STUDY ON FRUIT CHARACTERISTICS OF NETTED MUSKMELON (*Cucumis melo* L., 'DANDY-449' AS AFFECTED BY GIBBERELLIC ACID (GA_3) APPLICATION AND SCHEDULED IRRIGATION

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STUDY ON FRUIT CHARACTERISTICS OF NETTED MUSKMELON (*Cucumis melo* L., 'DANDY-449') AS AFFECTED BY SCHEDULED IRRIGATION AND GIBBERELLIC ACID (GA_3) APPLICATION

A thesis presented by

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to

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The thesis attached hereto, entitled "**Study on Fruit Characteristics of Netted Muskmelon (*Cucumis melo* L., 'Dandy-449') as Affected by Gibberellic Acid (GA_3) Application and Scheduled Irrigation** " was prepared under the direction of the chairperson of the candidate's Supervisory Committee and has been approved by all members of that committee and board of examiners as a partial fulfillment of the requirements for the degree of **MASTER OF AGRICULTURAL SCIENCE (HORTICULTURE AND AGRICULTURAL BIOTECHNOLOGY).**

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DECLARATION OF ORIGINALITY

This thesis represents the original works of the author, except where otherwise stated. It has not been submitted previously for a degree at any University.

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Date -

DEDICATED TO MY BELOVED PARENTS,
U THEIN WIN AND DAW THI THI SAN

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Study on Fruit Characteristics of Netted Muskmelon (*Cucumis melo* L., 'Dandy-449') as Affected by Gibberellic Acid (GA_3) Application and Scheduled Irrigation

ABSTRACT

Netted muskmelon (*Cucumis melo* L.) is a high demanding fruit for fresh consumption and juice. The producing areas of this fruit occupy Sagaing, Mandalay and Yangon Regions as majority in Myanmar. The fruit, however, does not consistently show desirable market quality in regards of fruit characteristics in response to improper irrigation during fruit growth in Myanmar. As a bridge to that problem, the study was conducted to investigate effects of gibberellic acid (GA_3) application and scheduled irrigation on fruit characteristics of netted muskmelon, cultivar 'Dandy-449'. Investigations were done in two experiments under plastic house condition. Fruit characteristics observed were fruit diameter, fruit weight, flesh thickness, cuticular membrane (CM) thickness, crack number (net) on fruit skin, Brix (total soluble solid), and seed weight.

Experiment 1 was focused on fruit characteristics in response to GA_3 concentrations (0, 10, 20, 30 and 40 ppm) and spray methods (foliar and fruit sprays) at 7 days after full bloom (DAFB), which was early fruit growth stage. Fruit diameter, fruit weight and flesh thickness of 'Dandy 449' showed remarkably increase by GA_3 40 ppm. GA_3 10 ppm application was as good as 40 ppm for those three characteristics of fruits. Crack number (netting) per unit area in fruit skin resulted in no effect by GA_3 application and scheduled irrigation. Nevertheless, GA_3 10 ppm sprayed to fruit tremendously increased CM thickness. That concentration of GA_3 also promoted Brix of the fruit. Based on the results, 10 ppm of GA_3 was considered an appropriate concentration for 'Dandy 449'.

Experiment 2 was proceeded focusing on fruit characteristics affected by GA_3 application (+ or -) and scheduled irrigation (optimum irrigation, OI or deficit irrigation, DI) at stage 1 and 3 of fruit growth. GA_3 application increased fruit diameter and fruit weight irrespective of scheduled irrigation. However, flesh thickness of the fruit did not show any effect of prescribed treatments at both of fruit growth stages. GA_3 application and scheduled irrigation increased CM thickness at stage 1, respectively, while the interactive effect of those treatments revealed by

increased CM thickness at stage 3. GA_3 application with respect to optimum irrigation accelerated increment of crack number (netting) per unit area in the fruit skin at stage 1, whereas only optimum irrigation promoted netting at stage 3. Again, optimum irrigation raised seed weight at stage 1, while GA_3 application increased seed weight at stage 3. Non- GA_3 application interactive with deficit irrigation promoted Brix of the fruit at stage 3. The results highlighted that most of fruit characteristics of netted muskmelon, 'Dandy 449', can be promoted by GA_3 application to fruit. Optimum irrigation could promote crack (net formation) on the fruit skin. Deficit irrigation at stage 3 of fruit growth enabled to increase Brix of the fruit.

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1. INTRODUCTION

Muskmelon (*Cucumis melo* L.) is a popular fruit widely used for fresh consumption and juice in Myanmar all round the year. It is extensively cultivated in 'Sagaing', 'Mandalay', 'Yangon' Regions and 'Shan' State. Amongst them, 'Chaung-Oo', 'Mon-Ywa' and 'Butalin' townships of 'Sagaing' Region are the most leading areas of muskmelon production. This fruit is produced with the focus of export market to China in dry season, while local market to 'Mandalay' and 'Yangon' in rainy season. Different cultivars of muskmelon with and without net in fruit skin are marketed to both destinations.

The cultivar with net in fruit skin (netted muskmelon) is a high-demanding one for its market-attractive appearance (Personal Communication, 2011). Nevertheless, it is a crack-prone cultivar when it is particularly produced in dry season. Cracks appear in the fruit skin: 1) during early stages of fruit development at the onset of netting and 2) during fruit maturation (Personal Communication, 2011). During these stages, high moisture level in the air or soil (dew or rain) following dry spell might induce cracks in the fruit skin.

The fruit skin is an envelope composed of cuticle, and epidermal and subepidermal cell layers (Sekse, 1995a,b). Fruit development (fruit size increment) applies tensile force to the fruit skin, in particular of the cuticle as the outermost coverage (Considine and Brown, 1981). If there is a mismatch between fruit size increment and cuticle development, the cuticle begins to rupture (Knoche and Peschel, 2007a; Knoche et al., 2004). As a consequence, water influx through the cuticular ruptures into the fruit degrades the walls of the epidermal cells and eventually there occurs fruit cracking. Cracks downgrade the fruit quality and are very uneconomical in sales. The cracks are also primary opening for fungal infection which can result in fruit rot.

The cuticle at the outermost part of the fruit skin is a hydrophobic layer composed of waxes, cutin and cellulose fibrils. Its development provides the fruit firmness. However, some fruits (European plum and sweet cherry) do not possess continual development of the cuticle (Knoche and Peschel, 2007a; Knoche et al., 2004). In such case, development of the cuticle is enhanced by external gibberellins (GAs) increases cuticle thickness in tomato (Knoche and Peschel, 2007b). The

thicker the cuticle, the more likely the fruit is resistant to crack. Nevertheless, there was no precise indication of the role of GAs in muskmelon reported yet.

Giving the proper amount of water to melon is crucial to get maximum crop yield. The more amount of irrigation water applied, the more muskmelon fruit yield (increasing fruit size, fruit weight and flesh thickness) can be realized (Zeng et al., 2009). However, excessive soil water can damage melon and cause fruit quality problems (Zeng et al., 2009). Deficit irrigation (DI) which can be defined as the practice given to fully-irrigated crops from which water is withheld during certain tolerance growth stages (Geerts and Raes, 2009). Although DI minimizes the evaporated water loss, DI followed by regular water uptake cannot control volume expansion of fruits (Geerts and Raes, 2009). In some cases, that can fracture the cuticle of the fruits and reduce economic profits.

In Myanmar, research work on quality fruit production of muskmelon under application of plant hormones and irrigation practices is relatively little developed. In order to provide feasible information, it was hypothesized that 1) concentration, method and GA_3 application can play a role in fruit growth and cuticle deposition of muskmelon and, 2) GA_3 application with scheduled irrigation during fruit growth would enhance fruit size and quality.

Thus, this study was carried out with the following objectives:

- to observe the role of GA_3 during fruit growth of netted muskmelon and
- to examine fruit characteristics of netted muskmelon affected by GA_3 application and scheduled irrigation during fruit growth

2. LITERATURE REVIEW

2.1 Botanical Characteristics of Muskmelon

Muskmelon (*Cucumis melo* L.) is classified under Cucurbitaceae family. The Cucurbitaceae family consists of mostly frost sensitive, principally tendril-bearing vine plants which are found in sub-tropical and tropical regions around the world. Optimum temperature for melon production is 34ºC and permissible range is from 10ºC to 45ºC (Baker and Reddy, 2001). Plants consist of a main or primary stem, from which lateral stems or branches are produced. In turn these branches may bear more lateral stems. Leaves are simple, either three or five lobed, and borne singly at the nodes. Coiled tendrils occur at leaf axils, and act to help the plant cling to trellises and other supports. The plant has a relatively strong tap root which is thought to penetrate to approximately 1m depth depending on soil type and irrigation arrangement (Mauynard, 2007; Long, 2005; Hector, 2005).

Sex expression in cucurbits is influenced by genetic, environmental, and hormonal factors. Monoecious strains of muskmelon bear staminate (male) and pistillate (female) flowers. Gynoecious strains normally produce only pistillate flowers. Staminate flowers occur in axillary clusters on the main stem and laterals, whilst ovary bearing flowers occur at the first node of each lateral branch (Long, 2005; Hector, 2005).

Fruits of muskmelon are generally classified as an indehiscent pepo, with three ovary sections or locules. There are different types of flesh color (orange, orange light or pink, green, white or even mixture of these colors), rind color (green, yellow, white, orange, red, gray or blend of these colors), rind texture (smooth, warty, striped, netted, rough or combination of these textures), form (round, flatten or elongated), and size (from 4 up to 200cm) (Kirkbride, 1993; Goldman, 2002). Muskmelons typically mature 42 to 46 days after pollination. When ripe, the melon rind changes from green to tan or yellow between the netting and they are harvested at full-slip; i.e., when the stem separates easily at the point of attachment. It should be store at 0ºC and 95% relative humidity (Andersen, 1914).

2.2 Fruit Development

Fruit development is a beautifully complex process that begins with the change from a vegetative to a floral meristem and ends with mature fruit and viable seed (Ozga and Dennis, 2003). The developmental processes of fruits from gynoeciums in higher plants can be divided into three distinct phases. The earliest phase involves the development of the ovary, generally referred to as fruit set. In the second phase, fruit growth is due primarily to cell division whereas in the third phase, growth occurs mainly by cell expansion until the fruit reaches its final size (Gillaspy et al., 1993).

In Cucurbit, fruit grows exponentially for a period after fruit set, and then the growth rate relatively slows. The increase in fruit size after pollination is largely as a result of cell expansion rather than an increase in the number of cells (Mauynard, 2007).

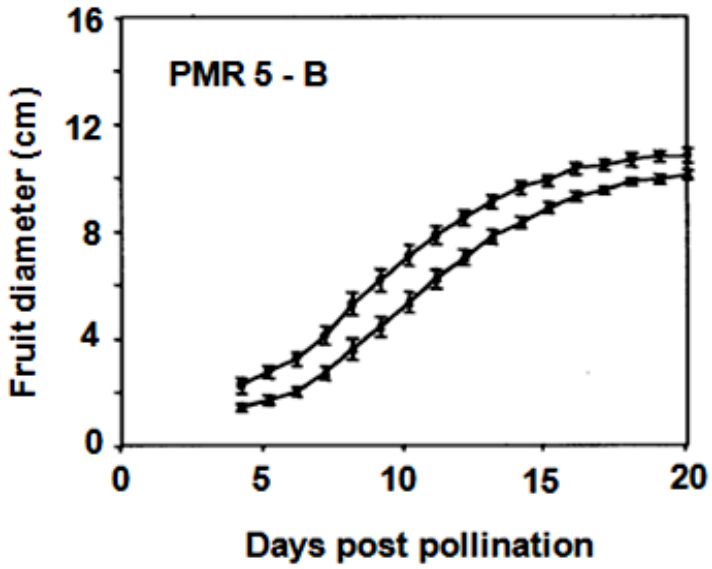

Figure 1 Growth patterns of netted muskmelon PMR5-B. (▲): the growth diameter of equatorial axis and (●): the growth diameter of longitudinal axis (Adapted from Keren- Keiserman et al., 2004)

Increase in pericarp thickness or fruit size increment was due to increase in the number of pericarp cells only occurred during the first few days after anthesis, where after growth slowed down. And also increased cell size occurred during the first 14 days after anthesis (DAA). The first sign of lenticle (net from peel) development was

observed at 14 DAA and they were well developed at 28 DAA. Towards the end of the fruit development period, fruit mass and volume increased at a lower rate but growth did not cease completely (Combrink et al., 2001; Keren-Keisermen et al., 2004). However, Mc Glasson and Pratt (1963) showed that the growth of the individual muskmelon fruit resembles a sigmoid curve, as found with other cucurbits.

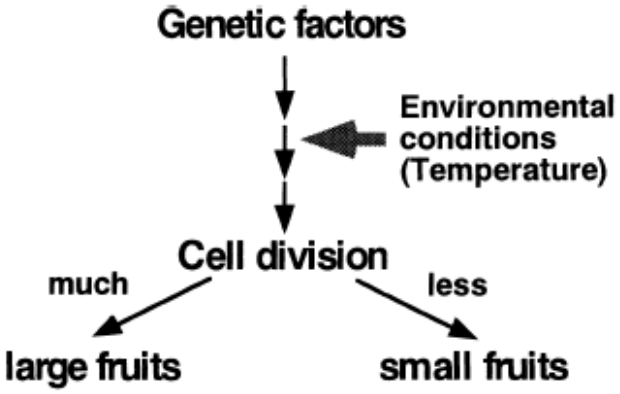

Figure 2 Schematic model showing that, the size of melon fruits is affected by their genetic factors which control cell divisions occurring in the fruit pericarp region (Katsumi et al., 1999)

As shown in the above model of melon fruit development (Figure 2), the size of muskmelon fruit is defined by the cell number in the pericarp, and the difference in the cell number is mediated by the different genetic factors controlling cell proliferation which is modified by temperature (Katsumi et al., 1999). The sink-source relationship is also regarded as an important factor in the determination of fruit size (Bohner and Bangerth, 1988).

2.3 Structure of Muskmelon Peripcarp

During muskmelon fruit development, the ovary becomes the pericarp, which consists of the hard rind (epicarp - the most-outside layer or peel of the frutis), and the soft middle layer (mesocarp - usually the major part of the fruit that is eaten) and the innermost layer containing seeds (endocarp) (Gillaspy et al., 1993). They reported that the pericarp is covered by a thin cuticle at the outermost layer that thickens as the fruit ages.

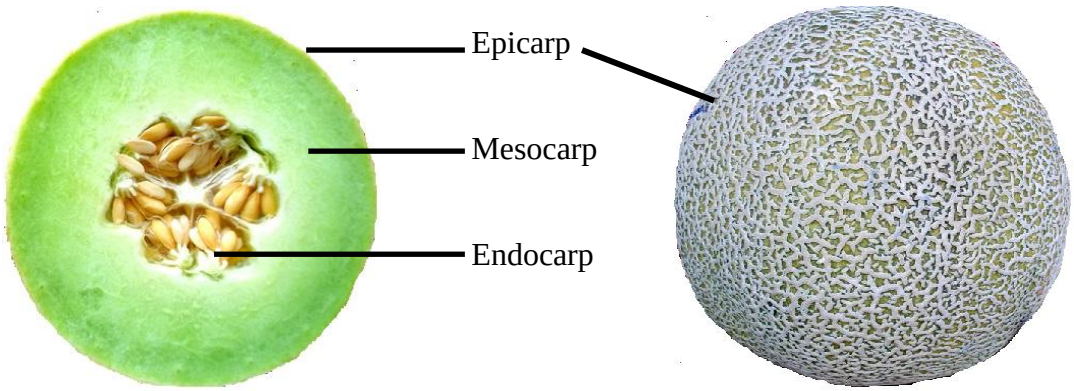

Plate 1 Muskmelon (*Cucumis melo* L.) pericarp in cross-section and fruit in side view

2.4 Rind Behavior of Muskmelon

In netted varieties, the fully developed net is important for market quality of fruit. Complete netting of the fruit is one of the factors that determine melon quality, and also serve as a protection against mechanical injury (pre- and postharvest). Furthermore, from the point of view of the consumer, the netting pattern is of aesthetic importance, and thus has a high market value (Keren Keiserman et al., 2004).

The development of the net tissue has been characterized as a response to cracking of the fruit surface (Meissner, 1952). The netting starts towards the end of the fruit-expansion stage. An immature net is a shallow, greenish, non-dried tissue that protrudes only slightly above the fruit surface. As the fruit enlarges, the cracks deepen and widen and disrupt both the cuticle and some subtending epidermal and hypodermal cells. Below these fissures, periderm cells start multiplying, and produce masses of cells with suberized walls that extend above the fruit surface (Natalie et al., 2008; Keren-Keiserman et al., 2004). Periderm is a secondary tissue made up of three cell types; phellem, phellogen and phelloderm (Natalie et al., 2008). Suberization has been suggested to be a fundamental process involved in wound-healing in plants (Dean and Kolattukudy, 1976). Hence, the cracks that appear in the melon rind can be viewed as wounds and the resultant net tissue as a healing periderm.

The cutinized membranes of suberized cell wall layers with wax depositions develop below the net fissures at the fruit ripening stage and can act as a barrier to moisture loss in muskmelon fruit (Puthmee et al., 2013) and penetration of pathogen (Natalie et al., 2008).

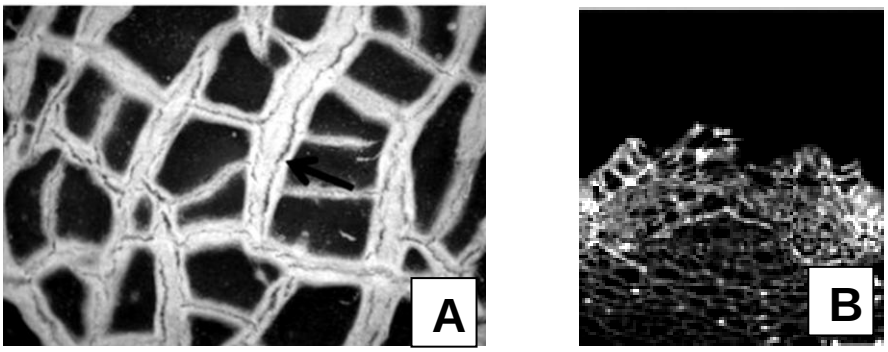

Plate 2 Development of net-like wound-induced periderm; (A) Natural rind (Puthmee et al., 2013), (B) Filling of the fissures with peridermal cells (Puthmee et al., 2013)

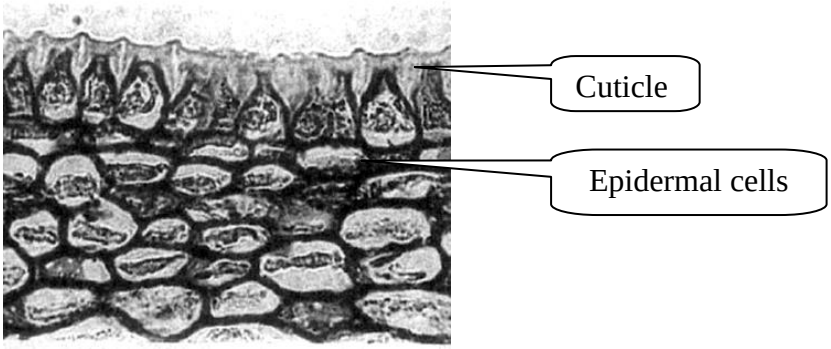

Plate 3 Epidermal tissues in the fruit-peel of 'Galia' melons at 42 days after anthesis (Combrink et al., 2001)

Combrink et al. (2001) founded that the fruit-peel surface increased during the first three days after anthesis without any change in epidermal cell size or shape that may of be ascribed to epidermal cell division only. During the following 11 days the walls of the epidermal cells enlarged significantly. At 14 days after anthesis a well-developed cuticle probably prevented further epidermal cell division and enlargement. The increase in fruit-peel area after this stage could have caused cracks in the epidermal layer.

2.5 Plant Cuticle and Its Function

The plant cuticle is an extracellular hydrophobic layer that covers the aerial epidermis of all land plants. The cuticle appears to play an important role in defining organ boundaries during development. The cuticle that typically consists of an external layer of epicuticular waxes and epicuticular wax crystals overlying a comparatively thin layer of lipids the cuticle proper that covers an inner layer of

waxes and fibrous polysaccharides embedded in a cutin matix (the cuticular layer). The cuticle proper and cuticular layer comprises the cuticular membrane (CM) (Yeats and Rose, 2013; Matas et al., 2004). Cuticles vary considerably in their architecture and, depending on species and ontogeny, differ dramatically in thickness, from the nanometer to the micrometer scale (Yeats and Rose, 2013; Dominguez et al., 2011).

The main function of cuticle is protection against water loss by controlling the movement of water between two compartments: (1) the outer cell wall of the epidermis and (2) the atmosphere adjacent to the plant, together with regulation of gas exchange (Yeats and Rose, 2013; Markus and Lukas, 2001). However, the cuticle has evolved other functions, such as protection against mechanical injury from the environment or in association with an attack of microorganisms or pests and attenuation of UV light absorption. Plant cuticles are composite structures, the main constituent being lipid polymer cutin and a variety of organic solvent-soluble lipids that are collectively termed waxes (Yeats and Rose, 2013).

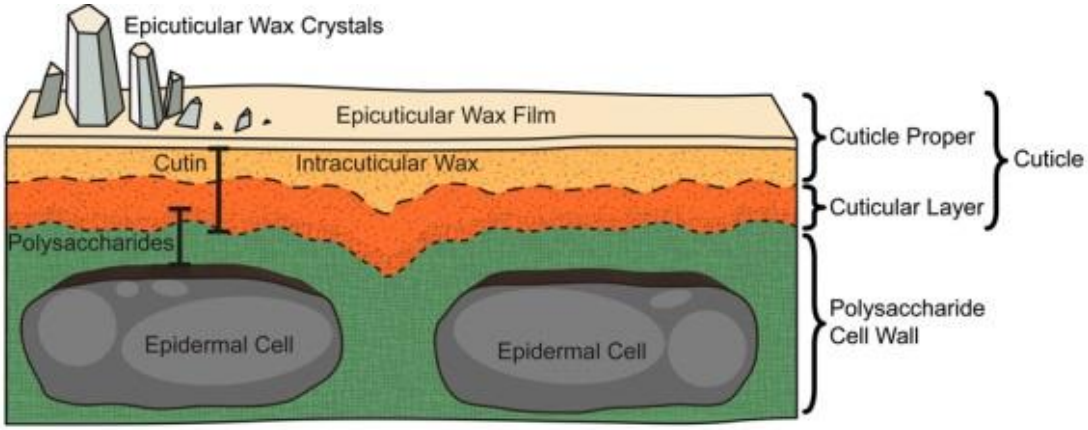

Plate 4 Schematic diagram highlighting the major structural features of the cuticle and underlying epidermal cell layer (Yeats and Rose, 2013)

Dominguez et al. (2011) concluded that during growth and development, the plant cuticle responds to some abiotic and hormone stresses by changes in cuticle thickness and deposition, and also by modifying the ratio of some cuticular components. Such alterations have been noticed, for example, as a result of the influence of plant hormone application, water deficiency and in association with iron deficiency chlorosis.

2.6 Role of Plant Growth Regulator (GA_3) on Fruit Size and Quality of Muskmelon

The plant hormones are signal molecules that regulate many processes of plant development, including fruit development leading to mature fruit and viable mature seed. As other growth hormones, gibberellins have been implicated at various stages of fruit development (Ozga and Dennis, 2003).

The plant growth regulators (PGR) act as messengers and are needed in small quantities at low concentrations. Generally their site of action and biosynthesis are different. Most of the PGR exhibit a broad spectrum and thus a single PGR may influence several entirely different processes (Tuan and Ruey, 2013). Among agricultural practices which may increase the fruit production and improve the quality of several other fruit crops are the applications of plant growth regulators, especially gibberellic acid (Chao and Lovatt, 2010).

Considering role of growth regulators at cellular levels in the processes of growth and development of a fruit from the ovary following pollination and fertilization, two important physiological processes occur- 1) increase in volume of the fruit and 2) movement of metabolites into the fruit (Deepthi, 2008).

Gibberellic acid has been used widely in horticultural crops for cracking of pomegranate fruit (Tuan and Ruey, 2013) and sweet cherry (Yildirim and Koyuncu, 2010), cuticle deposition in tomato fruit (Knoche and Peschel, 2007b). Melon fruits also improve their quality and delay senescence with pre-harvest GA_3 application (Georgia et al., 2008).

GA_3 attributed to increase in cell division and cell elongation (Taiz and Zeiger, 2006; Davies, 1988). According to the effects of increase cell division and cell elongation, the exogenous application of GA_3 enhanced fruit growth of tomato (Serrani et al., 2007) and increased fruit size of sweet cherry (Yildirim and Koyuncu, 2010). Gibberellic acids $50mgL^{-1}$ (50 ppm) promoted fruit diameter of apple uniform branches as compared to control (Moneruzzaman et al., 2011). Similarly, $GA_3$100µM application at anthesis of melon plants enhanced fruit diameter by 56% of the control (Georgia et al., 2008).

The maximum fruit weight was indicated with GA_3 (20 ppm) fruit dipping in muskmelon (Deepthi, 2008), spraying with 30 ppm GA_3 to small flower buds in apple (Tuan and Ruey, 2013), 15 ppm and 40 mg L^{-1} (40 ppm) GA_3 in sweet cherry (Yildirim and Koyuncu, 2010; Cline and Trought, 2007) when compared to control by the effects of cell elongation.

The flesh thickness was significantly influenced by application PGRs. The application of GA_3 at 30 ppm at small bud and petal fall stage of apple considerably increased flesh thickness after application compared to the untreated control (Tuan and Ruey, 2013). Similarly, the application of 30ppm GA_3 increased flesh thickness of melon fruits (Budiastuti et al., 2012).

Plant hormones especially auxins and gibberellin have the key role in ovary and ovules development following fertilization. After fertilization, the process of seed development comprising both endosperm proliferation and embryo growth showed regulation by gibberellins and other growth promoting hormones and resulted in increase in seed size and considerably seed weight (Ruan et al., 2012).

The cuticular membrane (CM) is a protective barrier against water loss and invasion of pathogens (Jeffree, 1996). To maintain these functions, CM required continual deposition during growth and development. A mismatch of CM deposition and fruit surface expansion during development had been implicated in a number of surface disorders (Knoche and Peschel, 2007a). GA_3 application was able to induce increase in CM deposition by promoting the main composer of cuticle that positively affected the thickness of the epiderm and cuticular layer of deep water rice internodes (Hoffmann and Kende, 1994), tomato (Knoche and Peschel, 2007b) and sweet cherry fruits (Cline and Trought, 2007).

Gibberellic acid could promote not only flower, fruit and seed development but also enhance the source potential and redistribution of photosynthates increasing sink strength. GA promoted sucrose synthesis within the leaf through their positive effect on fructose and sucrose phosphate synthase (Iqbal et al., 2011). In addition, GA stimulated the conversion of complex polysaccharides into simple sugars (Kher et al., 2005). The increased translocation of assimilates into the fruits in response to hormonal stimulation could be attributed to increase in total soluble solids (TSS) (Kaur, 2000).

Brix % or total soluble solid (TSS) content was considered an important quality parameter of any fruit. There was a significant increase in TSS content of fruits due to GA_3 application. The highest TSS content was recorded in GA_3 (60 ppm) foliar application followed by GA_3 (20 ppm) as muskmelon fruit dipping (Deepthi, 2008). The similar result of increased total soluble solids content were also found in the application of 30 ppm GA_3 spraying to apples at small flower bud stage (Tuan and Ruey, 2013).

2.7 Role of Irrigation Water to Fruit Size and Quality of Muskmelon

Total volume of irrigation water was highly influenced to fruit yield and its components (Fabeiro et al., 2002). Giving the right amount of water to irrigate the melon is crucial to get maximum crop yield and save more water for domestic and agricultural purposes. Excessive application of water can damage melon and cause fruit quality problems, leading to reduction of the melon fruit yield, lower fruit quality and lower plant resistance (especially to diseases) (Sensoy et al., 2007). Therefore, irrigation must be scheduled to avoid excessive irrigation that can lead to lower quality and plant disease (Zeng et al., 2009). Reducing the irrigated water without limitation may be lead to reduce the agriculture production. The solutions are to manage the way and amount of water application to the agricultural crops with maintaining the economic value of crop yield (Al-Mefleh et al., 2012).

Deficit irrigation has been widely investigated as a valuable and sustainable production strategy in dry regions. This practice aims to maximize water productivity and to stabilize yields. Deficit irrigation is successful in increasing water productivity for different crops without causing severe yield reduction (Geerts and Raes, 2009).

Water deficit during the blooming stage had the lowest production, at setting stage both quantity and quality, and at ripening stage principally to quality (sugar content) (Zeng et al., 2009).

Increasing the quantity of irrigation will increase the yield of field grown melon (Sensoy et al., 2007). High crop yield was found under the treatments using the greatest frequency and quantity of irrigation (Sensoy et al., 2007), while most of the fruit characteristics of melon were significantly affected by differences in irrigation treatments (Al-Mefleh et al., 2012).

The irrigation levels had a positive correlation when increasing irrigation level that could increase the rate of melon fruit growth (Al-Mefleh et al., 2012). When the more total irrigation water applied, the larger the fruit size (fruit diameter and length) in muskmelon (Zeng et al., 2009). Water stress had significant effects on fruit size. Average fruit size was lower for the fruit from the deficit irrigation than form the optimum or excess irrigation in peaches (Crisosto et al., 1994).

The more amount of irrigation water also produced the larger fruit weight and could increase flesh thickness. The more amount of irrigation water applied, the more muskmelon fruit yield can be got. The increase in muskmelon yield can be explained by the mean fruit weight (Zeng et al., 2009). The largest and the smallest flesh thickness values obtained from maximum water amount and minimum water amount. Cabello et al. (2009) reported that when the fruit, as a result of irrigation, varied in size, and accordingly did flesh thickness.

Water availability plays a major role in the regulation of seed development. Water relations of developing seeds were determined primarily by phloem transport and nutrients and accompanying water are imported into maternal seed tissues (Zhang et al., 2007). That influenced seed number at seed set and determines their final size (Zhang et al., 2007).

The regulation of cuticle biosynthesis is complex and involves interacting signaling networks associated with environmental stress responses (Yeats and Rose, 2013). Cutin and wax biosynthesis is the main regulators of cuticle development that was induced by water deficit. The fruit from the deficit irrigation treatment showed a thicker cuticle than fruit from the optimum and excess irrigation treatments (Crisosto et al., 1994).

Sugar content is an important factor to increase the flavor of muskmelon (Mirabad et al., 2013). The concentration of sugars in the fruit became higher with a lower irrigation intake (Fabeiro et al., 2002; Long et al., 2006). Crisosto et al. (1994) also reported that brix % was higher for peaches from the deficit irrigation than those from the optimum or excess irrigation. The soluble solid content improvement was resulted from a reduction of water import to the fruit, the same mechanism as with water stress (Al-Mefleh et al., 2012).

3. MATERIALS AND METHODS

The experiments were conducted at the Research Farm of Yezin Agricultural University (19˚ 15'N and 0˚ 7'E) from November, 2011 to March, 2012 (Experiment 1) and from December, 2013 to April, 2014 (Experiment 2). The muskmelon cultivar used in both of the experiments was netted muskmelon 'Dandy-449' (Aye Yar Waddy Seeds Co. Ltd., Myanmar) that has greenish yellow rind, greenish flesh and round shape. The plants were grown in polythene bags (28" x 17") containing growing medium (1:1 (v/v) proportion of cow dung and garden soil) under plastic house conditions in both experiments.

3.1 Experiment 1: Concentration and Spray Method of Gibberellic Acid (GA_3)

3.1.1 Experimental design

The experiment was conducted in two-factor factorial Completely Randomized Design.

Factor A: Gibberellic acid (GA_3) concentration (0, 10, 20, 30 and 40 ppm)

Factor B: Spray method (foliar spray and fruit spray)

Gibberellic acid was applied at predetermined early growth stage of muskmelon fruits. There were ten treatments in this experiment and 27 plants which bore 2 fruits per plant in each treatment out of 260 plants as total population.

3.1.2 Experimental procedure

3.1.2.1 Nursery practice

Firstly, the tested cultivar 'Dandy-449' seeds were germinated in closed carton box containing mixture of cow dung and garden soil at 32 ºC, and then transferred into plastic seedling trays of 50 cells filled with garden soil and peat moss in 1:1 proportion. The trays were watered regularly and the seedlings of two-true-leaf stage were transplanted to polythene bags in the experimental site with the spacing of 0.8 m within plants and 0.9 m within rows under plastic house conditions. One seedling was planted per pot. Irrigation was applied 500 ml in early growth stage of plants and 2000 ml in later growth stage of plants on alternate days, and weeding, pests and diseases control were done throughout the season as needed. Bagging the fruits was

also practiced to protect the damage of fruit fly. Fertilizer application rates (70 g of N, P and 140 g of K) were carried out throughout the growing season.

Plate 5 (A) Germinated muskmelon seeds, (B) Seedlings of muskmelon

3.1.2.2 Vine training and care

Stems of main vines were trained into vertical growth. Side-shoots up to 9^{th} node were removed allowing the side shoots from between 10^{th} -16^{th} nodes to bear the fruits as in farmers' practice; two fruit were retained on each plant for fruit characteristic measurements. Terminal shoots of main vines were cut off when there were 25 leaves on the main vine. Basal older leaves on main vine were removed when they got blocked ventilation at the basal part of the vines. Hand pollination was carried out to ensure pollination of flowers as bees were not available in the experimental site.

3.1.2.3 GA_3 preparation and spray

During stage 1 of predetermined fruit growth, up 20 days after full bloom (DAFB), different concentrations (0, 10, 20, 30 and 40 ppm) GA_3 were prepared by diluting with distilled water using gibberellic acid (GA_3 content > 90%, Sigma – Aldrich Chemie Gmbh Co., Germany). In the morning between 7:00-9:00 am, GA_3 was sprayed with a hand sprayer until runoff shielding the leaves in the vicinity with plastic sheet. On the other hand, the fruits were packed with plastic bag while the leaves were being sprayed. For the control treatment, the fruits were treated with distilled water only.

Plate 6 Application of gibberellic acid, (A) fruit spray; (B) foliar spray

3.1.2.4 Cuticular membrane (CM) isolation

Discs of 2 cm diameter were excised from the fruit skin with metal borer at the equatorial part of the fruit. According to Holloway and Baker (1968), the explants were isolated in 17 % of zinc chloride ($ZnCl_2$): 100 ml of concentrated hydrochloric acid (HCL) (17g/100ml) solution from adhering tissue. Extraction of CM was carried out at room temperature (25-30ºC) for 12 days. Following complete isolation, CM were thoroughly rinsed with distilled water and air-dried at ambient temperature.

Plate 7 Cuticular membrane (CM) isolation from muskmelon fruit skin (A) 2cm discs excised from fruit rind; (B) zinc chloride ($ZnCl_2$ -17 g): hydrochloric acid (HCL – 100 ml) solution; (C) isolated cuticular membrane (CM)

3.1.3 Data collection

The following data were recorded:

Frut diameter (cm)

Fresh fruit weight (kg)

Flesh thickness (cm)

Cuticular Membrane (CM) thickness

Crack number cm^{-2} fruit skin and

Brix % (Total Soluble Solids)

Fruit diameter was calculated from the following formula:

$C = \pi D$ (Geographic formula; www. Wikipeida.org)

Where, C = circumference of fruit

π = 3.14, and

D = diameter

Fruit circumference was measured at the equatorial region of the fruit.

All fresh weight of the fruits was measured by gravimetric means using digital balance (A & D Co Ltd., max 2000 g, Japan). The flesh thickness was measured as the distance between fruit exocarp and cavity border by ruler. Total soluble solids were measured with handheld refractrometer (Atago Co. Ltd, Japan). Fruits were cut into two halves and the juice sample was taken from the equatorial region of each fruit. The TSS was expressed in Brix (%). Number of cracks (nets) was counted in the fruit skin and calculated into number per 1 cm^2. Weights of isolated cuticular membranes were measured by gravimetric means with analytical balance (A & D Co Ltd., Japan) to calculate CM thickness according to Petracek and Bukovac (1995).

CM thickness calculation was as follows:

$$\frac{CM\ mass\ (g/m^2)}{Specific\ Density\ (1210 kg/m^2)} = CM\ thickness\ (\mu m)$$

3.1.4 Statistical analysis

ANOVA was done using Statistical Analysis System (SAS) software package (version 9.1). Significance of means was determined with Tukey's studentized test for two-way analysis, where appropriate one-way mean comparison was carried out at $\alpha = 0.05$.

3.2 Experiment 2: Gibberellic acid (GA_3) Application and Scheduled Irrigation at Fruit Growth Stage 1 and 3

3.2.1 Experimental design

Experiment 2 was conducted in two-factor factorial Completely Randomize design with 25 plants.

Factor A: GA_3 application (Plus and minus)

Factor B: Irrigation [Optimum (OI) and Deficit (DI)]

GA_3 10 ppm spraying to fruits was considered based on the results of experiment1.

3.2.2 Experimental procedure

In this experiment, tested cultivar 'Dandy-449' was also used and there were total population of 200 plants. Other procedures of this experiment were the same as conducted in the experiment 1.

3.2.2.1 GA_3 preparation and spray

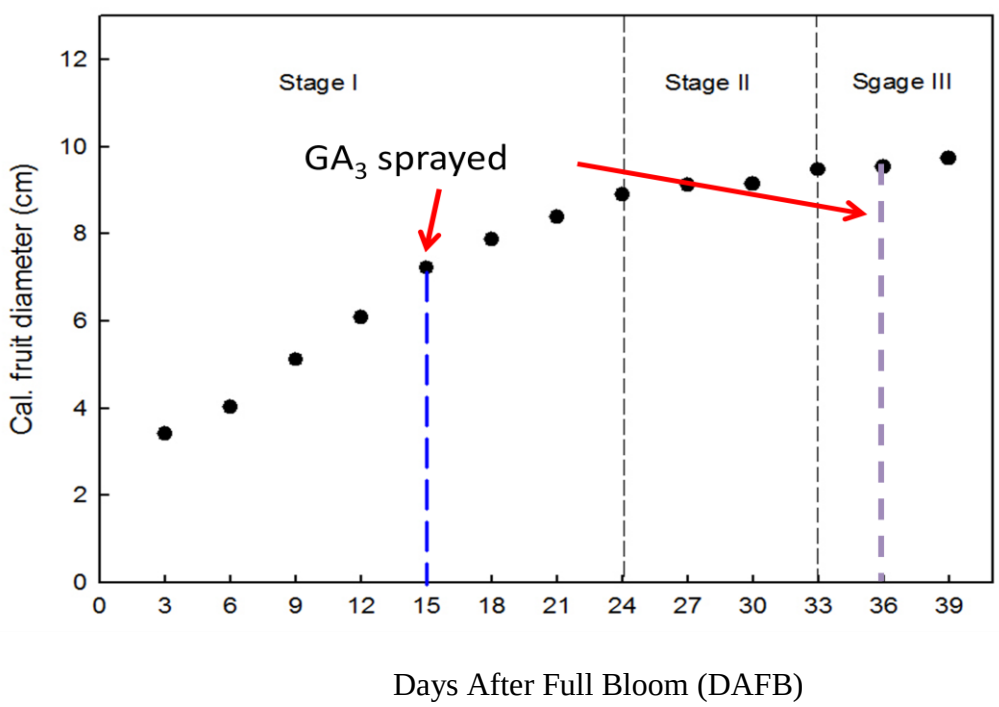

Figure 3 Fruit growth stages of 'Dandy-449', GA_3 applied at 15 DAFB (stage 1) and at 36 DAFB (stage 3)

The best concentration of 10 ppm gibberellic acid (GA_3 content > 90%, Sigma – Aldrich Chemie Gmbh Co., Germany) was applied to fruits at the stage 1 of fruit growth, at 15 days after full bloom (DAFB) and stage 3 of fruit growth, up to 36 DAFB as shown in above figure (according to the previous experiment).

3.2.2.2 Irrigation practices

Plants were irrigated by drip at one day interval throughout the growing season. The optimum irrigation was practiced as regular irrigation at one day intervals. Deficit irrigation was practiced for a few days until the plants showed leaf-drooping under no further irrigation. Immediately after deficit irrigation the plants were reirrigated.

Plate 8 Drip irrigation practice to muskmelon plants in experiment 2

3.2.3 Data collection

Although collection of harvest data of the experiment 2 was the same as in the experiment 1, the parameter of seed weight (seeds were separated from the other fruit components, sun dried and measured gravimetrically by digital balance) and fruit diameter as a fruit growth parameter at three-day intervals was added to record in experiment 2.

4. RESULTS AND DISCUSSION

4.1 Experiment 1 (2011-2012)

4.1.1 Fruit diameter

Effects of spray methods and different concentrations of GA_3 on fruit diameter are shown in Table 1. In spray methods, fruit spray resulted in larger fruit diameter (10.37 cm) than foliar spray (9.85 cm). GA_3 application with the concentrations of 10, 20, 30 and 40 ppm increased fruit diameter in contrast to no application (0 ppm). The more the GA_3 concentrations, the larger the fruit diameter is likely at harvest. Nevertheless, there was no significant difference in fruit diameter despite being increased in concentrations in this experiment. The 10 ppm of GA_3 was as good as 40 ppm in fruit diameter, 10.07 cm and 10.62 cm, respectively. No GA_3 application (0 ppm) indicated evidently reduced fruit diameter. There was no interactive effect of spray method and concentrations of GA_3 on fruit diameter of netted muskmelon, Dandy 449.

The increase in fruit diameter was recorded with GA_3 spraying (40 ppm). This could be attributed to the stimulatory effect of GA_3 on cell division and elongation (Davies, 1988; Taiz and Zeiger, 2006). Hence, the expansion rate of fruit mesocarp cells was greatly accelerated by the GA_3, consequently resulting in fruit diameter increment and fruit size as a whole.

GA_3 100 μM application at the time of anthesis attributed to increase fruit size of melon by 56 % of the control to the effect of increase cell division and elongation (Georgia et al., 2008). In apple, the application of 50 mgL^{-1} (50 ppm) GA_3 promoted increased fruit diameter as compared to control (Moneruzzaman et al., 2011) and the increased fruit diameter was also found with the application of GA_3 30 ppm at small bud and petal fall stage (Tuan and Ray, 2013).

4.1.2 Fruit weight

The data describe in Table 1 showed the effects of spray methods and GA_3 concentrations on fruit weight of Dandy 449. Among the GA_3 spray methods, the fruit weights of fruit spray and foliar spray were not significantly different from each other, 0.59 kg and 0.54 kg, respectively. Among the GA_3 concentration treatments the maximum fruit weight (kg) was recorded in GA_3 40 ppm (0.64 kg). The treatments

with GA_3 (30 ppm, 20 ppm and 10 ppm) with fruit weight of 0.58, 0.57, 0.56 kg, however, did not differ significantly from each other. Significantly lower fruit weight of 0.47 kg was recorded in control. There was no significant fruit weight by interaction between GA_3 concentration and spray methods.

The highest fruit weights were obtained from highest GA_3 concentration. The weight of the fruit is also determined by the number of cells and cell enlargement (Davies, 1988; Taiz and Zeiger, 2006). The effect of GA_3 on cell enlargement could enhance to increase in returns of fruit weight. The same has been reported by Deepthi, 2008, who concluded that the maximum fruit yield less weight was obtained from GA_3 (20 ppm) as fruit dipping followed by 60 ppm foliar application. Similarly, Tuan and Ruey (2013) reported that 30 ppm GA_3 spray to petal fall stage of apple had a much greater influence on fruit weight and the application of GA_3 at different concentrations in sweet cherry revealed that GA_3 (15ppm) produced highest fruit weight when compared to control (Yildirim and Koyuncu, 2010). And also, the application of GA_3 at 40 mg L^{-1} in sweet cherry increased fruit weight by 7% compared to untreated control fruit (Cline and Trought, 2007).

4.1.3 Flesh thickness

The effect of spray methods and different GA_3 concentrations on flesh thickness of fruits are described in Table 1. In spray methods of foliar and fruit spray, there was no significant difference of flesh thickness. Concentration of GA_3, 40 ppm, produced the highest flesh thickness (2.15 cm). This was followed by concentrations of 30 ppm, 10 ppm and 20 ppm in flesh thickness of 2.11 cm, 2.08 cm, and 2.00 cm, respectively in magnitude. This implied that flesh thickness of 40 ppm GA_3 treatment was not significantly different from 10, 20 and 30 ppm. It, however, evidently differed from no GA_3 application (0 ppm) which had the lowest magnitude of flesh thickness (1.91 cm). No GA_3 application (0 ppm) resulted in almost similar flesh thickness of 10, 20 and 30 ppm. As in fruit weight, the flesh thickness of 'Dandy-449' was not influenced by interactive effect of GA_3 concentrations and spray methods.

Hence, GA_3 spray considerable increased flesh thickness compared to the untreated control, according to the effect of increase cell number and elongation (Davies, 1988; Taiz and Zeiger, 2006). These results indicated that the application of GA_3 increased flesh thickness of melon fruits, that is, similarly, flesh thickness of

apple considerably increased by GA_3 30 ppm application at small bud and petal fall stage (Tuan and Ruey, 2013).

4.1.4 Crack number cm^{-2} fruit skin (Net)

Among the spray methods of foliar and fruit, there was also no significant difference in to crack number of the fruit skin (18.23 and 18.03, respectively) (Figure 4). In this study, spraying with GA_3 (10 ppm) resulted in the highest crack number on the fruit skin of (19.00) which was superior to all other treatments followed by GA_3 40 ppm (18.69), whereas the control produced the lowest crack number (17.28). The GA_3 30 ppm and 20 ppm applications resulted in crack number of 17.77 and 18.04 but did not differ significantly from each other. The crack (net) number per cm^2 fruit skin was not significantly difference between the applications by the interaction between the GA_3 concentration and spray methods.

The highest crack number was obtained even with the lowest GA_3 concentration of 10 ppm but the lowest crack number was found in control. In netted melon fruit, however, epidermal fracture of rind (netting) developed during early fruit development (Keren-Keiserman et al., 2004). Epidermal cells of *Cucumis* fruits have only a short period of cell division and expansion (Meissner, 1952). The combination of the characteristics of lower cell density and lower surface contact of the epidermal cell layer of the netted varieties might contribute to the weakness of epidermal layers thus increase its susceptibility to fracturing (Keren-Keiserman et al., 2004). Therefore, the number of cracks on fruit skin seemed to be increased by the effect of rapid cell division and expansion after gibberellic acid application in this study.

Table 1 Fruit diameter, fruit weight and flesh thickness of 'Dandy 449' as affected by different Gibberellic acid (GA_3) concentration and spray methods

Conc: (ppm)	Fruit Diameter (cm)			Fruit Weight (kg)			Flesh Thickness (cm)		
	Spray Method		Mean	Spray Method		Mean	Spray Method		Mean
	Foliar	Fruit		Foliar	Fruit		Foliar	Fruit	
0	8.99	9.71	9.35 B	0.413	0.527	0.47 B	1.93	1.89	1.91B
10	9.45	10.69	10.07 A	0.516	0.646	0.56 AB	2.04	2.12	2.08 AB
20	10.25	10.24	10.25 A	0.587	0.549	0.57 AB	2.03	1.97	2.00 AB
30	10.11	10.44	10.28 A	0.551	0.577	0.58 AB	2.03	2.19	2.11 AB
40	10.45	10.78	10.62 A	0.624	0.649	0.64 A	2.11	2.19	2.15 A
Mean	9.85 B	10.37 A		0.54 A	0.59 A		2.03 A	2.07 A	
Concentration (C)	**			**			*		
Method (M)	**			ns			ns		
C x M	ns			ns			ns		
CV%	7.41			23.80			11.86		

Means followed by the same lettter(s) in the same column show no significant difference at Tukey's Studentized range test, $\alpha = 0.05$; ** = highly significant, * = significant, ns= not significance

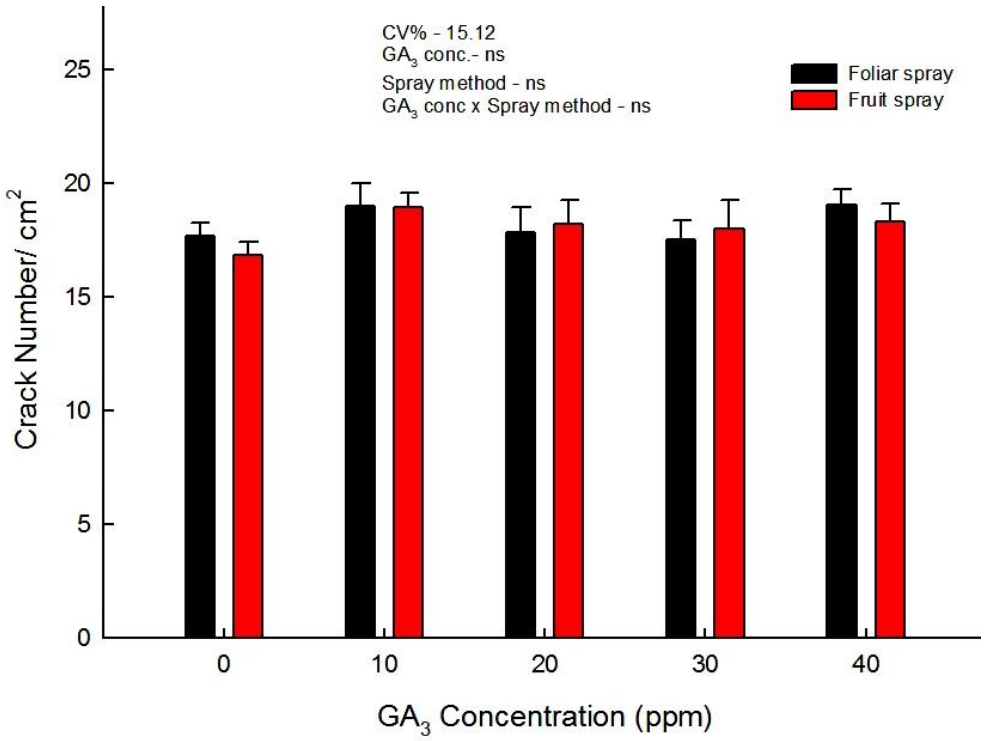

Figure 4 Crack number in fruit skin of 'Dandy 449' in response to different GA_3 concentrations interacting with spray methods; bars indicate mean ± SE, ns = not significant at Tukey 0.05 level.

4.1.5 Cuticular membrane (CM) thickness

Table 2 described the effect of spray methods and different GA_3 concentrations on cuticular membrane thickness of fruits. Foliar spray of GA_3 concentrations, 10, 20, 30 and 40 ppm increased the CM thickness of the fruit of 'Dandy-449' compared to 0 ppm. No remarkable difference of CM thickness resulted between 10, 20, 30 and 40 ppm concentrations of GA_3. In fruit spray, 10 ppm of GA_3 tremendously increased CM thickness followed by 20 and 30 ppm. The concentration of 40 ppm GA_3 could not increase more in CM thickness than 0 ppm. Cuticular membrane (CM) thickness of 'Dandy-449' indicated significant results by the interactions between the GA_3 concentrations and spray methods.

The application of gibberellins (GA_{4+7} & GA_3) increased CM mass per unit fruit surface area by promoting the main component of cuticle, cutin (Knoche and Peschel, 2007a). It was also reported that the GA_3 also stimulated elongation of deepwater rice internodes accompanied by a greatly accelerated rate of cutin biosynthesis (Hoffmann and Kende, 1994). The thickness of the epiderm and cuticular layer of the sweet cherry fruit and thus increased resistance against cracking was positively affected by the GA_3 applications (Cline and Trought, 2007). However, the mechanistic basis for the GA_3 effects has not been established.

Hormones are transported over short or long distances in the plant; for example, radiolabeled GA applied to fully expanded leaves moves down the stem and up to growing shoot apices. All hormones are translocated over short distances by diffusion and long distances by translocation through phloem to the targeted side (Lalit, 2002). When GA_3 applied to fruits, they directly diffused into the fruit and when applied to leaves, they translocated via the phloem. Therefore, in this study the highest CM thickness was obtained from the fruit spraying methods.

4.1.6 Brix % (Total soluble solids)

No significance of difference was found between foliar spray and fruit spray in Table 2. However, the significant results between GA_3 concentrations indicated that 10 ppm raised brix % of the fruit in contrast to 0 ppm. The bix % of 10 ppm GA_3 applied fruit was not statistically different from those of 20, 30 and 40 ppm although the magnitude of brix % of 10 ppm was likely to be higher than those of 20, 30 and 40 ppm. The later three concentrations did not show considerably difference from 0 ppm.

There was no interactive effect of GA_3 concentrations and spray methods in brix % of 'Dandy-449'.

Even if the low concentration of GA_3 enabled to increase the total soluble solids (TSS) content of the fruit, the increase in TSS could be attributed to the enhanced photosynthetic efficiency of the leaves and a possible increase in translocation of assimilates into the fruits in response to hormonal stimulation (Kaur, 2000). Moreover, GA_3 seemingly diverted more solids towards developing fruits by enhancing the source-sink balance (Kher et al., 2005).

The TSS content of the fruits increased with an increase in the concentration of GA_3. The highest TSS content was recorded in GA_3 (60 ppm) foliar application followed by GA_3 (20 ppm) as fruit dipping in melon (Deepthi, 2008). Moneruzzaman et al. (2011) also reported that the highest TSS was observed in 50 mgL^{-1}(50 ppm) GA_3 treated fruits of apple. The similar result of increased total soluble solids content were also found with the spraying of 30 ppm GA_3 spraying to apples (Tuan and Ruey, 2013).

Table 2 Cuticular membrane thickness and Brix % of 'Dandy 449' as affected by different Gibberellic acid (GA_3) concentration and spray methods

Conc: (ppm)	CM Thickness (μm)			Brix (%)		
	Spray Method		Mean	Spray Method		Mean
	Foliar	Fruit		Foliar	Fruit	
0	60.15 b	67.03 c	63.59 C	11.55	12.08	11.82B
10	75.92 a	109.39a	89.77 A	13.18	12.98	13.08 A
20	77.21 a	82.24 b	79.00 B	12.23	12.17	12.20 AB
30	79.52a	80.37 b	79.94 B	12.38	12.67	12.53 AB
40	78.90 a	57.19 c	66.69 C	13.03	12.37	12.70 AB
Mean	75.19 A	77.41A		12.48 A	12.45 A	
Concentrations (C)	**			**		
Methods (M)	ns			ns		
C x M	**			ns		
CV%	14.82			8.53		

Means followed by the same letter(s) in the same column show no significant difference at Tukey's Studentized range test, $\alpha = 0.05$; ** = highly significant, * = significant, ns= not significant, C = GA_3 concentration, M = spray method

4.2 Experiment 2 (2013-2014)

In this experiment, fruit characteristics of ‘Dandy 449’ were observed under gibberellic acid (+ and -) application and scheduled irrigation (optimum and deficit) at the fruit growth stage 1 and 3.

4.2.1 Fruit growth

Measurement on fruit growth of ‘Dandy-449’ was commenced at 7 days after full bloom. After full bloom, the fruit growth exhibited sigmoidal pattern under all treatments (Figure 5). The growth of the individual fruits started slowly at the beginning then speeded up in size increment until about 21 DAFB (stage 1). Afterword, the fruit growth stunted for about 7 to 10 days (20 to 31 DAFB, stage 2). Following stage 2, final swelling of fruit growth continued until harvest (stage 3). Fruit growth pattern of ‘Dandy-449’ was in line with the growth of Fuyu A and Natsu 4 muskmelon (Katsumi et al., 1999).

Netting began at the stylar end of the fruit around 14 DAFB and continued to the pedicle end. It took about 21 days (35 DAFB) to complete netting on the whole fruit skin. The netting in fruits showed the similar conditions in all treatments. Fruit diameter among pattern of fruit growth of ‘Dandy-449’ did not differ significantly among the treatments.

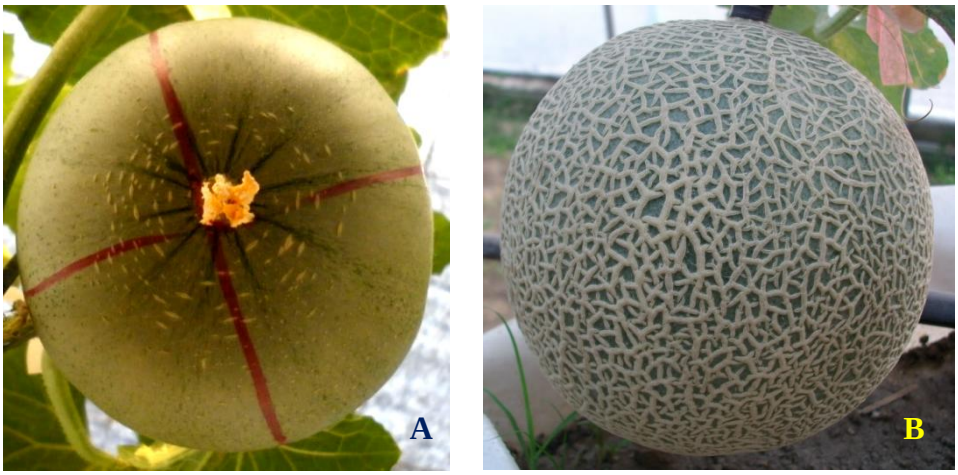

Plate 9 Net formation of ‘Dandy-449’, (A) Net beginning at the stylar end of the fruit at 14 DAFB (B) Full coverage of net on fruit skin at 35 DAFB (DAFB = Days After Full Bloom)

The common growth pattern of fruits of angiosperms in higher plants can be divided into three distinct phases that is a major issue in plant physiology (Gillaspy et al., 1993). The earliest phase (stage 1) is the period between flower development and rapid cell division of fruit. Fruit growth (stage 2) slow somewhat after rapid cell division in stage 1, this growth stage reflects endocarp or seed size increment in the ovary in most fruits species then the fruit growth resume as a final swelling until the time of harvest (biological maturity, stage 3). This growth of fruit refer to us sigmoid growth pattern. Current observation of fruit growth on ‘Dandy-449’, therefore, represented the above mentioned growth pattern.

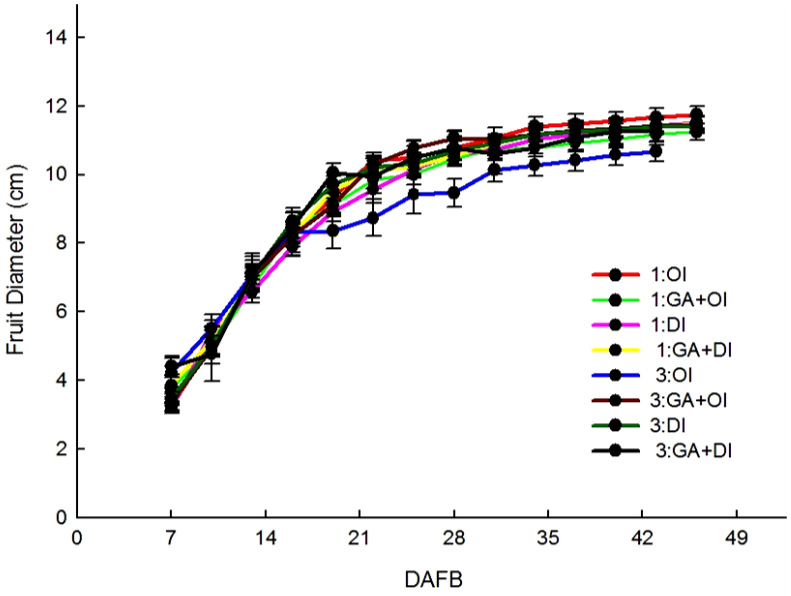

Figure 5 Time-course increment of fruit diameter (fruit growth) of netted muskmelon, 'Dandy-449' under GA_3 application and scheduled irrigation. Fruit diameter was measured at the equatorial portion of the fruit during fruit growth; the values at each are means of ± SE. (DAFB: days after full bloom; OI= optimum irrigation; DI= deficit irrigation; GA= gibberellic acid (GA_3))

4.2.2 Fruit diameter, fruit weight and flesh thickness

Fruit diameter of 'Dandy-449' showed no significant result between scheduled irrigation and GA_3 application at fruit growth stage 1 and 3 in Table 3. At stage 1, fruit diameter was not different between GA_3 application and no GA_3 application and between optimum irrigation and deficit irrigation. However, fruit diameter remarkably increased by the GA_3 application (12.30 cm) compared to no GA_3 application (11.39 cm) at stage 3. No difference in fruit diameter indicated between optimum irrigation and deficit irrigation at stage 1 and 3. There are no interactions between the treatments at fruit growth stage 1 and 3, respectively.

Table 3 also showed the effect of scheduled irrigation and GA_3 application on fruit weight at stage 1 and 3. At stage 1, the data showed no significant effect between GA_3 application (0.91 kg) and no GA_3 application (0.91 kg); between optimum irrigation (0.92 kg) and deficit irrigation (0.90 kg). At stage 3, considerable increased fruit weight of 0.94 kg was resulted under the effect of GA_3 application compared to no GA_3 application (0.83 kg). There was no significant in fruit weight by the effect of optimum irrigation and deficit irrigation. As in fruit diameter, fruit weight had no

significant increase by the interactive effect between GA_3 application and scheduled irrigation at fruit growth stage 1 and 3.

Flesh thickness indicated no significant results between GA_3 application (3.43 cm) and no GA_3 application (3.43 cm); between optimum irrigation (3.45 cm) and deficit irrigation (3.41 cm) at stage 1, respectively (Figure 6). Similarly, flesh thickness showed no significant difference between optimum irrigation (3.22 cm) and deficit irrigation (3.31 cm); between GA_3 application (3.25 cm) and no GA_3 application (3.29 cm) at stage 3. There was no interactive effect of GA_3 application and scheduled irrigation on flesh thickness of 'Dandy-449' in stage 1 and stage 3.

In this case, GA_3 application provided and evidenced of fruit diameter and fruit weight of 'Dandy-449' at stage 3, irrespective of optimum or deficit irrigation. As in experiment 1, this finding confirmed fruit diameter and fruit weight increment by GA_3 application to fruit as shown in experiment 1. Therefore, GA_3 supposedly attributed to cell enlargement of muskmelon fruit, consequently the whole fruit size increased. On the other hand, increased fruit diameter accordingly contributed to the whole fruit weight. GA_3 might contribute the effect to other fruit tissue the flesh (mesocarp). GA_3 application and also scheduled irrigation had no effect on the flesh thickness.

Table 3 Effects of GA_3 application and Scheduled irrigation on fruit diameter and weight in fruit growth stage 1 and stage 3 of 'Dandy 449'

Stage	GA_3	Fruit diameter (cm)			Fruit weight (kg)		
		Irrigation		Mean	Irrigation		Mean
		OI	DI		OI	DI	
1	+	11.97	11.99	11.98A	0.91	0.91	0.91A
	-	12.22	11.84	12.03A	0.93	0.90	0.91A
	Mean	12.09A	11.91A		0.92A	0.90A	
GA_3		ns			ns		
Irrigation		ns			ns		
GA_3 x Irrigation		ns			ns		
CV %		3.97			9.08		
3	+	12.22	12.37	12.30A	0.95	0.93	0.94A
	-	11.32	11.45	11.39B	0.80	0.86	0.83B
	Mean	11.77A	11.91A		0.88A	0.90A	
GA_3		**			**		
Irrigation		ns			ns		
GA_3 x Irrigation		ns			ns		
CV %		7.92			9.62		

Means followed by the same letter(s) in the same column show no significant difference at Tukey's Studentized range test, $\alpha = 0.05$; ** = highly significant, * = significant, ns= not significant, OI= optimum irrigation, DI= deficit irrigation.

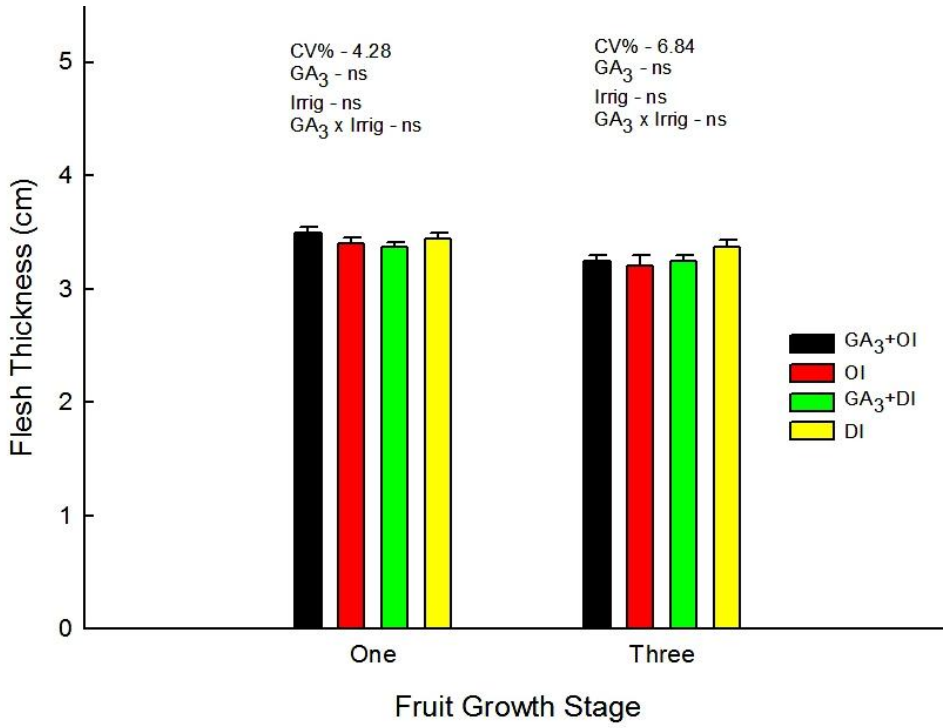

Figure 6 Flesh thickness of 'Dandy 449' under GA_3 application and scheduled irrigation at fruit growth stage 1 and stage 3; No significant result was indicated by GA_3 application, Scheduled Irrigation and GA_3 x Irrigation at α=0.05; OI= optimum irrigation, DI = deficit irrigation, ns = not significant

4.2.3 Crack number cm^{-2} in fruit skin (Net)

Crack number (net) in the fruit skin of 'Dandy-449' showed by the effect of GA_3 application and scheduled irrigation at stage 1and 3 in Table 4. At stage 1, crack number on the fruit skin increased (18.67 cm^{-2}) by GA_3 application under optimum irrigation compared to (17.32 cm^{-2}) no GA_3 application under optimum irrigation. GA_3 application also raise crack number on the fruit skin under deficit irrigation (17.40 cm^{-2}), whereas it was (16.24 cm^{-2}) by no GA_3 application under deficit irrigation. Significant result by interaction between the GA_3 application and scheduled irrigation was observed.

At stage 3, GA_3 application did not give significant difference in crack number in contrast to no GA_3 application. Nevertheless, optimum irrigation increased crack number (17.41cm^{-2}) in fruit skin, while deficit irrigation did (16.40 cm^{-2}). The interaction between GA_3 application and scheduled irrigation resulted in no considerable effect on crack number in the fruit skin of 'Dandy-449'.

At stage 1, GA_3 application enhanced crack number cm^{-2} in the fruit skin of 'Dandy 449' both under optimum irrigation and deficit irrigation. During stage 1, fruit growing in sigmoidal pattern possesses active cell division and expansion (Gillapsy et al., 1993), which contributes to rapid fruit size increment. Meanwhile, GA_3 application to fruits promoted seemingly cell enlargement which equivocally expanded fruit volume. In addition, water intake into the fruit supposedly provided surplus force to expand the volume. Thus, the cuticle in the fruit skin suffered from strain (deformation) and resulted in microcracks leading to macrocracks (visible cracks) eventually (Knoche and Peschel, 2006). These conditions revealed why crack number in fruit skin of 'Dandy 449' increased.

At stage 3, crack number cm^{-2} considerably increased under the effect of optimum irrigation. The netted muskmelon variety is coated with a well developed cuticular layer during the expansion phase, which might expose its cuticle to higher tensile strains (Keren-Keiserman et al., 2004). The epidermal fracture of rind (netting) resulted from increased turgor caused by ample amount of water uptake by the fruit (Knoche and Peschel, 2006). Moreover, cell expansion continuously caused epidermal fracture not only at the early fruit growth but at the later stage (Sekse, 1995a,b). The

number of cracks on fruit skin increased by rapid fruit growth rate supposedly relied on epidermal cracking and thus enhanced fruit netting (Combrink et al., 2001).

4.2.4 Cuticular Membrane (CM) thickness

The data describe in Table 4 showed by the effect of GA_3 application and scheduled irrigation on cuticular membrane thickness of fruits at stage 1 and 3. At stage 1, cuticular membrane of 'Dandy-449' increased under the GA_3 application of (52.86 μm) in contrast with no GA_3 application (40.68 μm). Considerable increament in CM thickness occurred under deficit irrigation (50. 29 μm) which was higher than CM thickness under optimum irrigation (42.23 μm). CM thickness accounted for no significant result by the interactive effect between GA_3 application and scheduled irrigation.

At stage 3, CM thickness was evidently higher (67.65 μm) by GA_3 application under optimum irrigation than CM thickness 43.88 μm of no GA_3 application under optimum irrigation. CM thickness significantly increased (71.20 μm) by GA_3 application under deficit irrigation, in comparison with CM thickness (62.90 μm) by no GA_3 application under deficit irrigation. There was interaction between GA_3 application and scheduled irrigation on CM thickness of 'Dandy 449'.

GA_3 application had positive effect on cuticular membrane (CM) by promoting its thickness both under optimum and deficit irrigations. This phenomenon particularly occurred at stage 3. GA_3 was considered to appeal deposition of cutin, the main component of cuticle in the epidermis. Cutin is synthesized by epidermal cells (Yeats and Rose, 2013). GA_3 application would enhance elongation of epidermal cells which in turn likely synthesized ample amount of cutin to be transported to the epidermis. GA_3 treated rice indicated thickened cuticle in the internode (Hoffmann and Kende, 1994). Similar result showed that GA_3 considerably increased CM deposition in tomato (Knoche and Peschel, 2007b).

Thickening of CM of 'Dandy 449' under deficit irrigation accounted for that water deficit promoted barrier properties of cuticle. Water deficit increased of cutin biosynthesis and wax deposition on the leaf surface of *Arabidopsis* (Yeats and Rose, 2013). Plant parts usually make resistant to water loss by enhancing cutin deposition when epidermis gets less water. In peaches, the deficit irrigation induced thicker cuticle (Crisosto et al., 1994).

Table 4 Effects of GA_3 application and Scheduled irrigation on CM thickness and crack no per cm square in fruit growth stage 1 and stage 3 of 'Dandy 449'

Stage	GA_3	CM thickness (µm)			Crack no cm^{-2}		
		Irrigation		Mean	Irrigation		Mean
		OI	DI		OI	DI	
1	+	49.98	55.40	52.86 A	18.67a	17.40a	17.91A
	-	37.74	45.19	40.68 B	17.32b	16.24b	16.89B
	Mean	42.23 B	50.29 A		17.89A	16.95B	
GA_3		**			**		
Irrigation		**			**		
GA_3 x Irrigation		ns			*		
CV %		9.31			6.72		
3	+	67.65a	71.20a	69.49 A	17.63	16.81	17.18A
	-	43.88b	62.90b	50.22 B	17.17	16.10	16.48A
	Mean	53.13 B	67.69 A		17.41A	16.40B	
GA_3		**			ns		
Irrigation		**			*		
GA_3 x Irrigation		**			ns		
CV %		9.47			8.43		

Means followed by the same letter(s) in the same column show no significant difference at Tukey's Studentized range test, α = 0.05; ** = highly significant, * = significant, ns= not significant, OI= optimum irrigation, DI= deficit irrigation.

4.2.5 Seed weight

The data in Table 5 showed the effect of GA_3 application and scheduled irrigation on seed weight of fruit at stage 1 and 3. In particular, seed weight of fruit under GA_3 application was not different from that under no GA_3 application at stage 1. Seed weight of fruit under optimum irrigation was significantly higher (13.37 g) than under deficit irrigation (11.38 g). Seed weight of 'Dandy-449' had no evidence by the interaction between GA_3 application and scheduled irrigation.

At stage 3, interaction between GA_3 application and scheduled irrigation also gave no remarkable effect on seed weight. GA_3 application notably increased seed weight (11.95 g), while no GA_3 application obtained (10.56 g), while optimum irrigation resulted in seed weight indifference from that by deficit irrigation.

At stage 1 seed weight increased under optimum irrigation accounted for the fact that rapidly dividing and growing cells of the fruit demanded for water as the fruit contained osmotically active substances (Matthwes and Shackel, 2005). During early fruit growth, the young seeds inside the fruit are heterotrophic organs which depend on nutrients imported from the plant for their development (Zhang et al., 2007). Plant nutrients are also osmotically-active substances accompanied by water through unloading into the seeds (Ruan et al., 2012; Zhang et al., 2007). Imported nutrients into the seeds determine seed number and final seed size (Zhang et al., 2007), thereby increasing seed weight of 'Dandy 449' under optimum irrigation.

At stage 3, GA_3 application produced larger seed weight of 'Dandy449' than no GA_3 application. It is likely due to the facts that 1) gibberellins are transferred to ovarian tissue and developing seeds from the pollen tube (Jong et al., 2009), and 2) exogenous application of GA_3 could enhance the GA concentration in young seeds. As a consequence, cell expansion of developing seeds equivocally might increase seed size, and then raise final seed weight of 'Dandy 449'. It agreed with seed weight increment by GA_3 application in sweet cherry fruits (Yildirim and Koyuncu, 2010).

4.2.6 Brix % (Total soluble solids)

Table 5 showed the effect of GA_3 application and scheduled irrigation on brix % of fruit at stage 1 and 3. At stage 1, brix % (Total soluble solids) of 'Dandy 449' had no effect by interaction between GA_3 application and scheduled irrigation. On the other hand, and also brix of fruit under GA_3 application was indifferent from that

under no GA_3 application; brix under optimum irrigation indifferent from that under deficit irrigation.

At stage 3, GA_3 application interacting with scheduled irrigation provided considerable effect on brix of fruit of 'Dandy449'. Brix of fruit by GA_3 application under optimum irrigation resulted in similar magnitude (11.23 %) to that by no GA_3 application under optimum irrigation (11.09 %). However, brix of fruit by GA_3 application under deficit irrigation (10.87 %) was inferior to that by GA_3 application under deficit irrigation (13.34 %). It accounted for higher brix% in no GA_3 application under deficit irrigation.

Increased brix of fruit without GA_3 application under deficit irrigation at stage 3 indicated vital role of water in total soluble solids level of fruit. However, it deviated from the fact that enhanced sink strength for photoassimilation by larger cell size of fruit by GA_3 (Iqbal et al., 2011). Despite lower brix% under GA_3 application and deficit irrigation at stage 1, significantly high brix% was obtained at stage 3. Therefore, in muskmelon 'Dandy 449', brix % of fruit was likely affected by irrigation system.

During final stage of fruit growth (stage 3), sweetness of fruit determined by the quantity of photoassimilates (hexoses and sucroses) (Muller et al., 2011) referred to as total soluble solids (brix) relied on water intake to the fruit (Matthews and Shackel, 2005). For being osmotically active solutes, increasing photoassimilates draw more water from the vascular flow of the plant (Taiz and Zieger, 2006). In other words, regular influx of water into the fruit would downgrade the sweetness of the fruit, diluting total soluble solids. Under deficit irrigation, on the other way round, total soluble solids concentration of fruit rises with reduced water content (Muller et al., 2011). This implied increased brix % of 'Dandy 449' under deficit irrigation with respect to no GA_3 application.

Table 5 Effects of GA_3 application and Scheduled irrigation on seed weight and brix % (Total soluble solids) in fruit growth stage 1 and stage 3 of 'Dandy 449'

Stage	GA_3	Seed weight (g)			Brix (%)		
		Irrigation		Mean	Irrigation		Mean
		OI	DI		OI	DI	
1	+	13.00	11.50	12.14 A	12.25	13.07	12.66 A
	-	13.70	11.25	12.36 A	12.05	12.41	12.22 A
	Mean	13.37 A	11.38 B		12.15 A	12.74 A	
GA_3		ns			ns		
Irrigation		**			ns		
GA_3 x Irrigation		ns			ns		
CV %		15.53			10.97		
3	+	11.36	12.60	11.95 A	11.23a	10.87b	11.05 B
	-	10.13	10.76	10.56 B	11.09a	13.34a	12.22 A
	Mean	10.84 A	11.44 A		11.16 B	12.11 A	
GA_3		*			*		
Irrigation		ns			**		
GA_3 x Irrigation		ns			**		
CV %		9.47			10.80		

Means followed by the same letter(s) in the same column show no significant difference at Tukey's Studentized range test, $\alpha = 0.05$; ** = highly significant, * = significant, ns = not significant, OI = optimum irrigation, DI = deficit irrigation.

5. CONCLUSION

Since muskmelon is a leading fruit in Myanmar fruit industry, its quality determines vulnerability to market price at local as well as oversea destination. Dozens of fruits turn to garbage for downgraded quality like unmarketable size and inferior appearance. In line with such problem, current research was emphasized on fruit characteristics of netted muskmelon by GA_3 application and scheduled irrigation.

In experiment 1, GA_3 concentrations of 0, 10, 20, 30, and 40 ppm were applied to leaf and fruit of muskmelon, Dandy 449'. GA_3 application with a minimum concentration of 10 ppm gave larger fruit diameters as good as with maximum concentration, 40 ppm, compared with no GA_3 application, 0 ppm. No significant interaction effect was observed between GA_3 concentration and spray method. Fruit weight and flesh thickness showed remarkable increment by GA_3 40 ppm which is superior to 0 ppm, while those two characteristics of fruit by 10 ppm of GA_3 did not diverge from those of 40 ppm. Crack number (netting) per unit area in fruit skin was effected neither by GA_3 concentration nor spray method and by both. Nevertheless, GA_3 10 ppm sprayed to fruit tremendously increased CM thickness compared to others. That concentration of GA_3 also promoted brix % (total soluble solids) of the fruit as in 20, 30 and 40 ppm. Based on the results, 10 ppm of GA_3 deserved comparatively to be considered an appropriate concentration if it was applied to fruit of 'Dandy 449'.

In experiment 2, 10 ppm GA_3 was applied to fruit under optimum irrigation and deficit irrigation at two fruit growth stages, one and three. GA_3 application enhanced fruit diameter and fruit weight, respectively, regardless of scheduled irrigation at stage 3. However, flesh thickness of the fruit did not show any effect of prescribed treatments at both of fruit growth stages. GA_3 application promoted CM thickness as in experiment 1, but deficit irrigation thickened the CM so as to prevent fruit water loss under limited water supply at stage 1. The interactive effect of prescribed treatments pointed out GA_3 application increased CM thickness both under optimum irrigation and deficit irrigation at stage 3. Increment in crack number (netting) per unit area in the fruit skin was accelerated by GA_3 application with respect to scheduled irrigation at stage 1. Independent increment of crack number per unit area in the fruit skin by optimum irrigation revealed that GA_3 application did not

affect netting at stage 3. Again, optimum irrigation raised seed weight of ‘Dandy 449’ at stage 1 independent of GA_3 application. At the opposite end, GA_3 application increased seed weight irrespective of scheduled irrigation at stage 3. No GA_3 application interactive with deficit irrigation promoted brix % (total soluble solids) of ‘Dandy 449’ at stage 3.

The results of present study highlighted that

- GA_3 application (10 ppm) provided comparative superiority in most of fruit characteristics of netted muskmelon, ‘Dandy 449’including of crack number (netting) with enormously thickened fruit cuticle in experiment 1
- GA_3 (10 ppm) application increased fruit diameter, fruit weight and seed weight without regards of scheduled irrigation, but increased the cuticle thickness and brix % of the fruit under deficit irrigation at fruit growth stage 3 and
- GA_3 application independently thickened the fruit cuticle, whereas deficit irrigation did alone at stage 1. Nevertheless, combined action of GA_3 application and scheduled irrigation enhanced netting (crack number per cm^{-2}) in fruit skin. Only optimum irrigation promoted seed weight of ‘Dandy 449’ when it was practiced at fruit growth stage 1.

It is suggested that the result of this experiment just stands as a door for further investigation on optimum water requirement of netted muskmelon for optimum fruit growth. In addition, it is wise to find out impacts of erratic irrigation during rapid fruit growth under high moisture condition.

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