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Manipulating tomato plant electric signaling system by microwave radiation to enhance crop productivity and nutritional value

Nilima Kumari, Shalini Verma, Vinay Sharma^{*}

Department of Bioscience and Biotechnology, Banasthali Vidyapith, P.O. Banasthali Vidyapith, Rajasthan 304022, India

ductivity.

ARTICLE INFO Keywords: Bioactive compounds β-1 3-Glucanase Lycopersicon esculentum Mill. Microwave radiation ABSTRACT The present study was undertaken to explore the stimulatory effect of microwave radiation on growth and development of Lycopersicon esculentum Mill. (tomato) plant. These signal stimuli propagate and encode information which alters and manipulate the plant signaling system. Two varieties of tomato seeds i.e. NS-585 and NS-2535 were exposed to microwave radiation at 9.3 GHz for 0–8 h where 0 h seeds were treated as control. Exposure to microwave radiation showed enhanced germination rate and speed germination index and increment in the β-1,3-glucanase activity which clearly suggested the stimulation of the germination rate and seedling vigour of the tomato seedlings. Microwave radiation showed increment in the bioactive compounds like polyphenol and flavonoid, chlorophyll, carotenoid and protein content of the tomato plant exposed for 4–5 h; further increase in the microwave dose reduced the growth and other biochemical content in the tomato plant. Growth

1. Introduction

The world population is steadily increasing and has reached 7 billion today ([Samir and Lutz, 2017\)](#page--1-0). It has been reported that around 805 million people worldwide suffer from food scarcity and over 2 billion people from micronutrient deficiencies ([Holdsworth and](#page--1-1) [Bricas, 2016](#page--1-1)). Among all the commonly consumed vegetables, tomato is the fourth most frequently consumed as it has high nutritive value together with other micronutrient benefits ([Canene-Adams et al., 2005](#page--1-2)). Tomato (Lycopersicon esculentum Mill.) is a major horticultural crop with an estimated global production of over 120 million metric tons. Impact of globalization on horticulture has caused an increasing demand for quality improvement and wider adoption of techniques to meet the high demand of food ([Passam et al., 2007\)](#page--1-3). To fulfill these agendas genetically modified crops (GMO-crops) were developed two decades ago but criticized due to various ethical, legal and economic questions on agriculture system ([Azadi and Ho, 2010\)](#page--1-4). Researchers are working in different fields to develop new and innovative techniques to develop crops which are environment-friendly, disease resistant and affordable with increased productivity and have high nutritive value and shelf life.

In recent years, application of microwave radiation is becoming

popular in food industries for drying, thawing, baking as well as for the inactivation of microorganisms [\(Woo et al., 2000](#page--1-1)). Apart from this, microwave radiation has also the potential to be applied for increasing the crop productivity and nutritional value. Although it has been reported that magnetic fields produce biological and physiological changes in cell structure of plant ([Pietruszewski et al., 2007\)](#page--1-5), there is nearly total lack of any data on microwave radiation exposure and its effects on the non-destructive changes and physiological stress responses of plants. Hence, it is important to explore the effect of artificially inducted electromagnetic radiation on agricultural productivity. It has been reported that microwave radiations do not cause any physical damage to plants but can cause some biological changes [\(Ng,](#page--1-6) [2003\)](#page--1-6). The exposure of seeds to this electromagnetic radiation causes many enzymatic reactions and can stimulate the biosynthesis of 'stress response' in cells ([Pietruszewski et al., 2007](#page--1-5)). In-vitro experiments have showed changes in the enzyme activity as well as stimulation of biosynthetic processes that involve free radicals and expedite the release of enzyme from bound state and release seeds from dormant state ([Ajikumar et al., 2008\)](#page--1-7). Radiation has shown the stimulating effect but the reaction is frequency, amplitude and time dependent [\(Pietruszewski](#page--1-5) [et al., 2007](#page--1-5)). Due to the sensitivity of plants to external stimuli, it is possible to manipulate the electric signaling of plants. Plants respond to

related modification in tomato seedlings were characterized using FT-IR spectroscopy which clearly confirmed the increase in pectin, lignin, protein, carbohydrate and lipid content in the microwave irradiated tomato seedlings. It is suggested that controlled and defined microwave radiation may help in enhancing crop pro-

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[⁎] Corresponding author at: Amity University Rajasthan, Jaipur, Rajasthan 304022, India. E-mail address: vinaysharma30@yahoo.co.uk (V. Sharma).

Fig. 1. Illustration of microwave exposure of tomato seeds using klystron based microwave bench.

external stimuli by various signaling pathways which operate among different plant parts ([Ilík et al., 2010; Mancuso and Mugnai, 2006](#page--1-8)). The plant electric signaling pathway usually transmits signals at high rate and on the basis of signal transmission rate and mode; these transmitting signals are divided into action, variation potential and system potential [\(Stahlberg and Cosgrove, 1997; Stankovic et al., 1997, Volkov](#page--1-9) [and Ranatunga, 2006; Zimmermann et al., 2009](#page--1-9)). These signals collectively propagate as the plant electric potential varies and transmits encoded information which is determined by the signal's shape ([Krol](#page--1-10) [et al., 2006; Fromm and Lautner, 2007](#page--1-10)). Therefore, if the plant electric potential is manipulated by any external stimuli, it is possible to transfer incorrect information. These signal stimuli propagate and encode information which alter or manipulate the plant electric potential ([Ilík et al., 2010; Zimmermann et al., 2009\)](#page--1-8). This alteration principle can be used to send incorrect information to disturb the electric signaling system of plant. Same principle was used by [Stankovi](#page--1-11)ć and [Davies \(1997\) and Volkov et al. \(2007\)](#page--1-11) for manipulating the plant electrical signaling system. Nowadays, the electromagnetic radiation in the environment has increased due to the massive use of mobile phones and wireless communication ([Hyland, 2005\)](#page--1-12). Radiations from different sources have greatly affected the ecosystem, plants and various organisms ([Roux et al., 2006; Sharma et al., 2009; Tkalec et al., 2007](#page--1-13)). Plant system is complex and therefore different biological processes will response in different manner to external electromagnetic field (EMF) stimulus. The knowledge of parameters of seeds and EMF is necessary to understand the correlation between physiology of growing seedlings and the electromagnetic stimuli. For example increasing magnetic field strength has found in improving the yield of wheat, soybean and cotton ([Senavirathna and Asaeda, 2014\)](#page--1-14). However evidence for such effects in plants is lacking and investigating this will broaden the understanding of the impact of extensive use of EMF as agronomic tool for crop productivity. Therefore, we conducted this study to investigate the effect of radiation in the range of wireless communication on Lycopersicon esculentum Mill. To the best of our knowledge, this study represents the first attempt to investigate the effect of radiation on plants.

2. Material and methods

2.1. Plant material

Two varieties of tomato seeds i.e. NS-585 and NS-2535 were procured from Namdhari seeds, Bengarulu, Karnataka. NS-585 is determinate type with acidic flat round shape fruit variety suitable for Rabi season and is intermediately resistant to tomato leaf curl virus whereas NS-2535 is fresh market - dual purpose determinate type with oval fruit shape suitable for Kharif season. Tomato seeds of both the varieties were surface sterilized in 0.1% (m/v) HgCl₂ for 2 min and then washed several times with distilled water. These sterilized tomato seeds were then soaked in distilled water for 12 h and used for exposure with microwave radiations. After this, the tomato seedlings were evenly distributed in petriplates placed inside the artificial faraday cage for microwave exposure. Filter paper was moistened with 3 ml of distilled water before microwave exposure. Immediately after the exposure to microwave radiation, seeds were germinated on filter paper in petri dishes with regular supply of 1/5 strength of Hoagland's solution ([Hoagland and Arnon, 1950\)](#page--1-15) in a growth chamber for 5 d. Then, healthy tomato seedlings were transferred to pots containing 200 g of sterilized soilrite and grown under a diurnal cycle of 9 h light and 15 h dark and the day/night temperature 25 ± 2 °C and 15 ± 2 °C respectively with relative humidity of 60/70% and watered regularly.

2.2. Exposure of tomato seeds with microwave radiations

The experiments were conducted in two independent sets with three replicates each. The imbibed seeds of the two varieties in triplicate each containing 50 seeds were exposed to microwave radiation for different time intervals, viz. 0 (control), 15, 30, 45, 60 min and further up to 12 h (microwave irradiated) at the interval of 1 h. Exposure of tomato seeds with microwave radiation was performed following the methodology of [Ragha et al. \(2011\).](#page--1-16) The exposure system contained: a Function Generator (MARS, ME957 5 Hz), a Klystron power supply (MARS, ME 6000 kp), a Klystron tube, Isolator, Frequency Meter, a Variable Attenuator, Detector Mount, VSWR Meter, Oscilloscope (MARS, ME3020 20 MHz CRO) and an artificial faraday cage, a closely shielded chamber with aluminum sheet of 2 mm for exposing tomato seeds evenly distributed over moistened filter paper in petriplates with the microwave radiation to create homogenous EMFs and to protect from external EMFs as shown in [Fig. 1](#page-1-0) ([Singh et al., 2012](#page--1-17)). At the beginning, during and till the end of exposure period, the temperature inside the chamber was measured using a thermometer. The temperature inside the chamber and on the seedlings surface did not vary more than \pm 0.1 °C. Seedlings surface temperature was measured as explained by [Oleskog](#page--1-18) [and Sahlén \(2000\)](#page--1-18). Temperature was measured after each hour of irradiation treatment using thermocouple, copper constantant type-T thermocouple 0.2 mm diameter by carefully placing the thermocouple over the non-micropylar region of the seed. The tomato seedlings were radiated to 9.3 GHz field with pulse modulation of 1.3 KHz and Field strength of 0.2 V m⁻¹ corresponding to the power flux density of 3.2 mW m−² . [Durney et al. \(1975\)](#page--1-19) have given the expression for the specific absorption rate (SAR) value for the prolate spheroid objects. According to this formulation, we calculated SAR value for both the NS-585 and NS-2535 varieties of tomato seed.

2.3. Evaluation of seed germination and seedling vigour

The irradiated tomato seeds were placed in petriplates with wet filter paper and covered with black blotter. The plates were placed in artificial growth chamber at 25 °C for 8 d with 9-h light/15-h dark photoperiod. The plates were observed daily for 8 d. The seeds were considered to be germinated when the length of root is similar to the seed length and length of shoot similar to half length of the seed. Germination percentage and germination index (GI) of seeds were calculated according to [Siddiqui and Al-Whaibi \(2014\).](#page--1-20)

2.4. $β-1$, 3-glucanase activity

2.4.1. Enzyme extraction

Microwave irradiated and non-irradiated tomato germinated seeds

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