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Peer review history for:

Ibrahim L. Accumulation of α -tocopherol in *Capsicum annuum* L. influenced by changes in water temperature. *S Afr J Sci.* 2026;122(3/4), Art. #23103. <https://doi.org/10.17159/sajs.2026/23103>

HOW TO CITE:

Accumulation of α -tocopherol in *Capsicum annuum* L. influenced by changes in water temperature [peer review history]. *S Afr J Sci.* 2026;122(3/4), Art. #23103. <https://doi.org/10.17159/sajs.2026/23103/peerreview>

The original manuscript for review is appended below.

Reviewer 1: Round 1

Date completed: 11 November 2025

Conflicts of interest: None

Recommendation:

Accept / Revisions required / Resubmit for review / Resubmit elsewhere / Decline / See comments

REVIEWER: Accept

Does the manuscript fall within the scope of SAJS?

Yes/No

REVIEWER: Yes

Is the manuscript written in a style suitable for a non-specialist and is it of wider interest than to specialists alone?

Yes/No

REVIEWER: Yes

Does the manuscript contain sufficient novel and significant information to justify publication?

Yes/No

REVIEWER: Yes

Do the Title and Abstract clearly and accurately reflect the content of the manuscript?

Yes/No

REVIEWER: Yes

Is the research problem significant and concisely stated?

Yes/No

REVIEWER: Yes

Are the methods described comprehensively?

Yes/No

REVIEWER: Yes

Is the statistical treatment appropriate?

Yes/No/Not applicable/Not qualified to judge

REVIEWER: Yes

Are the interpretations and conclusions justified by the research results?

Yes/Partly/No

REVIEWER: Yes

Please rate the manuscript on overall contribution to the field

Excellent/Good/Average/Below average/Poor

REVIEWER: Good

Should this manuscript be expanded and considered rather as a Research Article?

Yes/No
REVIEWER: Yes
Please rate the manuscript on language, grammar and tone Excellent/Good/Average/Below average/Poor
REVIEWER: Good
Is the manuscript succinct and free of repetition and redundancies? Yes/No
REVIEWER: Yes
The number of tables in the manuscript is Too few/Adequate/Too many/Not applicable
REVIEWER: Adequate
The number of figures in the manuscript is Too few/Adequate/Too many/Not applicable
REVIEWER: Adequate
Are the results and discussion confined to relevance to the objective(s)? Yes/No
REVIEWER: Yes
Is the supplementary material relevant and separated appropriately from the main document? Yes/No/Not applicable
REVIEWER: Yes
Please rate the manuscript on overall quality Excellent/Good/Average/Below average/Poor
REVIEWER: Good
Is appropriate and adequate reference made to other work in the field? Yes/No
REVIEWER: Yes
Is it stated that ethical approval was granted by an institutional ethics committee for studies involving human subjects and non-human vertebrates? Yes/No/Not applicable
REVIEWER: Yes
If accepted, would you recommend that the article receives priority publication? Yes/No
REVIEWER: Yes
Are you willing to review a revision of this manuscript? Yes/No
REVIEWER: No
With regard to our policy on ' <u>Publishing peer review reports</u> ', do you give us permission to publish your anonymised peer review report alongside the authors' response, as a supplementary file to the published article? Publication is voluntary and only with permission from both yourself and the author. Yes/No
REVIEWER: Yes
Comments to the Author: #22-32: I suggest that the Significance section should be a single paragraph. #126-127: The following recent references can further improve the quality of the report.
<ul style="list-style-type: none"> • Chouikhi, M., Ben Salem, A., Jebari, H., & Attia, H. (2024). Agronomic and functional quality traits in various pepper landraces: Carotenoids and tocopherols. <i>Plants</i>, 13(3), 412. • Niu, Y., & Liu, J. (2022). Vitamin E synthesis and response in plants: Mechanisms and roles in stress tolerance. <i>Frontiers in Plant Science</i>, 13, 875401. • Wen, Y., Zhang, D., & Li, H. (2023). Effects of drip irrigation timing and water temperature on greenhouse crop physiology and soil respiration. <i>Agricultural Water Management</i>, 286, 108389. • Zamljen, T., Hudina, M., & Stopar, M. (2024). Impact of irrigation water temperature on vegetable seedling quality and early growth. <i>Agronomy</i>, 14(2), 293.

Author response to Reviewer 1: Round 1

I suggest that the Significance section should be a single paragraph.

AUTHOR: The section was made to be a single paragraph.

The following recent references can further improve the quality of the report.

AUTHOR: Trying to provide additional information can alter the maximum number of word count to exceed 2000.

Reviewer 2: Rounds 1 and 2

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Bioaccumulation of α -tocopherol in chili (*Capsicum annum* L.) pepper fruits influenced by seasonal changes in water temperature

Abstract

A colder or warmer temperature below or above the optimum level can reduce the rate of photosynthesis in higher plants. This in turn, decreases metabolite precursor pools for the biosynthesis of secondary metabolites. The paper aimed to determine the effects of changes in water temperature because of seasonality on α -tocopherol bioaccumulation in chili pepper fruits from an aquaponic system. A portable dissolved oxygen meter was used to measure the water temperature levels. A high-performance liquid chromatography method was utilized to determine the levels of α -tocopherol bioaccumulation. The water temperature levels of the fish water in the winter ranged from 8.3–13.8 °C. However, in the summer, the values ranged from 20.2–25.8 °C. The water temperature levels were significantly differed ($p < 0.05$) between the winter and summer. The α -tocopherol bioaccumulation in chili pepper fruits in the winter and summer were $0.77 \pm 0.13\%$ and $1.64 \pm 0.21\%$, respectively. The bioaccumulation of α -tocopherol differed significantly ($p < 0.05$) between these two seasons. This study revealed that changes in water temperature levels due to seasonality have caused variations in α -tocopherol bioaccumulation in chili pepper fruits. In this research, the warmer season (summer) presented a higher bioaccumulation of α -tocopherol in the studied plant material. Hence, a better period for the production of this compound.

significance:

The research evaluated whether variations of seasonal temperature in South Africa affects α -tocopherol bioaccumulation in chili pepper fruits grown in the aquaponic greenhouse.

With South Africa diverse seasons and fresh water scarcity, the aquaponic technology could provide sustainable solution not only in this country but, in other Africa countries as well. The system integrates hydroponic plants and fish protein production, even with unfavourable climatic regime and limited fresh water.

The study determines the optimal temperature for increased α -tocopherol biosynthesis in higher plants, which allows the understanding of better climate for sustainable vitamin E source in developing worlds.

Keywords: Aquaponic system, bioaccumulation, fish water, portable dissolved oxygen meter, summer.

Introduction

Vitamin E is also known as tocochromanol. It is a vital micronutrient for humans.¹ The synthesis of vitamin E occurs in most photosynthetic plants and some algae.¹ There are four (α , β , γ , and δ) different forms of vitamin E.² Owing to its antioxidant activity, it can neutralize free radicals into less reactive compounds.³ Improper amounts of this vitamin in humans can cause many disease conditions such as cancer, Alzheimer's disease, and cardiovascular disease.⁴ It is mostly occurs in an unesterified form.⁵ In nature, α -tocopherol is the predominant and biologically active form of this vitamin.⁵

The two main environmental factors that can influence crop production are temperature and light intensity.⁶ A high or low temperature above or below the optimal level can decrease the photosynthetic rate in higher plants, which influences the precursor pool by reducing the biosynthesis of secondary metabolites.⁷ Higher heat stress induce the accumulation of tocopherols, especially α -tocopherol.⁸ An optimal level of α -tocopherol is required to maintain the stability of chloroplasts and protect photosystems from singlet oxygen savaging.⁸

Studies concerning effects of changes in water temperature (WRTE) because of seasonality on α -tocopherol bioaccumulation in chili pepper (CIPR) fruits are scarce. However, it was reported that climatic changes are associated with increased carotenoid production in plantains⁹. According to another report, seasonal shifts affect the levels of total phenolic and flavonoid in vegetable plants¹⁰. In addition, low temperatures and high humidity have been linked to reduced levels of chlorophyll a, b, and carotenoids in cucumber¹¹. Therefore, this study was conducted to determine the influence of seasonal variations in WRTE on the bioaccumulation of α -tocopherol CIPR fruits grown in an aquaponic system.

Materials and methods

Aquaponic system site

The aquaponic greenhouse was sited in Makhanda town (formerly Grahams town), Eastern Cape Province, South Africa. The system is exposed to only ambient sunlight and is set up as a coupled commercial system.

Water sample analysis

The WRTE levels of the fish water were measured directly from the tap using a portable dissolved oxygen meter. The tap provides the nutrient water to the CIPR plant growing on the gravel stone media bed. In the winter, the WRTE levels were determined from 29 June 2020 to 31 August 2020. The WRTE measurements in the summer started from 3 December 2020 to 4 February 2021. Each week, the WRTE was measured two times, Mondays and Thursdays throughout the study duration.

75 ***Plant material collection***

76 The CIPR fruits were collected from media bed of the system after eight weeks of WRTE
77 measurements. In the winter, the CIPR fruits were obtained on 31 August 2020. However, they were
78 collected on 4 February 2021, during the summer period.

79

80 ***Chemical reagents and apparatus***

81 The standard (α -tocopherol) was sourced from Sigma-Aldrich, USA; HPLC grade ethanol was
82 provided by Merck, Germany; ascorbic acid was obtained from Merck, South Africa; Whatman No.
83 1 filters were purchased from Whatman, England; and syringe filters were acquired from Goppingen,
84 Germany.

85

86 ***Sample extraction***

87 To 5.0 g of ground CIPR fruits powder, 0.1% ascorbic acid was added to reduce oxidation. Ethanol
88 (30 mL) was then added, vortexed for 10 min, and centrifuged at 3,500 rpm for 10 min. The extract
89 was filtered with Whatman No. 1 filter. The filtrate was then evaporated to dryness using rotary
90 evaporator under reduced pressure. The residue (4.0 mg) was suspended in ethanol (2.0 mL) and
91 filtered through a 0.22 μ m syringe filter. Finally, the filtrate was injected into the HPLC system for
92 chromatography.

93

94 ***Chromatographic conditions***

95 The HPLC system consisted of a connector, a pump, an auto-sampler, and a diode array detector.
96 The detection wavelength ranged from 190–800 nm. The data acquisition and analysis were carried
97 out with "LC Lab Solution" software. A Luna[®] column (150 \times 4.6 mm, 5 μ m C18 (2) 100A) carried
98 out the chromatographic separation. The mobile phase consisted of ethanol and water in a ratio of
99 97:3, v/v. The flow rate was 1.5 mL min⁻¹. The analyte injection volume was 12 μ L for 10 min run. At
100 a wavelength of 298 nm, the analyte was detected.

101

102 ***Statistical analysis***

103 The data analysis was carried out using Student's t-test with Microsoft Excel 365[®] (Microsoft
104 Corporation, New York, USA). The statistical level of significance was 5%.

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Results and discussion

Figures 1 and 2 depict the winter and summer WRTE values. In the winter, the minimum and maximum WRTE levels were 8.3 °C and 13.8 °C, respectively (Figure 1). In contrast, the lowest and highest WRTE values in the summer were 20.2 °C and 25.8 °C, respectively (Figure 2). The WRTE levels were detected to differ significantly ($p < 0.05$) between the two period.

The bioaccumulation of α -tocopherol in the CIPR fruits varied significantly ($p < 0.05$) between the winter ($0.77 \pm 0.13\%$) and summer ($1.64 \pm 0.21\%$) (Table 1). The summer results indicated greater bioaccumulation, however, it was lower in the winter (Table 1).

Plants are exposed to different kinds of abiotic factors that can influence their growth and productivity.¹² Cold or low-temperature stress prevents the growth of plants, resulting in reduced metabolite production.¹¹ However, in higher plants, heat stress was reported to increase accumulation of secondary metabolites¹³. Microorganisms in aquaponic systems are able to mineralize organic compounds if the WRTE is between 25–30 °C.¹⁴ The availability of nutrients to plants is optimal between 21–24 °C.¹⁵ Hence, in this study, WRTE levels of ± 25 °C in the summer were relatively optimal.

Reports on the effects of seasonal changes in WRTE levels on α -tocopherol bioaccumulation in CIPR fruits are lacking. However, α -tocopherol was reported to be present in tomatoes and papers¹⁶. In addition, temperatures ranging from 12–32 °C reportedly decrease the bioaccumulation of lycopene and carotenoids in tomatoes¹⁷. Furthermore, Siberian ginseng grown at an ambient temperature of 24 °C presented a relatively high total phenolic content¹⁸. In this study, the WRTE level detected during the summer increased the bioaccumulation of this metabolite. However, in the winter, a lower concentration was obtained, which might be the inability of this plant to withstand cold stress in the period.

Conclusion

The bioaccumulation of α -tocopherol in CIPR fruits was greater in the summer, however, interrupted in the winter. Thus, in higher plants, bioaccumulation of α -tocopherol can be advantageous for providing adaptive tolerance to temperature stress. Additional studies are needed to elucidate the mechanism of the biosynthesis.

Data availability

The data supporting the results of this study are available upon request to the corresponding author.

148

Funding

149 Not applicable

150

151

Declaration

152 The author declares to have no competing interests. There is no AI or LLM to declare.

153

154 [Ethical approval information was provided to the journal]

155

156 [This work is based on a thesis/dissertation]

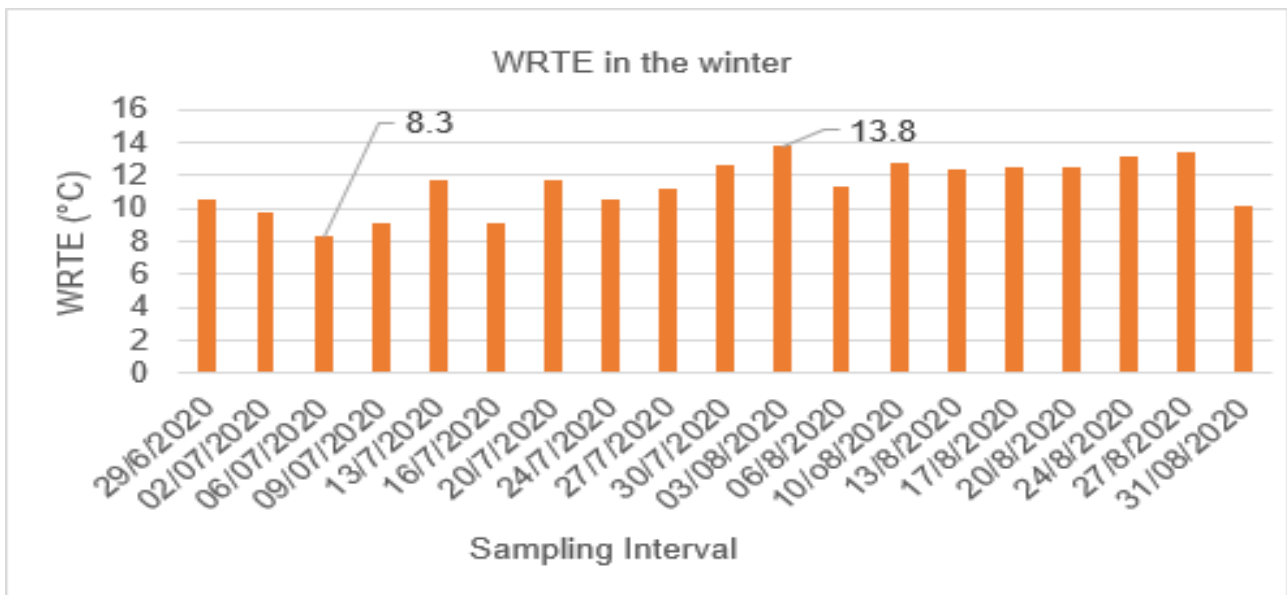
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158 [The figures/tables are provided on the page below the references]

References

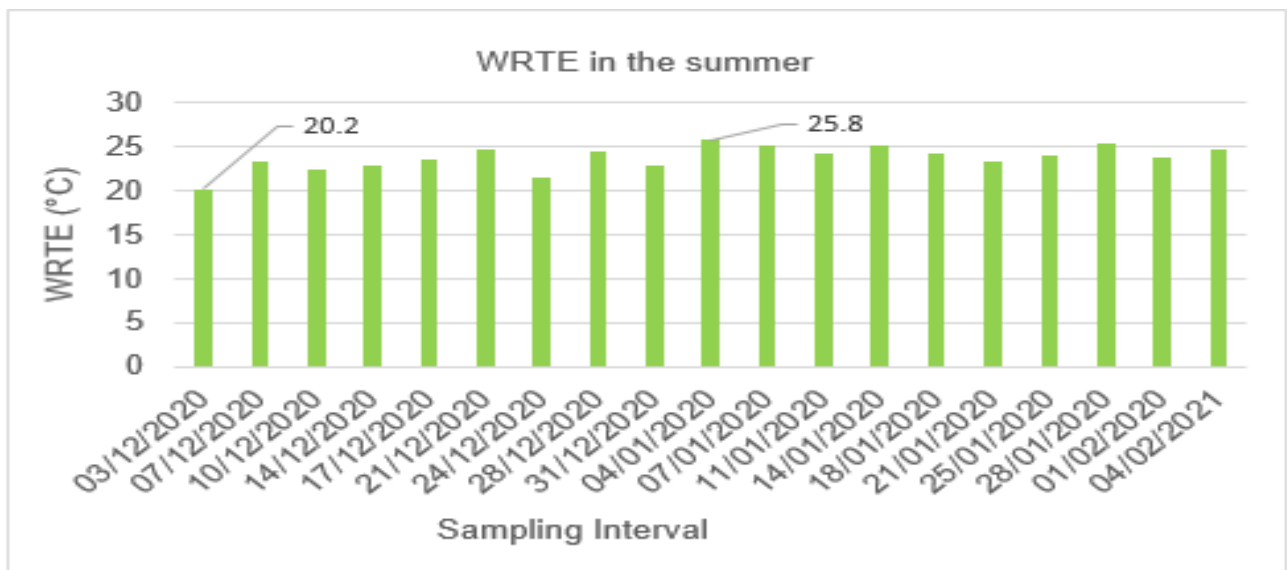
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233 **Figure 1.** WRTE levels in the winter. The highest and lowest WRTE values were 8.3 °C and 13.8 °C,
 234 respectively. WRTE, water temperature.
 235



236

237 **Figure 2.** WRTE levels in the summer., the maximum and minimum WRTEs levels were 20.2 °C and
 238 25.8 °C, respectively. WRTE, water temperature.
 239
 240

241 **Table 1.** Seasonal differences in α-tocopherol bioaccumulation in the chili pepper fruits.

Plant material	CIPR fruits, dw (n = 3)	
	Winter	Summer
α-tocopherol (%)	0.77±0.13*	1.64±0.21*

242 dw, dry weight; CIPR, chili pepper; n, number of replications; *, significant difference (p < 0.05).
 243