.541	0.607	0.647	0.598
7	0.399	0.504	0.496	0.466
GA ₃ mean	0.480	0.536	0.548	
LSD irrigation interval x GA ₃	0.040			

Table 5

The Effects of GA₃ and Melatonin Spraying under Different Watering Intervals on the Leaf Potassium (%)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	1.154	1.052	1.112	1.106	1.104
	100	1.161	1.041	1.102	1.101	
	200	1.173	1.058	1.081	1.104	
5 days	0	1.226	1.166	1.193	1.195	1.322*
	100	1.254	1.338	1.432	1.341	
	200	1.243	1.252	1.793*	1.429	
7 days	0	0.598	1.005	1.042	0.882	1.031
	100	0.961	1.135	1.246	1.114	
	200	1.140	1.013	1.136	1.096	
Melatonin mean		1.101	1.118	1.238		
LSD irrigation interval			0.051			
LSD GA ₃			0.051			
LSD melatonin			0.051			
LSD irrigation interval x GA ₃ x melatonin			0.153			

Table 6

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Leaf Potassium

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	0.993	1.074	1.116	1.061
100	1.125	1.171	1.260	1.322
200	1.185	1.107	1.337	1.031
Melatonin mean	1.101	1.118	1.238	
LSD GA ₃ x melatonin	0.088			

Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	1.162	1.050	1.098	1.104
5	1.241	1.252*	1.473	1.322
7	0.900	1.051	1.141	1.031
Melatonin mean	1.101	1.118	1.238	
LSD irrigation interval x melatonin	0.088			

Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	1.106	1.101	1.104	1.104
5	1.195	1.341*	1.429	1.322
7	0.882	1.114	1.096	1.031
GA ₃ mean	1.061	1.186	1.210	
LSD irrigation interval x GA ₃	0.088			

Table 7

The Effects of GA₃ and Melatonin Spraying under Different Watering Intervals on the Leaf Phenol (%)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	0.390	0.341	0.371	0.367	0.367
	100	0.393	0.339	0.367	0.366	
	200	0.399	0.347	0.357	0.368	
5 days	0	0.423	0.396	0.408	0.409	0.466*
	100	0.435	0.473	0.516	0.475	
	200	0.430	0.434	0.679*	0.515	
7 days	0	0.139	0.323	0.340	0.267	0.334
	100	0.303	0.382	0.432	0.372	
	200	0.384	0.326	0.382	0.364	
Melatonin mean		0.366	0.373	0.428		
LSD irrigation interval			0.24			
LSD GA ₃			0.24			
LSD melatonin			0.24			
LSD irrigation interval x GA ₃ x melatonin			0.74			

Table 8

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Leaf Phenols

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	0.993	1.074	1.116	1.061
100	1.125	1.171	1.260	1.322
200	1.185	1.107	1.337	1.031
Melatonin mean	1.101	1.118	1.238	
LSD GA ₃ x melatonin	0.42			

Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	0.394	0.342	0.365	0.367
5	0.429	0.434	0.534	0.466
7	0.275	0.344	0.384	0.415
Melatonin mean	0.366	0.373	0.384	
LSD irrigation interval x melatonin	0.42			

Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	0.367	0.366	0.368	0.367
5	0.409	0.475	0.515	0.466
7	0.267	0.372	0.364	0.415
GA ₃ mean	0.348	0.404	0.415	
LSD irrigation interval x GA ₃	0.42			

Table 9

The Effects of GA₃ and Melatonin Spraying under Water Stress Conditions on the Leaf Oil Content (%)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	0.128	0.116	0.123	0.122	0.122
	100	0.128	0.115	0.122	0.123	
	200	0.130	0.117	0.120	0.122	
5 days	0	0.136	0.129	0.132	0.132	0.146
	100	0.139	0.148	0.158	0.148	
	200	0.137	0.138	0.198	0.158	
7 days	0	0.066	0.111	0.115	0.098	0.114
	100	0.106	0.126	0.138	0.123	
	200	0.126	0.112	0.126	0.121	
Melatonin mean		0.122	0.124	0.137		
LSD irrigation interval			0.005			
LSD GA ₃			0.005			
LSD melatonin			0.005			
LSD irrigation interval x GA ₃ x melatonin			0.017			

Significant two-way interactions were observed, particularly in the combined application of irrigation regimes and hormonal sprays (Table 10). The 5-day irrigation interval supplemented with 20 mg/L melatonin (8.898 g per plant) and the 5-day irrigation interval combined with 200 mg/L GA₃ (8.487 g per plant) produced the highest yields among the two-factor treatments. In contrast, the lowest performance was recorded under the 7-day irrigation interval without hormone application (0 mg/L), yielding 4.027 g per plant. The three-way interaction demonstrated an even more pronounced effect (Table 11). The combination of a 5-day irrigation interval with 200 mg/L GA₃ and 40 mg/L melatonin yielded the greatest enhancement in volatile oil productivity, reaching 12.107 g per plant (Table 11). This value represents more than a five-fold increase compared to the minimum yield observed under the 7-day

irrigation interval without GA₃ or melatonin (2.232 g per plant). Overall, these results highlight the strong synergistic effect of GA₃ and melatonin in alleviating the adverse impact of water deficit on aromatic plant productivity.

The results across Tables 1-12 demonstrate that leaf nutrient status (N, P, K), total phenolics, and volatile oil metrics were optimized with a 5-day irrigation interval. This suggests that moderate irrigation frequency facilitates soil moisture availability, promoting nutrient solubility and uptake efficiency. In contrast, extended irrigation intervals (7-day intervals) are likely to trigger physiological shutdowns or increased root hydraulic resistance, severely hindering mineral absorption (Obidiegwu et al., 2015). The enhancement in chemical and oil profiles following gibberellic acid (GA₃) application is likely due to its role in stimulating vegetative biomass and increasing “sink strength.” By

Table 10

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Leaf Oil Content (%)

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	0.110	0.119	0.123	0.117
100	0.125	0.130	0.139	0.131
200	0.131	0.123	0.148	0.134
Melatonin mean	0.122	0.124	0.137	
LSD GA ₃ x melatonin		0.009		

Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	0.129	0.116	0.122	0.122
5	0.137	0.138	0.163	0.146
7	0.100	0.116	0.126	0.114
Melatonin mean	0.122	0.124	0.137	
LSD Irrigation interval x Melatonin		0.009		

Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	0.122	0.123	0.122	0.122
5	0.132	0.148	0.158	0.146
7	0.098	0.123	0.121	0.114
GA ₃ mean	0.117	0.131	0.134	
LSD Irrigation interval x GA ₃		0.009		

Table 11

The Effects of GA₃ and Melatonin Spraying under Water Stress Conditions on the on the Volatile Oil Yield (g per plant)

Irrigation interval	GA ₃ (mg/L)	Melatonin 0 mg/L	Melatonin 20 mg/L	Melatonin 40 mg/L	GA ₃ mean	Irrigation interval mean
3 days	0	5.853	5.078	5.593	5.508	5.524
	100	5.986	5.048	5.502	5.512	
	200	6.098	5.213	5.349	5.554	
5 days	0	6.514	6.030	6.268	6.271	7.421
	100	6.742	7.456	8.320	7.506	
	200	6.613	6.742	12.107*	8.487	
7 days	0	2.232	4.785	5.065	4.027	5.062
	100	4.505	5.800	6.709	5.671	
	200	5.818	4.856	5.786	5.486	
Melatonin mean		5.596	5.668	6.744		
LSD irrigation interval			0.419			
LSD GA ₃			0.419			
LSD melatonin			0.419			
LSD irrigation interval x GA ₃ x melatonin			1.259			

Table 12

Plant Growth Regulator and Irrigation Interval Interactions in Affecting Volatile Oil Yield (g per plant)

GA ₃ (mg/L)	Melatonin (mg/L)			GA ₃ mean
	0	20	40	
0	4.866	5.298	5.642	5.269
100	5.744	6.101	6.844	6.230
200	6.176	5.604	7.747	6.509
Melatonin mean	5.596	5.668	6.744*	
LSD GA ₃ x melatonin	0.727			
Irrigation intervals (days)	Melatonin (mg/L)			Irrigation interval mean
	0	20	40	
3	5.979	5.113	5.481	5.524
5	6.623	6.743	8.898	7.421
7	4.185	5.147	5.853	5.062
Melatonin mean	5.596	5.668	6.744	
LSD Irrigation interval x Melatonin	0.727			
Irrigation intervals (days)	GA ₃ (mg/L)			Irrigation interval mean
	0	100	200	
3	5.508	5.512	5.554	5.524
5	6.271	7.506	8.487	7.421
7	4.027	5.671	5.486	5.062
GA ₃ mean	5.269	6.230	6.509	
LSD Irrigation interval x GA ₃	0.727			

promoting cell elongation and metabolic activity, GA₃ facilitates the translocation of minerals from the rhizosphere to the foliar tissues (Maghsoudi et al., 2018).

The significant role of melatonin in increasing nutrient and oil content can be attributed to its function as a master physiological regulator. Melatonin enhances the plant's efficiency in performing vital metabolic functions and synergistically interacts with endogenous auxins to improve overall growth (Al-Saadi et al., 2022). Furthermore, melatonin is known to protect the photosynthetic apparatus from oxidative damage during water stress, ensuring that the carbon precursors required for volatile oil biosynthesis remain available even under moisture-limited conditions. The synergistic interaction of 5-day irrigation interval, 200 mg/L, and 40 mg/L GA₃ represents an integrated resilience strategy: 5-day irrigation interval provides the hydration necessary for cellular functions, GA₃ ensures the structural development of oil glands, and melatonin provides antioxidant protection and hormonal balance required to sustain secondary metabolism.

Conclusions

The results of this study demonstrate that the productivity and chemical composition of basil (*Ocimum basilicum* L.) are highly dependent on the interaction between irrigation management and exogenous hormonal application. A 5-day irrigation interval is optimal for maximizing leaf nutrient content (N, P, K), total phenolics, and volatile oil yield. While severe water stress (7-day irrigation interval) significantly hindered physiological performance, the application of 200 mg/L gibberellic acid and 40 mg/L melatonin served as effective mitigants. Specifically, the 5-day irrigation interval, 200 mg/L GA₃, and 40 mg/L melatonin interaction emerged as the superior treatment, yielding an oil yield of 12.107 g per plant. This study suggests that integrating GA₃ and melatonin into the cultivation regime can enhance the resilience of aromatic crops against water scarcity. For growers in arid regions like Iraq, adopting a moderate irrigation schedule (5-

day intervals) combined with foliar applications of GA₃ and melatonin offers a sustainable strategy to stabilize secondary metabolite production and optimize essential oil quality.

References

- Al-Dajwi, A. (2004). *Cultivation and production of ornamental plants and urban and flower coordination*. Madbouly Press.
- Al-Janabi, A. M. I., Al-Dulaimy, A. F., Sekhi, Y. S., Almohammed, O. H., & Al-Taey, D. K. (2024). Effect of salt stress on growth and yield of plants: A review. *IOP Conference Series: Earth and Environmental Science*, 1371(4), 042028.
- Al-Saadi, A. J. H., Al-Shammari, M. Z. F., Twaij, S. D., & Mohamed, B. M. R. (2022). Effect of interaction between gibberellic acid and urea fertilizer on some traits and yield components of fenugreek (*Trigonella foenum-graecum* L.). *Journal of Human and Social Sciences*, 30(4), 225–234.
- Al-Shahat, A. Z. N. (2006). *Volatile oils*. National Research Center, Dar for Publishing and Distribution.
- AOAC. (1980). *Official methods of analysis* (16th ed.). Association of Official Analytical Chemists.
- Arnao, M. B., & Hernández-Ruiz, J. (2018). The potential of phyto-melatonin as a nutraceutical. *Molecules*, 23(1), 238.
- Arya, V., & Thakur, N. (2012). Microscopic studies on *Ocimum* species. *Plant Sciences Feed*, 2(4), 56–58.
- Davies, P. J. (2012). *The plant hormones: Their nature, occurrence, and functions*. Martinus Nijhoff Publishers.
- Jaleel, C. A., Manivannan, P., Wahid, A., Farooq, M., Al-Juburi, H. J., Somasundaram, T., & Panneerselvam, R. (2009). Drought stress in plants: A review on morphological characteristics and pigment composition. *Journal of Agriculture and Biology*, 11, 100–105.
- Leung, A. Y., & Foster, S. (1996). *Encyclopedia of common natural ingredients used in food, drugs, and cosmetics* (2nd ed.). John Wiley and Sons.

- Maghsoudi, K., Arvin, M. J., & Ashraf, M. (2018). Mitigation of drought stress in wheat by foliar application of salicylic acid and gibberellic acid. *Journal of Plant Growth Regulation*, 37(2), 469–481. <https://doi.org/10.1007/s00344-017-9746-8>
- Mahadevan, A., & Sridhar, R. (1986). *Methods in physiological plant pathology* (3rd ed.). Sivakami Publications.
- Mamedov, N. (2012). Medical plants studies: History, challenges and prospective. *Medicinal & Aromatic Plants*, 1, 8.
- Manzoor, A., Asif, M., Khalid, S. H., Ullah Khan, I., & Asghar, S. (2023). Nanosizing of lavender, basil, and clove essential oils into microemulsions for enhanced antioxidant potential and antibacterial and antibiofilm activities. *ACS Omega*, 8(43), 40600–40612. <https://doi.org/10.1021/acsomega.3c05394>
- Mounira, G. M., Ahlem, Z., Mariem, B. A., Romdhane, M., Okla, M. K., Al-Hashimi, A., Alwase Y. A., Madnay M. M., AbdElgayed G., Asard H., Beemster G. T. S., & AbdElgawad H. (2022). Essential oil composition and antioxidant and antifungal activities of two varieties of *Ocimum basilicum* L. (Lamiaceae) at two phenological stages. *Agronomy*, 12(4), 825. <https://doi.org/10.3390/agronomy12040825>
- Obidiegwu, J., Bryan, G., Jones, G., & Prashar, A. (2015). Coping with drought: Stress and adaptive responses in potato and perspectives for improvement. *Frontiers in Plant Science*, 6, 542.
- Ren, J., Yang, X., Zhang, N., Feng, L., Ma, C., Wang, Y., Yang, Z., & Zhao, J. (2022). Melatonin alleviates aluminum-induced growth inhibition by modulating carbon and nitrogen metabolism, and reestablishing redox homeostasis in *Zea mays* L. *Journal of Hazardous Materials*, 423, 127159. <https://doi.org/10.1016/j.jhazmat.2021.127159>
- Ru, Y., Zhu, Y., Wang, X., Dong, Q., & Ma, Y. (2024). Edible antimicrobial yeast-based coating with basil essential oil for enhanced food safety. *Innovative Food Science & Emerging Technologies*, 93, 103612. <https://doi.org/10.1016/j.ifset.2024.103612>
- Shahrajabian, M. H., Sun, W., Cheng, Q., & Yang, L. (2021). Effect of water regime and harvest stage on essential oil accumulation in basil plant growing in sandy soil. *Irrigation Science*, 39(6), 727–739. <https://doi.org/10.1007/s00271-021-00719-1>
- Taiz, L., & Zeiger, E. (2010). *Plant physiology* (5th ed.). Sinauer Associates Inc.
- Zolfaghari, M., Nazeri, V., Sefidkon, F., & Rejali, F. (2013). Effect of arbuscular mycorrhizal fungi on plant growth and essential oil content and composition of *Ocimum basilicum* L. *Iranian Journal of Plant Physiology*, 3(2), 643–650.