# EVALUATION OF THE EFFECT OF PLANT GROWTH RETARDANTS ON VEGETATIVE GROWTH, YIELD COMPONENTS, SEED QUALITY AND CROP MATURITY OF THE KABULI CHICKPEA CULTIVAR CDC FRONTIER

A Thesis Submitted to the College of
Graduate Studies and Research
In Partial Fulfillment of the Requirements
For the Degree of Masters of Science
In the Department of Plant Sciences
University of Saskatchewan
Saskatoon, Saskatchewan

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#### **Abstract**

Chickpea production in the short growing season of the Canadian Prairies is still a challenging task due to excessive and continuous vegetative growth which often results in severe yield and quality reduction. This study examined the effects of three plant growth retardants (PGR), Chlormequat Chloride (CCC), Prohexadione Calcium and Trinexapac Ethyl applied during flowering stage on vegetative growth, seed quality, yield and crop maturity of the Kabuli chickpea cultivar CDC Frontier. Field experiments were conducted at Brooks and Bow Island in southern Alberta in the 2010 and 2011 growing seasons. Four concentrations of each PGR were applied at 10, 20 and 30 days after flowering (DAF) stages.

During the 2010 growing season the crop experienced above average moist and cooler temperature conditions. In contrast, later half of the 2011 growing season was above average dry and hot. None of the three PGR tested in this study had a significant effect on plant height at 30 days after treatments or on above ground biomass plant<sup>-1</sup> at harvest. Application of PGR had no significant effects on the number of seeds m<sup>-2</sup>, except at the Brooks rain-fed site in 2011 where the PGR treatment applied at 10 and 20 DAF increased the number of seeds m<sup>-2</sup> at harvest. An increase of 1000-seed weight of marketable seeds was obtained with Prohexadione Calcium and Trinexapac Ethyl applications at Bow Island, but the effects were not consistent across sites and years. Results suggested that the effect of PGR on 1000-seed weight of marketable seeds mainly depended upon the growing environment and the type of PGR. In general, PGR applications reduced the total and marketable seed yields. Application of Prohexadione Calcium and Trinexapac Ethyl at the Bow Island site delayed crop maturity in 2011. In contrast, the application of CCC at 6000 mg L<sup>-1</sup> at 20 DAF accelerated crop maturity at the Brooks irrigated site in 2011. In addition to this main study, the potential effects of Pyraclostrobin and Prothioconazole fungicides on the activities of the three PGR were compared by a separate experiment conducted at the Brooks irrigated site in 2011. The results of that study revealed that there were no significant differences in the effects of PGR on chickpea vegetative growth, seed yield parameters and maturity when they were applied as a mixture with either Pyraclostrobin or Prothioconazole fungicide.

In summary, results revealed that PGR applied during flowering stage were not effective on controlling vegetative growth of chickpea and did not improve seed yield and crop maturity. Their effects on yield-related traits were highly inconsistent. Thus, it can be concluded that the application of PGR is not a reliable agronomic option to handle the production issues associated with continues vegetative growth at the late reproductive stage of the chickpea cultivar CDC Frontier under the western Canadian growing conditions.

#### **ACKNOWLEDGEMENT**

I express my sincere gratitude to my co-supervisors, Dr. Manjula Bandara and Dr. Bunyamin Taran for their guidance, encouragement and assistance during this study. I appreciate their trust on my capability to conduct this research.

I would also like to extend my appreciation to my committee members, Dr. Bruce Coulman (Chair), Dr. Rosalind A. Bueckert and Dr. Sabine Banniza for their suggestions and constructive criticism. I was truly privileged to have the opportunity to work with them. I would like to thank Mr. William May for serving as the external examiner and also for his valuable suggestions.

I acknowledge the excellent technical assistance provided by Art Kruger, Kathie Davidson, Stephanie Mather, Don Elmer, Kara MacAulay and Eldon Graham at Crop Diversification Center South at Brooks, Alberta during the field studies. I also wish to acknowledge Brent Barlow and field staff of Crop Development Center, Saskatoon for the assistance provided during sample processing.

I gratefully acknowledge Canadian Agricultural Adaptation Program, Alberta Pulse Growers Commission and Saskatchewan Pulse growers for financial support for this project and Alberta Agriculture and Rural Development for providing all facilities for this study.

I would like to thank graduate students of the Department for being my wonderful friends. I am grateful to my family for their understanding and support during my study.

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#### **List of Abbreviations**

a.i. Active ingredient

ABA Abscisic acid

ACC 1-aminocyclopropane—carboxylic acid

CCC Chlormequat Chloride

CHU Crop heat units

DAF Days after flowering

DAS Days after seeding

DAT Days after treatments

DF Degree of freedom

GA Gibberellic acid

IAA Indole-3-acetic acid

LSD Fisher's least significance difference

LTA Long term average

N:P:K Nitrogen: phosphorous: potassium

NO<sub>3</sub>-N available nitrate-nitrogen

PGR Plant growth retardants

TIBA Tri-iodobenzoic acid

#### 1. Introduction

Chickpea (*Cicer arietinum* L.) is the world's second largest food legume crop in terms of total production (10.9 million tonnes), which is only next to dry beans (FAO STAT, 2012). In 2010, about 55 countries produced chickpeas representing almost all geographical regions of the world (FAO STAT, 2012).

Chickpea is an important source of protein, carbohydrates, dietary fibre and essential micronutrients for human diets (De Falco et al., 2010). It is an important high quality protein and carbohydrate source for a large population in developing countries (Kumar and Abbo, 2001). Therefore, an increase in chickpea production is vital to feed rapidly increasing world population. The inclusion of chickpea in crop rotations offers economic and agronomic benefits to the producers, who traditionally practice cereal-based cropping systems on the Canadian Prairies. Chickpea fixes atmospheric nitrogen and provides most of its own nitrogen requirements, thus reduces fertilizer requirements (Doughton et al., 1993). Pulse crops such as chickpea increase soil nitrogen fertility, add high quality organic matter to the soil, break disease cycles of certain cereal crops and increase water and nutrient use efficiency (Siddique et al., 2012).

Despite the many advantages of growing chickpea, its production on the Canadian Prairies faces unique challenges. In Canada, chickpea production is mainly confined to the south-western region of Saskatchewan and the south-eastern region of Alberta. The average frost free days in these areas (92 to 115 days) are critically close to the minimum chickpea crop duration. As a crop with an indeterminate growth habit, chickpea continues vegetative growth during the generative stage instead of exclusively setting pods under favourable conditions (Gan et al., 2008). The end of the western Canadian growing season is characterized by declining temperatures and often wet conditions which are highly conducive for vegetative growth. These conditions contribute to the extended crop growth, thus delay the crop maturity. Prolonged crop growth increases the risk of frost damage to the immature pods and seeds at the end of the growing season resulting in significant yield losses or seed quality reductions (Anbessa et al., 2007a). The late and uneven crop maturation is a major impediment for high quality chickpea production on the Canadian Prairies.

Developing early maturing cultivars with determinate growth habit would be the long term solution for maturity-related issues in chickpea. Development of agronomic practices in parallel with breeding efforts can play an important role since these practices could be flexible with changing conditions and location specific requirements. Effects of the seed bed conditions (Gan et al., 2009b), nitrogen fertilizer management (Zakeri et al., 2012), application of desiccants (Choudhry, 2012) and different harvesting techniques (Gan et al., 2008) have been evaluated under western Canadian conditions to control the excessive vegetative growth and to influence the maturity of indeterminate pulse crops including chickpea. However, only limited success has been achieved.

Plant growth retardants (PGR) are substances which are used in agriculture primarily to control plant growth. In most cases PGR inhibit the biosynthesis of active gibberellins which are an essential group of plant hormones for the growth process (Rademacher, 2000; Hedden et al., 2010). PGR application can reduce plant height and alter the partition of assimilates towards the grain yield of wheat (Shekoofa and Emam, 2008). In case of cotton, PGR such as Mepiquat Chloride minimize excessive growth, accelerate and synchronize ball maturity (Rademacher, 1991). Yadev and Bharud (2009) indicated that plant growth retardant, Cycocel applied at flower initiation stage, improved chickpea yield components which resulted in a 16% increase of seed yield. Application of PGR caused an increase of seed yield and pod diameter of an early sawn chickpea crop (Brar et al., 1992). This indicates that PGR may have the potential to control vegetative growth of chickpea during reproductive stage and to increase the partition of assimilates toward pod development which would be critical under the growing conditions on the Canadian Prairies. However, information on this aspect is lacking. This study was designed to evaluate the effects of PGR on growth, crop maturity and seed yield of chickpea under western Canadian growing conditions.

Three commercially available PGR, namely Prohexadione Calcium, Chlormequat Chloride (CCC) and Trinexapac Ethyl were used for the study. These PGR are currently used in Europe, the United States and Canada to control lodging in cereal grain crops (Rajala and Peltonen-Sainio, 2002), to control vegetative growth of orchards (Rademacher et al., 2006), to increase the row visibility of peanuts (Beam et al., 2002), to increase the performances of ornamental plants (Leclerc et al., 2006; Sahi, 2009) and to control growth of turf grasses (Lickfeldt et al., 2001;

McCullough et al., 2006). These three PGR can be generally categorized as Gibberellins Biosynthesis Inhibitors based on their mode of action (Rademacher, 2000).

PGR application during the reproductive stage is expected to minimize the vegetative growth of chickpea by reducing the levels of active gibberellins in plants. As a species with a strong indeterminate growth habit, vegetative and reproductive organs compete for assimilates at the reproductive stage in chickpea (Chopra and Sinha, 1987). Therefore, regulating vegetative growth is expected to have beneficial effects on the seed yield.

This study was conducted to test the following hypotheses:

- PGR will control excessive vegetative growth (plant height and above ground biomass)
   of chickpea when applied at the reproductive stage.
- Controlled vegetative growth at the reproductive stage will be beneficial for the seed development due to enhanced of assimilates partition for reproductive organs.
- PGR will synchronize seed maturity and reduce the proportion of green and immature seeds at harvest.

The main objectives of this study were:

- 1. To evaluate the effects of PGR applied at different growth stages on the growth of the Kabuli chickpea cultivar CDC Frontier during the reproductive stage.
- 2. To evaluate the effects of PGR on seed yield and quality of Kabuli chickpea cultivar CDC Frontier.
- 3. To evaluate the use of PGR to promote uniform seed maturity of Kabuli chickpea cultivar CDC Frontier.

The ultimate goal of the study was to find a practical solution to maturity-related issues of Canadian chickpea production, specifically the study attempted to find a solution to control the excessive vegetative growth in chickpea.

A secondary experiment was conducted to examine whether fungicides Pyraclostrobin (strobilurin group) and Prothioconazole (triazole group) interfere differently with PGR under field conditions. These fungicides are extensively used in western Canada to control Ascochyta blight in chickpea caused by *Ascochyta rabiei*. Previous reports indicated that fungicides

belonging to the strobilurin group have plant growth enhancement effects through their side activities on plant hormone levels, plant physiological processes such as nitrogen reductation and oxidative stress prevention (Grossmann and Retzlaff, 1997; Wu and von Tiedemann, 2001; Koehle et al., 2002). In contrast with the growth enhancement effects of strobilurin group of fungicides, the trizole type of fungicides often retards plant growth as a side effect of their activity on sterol biosynthesis (Rademacher, 2000). This indicates that Pyraclostrobin and Prothioconazole might interfere with the activities of PGR, thus the use of these fungicides can indirectly affect the results of the main study. The main objective of this secondary study was to compare the effects of Pyraclostrobin and Prothioconazole on the activities of CCC, Prohexadione Calcium and Trinexapac Ethyl on growth, yield components and maturity of the Kabuli chickpea cultivar CDC Frontier.

#### 2. Literature Review

#### 2.1 Chickpea

Chickpea (*Cicer arietinum* L.) has a long history of use as human food. First evidence of use of chickpeas goes back to the 8<sup>th</sup> millennium BC (Tanno and Willcox, 2006). Chickpea is grown in countries across all continents except, Antarctica, and has adapted to a wide range of climatic conditions, geographical regions, and cropping systems (Berger and Turner, 2007).

Chickpea belongs to the Family Fabaceae, Sub family Papillionaceae and Tribe Cicereae. Cultivated chickpea is originated in the south eastern part of Turkey from its wild progenitor Cicer reticulatum (Ladizinsky and Adler, 1976). There are two types of chickpea i.e. Desi and Kabuli. The Desi type has small and brown coloured seeds. The Kabuli type has large whitish seeds (Reddy et al., 2007). It is difficult to give an average plant height for chickpea as it depends on the agro-climatic conditions. In general, plant height of chickpeas varies from 20 cm to 100 cm. Chickpea plant development largely depend on the environment conditions. At favourable soil moisture conditions a tremendous plant growth can be expected. Chickpea has a well-developed root system and a strong stem with primary, secondary and tertiary branches. Tertiary branches are formed from the buds of secondary branches and not very important for the yield (Cubero, 1987). Most chickpea varieties have fern type leaves, but some Kabuli varieties have single (unifoliate) leaves. Leaves are borne at each node and usually have 11 to 13 leaflets in a single leaf. Single flowers are borne in axillary racemes. A purple colour corolla is characteristic for Desi types whereas Kabuli types have white colour corollas. The whole plant surface, except corolla is covered by glandular and non-glandular hairs. Chickpea has inflated oblong or ovate shape pods. The majority of pods are located on the primary branches. Seeds can be angular, owl or nearly round shaped and are characterised by a beak (Singh, 1997).

Chickpea plays an important role in crop rotation systems on the Canadian Prairies. Chickpeas provide most of their nitrogen requirement through the symbiotic nitrogen fixation with rhizobium. Thus, the inclusion of chickpea in crop rotation reduces nitrogen fertilizer inputs which are beneficial to producers and the environment (Kumar and Abbo, 2001). According to Aslam et al. (2003) chickpea can fix up to 187 kg ha<sup>-1</sup> of atmospheric nitrogen in a single growing period. Fatima et al. (2008) indicated that the use of chickpea in rotation could result in

a positive nitrogen balance (35-38 kg ha<sup>-1</sup>), which enriched the soil nitrogen. They further revealed that an 11% yield increase in the subsequent wheat crop compared to a continuous wheat monoculture. Improved soil nitrogen fertility by the previous chickpea crop would mainly contributed for this yield increase (Fatima et al., 2008). A significant increase of grain yield and grain protein content of durum wheat was also observed when it was grown immediately after pulse crops including chickpea (Gan et al., 2003b).

Chickpea has a well-developed deep root system and a large proportion of fine roots in the lower soil profile. This enables chickpea plants to utilize soil water stored in both deeper and shallower soil profiles. In addition, chickpea has the ability to increase the proportion of roots in deeper areas of the soil when the plant encounters water deficit. Therefore, chickpea is a well suited crop for rain-fed agriculture in semi-arid areas of the northern Great Plains (Benjamin and Nielsen, 2006). Chickpea has larger root diameter which is important in relation to the physical properties of the soil compared to cereals and oil seed crops (Liu et al., 2010).

#### 2.2 Challenges for chickpea production on the Canadian Prairies

Commercial chickpea production in Canada started in mid 1990's with a small acreage in the province of Saskatchewan. Within a short period, Canadian chickpea production substantially increased in Saskatchewan and Alberta. However, after reaching the maximum production level (0.45 million tonnes) in 2001, Canadian chickpea production declined (Statistics Canada, 2012). Several chickpea varieties belonging to both Kabuli and Desi types are grown in western Canada. Generally these varieties take 49 to 57 days to flower. Crop maturity can be varied, however late and medium maturing varieties are among the chickpea varieties available in western Canada (Saskatchewan Agriculture, 2012).

Chickpea production area on the Canadian Prairies has been limited by biotic and abiotic stresses. The main biotic constraint for chickpea production in western Canada is Ascochyta blight caused by the fungus *Ascochyta rabiei* Pass. Lab. (Chongo et al., 2003; Gan et al., 2009a; Banniza et al., 2011). The major abiotic limitation is the short growing season of western Canada which affects the maturity of chickpea. Chickpea has a strong indeterminate growth habit. Favourable growing conditions such as mild temperatures and high soil moisture conditions extend the vegetative state. Unlike in the traditional chickpea growing areas, cool temperatures and late summer precipitations often occur in the northern Great Plains. These conditions

prolong the crop maturity by promoting secondary vegetative growth simultaneously with the growth of new flowers and pods, consequently there is an increased risk of frost damage to the crop at the end of growing season. This downgrades the quality of seed yield due to higher proportions of green and immature seeds. Moreover, the total yield can be loss under these adverse climatic conditions (Miller et al., 2002; Anbessa et al., 2007a; Angadi et al., 2008). A mild drought stress alters assimilate allocation of indeterminate pulse crops in favour of seed development from vegetative development (Turner, 1996). Chickpea has to be subjected to abiotic stress conditions such as drought or heat stress to shift the crop from vegetative state to the reproductive phase (Anonymous, 2012). However, the climatic conditions in the northern Great Plains are often characterized by highly fluctuating and unpredictable precipitation (Padbury et al., 2002). Accordingly, late season hot and dry conditions cannot always be guaranteed in the short growing season of the Canadian Prairies. Based on the long term weather data in southern Saskatchewan, Gan et al. (2009b) indicated that the possibility of exposure of the pre-mature chickpea crop to the onset of frost was as high as one third of the time due to excessive moisture and low temperature at the later part of the growing season. Therefore, finding a solution to the delayed and uneven crop maturity due to indeterminate growth habit of chickpea is crucial to the future expansion of this important crop in western Canada.

# 2.3 Potential solutions for the uneven crop maturity of Chickpea under the western Canadian growing conditions

The permanent solution for maturity related issue of chickpea would be developing new varieties with more determinate growth habit. However, Rheenen et al. (1994) indicated that the determinate growth character in chickpea germplasm was very rare to non-existence. Rheenen et al. (1994) attempted to introduce determinate traits into the chickpea variety ICCV6 through mutation breeding. They were able to produce determinate plants, but all of them were female sterile. Under the Saskatchewan growing conditions, however, these determinate lines changed the growth habits becoming indeterminate plants (Anbessa et al., 2007b).

Using conventional breeding techniques, a determinate Desi genotype, BGD 9971 was developed recently (Hegde, 2011). This development will be useful for stabilizing chickpea productivity and product quality in the cooler climates with high fertility and moisture conditions. The characteristics of this determinate genotype were compared with two

indeterminate Desi and Kabuli genotypes. The determinate genotype had a bushy compact plant structure with short primary and secondary branches terminated with a flower bud or fully opened flower. It was slightly late for flowering and maturity. The number of pods per plant was low in determinate genotype but it had more seeds (1-4) per pod. Therefore, it had a higher number of seeds per plant (88 vs. 55 and 65) than the traditional indeterminate varieties. Hundred seed weight (18.7 g) was also slightly lower than that of the indeterminate genotypes. The determinate trait was governed by two recessive genes designated as *dt1* and *dt2* (Hegde, 2011).

Besides breeding techniques, a limited number of agronomic studies have been conducted so far to find possibilities of synchronizing the maturity of the chickpea crop on the Canadian Prairies. Chickpea maturity is affected by seed bed conditions and crop management practices. Chickpea grown in wheat and barley stubbles with a moderate nitrogen fertilizer (28-84 kg ha<sup>-1</sup>) and without adding *Rhizobium* inoculant, matured 15 days earlier than chickpea grown in conventional summer fallow. However, the treatments were effective only in those years that had climatic conditions conductive to vegetative growth (low temperatures with high rainfall throughout the growing season). Moreover, about 90% of the variation in maturity was associated with environment conditions whereas the treatment accounted only for the remaining 10% (Gan et al., 2009b).

Lentil is another important pulse crop which has an indeterminate growth habit. It is extensively grown in western Canada and faces similar problems associated with indeterminacy coupled with delayed maturity as in the case of chickpea. Therefore, any effective crop management practice to control continuous vegetative growth or to synchronize maturity would be applicable to the other crops with necessary adjustments. Zakeri et al. (2012) investigated the possibility of controlling vegetative growth and accelerate maturity of lentils at the reproductive stage using nitrogen fertilizer management. The underlying idea was to limit nitrogen supply to the early stages of vegetative growth. When providing additional nitrogen fertilizer, the crop utilizes the external source and reduces its capacity of nitrogen fixation. This would create a nitrogen deficiency towards the end of the growing season. The end season nitrogen deficiency can influence the maturation of the crop. However, the results have not supported this hypothesis. Lentils grown with nitrogen fertilizer (50 kg ha<sup>-1</sup>) did not differ from a crop relying on its own

nitrogen fixation and a crop grown without nitrogen fertilizer or inoculants for earliness, growth and yield parameters considered (Zakeri et al., 2012).

The use of crop desiccants, mainly Diquat Dibromide prior to harvest to reduce the moisture content of seeds, to desiccate green weeds and crop foliage to facilitate harvest is a common practice for chickpea and lentil in western Canada. Choudhry (2012) examined the potential use of low rates of Diquat to control the continuous vegetative growth and to enhance the maturity of lentils. In this study, Diquat was applied at 0.425 (a.i.) L ha<sup>-1</sup> and 0.85 (a.i.) L ha<sup>-1</sup> on lentil at one or three weeks after flowering. The applied rates of Diquat were a quarter and a half of the recommended rates to desiccate lentil. The treatments were able to control the vegetative growth, but with a yield penalty. Moreover, the treatments had no impact on maturity except at one occasion, the treatments extended the maturity.

Gan et al. (2008) evaluated the effect of harvest management practices on seed yield, seed quality (seed colour, shrivelled and green seeds, seed diseases), harvest index and straw quality parameters (as a livestock feed) of chickpea variety CDC Yuma under western Canadian conditions. Results indicated that these parameters were highly affected by the late and uneven maturity of chickpea under the climatic conditions of a short growing season of western Canada. Therefore, better harvest management practices would facilitate the timely maturity of the crop thus may increase the yield and seed quality. The treatments evaluated in the study were (i) direct combine when plots were sufficiently dry for threshing (ii) desiccation with Diquat [1.7] (a.i.) L ha<sup>-1</sup>] when 80% pods turned tan colour (iii) application of a low rate of Glyphosate [250 (a.i.) g ha<sup>-1</sup>] when the bottom pods turned tan colour (iv) swathing when 80% of pods turned tan colour (v) swathing bottom pods turned tan colour. It was revealed that the direct combine method was superior over all the other methods tested in this study. Desiccation with Diquat or Glyphosate treatments did not influence crop maturity, seed yield or quality characters, but tended to reduce the seed weight. Both swathing treatments reduced seed yield, seed size and downgraded seed quality traits significantly, suggesting that this technique would not be acceptable for Canadian chickpea production. The best seed yield, seed quality and harvest index were obtained with direct combining of the naturally matured crop. It emphasized the dominating effects of environmental conditions on chickpea maturity over the harvesting management practices.

Anbessa et al. (2007b) studied the pattern of post flowering dry matter accumulation, partitioning of dry matter to the vegetative and reproductive parts and the relationship of dry matter partition pattern with crop maturity of five chickpea genotypes under western Canadian growing conditions. The results revealed that the post flowering total dry matter accumulation was not related to the crop maturity i.e. genotypes gaining different post flowering dry weights were not different for the maturity duration. However, the genotypes which partitioned dry matter preferentially to the pods at higher proportions at the late reproductive stage matured earlier than the rest of the genotypes. Accordingly, the rate of partitioning assimilates to the reproductive organs (allometric partitioning coefficient) and the pod harvest index were negatively associated with the days to maturity.

# 2.4 Potential of using plant growth retardants to synchronize the seed maturity of chickpea

The agronomic studies conducted so far to address the late and uneven crop maturity of chickpea or lentil under western Canadian growing conditions (Gan et al., 2008; Gan et al., 2009; Choudhry, 2012; Zakeri et al., 2012) were based on two broad principles which were closely inter-connected; these are (i) cessation of vegetative growth at the reproductive phase of the crop and (ii) alteration of the pattern of assimilates allocation preferentially to the reproductive organs. Plant growth regulators are capable of addressing these two objectives simultaneously. Control of vegetative growth, adjusting perennial plants to annual cycles, managing the partition ratio between vegetative growth and reproduction, reducing the cost of pruning and maintaining compact plant structures are among the substantial uses of plant growth regulators (Rademarcher, 2000). Therefore, evaluation of the effect of plant growth regulators on maturityrelated issues of chickpea represents a gap in agronomic studies on the Canadian Prairies. The majority of plant growth regulators used in the agricultural industry are plant growth inhibitors. Since the majority of plant growth regulators used commercially are PGR, the word 'regulators' is generally used for retardants. Basically, these compounds interfere with the natural hormone balance of the plants. Further, gibberellins are the main target group of plant hormones among the substances used to control plant growth. In general, they interfere with different steps of the gibberellin biosynthesis pathway by inhibiting the process.

#### 2.5 Role of plant hormones

Plant hormones are natural organic substances synthesized in plants. Their functions are influential for many processes which are essential for plant survival and the whole plant development. The activities of plant hormones may be localized to the site of synthesis or they can be transported and activate a response away from their site of synthesis (Davis, 2004). Plant development is an outcome of coordinated gene expressions. Coordination of gene signalling is partly directed by plant hormones (Chandler, 2009). The vast majority of plant organs are developed after germination. Being sessile organisms, plants have no option but to adapt to their surrounding environment. Therefore, this post-emergence development mode enables to integrate environmental inputs to a plant's genetic program, which is vital to their survival. Plant hormones play a central role of integrating environmental cues to plants' genetic program (Santner and Estelle, 2009; Depuydt and Hardtke, 2011). Auxins, abscisic acid, cytokinins, gibberellins, ethylene, brassinosteroids, jasmonic acid, salicylic acid, nitric oxide and strigolactones are the compounds currently identified as plant hormones (Santner and Estelle, 2009).

#### 2.6 Role of Gibberellins in plants

Gibberellins were first identified by Japanese scientists who worked with "bakanae" disease of rice. The name "Gibberellin" came into use by 1935 after the isolation of the pure form of noncrystalline solids which had high growth promoting activity (Tamura, 1991). The well-known function of gibberellins in plants is in stem elongation. In addition, stimulation of flowering in some species, induction of bolting in long day plants, induction of seed germination, promotion of fruit setting and fruit growth are among the main effects of gibberellins (Taiz and Zeiger, 2002; Davis, 2004).

#### 2.6.1 Gibberellin biosynthesis

Gibberellins biosynthesis in higher plants can be categorized in to three stages;

- (i) Biosynthesis of ent-kaurene
- (ii) Formation of  $GA_{12}$  from ent-kaurene
- (iii) Formation of Carbon 20 and Carbon 19 structure Gibberellins

During the first stage, geranylgeranyl diphosphate (GGDP) is synthesised in plastids. This GGDP is converted to ent-coplyl diphophate and then to ent-kaurene by the activity of ent-copallyl diphosphate synthase and ent-kaurene synthase enzymes.

The second stage of the gibberellin biosynthesis takes place in the endoplasmic reticulum. During this stage, ent-kaurenic acid is formed through the stepwise oxidation of ent-kaurene. The oxidation of ent-kaurene is catalyzed by ent-kaurene oxidase enzyme. Further oxidation and a ring contraction (B ring) of the molecule convert ent-kaureneic acid to  $GA_{12}$ .  $GA_{12}$  is the first gibberellin product of the gibberellin biosynthesis pathway and it is the precursor of all other forms of gibberellins in subsequent reactions.  $GA_{12}$  can be further hydroxilated to  $GA_{53}$  during this stage.

At the third stage,  $GA_{12}$  or  $GA_{53}$  is subjected to series of oxidations which finally cause the formation of various Gibberellin forms.  $GA_{20}$  oxidase,  $GA_3$  oxidase and  $GA_2$  oxidase enzymes (dioxygenase enzymes) catalysed this oxidation steps while the 2-oxoglutarate and molecular oxygen act as the substrate for these reactions. This third stage is occurring in the cytosol. The activities of the above enzymes are key to convert different gibberellin molecules into biologically active and biologically inactive forms. Oxidations of the  $20^{th}$  carbon atom and the  $3\beta$  position of the GA molecules are the main requirements for biological activity. The activities of  $GA_{20}$  oxidase and  $GA_3$  oxidase enzymes fulfil these requirements respectively. Oxidation of the  $2\beta$  position of GA molecules through the activity of  $GA_2$  oxidase makes them biologically inactive (Olszewski et al., 2002; Taiz and Zeiger, 2002).

#### 2.7 Plant growth regulators

Plant growth regulators are the substances which can influence, retard or modify any physiological process in plants, excluding nutrients (Basra, 2000). Therefore, a wide variety of growth promoting and retarding substances including plant hormones are classified as plant growth regulators. They are being used for a wide range of crop management practices throughout the world. However their contribution to the agro-chemical market is small i.e. below 5% (Rademacher, 1991 and 2000). Gibberellin biosynthesis inhibitors or PGR are the leading group of the plant growth regulators by market share and used area (Rademacher, 2010). A list of

commercially used plant growth regulators with their well-known applications in agriculture and horticulture industry is included in Appendix 1.

#### 2.7.1 Plant growth retardants

PGR are synthetic substances primarily used to control vegetative growth of the plants. They are antagonistic to gibberellins thus reduce the rate of cell elongation and cell division. Except for the reduction of vegetative growth, application of PGR does not affect many other development processes of plants. Therefore, they are mainly being used to adjust plant growth in a desired way (Rademacher, 2000). Based on the mode of action on Gibberellin biosynthesis pathway, PGR can be categorized in to four groups (Rademacher, 2000).

- (i) Onium compounds
- (ii) Compounds with a Nitrogen containing heterocycle
- (iii) Structural mimics of 2-oxoglutaric acid
- (iv) 16, 17-dihydro GAs

Figure 2.1 illustrates the well-known PGR belonging to each group and their main sites of activity in GA biosynthesis pathway.

PGR are primarily used to control plant height of plant species by arresting stem elongation. In cereal grain production in Europe, PGR are traditionally used to reduce lodging by shortening and thickening stems. In northern Europe especially where high moisture conditions at the end of the growing season frequently occur, PGR are commonly used to control lodging caused by excessive growth (Rajala and Peltonen-Sainio, 2000). Rajala and Peltonen-Sainio (2000) also stated that PGR may increase yield of cereal crops due to the partitioning of surplus assimilates to the yield components instead of vegetative growth and other direct or indirect effects on crop characteristics. In the ornamental plant industry, PGR play an important role in controlling plant growth. Usually these plants are grown in containers at the initial stages and provided with high fertility and abundant moisture to enhance plant structure. In most cases, long photoperiods and high temperatures are enforced to control flowering in plant species. These conditions result in rapid stem elongation. Consequently, plants will be taller than desired.

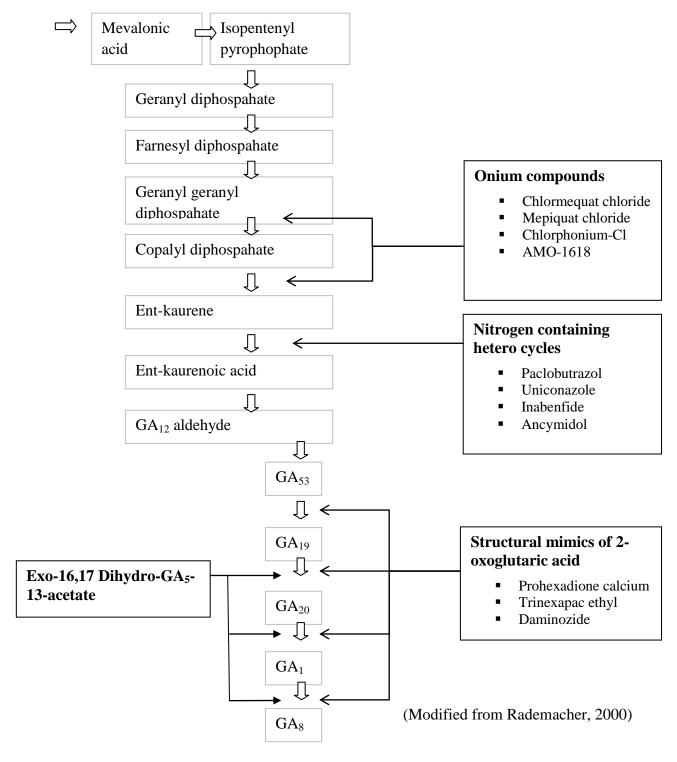


Figure 2.1: Mode of action of different groups of plant growth retardants on gibberellin biosynthesis pathway.

Therefore, PGR are used to obtain the required height levels for a wide range of ornamental plants with different growth habits (Gent and McAvoy, 2000). Furthermore, PGR are used for many other crops for the direct and indirect benefit of the controlled vegetative growth.

#### 2.8 Plant growth retardants used in this study

#### **2.8.1** Chlormequat Chloride (CCC)

CCC is chemically known as (2- chloroethyl) trimethylammonium chloride which was developed in the 1960s. Initial experiments revealed that CCC was the opposite of growth alternations achieved with gibberellins. The activity of CCC was reversed by applying gibberellins. Short plants with shorter internodes, thick stems and much greener leaves were the features of the treated plants. It also inhibited seed germination (Tolbert, 1960 and 1961). CCC can be absorbed by aerial parts of the plants or by roots. When applied to the soil, about 90% CCC was recovered from roots while the rest was recovered from other plant parts in treated wheat plants after six hours. Seven and half days after the application, 50% of CCC by total absorption was detected in plants in its original form. This amount dropped to 30% after 14 days. CCC was metabolized in plants into choline followed by betaine, glycine and serine (Dekhuijzen and Vonk, 1974).

#### 2.8.2 Prohexadione Calcium

Prohexadione Calcium was originally developed in Japan in early 1980's and it is a derivative of acylcyclohexanedione. Its systematic chemical name is calcium 3-hydroxy-5-oxo-4-propionyl-cyclohex-3-ene-1-carboxylate (Halbwirth et al., 2006). Initially it was developed to control the growth of rice and later its use has been expanded to other crops including apple, cereals and peanuts. It is a structural mimic of 2-oxoglutaric acid which is the co-substrate of dioxygenase enzymes. Therefore, Prohexadione Calcium blocks the activity of dioxygenases required for the formation of biologically active and inactive forms of gibberellins in the later stages of gibberellin biosynthesis (Rademacher, 2009). Foliar applied Prohexadione Calcium on apple plants translocated acropetally to the growing points. Minimum basipetal movement was detected. In higher plants, Prohexadione Calcium is metabolised to naturally-occurring tricarballylic acid (Evans et al., 1997; Rademacher, 2009). Prohexadione Calcium exhibits very low susceptibility for crop residuals and low mammalian toxicity. It rapidly depletes in the soil (Winkler, 1997). In North America, Prohexadione Calcium was initially introduced for growth

control of apple. Application of Prohexadione Calcium at 250 mg L<sup>-1</sup> at 7 to 21 days after the petal fall stage of apple has reduced vegetative growth with a range of 47% to 67%. Consequently, pruning weight (weight of removed parts) and pruning time was reduced. Fruit set and fruit quality were not affected (Unrath, 1997).

#### 2.8.3 Trinexapac Ethyl

Trinexapac Ethyl is also a derivative of acylcyclohexanedione and was developed parallel with Prohexadione Calcium. Both substances have similar mode of action on the gibberellin biosynthesis pathway. Trinexapac Ethyl was primarily intended for use as an anti-lodging agent for small grain crops and for use as a turf grass growth regulator (Griggs et al., 1991; Rademacher and Bucci, 2002; Rademacher, 2009). Trinexapac Ethyl is chemically known as 3-hydroxy-5-oxo-4-cyclopropanecarbonyl-cyclohex-3-ene-1-carboxylic acid ethyl ester (Halbwirth et al., 2006). It is rapidly metabolised within plants into its free acid form (CGA-179500) which is primarily responsible for its biological activity (Anonymous, 2001). In North America, it was initially used for growth control of turf grass. Up to 94% of Trinexapac Ethyl was absorbed by the areal parts (leaf sheath and leaf blades) of Kentucky blue grass within 2 hours after application. However, absorption by roots was very low (5%) even 24 hours after the application. Further, Trinexapac Ethyl can be translocated acropetally or basipetally within plants. Half of the Trinexapac Ethyl applied on the plant base was translocated acropetally to the plant foliage within 24 hours. It was however, predominantly translocated basipetally when applied on leaf blades (Penner and Fagerness, 1998).

#### 2.9 Use of plant growth retardants in cotton

The main intention of the use of PGR in this study was to control vegetative growth of chickpea at the reproductive stage to allow for more assimilate translocation towards the reproductive organs. It was assumed that this would increase seed size and uniformity of seed maturity. PGR are being widely used in cotton production to achieve objectives similar to those of the present study.

Cotton has an indeterminate growth habit. Vegetative growth of cotton often continues later in the season under favourable growing conditions (Cothren and Oosterhuis, 2010). This late season excessive vegetative growth causes several production problems such as delayed maturity, fruit

abortions and harvest difficulties. PGR are commonly used in cotton production to control vegetative growth at agronomically desired growth stages. As a result of retarded vegetative growth, more resources are diverted to the first position ball (early set balls) development which can influence ball retention and timely maturity (Jost et al., 2006).

Application of Mepiquat Chloride as a split application regime on cotton plants, reduced plant height effectively by 33% averaged across different row widths (Gwathmey and Clement, 2010). The treatment application time varied within three years of the experiment. A first round of Mepiquat Chloride was applied 43 to 56 days after planting. The second and third rounds were applied at 8 to 9 and 14 to 25 days after the first application, respectively. In addition to a reduction in plant height, Mepiquat Chloride increased ball density on the lower section of the plants, diverted more photosynthates to the balls and synchronized ball maturity (Gwathmey and Clement, 2010).

Application of Chlorocholine Chloride was effective in controlling vegetative growth of late planted cotton at Rio Dulce area in Argentina. Late-planted cotton in that area has a narrow window to complete the production cycle before frost of the advancing winter. In general, the growth cycle of late-planted cotton is extended due to the excessive vegetative growth in the first half of the season and then due to the declining temperatures. Chlorocholine Chloride applied at two growth stages (based on mean inter node length) effectively reduced plant height, node number, inter node length and aerial biomass with a substantial increase of yield by 35% due to the increased ball weight and ball number (Mondino et al., 2004).

A major challenge of cotton production in the northern cotton belt of the United States is to have the crop mature within the short growing period before the beginning of the cold weather. The crop needs to shift from vegetative to reproductive growth in mid-season to provide adequate time for balls to mature. PGR, particularly Mepiquat Chloride, are a major crop management tool used to influence cotton maturity in that region. A single application of the recommended rate at 65 to 80 days after planting and multiple applications of a low rate of Mepiquat Chloride onwards 51 days after planting significantly reduced plant height and hastened the progress of flowering. Multiple applications were more influential on the progress of flowering and reduction of plant height of more indeterminate cultivars. In general, lint yield was not affected by Mepiquat Chloride, but lint yield tended to decrease in more determinate and early maturing

cultivars. In addition, Mepiquat Chloride significantly influenced earliness. The effects were more prominent on more indeterminate cultivars (Gwathmey and Craig, 2003).

#### 2.10 Use of plant growth retardants in pulse crops

Pulse crops are an integral part of cropping systems in the south Asian region. Increasing the productivity of food legume crops is a crucial requirement in that region due to the limited arable lands for crop production. Yadev and Bharud (2009) conducted a field study to examine the effects of various plant growth regulators on yield and yield components of Kabuli chickpea variety 'Virat'. Treatments included 10 ppm of Gibberellic Acid , 20 ppm of Naphthalene Acetic Acid , 25 ppm of Benzyl Adenine, 25 ppm of Cycocel and two commercial products called "Bioforce" and "Biopower" at 2mL L<sup>-1</sup> each and were sprayed four times at 10-day intervals from the initiation of flowering. The Cycocel treatment increased seed yield of chickpea by 16 % over the control and improved all yield components, such as number of pods plant<sup>-1</sup>, number of seeds plant<sup>-1</sup>, 100-seed weight and harvest index.

Brar et al. (1992) reported that plant growth regulators had a significant effect on grain production and dry matter partition of late- and early-sown chickpea in northern India. Early- and late-sown Desi chickpea variety GL769 and Kabuli chickpea variety L550 were treated with two PGR (Maleic Hydrazide at 200 ppm and Cycocel at160 ppm) and a growth enhancing substance, Kinetin at 10 ppm and 20 ppm at flower initiation stage. The effects of plant growth regulators were dependent on the sowing date, but not on the genotype. PGR (Maleic Hydrazide and Cycocel) significantly increased the yield of early-sown crops whereas Kinetin significantly increased the yield of late-sown crops. In addition, Cycocel and Maleic Hydrazide increased pod diameter. A slight increase of stem diameter and a slight decrease in leaf diameter were also obtained with the PGR.

Bora and Sarma (2006) stated that pea seeds soaked in different concentrations (10, 100, 250, 500 and 1000 mg L<sup>-1</sup>) of CCC solutions for 12 hours before sowing showed significant changes in growth and yield parameters. Increasing concentrations of CCC consistently reduced shoot length measured at three-day intervals up to 19 days after sowing. In addition, CCC increased branching, leaf chlorophyll content, number of flowers, pods per plant, seed weight and seed protein content. They suggested that controlling excessive vegetative growth would be beneficial

for synchronisation of flower initiation and pod development. However, the seeds treated with similar concentrations of Gibberellic Acid (GA<sub>3</sub>) also had a positive effect on selected yield parameters such as number of pods plant<sup>-1</sup> and mean seed weight. Both chemicals (GA<sub>3</sub> and CCC) increased the productivity of the crop, but the maximum productivity was dependent on the variety.

Sharma and Lashkari (2009) reported that seed yield and yield quality of cluster bean (*Cyamopsis tetragonaloba* L.) could be improved by CCC application. Cluster bean cultivar Pusanvibahar treated with different concentrations (1000, 1500 and 2000 ppm) of CCC had more branches, higher pod weights, higher pod yield and higher pod crude protein contents. The lowest plant heights and the highest seed yield were obtained with the application of CCC at 2000 ppm concentration (Sharma and Lashkari, 2009).

According to Devi et al. (2011), critical growth and yield parameters of soybean can be altered by plant growth regulators. Salicylic Acid (50 ppm), Ethrel (200 ppm) and CCC (500 ppm) were applied at flower initiation stage [40 days after seeding (DAS)], pod initiation stage (60 DAS) and both stages (40 DAS+60 DAS) on soybean. All plant growth regulators significantly increased number of branches plant<sup>-1</sup>, plant dry weight at 75 DAS, leaf area index at 75 DAS, leaf chlorophyll content, leaf carotenoids content, number of pods plant<sup>-1</sup>, 100-seed weight, number of seeds pod<sup>-1</sup>, seed protein content, seed oil content, seed yield and harvest index regardless the growth stage. The maximum growth and yield enhancement was associated with the Ethrel treatment applied at both growth stages (40 DAS+60 DAS). Multiple application of CCC (40 DAS+60 DAS) was superior for leaf chlorophyll and carotenoids contents. Increased branches and chlorophyll content by plant growth regulators could enhance the photosynthesis and transfer of assimilates to the seeds. These changes resulted in higher yield (Devi et al., 2011).

PGR could influence the partitioning of assimilates towards the reproductive growth of soybean. Semi determinate soybean genotype MACS-124 and determinate soybean genotype JS-335 were treated with PGR; Cycocel (250 and 500 ppm), Mepiquat Chloride (500 and 1000 ppm), Triiodobenzoic acid (TIBA; 50 and 100 ppm) and a growth enhancer, Kinetin (25 and 50 ppm) at the flowering stage (40 DAS). The total dry weight of the plants at harvest was significantly increased by all plant growth regulators whereas the plants treated with PGR (Cycocel, Mepiquat

Chloride and TIBA) had higher total dry weights compared to the Kinetin treatments. Comparable with the total dry weight, the dry weight of reproductive organs (from 70 DAS to harvest) were significantly increased by PGR. In addition, determinate genotypes accumulated more dry matter at the reproductive stage regardless of the type or the concentration of PGR. Results indicated that PGR improved the efficiency in the utilization of resources and enhanced source supply to the sinks (Kumar et al., 2006).

#### 2.11 Effects of fungicides on plant growth and development

Pyraclostrobin and Prothioconazole are commonly used fungicides in chickpea production in North America to control Ascochyta blight. Pyraclostrobin belongs to the strobilurin fungicide group whereas Prothioconazole belongs to the triazole fungicide group. In addition to the effects directly related to the disease control, fungicides belonging to the strobilurin group can influence growth and yield of the treated plants by affecting some physiological processes.

Koehle et al. (2002) discussed the effects of Pyraclostrobin on the physiology of wheat plants. Pyraclostrobin enhanced yield and biomass of the treated wheat plants. This was mainly due to an increase in nitrogen uptake and nitrogen reduction. In addition, Pyraclostrobin affected the levels of plant hormones. It reduced ethylene biosynthesis by decreasing 1-aminocyclopropane – carboxylic acid (ACC) synthase activity. Furthermore, it increased the levels of Indole-3-acetic acid (IAA) and abscisic acid (ABA). The increased in IAA levels are believed to be a result of Pyraclostrobin metabolism within plants. Plant senescence was delayed by Pyraclostrobin, which is mainly due to the reduction of ethylene and increase of IAA levels. Increased ABA levels can enhance the stress tolerance capacity of plants. Pyraclostrobin also improved the anti-oxidative capacity of barley plants by increasing the activity of anti-oxidative enzymes (Koehle et al., 2002).

Grossmann and Retzlaff (1997) studied the effects of Kresoxim Methyl (a fungicide belonging to the strobilurin group) on the physiological processes of wheat under laboratory conditions. Kresoxim Methyl increased cytokinins (113 to 160 %) and reduced ACC levels up to 50%. ACC is the precursor for ethylene, thus ethylene levels were also reduced by 36%. Plant fresh weight was increased by 12% at six days after treatments. In addition, Kresoxim Methyl delayed leaf senescence and slightly increased IAA, ABA and gibberellin (GA<sub>1</sub>) levels. Due to these changes,

enhanced growth performances and higher yield can be obtained besides the fungicidal activity of Kresoxim Methyl.

Rademacher (2000) indicated that the triazole type of fungicides often suppress plant growth as a side effect of their activity on sterol biosynthesis. Therefore, in contrast to the growth enhancing effects of Pyraclostrobin, the use of Prothioconazole (a triazole group of fungicide) to control Ascochyta blight in chickpea might suppress the growth of the treated plants.

#### 3. Materials and methods

#### 3.1 Study of the influences of fungicides

This small scale study was conducted under supplementary irrigation at Brooks in 2011. The treatments were arranged in a Randomized Complete Block Design with four replicates. Comparable with the main study, an individual plot (experimental unit) included four 6-m long rows spaced 30 cm apart. Site description, crop establishment, agronomic practices, data collection and analysis of this study were identical to the main study in 2011 and will be discussed in the sections 3.2, 3.3, 3.5 and 3.7, respectively. Table 3.1 summarises the phenological events and crop management practices relevant to this study.

Table 3.1: Dates of main phenological events and crop management practices of the study conducted to evaluate the influence of two fungicides on the activity of plant growth retardants at Brooks in 2011

Phenological event/crop management practice	Date
Seeding	06 May (126)
50% seedling emergence	22 May (142)
50% flowering	02 July (183)
Irrigation 35 mm	06 July (187)
40 mm	09 July (190)
Diquat dibromide – 1.7 L ha <sup>-1</sup>	15 Sept (258)
Harvesting	21 Sept (264)
Fungicide applications	
Chlorothalonil - 3.5 L ha <sup>-1</sup>	22 June (173)
Pyraclostrobin - 500 mLha <sup>-1</sup> + Boscalid - 395 mL ha <sup>-1</sup>	29 June (180)
	06 July (187)

Table 3.2 outlines the treatment information for this study. Boscalid was always applied with Pyraclostrobin as it is an integral part of the commercial product recommended for Ascochyta blight control of chickpea (Headline Duo<sup>®</sup>, BASF Canada).

Table 3.2: Concentrations and combinations of fungicides and plant growth retardants applied as treatments for fungicide-plant growth retardant study in 2011

Treatment	Concentration of	Concentration of
	PGR in mixture	fungicide in mixture
1. (Pyraclostrobin+ Boscalid)	-	500 mL ha <sup>-1</sup> + 395 g ha <sup>-1</sup>
2. Prothioconazole	-	370 mL ha <sup>-1</sup>
3. (Pyraclostrobin+ Boscalid) + Prohexadione Calcium	1500 mg L <sup>-1</sup>	
4. (Pyraclostrobin+ Boscalid) + Trinexapac Ethyl	2000 mg L <sup>-1</sup>	500 mL ha <sup>-1</sup> + 395 g ha <sup>-1</sup>
5. (Pyraclostrobin+ Boscalid) + CCC	$6000 \text{ mg L}^{-1}$	
6. Prothioconazole + Prohexadione Calcium	1500 mg L <sup>-1</sup>	
7. Prothioconazole + Trinexapac Ethyl	$2000~\text{mg}~\text{L}^{1}$	370 mL ha <sup>-1</sup>
8. Prothioconazole + CCC	6000 mg L <sup>-1</sup>	

Note: Pyraclostrobin+Boscalid were applied twice to all experimental units before this treatment application to keep ascochyta blight disease under control. Treatments listed in this table excluded that prior application of Pyraclostrobin + Boscalid.

Treatments were applied once at the 20 DAF growth stage on 21 July 2011 with similar equipment used in the main study. The mixed procedure of the Statistical Analysis System (SAS) version 9.2 (SAS Institute Inc., Cary, NC, USA) was used for data analysis. Treatments as stated in Table 3.2 were considered as fixed factors, whereas replicates were considered as random effects for the analysis. Means were compared by pre-determined contrasts and the differences were declared significant at p< 0.05.

The original plan was not to apply Pyraclostrobin to this experiment other than with treatments. Chlorothalonil was applied for all plots before flowering as a preventive measure of Ascochyta blight. However, about 4 days after the Chlorothalonil application, plants were exposed to a storm with severe wind which resulted in damages to aerial parts. Then, a rapidly spreading Ascochyta blight epidemic occurred. Therefore, all experiment units (plots) were treated with Pyraclostrobin + Boscalid at recommended rates twice, 7 days apart to avoid complete crop failure.

#### 3.2 Test site description

Field experiments were conducted in 2010 and 2011 growing seasons at two locations in southern Alberta, one at the research site of the Crop Diversification Centre South, Brooks (50° 33′ 51″ N and 111° 53′ 56″ W, Elevation 758 m) and the other at the Bow Island Sub

Station near Bow Island (49° 52′ 3″ N and 111° 22′ 46″ W; Elevation 817 m). At the Brooks test site, one experiment was conducted with supplementary irrigation to maintain the soil moisture levels of the test site over 50% of field capacity until the crop reached 50% pod set, and the other experiment was conducted under rain-fed conditions. The experiment at the Bow Island test site was designed to have supplementary irrigation, however, due to excessive rainfall received during the growing seasons in 2010 and 2011, soil moisture levels at the test site were > 50 % of field capacity for almost the entire growing season, therefore, supplementary irrigation was not required in both years.

The main soil type at the Brooks test site is Orthic Brown Chernozemic with silty loam surface whereas the Bow Island site has a medium textured Orthic Brown Chernozemic soil (Alberta Soil Information Viewer, 2012). The physical and chemical status of the soils at test sites in early spring of 2010 and 2011 growing seasons are given in Table 3.3.

The soil pH value at test sites ranged from 7.5 to 8.8 in the 0 - 60 cm soil depth indicating the soil was slightly alkaline. The soil organic matter content of the top soil layer (0-15 cm) varied from 1.2% to 1.7%. The average soil organic matter content of Brown Chernozemic soil region in Canada is 2.5% – 3.4% (Huffman et al., 2012). This indicates the soil of the test sites were relatively low in organic matter. The available nitrate-nitrogen (NO<sub>3</sub>-N) levels varied with soil depths at both test sites (Table 3.3). In 2011, the Brooks site had very low available NO<sub>3</sub>-N contents in entire soil depth (0 - 60 cm). However, nitrogen fertilizer is not generally recommended for chickpea under western Canadian growing conditions when using the proper inoculant for symbiotic nitrogen fixation (Walley, 1999). A specific guideline for the other major nutrient (phosphorous, potassium and sulphur) requirement for chickpea production in western Canada is not available.

Table 3.3: Physical and chemical properties of soils at the Brooks and Bow Island test sites in 2010 and 2011

		Brooks					Bow Island					
Physical and chemical		2010			2011			2010			2011	
characteristics	0-15cm#	15-30cm	30-60cm	0-15cm	15-30cm	30-60cm	0-15cm	15-30cm	30-60cm	0-15cm	15-30cm	30-60cm
pН	7.9	8	8	7.6	7.9	8.2	7.5	7.5	8	7.7	8.1	8.8
EC <sup>X</sup> (mS cm <sup>-1</sup> )	0.52	0.43	2.23	0.66	1.18	2.1	0.37	2.37	2.02	0.68	0.6	0.99
Organic matter												
content (%)	1.5	-	-	<1.2	-	-	1.4	-	-	1.7	-	-
Nitrate-nitrogen												
(kg ha <sup>-1</sup> )	24.6	22.4	44.8	10.1	5.6	<11.2	24.6	20.2	33.6	32.5	16.8	15.7
Phosphorus (kg ha <sup>-1</sup> )	199.3	-	-	65	-	-	159.0	-	-	147.8	-	-
Potassium												
(kg ha <sup>-1</sup> )	2240	-	-	370	-	-	1568	-	-	985.6	-	-
Sulphate sulphur												
(kg ha <sup>-1</sup> )	61.6	<28	>1120	94.1	>224	>448	<28	>560	>1120	23.5	<11.2	179.2

<sup>\*</sup> Soil depth; \* Electrical conductivity; (-) not collected.

In general, 40-60 kg ha<sup>-1</sup> of phosphorus, 17-25 kg ha<sup>-1</sup> of potassium under low potassium conditions and 20 kg ha<sup>-1</sup> of sulphur for sulphur deficient soils are recommended for chickpea seed production (Gaur et al., 2010). As indicated in Table 3.3, the soil of the test sites contained these nutrients at levels above those required. In the 2010 growing season, to avoid potential nutrient deficiencies, fertilizer mixture of 11:52:0 (N:P:K) was evenly applied at a rate of 46 kg ha<sup>-1</sup> on May 09 at the Bow Island test site.

#### 3.3 Crop establishment of the test sites

The late-maturing medium-seeded (mean seed weight of 367 mg seed <sup>-1</sup>) type Kabuli chickpea cultivar CDC Frontier was used for this study. The average plant height of this cultivar is 40 cm. In general, the plants takes about 53 days to flower and 97 days to mature with an average seed yield 1,936 kg ha<sup>-1</sup> in the Brown Soil Zones of Alberta and Saskatchewan (Warkentin et al., 2005).

Seeds were tested for Ascochyta blight infection and viability by a Canadian Food Inspection Agency's accredited laboratory (Parkland Laboratories, Red Deer, AB) prior to seeding. The test results revealed that the seeds were free of *Ascochyta* with a germination rate of 82%. For precautionary purpose, however, the seeds were treated with the mixture of Fludioxonil and Mefenoxam [Apron Maxx®, Syngenta Crop Protection Inc, 2.31% and 3.46% active ingredient (a.i.)] at a rate of 325 mL 100 kg <sup>-1</sup>. A granular inoculant (Nodulator®, Becker Underwood Canada Ltd.) containing *Mesorhizobium ciceri* nitrogen-fixing bacteria was applied with the seed at a rate 5.6 kg ha<sup>-1</sup>. Plots were seeded at a density of 55 seeds m<sup>-2</sup> with a double disc drill. Each plot consisted of four 6-m long rows spaced 30 cm apart with a total plot area of 7.2 m<sup>2</sup>.

#### 3.4 Treatments

Three commercially available PGR, namely Prohexadione Calcium (Apogee<sup>®</sup>, BASF Canada Inc., 27.5% a.i.), CCC [Cycocel<sup>®</sup>, BASF Canada Inc., 460 g (a.i). L<sup>-1</sup>) and Trinexapac Ethyl [Palisade<sup>®</sup>, Syngenta Crop Protection Inc., 250 g (a.i) L<sup>-1</sup> used in 2010 and 120 g (a.i) L<sup>-1</sup> used in 2011] were applied at four concentrations at three growth stages [10 days after 1<sup>st</sup> flowering (10 DAF), 20 DAF and 30 DAF]. Prohexadione Calcium was applied at 750, 1500, 3000 and 4500 mg (a.i.) L<sup>-1</sup> concentrations. CCC was applied at 1000, 2000, 4000 and 6000 mg (a.i) L<sup>-1</sup>. In 2010, Trinexapac Ethyl was originally scheduled to be applied at 1000, 2000, 4000 and 6000 mg

(a.i.) L<sup>-1</sup> concentrations. However, the actual concentrations applied in 2010 were more than twice the planned concentration due to a miscommunication on the active ingredient concentration of the original product received. Consequently, concentrations applied in 2010 were 2083, 4167, 8333 and 12498 mg (a.i.) L<sup>-1</sup>. In 2011, however, Trinexapac Ethyl was applied as planned. A summary of the PGR concentrations and the application volume at each test site is given in Table 3.4.

Table 3.4: Concentrations and application rate of three PGR used in the field studies in 2010 and 2011

Tyl	pe of Plant growth retai	rdant								
Prohexadione Calcium	CCC	Trinexap	ac Ethyl							
(2010/2011)	(2010/2011)	2010	2011							
Concentration [mg (a.i.) L <sup>-1</sup> ]										
750	1,000	2,083	1,000							
1,500	2,000	4,167	2,000							
3,000	4,000	8,333	4,000							
4,500	6,000	12,498	6,000							
Plant growth retardant solution volume (L ha <sup>-1</sup> )										
225	225	225	225							

Note: a.i. = active ingredient.

#### 3.5 Agronomic practices

A summary of dates and treatment rates, herbicide, fungicide, desiccant applications and the dates of crop phenological events at each test site are given in Table 3.5. For broad-leaf weed control, Ethalfluralin [Edge<sup>®</sup>, Dow Agro Sciences Canada Inc., 5% (a.i.)] was applied in the fall of 2009 and 2010 at the Brooks test site, whereas Trifluralin [Rival<sup>®</sup>, Nufarm Agriculture Inc., 500 g (a.i.) L<sup>-1</sup>] was applied in early spring of 2010 and 2011 at the Bow Island test site. In addition, several rounds of manual weeding were carried out at all sites. At the vegetative stage, just prior to flowering, fungicides Azoxystrobin [Quadris<sup>®</sup>, Syngenta Crop Protection Inc., 250 g (a.i.) L<sup>-1</sup>] and Chlorothalonil [Bravo<sup>®</sup>, Syngenta Crop Protection Inc., 720 g (a.i) L<sup>-1</sup>] were applied at recommended rates as a precautionary measure against Ascochyta blight. Azoxystrobin was applied only in 2010.

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Table 3.5: Dates of major phenological events, treatment applications and crop management practices at test sites during the 2010 and 2011 growing seasons

Dhanalagiaal ayant/	Brooks 1	<b>Irrigated</b>	Brook	s Rain-fed	Bo	w Island
Phenological event/ treatment	2010	2011	2010	2011	2010	2011
ti cutilicit			Date of e	vent		
Fertilizer applications – (N:P:K: at 11:52:0 – 46 kg ha <sup>-1</sup> )	-	-	-	-	09 May (129) <sup>#</sup>	-
Seeding	20 May (140)	06 May (126)	20 May (140)	06 May (126)	19 May (139)	12 May (132)
50% seedling emergence	05 June (156)	22 May (142)	05 June (156)	22 May (142)	05 June (156)	05 June (156)
50% flowering	11 July (192)	02 July (183)	11 July (192)	02 July (183)	14 July (195)	08 July (189)
PGR treatment 1 (10 DAF)!	21 July (202)	11 July (192)	21 July (202)	11 July (192)	21 July (202)	18 July (199)
PGR treatment 2 (20 DAF)	03 Aug (215)	21 July (202)	03 Aug (215)	21 July (202)	03 Aug (215)	28 July (209)
PGR treatment 3 (30 DAF)	16 Aug (228)	02 Aug (214)	16 Aug (228)	02 Aug (214)	16 Aug (228)	08 Aug (220)
Ethalfluralin (Edge®) – 9 kg ha <sup>-1</sup>	20 Oct 2009	07 Oct 2010	20 Oct 2009	07 Oct 2010		-
Trifluralin (Rival®) - 2.25 L ha <sup>-1</sup>	-	-	-	-	14 May 2010	10 May 2011
Azoxystrobin (Quadris®) – 500 mL ha <sup>-1</sup>	23 Jun (174)	-	23 June (174)	-	23 June (174)	-
Chlorothalonil (Bravo®) – 3.5 L ha <sup>-1</sup>	08 July (189)	22 June (173)	08 July (189)	22 June (173)	08 July (189)	23 June (174)
Pyraclostrobin (Headline®) - 500 mLha <sup>-1</sup> + Boscalid (Lance®) - 395 mL ha <sup>-1</sup>	21 July (202) 16 Aug (228)	29 June (180) 06 July (187)	21 July (202) 16 Aug (228)	29 June (180) 06 July (187)	21 July (202) 16 Aug (228)	30 June (181) 08 July (189)
Prothioconazole (Proline®) - 500 mL ha <sup>-1</sup>	03 Aug (215)	11-13 July (192-194) 21-22 July (202-203) 02-05 Aug (214-217)	03 Aug (215)	11-13 July (192-194) 21-22 July (202-203) 02-05 Aug (214-217)	03 Aug (215)	18-20 July (199-201) 28-29 July (209-210) 08-10 Aug (220-222)
Irrigation	27 July (208) - 35mm 28 July (209) - 20mm	06 July (187) -35mm 09 July (190)-40mm	-	-	-	-
Diquat dibromide (Reglone®) – 1.7 L ha <sup>-1</sup>	Oct 12 (285)	15 Sept (258)	Oct 12 (285)	15 Sept (258)	Oct 13 (286)	-
Harvesting	01 Nov (305)	21 Sept (264)	23 Oct (296)	23 Sept (266)	20 Oct (293)	12 Oct (285)

DAF= Days after 1<sup>st</sup> flowering; \*Day number of the year is shown within parenthesis.

In general, post flowering Ascochyta blight was controlled by applying Pyraclostrobin [Headline<sup>®</sup>, BASF Canada Inc., 23.6% (a.i.)] with Boscalid [Lance<sup>®</sup>, BASF Canada Inc., 70% (a.i.)], and Prothioconazole [Proline<sup>®</sup>, Bayer Crop Science Inc., 480 g (a.i.) L<sup>-1</sup>]. Both fungicides were applied at recommended rates as a mixture with the PGR treatments or alone 1 - 2 days after the PGR treatments. To control a severe Ascochyta blight epidemic in 2011, the recommended mixture of Pyraclostrobin and Boscalid was applied twice prior to the PGR treatment application. A crop desiccant, Diquat Dibromide [Reglone<sup>®</sup>, Syngenta Crop Protection Inc., 240 g (a.i.) L<sup>-1</sup>] was applied at all test sites except the Bow Island site in 2011 to facilitate the harvest operations.

#### 3.6 Treatment arrangement and experimental design

In 2010, treatments were assigned as a split-plot in a Randomized Complete Block Design with six replicates in each experiment. The growth stage was considered as the main plot and PGR types and concentrations were collectively considered as the sub plot. For each growth stage, an untreated plot was included as control. In 2011, field experiments were modified as a split-split-plot in a Randomized Complete Block Design to manage possible special variation within a replication due to relatively long replication size. The growth stage, PGR type and PGR concentration were considered as main plot, sub-plot and sub-sub plot, respectively. An untreated plot was included for each PGR type, to serve as control. Six replicates were also maintained at each test site in 2011.

#### 3.7 Data collections and analysis

Plant stand at each experimental unit was estimated by counting the number of plants within one meter length of the two middle rows at 1 to 2 weeks after emergence. Treatment effects on plant height, above ground biomass plant<sup>-1</sup>, crop maturity, harvest index, seed yield, 1000-seed weight of marketable seeds, number and weight of seeds plant<sup>-1</sup>, number of seeds m<sup>-2</sup> and seed quality (percentage marketable seed) of CDC Frontier chickpea were evaluated by collecting and analyzing the data as described below.

Plant height of each plot (experimental unit) at both test sites was collected 20 and 30 days after treatments. Ten or five randomly selected plants from the middle rows of each experimental unit were used to determine plant heights in 2010 and 2011, respectively.

Five randomly selected plants from the middle rows of each experiment unit were harvested 3 - 4 days prior to crop harvest at each test site, and those plants were used to determine the number of seeds, total seed dry weight and the total above ground biomass dry weight per plant. Plant samples were placed in a drier at 39 - 42 °C for about a week until a constant dry weight was reached, prior to determination of biomass and seed dry weights. This information was used to calculate the harvest index and the number of seeds m<sup>-2</sup> as follows.

- Harvest index = (Total seed weight / Total aboveground biomass weight) x 100
- Number of seeds  $m^{-2}$  = number of seeds plant<sup>-1</sup> x number of plants  $m^{-2}$

The maturity rating scale (1-10) developed by Gan et al. (2009b) for the assessment of chickpea crop maturity was used to evaluate crop maturity of each plot before harvest. On this scale, maturity is rated based on percentage colour change of plants in a plot. For example, 10% of plants in a plot that turned to a fully brown colour is equivalent to a rating of 1 whereas 100% plants with brown colour change is equivalent to 10. At crop maturity, after eliminating borders, individual plot areas of 5.4 m<sup>2</sup> (4.5 m x 1.2 m) were harvested with a plot harvester (Wintersteiger Nurserymaster 2000; Salt Lake, Utah, USA.). Harvested samples were placed in a drier at 39 - 42°C for about a week prior to determining the plot seed weights. Total seed yield of individual plots were converted to kg ha<sup>-1</sup>.

Two hundred and fifty seeds, randomly sampled from marketable seeds of individual plots at each test site were used to determine the 1000-seed weight. A sub-sample of 500 g from each individual plot containing green and immature seeds was cleaned using a seed blower. During this cleaning process, immature/green and small seed (<7 mm diameter) were removed. Good appearance (high quality) seeds with >7 mm diameter were considered 'Marketable seeds'. The percentage of marketable seeds and marketable seed yield were calculated as follows.

- Marketable seed (%) = [weight of marketable seeds (g) / 500 g] x 100
- Marketable seed yield (kg ha<sup>-1</sup>) = total seed yield (kg ha<sup>-1</sup>) x marketable seed %

#### 3.8 Statistical analysis

Data analyses were done using Statistical Analysis System (SAS) version 9.2 (SAS Institute Inc., Cary, NC, USA). A mixed model was used analysis the pooled data related to CCC and

Prohexadione Calcium considering site-year, growth stage and plant growth retardants (concentrations of CCC and Prohexadione Calcium collectively considered as plant growth retardants) as fixed factors whereas effects of replication and interaction of growth stage and replication nested in site-year as random factors. Pooled data related to Trinexapac Ethyl was analysed separately from the other two PGR due to the difference of concentrations within two years. Test site, growth stage and Trinexapac Ethyl concentration was considered as fixed factors whereas replication and interaction of growth stage with replication nested in test site were considered as random factors in the mixed model used to analyse the pooled data related to Trinexapac Ethyl. This initial analysis of pooled data revealed that, the treatment effects were significantly dependent on the growing environments (year-site). Therefore, the data of each test site in each year were analysed separately for further presentations. Since this study can be considered as the initial step of evaluating the effects of PGR on pulse crops under western Canadian growing conditions, it would be useful to report behaviour of these chemicals within specific growing conditions such as wet and dry years, irrigated and rain-fed conditions and in the areas that more heat units accumulate within a growing season. In the mixed model used to analysis the data of each year at each test site, growth stages and plant growth retardant concentrations were considered fixed, whereas replicate and growth stages in replicate effects were considered random. Due to the heterogeneity of data distribution among years and among test sites, they were not used as fixed factors in a single mixed model. Treatment effects were declared as significant at p< 0.05 level. Treatment means were compared using Fisher's least significance difference (LSD) test.

In addition, whenever the growth stage x plant growth retardant concentration interaction was significant, a pre-planned (*a priori*) trend analysis was conducted to determine the relationship between concentration of each plant growth retardant and dependent variables separately at each growth stage. Coefficients used for linear and quadratic contrasts were calculated using Proc ILM of the SAS program.

#### 4. Results

#### **4.1 Climatic Conditions**

Due to excessive moisture and cool conditions in 2010, seeding was delayed up to the 20<sup>th</sup> of May at both test sites. In 2010, the crop experienced below average temperature and above average precipitation throughout the growing period (Table 4.1).

Table 4.1: Monthly mean temperatures and precipitation at test sites from April to October in 2010 and 2011

		Brooks						Bow Island					
	Me	an tem	p. °C	Prec	ipitation	(mm)	Mear	temp.	°C	Precipitation (mm)		(mm)	
Month	2010	2011	$LTA^*$	2010	2011	LTA*	2010	2011	${\rm LTA}^*$	2010	2011	LTA*	
April	5.7	3.8	5.8	41.8	20.0	25.4	6.3	4.4	6.0	60.7	38.6	17.7	
May	8.8	10.7	11.3	88.6	25.2	31.8	9.0	11.0	11.6	118.8	50.3	32.7	
June	14.5	14.5	15.6	87.6	81.2	78.0	15.3	15.5	15.4	114.5	69.1	80.4	
July	17.6	18.0	19.6	35.2	31.7	26.5	18.3	18.8	19.4	24.5	28.3	10.9	
Aug.	16.5	18.1	18.3	32.6	25.2	38.0	17.2	19.0	18.4	40.2	12.5	27.4	
Sep.	11.0	15.4	12.5	45.0	1.9	32.2	11.8	16.4	13.2	38.8	5.1	21.3	
Oct.	7.3	6.7	5.8	6.0	13.2	12.9	8.6	7.9	6.7	7.9	31.1	12.3	
Average	11.6	12.5	12.7	48.1	28.3	35.0	12.3	13.3	13.0	57.9	33.6	28.9	

<sup>\*</sup>LTA: long term average (1997 to 2007).

In 2010, the precipitation received between May and September at the Brooks test site was 40% higher than the long term average for that area. The soil moisture levels were much higher at the Bow Island test site, since it received 95% higher precipitation than the long term average during the same period (Table 4.1). The accumulated crop heat units (CHU) from May  $15^{th}$  to the first killing frost (first occurrence of  $\leq$  -2°C at the end of growing period) revealed that the 2010 was a below average cooler growing season (Table 4.2).

The cooler conditions combined with the high soil moisture levels were favourable for the continuous vegetative growth of chickpea in 2010. The occurrence of first frost was recorded on the 18<sup>th</sup> September in 2010 at the Brooks test site that resulted in a cessation of crop growth. The conditions were much better at the Bow Island test site where the first frost occurred almost one month later than at the Brooks site (on 15<sup>th</sup> October, 2010).

Table 4.2: Cumulative crop heat units accumulated during the period from May 15<sup>th</sup> to the first killing frost<sup>@</sup> at Brooks and Bow Island during the 2010 and 2011 growing seasons

Long term average*	Brook		Bow Island 2505		
	2010 2011		2010	2011	
May <sup>#</sup>	164	184	172	191	
June	633	659	673	719	
July	1254	1293	1333	1384	
August	1825	1914	1929	2038	
September	1996	2365	2254	2550	
1 <sup>st</sup> Killing frost!	1996	2365	2460	2680	

<sup>\*</sup>Average of 1971to 2009 period (Source: Alberta Agriculture and Rural development, 2012);

On average, the 2010 growing season was a cooler, wetter and shorter growing season than the long-term average that adversely affected the chickpea crop. Consequently, a high degree of variation was recorded for the growth and yield parameters in 2010. In addition, the crop did not reach the physiological maturity before the occurrence of frost. Therefore, crop maturity data were not collected in 2010.

The crop experienced slightly higher precipitation and cooler temperature conditions compared to the long-term average during the vegetative period of 2011 at both test sites (Table 4.1). However, during the late reproductive period (August and September) both test sites received less rainfall. In addition, higher amounts of crop heat units were accumulated throughout the entire growing season in 2011 compared to the previous year (Table 4.2). Especially, the heat unit accumulation in 2011 was much higher at the end of the season than in 2010. The first frost at the Brooks and Bow Island test sites occurred on September 29<sup>th</sup> and October 14<sup>th</sup> in 2011, respectively. The growing period in 2011 was at least two weeks longer than that of 2010. The occurrence of dry and warm conditions, during the late reproductive period at test sites in 2011, accelerated the crop maturing process. The 2011 growing season was long enough to chickpea to complete the maturity.

<sup>#</sup> Starting date: 15<sup>th</sup> of May; <sup>@</sup>First occurrence of minimum temperature < -2<sup>0</sup>C at the end of growing season.

# 4.2 Evaluation of the influence of two fungicides on the effects of plant growth retardants

Throughout the study period, multiple applications of Pyraclostrobin and Prothioconazole were used to control Ascochyta blight of the crop. The main objective of this study was to examine if there was any difference of the activities of PGR on chickpea growth, yield parameters and maturity when they were applied either with Pyraclostrobin or Prothioconazole. Therefore, the effects of each plant growth retardant on these parameters in the presence of Pyraclostrobin or Prothioconazole were compared using orthogonal contrasts. In addition, sole application of fungicides was compared with each other to determine the possible differences on growth and yield parameters.

The results of the statistical analysis revealed that each plant growth retardant had statistically comparable effects on vegetative growth, yield components, seed yield, seed quality and crop maturity, when they were applied as a mixture with Pyraclostrobin or Prothioconazole (Table 4.3).

Table 4.3: Comparison of the effects of two fungicides on the activity of plant growth retardants on chickpea vegetative growth, yield components, total seed yield, seed quality and maturity; results were presented as probability values from predetermined contrasts

Contrast	Plant height at 30DAT <sup>@</sup> (cm)	Above ground biomass plant <sup>-1</sup>	Number of seeds m <sup>-2</sup>	1000- seed weight (g)	Total seed yield (Kg ha <sup>-1</sup> )	Marketa- ble seed yield (Kg ha <sup>-1</sup> )	Harvest index	Maturity (1 – 10)
		(g)						
Trt.# 1 vs.	0.78 ns	0.42 ns	0.41 ns	0.59 ns	0.97 ns	0.99 ns	0.69 ns	1.0 ns
Trt. 2								
Trt. 5 vs.	0.84 ns	$0.14^{\mathrm{ns}}$	$0.20^{\mathrm{ns}}$	$0.18^{\mathrm{ns}}$	0.66 ns	$0.72^{\mathrm{ns}}$	$0.84^{\rm  ns}$	0.06 ns
Trt. 8								
Trt. 3 vs.	$0.13^{\text{ ns}}$	$0.92^{\mathrm{ns}}$	$0.73^{\text{ ns}}$	0.81 ns	$0.59^{\mathrm{ns}}$	$0.42^{\mathrm{ns}}$	$0.76^{\mathrm{ns}}$	0.22 ns
Trt. 6								
Trt. 4 vs.	0.57 ns	$0.99^{\rm ns}$	$0.98^{\mathrm{ns}}$	$0.17^{\mathrm{ns}}$	0.51 ns	$0.47^{\rm ns}$	1.0 ns	$0.34^{\mathrm{ns}}$
Trt. 7								

ns = non-significant at p≤0.05; <sup>@</sup> Days after treatments; <sup>#</sup> Treatment; Trt.1- (Pyraclostrobin+ Boscalid); Trt.2 − Prothioconazole; Trt.3 - (Pyraclostrobin+ Boscalid) + Prohexadione Calcium; Trt.4 - (Pyraclostrobin+ Boscalid) + Trinexapac Ethyl; Trt.5 - (Pyraclostrobin+ Boscalid) + CCC; Trt.6 - Prothioconazole + Prohexadione Calcium; Trt.7 - Prothioconazole + Trinexapac Ethyl; Trt.8 - Prothioconazole + CCC.

Thus, this study confirmed that the activity of CCC, Prohexadione Calcium and Trinexapac Ethyl on chickpea vegetative growth, yield parameters and maturity were not different with the main systemic fungicides (Pyraclostrobin or Prothioconazole) used to control Ascochyta blight. However, due to the practical difficulty of maintaining a disease free control plot without application of one of these fungicides, it was not possible to evaluate the potential interaction of PGR with these two fungicides from this study. Therefore, a comprehensive study is necessary to evaluate that how the side effects of these fungicides affect on growth retarding properties of PGR. Since the similar concentrations of PGR were evaluated under different growing environments in the main study, treatment effects on chickpea growth and yield parameters were not a matter of interest in this study. However, a summary of treatment effects of this study was included in appendix 2.

# 4.3 Evaluation of the effects of plant growth retardants on vegetative growth, yield components, seed quality and crop maturity of the Kabuli chickpea cultivar CDC Frontier

#### 4.3.1 Analysis of pooled data

Analysis of pooled data of 2010 and 2011 at all locations revealed that the growing environment (site x year) had highly significant effects on all growth and yield parameters (Table 4.4).

Table 4.4: Summary analysis of variance (p values) for the effects of growing environment, growth stage, Prohexadione Calcium and Chlormequat Chloride on growth, seed yield and yield components of the Kabuli chickpea cultivar CDC Frontier

Effect	DF	Plant height at 30DAT <sup>#</sup> (cm)	Above ground biomass (g plant <sup>-1</sup> )	No. of seeds m <sup>-2</sup>	1000-seed weight (g)	Total seed yield (kg ha <sup>-1</sup> )	Marketa- ble seed yield (kg ha <sup>-1</sup> )	Harvest index (%)
Site-year (SY)	5	<0.01**	<0.01**	<0.01**	<0.01**	<0.01**	<0.01**	<0.01**
Growth stage (GS)	2	0.96 <sup>ns</sup>	0.18 ns	0.16 <sup>ns</sup>	$0.90^{\mathrm{ns}}$	0.28 ns	0.59 <sup>ns</sup>	0.10 ns
Plant growth retardant (PGR)	9	0.14 <sup>ns</sup>	0.53 <sup>ns</sup>	0.02*	0.25 ns	<0.01**	<0.01**	0.30 <sup>ns</sup>
SY x GS	10	0.99 ns	0.34 <sup>ns</sup>	0.06 ns	$0.01^{*}$	0.03*	0.64 <sup>ns</sup>	<0.01**
SY x PGR	45	0.88 ns	$0.04^{*}$	<0.01**	<0.01**	$0.04^{*}$	<0.01**	0.03*
GS x PGR	18	0.11 <sup>ns</sup>	0.69 <sup>ns</sup>	<0.01**	<0.01**	<0.01**	<0.01**	<0.01**
SY x GS x PGR	90	0.99 <sup>ns</sup>	0.65 <sup>ns</sup>	<0.01**	0.13 <sup>ns</sup>	<0.01**	0.01**	$0.02^{*}$

ns = non-significant at p $\leq$ 0.05; \*significant at p $\leq$ 0.05; \*significant at p $\leq$ 0.01; \*Days after treatments.

Due to the differences in concentrations of Trinexapac Ethyl between 2010 and 2011, Trinexapac Ethyl treatment was excluded from this combined analysis. Therefore, the effect of Trinexapac Ethyl was separately analysed for each year across the test locations (Table 4.5).

Results of the combined analysis revealed that CCC or Prohexadione Calcium had no significant effect on plant height and above ground biomass plant<sup>-1</sup> at any growth stage across the growing environments. In contrast, CCC and Prohexadione Calcium effects on number of seeds m<sup>-2</sup>, total seed yield and marketable seed yield were significant across the growth stages. Growth stage had no effect on any parameter considered in this study. However, the growth stage with two plant growth retardant concentration interaction was highly significant for seed yield parameters (number of seeds m<sup>-2</sup>, 1000-seed weight, total seed yield, marketable seed yield and harvest index). In addition, the three way interactions of environment (site-year), growth stage and plant growth retardant concentration, were significant for seeds m<sup>-2</sup>, total seed yield, marketable seed yield and harvest index (Table 4.4).

Analysis of pooled data of test sites in each year for Trinexapac Ethyl revealed that 1000- seed weight, total seed yield and harvest index were significantly different among test sites in 2010 (Table 4.5). On average, growth stage had significant effects on yield parameters (number of seeds m<sup>-2</sup>, 1000-seed weight, total seed yield, marketable seed yield and harvest index) in 2010.

The interaction between Trinexapac Ethyl concentration and growth stage was significant for the number of seeds m<sup>-2</sup>, 1000-seed weight, total seed yield, marketable seed yield and harvest index in 2010, suggesting that the effect of Trinexapac Ethyl on yield parameters was mainly dependent upon the concentration and the crop growth stage in that year.

All growth and yield parameters except marketable seed yield were significantly different among the test sites in 2011. In addition, crop growth stage had significant effects on 1000- seed weight, total seed yield and marketable seed yield in 2011. On average, Trinexapac Ethyl significantly affected total seed yield and marketable seed yield.

Table 4.5: Summary of analysis of variance (p values) for the effects of test site (TS), growth stage (GS), concentration Trinexapac Ethyl (TE) on crop growth, seed yield and yield components of the Kabuli chickpea cultivar CDC Frontier in 2010 and 2011

Effect	DF	Plant	Above	No. of	1000-	Total	Marketa-	Harvest
		height at	ground	seeds m <sup>-2</sup>	seed	seed	ble seed	index
		30DAT#	biomass		weight	yield	yield	
		(cm)	(g)		(g)	(kg ha <sup>-1</sup> )	(kg ha <sup>-1</sup> )	
				201				_
TS	2	$0.11^{ns}$	$0.14^{\mathrm{ns}}$	0.11 ns	$0.04^{*}$	$0.03^{*}$	0.17 ns	$0.03^{*}$
GS	2	0.53 ns	0.83 ns	<0.01**	<0.01**	< 0.01**	$0.04^{*}$	<0.01**
TE	4	$0.30^{\mathrm{ns}}$	< 0.01**	$0.75^{\rm ns}$	$0.40^{\mathrm{ns}}$	$0.37^{\mathrm{ns}}$	0.46 ns	$0.74^{\mathrm{ns}}$
TS x GS	4	0.95 ns	0.98 ns	$0.30^{\mathrm{ns}}$	<0.01**	$0.04^{*}$	0.53 ns	$0.03^{*}$
TS x TE	8	$0.82^{\mathrm{ns}}$	$0.41^{\text{ ns}}$	$0.03$ $^*$	$0.80^{\mathrm{ns}}$	$0.14^{\rm  ns}$	$0.18^{\text{ns}}$	$0.16^{\mathrm{ns}}$
GS x TE	8	0.31 ns	$0.06^{\mathrm{ns}}$	<0.01**	$0.03^{*}$	< 0.01**	< 0.01**	$0.05^{\rm ns}$
TS x GS x TE	16	0.96 ns	$0.42^{\mathrm{ns}}$	0.93 ns	$0.59^{\mathrm{ns}}$	$0.17^{\mathrm{ns}}$	0.23 ns	$0.69^{\mathrm{ns}}$
					-2011			
TS	2	< 0.01**	< 0.01**	<0.01**	<0.01**	<0.01**	$0.42^{\mathrm{ns}}$	<0.01**
GS	2	$0.89^{\mathrm{ns}}$	0.66 ns	$0.97^{\mathrm{ns}}$	<0.01**	< 0.01**	< 0.01**	$0.20^{\mathrm{ns}}$
TE	4	$0.07^{\rm ns}$	$0.46^{\mathrm{ns}}$	0.91 ns	$0.14^{\mathrm{ns}}$	< 0.01**	< 0.01**	$0.59^{\mathrm{ns}}$
TS x GS	4	0.61 ns	$0.14^{\mathrm{ns}}$	$0.47^{\mathrm{ns}}$	<0.01**	$0.26^{\mathrm{ns}}$	$0.01^*$	$0.15^{\rm ns}$
TS x TE	8	$0.87^{\rm ns}$	$0.39^{\mathrm{ns}}$	0.25 ns	<0.01**	$0.22^{\mathrm{ns}}$	< 0.01**	0.41 ns
GS x TE	8	$0.70^{\rm  ns}$	$0.36^{\mathrm{ns}}$	0.93 ns	<0.01**	$0.09^{\mathrm{ns}}$	< 0.01**	$0.75^{\rm ns}$
TS x GS x TE	16	0.79 ns	0.58 ns	0.71 ns	0.03*	<0.01**	<0.01**	0.23 ns

ns = non-significant at p $\le$ 0.05; \*significant at p $\le$ 0.05; \*significant at p $\le$ 0.01; \*Days after treatments.

The crop growth stage x Trinexapac Ethyl concentration interaction was significant for 1000-seed weight and marketable seed yield in 2011. In addition, the test site x crop growth stage x Trinexapac Ethyl concentration interaction was significant for 1000-seed weight, total seed yield and marketable seed yield in 2011 (Table 4.5). As indicated in Tables 4.4 and 4.5, most of the parameters considered in this study were significantly different among the growing environments (site x year). Therefore, further discussion on the results is based on each location in each year.

#### 4.3.2 Plant height at 30 days after treatments

Plant height at 30 DAT was not affected by the treatments in each 2010 and 2011 growing seasons. The highest mean plant height was noticed at the Brooks irrigated site in both years (Table 4.6). Plant heights in 2010 and 2011 at each test site were compared by a two sample t test. The result revealed that the plant heights of 2010 and 2011 were significantly different (at p

 $\leq$  0.01) at all three test sites. The favourable growing conditions experienced throughout the 2010 growing season would be the reason for taller plants observed across test sites in that year.

Table 4.6: Mean plant height at 30 days after treatments of the Kabuli chickpea cultivar CDC Frontier at three test sites in 2010 and 2011

	Bow Island	Brooks Rain-fed	Brooks Irrigated
		2010	
Mean plant height (cm)	65	69	76
Standard error	0.6	0.8	0.8
CV %	14	20	17
		2011	
Mean plant height (cm)	53	47	59
Standard error	0.4	0.2	0.3
CV %	12	8	9

#### 4.3.3 Results of the 2010 field experiments

Due to the occurrence of extreme growing conditions (cooler and wetter soil conditions) in 2010, abnormal and uneven chickpea growth was observed within and among test sites. Consequently higher levels of variation within treatments occurred for most of the traits measured in this study. Except for the plant height and 1000-seed weight, the coefficient of variation of the other traits was higher than 30% at all test locations in 2010 (Table 4.7).

Table 4.7: Means and coefficient of variations (CV) of the traits which had a CV higher than 30% of the Kabuli chickpea cultivar CDC Frontier at three test sites in 2010

	Test site	Mean	Std. error	CV%
	Bow Island	24.5 g	0.67	41.8
Above ground biomass	Brooks rain-fed	22.3 g	0.55	37.9
	Brooks irrigated	29.4 g	0.66	34.3
	Bow Island	1357	37.8	45.7
Number of seeds m <sup>-2</sup>	Brooks rain-fed	1254	42.4	55.5
	Brooks irrigated	819	50.5	95.9
	Bow Island	3105 kg ha <sup>-1</sup>	110.6	54.5
Total seed yield	Brooks rain-fed	3422 kg ha <sup>-1</sup>	124.4	55.6
	Brooks irrigated	1237 kg ha <sup>-1</sup>	87.6	108.4
	Bow Island	1778 kg ha <sup>-1</sup>	125.3	107.4
Marketable seed yield	Brooks rain-fed	1850 kg ha <sup>-1</sup>	139.5	115.4
	Brooks irrigated	307 kg ha <sup>-1</sup>	39.5	195.3
	Bow Island	0.34	0.01	48.4
Harvest index	Brooks rain-fed	0.34	0.01	56.1
	Brooks irrigated	0.14	0.01	108.8

Therefore, presentation of the results of statistical analysis was limited to 1000-seed weight in 2010. Even though a much lower coefficient of variation (< 30%) was obtained for plant height at 30 DAT, the plant height variation was not due to the treatment effects.

#### 4.3.3.1 1000- seed weight of marketable seeds

The crop growth stage had a significant effect on 1000-seed weight regardless the PGR type and concentration at the Bow Island test site. In addition, the interaction between PGR concentration and growth stage was also significant for 1000-seed weight at Bow Island (Table 4.8).

Table 4.8: Summary of analysis of variance (p values) for the effects of growth stage and plant growth retardant on plant height at 30 days after treatments and 1000-seed weight of marketable seeds of the Kabuli chickpea cultivar CDC Frontier at three test sites in 2010

		Bow 1	Bow Island		Rain-fed	Brooks Irrigated	
Factor	DF	Plant	1000-seed	Plant	1000-seed	Plant	1000-seed
		height at	weight	height at	weight	height at	weight
		30 DAT!		30 DAT		30 DAT	
Growth stage	2	0.60 <sup>ns</sup>	0.03*	0.98 <sup>ns</sup>	0.39 <sup>ns</sup>	0.99 <sup>ns</sup>	<0.01**
PGR concentration	12	$0.90^{ns}$	$0.13^{ns}$	$0.09^{ns}$	$0.35^{ns}$	$0.61^{ns}$	$0.56^{\text{ns}}$
Growth stage x PGR	24	$0.86^{\mathrm{ns}}$	$0.03^*$	$0.87^{ns}$	$0.11^{ns}$	$0.96^{\text{ns}}$	$0.10^{ns}$

DAT' = Days after treatment; ns = non-significant at  $p \le 0.05$ ; \*significant at  $p \le 0.05$ ; \*significant at  $p \le 0.01$ ; PGR= Plant growth retardants.

The largest 1000-seed weight (311 g) at Bow Island was obtained at 30 DAF treatments and this was significantly higher than that of 20 DAF (278 g) and 10 DAF (268 g). Application of Prohexadione Calcium at 3000 mg L<sup>-1</sup> at 10 DAF reduced 1000-seed weight significantly compared to the untreated control and those treated with 750 and 4500 mg L<sup>-1</sup> at the same growth stage (Table 4.9). This reduction at 3000 mg L<sup>-1</sup> contributed for significant linear and quadratic trends at 10 DAF. Application of Prohexadione Calcium was not statistically significant on 1000-seed weight at 20 DAF at Bow Island. However, a significant linear trend was noticed at the same growth stage (Table 4.9).

Irrespective of crop growth, increasing concentration of CCC from 1000 to 6000 mg L<sup>-</sup>had no significant effect on 1000-seed weight at Bow Island. Application of Trinexapac Ethyl at 10 DAF had no significant effect on 1000-seed weight, but contributed for a significant negative

linear trend on 1000-seed weight (Table 4.9). Trinexapac ethyl applied at 8,333 mg L<sup>-1</sup> at 20 DAF significantly increased (30%) 1000-seed weight compared to the untreated control.

Table 4.9: Effect of concentration of three plant growth retardants applied at three crop growth stages on 1000-seed weight of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2010

	PGR!	Growth	stage at PGR app	olied (GS)	
Type	Concentration (mg L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
			1000-seed	weight (g)	
	Control (0)	275	239	310	275
Duchanadiana	750	299	297	316	304
Prohexadione Calcium (PC)	1500	238	275	318	277
	3000	184	273	327	261
	4500	271	300	320	297
Mean of GS for P	rohexadione Calcium	248	286	320	285
	Control (0)	275	239	310	275
Chlamman	1000	281	256	331	289
Chlorida (CCC)	2000	256	314	272	281
Chloride (CCC)	4000	318	256	322	299
	6000	299	274	362	312
Mean of GS for C	hlormequat Chloride	289	275	322	295
	Control (0)	275	239	310	275
Trimeren - Etherl	2080	241	271	326	279
Trinexapac Ethyl	4167	303	278	291	291
(TE)	8333	263	310	295	289
	12498	252	267	252	257
Mean of GS for T	rinexapac Ethyl	265	282	291	279
-	stage across all PGR	269	273	311	284
LSD (p=0.05) for	Growth stage (GS)	31			
for	PGR	-			
for	GS x PGR	63			
Coefficient of vari	iation (%)	24			
	Probab	oility values for trend	•		
PC –Linear		<0.01**	0.02*	0.95 ns	
PC-Quadratic		<0.01**	0.28 ns	0.66 ns	
CCC- Linear		$0.44^{\text{ns}}$	0.27 ns	0.71 <sup>ns</sup>	
CCC-Quadratic		0.26 ns	0.69 ns	$0.17^{\rm ns}$	
TE-Linear		<0.01**	0.17 ns	$0.61^{\text{ns}}$	
TE-Quadratic		$0.15^{ns}$	0.31 <sup>ns</sup>	$0.32^{\mathrm{ns}}$	

PGR = Plant growth retardants;  $^{\text{@}}DAF = Days$  after  $1^{\text{st}}$  flowering;  $^{\text{ns}}$  non-significant at p $\leq$ 0.05; \*significant at p $\leq$ 0.05 level; \*\*significant at p $\leq$ 0.01 level.

Irrespective of growth stage and concentration, none of the growth retardants had a significant effect on 1000-seed weight of CDC Frontier chickpea at the Brooks rain-fed test site in 2010

(Table 4.8). The mean 1000-seed weight observed at the Brooks rain-fed site in 2010 was 284g  $\pm 5$ .

On average, growth stage at PGR application time had a significant effect on 1000-seed weight at the Brooks irrigated site in 2010 (Table 4.8). Treatments applied at 30 DAF resulted in a significant reduction in 1000-seed weight compared to that of 10 DAF, but the effects of treatments applied at 10 DAF and 20 DAF on 1000-seed weight was comparable at the Brooks irrigated test site (Figure 4.1).

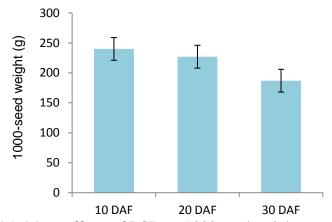


Figure 4.1: Mean effects of PGR on 1000-seed weight across different growth stages of the Kabuli chickpea cultivar CDC Frontier under irrigated conditions at Brooks in 2010. Error bars represent 1.96 times of standard error for each direction.

#### 4.3.4 Results of the 2011 field experiments

### 4.3.4.1 Above ground biomass plant<sup>-1</sup>

Above ground biomass plant<sup>-1</sup> differed significantly among growth stages, however the effects of PGR or the interaction of PGR and growth stage at Bow Island had no significant effect on that (Table 4.10).

The mean above ground biomass plant<sup>-1</sup> at the Brooks rain-fed and irrigated sites were 19 g  $\pm 0.2$  and 26 g  $\pm 0.4$ , respectively. Treatment effects were not significant on above ground biomass plant<sup>-1</sup> at both rain-fed and irrigated test sites at Brooks in 2011 (Table 4.10).

Table 4.10: Summary of analysis of variance (p values) for the effects of crop growth stage of treatment applied (GS), different concentrations of plant growth retardants (PGR) and their interactions on growth and yield parameters of the Kabuli chickpea cultivar CDC Frontier at three test sites in 2011

Effect	DF	Plant height at 30DAT <sup>#</sup> (cm)	Above ground biomass plant <sup>-1</sup> (g)	No. of seeds m <sup>-2</sup>	1000 seed weight (g)	Total seed Yield (kg ha <sup>-1</sup> )	Market- able seed yield (kg ha <sup>-1</sup> )	Harvest index	Maturity (1 – 10)
					Bow Is	sland			
GS	2	0.63 <sup>ns</sup>	$0.04^{*}$	0.21 ns	<0.01**	$0.06^{\text{ns}}$	<0.01**	$0.03^{*}$	<0.01**
PGR	14	$0.99^{ns}$	$0.14^{ns}$	0.11 ns	<0.01**	<0.01**	<0.01**	0.11 <sup>ns</sup>	<0.01**
GS x PGR	28	$0.29^{ns}$	$0.33^{ns}$	0.18 ns	<0.01**	<0.01**	<0.01**	$0.22^{ns}$	$0.36^{\text{ns}}$
					Brooks Ra	ain-fed			
GS	2	0.71 <sup>ns</sup>	0.16 <sup>ns</sup>	<0.01**	<0.01**	$0.06^{\text{ns}}$	$0.06^{\text{ns}}$	$0.22^{ns}$	0.20 <sup>ns</sup>
PGR	14	$0.44^{\text{ns}}$	$0.16^{\text{ns}}$	$0.15^{\text{ns}}$	<0.01**	$0.38^{\text{ns}}$	$0.26^{\text{ns}}$	$0.28^{ns}$	$0.36^{\text{ns}}$
GS x PGR	28	$0.38^{\text{ns}}$	$0.84^{ns}$	$0.21^{ns}$	$0.22^{ns}$	<0.01**	<0.01**	$0.02^*$	$0.07^{\text{ns}}$
					Brooks	Irrigated			
GS	2	$0.82^{ns}$	$0.47^{ns}$	$0.60^{ns}$	$0.24^{\text{ns}}$	$0.12^{ns}$	0. 79 <sup>ns</sup>	0.58 <sup>ns</sup>	0.25 <sup>ns</sup>
PGR	14	$0.16^{\text{ns}}$	$0.09^{ns}$	$0.22^{\text{ns}}$	0.26 <sup>ns</sup>	<0.01**	<0.01**	0.53 <sup>ns</sup>	$0.78^{\text{ns}}$
GS x PGR	28	$0.79^{ns}$	$0.55^{\text{ns}}$	$0.94^{\text{ns}}$	$0.28^{\text{ns}}$	$0.53^{\text{ns}}$	$0.42^{ns}$	0.81 <sup>ns</sup>	<0.01**

<sup>&</sup>lt;sup>#</sup> Days after treatments; ns = non-significant at p≤0.05; \*\*significant at p≤0.05; \*\*significant at p≤0.01.

Above ground biomass plant<sup>-1</sup> was significantly higher at 10 DAF compared to that of at 30 DAF treatment application at Bow Island (Figure 4.2). On average, PGR reduced the above ground biomass plant<sup>-1</sup> when applied at much later stages after flowering irrespective of the type and concentration used.

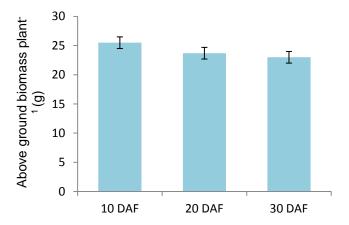


Figure 4.2: Mean effects of PGR on above ground biomass plant<sup>-1</sup> across different growth stages of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011. Error bars represent 1.96 times of standard error for each direction.

## 4.3.4.2 Number of seeds per unit area (m<sup>-2</sup>)

On average, the highest number of seeds  $m^{-2}$  (1365 ± 22) was obtained at Bow Island in 2011. About 917 ± 15 and 1213 ± 25 of number of seeds  $m^{-2}$  were noticed at the Brooks rain-fed and irrigated test sites, respectively. PGR and crop growth stages did not significantly affect the number of seeds  $m^{-2}$  at the Bow Island and Brooks irrigated test sites (Table 4.10). On average, the application of PGR at 10 and 20 DAF produced comparable numbers of seeds  $m^{-2}$  at the Brooks rain-fed test site. In addition, number of seeds  $m^{-2}$  obtained at 10 and 20 DAF at the Brooks rain-fed test site was significantly higher than that of 30 DAF (Figure 4.3).

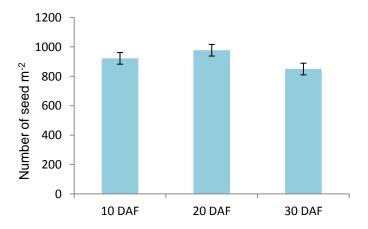


Figure 4.3: Mean effects of PGR on number of seeds m<sup>-2</sup> across different growth stages of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011. Error bars represent 1.96 times of standard error for each direction.

#### 4.3.4.3 1000-seed weight of marketable seeds

On average, PGR applied at 10 DAF and 20 DAF produced a significantly higher 1000-seed weight compared to that of 30 DAF at the Bow Island test site (Table 4.11). The significant interaction between growth stage and plant growth retardant concentration suggested that the effects of plant growth retardant concentration on 1000-seed weight varied with the growth stage of the crop at Bow Island (Table 4.10).

Table 4.11: Effect of concentration of three plant growth retardants applied at three crop growth stages on 1000-seed weight of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011

	PGR <sup>!</sup>	Growth	stage at PGR ap	plied (GS)	
Type	Concentration (mg L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
			1000-seed	weight (g)	
	Control (0)	358	356	354	356
Prohexadione	750	377	385	361	374
	1500	386	390	367	381
Calcium (PC)	3000	403	398	364	388
	4500	395	393	371	386
Mean of GS for Pr	ohexadione Calcium	390	392	366	382
	Control (0)	351	353	352	352
Chlormoquat	1000	354	354	342	350
Chloride (CCC)	2000	359	354	345	353
Chloride (CCC)	4000	347	350	354	350
	6000	350	358	352	353
Mean of GS for C	hlormequat Chloride	353	354	348	352
	Control (0)	355	359	351	355
Trimovomoo Ethyd	1000	382	383	367	377
Trinexapac Ethyl	2000	386	397	369	384
(TE)	4000	386	403	371	387
	6000	384	417	364	388
Mean of GS for Tr	rinexapac Ethyl	384	400	368	384
Mean for growth s	tage across all PGR	372	377	359	369
LSD (p=0.05) for	Growth stage (GS)	8			
for	PGR	7			
for	GS x PGR	13			
Coefficient of vari	ation (%)	9.			
		lity values for trend	analysis		
PC –Linear		<0.01**	<0.01**	<0.01**	
PC-Quadratic		<0.01**	<0.01**	$0.31^{\rm ns}$	
CCC- Linear		$0.44^{ns}$	$0.71^{\rm ns}$	$0.13^{\rm ns}$	
CCC-Quadratic		$0.67^{ns}$	$0.49^{\rm ns}$	$0.33^{\rm ns}$	
TE-Linear		<0.01**	<0.01**	$0.06^{\text{ns}}$	
TE-Quadratic		<0.01**	<0.01**	<0.01**	

PGR = Plant growth retardants;  $^{\text{@}}$ DAF = Days after 1<sup>st</sup> flowering;  $^{\text{ns}}$ non-significant at p $\leq$ 0.05; \*significant at p $\leq$ 0.05 level; \*\*significant at p $\leq$ 0.01 level.

Application of Prohexadione Calcium (750 to 4500 mg L<sup>-1</sup>) at 10 DAF increased 1000-seed weight significantly compared to the untreated control (Table 4.11). The highest 1000-seed weight with Prohexadione Calcium at 10 DAF (403 g) was noticed at 3000 mg L<sup>-1</sup>. The effect of Prohexadione Calcium on 1000-seed weight at 20 DAF application was statistically comparable with that of 10 DAF. All Prohexadione Calcium treatments applied at 20 DAF significantly increased 1000-seed weight with the maximum increase (398 g) at 3000 mg L<sup>-1</sup>. Compared to the earlier application stages (10 and 20 DAF), the effect of Prohexadione Calcium was much lower at 30 DAF. Only 1500 and 4500 mg L<sup>-1</sup> caused significant increases of 1000-seed weight at 30 DAF application. Increasing concentrations of Prohexadione Calcium applied at 10 and 20 DAF resulted in a significant linear increase in 1000-seed weight up to 3000 mg L<sup>-1</sup> and further increase in concentration tended to have negative effects on 1000-seed weight. Consequently, the quadratic effect was also significant at 10 and 20 DAF. Only the linear effect on 1000-seed weight was significant (p<0.01) at 30 DAF (Table 4.11).

Irrespective of concentration and growth stage of application, CCC had no significant effect on 1000-seed weight at Bow Island. Applications of Trinexapac Ethyl (1000 - 6000 mg L<sup>-1</sup>) increased 1000-seed weight significantly compared to the respective untreated controls at all three application stages. At 10 DAF, Trinexapac Ethyl at 1000 mg L<sup>-1</sup> increased 1000- weight by 7.6% (382 g vs. 355 g) over the untreated control, but further increase in concentration had no significant effect on seed size. The maximum positive effects of Trinexapac Ethyl were obtained at 20 DAF application, where the increasing concentrations caused consistent significant increase of 1000-seed weight. Consequently, the largest seed size was noticed at 6000 mg L<sup>-1</sup> (417 g) at the Bow Island test site (Table 4.11). Comparable with the 10 DAF application, the increasing concentration had no significant effect on 1000-seed weight at the 30 DAF application. Even though the effect of increasing Trinexapac Ethyl concentration was not significant at the 10 DAF and 30 DAF applications, the quadratic trend was significant at all three application stages at Bow Island. In addition, the effect of increasing Trinexapac Ethyl concentrations on 1000-seed weight contributed to significant linear trends at 10 DAF and 20 DAF.

Growth stage and PGR had a significant effect ( $p \le 0.01$ ) on 1000-seed weight at the Brooks rainfed test site, but the growth stage x PGR interaction was not significant (Table 4.10). Application of PGR at 10 DAF caused a highly significant reduction of 1000-seed weight which was 3.6%

(416 g vs. 431 g) and 3.4% (416 g vs. 430 g) lower than that at 20 DAF and 30 DAF, respectively (Figure 4.4). 1000-seed weights were not significantly different at 20 and 30 DAF treatment applications.

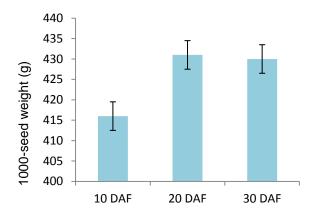


Figure 4.4: Mean effects of PGR on 1000-seed weight across different growth stages of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011. Error bars represent 1.96 times of standard error for each direction.

On average, different concentrations of Prohexadione Calcium and CCC had no significant effect on 1000-seed weight at the Brooks rain-fed test site (Table 4.12). On the other hand the application of Trinexapac Ethyl significantly reduced 1000-seed weight over the untreated control regardless the crop growth stage. However, there were no differences in 1000-seed weight among different concentrations (1000 to 6000 mg L<sup>-1</sup>) of Trinexapac Ethyl (Table 4.12).

Table 4.12: Effect of concentration of three plant growth retardants on 1000-seed weight of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011

Plant growth retardant	Concentration (mg L <sup>-1</sup> )	1000-seed weight (g)
	Control (0)	432 <sup>A</sup>
	750	428 <sup>A-C</sup>
Prohexadione Calcium	1500	426 <sup>A-C</sup>
	3000	432 <sup>A</sup>
	4500	429 <sup>AB</sup>
	Control (0)	431 <sup>A</sup>
	1000	432 <sup>A</sup>
Chlormequat Chloride	2000	425 <sup>A-C</sup>
_	4000	429 <sup>AB</sup>
	6000	$430^{AB}$
	Control (0)	425 <sup>A-C</sup>
	1000	421 <sup>B-D</sup>
Trinexapac Ethyl	2000	419 <sup>CD</sup>
	4000	$414^{\mathrm{D}}$
	6000	414 <sup>D</sup>

Note: Values of 1000-seed weight followed by the same letter are not significantly different based on LSD at P = 0.05 level.

The lowest mean 1000-seed weight among test sites (360 g  $\pm$ 1) in 2011 was found at the Brooks irrigated site while 1000-seed weight was unaffected by treatments at the Brooks irrigated site (Table 4.10).

#### 4.3.4.4 Total seed yield

PGR concentrations and their interaction with growth stage had a highly significant effect on total seed yield at the Bow Island test site (Table 4.10). On average, an increasing concentrations of Prohexadione Calcium (1500, 3000 and 4500 mg L<sup>-1</sup>) and Trinexapac Ethyl (4000 and 6000 mg L<sup>-1</sup>) reduced total seed yield significantly compared to the untreated control. However, the significant growth stage x PGR interaction for total seed yield indicated that the effect of each plant growth retardant depends on the concentration and the growth stage at Bow Island. The effects of the increasing concentrations of Prohexadione Calcium and Trinexapac Ethyl on total seed yield followed the linear and quadratic models at 10 and 20 DAF (Table 4.13).

Prohexadione Calcium applied at 10 and 20 DAF growth stages significantly reduced total seed yield (11 to 22%) compared to the untreated control except at the lowest (750 mg L<sup>-1</sup>) concentration. It had no significant effect on total seed yield when applied at 30 DAF (Table 4.13). The differences in total seed yield among different concentrations were not significant at both 10 and 20 DAF. The adverse effects of Prohexadione Calcium on total seed yield resulted in a significant negative linear trend at 10 DAF and significant linear and quadratic trends at 20 DAF.

The application of CCC at 1000 to 6000 mg L<sup>-1</sup> concentrations had no significant effect on total seed yield at any growth stage at Bow Island. Trinexapac Ethyl applied at 4000 and 6000 mg L<sup>-1</sup> at 10 DAF significantly reduced total seed yield compared to the respective untreated control and the application at low concentration (1000 mg L<sup>-1</sup>). The total seed yield that was obtained with the application of 6000 mg L<sup>-1</sup> Trinexapac Ethyl at 20 DAF (5024 kg ha<sup>-1</sup>) was significantly lower than that of untreated control or at lower concentrations (1000 and 2000 mg L<sup>-1</sup>; Table 4.13). Consequently, the linear effect of the Trinexapac Ethyl concentrations at 10 and 20 DAF was significant at Bow Island. In contrast, Trinexapac Ethyl applied at 30 DAF had no significant effect on total seed yield. The quadratic effect of the Trinexapac Ethyl treatment was significant only at 30 DAF application at Bow Island (Table 4.13).

Table 4.13: Effect of concentration of three plant growth retardants applied at three crop growth stages on total seed yield of the Kabuli chickpea cultivar CDC Frontier under at Bow Island in 2011

	PGR <sup>!</sup>		stage at PGR app	lied (GS)	
Type	Concentration (mg L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
	-		Total seed y	ield (kg ha <sup>-1</sup> )	
	Control (0)	6610	6167	5893	6223
D., 1, 4'	750	6296	5607	5966	5956
Prohexadione	1500	5875	5363	6086	5775
Calcium (PC)	3000	5534	4815	5913	5421
	4500	5286	5121	5878	5428
Mean of GS for Pro	ohexadione Calcium	5748	5227	5961	5645
	Control (0)	5815	6030	6130	5992
Chlormaguet	1000	6323	6200	6229	6251
Chlormequat Chloride (CCC)	2000	6087	6143	5962	6064
Cilioride (CCC)	4000	5609	6220	6007	5945
	6000	6179	6086	6169	6145
Mean of GS for Ch	lormequat Chloride	6049	6162	6092	6101
	Control (0)	5685	6201	6172	6019
Trinavanaa Ethyl	1000	5629	5995	6554	6059
Trinexapac Ethyl (TE)	2000	5135	5866	6546	5849
(1E)	4000	4616	5585	6321	5507
	6000	4689	5024	6264	5326
Mean of GS for Tr	inexapac Ethyl	5017	5618	6421	5685
Mean for growth st	tage across all PGR	5691	5762	6139	5864
LSD (p=0.05) for 0	Growth stage (GS)	-			
for 1	PGR	417			
for	GS x PGR	722			
Coefficient of varia	ation (%)	13.7			
	Probabi	lity values for trend	analysis		
PC -Linear		<0.01**	<0.01**	$0.69^{ns}$	
PC-Quadratic		$0.25^{\mathrm{ns}}$	$0.02^*$	$0.39^{ns}$	
CCC- Linear		$0.96^{\mathrm{ns}}$	$0.85^{\mathrm{ns}}$	$0.89^{ns}$	
CCC-Quadratic		$0.33^{\text{ns}}$	$0.31^{\text{ns}}$	$0.29^{\rm ns}$	
TE-Linear		<0.01**	<0.01**	$0.48^{ns}$	
TE-Quadratic		$0.28^{\text{ns}}$	$0.41^{\text{ns}}$	$0.02^*$	

 $^{1}$ PGR = Plant growth retardants;  $^{(0)}$ DAF = Days after 1<sup>st</sup> flowering;  $^{ns}$ non-significant at p<0.05; \*significant at p<0.05 level; \*\*significant at p<0.01 level.

The growth stage x PGR interaction for total seed yield (p <0.01) was significant at the Brooks rain-fed site although the main effects were not significant (Table 4.10).

The effects of the application of Prohexadione Calcium from 750 to 4500 mg  $L^{-1}$  and CCC from 1000 to 6000 mg  $L^{-1}$  on total seed yield were not significant across the three crop growth stages at the Brooks rain-fed site (Table 4.14). Trinexapac Ethyl applied at 4000 mg  $L^{-1}$  at 20 DAF and 6000 mg  $L^{-1}$  at 30 DAF significantly reduced total seed yield over the respective untreated controls. In addition, the increasing concentrations of Trinexapac Ethyl from 0 to 6000 mg  $L^{-1}$ 

resulted in significant linear and quadratic effects on total seed yield when applied at 10 and 20 DAF. Increasing concentration of the same compound caused a significant linear reduction in total seed yield when applied at 30 DAF (Table 4.14).

Table 4.14: Effect of concentration of three plant growth retardants applied at three crop growth stages on total seed yield of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011

	PGR <sup>!</sup>	Growth	stage at PGR app	olied (GS)	
Type	Concentration (mg L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
• • • • • • • • • • • • • • • • • • • •			Total seed yi	ield (kg ha <sup>-1</sup> )	
	Control (0)	5482	4978	5014	5158
D 1 1'	750	5185	5167	5063	5138
Prohexadione Calcium (PC)	1500	5277	5245	4861	5128
Calcium (PC)	3000	5317	5261	4892	5157
	4500	5468	5055	4958	5160
Mean of GS for Prol	nexadione Calcium	5312	5182	4943	5146
	Control (0)	5521	5479	4729	5243
Chlamaayat	1000	5336	5294	4773	5134
Chlorida (CCC)	2000	5126	5588	4682	5132
Chloride (CCC)	4000	5306	5763	4614	5228
	6000	5169	5601	4750	5173
Mean of GS for Chle	ormequat Chloride	5234	5561	4705	5167
	Control (0)	5091	5503	5570	5388
Trinarianaa Ethiil	1000	5338	5593	5152	5361
Trinexapac Ethyl	2000	5282	5578	5322	5394
(TE)	4000	5013	4909	5140	5021
	6000	4718	5378	4988	5028
Mean of GS for Trin	nexapac Ethyl	5088	5364	5150	5201
Mean for growth sta	ge across all PGR	5242	5359	4967	5190
LSD (p=0.05) for Gr	rowth stage (GS)	-			
for Po	GR	-			
for G	S x PGR	535			
Coefficient of variat	ion (%)	12.2			
	* *	ty values for trend	l analysis		
PC –Linear		$0.62^{\text{ns}}$	0.83 ns	$0.55^{\rm ns}$	
PC-Quadratic		$0.15^{\rm ns}$	0.21 ns	$0.42^{\mathrm{ns}}$	
CCC- Linear		$0.13^{\text{ ns}}$	$0.13^{\text{ ns}}$	0.81 ns	
CCC-Quadratic		$0.31^{\text{ ns}}$	$0.42^{\text{ ns}}$	$0.47^{\text{ ns}}$	
TE-Linear		<0.01**	$0.09^{\text{ ns}}$	$0.02$ $^*$	
TE-Quadratic		$0.04$ $^*$	$0.29^{\text{ ns}}$	$0.62^{\rm ns}$	

 $^{1}$ PGR = Plant growth retardants;  $^{@}$ DAF = Days after 1<sup>st</sup> flowering;  $^{ns}$ non-significant at p≤0.05; \*significant at p<0.05 level; \*\*significant at p<0.01 level.

Total seed yield at the Brooks irrigated test site was significantly different among different concentrations of PGR while the effects of growth stage and the interaction of growth stage with PGR were not significant (Table 4.10). Regardless the crop growth stage, the highest

Prohexadione Calcium concentration (4500 mg L<sup>-1</sup>) significantly reduced total seed yield (6%) compared to the untreated control whereas the effect of the other concentrations was not significant (Table 4.15).

Table 4.15: Effect of concentration of three plant growth retardants on total seed yield of the Kabuli chickpea cultivar CDC Frontier under irrigated conditions at Brooks in 2011

Plant growth retardant	Concentration	Total seed yield (kg ha <sup>-1</sup> )
	Control (0)	5285 <sup>A-C</sup>
	750	5227 <sup>A-D</sup>
Prohexadione Calcium	1500	$5039^{ ext{B-E}}$
	3000	5014 <sup>C-E</sup>
	4500	4963 <sup>DE</sup>
	Control (0)	5205 <sup>A-D</sup>
	1000	5282 <sup>A-C</sup>
Chlormequat Chloride	2000	5275 <sup>A-C</sup>
	4000	5295 <sup>A-C</sup>
	6000	5421 <sup>A</sup>
	Control (0)	5206 <sup>A-D</sup>
	1000	$5327^{\mathrm{AB}}$
Trinexapac Ethyl	2000	$5034^{\text{B-E}}$
	4000	4942 <sup>DE</sup>
	6000	4851 <sup>E</sup>

Note: Values of total seed yield assigned by the same are not significantly different based on LSD at p = 0.05 level.

There were no significant effects of CCC application at 1000 to 6000 mg L<sup>-1</sup> across crop growth stages on total seed yield at the Brooks irrigated site. The average total seed yield obtained with the application of Trinexapac Ethyl at 6000 mg L<sup>-1</sup> was 4851 kg ha<sup>-1</sup> which was significantly lower than the untreated control (5206 kg ha<sup>-1</sup>). In addition, the total seed yields obtained with the application of 4000 and 6000 mg L<sup>-1</sup> Trinexapac Ethyl were significantly lower than that of 1000 mg L<sup>-1</sup> concentration at the Brooks irrigated site (Table 4.15).

#### 4.3.4.5 Marketable seed yield

PGR, crop growth stage and the interaction of crop growth stage and PGR had highly significant effects on marketable seed yield at the Bow Island test site in 2011 (Table 4.10). The marketable seed yield measured from PGR application at 10 DAF was significantly lower than that of 30 DAF at Bow Island. On average, Prohexadione Calcium and Trinexapac Ethyl caused significant reduction in marketable seed yield compared to the untreated and CCC treated plants (Table

4.16). In addition, the plant growth retardant concentration by growth stage interaction for marketable seed yield was also significant at the Bow Island site in 2011 (Table 4.10).

Table 4.16: Effect of concentration of three plant growth regulators applied at three crop growth stages on marketable seed yield of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011

	PGR <sup>!</sup>		age at PGR appli	ed (GS)	
Type	Concentration (mg.L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
	-		Marketable see	d yield (kg ha <sup>-1</sup> )	
	Control (0)	5903	5521	5374	5599
D 1 1'	750	4965	4539	5360	4955
Prohexadione Calcium (PC)	1500	4318	4341	5409	4689
Calcium (PC)	3000	3743	3804	5383	4310
	4500	3502	4313	30 DAF    yield (kg ha <sup>-1</sup> )   5374	4384
Mean of GS for Prol	nexadione Calcium	4132	4249	5373	4585
	Control (0)	5060	5569	5616	5415
Chlormoquat	1000	5662	5702	5785	5716
Chlormequat Chloride (CCC)	2000	5477	5684	5391	5517
Chioride (CCC)	4000	4995	5667	5315	5326
	6000	5467	5683	5582	5577
Mean of GS for Chlo	ormequat Chloride	5400	5684	5518	5534
	Control (0)	4948	5824	5738	5503
Trimarrama a Ethril	1000	4465	5378	6021	5288
Trinexapac Ethyl	2000	3813	5272	6072	5052
(TE)	4000	0 4465 5378 6021 0 3813 5272 6072 0 3075 4734 5851	4553		
	6000	2435	4152	5913	4167
Mean of GS for Trin	nexapac Ethyl	3447	4884	5964	4765
Mean for growth sta	ge across all PGR	4522	5079	5610	5070
LSD (p=0.05) for G	rowth stage (GS)	504			
for Po	GR	562			
for G	S x PGR	973			
Coefficient of variat	ion (%)	16.7			
	Probabili	ity values for trend	analysis		
PC –Linear		<0.01**	<0.01**	$0.89^{ns}$	
PC-Quadratic		$0.02^*$	<0.01**		
CCC- Linear		$0.93^{ns}$	$0.68^{\text{ns}}$	$0.36^{\rm ns}$	
CCC-Quadratic		$0.94^{\text{ns}}$	$0.65^{\text{ns}}$	$0.16^{\rm ns}$	
TE-Linear		<0.01**	<0.01**		
TE-Quadratic		$0.55^{\rm ns}$	$0.99^{\rm ns}$	$0.16^{ns}$	

PGR = Plant growth retardants; <sup>®</sup>DAF = Days after 1<sup>st</sup> flowering; <sup>ns</sup>non-significant at p≤0.05; \*significant at p<0.05 level; \*\*significant at p<0.01 level.

Application of Prohexadione Calcium at 10 DAF resulted in significant marketable yield reduction (16% to 41%) compared to the untreated check, except at 750 mg L<sup>-1</sup> (Table 4.16). Marketable seed yields at higher Prohexadione Calcium concentrations (3000 and 4500 mg L<sup>-1</sup>) were also significantly lower than that of 750 mg L<sup>-1</sup> at this application stage. The Prohexadione Calcium concentrations had significant linear and quadratic effects on marketable seed yield at

10 DAF application. This negative effect of Prohexadione Calcium was continued when the same concentrations were applied at 20 DAF.

At 20 DAF, depending upon the concentration, Prohexadione Calcium (750 to 4500 mg L<sup>-1</sup>) significantly reduced marketable seed yield by17% to 31% over the untreated control. However, the differences among concentrations were not significant at 20 DAF. Comparable with 10 DAF, the linear and quadratic trends were also significant at 20 DAF. Prohexadione Calcium had no significant effect on marketable seed yield at 30 DAF.

CCC (1000 to 6000 mg L<sup>-1</sup>) had no significant effect on marketable seed yield at any growth stage at the Bow Island test site (Table 4.16). Except at the lowest concentration (1000 mg L<sup>-1</sup>), Trinexapac Ethyl applied at 10 DAF significantly reduced marketable seed yield compared to the untreated control. The marketable seed yield under 6000 mg L<sup>-1</sup> Trinexapac Ethyl (2435 kg ha<sup>-1</sup>) at 10 DAF was the lowest at Bow Island in 2011 (Table 4.16). The strong negative effect of Trinexapac Ethyl on marketable seed yield at Bow Island was also observed at 20 DAF applications. Due to this adverse effect of Trinexapac Ethyl on marketable seed yield at the higher concentrations, the negative linear trend was highly significant (<0.01 level) at 10 and 20 DAF applications. In contrast, Trinexapac Ethyl applied at 30 DAF had no significant effect on marketable seed yield at Bow Island.

The PGR concentration by growth stage interaction for marketable seed yield was highly significant (p<0.01) even though the main effects of the treatments were not significant at the Brooks rain-fed test site (Table 4.10). Prohexadione Calcium (750 to 4500 mg L<sup>-1</sup>) and CCC (1000 to 6000 mg L<sup>-1</sup>) had no significant effects on marketable seed yield compared to the respective untreated controls at all three growth stages (Table 4.17). Trinexapac Ethyl significantly reduced marketable seed yield at 4000 and 6000 mg L<sup>-1</sup> when it was applied at 20 DAF and 30 DAF stages, respectively. Trinexapac Ethyl also had significant linear and quadratic trends on marketable seed yield at 10 DAF, but only a significant linear trend at 30 DAF (Table 4.17). Treatment effects on seed yield and marketable seed yield of the Brooks rain-fed test site were almost comparable. This was mainly due to the presence of a lower percentage of green and immature seeds in the final harvest.

Table 4.17: Effect of concentration of three plant growth retardants applied at three crop growth stages on marketable seed yield of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011

	PGR <sup>!</sup>		stage at PGR app	lied (GS)	
Type	Concentration (mg L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
			Marketable see	d yield (kg ha <sup>-1</sup> )	
	Control (0)	5417	4922	4959	5099
Prohexadione	750	5122	5090	4999	5070
Calcium (PC)	1500	5199	5146	4791	5045
Calcium (FC)	3000	5253	5138	4813	5068
	4500 5295	4915	4896	5035	
Mean of GS for Prol	nexadione Calcium	5217	5072	4875	5055
	Control (0)	5455	5404	4673	5177
Chlormequat	1000	5278	5220	4713	5070
Chloride (CCC)	2000	5072	5499	4632	5068
Cilioride (CCC)	4000	5230	5690	4563	5161
	6000	5090	5501	4693	5095
Mean of GS for Chlo	ormequat Chloride	5167	5477	4650	5098
	Control (0)	5019	5435	5506	5320
Trinavanaa Ethyl	1000	5270	5506	5094	5290
Trinexapac Ethyl (TE)	2000	5186	5457	5250	5298
(1E)	4000	4907	4793	5075	4925
	6000	4587	5277	4904	4923
Mean of GS for Trin	exapac Ethyl	4987	5258	5081	5109
Mean for growth sta	ge across all PGR	5159	5266	4904	5110
LSD (p=0.05) for Gr	rowth stage (GS)	=			
for Po	GR	-			
for G	S x PGR	527			
Coefficient of variat	ion (%)	12.1			
	Probabili	ty values for trend	l analysis		
PC -Linear		$0.95^{\rm ns}$	$0.89^{\rm ns}$	$0.51^{ns}$	
PC-Quadratic		$0.31^{ns}$	$0.23^{ns}$	$0.34^{ns}$	
CCC- Linear		$0.09^{ns}$	$0.14^{ns}$	$0.82^{ns}$	
CCC-Quadratic		$0.33^{\text{ns}}$	$0.37^{ns}$	$0.49^{ns}$	
TE-Linear		<0.01**	$0.06^{\text{ns}}$	$0.02^*$	
TE-Quadratic		$0.04^{*}$	$0.20^{ns}$	$0.65^{ns}$	

PGR = Plant growth retardants;  $^{\text{@}}$ DAF = Days after 1<sup>st</sup> flowering;  $^{\text{ns}}$ non-significant at p≤0.05; \*significant at p<0.05 level; \*\*significant at p<0.01 level.

There were significant differences in marketable seed yield across different concentrations of PGR at the Brooks irrigated site regardless the crop growth stage (Table 4.10). Except at the lowest concentration (750 mg L<sup>-1</sup>), Prohexadione Calcium significantly reduced marketable seed yield by 6 to 7% over the untreated control at the Brooks irrigated site (Table 4.18).

Table 4.18: Effect of concentration of three plant growth retardants on Marketable seed yield of the Kabuli chickpea cultivar CDC Frontier under irrigated conditions at Brooks in 2011

Plant growth retardant	Concentration	Marketable Seed
		yield (kg ha <sup>-1</sup> )
	Control (0)	5145 <sup>AB</sup>
	750	5041 <sup>A-D</sup>
Prohexadione Calcium	1500	4825 <sup>C-E</sup>
	3000	$4781^{\mathrm{DE}}$
	4500	4791 <sup>C-E</sup>
	Control (0)	5083 <sup>A-D</sup>
	1000	$5148^{AB}$
Chlormequat Chloride	2000	$5142^{AB}$
•	4000	5169 <sup>A</sup>
	6000	5297 <sup>A</sup>
	Control (0)	5087 <sup>A-C</sup>
	1000	5155 <sup>AB</sup>
Trinexapac Ethyl	2000	$4854^{ ext{B-E}}$
-	4000	$4678^{\mathrm{E}}$
	6000	$4578^{\mathrm{E}}$

Note: Values of marketable seed yield assigned by the same are not significantly different based on LSD at p = 0.05 level.

The insignificant differences of marketable seed yield among Prohexadione Calcium concentrations suggested that the reduction of marketable seed yield was not depended on the concentration. CCC applied at 1000 to 6000 mg L<sup>-1</sup> was not significant on marketable seed yield averaged across three crop growth stages at the Brooks irrigated site.

On average, Trinexapac Ethyl reduced marketable seed yield by 5% to 10% (233 kg ha<sup>-1</sup> to 508kg ha<sup>-1</sup>) over the untreated control at the Brooks irrigated site. The reductions in marketable seed yield at higher concentrations (4000 and 6000 mg L<sup>-1</sup>) were significant as compared to that of untreated control or 1000 mg L<sup>-1</sup> concentration (Table 4.18).

#### 4.3.4.6 Harvest index

Growth stage at which the PGR were applied had a significant effect on harvest index, but the main effects of PGR or the interaction of PGR with growth stages were not significant at Bow Island (Table 4.10). On average, PGR applied at 10 DAF significantly reduced harvest index compared to the application at 20 DAF at Bow Island (Figure 4.5). However, the harvest indices determined at 10 DAF and 30 DAF were statistically comparable. The highest harvest index for this test site (55) was observed when PGR were applied at 20 DAF.

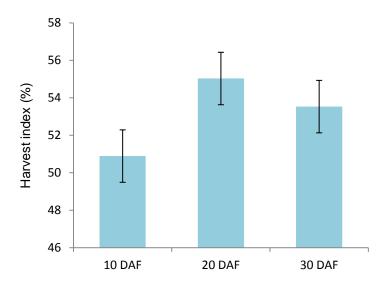


Figure.4.5: Mean effects of PGR on harvest index across different growth stages of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011. Error bars represent 1.96 times of standard error for each direction.

Main effects of growth stage and PGR were not significant on harvest index at the Brooks rainfed test site. However, their interaction effects on harvest index were significant (Table 4.10). The highest harvest index of the Brooks rain-fed test site (0.56) was recorded with 2000 mg L<sup>-1</sup> CCC concentration applied at 20 DAF while the lowest value (0.50) shared by Prohexadione Calcium at 4500 mg L<sup>-1</sup> applied at 20 DAF, Prohexadione Calcium at 3000 mg L<sup>-1</sup> applied at 30 DAF and the untreated control at sub-plot Prohexadione Calcium at 20 DAF growth stage (Table 4.19).

Increasing concentrations of Prohexadione Calcium from 750 to 4500 mg L<sup>-1</sup> at 10 DAF had no significant effect on harvest index. The lowest Prohexadione Calcium concentration (750 mg L<sup>-1</sup>) applied at 20 DAF significantly increased harvest index compared to the untreated control. However, the harvest indices among the concentrations were statistically comparable at this stage (Table 4.19).

Table 4.19: Effect of concentration of three plant growth retardants applied at three crop growth stages on harvest index of the Kabuli chickpea cultivar CDC Frontier under rain-fed conditions at Brooks in 2011

	PGR <sup>!</sup>	Growtl	n stage at PGR ap	plied (GS)	
Type	Concentration (mg.L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
	-		Harvest	index %	
	Control (0)	54	50	53	52
D., . l	750	53	53	51	52
Prohexadione Calcium (PC)	1500	55	52	53	53
Calcium (PC)	3000	54	52	50	52
	4500	55	51	52	52
Mean of GS for Pro	hexadione Calcium	54	52	52	53
	Control (0)	53	53	52	53
Chlormaquat	1000	55	52	52	53
Chlormequat Chloride (CCC)	2000	54	56	53	54
Chloride (CCC)	4000	53	55	52	53
	6000	54	53	53	53
Mean of GS for Chl	ormequat Chloride	54	54	52	53
	Control (0)	53	52	53	53
Trinovonoo Ethyl	1000	52	53	53	53
Trinexapac Ethyl (TE)	2000	53	53	54	53
(IE)	4000	52	52	52	52
	6000	52	54	53	53
Mean of GS for Trir	nexapac Ethyl	52	53	53	53
Mean for growth sta	ge across all PGR	53	53	52	53
LSD (p=0.05) for G	rowth stage (GS)	=			
for P	GR	-			
for G	S x PGR	2.8			
Coefficient of variat	ion (%)	5.4			
	Probabili	ty values for trend	analysis		
PC -Linear		0.51 <sup>ns</sup>	$0.31^{\text{ns}}$	$0.24^{ns}$	
PC-Quadratic		$0.59^{ns}$	$0.04^{*}$	$0.15^{ns}$	
CCC- Linear		$0.76^{\text{ns}}$	$0.45^{\rm ns}$	$0.49^{ns}$	
CCC-Quadratic		$0.78^{ns}$	$0.02^*$	$0.89^{ns}$	
TE-Linear		$0.39^{ns}$	$0.79^{ns}$	$0.37^{ns}$	
TE-Quadratic		$0.88^{ns}$	$0.81^{\text{ns}}$	$0.68^{\text{ns}}$	

 $^{1}$ PGR = Plant growth retardants;  $^{@}$ DAF = Days after 1<sup>st</sup> flowering;  $^{ns}$ non-significant at p≤0.05; \*significant at p<0.05 level.

Increasing Prohexadione Calcium concentrations caused a significant quadratic effect on harvest index at 20 DAF. This was mainly due to the increase of harvest index at 750 mg L<sup>-1</sup> concentration; however, the application of Prohexadione Calcium at 3000 mg L<sup>-1</sup> concentration at 30 DAF caused a significant reduction of harvest index at the Brooks rain-fed test site.

Increasing concentration of CCC (1000 to 6000 mg L<sup>-1</sup>) had no significant effect on harvest index at 10 and 30 DAF applications. However, at 20 DAF application, CCC at 2000 mg L<sup>-1</sup> concentration significantly increased harvest index over the untreated control. Overall, this

increase also led to a significant quadratic effect (Table 4.19). Trinexapac Ethyl (1000 to 6000 mg L<sup>-1</sup>) had no significant effect on harvest index at the Brooks rain-fed test site.

Neither treatments nor their interactions were significant for harvest index at the Brooks irrigated test site (Table 4.10). The harvest index (overall average) obtained at this test site (0.41) was the lowest in 2011 growing season compared to the Bow Island (0.53) and Brooks rain-fed (0.53) test sites.

#### 4.3.4.7 Crop maturity

Crop maturity was significantly different across growth stages and different plant growth retardant concentrations at the Bow Island test site in 2011. However, their interaction effects were not significant for maturity (Table 4.10). On average, crop maturity was significantly delayed when the plants were treated with PGR at 10 DAF compared to the applications at later growth stages (20 and 30 DAF) at Bow Island (Figure 4.6).

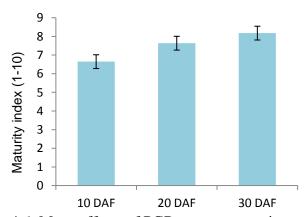


Figure 4.6: Mean effects of PGR on crop maturity across different growth stages of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011. Error bars represent 1.96 times of standard error for each direction.

Application of Prohexadione Calcium at 750 to 4500 mg L<sup>-1</sup> significantly delayed crop maturity compared to the untreated plants regardless of the growth stage when they were applied at Bow Island (Table 4.20). The maturity ratings among different concentrations, however, were not significantly different. CCC did not change the maturity of the crop at Bow Island.

Table 4.20: Effect of concentration of three plant growth retardants on the crop maturity of the Kabuli chickpea cultivar CDC Frontier at Bow Island in 2011

Plant growth retardant	Concentration	Maturity index (1 – 10)
	Control (0)	8.1 <sup>A</sup>
	750	7.1 <sup>B-D</sup>
Prohexadione Calcium	1500	$6.9^{ ext{B-D}}$
	3000	$6.8^{\mathrm{CD}}$
	4500	$6.6^{\mathrm{D}}$
	Control (0)	8.2 <sup>A</sup>
	1000	8.1 <sup>A</sup>
Chlormequat Chloride	2000	$8.2^{A}$
•	4000	$7.8^{AB}$
	6000	8.1 <sup>A</sup>
	Control (0)	8.1 <sup>A</sup>
	1000	$7.6^{\text{A-D}}$
Trinexapac Ethyl	2000	7.4 <sup>A-D</sup>
1	4000	$6.8C^{D}$
	6000	$6.8C^{D}$

Note: Values of maturity index assigned by the same are not significantly different based on LSD at p = 0.05 level.

On average, the applications of Trinexapac Ethyl at 4000 and 6000 mg L<sup>-1</sup> significantly delayed maturity over the untreated control. The maturity ratings of the plants treated with different concentrations of Trinexapac Ethyl were comparable with those treated with Prohexadione Calcium, (Table 4.20).

Crop maturity at the Brooks rain-fed sites was not affected by either growth stage or PGR applications or treatment combinations in the 2011 growing season (Table 4.10). The average maturity index obtained at the Brooks rain-fed site was  $5.9 \pm 0.09$ .

The interaction effects between plant growth retardant and growth stage were significant for crop maturity at the Brooks irrigated test site in 2011 (Table 4.10). However, only the 6000 mg L<sup>-1</sup> concentration of CCC applied at 20 DAF significantly delayed crop maturity compared to the corresponding untreated control (Table 4.21). In addition, increasing Trinexapac Ethyl concentrations (1000 to 6000 mg L<sup>-1</sup>) applied at 10 DAF showed a significant negative linear effect on maturity index.

Table 4.21: Effect of concentrations of three plant growth retardants at three crop growth stages on maturity index of the Kabuli chickpea cultivar CDC Frontier under irrigated conditions at Brooks in 2011

	PGR <sup>!</sup>	Growth	n stage at PGR ap	plied (GS)	
Type	Concentration (mg.L <sup>-1</sup> )	10 DAF <sup>@</sup>	20 DAF	30 DAF	Mean
			Maturi	ty index	
	Control (0)	5.8	7.0	6.4	6.4
D h d'	750	5.7	6.7	6.0	6.1
Prohexadione Calcium (PC)	1500	5.2	6.9	6.1	6.1
Calcium (PC)	3000	5.0	6.8	6.8	6.2
	4500 5.9 6.8	6.8	6.0	6.2	
Mean of GS for Prol	nexadione Calcium	5.4	6.8	6.2	6.1
	Control (0)	6.6	5.9	6.9	6.5
Chlormoquot	1000	6.8	6.3	6.5	6.5
Chlormequat Chloride (CCC)	2000	6.3	6.4	6.7	6.5
Chioride (CCC)	4000	6.8	6.1	6.5	6.5
	6000	6.5	6.8	6.7	6.7
Mean of GS for Chlo	ormequat Chloride	6.6	6.4	6.6	6.5
	Control (0)	6.7	6.2	6.2	6.4
Trinexapac Ethyl	1000	6.7	6.2	6.2	6.4
(TE)	2000	6.3	6.6	6.7	6.5
(1E)	4000	6.2	6.5	6.3	6.3
	6000	6.1	6.3	6.4	6.3
Mean of GS for Trin	exapac Ethyl	6.3	6.4	6.4	6.4
Mean for growth sta	ge across all PGR	6.2	6.5	6.4	6.4
LSD (p=0.05) for G	rowth stage (GS)	-			
for Po	GR	-			
for G	S x PGR	0.9			
Coefficient of variat	ion (%)	14.6			
	Probabil	ity values for trend	d analysis		
PC -Linear		1.00 <sup>ns</sup>	$0.58^{\mathrm{ns}}$	$0.95^{ns}$	
PC-Quadratic		0.08 ns	$0.39^{\text{ ns}}$	$0.55^{\text{ ns}}$	
CCC- Linear		0.85 ns	$0.22^{ns}$	$0.71^{\text{ns}}$	
CCC-Quadratic		0.91 <sup>ns</sup>	$0.86^{\mathrm{ns}}$	$0.50^{\rm ns}$	
TE-Linear		<0.01**	$0.17^{\text{ ns}}$	$0.58^{ns}$	
TE-Quadratic		$0.39^{\mathrm{ns}}$	$0.02$ $^*$	0.52 ns	

 $^{1}$ PGR = Plant growth retardants;  $^{@}$ DAF = Days after 1<sup>st</sup> flowering;  $^{ns}$ non-significant at p≤0.05; \*significant at p<0.05 level; \*\*significant at p<0.01 level.

In contrast, Trinexapac Ethyl concentrations applied at 20 DAF had significant quadratic effects on maturity rating with a plateau was observed at 2000 mg L<sup>-1</sup> concentration (Table 4.21).

## 5. Discussion

## 5.1 Effects of fungicides on the activities of plant growth retardants

Application of several rounds of fungicides to control Ascochyta blight is a common practice for chickpea production in western Canadian. Increasing the number of fungicide applications is often necessary for chickpea under high Ascochyta blight pressure (Banniza et al., 2011). Pyraclostrobin (strobilurin group) and Prothioconazole (triazole group) are two of the main fungicides used in western Canada to control Ascochyta blight incidents. In addition to controlling fungal diseases, the fungicides belonging to these groups may affect the growth and yield of the treated plants (Rademacher, 2000; Koehle et al., 2002; Lima et al., 2012). Pyraclostrobin and Prothioconazole were used several times to control Ascochyta blight in the present study in both 2010 and 2011. Since this study mainly focused on the effect of PGR on growth and yield parameters of chickpea, the possible influences of these fungicides on the some measurements may affect the validity of the results. However, any effective PGR treatment must work on chickpea with fungicide applications to adapt as a feasible crop management practice since chickpea in western Canada often get fungicide applications to control Ascochyta blight. The secondary study specifically designed to compare the possible differences of Pyraclostrobin and Prothioconazole with PGR on the dependant variables of the main study confirmed that PGR had similar effects on plant height, above ground biomass plant<sup>-1</sup>, number of seeds m<sup>-2</sup>, 1000seed weight, total seed yield, marketable seed yield, harvest index and crop maturity of CDC Frontier chickpea when they applied with each of these two fungicides. However, the application of Pyraclostrobin prior to PGR application to prevent complete crop failure may affect the results of the secondary experiment. Under practical field conditions in Brooks, it was difficult protect chickpea crop from Ascochyta blight without multiple applications of fungicides. Further studies are necessary to evaluate the side effects of fungicides on chickpea growth.

## 5.2 Effects of plant growth retardants on vegetative growth

Production of high quality chickpea seeds in western Canada where relatively short growing conditions prevail is challenged by genetic and environment-related issues. Being an indeterminate plant species, control of continues vegetative growth in chickpea is vital for

uniform crop maturity, which would have direct impact on overall production of high quality seeds (Gan et al., 2009b). High soil moisture conditions, particularly in the latter part of the growing season, encourage excessive secondary vegetative growth. Consequently, the chickpea crop often does not reach full maturity at the end of the growing season. This situation causes a dramatic reduction in seed yield and seed quality, as a result of increased proportion of green or immature seeds. The first objective of the study was to investigate the ability of PGR to control excessive vegetative growth of chickpea cultivar CDC Frontier at the reproductive phase. Plant heights measured 30 days after plant growth retardant treatment and above ground biomass per plant at harvest were considered reliable indicators for assessing vegetative growth of the chickpea crop. All three PGR used in this study (CCC, Prohexadione Calcium and Trinexapac Ethyl) did not affect these two parameters significantly. This was in contradiction with strong growth retarding effects of these chemicals on a wide range of other crops including apple (Unrath, 1997; Cline et al., 2008), rice (Na, et al., 2011), peanut (Beam et al., 2002), tomato (Altintas, 2011), wheat (Espindula et al., 2009; Grijalva-Contreras et al., 2012), lawn grasses (Jankowski et al., 2012) and sunflower (Spitzer et al., 2011). In fact, the height of the plants treated with Prohexadione Calcium and Trinexapac Ethyl were suppressed within the first two weeks after the treatments, but the plants recovered within the next two week period. Similar observations have been reported by Reekie et al. (2005) for strawberry. Stem and leaf weight of Prohexadione Calcium-treated strawberry plants were significantly lower than the untreated plants 28 days after treatment, but statistically comparable after 42 days. In addition, Prohexadione Calcium reduced the plant height measured 14 days after the application, but the height difference was gradually diminished after the first two weeks (Reekie at al., 2005). Smith et al. (1982) indicated that CCC was ineffective against stem growth of soybean. Over 80% of leaf applied CCC on soybean remained in the same leaf 14 days after the application suggesting that the ineffective translocation could be the reason for inactivity. However, another growth retarding substance 'BTS 44584<sup>‡</sup>' used in the same study had similar translocation pattern in soybean, but significantly reduced the stem elongation. It indicates that the expected outcome of PGR applications depend on the type of plant growth retardant and the plant species.

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<sup>&</sup>lt;sup>‡</sup> (S-2,5-dimethyl-4-pentamethylenecarbamoyloxyphenyl-SS-dimethylsulphonium p-toluenesulphonate) – an anti gibberellin compound

The reason for the inactivity or the inability of maintaining the growth suppressing effect of PGR over long periods in chickpea plants was difficult to determine based on data collected in the present study. The absorption, translocation and the effect on plant hormone levels by any PGR application on chickpea plants at certain time period have to be further investigated by bioassays. In addition, most studies conducted in the past have used multiple applications of PGR for regulating the plant height (Lickfeldt et al., 2001; Koutroubas et al., 2004; Cline et al., 2008; Jordan et al., 2008). Yadev and Bharud (2009) applied several plant growth regulators on chickpea including Cycocel which was also used in the present study, as four split applications after flower initiation. However, in that study, treatment effects on growth parameters such as plant height were not evaluated. In the present study we used only one application of PGR at different growth stages, as multiple applications were assumed not to be economically viable cultural practice for chickpea.

## 5.3 Effects of plant growth retardants on yield components

PGR are primarily used to control the vegetative growth of plants. Reduced demand for assimilates for vegetative growth is expected as a result of the growth suppression effect by PGR, thus they may contribute to an increase in harvestable yield by improving dry matter partitioning into reproductive organs (Rajala and Peltonen-Sainio, 2000). Improvement in seed yield and seed quality of plant growth retardant-treated chickpea was also expected from the present study, as a result of controlled excessive vegetative growth. However, the results of the current study revealed that PGR were not able to improve seed yield or 1000-seed weight, except at the Bow Island test site which will be discussed later.

Comparison of growth and yield parameters of the Brooks rain-fed and irrigated sites in 2011 clearly indicated the tendency of the chickpea crop for vegetative growth when the conditions are favourable. Compared to the Brooks ran-fed site, higher growth indicators at the Brooks irrigated site i.e. higher plant height (59 vs. 47 cm), higher above ground biomass plant<sup>-1</sup> (26 vs. 19 g) and lower harvest index (41 vs. 53) suggested that the crop tended to give less priority to reproductive development at elevated soil moisture conditions. Number of seeds m<sup>-2</sup> increased with the higher vegetative development at the irrigated site (1213 vs. 917 seeds m<sup>-2</sup>). The increase of vegetative growth generally comprises increased main stem and branch development. Increased branches and number of nodes will increase the pod bearing sites of the plant, thus the

increased number of seeds m<sup>-2</sup> at an irrigated site can be expected. However, 1000-seed weight was noticeably reduced (360 g vs. 426 g) with the increase in branches and number of nodes. These observations are in agreement with the response of chickpea genotypes to irrigation revealed by Bakhsh et al. (2007). Eight chickpea genotypes grown under irrigated conditions had 74% higher plant height and 36% higher dry weight plant<sup>-1</sup> compared to the same parameters under rain-fed conditions. In addition, the irrigation treatment increased pods plant<sup>-1</sup> by 47%, but the mean seed weight was reduced by 16% simultaneously. The reduced seed size (mean seed weight) was compensated by increased number of pods plant<sup>-1</sup>, thus the seed yield plant<sup>-1</sup> was increased by 17% under the irrigated conditions (Bakhsh et al., 2007). However, in the present study, seed yield did not benefit by the increased number of seeds m<sup>-2</sup> at the irrigated site. Moreover, under the highly favourable growing conditions in 2010, lower 1000-seed weight and seed yield also suggest that the excessive vegetative development of chickpea plants occurred on the expense of seed development during the relatively short growing period in western Canada. This is in contradiction with the beneficial effects of favourable growing conditions on chickpea yield components and seed yield in traditional chickpea growing areas where the crop is grown under continuously depleting soil moisture conditions. A significant increase of seeds plant<sup>-1</sup>, mean seed weight, biological yield, seed yield and harvest index were obtained regardless of the chickpea genotype under irrigated conditions in the areas usually experiencing terminal drought conditions (Kumar et al., 2012; Ghassemi-Golezani et al., 2013). Therefore, a proper understanding of the effect of vegetative development (especially branches) on chickpea yield components under western Canadian growing conditions is highly important to interpret the effect of PGR on the same parameters.

Zali et al. (2011) indicated that about 96% of the variation of chickpea seed yield was contributed by number of seeds per plant and mean seed weight. Number of primary and secondary branches increases the pod bearing sites of chickpea plants. Therefore, a plant structure with more branches will increase the chickpea seed yield (Zali et al., 2011). However, the cultivar used in this study (CDC Frontier) did not support this idea. In both the 2010 and 2011 growing seasons, several primary branches developed sequentially at the basal nodes with the progress of main stem development (more primary branches were observed in 2010 growing season). When the crop reached full canopy closure (around 35 to 40 days after emergence) the branches below the top canopy surface received little sunlight. In both years plants started to

flower at the 16 to 18 node stages on the main stem and progressed upward on the each node of the main stem and apical branches (branches formed after flowering). However, flowering started in basal branches slightly later and did not synchronize with the flowering order of the main stem and apical branches. In addition, many flowers in basal branches did not develop into pods. Consequently, pods belonging to different development stages could be detected all over the plant at the later stages of the reproductive phase and a higher variation was attached to the seed number per unit area. Gan et al. (2003) also indicated that new pods continuously emerged throughout the reproductive period (flowering to maturity) of chickpea, thus the length of the reproductive period for a given chickpea variety in a particular growing season on the Canadian Prairies mainly depends on the existing environment conditions.

Siddique et al. (1984) studied the contribution of branches of chickpea to seed yield under different plant densities in a Mediterranean type climate in Western Australia. They indicated that the number of branches did not depend on the plant density, but branches developed faster at lower densities. Main stem had higher dry matter, more flowers and better pod set specially at a higher density (50 plants m<sup>-2</sup>). Consequently, a sharp decrease in harvest index was noticed between main stem and basal branches. The highest biological yield was obtained at higher plant densities, but the seed yield was not different due to the low productivity of the basal branches. The results suggested that seed yield would be increased with none or sparsely branching plant structure at higher densities. Siddique and Sedgley (1985) further investigated this issue by debranching all basal branches except the branch started at the 1<sup>st</sup> node (branch 1) at a higher plant density (70 plants m<sup>-2</sup>). Debranching increased the seed yield and harvest index by 39% and 31%, respectively compared to the freely branched control plants whereas the biological yield was not much affected by debranching. Main stem and branch one had 180% more pods compared to the control and it more than compensated for the loss of seeds in removed branches. Based on these observations, a chickpea plant which has the main stem and one primary branch was proposed as an ideotype for that short season environment. Even though the climatic conditions of western Canadian short growing season is different, a similar plant structure would be useful to stabilise the number of seeds per unit area and the uniformity of seed maturity.

Sufficient data to study the effect of PGR on branching could not be collected in this study. However, application of Prohexadione Calcium and Trinexapac Ethyl caused an increase in branching (data not shown). Suppressed apical dominance by these two chemicals would enhance the lateral growth. This is in agreement with the observations made by Kumar et al. (2012) on the effect of CCC on tomato. In that study, CCC increased branching of the treated tomato plants. Garai and Datta (1999) also indicated that CCC and Prohexadione Calcium significantly increased the number of primary branches of sesame (*Sesamum indicum L. cv.* Rama).

The productivity of increased branches on seed yield in this study was highly variable and difficult to predict. This situation would lead to the inconsistent and adverse effect of PGR observed on the total seed yield and marketable seed yield. A similar observation was reported by Leitch and Kurt (1999), for linseed (*Linum usitatissimum*; Flax). Linseed plants treated with PGR (CCC and Ethephon) significantly increased tillers (number of stems per unit area), but decreased the seed yield. They suggested that the increased tillers adversely affected the main stem development and contribution of tillers to the seed yield could not compensate for the reduced seed production of main stem.

In 2011, the lowest number of seeds m<sup>-2</sup> and the highest 1000-seed weight (the maximum seed size) were obtained at the Brooks rain-fed site. Specifically, 1000-seed weight obtained at the Brooks rain-fed site was 66 g and 57 g higher than that of the Brooks irrigated and Bow Island sites, respectively. Gan et al. (2003a) stated that chickpea mean seed weight is a flexible contributor to seed yield unlike the stable kernel weight of cereal crop cultivar across different growing environments. Measuring of assimilates accumulation in sink organs by reducing the number of sink organs in a plant is one way to estimate the potential sink strength since increased number of reproductive sinks can decrease the assimilate partition into individual reproductive sinks (Marcelis, 1996). In that context, the elevated 1000-seed weight at the Brooks rain-fed site under lower number of seeds per unit area indicated that the reproductive sinks of chickpea (seeds) are capable to attract more assimilates if the source supply is not limited. Then, one important question arising would be the possibility of using PGR as a strategy to utilize this sink capacity. Prohexadione Calcium and Trinexapac Ethyl were able to increase 1000-seed weight at the Bow Island site in 2010 (only at 20 DAF) and 2011. The increased 1000-seed

weight is in agreement with the previous study on lentil as reported by Ginnakoula et al. (2012). In that study, the application of Prohexadione Calcium (100 mg L<sup>-1</sup>) twice at 14-day intervals beginning with the 5-7 leaves stage increased 1000-seed weight of lentil by 16% over the untreated control. Except at the Bow Island site, PGR did not positively affect the 1000-seed weight. At the Bow Island site, more heat units were accumulated during the growing season compared to the Brooks test sites (465 and 315 in 2010 and 2011, respectively). Moreover, the frost free period at Bow Island was at least two weeks longer than Brooks. This provided more time for seed to mature during the reproductive phase. Pod development and mean seed weight of kabuli chickpea can be increased by prolonging the reproductive period (Gan et al., 2003a). In addition, the highest increases of 1000-seed weight by these two chemicals were obtained in the year when the crop experienced above average dry conditions at the late reproductive period (about 60% less rainfall received at both locations during August and September in 2011compared to LTA). These conditions could be the main reasons for the difference of the effects of Prohexadione Calcium and Trinexapac Ethyl on 1000-seed weight determined at the Bow Island site. However, a major constraint for Canadian chickpea production is the difficulty to guarantee higher temperatures and dry conditions at the end of the growing season. In addition, the total seed yield was not benefited by the increased 1000-seed weight with Prohexadione Calcium and Trinexapac Ethyl at the Bow Island site. Therefore, it can be concluded that PGR are not a reliable tool to utilize the potential sink strength of chickpea under current conditions.

Number of seeds per unit area was not affected by PGR treatment at Bow Island. Therefore, the increase of 1000-seed weight with Prohexadione Calcium and Trinexapac Ethyl, conflicts with the significant negative effects of same chemicals on total seed yield at same site. Since one of main concern of this study was quality and uniformity of seeds, the 1000-seed weight was calculated only in marketable seed yield. Due to this reason, 1000-seed weight cannot be considered as a true yield component in this study. Therefore, it is difficult to explain the possible reasons for this conflicting effect of 1000-seed weight on total seed yield at Bow Island by the data collected in this study.

Results of the present study contradict with the positive effects of PGR on chickpea seed yield and seed quality as revealed by Yadev and Bharud (2009) and Brar et al. (1992). Control of

excessive vegetative development was not a matter of interest for both of those studies. Moreover, low concentrations of PGR ( $\geq$  200 ppm) were used in both studies which could only resulted in a slight modification of plant hormone levels. However, the objectives aimed to achieve in the present study was different from uses of PGR in south Asian region, thus higher concentrations of PGR were used to control excessive vegetative development at reproductive stage. In addition, differences of growing environments including climatic conditions and varieties used in that region could largely behind the different effects of PGR on chickpea yield components as reported by Yadev and Bharud (2009) and Brar et al. (1992).

# 5.4 Effects of plant growth retardants on crop maturity

The results of this study revealed that applications of Prohexadione Calcium and Trinexapac Ethyl tend to delay the crop maturity at the Bow Island and Brooks irrigated test sites. Rademacher (2000) indicated that senescence of the plants treated with growth retardants can be delayed mainly due to the increased cytokinin levels as a side effect growth retardation process. Grijalva-Contreras et al. (2012) also reported that Trinexapac Ethyl at150 g (a.i.) ha<sup>-1</sup> delayed grain maturity of wheat by 4 to 7 days.

Marketable seed yield was used in this study as an indicator for the uniformity of seed. On average, application of PGR caused a reduction in marketable seed yield. Specifically Prohexadione Calcium and Trinexapac Ethyl caused significant reduction in marketable seed yield at the Bow Island and Brooks irrigated test sites in 2011. This suggests that these chemicals can further prolong the seed maturing process.

Cotton cultivation is the most promising production system which uses PGR to enhance the uniform maturity of economic yield (balls). Therefore, it is important to compare the results of this study with the uses of these substances in cotton industry to understand the possible reasons for their negative effect on chickpea seed maturity. The reproductive organs of cotton plant develop in an ordered pattern. Flowers and balls initiate at the lower section of the plant and then with the progress of the season, proceed upward and expand to more distal positions. When ball development starts, they become stronger sinks for assimilates. Their collective demand for assimilates increases with the advancement of the season, thus less resources are available for new vegetative growth. Eventually, all plant energy is used by the existing balls and ceases new

flower development. This point is generally called as 'cut-out' (Ritchie et al., 2004). PGR are commonly use to manipulate this process at agronomically desired positions, thus early set balls get more resources for their development. The influence of PGR on uniform ball maturity is discussed under the section 2.9.

Freely branching chickpea cultivar CDC Frontier does not have this type of orderly flowering and pod development pattern. Moreover, the influence of PGR on lateral growth can further complicate the situation. Therefore, the effect of PGR was not predictable for pod development as in the case of cotton production.

## 5.5 Possible reasons for the inconsistent effects of plant growth retardants

In general, PGR had highly inconsistent effects especially on the yield components of this study. Some biotic and abiotic factors which could not be controlled during the field experiments may have contributed at least for a part of the inconsistency from PGR applications. The growth control of chickpea plants was expected to be achieved by reducing the bio active gibberellins levels through the activity of PGR. However, a specific hormone level in a plant is a result of a dynamic process which can be governed by many other factors. Jaillais and Chory, (2010) indicated that plant growth is a complex process which involves several plant hormones. Biosynthesis and perception of plant hormones is not only an inherent process of plants, but external environmental factors convey the inputs to it (Depuydt and Hardtke, 2011). Plants respond to stress conditions by reducing their bio-active gibberellins levels (Weiss and Ori, 2007; Yamaguchi, 2008). Therefore, the effects of reduced levels of gibberellins by PGR can be highly depended on the prevailing environmental conditions at the time of application.

Plants in the trials encountered highly variable growing conditions within the two growing seasons of this study. At the Brooks test sites, a highly impermeable hard soil layer laid beneath the first 30 cm top soil layer. When the site received even a moderate amount of rainfall, an excessive moisture level was observed in the top soil layer for several days, probably due the poor drainage of hard layer beneath it. In contrast, during a short dry spell (around 2 weeks) the top soil layer quickly dried, thus plants started to show stress signs. Both excessive moisture and water deficit conditions, can significantly affect the growth and yield of chickpea. Transient subsurface water logging conditions at the vegetative stage significantly affected on growth and

yield parameters of Kabuli chickpea (Palta et al., 2010). It inhibited root growth and substantially reduced root dry matter. Consequently, leaf area and shoot dry weight were reduced by 70% and 56% respectively. Reduced branching mainly contributed to the lower shoot dry weight. About 55% reduction of seed yield and a significant reduction of harvest index were also noticed under transient water logging conditions (Palta et al., 2010). Water deficit conditions significantly reduce vegetative growth, seed yield and harvest index of Kabuli chickpea (Anwar et al., 2003; Ghassemi-Golezani et al., 2013; Kumar et al., 2013). In addition to the direct effects of these conditions on dependant variables of this study, they also can affect the endogenous plant hormone levels in plants. Therefore, exogenously induced gibberellins reduction can complicate the situation by adding more variation in to that.

Towards the end of the 2010 and 2011 growing seasons, chickpea root rot caused by *Fusarium spp.* and *Rhizoctonia solani* was spotted at the Brooks test sites. In addition, the grey and white mold complex caused by *Sclerotinia sclerotiorum* and *Botrytis cinerea* was noticed during late reproductive period at the Brooks test sites in 2010. The effects of these diseases on growth and yield parameters were very difficult to assess since the symptoms of the affected plants were difficult to distinguish from natural senescence. Chang et al. (2004) revealed that chickpea root rot caused by *Rhizoctonia solani* reduced the capacity of root system, thus affect the water and nutrient uptake of the plants. Consequently, the disease reduces shoot growth, shoot height, plant stand and seed yield. In addition it can affect the uniformity of crop growth stage (Chang et al., 2004). Substantial yield losses, heavy mortality of flowers, poor pod formation and shrivelled pods are common consequences of Botrytis gray mold in chickpea (Pande et al., 2001). It indicates that, these diseases directly affect the dependent variables of this study. However, their influence on the total variation could not be identified separately.

Previous studies also revealed the inconsistent effects of PGR especially on yield parameters of cereal grain crops. Nafziger et al. (1986) evaluated three PGR, Mefluidide, Ethephon and CCC on plant height, and grain yield of five winter wheat cultivars. Treatments were applied at pseudo-stem erected stage (Feeks scale -5). PGR were able to control plant height, but their effect on seed yield was highly variable. Mefluidide significantly reduced grain yield (by 43%), but CCC was not affected on grain yield. Ethephon reduced grain yield only at a higher rate (0.56 kg ha<sup>-1</sup>). By reviewing the results of many studies on the uses of PGR on manipulating the

yield potential in cereal crops, Rajala and Peltonen-Sainio (2000) stated that, in most of cases PGR are effective in controlling plant height but for many other characters such as root growth, tillering, grain yield components and days to maturity, their effect is highly inconsistent, thus conflicting results may be easily obtained. Further, the effects on PGR can be depended on crop growth stage, interaction with partitioning of current assimilates, interaction with mobilization of carbohydrates, weather conditions and genotype differences (Rajala and Peltonen-Sainio, 2000). In some cases, reasons for conflicting effects of PGR were difficult to determine even by bioassays. Yokota et al. (1991) evaluated two isomers of PGR, Uniconazole (S-Uniconazole and R-Uniconazole) on growth parameters (plant height, leaf number, stem weight and leaf area) of pea (Pisum sativum) plants. Both isomers effectively suppressed the plant growth, but S-Uniconazole was more effective on all the parameters considered. However, R-Uniconazole reduced endogenous active gibberellins, brassinosteroids and sterols levels in plants more effectively than S-Uniconazole. Cytokinin and ethylene levels were similarly affected by two isomers, thus the higher activity of S-Uniconazole contradicted with the results of bioassays. It can be assumed that, unknown substances could involve with the growth retarding activity of S-Uniconazole (Yokota et al., 1991).

#### Conclusion

Growing conditions of the two years of the field experiments were different from each other. The 2010 was marked with an above average wet and below average cool growing season. Thus, the crop experienced an enhanced vegetative growth and could not attain the physiological maturity. In contrast, the climatic conditions in 2011 growing season was above average warm and dry specially at the late reproductive period, therefore, the effect of growth control would be minimal. However, in both growing seasons, PGR did not control the vegetative growth effectively.

Improvement of seed quality, seed yield and seed maturity was expected as the results of the retarded vegetative growth at the reproductive phase of the crop. The negative and inconsistent effects of PGR on 1000-seed weight, seed yield and marketable seed yield indicated that their effects on these parameters were complex. In fact, vegetative growth was not effectively controlled by PGR, thus positive impacts of controlled vegetative growth on yield parameters cannot be expected as predicted.

PGR cannot be recommended as an agronomical tool to manage the problems associated with the excessive vegetative growth of chickpea for a cultivar like CDC Frontier due to their limited capability of controlling excessive vegetative growth and unpredictable nature of effects on yield components revealed by this study.

#### **Future research**

- 1. Branches are major component of chickpea biological yield. Free branching habit of chickpea would be a major constraint in controlling chickpea vegetative growth by agronomic practices. In addition, they might have direct influence on flowering and pod set. Estimation of the effect of branching on flowering and pod set pattern, seed yield and uniformity of seed maturity would be highly useful to find a solution for maturity-related issue of chickpea and variety selection for different agro-climatic zones.
- 2. Even though PGR were not effective in vegetative growth control of chickpea in this study, their usefulness on this matter cannot be totally rejected from a single study. Their ability of controlling vegetative growth for a certain period during the reproductive stage and their influence to increase the apical branches could be utilized to increase the seed yield. Studying gibberellins and other plant hormone changes after PGR application at several growth stages and PGR distribution among different plant organs at different time intervals after applications would be highly useful for future studies on utilization of PGR on chickpea.
- 3. Fungicide applications to manage Ascochyta blight is a common practice of western Canadian chickpea producers. Proper evaluation of the impacts of commonly used fungicides on chickpea growth, senescence, hormone levels and stress tolerance is a timely requirement for further development of Canadian chickpea production.

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# **Appendices**

Appendix 1: Commercially available plant growth regulators and their main applications

Туре	Active ingredient	Main applications				
Auxin	Indol-3ylbutyric acid	Promote rooting of cuttings				
	Carbaryl	Regulate fruit setting in apple				
Auxin transport inhibitor	Cyclanilide	Regulate growth and ball opening in cotton				
Gibberellin	$GA_3$	Enhance fruit quality in orchards, improve				
		seedling establishment of rice				
	$GA_4$	Minimize russeting in apple				
Inhibitors of Gibberellin	Chlormequat chloride	Control growth in cereal crops and				
biosynthesis	_	ornamentals				
	Mepiquat chloride	Control growth in cereals, cotton and oil				
		seed rape				
	Paclobutrazol / Uniconazole	Control growth in fruit trees, ornamentals				
		and rice				
	Tebuconazole	Control growth in oil seed rape				
	Metconazole	Control growth in oil seed rape and				
		ornamentals				
	Inabenfide	Control growth in rice				
	Daminozide	Control growth in ornamentals				
	Trinexapac ethyl	Control growth in cereals, turf grass, oil				
		seed rape and rice				
	Prohexadione calcium	Control growth in apple, pear, peanut,				
		cereals and turf grass				
Cytokinin	Benzyladenine	Improve fruit size and thinning in apple				
	Thidiazuron	Defoliation in cotton				
	Chlorflorfenuron	Improve fruit size in grapes, kiwi and other				
		fruits				
Ethylene releaser	Ethephon	Induction of fruit ripening, induction of				
		flowering in pineapple, stimulation of latex				
		flow of rubber, opening cotton ball, growth				
		control in cereals				
Inhibitor of ethylene	Aviglycine	Delay fruit ripening in apple				
biosynthesis						
Inhibitor of ethylene action	Silverthiosulfate	Delay senescence in ornamentals				
	1-methylcycloprpene	Delay ripening in fruit, ornamentals and				
		vegetables				
Other	Hydrogen cynamide	Dormancy break in fruit trees and grape				
		vines				
	Maleic hydrazide	Sucker control in tobacco				
	Mefluidide	Growth control in turf grasses and				
		ornamentals				
	Dikegulac	Pinching in ornamentals				
	Chlorpropham carvone	Inhibit potato sprouting				

Source: Rademacher (2010).

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Appendix 2: Effects of the fungicide-plant growth retardants mixtures applied at 20 days after flowering on vegetative growth, yield components, seed yield, seed quality and maturity of the Kabuli chickpea cultivar CDC Frontier under irrigated conditions at Brooks in 2011

Treatment	Plant	Above	Number	1000-seed	Total seed	Marketa-	Harvest	Maturity
	height at	ground	of seeds	weight (g)	yield (kg	ble seed	index	(1 - 10)
	30 DAT <sup>!</sup>	biomass	m <sup>-2</sup>		ha <sup>-1</sup> )	yield		
	(cm)	plant <sup>-1</sup> (g)				(kg ha <sup>-1</sup> )		
(Pyraclostrobin+ Boscalid)	61 <sup>A</sup>	25	1255	353	5409	5225	0.39	6.3
Prothioconazole	61 <sup>AB</sup>	30	1855	358	5396	5228	0.38	6.3
(Pyraclostrobin+ Boscalid) + Prohexadione Calcium	$57^{\text{B-D}}$	27	1406	358	5248	5046	0.37	6.4
(Pyraclostrobin+ Boscalid) + Trinexapac Ethyl	55 <sup>CD</sup>	24	1203	361	5273	4964	0.39	6.3
(Pyraclostrobin+ Boscalid) + Chlormequat Chloride	$60^{AB}$	35	2409	363	5339	5145	0.38	6.1
Prothioconazole + Prohexadione Calcium	$60^{AB}$	27	1653	360	5073	4756	0.36	6.3
Prothioconazole + Trinexapac Ethyl	54 <sup>D</sup>	24	1226	349	5491	5221	0.39	6.4
Prothioconazole + Chlormequat Chloride	59 <sup>A-C</sup>	26	1456	351	5484	5274	0.38	6.4
Coefficient of variation %	6.2	29.1	62.2	3.6	14.5	15.7	11.9	4.1

DAT = Days after treatments; Values of each column not followed by letters or followed by the same letter are not significantly different based on LSD at p = 0.05 level.