# DIFFERENTIAL RESPONSE OF INDIAN MUSTARD (*Brassica juncea* L. Czern & Coss) GENOTYPES UNDER RAINFED AND IRRIGATED CONDITIONS

**Thesis**

**Submitted to the Punjab Agricultural University**
**in partial fulfillment of the requirements**
**for the degree of**

**MASTER OF SCIENCE**
**in**
**BOTANY**
**(Minor Subject: Biochemistry)**

**By**

**Rhythm**
**(L-2017-BS-259-M)**

**Department of Botany**
**College of Basic Sciences and Humanities**


**LUDHIANA-141 004**
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## CERTIFICATE – I

This is to certify that the thesis entitled **"Differential response of Indian mustard (*Brassica juncea* L. Czern & Coss) genotypes under rainfed and irrigated conditions"** submitted for the degree of **Master of Science** in the subject of **Botany** (Minor subject: **Biochemistry**) of the Punjab Agricultural University, Ludhiana, is a bonafide research work carried out by **Rhythm** (**L-2017-BS-259-M**) under my supervision and that no part of this thesis has been submitted for any other degree.

The assistance and help received during the course of investigation have been fully acknowledged.

**(Dr. Pushp Sharma)**
**Major Advisor**
**Principal Plant Physiologist (O)**
**Department of Plant Breeding and Genetics,**
**Punjab Agricultural University**
**Ludhiana-141 004, Punjab, India**

## CERTIFICATE – II

This is to certify that the thesis entitled **"Differential response of Indian mustard (*Brassica juncea* L. Czern & Coss) genotypes under rainfed and irrigated conditions"** submitted by **Rhythm** (**L-2017-BS-259-M**) to the Punjab Agricultural University, Ludhiana, in partial fulfillment of the requirements for the degree of **Master of Science** in the subject of **Botany** (Minor subject: **Biochemistry**) has been approved by the Student's Advisory Committee after an oral examination on the same.

**(Dr. Pushp Sharma)**
**Major Advisor**

**(Dr. P.C. Sharma)**
**External Examiner**
**Director**
**CSSRI, Karnal**

**Head of the Department**

**(Dr. Gurinder Kaur Sangha)**
**Dean Postgraduate Studies**

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***Place:***

***Date:*** ***(Rhythm)***

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## ABSTRACT

The present investigation was carried out to assess the differential response of *Brassica juncea* genotypes under rainfed and irrigated conditions. Only pre-sowing irrigation, referred as rainfed ($I_0$) and two irrigations (35 and 65 DAS), referred as irrigated ($I_2$) module. Durations of flowering and siliquing along with reproductive phase were shorter under rainfed/stressed condition. Physiological parameters like SPAD value, photosynthetic pigments, leaf area, specific leaf area and specific leaf weight declined significantly due to moisture stress. Decline in RWC was 9.0%, LWR 11.4%, membrane stability 25.3% however, RSD, WSD, membrane injury and MDA increased by 21.6%, 24.4%, 10.3% and 7.9% respectively under moisture stress. Catalase, superoxide dismutase and peroxidase activity increased imparting protection under stress. Osmoprotectants particularly TSS (29.3%), RS (53.8%) and proline (23.7%) increased to appreciable extent. Genotypes suffered decline in growth parameters, yield components and seed yield due to moisture stress as compared to non-stressed/irrigated condition. However, variation existed within genotypes for different studied traits. Banding pattern for protein by SDS-PAGE showed similar protein bands in PBR 422 under moisture stress and irrigated module. Negative correlation existed between DTE and DSI for biomass (r= -0.857**) and seed yield (r= -0.999**), however positive association existed between DTE and DTI for biomass (r= 0.791**) and seed yield (r= 0.679*). Seed yield was positively associated with initiation and completion of flowering, LA, leaf width, LWR, plant height, seed size, BY and HI. Drought resistant parameters and lesser yield reduction identified RB-50, RH 406, PBR 422, CSR 1163 and PBR 357 as promising genotypes.

**Keywords:** Moisture stress, antioxidative enzymes, osmoprotectants, seed yield and stress indices

**Signature of Major Advisor** **Signature of the Student**

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| **ਖੋਜ ਪ੍ਰਬੰਧ ਦਾ ਸਿਰਲੇਖ** | : | ਸੋਕੇ ਵਾਲੇ ਅਤੇ ਸਿੰਚਤ ਹਲਾਤਾਂ ਅਧੀਨ ਸਰ੍ਹੋਂ (*Brassica juncea* L. Czern & Coss) ਦੇ ਜੀਨੋਟਾਈਪਾਂ ਦੀ ਵੱਖੋ-ਵੱਖਰੀ ਪ੍ਰਤੀਕਿਰਿਆ ਦਾ ਮੁਲਾਂਕਣ |
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## ਸਾਰ ਅੰਸ਼


ਮੌਜੂਦਾ ਅਧਿਐਨ, ਵਰਖਾ ਨਾਲ ਅਤੇ ਸਿੰਚਤ ਹਲਾਤਾਂ ਅਧੀਨ ਸਰ੍ਹੋਂ (*ਬ੍ਰੈਸਿਕਾ ਜੰਸੀਆ*) ਦੇ ਜੀਨੋਟਾਈਪਾਂ ਦੀ ਪ੍ਰਤੀਕਿਰਿਆ ਦਾ ਮੁਲਾਂਕਣ ਕਰਨ ਲਈ ਕੀਤਾ ਗਿਆ। ਇਸ ਅਧਿਐਨ ਅਧੀਨ ਰੋਣੀ ਮਗਰੋਂ ਬੀਜਾਈ ਕਰਕੇ, ਸੋਕਾ ($I_0$) ਅਤੇ ਬੀਜਾਈ ਦੇ 35 ਅਤੇ 65 ਦਿਨਾਂ ਮਗਰੋਂ ਦੋ ਸਿੰਚਈਆਂ ($I_2$) ਦੇ ਉਪਰਲੇ ਕੀਤੇ ਗਏ। ਬਰਾਨੀ/ਸੋਕਾ ਹਲਾਤਾਂ ਵਿੱਚ ਪ੍ਰਜਨਣ ਪੜਾਅ, ਫੁੱਲਾਂ ਅਤੇ ਫ਼ਲੀਆਂ ਦੇ ਬਣਨ ਦੇ ਅੰਤਰਾਲ ਛੋਟੇ ਸਨ। ਸੋਕੇ ਕਾਰਨ ਫਿਜ਼ੀਓਲਾਜੀਕਲ ਮਾਪਦੰਡਾਂ ਜਿਵੇਂ ਕਿ SPAD, ਪ੍ਰਕਾਸ਼ ਸੰਸਲੇਸ਼ਣ ਪਦਾਰਥ, LA, SLA ਅਤੇ SLW ਵਿੱਚ ਕਮੀ ਆਈ। ਬਰਾਨੀ ਹਲਾਤਾਂ ਅਧੀਨ RWC, LWR ਅਤੇ ਮੈਂਬਰੇਨ ਦੀ ਸਥਿਰਤਾ ਵਿੱਚ ਕ੍ਰਮਵਾਰ 9.0%, 11.4% ਅਤੇ 25.3% ਦੀ ਕਮੀ ਆਈ ਜਦੋਂਕਿ RSD, WSD, ਮੈਂਬਰੇਨ ਦੀ ਸਥਿਰਤਾ ਅਤੇ MDA ਵਿੱਚ ਕ੍ਰਮਵਾਰ 21.6%, 24.4%, 10.3% ਅਤੇ 7.9% ਦਾ ਵਾਧਾ ਹੋਇਆ। ਐਂਟੀਆਕਸੀਡੇਟਿਵ ਇੰਜ਼ਾਈਮ ਦੀ ਮਾਤਰਾ ਵਿੱਚ ਵਾਧਾ ਹੋਇਆ ਜਿਸ ਨਾਲ ਫ਼ਸਲ ਨੂੰ ਸੋਕੇ ਦਾ ਟਾਕਰਾ ਕਰਨ ਵਿੱਚ ਸਹਾਇਤਾ ਮਿਲੀ। ਓਸਮੋਪ੍ਰੋਟੈਕਟੇਂਟਸ ਖਾਸਤੌਰ ਤੇ TSS (29.3%), RS (53.8%) ਅਤੇ ਪ੍ਰੋਲੀਨ (23.7%) ਦੀ ਮਾਤਰਾ ਵਿੱਚ ਵਾਧਾ ਹੋਇਆ। ਸਿੰਚਾਈ ਵਾਲੇ ਹਲਾਤਾਂ ਦੇ ਮੁਕਾਬਲੇ ਸੋਕੇ ਵਾਲੇ ਉਪਚਾਰ ਅਧੀਨ ਬੀਜੇ ਗਏ ਜੀਨੋਟਾਈਪਾਂ ਦੇ ਵਿਕਾਸ ਦੇ ਮਾਪਦੰਡਾਂ, ਝਾੜ ਦੇ ਮਾਪਦੰਡ ਅਤੇ ਝਾੜ ਵਿੱਚ ਕਮੀ ਆਈ। ਹਾਲਾਂਕਿ, ਅਧਿਐਨ ਅਧੀਨ ਵੱਖੋ-ਵੱਖਰੇ ਗੁਣਾਂ ਲਈ ਜੀਨੋਟਾਈਪਾਂ ਵਿੱਚ ਵਿਭਿੰਨਤਾ ਦਰਜ ਕੀਤੀ ਗਈ। SDS-PAGE ਦੁਆਰਾ ਪ੍ਰੋਟੀਨ ਦੇ ਬੈਂਡਿੰਗ ਪੈਟ੍ਰਨ ਤੋਂ ਘੱਟ ਨਮੀ ਅਤੇ ਸਿੰਚਤ ਹਲਾਤਾਂ ਅਧੀਨ PBR 422 ਜੀਨੋਟਾਈਪ ਵਿੱਚ ਇੱਕੋ ਤਰ੍ਹਾਂ ਦੇ ਪ੍ਰੋਟੀਨ ਬੈਂਡ ਦੇਖਣ ਨੂੰ ਮਿਲੇ। ਬਾਇਓਮਾਸ (r= -0.857**) ਅਤੇ ਬੀਜ ਦੇ ਝਾੜ (r= -0.999**) ਲਈ DSI ਅਤੇ DTE ਦੇ ਨਾਲ ਨਾਕਾਰਆਤਮਕ ਸੰਬੰਧ ਅਤੇ ਬਾਇਓਮਾਸ (r= 0.791**) ਅਤੇ ਬੀਜ ਦੇ ਝਾੜ (r= 0.679**) ਲਈ DTI ਅਤੇ DTE ਦੇ ਨਾਲ ਸਾਕਾਰਆਤਮਕ ਸੰਬੰਧ ਦਰਜ ਕੀਤਾ ਗਿਆ। ਫ਼ੁੱਲ ਪੈਣ ਦੇ ਸ਼ੁਰੂ ਹੋਣ ਅਤੇ ਫ਼ੁੱਲਾਂ ਦੇ ਪੂਰੀ ਤਰ੍ਹਾਂ ਖਿੜਣ, LA, ਪੱਤੇ ਦੀ ਚੌੜਾਈ, LWR, ਪੌਦੇ ਦੀ ਉਚਾਈ, ਬੀਜ ਦੇ ਮਾਪ, BY ਅਤੇ HI ਨਾਲ ਬੀਜ ਦੇ ਝਾੜ ਦਾ ਸਾਕਾਰਆਤਮਕ ਸੰਬੰਧ ਸੀ। RB-50, RH 406, PBR 422, CSR 1163 ਅਤੇ PBR 357 ਜੀਨੋਟਾਈਪ, ਸੋਕੇ ਦੇ ਹਲਾਤਾਂ ਦਾ ਟਾਕਰਾ ਕਰਨ ਵਾਲੇ ਮਾਦਪਦੰਡਾਂ ਅਤੇ ਝਾੜ ਦੇ ਘੱਟ ਨੁਕਸਾਨ ਦੇ ਲਿਹਾਜ਼ ਨਾਲ ਵਧੀਆ ਪਾਏ ਗਏ।



**ਮੁੱਖ ਸ਼ਬਦ:** ਸੋਕਾ, ਐਂਟੀਆਕਸੀਡੇਟਿਵ ਇੰਜ਼ਾਈਮ, ਓਸਮੋਪ੍ਰੋਟੈਕਟੇਂਟਸ, ਝਾੜ, ਤਨਾਅ ਸੂਚਕਾਂਕ


ਮੁੱਖ ਸਲਾਹਕਾਰ ਦੇ ਹਸਤਾਖਰ

ਵਿੱਦਿਆਰਥੀ ਦੇ ਹਸਤਾਖਰ

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**CHAPTER-I**

## INTRODUCTION

Oleiferous Brassica, commonly known as rapeseed-mustard, is one of the economically essential agricultural commodities grown in more than 50 countries of Europe, Asia, America and Australia (Singh *et al* 2014). Seven kinds of oilseed crops are grown in the world which are coconut, cottonseed, palm, peanut, rapeseed, soybean and sunflower. Out of these soybean ranks first and rapeseed second on the basis of worldwide production i.e. 360.08 mtonnes and 70.91 mtonnes respectively (Anonymous 2019).

India's agricultural economy is majorly contributed by oilseeds and ranks second after food grains (Rathore *et al* 2018). Oilseeds account for nearly 5% of gross national product and 10% of rest of the agricultural products (Verma *et al* 2018). In India, the productivity of oilseeds is just 50–60% of the world average and for 14-15% import, Rs 75000 crores are being drained out of the nation to meet the requirement of edible oils. Even though the oilseeds production in India has significantly increased due to high yielding varieties but the demand is constantly increasing (Rathore *et al* 2018). Rapeseed mustard is the second most important oilseed crop of India after groundnut. Its area under cultivation, production and productivity in India is reported to be 6.07 mha, 7.91 mtonnes and 1134 kg/ha respectively in year 2016-17 and its production in 2018-19 is estimated to be 8.39 mtonnes (Indiastat 2017). In Punjab, rapeseed-mustard was cultivated under 40,000 ha area and average yield was 52 kg/ha in 2017-18 (Anonymous 2018).

In India, Brassica crops are grown in various agro/climatic conditions i.e. from north-east/north-west to down south under timely/late sown, irrigated/ rainfed, mixed cropping and saline soils (Anonymous 2017). Genus Brassica of family Brassicaceae comprises 100 species which includes mainly rapeseed (*Brassica napus*), cabbage (*Brassica oleracea*), mustard (*Brassica juncea*) and turnip (*Brassica rapa*) (Hosaini *et al* 2009). These are mostly grown for oil, vegetables, condiments and fodder.

Mustard in India is grown in rabi season and it is third most important source of vegetable oil in the world (Jat *et al* 2018). Mustard oil is used for both edible and non-edible purpose. Mustard seeds have 25-45% oil content and characterised by its tempting flavour and preservative value and also for moderating food. The seed and oil of mustard have a peculiar pungency due to presence of glucosinolate and its hydrolysis products such as allyl isothiocynate (0.30-0.35%) making it suitable to use as condiment in the preparation of pickles and for flavouring curries and vegetables (Godara *et al* 2016). The oil is utilized in preparation of hair oils and medicines. Moreover, its oil cake makes an important cattle feed and manure (Rakow and Raney 2003). The leaves of mustard at vegetative stage are consumed as green vegetables because it provides adequate sulphur and other minerals in the diet. Canola oil from canola mustard varieties are helpful in preventing heart diseases as it

contains very small level of artery clogging saturated fatty acids and large level of omega-3-fatty acid. These omega-3 rich foods nurture the brain which improves memory and also cognitive function (Sodani 2015).

Above 90% area under oilseed Brassica is covered by Indian mustard (*Brassica juncea*) due to its relative tolerance to biotic and abiotic stresses as compared to other oilseed Brassica species (Sharma and Sardana 2013). The average input of rapeseed-mustard to the production and total oilseed acreage in India is 22.6% and 22.2%, respectively (Rathore *et al* 2018). The productivity of mustard in India is poorer than other developed nations because of imbalanced fertilization with poor water management (Jat *et al* 2018). Rapeseed production is higher in Europe and North America among other oilseeds, whereas Indian mustard is largely produced in India and North Africa (Singh and Singh 2018).

The mustard crop is grown largely under the arid and semi-arid areas in India and these areas are now under several risks of depleted water reserve, increasing population pressure, degradation of other natural resource base and above all the anthropogenic warming of the climate. So, despite being one of the largest acreage in India, yield of mustard is comparatively lower (Rana and Chaudhary 2013). This crop has the potential of 25-35 q/ha productivity but the poor management of resources used in its cultivation leads to the decline of average productivity to about 11.0 q/ha. Irrigation scheduling is essential factor influencing the mustard yield because excessive water application leads to its wastage, whereas restricted water application may lead to decrease in yield. (Jat *et al* 2018).

Abiotic stresses are the main reason which affects crop productivity, morpho-physiological and biochemical properties of all Brassica species (Jaleel *et al* 2009). Yield of crop is reduced by number of factors but drought is the most common factor associated with the decreased production of crops. Drought is a period with low average precipitation leading to water deficit of an area for long time. In plants, water stress affects several biochemical and physiological processes like respiration, photosynthesis, ion uptake capacity, nutrient metabolism etc. which are directly linked with the productivity of the plants (Kumari *et al* 2018a). Under water deficit conditions, the amount of reactive oxygen species (ROS) increases leading to oxidative stress. ROS are partially reduced or excited forms of atmospheric oxygen (Schneider *et al* 2019). The elevated levels of ROS in cells are very reactive and toxic causing molecular and cellular damage which can further lead to cell death. To ameliorate the oxidative stress, plants have antioxidative defence systems which include enzymatic as well as non-enzymatic antioxidants. The increased levels of these antioxidative molecules during stress can prevent membrane injury, cellular damage and therefore their concentrations are important parameters to determine the extent of drought tolerance in different genotypes (Meena and Kaur 2019). Drought stress causes extensive decline in crop production (Nasri *et al* 2008). Under stress, plants fail to express their full genetic yield

potential (Schneider *et al* 2019). Drought condition during flowering stage leads to increase in erucic acid and glucosinolate content affecting goodness of oil of *Brassica napus* (Ullah *et al* 2012). While other oil components of oilseeds are reduced during water deficit condition (Zhang *et al* 2014). Water deficit during and after flowering stage poses a more harmful effect on seed yield because of vulnerability of pollen growth, floral development and fertilization (Faraji *et al* 2009). However, acute moisture stress can even terminate the photosynthesis which results in plant death. Due to rise in global warming, water shortage is increasing at an alarming rate. Therefore, attempts should be made to increase the crop yield by 40% till 2025 in the areas with limited water availability. The present crop system is facing a big challenge of feeding the increasing population which can be overcome by increasing the crop production. So, to prevent crop from the loss caused by frequent drought, development of tolerant varieties is the present demand (Kumari *et al* 2018a). Since, there are vast variations in the climate and soil structure in the mustard growing regions of India, development of drought tolerant cultivars specific to different agro-climatic regions are important and need of the hour. Tolerant varieties have numerous methods of stress avoidance and tolerance to cope up with drought stress (Nasir *et al* 2019). Improved varieties of Indian mustard which are tolerant to biotic and abiotic stresses maintains high level of seed and oil meal quality (Rathore *et al* 2018). Therefore, the present study will help to identify donor lines for breeding drought tolerant varieties. This study will cover the knowledge gap of assessing the impact of irrigation and moisture stress on productivity of Indian mustard. So the main objectives of the present investigation are:

- To study the identified physiological traits in response to drought in *Brassica juncea*
- To study the impact of drought on anti-oxidative enzymes conferring tolerance to moisture stress in Indian mustard

## CHAPTER-II

## REVIEW OF LITERATURE

Water scarcity is the major limitation to agricultural production in several countries worldwide affecting the growth, production and quality of crops (Ahmad *et al* 2018). It affects various morphological, physiological and biochemical traits in plants. Water deficiency in plants can also hasten the process of switching from vegetative stage to reproductive stage. Drought stress in plants may lead to various physiological disorders such as decline in transpiration and photosynthetic rates (Sarker *et al* 2005). Further, there is a substantial reduction in water content, growth parameters and chlorophyll content (Jan *et al* 2017). Moreover, under water stress conditions, the consumption of $CO_2$ is ceased because of the closed stomata, and thus accumulation of excessive oxygen inside the stomata leads to the manufacture of the reactive oxygen species (ROS). This increase in ROS further leads to rupturing of cell membrane and thus making it more permeable; disturbing the rate of photosynthesis, respiration and growth of plant (Ahmad *et al* 2018). The reactive oxygen species extremely damage the construction of many cellular constituents as well, such as lipids, nucleic acids, proteins and carbohydrates (Waraich *et al* 2011).

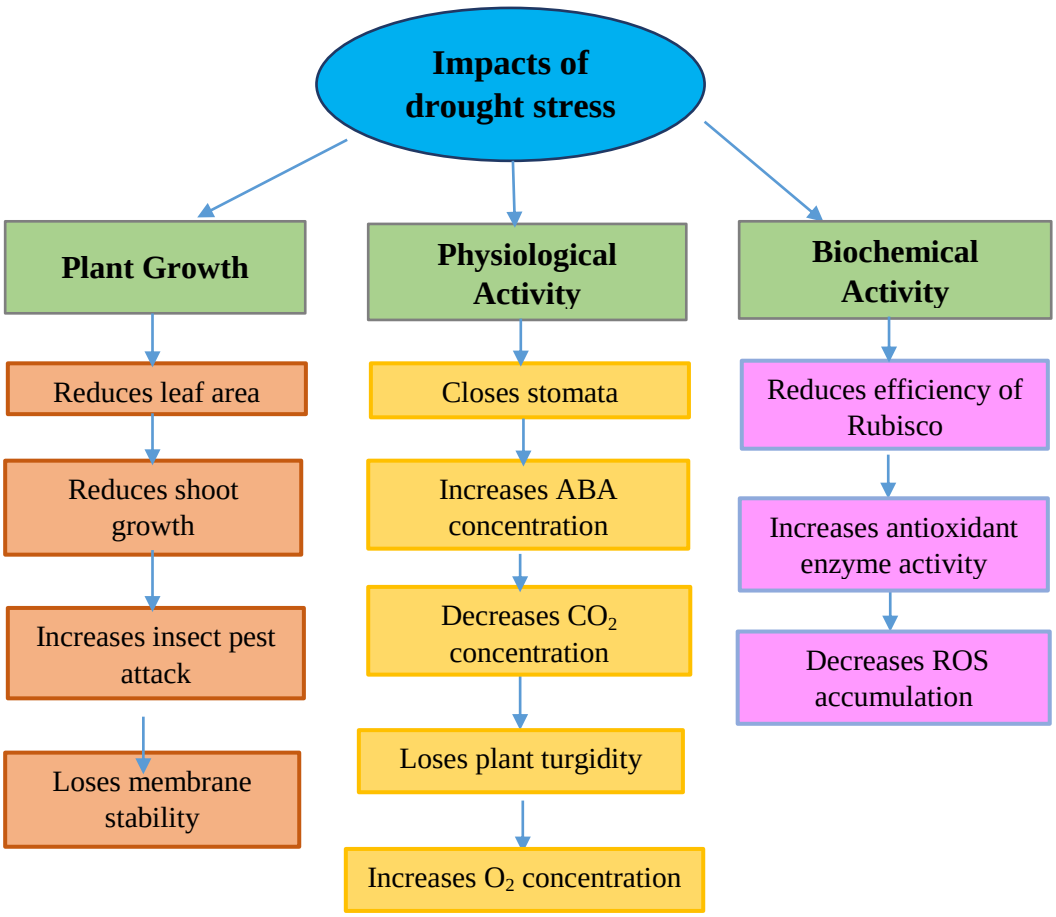



Fig. 1: Impacts of drought on plant growth, physiology and biochemical activity (Ahmad *et al* 2018)

Due to sessile nature of plants, they employ various strategies for stress tolerance which are in-built in plants. Therefore, plants are provided with several adaptation mechanisms to survive under drought condition.

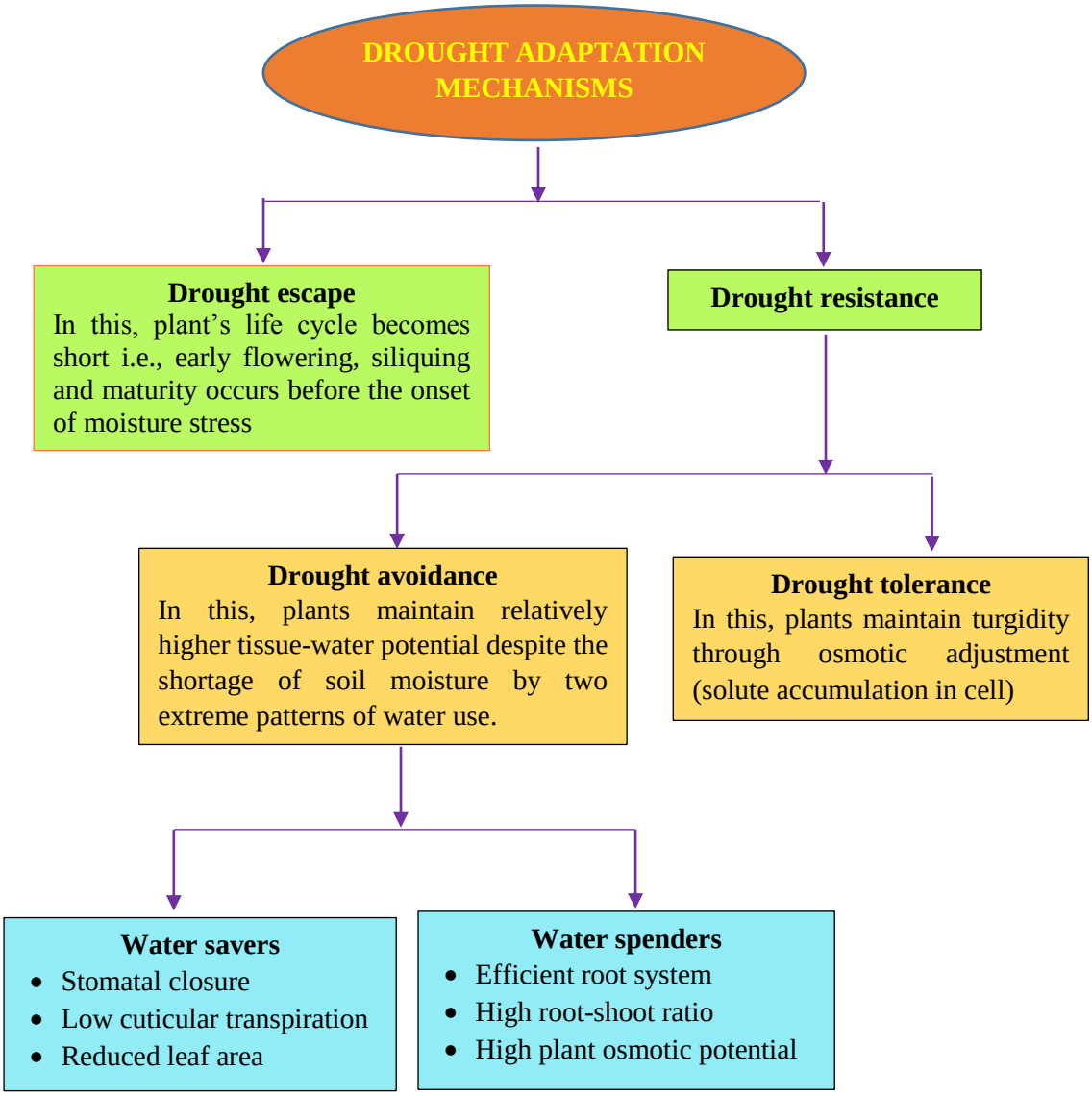



Fig. 2: Adaptation mechanism of plants to cope up with drought stress (Gautam and Bana 2014)

There is a large variation among species for drought tolerance and associated traits which will help breeders in the selection of the tolerant germplasm (Majidi *et al* 2015). Mustard crop is affected by drought of varying intensities, so immediate actions should be taken for drought proofing of mustard (Singh *et al* 2018).

The relationship between the morpho-physiological traits associated with drought tolerance is very much important in selecting suitable selection criterion for drought tolerance. The relevant literature pertaining to the present investigation is reviewed under the following heads:

- Phenological traits
- Physiological traits

- Biochemical parameters
- Growth parameters
- Yield and yield attributes

**Phenological traits**

Crop development is defined as the occurrence of different phenological stages successively with time under field conditions and these stages are significantly affected by different water regimes (Mabhaudhi and Modi 2013). In *Brassica juncea*, phenological stages include vegetative, flowering, siliquing stage and then maturity. Drought stress affects crop phenology and can stimulate plant development. Thus, plant switches swiftly from its vegetative stage to its reproductive stage (Kumari *et al* 2018a). Phenological traits like days to initiation of flowering, flowering completion, initiation of siliqua, siliquing completion and days to maturity are seriously affected by drought stress. Although, early flowering and early maturity are a part of drought avoidance mechanism in crop species but it has negative impact on productivity (Cheema and Sadaqat 2004).

Flowering period is the most important period which affects yield. There is an extreme decline in photosynthesis with the commencement of flowering period in plants and within this period, plant shows severe sensitivities to environmental stresses (Khayat *et al* 2018). Sharma and Sardana (2016) studied the effect of climate change on phenological behaviour of Brassica species by comparing two consecutive crop years and observed that mean days to 50% flowering and days to 100% flowering were higher during dry year and days to maturity were higher during wet year.

Faster phenological development is helpful in acquiring avoidance mechanism in case of terminal drought. However, selection based on earliness is complex because of continuous siliqua formation till maturity. Moreover, drought tolerance is related to early flowering and partitioning of dry matter to reproductive parts. So, selection of variety is easy as early maturing varieties can combat with drought stress. But selection of drought tolerant genotypes cannot be made on the basis of plant maturity only, as the maturity is severely affected by water deficit environments (Singh and Singh 2018).

A number of studies cited in the literature suggested that both biotic and abiotic stress factors played a key role in controlling the transition to flowering. Water stress caused an early arrest of floral development which further caused infertility (Su *et al* 2013). Plants survived under moisture stress by accelerating the flowering process and this mechanism is termed as ‘drought escape’ (Bernal *et al* 2011). In *Arabidopsis*, drought stress hastened flowering under long days but slowed flowering under short days because under long days, drought stress activated floral promoters and under short days, it activated floral repressors (Riboni *et al* 2013). However, in *Arabidopsis* it was observed that early flowering correlated

with more biomass when plants were susceptible to terminal drought stress, indicating this as an adaptation towards the drought escape response (Kenney *et al* 2014). In a similar study on *Arabidopsis* it was postulated that earlier flowering, which confirmed survival under severe terminal stress, may decrease plant yield under mild prolonged stress conditions (Schmalenbach *et al* 2014). Days to 50% flowering were lower under drought as compared to control in Indian mustard as reported by Sodani *et al* (2017). Ihsan *et al* (2016) studied the effect of drought stress (100, 75 and 50% in relation to field capacity) on developmental stages of wheat highlighting that drought stress affected each developmental stage i.e., germination, tillering, booting, heading, anthesis and maturity by reducing their durations. Birunara *et al* (2011) reported in canola that out of the three water availability treatments imposed at flowering stage i.e., 30%, 60% and 70% available water content (AWC), 30% AWC showed higher difference from control (Field capacity) which was of 155 degree days from 50% flowering until 50% siliquing in main stem. The days to flowering, end of flowering and seed maturity reduced by 3.27%, 1.71% and 1.29% under drought stress over normal whereas flowering duration increased by 9.14% under stress over normal condition in rapeseed (Zirgoli and Kahrizi 2015).

Water stress at pre-anthesis stage in cereals, declined the time to anthesis, however, stress after anthesis affected the grain filling period (Fahad *et al* 2017). Flower initiation occurred 15 days earlier under rainfed condition as compared to irrigated condition as reported in groundnut by Mabhaudhi and Modi (2013). Days to maturity decreased significantly (9.1-28.1 days) when drought stress was applied at 75% podding stage. However, the effect was more when drought stress was combined with heat stress (Sehgal *et al* (2017). Tesfamariam *et al* (2010) observed in his study on canola that water stress during flowering stage led to 127 growing degree days (GDD) earlier crop maturation. Mustafa *et al* (2018) also reported earlier maturation of rainfed over irrigated canola genotypes and found canola quality *B. juncea* lines ZBJ-06012 and ZBJ-08061 as the early maturing lines. Moderate drought treatments applied both at tillering and jointing stages in wheat had no significant effect on days to anthesis, grain filling duration, dry matter and grain yield reduction however severe drought at both stages reduced anthesis and grain filling duration significantly (Abid *et al* 2018).

**Physiological traits**

**(i) Plant water status**

Water deficit condition led to many morphological, physiological and biochemical responses in plants out of which some changes act as indicators of drought tolerance (Jan *et al* 2017). Relative water content (RWC) measures the extent to which a plant can tolerate water deficiency, so it is a better indicator of moisture stress than other parameters. On the other hand, water saturation deficit (WSD) is the deviation of water content from the leaf compared

to the saturation level of that leaf at a particular time. High water saturation deficit indicated that plants are subjected to a greater degree of water deficit. Moreover, leaf water retention (LWR) is the measure of how much water a leaf can retain (Tasmina *et al* 2016). Further, relative saturation deficit (RSD) is the amount of water lost through transpiration in the form of vapours. In an experiment conducted by Aldesuquy *et al* (2014), RWC decreased and water saturation deficit increased in flag leaf of wheat cultivars with the imposition of water stress. Similar decline in RWC was observed by Molnar *et al* (2002) in wheat. Decline in RWC and water potential due to water deficit led to closure of stomata, wilting, reduced chlorophyll content and thereby reducing the growth (Kaur and Sharma 2015c). A significant decrease in leaf relative water content and stomatal conductance occured when irrigation was stopped after 26 days in canola cultivars (Jamshidi-Zinab *et al* 2015). Further, relative water content (RWC) and stress tolerance index (STI) had appreciable complementarity with each other whereas RWC had low heritability (Majidi *et al* 2015).

Plants acclimatize to drier environments by minimizing the transpiring leaf surface (i.e., smaller leaves) or by altering relative rates of gas exchange, maximizing the ratio of carbon gain to water loss, it is known as water-use efficiency (WUE). At the leaf level, WUE is described as the net amount of $CO_2$ fixed per unit of water transpired (A/E), referred as instantaneous water-use efficiency (Ferguson *et al* 2017). WUE is often cited as a drought adaptation attribute by McKay *et al* (2008), but actually assesses only how much water a plant needs to yield biomass. However, genotypes tolerant to water limited conditions sustain higher water use efficiency (WUE) by cutting down water loss (Ahmad *et al* 2018). In *B. juncea* water deficit reduced water potential and relative water content of leaf, leading to higher osmotic adjustment and larger root growth. Therefore, the plants explored deeper and wider into the soil for water resulting in better yield attributes and ultimately seed yield. According to Kamoshita *et al* (2008), plant water status regulated performance of crop under water stress rather than plant function. Therefore, the genotypes that retain higher leaf water potential and relative water content are drought tolerant because of their higher internal water status. Farshad *et al* (2018) reported that the RWC decreased by 21.9% under limited irrigation in comparison to full irrigation in sunflower. Earlier, Hossain *et al* (2010) reported the reduction in relative humidity of leaves in sunflower under drought stress, but the degree of reduction was less in tolerant genotypes. Moreover, Rana and Chaudhary (2013) also reported the decrease in relative water content in water stressed Brassica species than unstressed which was later supported by Eslam *et al* (2017) in *B. napus.* Recently, Bhuiyan *et al* (2019) observed 23% decrease in relative water content under polyethylene glycol (PEG) induced drought stress in rapeseed. Relative water content may provide an equilibrium between the plant water content and leaf transpiration rate more productively than the other constituents reflecting the water relations (Assah *et al* 2015). Therefore, it has been regarded

as a good indicator to reveal the plant and leaf water status (Khan *et al* 2016).

Relative water content, water saturation deficit and leaf water retention (LWR) were considered as indicators of drought response. High RWC was considered to manage drought stress whereas low WSD and high LWR capacity indicated better response towards stress in *B. carinata* genotypes under drought stress (Lohani *et al* 2019). Ram *et al* (2016) observed the decrease in LWR under heat stress and considered it as a major physiological parameter for selecting high yielding Indian mustard genotypes. Under drought stress, RWC decreased and WSD increased in flag leaf of wheat (Aldesuquy *et al* 2014). RSD increased in wheat leaves exposed to water deficiency (Dedio 1975). However, tolerant genotypes faced lesser loss of water under water stress with low WSD and RSD as reported in Barley (Zhang *et al* 2015). Under shading stress also lower RWC and higher WSD, RSD and initial water content was observed in *B. juncea* genotypes by Kaur (2018). Moreover, WSD increased with increased salinity in castor bean as reported by Lima *et al* (2019). Further, WSD was more under irrigation given after 150 mm of evaporation than control in all genotypes of canola except Elite cultivar (Sepehri *et al* 2011). Under water deficit conditions, water retention capacity also decreased in wheat leaves at all growth stages (Tasmina *et al* 2016).

**(ii) SPAD and photosynthetic pigments**

The SPAD meter is used to determine the chlorophyll content measuring the absorbance of the leaf. SPAD (Soil-Plant Analysis Development) index is the ratio between thickness of leaf determined by the transmission of light in the IR range and leaf greenness determined by the transmission of light in the red light range. The Soil-Plant Analysis Development (SPAD) unit of Minolta Camera Co, Japan has developed SPAD 502 chlorophyll meter as a hand held self-convenient and light weight device for non-destructive estimation of amount of chlorophyll present in leaves which is estimated using SPAD values. Kaur and Sharma (2015c) studied variability in SPAD values of *B. juncea* and *B. napus* genotypes grown under different irrigation levels and recorded SPAD values at 65, 90 and 120 DAS. Genotypes differed significantly under moisture stress, restricted moisture at 65 days and genotypes and irrigation levels both showed significant difference in SPAD values at 90 days while no significant variation was observed in SPAD values at 120 days. Higher seed yield was associated with more SPAD values as reported by Sharma and Sardana (2016) in Brassicas and decreased under rainfed conditions. Similar results have been reported by Kumari *et al* (2019) in Indian mustard. Contradictory results under stress have been reported in rice, where SPAD values increased (Barnaby *et al* 2019).

Drought results in disorganization of thylakoid membranes resulting in decrease of chlorophyll contents and other pigments (Ashraf and Harris 2013). According to Ashraf *et al* (2013), decrease in chlorophyllase activity also caused degradation of chlorophyll under drought stress. Efeoglu *et al* (2009) observed that synthesis of carotenoid pigment increased

under drought conditions which further protected against oxidative damage. In Brassica species, moderate drought stress enhanced chla/chlb ratio whereas with severe stress the ratio decreased as observed by Majidi *et al* (2015) and same was later confirmed by Singh and Singh (2018). Under drought stress, photosynthesis is mainly affected by lesser leaf expansion, leaf senescence and improper functioning of the photosynthetic machinery and also by closure of stomata as $CO_2$ assimilation decreased and plant became more prone to photo damage (Fahad *et al* 2017). Chlorophyll content alterations can be identified as a main component influencing drought tolerance in Brassica (Majidi *et al* 2015).

Water stress destructed the thylakoid membranes and the photosynthetic pigments (Anjum *et al* 2011) and chlorophyll contents declined as observed by Din *et al* (2011) in canola, Hassan *et al* (2015) in cherry tomato and later by Dogra *et al* (2018) in *B. juncea*. Concentration of chlorophyll a was more than chlorophyll b in water stressed plants as reported by Jain *et al* (2010). Recently, Bhuiyan *et al* (2019) observed a significant decline in chl a, chl b and total chlorophyll by 55%, 49% and 50% in rapeseed seedlings. The impairment of photosynthesis under moisture stress can be subjected to many physiological reasons which are stomatal closure and decreased stomatal conductance, reduced synthesis of RuBisCO which in turn is due to reduction in its small subunits or binding of inhibitors like 2-carboxy-D-arabinitol 1-phosphate to the catalytic site of Rubisco thus damaging Rubisco activity. Decline of photosynthetic process can also be due to solute accumulation in cytoplasm leading to ion toxicity, as a result of which the enzymes involved in photosynthesis become inactive (Fahad *et al* 2017). Increase in concentration of reactive oxygen species in response to drought stress damaged the chloroplasts and thus reduced leaf chlorophyll content (Gill and Tuteja 2010). The damaging effect of drought stress on canola cultivars had been studied by Moaveni *et al* (2010) where a declining trend in chlorophyll content and photosynthetic rate was observed under water deficit as compared to control conditions. Similar findings have been reported by Sabagh *et al* (2016) in soybean. Similar decreasing trend of chlorophyll content in *B. napus* was found by Shekari *et al* (2015) in *B. napus*, further interactive effects of stress time and levels were significant on chla and b content, the Chla/Chlb and total chlorophyll content. Abid *et al* (2018) studied the effect of drought stress in wheat cultivars and found that carotenoid content was more reduced in severe stress than in moderate and normal or well-watered plants. Further the reduction was significantly greater in sensitive cultivar than tolerant cultivar, and this change was more prominent at jointing stage.

**(iii) Leaf characteristics**

Total dry matter (TDM), leaf area index (LAI) and specific leaf weight (SLW) were profusely diverse among the rapeseed cultivars and were considerably influenced by water deficit stress (Moaveni *et al* 2010). Specific leaf weight is defined as the ratio of leaf dry

weight to leaf area, which shows that it is the reciprocal of specific leaf area (leaf area / leaf dry weight) (Amanullah 2015). Leaf area index (LAI) and specific leaf weight (SLW) are important foliar traits that impacted light harvesting capacity and photosynthetic potential of leaves (Niinemets and Sacks 2006). Water stress could hasten leaf senescence and slow down leaf development, and also lead to symptoms of leaf wilting in severe drought conditions (Albert *et al* 2012). Decrease in photosynthetically active leaf area led to reduction in growth of canola cultivars under stimulated water stress conditions as reported by Lawlor (2002). Similarly, a study was conducted by Raza *et al* (2015) in rape plants, where a large decrease in leaf area was observed at every growth stage under drought stress as compared to control treatment. Similar trend was observed by Rana and Chaudhary (2013) in leaf area of *B. carinata* and *B. napus* at all growth stages and also by Amira and Qados (2014) in soybean and Madhusudhan and Sudhakar (2014) in groundnut.

The reduction in leaf area helps the plant to cope up with water stress by decreasing the surface of water loss from leaf (Vurayai *et al* 2011, Fathi and Tari 2016). However, a negative correlation between specific leaf area (SLA) and specific leaf weight (SLW) was reported by Kaur and Sharma (2015b). Both number of leaves and the size of individual leaf were reduced under drought conditions. The expansion of the leaf normally depends upon the turgor pressure and the supply of assimilates. Reduced turgor pressure and slow rate of photosynthesis under drought conditions mainly limit the leaf expansion (Fahad *et al* 2017). In *Brassica juncea* genotypes, genetic variability was studied under irrigated and non-irrigated conditions and it was found that specific leaf weight decreased in non-irrigated conditions in all genotypes (Chandra *et al* 2018). Mabhaudhi and Modi (2013) reported that leaf number reduced in rainfed condition to irrigated condition in Bambara groundnut landraces. Similarly, Germchi *et al* (2010) observed the decrease in leaf number as well as leaf area with the decrease in availability of water in soil in *B. napus* and earlier reported by Naderikharaji *et al* (2008). Water deficit decreased leaf number, leaf area, leaf area ratio (leaf area: plant dry weight and raised specific leaf weight and leaf weight ratio in *B. napus* seedlings (Qaderi *et al* 2012).

**(iv) Membrane stability index**

Cell membrane stability, reciprocal to cell membrane injury is a physiological index widely used for the evaluation of drought tolerance. The cellular membrane dysfunction due to stress is expressed by increased permeability and leakage of ions, which can be readily measured by the efflux of electrolytes, and may be used as a tolerance index for drought stress (Meena and Kaur 2019). Drought stress decreased nutrient uptake by roots and transport from roots to shoots, due to limited transpiration and hindered active transport and membrane permeability (Yuncai and Schmidhalter 2005). Godara *et al* (2017) studied the membrane stability at three sowing dates in Indian mustard and reported lesser MSI in late planting than

early sown mustard. However, the spray of salicylic acid increased MSI at all growth stages at three sowing dates. Low leaf water potential in non-irrigated condition led to decrease in membrane stability index in Indian mustard genotypes (Chandra *et al* 2018). There was more reduction of MSI in severe drought stress than in moderate stress. Drought tolerant plants retained significantly greater membrane stability index and lower membrane injury as compared to the sensitive cultivar as found in wheat (Abid *et al* 2018) and in lentil (Sehgal *et al* 2017).

**(v) Canopy temperature and Canopy air temperature differential**

Infrared canopy temperature is a proficient tool for quick, nondestructive monitoring of whole-plant response to any type of stress. The amount of cooling reveals the rate of evaporation on the surface of plant canopy (Kaur *et al* 2018). Canopy temperature (CT) is the most reliable physiological characteristic to screen drought tolerant genotypes. Eslam *et al* (2017) found that canopy temperature increased significantly under drought stress in *B. napus*. Pandey *et al* (2017) observed decrease in canopy temperature depression under drought stress in both glasshouse and field experiments of juncea canola. Moreover, canopy air temperature differential (CATD) was positively related to seed yield. Under drought stress, cooler canopy temperature is an outcome of improved stomatal conductance (Manavalan & Nguyen 2012). Plants with more stomatal conductance possess high transpiration rate, which leads to cooler canopy temperature. Therefore, canopy temperature and stomatal conductance are directly related to each other (Singh and Singh 2018). In wheat varieties increased canopy temperature observed under water stress conditions, inferred that it was due to increased respiration and decreased transpiration which was further due to stomatal closure (Tasmina *et al* 2016).

**Biochemical parameters**

**(i) Antioxidative enzymes**

Moisture stress led to the overproduction of reactive oxygen species (ROS) in plants which are highly toxic, reactive and cause damage to lipids, carbohydrates, proteins and nucleic acids which eventually led to oxidative stress. The reactive oxygen species (ROS) are mainly synthesized in the chloroplast and also in mitochondria by the reaction of oxygen with the components of electron transport chain (Fahad *et al* 2017). The ROS consists of both free radical ($OH^-$, hydroxyl radical; $O_2^-$, superoxide radicals; $RO^-$, alkoxy radicals and $HO_2^-$, perhydroxy radical) and non-radical (molecular) forms ($H_2O_2$, hydrogen peroxide and $^1O_2$, singlet oxygen).

Plants acquire an antioxidative system consisting of enzymatic and non-enzymatic components for the protection against detrimental effects of reactive oxygen species. Enzymatic system includes superoxide dismutase (SOD), catalase (CAT), peroxidase (POD), glutathione reductase (GR) and ascorbate peroxidase (APX); non-enzymatic system

comprises lipid soluble antioxidants such as carotenoids and water soluble molecules (ascorbate and glutathione) (Kumari *et al* 2018b). Superoxide dismutase (SOD) is the enzyme which comes first for protection against ROS, as it dismutates $O_2^-$ to $H_2O_2$ and $O_2$. Further, ascorbate peroxidase (APX), guaiacol peroxidase (POD) and catalase (CAT) are enzymes that catalyze the conversion of $H_2O_2$ to $O_2$ and $H_2O$ (Gratao *et al* 2005). They constitute the second defense against ROS.

Drought stress conversely increased the activities of superoxide dismutase (SOD) and guaiacol peroxidase whereas it reduced catalase activity as revealed by Abedi and Pakniyat (2010) about antioxidative enzyme activities and their isozyme patterns in oilseed rape seedlings (*Brassica napus* L.). Due to ROS production, seed yield lowered in drought affected plants (Kumari *et al* 2018b). ABA increased in water deficit conditions leading to closure of stomata, increased production of ROS like superoxide thus damaging plant by oxidizing lipids, photosynthetic pigments, nucleic acids and other cell organelles in rape plants (Kheradmand *et al* 2014).

Catalase (CAT) destroys free radicals, which damages the structure and composition of cells. Catalase activity was affected by the interaction between genotype and drought stress levels. In a study about irrigation treatments on canola genotypes, it was observed that activities of catalase and peroxidase was maximum when irrigation was stopped at flowering stage and lesser under normal irrigation (Godarzi *et al* 2017). CAT is a main enzyme to eliminate $H_2O_2$ in the mitochondrion and microbody and thus help in ameliorating the detrimental effects of oxidative stress (Shigeoka *et al* 2002). Increase in catalase activity increased tolerance to oxidative stress by providing an energy efficient mechanism to remove hydrogen peroxide ($H_2O_2$) from the plant cell by decomposing hydrogen peroxide to water and molecular oxygen without consuming reductants (Ahmad *et al* 2010). Moreover, it was observed that antioxidative enzymes which are ascorbate peroxidase (APX) glutathione reductase (GR) superoxide dismutase (SOD) and peroxidase (POX) exhibited higher basal activities in leaves of drought-tolerant variety (RH 0406) relative to drought-sensitive variety (RH 0749) of Indian mustard (Kumari *et al* 2018b). Catalase (CAT) activity increased in wheat plants when exposed to drought stress as reported by Stoilova *et al* (2010). However, water stress may also lead to a decreased CAT activity as reported by Sharma and Dubey (2005) in rice seedlings.

Moisture stress improved the activity of antioxidative enzymes, catalase and peroxidase in canola genotypes, but was variable among genotypes, as the genotypes that have greater yields under stress conditions had higher levels of enzymatic activity (Hosseini and Hassibi 2011). In Maize, Shafiq *et al* (2019) observed increase in activities of CAT, SOD and POD with increase in levels of water deficit conditions (100%, 75% and 60% of field

capacity). In an experiment performed by Farshad *et al* (2018) in sunflower inbred lines, the catalase and peroxidase activity in limited irrigation condition increased by 87.9% and 48.87% in comparison to irrigated conditions. Mirzaee *et al* (2013) determined enzyme activities of canola cultivars (SLM046 and Hyola 308) at variable concentrations of polyethylene glycol 6000 which resulted in increased antioxidative enzyme activities of roots and shoots in both the canola cultivars. Moreover, enzyme activities were higher in SLM046 than Hyola308, therefore, SLM046 was more tolerant to water stress.

**(ii) Total soluble proteins**

Protein content decreased as deficiency of water increased. In a study on sunflower inbred lines, it was observed that the protein content decreased under water limited conditions in comparison with full irrigated conditions. Drought stress is known to induce the degradation of proteins and accumulation of free amino acids to keep and adjust the cells' osmotic pressure and thus reduced the synthesis of proteins in plants (Farshad *et al* 2018). The soluble protein content decreased and free amino acids and proline increased in wheat under stress condition. Moreover, the sensitive cultivar showed higher reduction in soluble protein than the tolerant cultivar (Abid *et al* 2018). Water deficit decreased soluble proteins because of acute fall in the rate of photosynthesis (Khanum *et al* 2019). Rezayian *et al* (2018) reported the effect of drought stress on protein content of two cultivars of canola (RGS003 and Sarigol) at polyethylene glycol (PEG) concentrations of 0%, 5%, 10% and 15%. Protein content of Sarigol increased at all intensities of drought whereas in RGS003 it increased at 15% of PEG only. Rapid recovery of plant metabolism from stress and osmotic adjustment was promoted by accumulation of amino acids which was associated with storage of available substrate for protein synthesis (Zandalinas *et al* 2018).

**(iii) Proline**

Osmotic adjustment is a crucial crop protection mechanism, abided by suitable solutes originated in response to predominant stress conditions, to protect enzyme and membrane structures. These compounds are assembled in enormous amount in response to stress and work as membrane osmoprotectants to prevent the disintegration of proteins. Sugars, amino acids (proline and glycine betaine), glycerol sugar alcohols (mannitol), and additional low molecular weight metabolites are included in osmoprotectants.

Proline is the most compatible osmolyte conferring tolerance to water stress. It regulated and initiated multiple response through free radical scavenging and balanced the cellular oxidation reduction potential which act as an important indicator to overcome environmental stress in plants (Ahmad *et al* 2018). Production of compatible solutes is an important mechanism of drought stress tolerance in many species, it was proved by some previous studies which discovered the significant increase of proline content in drought

conditions in the *B. napus* varieties under drought stress (Zhu *et al* 2015).

Osmoregulation an important part of the drought tolerance mechanism in plants (Omidi 2010) which under drought stress prevented the decline in photosynthesis and yield efficiency in sunflower (Rauf 2008). Proline has the ability to oppose oxidative stress, an important strategy to overcome adverse effects of moisture stress (Vendruscolo *et al* 2007; Szabo and Savoure 2010). In a study, proline accumulated in cytoplasm of cells to prevent wheat plants from drastic effects of drought (Bajji *et al* 2001). Leaf free proline content increased in water deficit conditions (60% and 75% of field capacity) over normal condition (100% of field capacity) as reported by Shafiq *et al* (2019) in maize. Proline content has positive correlation with drought conditions and showed positive association with seed yield (Majidi *et al* 2015). The organic and inorganic solutes thus accumulated raised the osmotic pressure in the cytosol, thereby maintaining cellular turgor and a driving force for water uptake. Restoration of water deficit stress has been indicated to be accountable to the raised antioxidant system (Fariduddin *et al* 2009). The proline content was higher in tolerant variety than in sensitive variety under stress conditions. It can be inferred that leaves of drought-tolerant variety (RH 0406) had greater capacity to perform reaction of antioxidative pathways under drought stress to control drought-induced oxidative stress (Kumari *et al* 2018b).

Glasshouse and field experiments of *B. juncea* and canola hybrids along with parental lines under moisture stress were conducted by Pandey *et al* (2017) which revealed that the leaf proline concentration rose under water deficit condition which was more in hybrids as compared to its parental lines. Chandra *et al* (2018) reported higher proline content in non-irrigated condition than in irrigated one in *B. juncea* and more increase was found in its drought-tolerant genotype Rajendra Suphlam, to adjust the redox potential as an energy source, and to eliminate the active oxygen species, which further provides the conditions required for continued absorption of water from root. Farshad *et al* (2018) reported that the proline content in limited irrigation increased by 43.5% as compared to the rise in full irrigation. Increased proline protected the plant cells from collapsing by increasing the osmotic pressure (Cechin *et al* 2010). Accumulation of proline relieved the cell from osmotic stress, and also from excess ammonia and thus causing stabilization of proteins and membranes. Moreover, it increased the stability of certain cytoplasmic and mitochondrial enzymes (Sabagh *et al* 2019).

**(iv) Sugar content**

The increase in production of soluble sugars under drought stress is due to degradation of starch which is attributed to amylase activity (Vaezi 2005). In wheat, maximum content of total soluble sugar was observed under limited irrigation (49 mg $g^{-1}$ of dry weight) as compared to irrigated condition (Qayyum *et al* 2011). Similarly, reducing

sugars also increased under non- irrigated condition as reported by Khan and Naqvi (2012) in wheat. Kaur and Sharma (2015a) observed increase in contents of total, reducing and non-reducing sugars under moisture stress over irrigated condition. In canola cultivars - Sarigol and RSG003, Sarigol had maximum soluble sugar content under drought stress imparting tolerance (Rezayian *et al* 2018). The concentrations of trehalose, glucose, fructose and sucrose were also increased in *B. napus* in water deficit condition (Muller *et al* 2012). With the increase of days exposed to drought stress, total soluble carbohydrates increasd in the leaves of *B. juncea* as observed by Dogra *et al* (2018).

**(v) Malondialdehyde content**

Plant membranes are the first line of defense and easily targeted by abiotic stresses. Under drought stress, plant cell membrane gets damaged rapidly. This injury of membrane is due to increased production of free radicals, which leads to lipid peroxidation. Destruction to fatty acids of membrane could yield small fragments of hydrocarbon, out of which one is malondialdehyde (MDA) (Khan *et al* 2016). Therefore, MDA is the ultimate product of plant cell membrane lipid peroxidation and one of the important indication of membrane leakage (Cunhua *et al* 2010). In response to increased ROS, an increased content of malondialdehyde has been observed in many plants (Moller *et al* 2007). Sharma *et al* (2012) reported that as the activity of antioxidative enzymes increased, the concentration of MDA decreased in the plant. The effect of drought stress on seedling of canola cultivar (SLM046 and Hyola 308) had been studied by Mirzaee *et al* (2013) and results revealed that increased PEG concentration increased the content of malondialdehyde. In tomato, highest MDA content was observed under 40% field capacity as compared to 100% and 60% field capacity (Noori *et al* 2018). Similar observations were recorded by Shafiq *et al* (2019) in maize crop. Recently, Bhuiyan *et al* (2019) observed the increase in MDA content under drought stress in rapeseed genotypes. Abid *et al* (2018) reported that drought sensitive plants contained more $O_2^{\bullet-}$, $H_2O_2$ and MDA content than tolerant plants.

**Growth parameters**

Early cessation of rainfall under rainfed condition affects growth of plant and grain filling, leading to fewer productive branches, reduced pod size and seed weight and ultimately poor seed productivity (Rathore *et al* 2018). Plant growth occurs by cell division, enlargement, and differentiation. Water deficit conditions impair mitosis and cell elongation which leads to poor growth of plant (Hussain *et al* 2008). Similar results have been reported in canola by Ashraf *et al* (2013). Further, cell growth slows down mainly due to the loss of turgor (Fahad *et al* 2017). Growth parameters like plant height, number of branches per plant and shoot dry weight were higher in adequately watered plants as compared to mild and severe drought imposition in *B. napus* (Mehanna *et al* 2013, Hadi *et al* 2014). Similar

reduction in growth traits was also observed under rainfed condition over irrigated in Indian mustard (Singh *et al* 2018). In canola cultivars, water limited conditions reduced shoot length and biomass (Ashraf *et al* 2013).

Moisture stress affects growth of mustard in different ways like reduction in plant height, cell division and expansion, low root to shoot ratio, less leaf growth, leaf area, number of nodes, number of branches, number of seeds per pod and eventually low yield (Raza *et al* 2017). Similar findings have been reported by Jat *et al* (2018) in Indian mustard by giving different irrigations (no irrigation, 0.4 IW/CPE, 0.6 IW/CPE, 0.7 IW/CPE and 0.8 IW/CPE) and recorded higher value of growth parameters with 0.8 IW/CPE. Drought stress imposed during seed filling stage resulted in significant decreased plant height and direct impact was evident on seed yield (Eslam *et al* 2017). Plant height and number of primary branches varied significantly within the cultivars of different Brassica species as reported by Sharma and Sardana (2016) with the significant effect of environment on the growth parameters as mean plant height increased by 5.2% while main shoot length, number of primary branches and secondary branches per plant decreased by 35.5%, 9.5% and 13.9% respectively in wet year as compared to dry year. Results in Ethiopian mustard (*B. carinata*) cultivars revealed that plant height, above ground biomass and leaf area index were maximum with three irrigations and declined with two irrigations and one irrigation respectively (Verma *et al* 2018). Water deficit stress had a significant negative impact leaf area, main inflorescence length, plant height and stem dry weight in water regime of 30% available water content during siliquae development stage of rapeseed (Germchi *et al* 2010).

The increase in growth can be attributed to the time and sufficient amount of irrigation which increased cell turgidity, cell enlargement and meristematic activity which led to higher rate of photosynthesis and thus, increased growth of plant (Verma *et al* 2018). Significant reduction in plant height, leaf size, and the stem girth was observed by Khan *et al* (2015) in maize cultivars grown under water limiting conditions. Plant growth is affected by reduction in nutrient uptake and hindrance in transport of photosynthates due to limited transpiration rates, membrane permeability and impaired active transport under drought stress by Silva *et al* (2011). The major outcomes of shortage of water in canola was decreased plant height, number of branches, pod length, 1000-grain weight and yield which further hampered quality parameters (Istanbulluoglu *et al* 2010). The growth parameters under limited irrigated conditions in sunflower depended on the balance between water status of plants, rate of photosynthesis, osmoregulation, chlorophyll index and fluorescence (Farshad *et al* 2018). The reduced plant growth was an adaptive response to stress rather than as a secondary consequence of deficiency of resources (Rollins *et al* 2013).

**Yield and yield attributes**

Yield possesses many component characters which conclusively result in a remarkably flexible yield structure and hence is a very complex trait (Meena *et al* 2014). Yield is fundamentally the complex fusion of the diverse physiological processes. Water stress poses negative effects on most of these physiological processes and hence ultimately on yield. Loss of yield mainly depend upon the intensity of stress and the stage of plant (Fahad *et al* 2017). Water stress at flowering stage decreased seed yield, number of siliquae per plant and the biological yield of important rapeseed cultivars (Sinaki *et al* 2007). This was further supported by the study of Nasri *et al* (2008) in rapeseed cultivars where water deficit decreased the number of siliquae per plant, number of seeds per siliqua, 1000-seed weight, seed oil content and oil yield of rapeseed cultivars. Drought stress imposed at different growth stages of canola resulted in maximum loss during the flowering stage (Din *et al* 2011). The effect of different irrigation scheduling on seed yield of *Brassica campestris* var. toria was observed by Deka *et al* (2018) and recorded higher seed yield on application of 6 cm irrigation at both 25 and 50 DAS that is 16.60, 23.11 and 37.10 percentage higher over the other irrigation schedules i.e., 6 cm irrigation at siliqua formation stage (50 DAS), 6 cm irrigation at pre flowering stage (25 DAS) and rainfed respectively.

Rapeseed genotypes (*Brassica napus* L.) under moisture stress showed a significant decline in the number of siliquae per plant, 1000-seeds weight and seed yield during the seed filling stage (Eslam *et al* 2017). Deviations in yield attributes of Indian mustard under rainfed and irrigated conditions as reported by Singh *et al* (2018) showed that value of phenotypic coefficient of variation (PCV) was more than the genotypic coefficient of variation for all traits under both environmental conditions, signifying the role of environment in expression of these characters. Mean relative yield under irrigated condition was more than rainfed condition in Indian mustard genotypes (Singh *et al* 2018). Under various irrigation scheduling Jat *et al* (2018) demonstrated that the scarcity of water significantly reduced yield attributes like number of siliquae/plant, length of siliqua, number of seeds/siliqua, 1000-seed weight and seed yield in *B. juncea* and recently by Kumari *et al* (2019) under irrigated and rainfed conditions. Similar findings have been endorsed in Brassica species subjected to increasing water deficit during the reproductive stages, the varieties with high osmotic adjustment had lesser effect on yield under stress (Blum 2017). Drought stress caused a significant reduction of seed oil yield compared with irrigation with held at flowering and grain filling stages, in which oil yield was reduced to 40% and 21% respectively in *B. juncea* genotypes (Chandra *et al* 2018).

Drought stress affected the growth, development and physiological processes of the plant which further reduced biomass and eventually grain and oil yield due to decrease of

number and size of the seeds (Pradhan *et al* 2014). According to Sehgal *et al* (2017) the high temperatures in drought stressed lentil plants at the time of seed filling caused a drastic reduction in seed quality and quantity which was due to the reduced supply of sucrose to the developing seeds leading to decrease in size and number of developed seeds whereas increase in number of shrivelled seeds. Moisture status greatly influenced the mechanism of synthesis and accumulation of various seed reserves and deficiency of water at this stage disrupted the seed filling (Ochatt 2015). Ethiopian mustard cultivars with different irrigation levels i.e., 3 irrigations, 2 irrigations and one irrigation and recorded that yield attributes increased with increase in number of irrigations (Verma *et al* 2018).

Positive and significant association of WUE with total dry matter (r= 0.632**) and seed yield (r= 0.712**) was observed in Brassica genotypes under rainfed condition (Singh *et al* 2009). In canola, Ashraf and Harris (2013) studied the effect of moisture stress by checking irrigation at flowering and grain filling stages which caused 35% and 18% yield loss respectively. A significant loss in number of seeds per siliquae, number of siliquae per plant, 1000-seed weight, seed production, seed oil content and oil production due to moisture stress was reported by Lakhdar *et al* (2009) and later confirmed by Shirani-Rad (2012). In a study conducted by Nejat and Mantri (2017), seed yield of different cultivars of *B. rapa* and *B. napus* were significantly affected by drought stress at ripening stage. A significant decline in the grain yield of barley (*Hordeum vulgare* L.) was observed due to less number of fertile tillers and grains along with low 1000 grain weight (Fahad *et al* 2017) under drought conditions. Naderikharaji *et al* (2008) reported the decline in seed yield, 100 seed weight, siliquae length, seeds/siliqua and no. of siliquae per plant with the decrease in proportion of water available to plants (75% FC, 50% FC and 25% FC) over control in *B. napus*. Similar trend was observed by Moaveni *et al* (2010) under drought stress as well as by Sodani *et al* (2017) in Indian mustard. The reducing trend of pod number and seeds/pod, 1000 seed weight and seed yield was also observed by Raza *et al* (2015).

The decrease of biomass, yield and harvest index was reported in groundnut landraces under drought condition over normal condition (Mabhaudhi and Modi 2013). Reproductive stage is vulnerable to drought stress as it reduced the yield components in canola (Ghobadi *et al* 2006). In canola, drought stress applied at flowering stage reduced leaf relative water content ($RWC_{leaf}$) and stomatal conductance, which further reduced yield as evaluated by number of siliquae per plant and number of grains per pod. Thus, seed yield in canola decreased, even by a short period of soil moisture stress, during reproductive stages (Jamshidi-Zinab *et al* 2015). Yield and yield attributes of canola were negatively affected by water deficit stress, as with the decline in number of siliquae per plant, plant height and plant weight (Sabagh *et al* 2017, Kandil *et al* 2017).

**Oil Content**

Moisture stress reduced seed oil content and seed yield to the time of 2.6% and 25%, respectively in *B. napus* according to Shekari *et al* (2015). Flowering stage was the most sensitive stage for drought injury which resulted in an extreme loss in seed yield (29.5%) as well as oil yield (31.7%) in *B. napus* (Ali *et al* 2017), earlier in soybean (Hosseini and Hassibi 2011) and in canola at flower budding stage moisture stress decreased oil content (Tesfamariam *et al* 2010).

However, contradictory results have been reported by Deka *et al* (2018) and Verma *et al* (2018) where no significant effect on oil content was observed under water deficit condition. Jat *et al* (2018) reported higher oil content and oil yield of *B. juncea* under 0.7 IW/CPE and 0.8 IW/CPE irrigation scheduling as compared to 0.4 IW/CPE and 0.6 IW/CPE. Similarly, Germchi *et al* (2010) reported the negative impact of water deficit stress on oil content of *B. napus* and the lower value was observed at flower bud formation stage. Later similar findings were recorded in *B. napus* by (Shekari *et al* 2015).

**Susceptibility and tolerance indices**

Different indices are used to measure the stress level experienced by a crop and associated seed yield which are based on both plant and soil water staus (Lipiec *et al* 2013). Drought susceptibility index (DSI) is a criterion of drought tolerance and its lowest value indicates highest leel of drought tolerance and vice versa. DSI is a ratio, thus a genotype with significantly lower seed yield under drought condition can also have lower DSI value. Therefore, genotypes having lower DSI values alongwith higher seed yield were selected as drought tolerant genotypes by Singh *et al* (2018). Among *B. carinata* varieties, Jayanti was highly tolerant to water stress with lower DSI value and among *B. napus* varieties Sheetal was highly tolerant due to lower DSI value as reported by Rana and Chaudhary (2013). In the study of Dogra *et al* (2018) on *Brassica juncea*, water stress was imposed at three stages-branch initiation, flower initiation and siliquae formation stages and observed that DSI showed decreasing trend in all genotypes from branch initiation to siliquae formation stage. However, out of genotypes, Kranti showed significantly lower value of DSI. DSI of biomass and seed yield were negatively associated with DTE under both one and two irrigations in both *B. juncea* and *B. napus* (Kaur 2012). Stress tolerance index and stress susceptibility index had highly negative correlation under each saline level in *B. juncea* as observed by Kannu Priya (2019). Similarly, Sharma and Sardana (2013) studied about heat resistant parameters associated with growth traits, heat tolerance efficiency and their correlation with seed yield in Indian mustard. Chauhan *et al* (2007) also calculated DSI values for seed yield and other related traits to find the relative tolerance of Indian mustard genotypes under watered and drought conditions.

**SDS PAGE**

Under abiotic stress conditions, plants' response is highly complex and involve drastic changes in the protein profiles (Qazi *et al* 2019). These proteins might play a role in antioxidative defence, heat shock, metal binding, signal transduction, antifreezing or osmolyte synthesis (Qureshi *et al* 2007). Under drought stress, several genes are induced which further synthesise the following proteins- late embryogenesis abundant proteins (LEA), heat shock proteins (HSPs), lipid transfer proteins (LTPs), protein phosphatases and protein kinases (Qazi *et al* 2019). SDS-PAGE is the technique employed to detect quantitative and qualitative changes in proteins (Qureshi *et al* 2007).

Lee *et al* (2016) reported that rubisco protein patterns differed considerably between different varieties of *B. napus* and further observed that lower intensity bands of larger and smaller subunit were found under drought stressed varieties of *B. napus* as compared to irrigated ones and the same was later confirmed by Khan *et al* (2016) in *B. napus* seedlings.The comparative proteomic analysis of *B. juncea* leaves under salinity stress led to the identification of 42 differentially-expressed proteins, out of which 33 increased in their intensity while 15 were down regulated under salt stress. Moreover, several novel proteins such as PT4 transporter, PII-like protein, SOS2, oxygen- evolving enhancer protein 1 and rubisco activase, along with other differentially-expressed proteins were recognised, which were associated with plants'response under salt stress and provided new ways for increasing salt tolerance in Indian mustard (Yousuf *et al* 2016). In a study, it was shown that LEA4-1 protein was induced in vegetative tissues of *Brassica napus* and related species by ABA and abiotic stresses as it had important role in conferring tolerance to abiotic stresses like cold, drought, salt, heat and osmotic stresses (Dalal *et al* 2009). In a study, Toosi *et al* (2011) studied the expression and quantity of several proteins at different stages of *B. juncea* var. Ensabi and observed that the seed protein S8 at 29 kDa was expressed at all stages in shoot samples and proteins S5 (54 kDa) and S10 (23 kDa) and were expressed in both root and shoot samples at all stages.

**CHAPTER-III**

# MATERIALS AND METHODS

The field experiments were conducted at the research farm of Oilseed section, Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana. The present investigation entitled, "Differential response of Indian mustard (*Brassica juncea* L. Czern & Coss) genotypes under rainfed and irrigated conditions" has been conducted in 2017-18 under three replications. The genotypes taken for present study were JC 210-335, CJRD 1261, RB-50, RH 406, PBR 422, ELM 38, CSR 1163, IAN, MCN 09-40, MLM 41-13-2, PBR 357 and RH 1518 procured from Oilseed section, Department of Plant Breeding and Genetics. The mustard crop was sown in Factorial Randomized Block Design (RBD) on 10$^{th}$ November, 2017. Physiological and biochemical experiments were performed in laboratories of Oilseed section.

**Location**

The experimental site is located at 30°56' N latitude and 75$^{o}$48' E longitude and at an altitude of 247 meters above the mean sea level.

**Treatments:** Irrigation modules comprised

a) $I_0$, only pre-sowing irrigation, referred as rainfed (RF)

b) $I_2$, two irrigations, first at 35 and second at 65 days after sowing, referred as irrigated (IR)

**Climate**

Ludhiana is a sub-tropical region having semi-arid climate with hot and dry period during April to June and further hot and humid period during July to September and cold period in the months of December and January. There is a substantial rise and drop of temperature during different months of the year. Temperature often exceeds 38$^{o}$C during summer and sometimes reaches 45$^{o}$C with dry spell during May and June. Minimum temperature falls below 0.5$^{o}$C with some frosty interval during the winters of December and January. The mean annual rainfall is 650 mm, about three-fourth of which is the effect of south-west monsoon during July-September. In the winter months of December, January and February, rains meagerly occur.

The meteorological data recorded during standard meteoreological weeks (SMWs) of the crop growing season (rabi 2017-18) obtained from meteorological observatory of the Department of Climate Change and Agricultural Meteorology, Punjab Agricultural University, Ludhiana which is situated at a distance of about 200 meters from the experimental site is depicted in Fig. 3 and Appendix-I. The temperature means reported during second week of November, 2017 (45$^{th}$ SMW) to second week of April, 2018 (15$^{th}$ SMW) ranged between 10.7$^{o}$C in the 1$^{st}$ SMW (1-7 January) to 27.6$^{o}$C in the 14$^{th}$ SMW (2-8

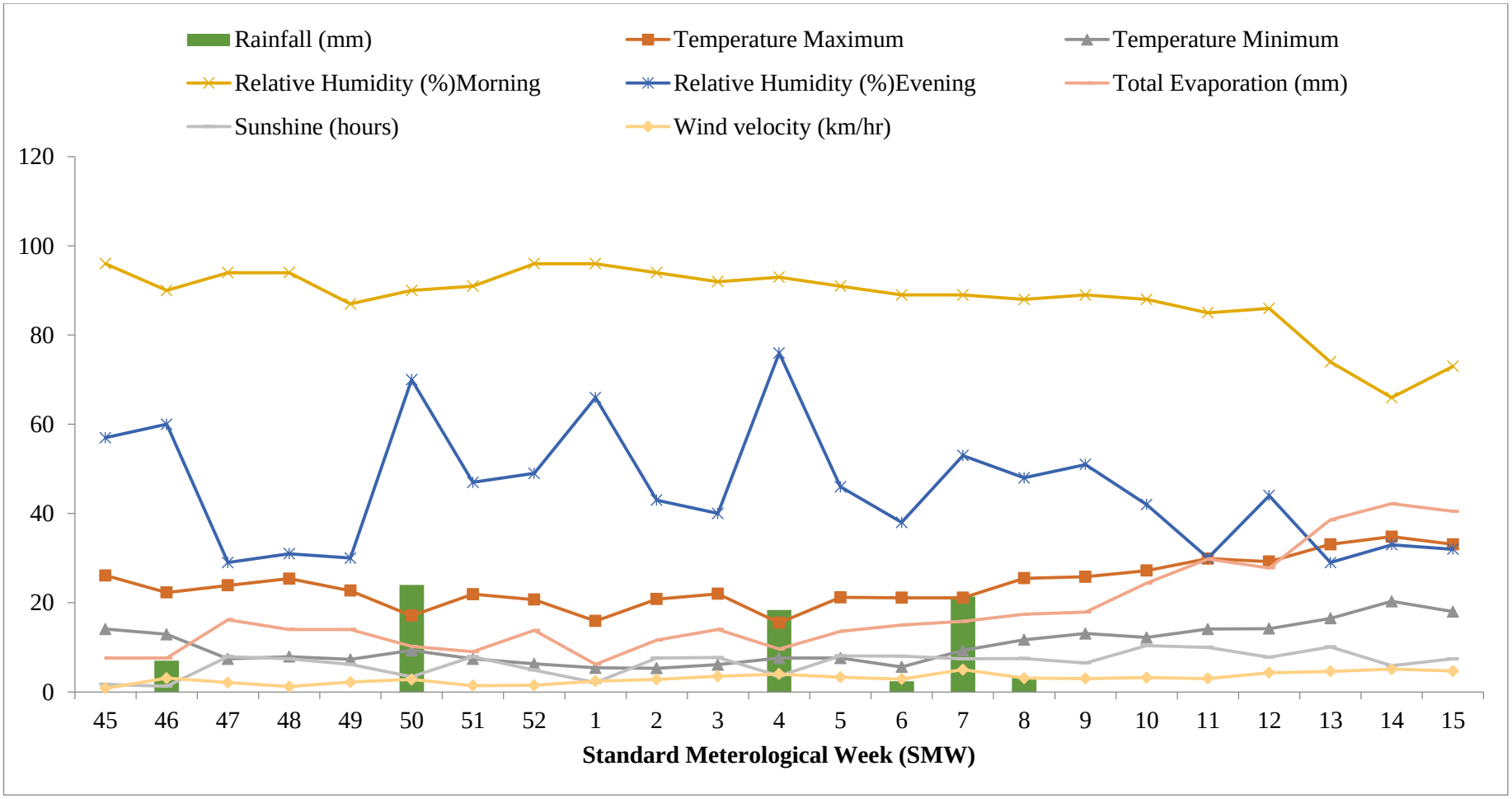



Fig. 3: Mean weakly meteorological data during the crop season (2017-18) in the Department of Climate Change and Agricultural Meteorology, PAU, Ludhiana

April). The minimum weekly temperature ranged from 5.3°C during period of crop growth in the 2nd SMW (8-14 January) to 20.3°C in 14th SMW (2-8 April) while the maximum weekly temperature ranged from 15.5°C in 4th SMW (22-28 January) to 34.8°C in 14th SMW (2-8 April). The relative humidity varied from 50-85% during crop growth period. The relative humidity varied from 66% to 96% in the morning and from 29 to 76% in the evening. Maximum rainfall of 24.0 mm was received during 50th SMW (10-16 December) and minimum of 2.4 mm during 6th SMW (5-11 February). The maximum weekly evaporation (42.2 mm) was recorded in 14th SMW (2-8 April) whereas minimum evaporation (6.2 mm) was recorded in 1st SMW (1-7 January). Daily mean sunshine hours ranged from 1.3 hours in 46th SMW (12-18 November) to 10.4 hours in 10th SMW (5-11 March). Daily mean wind velocity ranged from 0.8 km/hr in 45th SMW (5-11 November, 2017) to 5.1 km/hr in 14th SMW (2-8 April, 2018).

**Following are the observations recorded during crop growth and development**

**Phenology**

**Days to flower initiation**

The date on which first fully developed flower was observed in each plot under each treatment was noted. Later, the number of days from sowing date to the noted date was counted and given as number of days required for flower initiation.

**Days to 50% flowering**

The date on which fully developed flowers seemed on half the number of plants in each plot was noted. Similar method was followed for both the treatments. The number of days counted from sowing to the noted date was known as the number of days taken for 50% flowering.

**Days to flowering completion**

The date of appearance of fully opened flowers on all the plants of each plot in each treatment was noted. The number of days from sowing to the noted date was counted and was called the number of days taken for flowering completion.

**Flowering period**

The period in days from flower initiation to flowering completion was described as flowering period.

**Days to initiation of siliqua**

The date on which at least one flower in each plot was converted to siliqua was recorded. Days to initiation of siliqua was computed from date of sowing upto this noted date.

**Days to 50% siliquing**

The date when atleast half of the plants in each plot produced siliquae was noted. Days to 50% siliquing was computed from the date of sowing.

**Days to siliquing completion**

The date was noted when siliquae appeared on all plants of the plot. Days from sowing to completion were counted and were termed as days to siliquing completion

**Siliquing duration**

The period of days from siliqua initiation to siliquing completion is termed as siliquing duration.

**Reproductive phase**

Reproductive phase (days) is the number of days counted from initiation of flowering to siliquing completion.

**Days to maturity**

The maturity of crop was referred to the stage when plant stem and branches turned pale yellow to brown, siliquae became lemon yellow and seeds have become brown to brown-black in colour. The number of days from sowing date to this date was counted and considered as days to maturity

**Physiological traits**

Maximum expression occurs at flowering stage, so the following physiological traits were recorded at this stage. Crop is physiologically at its best at flowering stage. So, the important physiological traits were recorded at this stage under irrigated and rainfed modules.

**Chlorophyll content (Hiscox and Israelstam 1979)**

**Reagents:**

(i) Dimethyl sulphoxide (DMSO)

**Procedure:** Leaf samples of 0.1g were placed in vial containing 5 ml of DMSO. Vials were then kept into the water bath at 50ºC for 2 hours 30 minutes. Absorbance was recorded at 645nm and 663nm. The concentration of chlorophyll a, b and total chlorophyll were calculated by using Arnon's equations:

$$\text{Chl a (mg/g FW)} = 12.7 \times A_{663} - 2.69 \times A_{645} \times \frac{\text{Volume}}{1000 \times \text{Weight}}$$

$$\text{Chl b (mg/g FW)} = 22.9 \times A_{645} - 4.68 \times A_{663} \times \frac{\text{Volume}}{1000 \times \text{Weight}}$$

$$\text{Total Chl (mg/g FW)} = 20.2 \times A_{645} + 8.02 \times A_{663} \times \frac{\text{Volume}}{1000 \times \text{Weight}}$$

(A = Absorbance at respective wavelength)

**Carotenoids content (Kirk and Allen 1965)**

The same chlorophyll extract was measured at 480 nm by using UV 2600 spectrophotometer (Techcomp) to estimate the carotenoid content.

$$\text{Carotenoids (mg/g FW)} = \frac{1000 \times A_{480} - 1.29 \times \text{Chl a} - 53.78 \times \text{Chl b}}{220} \times \frac{\text{Volume}}{1000 \times \text{Weight}}$$

Where, A = Absorbance at respective wave length

**SPAD-chlorophyll values**

The Soil Plant Analysis Development (SPAD), a unit of Minolta Camera Co. of Japan has developed SPAD 502 Chlorophyll Meter which is a light weight and hand held self-convenient device for easy and non-destructive estimation of chlorophyll content of leaves. For each observation, third and fourth leaf from top of random ten plants were selected from each plot. The average of ten readings was reported as SPAD value. Midrib of the leaf was avoided carefully under sample area (sensor) of the instrument.

**Canopy air temperature differential (CATD) (Reynolds *et al* 1997)**

Canopy temperature was measured by directly imposing a beam on plant canopy using Everest Interscience Inc. Infrared thermometer, USA (Model no. 6110.4ZL) and CATD was calculated using formula:

CATD (°C) = Canopy temperature (°C) – ambient temperature (°C)

**Plant water status (Weatherley 1950)**

Fresh weight (FW) of 5 excised leaf discs was noted and then kept to rehydrate in 10 mL distilled water for 4 hours at room temperature. Saturated weight (SW) of the discs was recorded. The discs were later on dried for 48 hours at 60°C-70°C in an oven. The dry weight (DW) of leaf discs was recorded. The relative leaf water content, relative saturation deficit and water saturation deficit was calculated using formula given by Weatherley (1950) and Barrs (1968):

$$\text{Relative water content (RWC \%)} = \frac{FW - DW}{SW - DW} \times 100$$

$$\text{Relative saturation deficit (RSD \%)} = \frac{SW - FW}{SW} \times 100$$

$$\text{Water saturation deficit (WSD \%)} = \frac{SW - FW}{SW - DW} \times 100$$

(FW= fresh weight; SW= saturated weight; DW= dry weight)

**Leaf water retention (Sangakkara *et al* 1996)**

3rd or 4th leaf was sampled and weighed as fresh weight. Then the leaf was kept in shade for 4 hours and than weighed to record decrease in weight. After this, the leaf was dried for 48 hours at 60°C-70°C in an oven and dry weight (DW) was noted. Leaf water retention was computed by the following formula.

$$\text{Leaf water retention (LWR \%)} = \left[1 - \left(\frac{\text{Fresh weight - Weight after 4 hrs.}}{\text{Fresh weight}}\right)\right] \times 100$$

**Leaf traits**

**Number of leaves**

Number of leaves were counted of 10 random plants of each plot. Their mean value was calculated and referred as number of leaves per plant of that plot.

**Leaf area, specific leaf area and specific leaf weight**

Leaf length (cm), width (cm) and area ($cm^2$) were measured by area meter AM 300 (Bioscientific Ltd.). Fresh weight and dry weight of those leaf samples were taken. Based on leaf area and dry matter accumulation, following parameters were computed:

$$\text{Specific leaf area (SLA)} = \frac{\text{Leaf area } (cm^2)}{\text{Dry weight (mg)}}$$

$$\text{Specific leaf weight (SLW)} = \frac{\text{Dry weight (mg)}}{\text{Leaf area } (cm^2)}$$

**Membrane Stability Index (Premchandra *et al* 1990)**

Leaf membrane stability index (MSI) was determined from third or fourth leaves of main raceme. 0.1 g of fresh leaf tissue was placed in test tube containing 10ml of distilled water. Incubation was provided for 4 hrs at room temperature. The electrical conductivity of the water containing the sample was measured using conductivity meter and termed as C1. Then the test tubes were put in boiling water for 1 hour and after that it was cooled. Then again electrical conductivity was measured which was termed as C2. Leaf membrane stability and membrane injury was calculated using the following formula:

$$\text{Membrane stability (\%)} = [1- C1/ C2] \times 100$$

$$\text{Membrane injury (\%)} = 100- \text{Membrane stability}$$

**Biochemical parameters**

**Total soluble sugars (Dubois *et al* 1956)**

Sugars mixed with concentrated sulphuric acid leads to the formation of dehydration products which are furfural or 5-hydroxymethyl furfural. These products further react with phenols to serve as chromophore and forms orange-yellow colour.

**Extraction**: 0.1 g of sample was homogenized in 3 ml of 70% ethanol initially and centrifuged at 5000 rpm for 10 minutes. The residue was re-centrifuged by adding 2 ml ethanol to assure complete extraction. Supernatants were then pooled and utilized for estimation of total sugars and reducing sugars.

**Reagents**

i. 70% ethanol
ii. Concentrated $H_2SO_4$
iii. 5% phenol

**Estimation:** To 0.1 ml of extract, 1 ml of 5% phenol was added and then waited for 10 minutes. After that, 2.5 ml of concentrated $H_2SO_4$ was added. To ensure proper mixing of solutions, it was poured directly in the middle of test tube directly. The test tubes were cooled for 20 minutes at room temperature. The absorbance was read at 490 nm taking blank as the reaction mixture without supernatant. The total sugar content was calculated from the standard which is glucose given in the graph below using standard value (0.08 mg= 0.8120 O.D.).

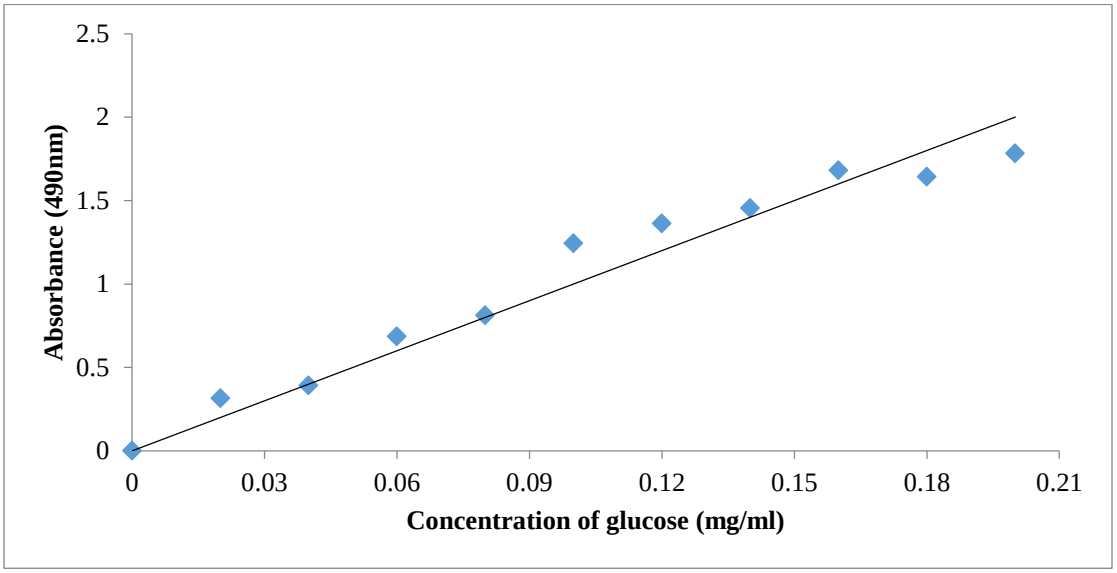



Fig. 4: Standard curve of total soluble sugars using glucose as standard

**Calculation:**

Total soluble sugar= $\frac{\text{Conc. of std.} \times \text{O.D. of sample} \times \text{total volume of extract (ml)}}{\text{O.D. of std.} \times \text{volume taken for estimation (ml)} \times \text{weight of tissue (g)}}$ (mg/g DW)

**Reducing sugars (Nelson 1944)**

**Reagents:**

A. Dissolved 25 g of Sodium potassium tartarate, 25 g of anhydrous sodium carbonate, 20 g of sodium bicarbonate, 200 g of anhydrous sodium sulphate were dissolved in 800 ml of distilled water and final volume made upto 1000 ml.

B. 15g of copper sulphate was dissolved in 100 ml of distilled water and 2-3 drops of conc. $H_2SO_4$ were added.

C. Reagent C was prepared afresh by mixing both reagent A and reagent B in 25:1 ratio (v/v).

D. Arsenomolybdate reagent: 25 g of ammonium molybdate was dissolved in 450 ml of distilled water and 25 ml of conc. $H_2SO_4$ was added gradually by stirring. 2.5 g of sodium arsenate was dissolved in 25 ml of distilled water separately. Both solutions were mixed and its volume was made upto 500 ml by adding distilled water. This solution was freshly made before use and was stored in brown bottle.

E. **Estimation:** To 0.1ml of sugar extract, distilled water was added and final volume made

to 1ml. Then 1 ml of newly prepared reagent C was added. The tubes kept in water bath at 60°C-70°C for 20 minutes and later cooled at room temperature. After that 1ml of reagent D (arsenomolybdate reagent) was added to the tubes and then 5ml of distilled water was added. Intensity of bluish-green colour so developed was recorded at 520 nm against reagent blank.

**Calculation:**

$$\text{Reducing sugar content (mg/g DW)} = \frac{\text{Conc. of std.} \times \text{O.D. of sample} \times \text{Total vol. of extract (ml)}}{\text{O.D. of std.} \times \text{vol. taken for estimation (ml)} \times \text{Wt. of tissue (g)}}$$

**Non-reducing sugars**

Non-reducing sugar content was determined by substracting the above calculated reducing sugar content from total soluble sugar content and was expressed in mg/g DW.

**Calculation:**

Non-reducing sugar = Total soluble sugar – Reducing sugar

(mg/g DW) (mg/g DW) (mg/g DW)

**Antioxidative enzymes**

Under stress, overproduction of reactive oxygen species (ROS) occur which are highly toxic and leads to cellular damage. To combat these ROS, antioxidative mechanisms operate in the form of enzymes and biomolecules. Enzymes were extracted at 4ºC to minimize denaturation.

**Superoxide Dismutase (EC 1.15.1.1) (Marklund and Marklund 1974)**

Superoxide dismutase (SOD) catalyzes the disproportionation of superoxide anion ($O_2^-$) to hydrogen peroxide and oxygen (molecular form).

$$O_2^- + O_2^- + 2H^+ \xrightarrow{\text{SOD}} H_2O_2 + O_2$$

**Extraction:** Weighed 0.2 g of fresh sample and extracted with 2 ml of 0.1 M sodium phosphate buffer (pH 7.5). The extract was centrifuged at 10,000 rpm for 20 minutes. Supernatant was used for estimation of superoxide dismutase, peroxidase and catalase.

**Reagents:**

(i) 6 mM Pyragallol (Fresh solution was prepared for assay)

(ii) 0.1M Tris-HCl buffer

(iii) 6 mM EDTA

**Estimation:** To a cuvette, 1.5 ml of 0.1 M Tris-HCl buffer (8.2 pH), 0.5 ml of 6 mM EDTA, 1ml of 6 mM pyragallol solution and further 0.1 ml of enzyme extract were added. The reaction mixture without the extract was taken as control. Absorbance was read at 420 nm after an interval of 30 seconds till 2.5 minutes. A unit of enzyme activity has been described as the amount of enzyme causing 50% inhibition of auto-oxidation of pyragallol observed in

blank. SOD activity has been defined as change in enzyme activity/min/g FW.

**Calculation:**

Change in absorbance (ΔA/min) = Maximum OD - Minimum OD / Time interval

Percent inhibition (%) = $\frac{\Delta A/min\ (sample) - \Delta A/min\ (control)}{\Delta A\ (sample)} \times 100 = X\ \%$ (say)

Calculate unit activity which causes 50% inhibition, say (Y unit).

50% inhibition= 1 unit

Therefore, X % inhibition= (1/50) × X = Y units

**Enzyme activity (EA/min/g FW)** = $\frac{Y\ units \times total\ volume\ of\ extract\ (ml)}{volume\ taken\ for\ estimation\ (ml) \times weight\ of\ tissue\ (g)}$

**Peroxidase (EC 1.11.1.7) (Shannon *et al* 1966)**

Peroxidase (POD) remove the excess of hydrogen peroxide from cytosol of the cell. They are not specific in utilizing electron donor for oxidation of $H_2O_2$.

Guaiacol + $H_2O_2$ $\xrightarrow{POD}$ $H_2O$ + Tetrahydroguaiacol

**Reagents:**

(i) 0.05 M guaiacol prepared in 0.1 M sodium phosphate buffer (pH 7.5)

(ii) 0.8 M $H_2O_2$

**Estimation:** The reaction mixture contained 3 ml of 0.05 M guaiacol formed in 0.1 M sodium phosphate buffer (pH 7.5), 0.1 ml of enzyme extract and 0.1 ml of 0.8 M $H_2O_2$. The reaction mixture without enzyme extract was measured as blank. The reaction was initiated by adding $H_2O_2$ and rate of change in absorbance was observed at 470 nm for 2.5 minutes at an interval of 30 seconds. POD activity has been expressed as change in EA/min/g FW or millimoles of enzyme activity/min/g FW.

**Calculation:**

Change in absorbance (ΔA/min) = Maximum OD - Minimum OD/ Time interval

Enzyme activity (mmoles/min/g FW) = $\frac{\Delta A/min \times total\ volume\ of\ extract\ (ml)}{26.6\ mM^{-1} \times volume\ taken\ for\ estimation\ (ml) \times weight\ of\ tissue\ (g)}$

**Catalase (EC 1.11.1.6) (Chance and Maehley 1955)**

Catalase (CAT) is able to use one molecule of $H_2O_2$ as substrate or electron donor and another molecule of $H_2O_2$ as oxidant or electron acceptor.

$$2\ H_2O_2 \xrightarrow{CAT} 2H_2O + O_2$$

**Reagents**

(i) 50 mM sodium phosphate buffer (pH 7.5)

(ii) $H_2O_2$ solution: 0.2 ml of $H_2O_2$ was dissolved in 50 ml with 50 mM sodium phosphate buffer (pH 7.5)

**Estimation:** In spectrophotometric cuvette of quartz, 1.8 ml of 50 mM sodium phosphate buffer (pH 7.5) and 0.1 ml of enzyme extract was added. The reaction was started by adding 1 ml of $H_2O_2$. Utilization of $H_2O_2$ was reported at intervals of 30 seconds for 2.5 minutes by measuring the decrease in absorbance at 240 nm. CAT activity was defined as mmoles of $H_2O_2$ decomposed/min/g FW or enzyme activity/min/g FW.

**Calculation:**

Change in absorbance (ΔA/min) = Maximum OD - Minimum OD/ Time interval

$$\text{Enzyme activity (mmoles/min/g FW)} = \frac{\Delta A/\text{min} \times \text{total volume of extract}}{0.039 mM^{-1} \times \text{volume taken for estimation} \times \text{weight of tissue (g)}}$$


UV 2600 spectrophotometer (Techcomp)

**Total soluble protein (Lowry *et al* 1951)**

**Reagents for extraction:** 0.1 M Sodium Phosphate buffer (pH 7.5) which is prepared as -

a) 0.1M Sodium dihydrogen phosphate (Monobasic) = 1.56 g/100 ml
b) 0.1M Disodium hydrogen phosphate (Dibasic) = 1.42 g/100 ml

**Extraction:** Weighed 0.2 g leaf sample, macerated in pestle and mortar in 2 ml of Sodium phosphate buffer (pH 7.5) and transferred the material to centrifuge tubes. Centrifuged the homogenate at 10000 rpm for 20 minutes and collected the supernatant.

**Reagents for estimation:**

1. A: 4 % Sodium carbonate (4 g/100 ml) in 0.2 N NaOH (0.8 g/100 ml)
2. B: 1% $CuSO_4.5H_2O$ (1g/100 ml) in 2% sodium potassium tartrate (2 g/100 ml)
3. Mixed reagents A and B in ratio of 50:1 to get reagent C
4. Folin-Ciocalteau's reagent diluted with distilled water in 1:1 ratio (freshly prepared just before use)

**Estimation:** 0.1 ml of supernatant was taken and diluted to 1.0ml with distilled water. 5ml of reagent C was added in each tube, the contents were shaken and after 10 minutes, 0.5ml of Folin-Ciocalteau's reagent was added. Contents were shaken and after 30 minutes, absorbance was read at 520 nm, using UV 2600 spectrophotometer (Techcomp). The concentration of protein samples was calculated from the standard curve of BSA using standard value (0.06 mg= 0.12 OD).

**Calculation:**

$$\text{Total soluble proteins (mg/g FW)} = \frac{\text{Conc. of std.} \times \text{O.D. of sample} \times \text{total volume of extract (ml)}}{\text{O.D. of std.} \times \text{fresh weight (g)} \times \text{aliquot taken (ml)}}$$

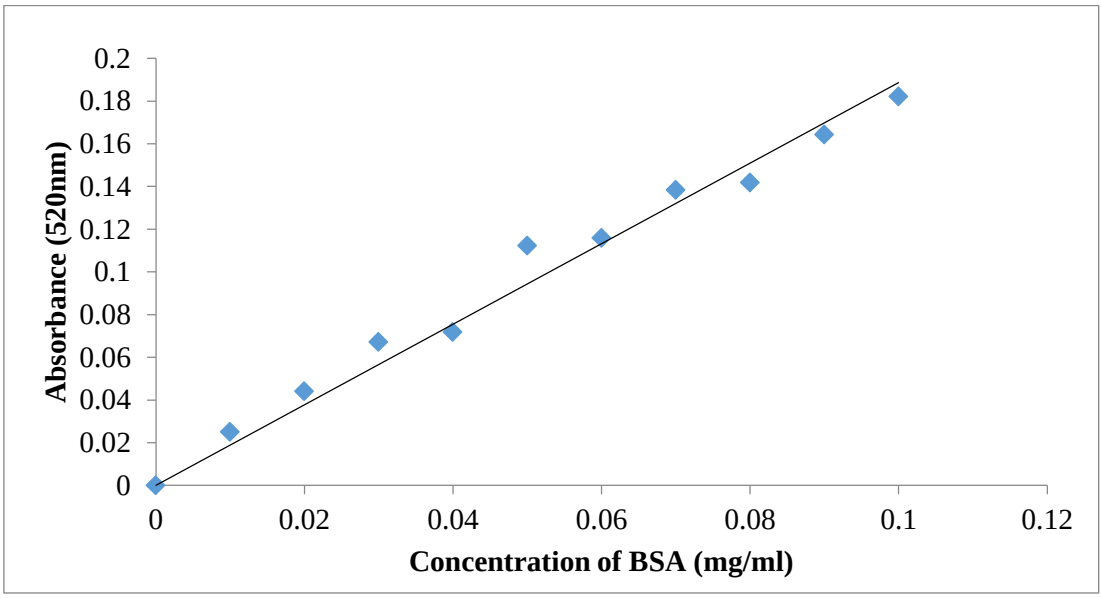



Fig. 5: Standard curve of BSA for the estimation of protein content

**Proline (Bates *et al* 1973)**

**Extraction:** 0.2 g of leaf tissue was extracted with 10 ml of 3% sulphosalycilic acid and centrifuged at 3000 rpm for 10 minutes and then supernatant was poured to fresh tube.

**Reagents:**

a) 3% sulphosalycilic acid
b) Acid ninhydrin reagent: Mixed 30 ml of glacial acetic acid and 20 ml of 6 M orthophosphoric acid, 1.25 g of ninhydrin added to it.
c) Benzene
d) 6 M orthophosphoric acid: Added 39.76 ml of orthophosphoric acid in 60.24 ml of distilled water
e) Glacial acetic acid

**Estimation:** 0.5 ml of supernatant was taken to fresh test tube, added to it 2 ml of 6 M orthophosphoric acid, 2 ml of acid ninhydrin and 2 ml of glacial acetic acid. The reaction mixture kept in boiling water bath for 45 minutes. After that, it was cooled at room temperature and 4 ml of benzene was added to it. Further, the mixture was vortexed. Two

separate layers were observed in the reaction mixture, out of these the chromophore containing pink layer on the upper side was collected. Absorbance was read at 520 nm using pure benzene as blank.

**Calculation:**

$$\text{Proline content (mg/g DW)} = \frac{\text{Conc. of std.} \times \text{O.D. of sample} \times \text{total volume of extract (ml)}}{\text{O.D. of std.} \times \text{volume taken for estimation (ml)} \times \text{weight of tissue (g)}}$$

**Malondialdehyde (Heath and Packer 1968)**

Lipid peroxidation was measured in terms of malondialdehyde (MDA) content. Lipid peroxidation products are considered useful and reliable indication of oxidative damage ocuured due to attack of reactive oxygen species on membrane. MDA is a secondary end product of oxidative polyunsaturated fatty acids and react with TBA to yield a pinkish red chromatogen with maximal absorbance at 532 nm.

**Extraction**: Weighed 0.2 g tissue, homogenized with 2 ml of 5% trichloroacetic acid (TCA) and centrifuged at 15,000 rpm for 15 minutes Supernatant was transferred.

**Reagents:**

(i) TBA-TCA solution: Mixed 0.5 g Thiobarbutaric acid (TBA) in 20% Trichloro acetic acid (TCA) solution with the help of NaOH solution

(ii) 5% Trichloroacetic acid (TCA) for extraction

**Estimation:** 1ml of TBA-TCA solution was added to 1 ml of supernatant. The mixture was heated for 30 min in a water bath at 95°C. After heating the mixture was put on ice. The samples were then centrifuged at 10,000 rpm for 10-15 minutes. Absorbance of the filtrate was read at 532 nm and 600 nm on UV spectrophotometer using TBA-TCA solution as blank. The result was expressed as µmoles MDA/g FW.

**Calculation:**

$$\text{MDA content} = \frac{\Delta A \times \text{total volume of extract (ml)} \times 1000}{\text{extinction coefficient} \times \text{volume of aliquot (ml)} \times \text{wt. of tissue (g)}}$$

Where, extinction coefficient = 155 $mM^{-1}cm^{-1}$

**Yield attributes**

At physiological maturity, five plants of each genotype were selected randomly from each treatment per replication to record yield attributes.

**Plant height**

At physiological maturity stage, plant height was measured from the base of plant to the tip of the main shoot. The values from five plants from each treatment were averaged and represented as mean of plant height.

**Main shoot length**

Main shoot length was measured from the base of last formed branch on the main

shoot to the tip of the main shoot at its physiological maturity. The values from five plants from each treatment were averaged and represented as mean of main shoot length.

**Number of primary and secondary branches per plant**

At physiological maturity, 5 plants from each treatment per replication were selected and number of primary branches and secondary branches of each plant were counted. The values of 5 plants were averaged and considered as mean number of primary and secondary branhes.

**Siliquae on main shoot and total siliquae per plant**

At physiological maturity, 5 plants from each treatment per replication were selected and siliquae on main shoot and total siliquae of each plant were counted. The values of 5 plants were averaged and considered as mean number of siliquae on main shoot and total siliquae per plant.

**1000 Seed weight**

After threshing the crop, a representative sample of seeds was obtained from bulk of the whole plant. One thousand seeds were counted and weighed to record seed weight in grams.

**Siliqua length and seed filling**

Before harvesting, 25 siliquae were collected randomly from each treatment, replication wise. Length of the siliqua was measured with the scale and averaged to represent the mean siliqua length in cms. After measuring the siliqua length, the number of shrivelled and developed seeds were counted. From this, mean number of shrivelled seeds per siliqua and developed seeds per siliqua were calculated and expressed in percentage.

**Biological yield**

After thorough drying and before threshing of the harvested crop, the biological yield consisted of the seed and stover from each treatment per net plot, that was weighed and expressed as kilogram per hectare (kg/ha).

**Seed yield**

Seeds obtained after threshing of the dried produce per net plot was cleaned and weighed to give seed yield which was converted to kg/ha.

**Harvest index**

Harvest index is expressed as ratio of seed yield to biological yield.

**Nuclear magnetic resonance (NMR)**

Oil content in seed samples was estimated by Nuclear Magnetic Resonance (NMR) method. Standardized the Newport Analyzer (Model MKIII A) with standard sample (4g seeds with a known oil content) and brought the read out values at the desired oil percent by

operating the instrument at the following conditions: gate width 1.5 gs; Rf level = 100 µ Amp; Integration time = 32 seconds. The dried seeds of the unknown sample were weighed down (4g) and the value of oil content was noted. Thus, the oil content (%) in seeds was obtained directly.

**Drought susceptibility index (Fischer and Maurer, 1978)**

Drought susceptibility index was measured on the basis of seed yield under stressed (rainfed) and non-stressed condition (irrigated) using following formula:

$$DSI = \frac{1-(Ys_i/Yp_i)}{1-(Ys/Yp)}$$

**Drought tolerance index (Fernandez 1992)**

Drought tolerance index was measured based on seed yield under stressed (rainfed) and non-stressed condition (irrigated) by using following formula:

$$DTI = \frac{Ys_i \times Yp_i}{(Yp)^2}$$

**Drought tolerance efficiency (Fischer and Wood, 1981)**

$DTE = (Ys_i / Yp_i) \times 100$

Where;

$Yp_i$: Seed yield of each genotype under non-stressed/irrigated condition

$Ys_i$: Seed yield of each genotype under stressed/rainfed condition

Yp: Mean of yield of all genotypes under non-stressed/irrigated condition

Ys: Mean of yield of all genotypes under stressed/rainfed condition

**Sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) (Laemmli 1970)**

**Extraction:** Protein from the fresh leaves (0.2g) was extracted in 2 ml of 20 mM Tris-HCL buffer containing 0.5% NaCl (pH 7.5). The extract was centrifuged at 8000 rpm for 20 minutes. The supernatant was used for protein estimation with the method described by Lowry *et al* (1951). A part of the supernatant was used for SDS PAGE.

**Reagent for SDS-PAGE**

1. **Stock solutions**

   a.) **Acrylamide-Bisacrylamide stock solution (30%)**

   Dissolve 14.6 g of acrylamide and 0.4 g of bis-acrylamide in 50 ml of distilled water. This solution was stored in brown bottle at 4° C.

   b.) **Resolving gel buffer (1.5 M Tris-HCL, pH 8.8)**

   Dissolve 18.17 g of Tris Base in 80 ml of distilled water. The pH was adjusted with HCL and final volume was made up to 100 ml.

   c.) **Stacking gel buffer (0.5 M Tris-HCL, pH 6.8)**

Dissolve 6.05 g of Tris Base in 80 ml of distilled water. The pH was adjusted with HCL and its final volume was made up to 100 ml.

**d.) 10% Sodium dodecyl sulphate (SDS)**

Dissolve 1 g of SDS in 10 ml of distilled water.

**e.) 10% Ammonium persulphate (APS) (freshly prepared in eppendrof)**

Dissolve 0.2 g of APS in 2 ml of distilled water

**f.) N, N, N', N'-tetramethylethylenediamine (TEMED)**

**g.) Running Buffer 1X**

Dissolve 3.03 g of Tris Base, 14.4 glycine and 1 g of SDS in 1 L of distilled water.

**h.) Staining solution**

To make 100 ml of staining solution add 50 ml of water, 40 ml of methanol, 10 ml of acetic acid and 125 mg of coomassie brilliant blue (CBB-R-250)

**i.) Destaning solution**

Mix 200 ml of methanol and 35 ml of acetic acid. The final volume was made up to 500 ml with distilled water.

**j.) Sample dye (4X)**

To make 10 ml of sample dye, add 2.8 ml of Tris Base (pH 6.8), 4.48 ml of glycerol, 1.2 gm of SDS, 12 mg of bromophenol blue, 2 ml of mercaptoethanol and 3.6 ml of distilled water.

**Working solution**

| **Solution** | **Resolving gel (12%)** | **Stacking gel (4%)** |
|---|---|---|
| Distilled water | 1.57 ml | 1.48 ml |
| Tris Base (pH 8.8) | 1.25 ml | _ |
| Tris Base (pH 6.8) | _ | 625 µl |
| Acrylamide-Bisacrylamide | 2.08 ml | 325 µl |
| SDS | 50 µl | 25 µl |
| APS | 37.5 µl | 37.5 µl |
| TEMED | 6 µl | 8 µl |

**Gel preparation**

The above solution was prepared one by one. The stacking should be added after the polymerization of resolving gel. Check the leakage before gel casting between plates. Avoid bubbles between the resolving and stacking gel and also between the wells.

**Sample preparation and loading**

1. The above prepared protein samples containing a known amount of protein was

mixed with an equal sample dye volume respectively and boiled for 3 minutes.

2. The samples alongwith molecular weight marker were loaded on to the wells.
3. The electrophoresis was run at constant voltage of 70 V until the sample travel through the stacking gel. The voltage was increased to 100 V when the bromophenol blue moved into resolving gel and continued till the dye reached at the bottom of gel.

After completion of electrophoresis, immerse the gel in staining solution for overnight or for 12 hours in dark conditions to avoid crystallization. Destaining was done by immersing gel in destaining solution for 24 hours.

**Statistical analysis**

In order to test the relative performance of genotypes and the significance of treatments the data recorded in the field and laboratory at various crop growth stages were statistically analysed using computer programme CPCS (2008). The correlation coefficient was statistically analysed using OPSTAT software.

**CHAPTER-IV**

## RESULTS AND DISCUSSION

The results of present study entitled "Differential response of Indian mustard (*Brassica juncea* L. Czern & Coss) genotypes under rainfed and irrigated conditions" are discussed in this chapter under the following headings:

### Phenological traits

In the present study, an adverse effect of water deficit was seen on the phenophases of Indian mustard genotypes. A significant decrease in days to flowering, siliquing, reproductive phase and days to maturity was observed under rainfed condition as compared to irrigated condition. Moreover, interactions (I×G) were also significant.

### Flowering behaviour

### Initiation of flowering

Flowering initiation occurred on average 1.6 days earlier under rainfed than irrigated condition (Table 1). However, under irrigated condition JC 210-335 took lesser days (48.7) and RH 406 took more days (66.0) for flower initiation whereas under rainfed condition, JC 210-335 took lesser days (45.0) and RH 406 took more days (64.7). Genotypic mean indicated variability for flower initiation from 46.8 days in JC 210-335 to 65.3 days in RH 406. In a similar study, Birunara *et al* (2011) observed in *Brassica napus* that water deficit condition applied at flowering stage reduced the time of onset of flowering which led to diminished time period between flowering and siliquae formation. Similar trend was observed in groundnut by Mabhaudhi and Modi (2013).

### 50% flowering

50% flowering on average occurred 2.1 days earlier under rainfed as compared to irrigated condition (Table 1). Under irrigated condition, 50% flowering in JC 210-335 took lesser days (61.7) and MCN 09-40 took more days (76.0) while under rainfed condition, JC 210-335 took lesser days (58.0) and both MCN 09-40 and MLM 41-13-2 took more days (73.3). From genotypic mean, it was observed that days to 50% flowering ranged from 59.8 (JC 210-335) to 74.7 (MCN 09-40). Similarly, Sodani *et al* (2017) observed lesser days to 50% flowering under drought condition as compared to control and reduction was highest in NRCDR-02 (31.7%) and lowest in URVASHI (1.3%). Similar trend was observed by Sharma and Sardana (2016) in Brassica species.

### Flowering completion

Flowering completion on average took 5 days lesser under rainfed over irrigated condition (Table 1). Under irrigated condition, days to 100% flowering ranged from 76.0 (RH 1518) to 92.3 (RH 406) whereas under rainfed condition range was 67.7 (JC 210-335) to 85.7 (MLM 41-13-2) days. Genotypic mean ranged from 72.5 (JC 210-335) to 88.8 (MLM 41-13-

2) days. Similarly, Zirgoli and Kahrizi (2015) reported the significant decline by 1.71% in mean number of days to end of flowering in rapeseed genotypes under drought over normal condition. Completion of flowering was delayed by shading in *B. juncea* (Kaur 2018).

**Flowering duration**

Flowering duration declined by 3.3 days on average under rainfed condition as compared to irrigated condition (Table 1; Fig. 6). Under irrigated condition, flowering duration was shorter in RH 1518 (17.0 days) and longer in CJRD 1261 (35.0 days) while under rainfed condition, it was shorter in RH 1518 (16.0 days) and longer in both ELM 38 and MLM 41-13-2 (24.7 days). Genotypic mean indicated that flowering duration ranged from 16.5 days (RH 1518) to 29.0 days (CJRD 1261). Mabhaudhi and Modi (2013) observed that Bambara groundnut landraces had shorter flowering duration under rainfed relative to irrigated condition. Kannu Priya (2019) reported the shortening of flowering duration in *B. juncea* in response to salt stress.

**Siliquing behaviour**

**Initiation of siliqua**

Moisture stress led to decrease in days to initiation of siliqua (Table 2). Under rainfed condition, initiation of siliqua occurred on an average of 1.2 days earlier than irrigated condition. Under irrigated condition, JC 210-335 (58.0) took lesser days and RH 406 (70.0) took more days out of different genotypes and under rainfed condition, JC 210 -335 (53.3) took lesser days and both RB-50 and RH 406 (69.0) took more days for initiation of siliqua. Genotypic mean ranged from 55.7 (JC 210-335) to 69.5 (RH 406) days (Table 2). Similarly, Kannu Priya (2019) reported the decline in days to initiation of siliquing in *B. juncea* under salt stress. However, *B. juncea* genotypes under shading stress took more days for siliqua initiation (Kaur 2018).

**50% siliquing**

50% siliquing on an average occurred 5.3 days earlier under rainfed as compared to irrigated condition (Table 2). Under irrigated condition, 50% siliquing in RH 1518 took lesser days (75.7) and MLM 41-13-2 took more days (98.7) while under rainfed condition, JC 210-335 took lesser days (74.3) and MLM 41-13-2 took more days (92.0). From genotypic mean, it was observed that days to 50% siliquing ranged from 75.3 (RH 1518) to 95.3 (MLM 41-13-2). Similar trend of 50% siliquing was reported by Birunara *et al* (2011) in canola under different irrigation treatments. However, Kaur (2016) observed decreased days to 50% siliquae formation under late sown *B. carinata*.

Table 1: Effect of moisture stress on flowering behaviour (days) of *B. juncea* genotypes

| Genotypes/ Treatment | Initiation of flowering | | | 50% flowering | | | Flowering completion | | | Flowering duration | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** |
| RH 1518 | 60.0±0.6 | 59.3±0.3 | **59.7** | 68.7±0.9 | 67.7±0.3 | **68.2** | 76.0±0.1 | 74.3±1.5 | **75.2** | 17.0±0.6 | 16.0±0.6 | **16.5** |
| JC 210-335 | 48.7±0.3 | 45.0±0.4 | **46.8** | 61.7±0.3 | 58.0±0.1 | **59.8** | 77.3±1.7 | 67.7±0.7 | **72.5** | 29.7±0.6 | 23.7±0.7 | **26.7** |
| CJRD 1261 | 54.7±0.7 | 54.3±0.7 | **54.5** | 68.3±0.3 | 66.7±0.3 | **67.5** | 88.7±0.7 | 76.3±1.8 | **82.5** | 35.0±0.1 | 23.0±0.1 | **29.0** |
| RB-50 | 64.3±0.3 | 63.7±0.3 | **64.0** | 71.3±0.3 | 70.3±0.3 | **70.8** | 82.3±3.2 | 80.3±1.7 | **81.3** | 19.0±0.1 | 17.7±0.7 | **18.3** |
| RH 406 | 66.0±0.2 | 64.7±0.3 | **65.3** | 72.7±0.3 | 72.0±0.6 | **72.3** | 92.3±0.7 | 82.3±1.9 | **87.3** | 27.3±0.3 | 18.7±0.7 | **23.0** |
| PBR 422 | 64.0±0.1 | 61.7±0.3 | **62.8** | 74.3±0.3 | 69.7±0.9 | **72.0** | 86.3±0.3 | 83.3±2.4 | **84.8** | 23.3±0.3 | 22.7±0.7 | **23.0** |
| ELM 38 | 60.0±0.3 | 59.7±0.3 | **59.8** | 72.7±0.7 | 68.0±1.5 | **70.3** | 86.3±0.3 | 83.3±2.3 | **84.8** | 27.3±0.3 | 24.7±0.7 | **26.0** |
| CSR 1163 | 62.3±0.3 | 60.7±0.7 | **61.5** | 70.0±0.1 | 69.0±0.1 | **69.5** | 82.7±0.7 | 79.3±2.3 | **81.0** | 21.3±0.3 | 19.7±0.3 | **20.5** |
| IAN | 61.3±0.7 | 59.0±0.5 | **60.2** | 67.7±0.3 | 67.3±0.3 | **67.5** | 78.7±1.8 | 76.0±0.1 | **77.3** | 18.3±0.3 | 18.0±0.1 | **18.2** |
| MCN 09-40 | 64.7±0.3 | 62.0±0.1 | **63.3** | 76.0±0.1 | 73.3±0.3 | **74.7** | 87.0±1.0 | 83.7±0.9 | **85.3** | 23.3±0.3 | 22.7±0.3 | **23.0** |
| MLM 41-13-2 | 63.0±0.6 | 62.0±0.2 | **62.5** | 74.7±0.3 | 73.3±0.3 | **74.0** | 92.0±0.1 | 85.7±2.4 | **88.8** | 30.0±0.6 | 24.7±0.3 | **27.3** |
| PBR 357 | 65.0±0.6 | 63.3±0.7 | **64.2** | 73.0±0.1 | 70.7±0.3 | **71.8** | 86.3±0.7 | 83.7±4.3 | **85.0** | 22.3±0.3 | 21.3±0.7 | **21.8** |
| **Average** | **61.2±0.5** | **59.6±0.4** | | **70.9±0.4** | **68.8±0.4** | | **84.7±0.9** | **79.7±1.8** | | **24.5±0.3** | **21.1±0.5** | |
| CD (p=0.05) | I= 0.344, G=0.842, I×G= 1.191 | | | I= 0.421, G= 1.031, I×G= 1.459 | | | I= 1.448, G= 3.547, I×G= 5.017 | | | I= 1.456, G= 3.567, I×G= 5.044 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

Table 2: Effect of moisture stress on siliquing behaviour (days) of *B. juncea* genotypes

| Genotypes/ Treatment | Initiation of siliqua | | | 50% siliquing | | | Siliquing completion | | | Siliquing duration | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** |
| RH 1518 | 66.0±0.1 | 65.3±0.3 | **65.7** | 75.7±1.3 | 75.0±0.6 | **75.3** | 111.3±0.7 | 109.7±0.7 | **110.5** | 46.3±0.7 | 45.3±0.3 | **45.8** |
| JC 210-335 | 58.0±0.2 | 53.3±0.3 | **55.7** | 85.0±2.3 | 74.3±1.3 | **79.7** | 109.7±0.9 | 104.3±0.3 | **107.0** | 52.6±0.3 | 52.0±0.1 | **52.3** |
| CJRD 1261 | 61.3±0.3 | 60.7±0.3 | **61.0** | 92.7±2.8 | 87.0±2.1 | **89.8** | 120.7±0.3 | 119.7±0.3 | **120.2** | 60.3±0.3 | 60.0±0.6 | **60.2** |
| RB-50 | 69.3±0.7 | 69.0±0.6 | **69.2** | 88.3±3.0 | 85.7±1.8 | **87.0** | 116.3±0.3 | 112.0±0.1 | **114.2** | 48.0±0.6 | 44.0±0.6 | **46.0** |
| RH 406 | 70.0±0.1 | 69.0±0.6 | **69.5** | 97.3±0.9 | 89.3±2.4 | **93.3** | 116.7±0.3 | 110.7±0.3 | **113.7** | 47.7±0.3 | 42.7±0.3 | **45.2** |
| PBR 422 | 68.3±0.3 | 67.7±0.3 | **68.0** | 92.3±0.3 | 88.0±2.6 | **90.2** | 119.7±0.3 | 112.7±0.7 | **116.2** | 52.3±0.3 | 46.0±0.6 | **49.2** |
| ELM 38 | 67.7±0.3 | 65.7±0.7 | **66.7** | 91.7±0.9 | 89.3±1.8 | **90.5** | 122.7±0.3 | 117.0±0.6 | **119.8** | 56.0±0.1 | 52.3±0.3 | **54.2** |
| CSR 1163 | 68.0±0.6 | 67.0±0.1 | **67.5** | 86.0±2.1 | 83.3±4.3 | **84.7** | 118.7±0.3 | 111.0±0.6 | **114.8** | 51.7±0.9 | 45.0±0.6 | **48.3** |
| IAN | 66.0±0.3 | 64.0±0.2 | **65.0** | 98.0±1.5 | 81.0±0.6 | **89.5** | 125.3±0.7 | 122.7±0.3 | **124.0** | 60.3±0.3 | 59.7±0.3 | **60.0** |
| MCN 09-40 | 68.3±0.7 | 67.7±0.3 | **68.0** | 91.0±2.3 | 90.0±2.1 | **90.5** | 120.0±0.1 | 113.3±0.3 | **116.7** | 52.7±0.3 | 46.7±0.3 | **49.7** |
| MLM 41-13-2 | 68.7±0.3 | 68.0±0.1 | **68.3** | 98.7±0.7 | 92.0±2.5 | **95.3** | 122.7±0.1 | 121.0±0.6 | **122.0** | 55.0±0.6 | 54.0±0.6 | **54.5** |
| PBR 357 | 69.0±0.1 | 68.3±0.3 | **68.7** | 92.0±2.1 | 89.3±3.7 | **90.7** | 121.7±0.3 | 115.0±0.1 | **118.3** | 53.7±0.3 | 47.7±0.3 | **50.7** |
| **Average** | **66.7±0.4** | **65.5±0.4** | | **90.7±1.7** | **85.4±2.1** | | **118.7±0.4** | **114.1±0.4** | | **53.1±0.4** | **49.6±0.4** | |
| CD (p=0.05) | I= 0.311, G= 0.761, I×G= 1.077 | | | I= 1.784, G= 4.370, I×G= 6.180 | | | I= 0.377, G= 0.923, I×G= 1.306 | | | I= 0.476, G= 1.167, I×G= 1.651 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

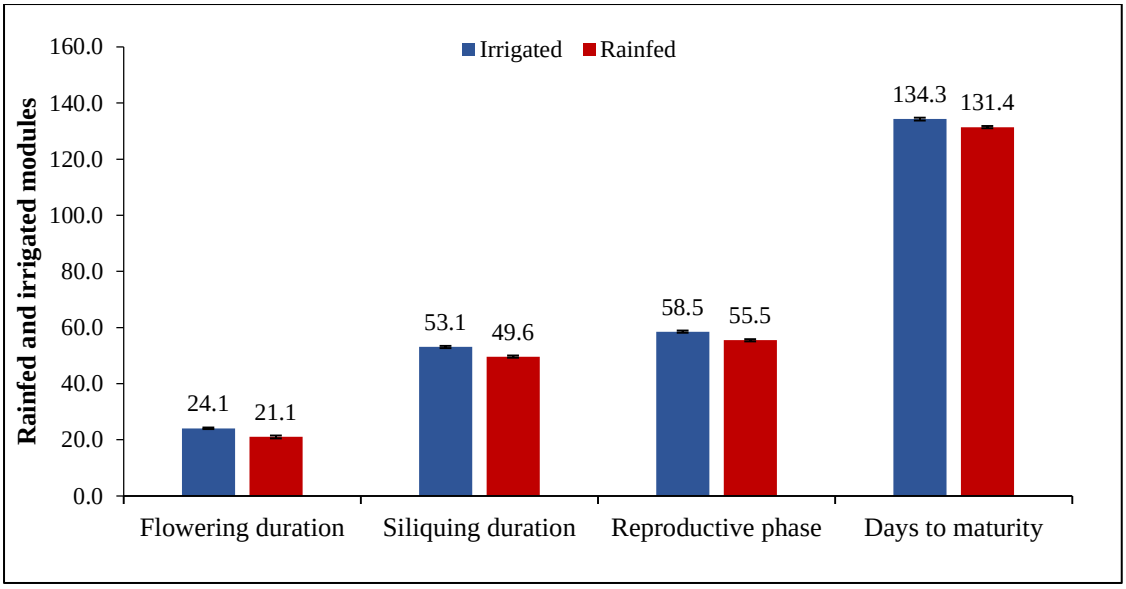



Fig. 6: Effect of moisture stress on mean phenophases in *B. juncea*

**Siliquing completion**

Average siliquing completion indicated that completion of siliquae formation took 5.3 days earlier under rainfed than irrigated condition (Table 2). Under irrigated condition, days to 100% siliquing ranged from 109.7 (JC 210-335) to 125.3 (IAN) and under rainfed condition, from 104.3 (JC 210-335) to 122.7 (IAN). Genotypic mean showed the variation among genotypes from 107.0 (JC 210-335) to 124.0 (IAN) days. Sharma and Sardana (2013) reported the decrease in days to 100% siliquae formation under late sown (105.9 days) as compared to timely sown (110.3 days) *Brassica juncea* and later by Kaur (2016) in *B. carinata*.

**Siliquing duration**

Siliquing duration on an average was 3.5 days shorter under rainfed (49.6) than irrigated (53.1) condition. The range of siliquing duration was 46.3 (RH 1518) to 60.3 days (CJRD 1261 and IAN) days under irrigated and from 42.7 (RH 406) to 60.0 days (CJRD 1261) under rainfed conditions. Genotypic mean showed that RH 406 had shortest (45.2) and CJRD 1261 had longest (60.2) siliquing duration (Table 2; Fig. 6). Similarly, Sehgal *et al* (2017) reported the decrease in podding duration under drought as well as heat stress in comparison to control in lentil genotypes. Kaur (2018) also reported decline in siliquing duration in *B. juncea* genotypes under shading stress.

**Reproductive phase**

Earlier flowering and siliquing behaviour of Indian mustard genotypes under rainfed condition/moisture stress led to shortening of flowering and siliquing duration and thus, shortening of reproductive phase. On an average, reproductive phase was 3.1 days shorter under rainfed as compared to irrigated condition. Reproductive phase ranged from 51.7 (RH

406) to 67.0 (CJRD 1261) days under irrigated condition and from 47.0 (RH 406) to 66.3 (CJRD 1261) days under rainfed condition. From the genotypic mean, it was concluded that RH 406 (49.3) had shortest and CJRD 1261 (66.7) had longest reproductive phase (Table 3; Fig. 6). Kumar *et al* (2017b) observed shorter reproductive phase under late sown condition in Indian mustard genotypes and also reported earlier by Kaur (2016) in *B. carinata*. Similarly, the reproductive phase of *B. juncea* shortened with increasing salinity levels as observed by Kannu Priya (2019).

Table 3: Effect of moisture stress on reproductive phase (days) and days to maturity

| **Genotypes/ Treatment** | **Reproductive phase** | | | **Days to maturity** | | |
|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Mean** | **Irrigated** | **Rainfed** | **Mean** |
| RH 1518 | 52.3±1.1 | 51.3±1.1 | **51.8** | 133.7±2.3 | 131.0±0.1 | **132.3** |
| JC 210-335 | 62.0±0.7 | 60.3±0.4 | **61.2** | 131.3±0.3 | 130.7±0.7 | **131.0** |
| CJRD 1261 | 67.0±0.7 | 66.3±1.1 | **66.7** | 134.0±0.2 | 130.3±0.3 | **132.2** |
| RB-50 | 53.0±0.7 | 49.3±0.4 | **51.2** | 137.3±0.3 | 131.7±0.7 | **134.5** |
| RH 406 | 51.7±0.4 | 47.0±0.7 | **49.3** | 133.7±0.3 | 131.3±0.3 | **132.5** |
| PBR 422 | 56.7±0.4 | 52.0±0.7 | **54.3** | 136.0±0.2 | 134.7±0.3 | **135.3** |
| ELM 38 | 63.7±0.4 | 58.3±0.8 | **61.0** | 136.7±0.3 | 129.7±0.3 | **133.2** |
| CSR 1163 | 57.3±0.8 | 51.3±1.5 | **54.3** | 132.3±0.3 | 131.3±0.3 | **131.8** |
| IAN | 65.0±1.4 | 64.7±0.4 | **64.8** | 133.7±0.3 | 130.7±0.3 | **132.2** |
| MCN 09-40 | 56.3±0.4 | 52.3±0.4 | **54.3** | 134.7±0.3 | 133.3±0.3 | **134.0** |
| MLM 41-13-2 | 60.7±0.8 | 60.0±0.7 | **60.3** | 132.0±0.6 | 130.0±0.6 | **131.0** |
| PBR 357 | 57.7±0.4 | 52.7±0.8 | **55.2** | 136.7±0.3 | 132.0±0.6 | **134.3** |
| **Average** | **58.6±0.7** | **55.5±0.8** | | **134.3±0.5** | **131.4±0.9** | |
| CD (p=0.05) | I= 0.528, G= 1.293, I×G= 1.829 | | | I= 0.510,G= 1.250, I×G= 1.767 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

**Days to maturity**

Under rainfed condition, genotypes matured on an average 2.9 days earlier than irrigated condition. Days to maturity ranged from 131.3 (JC 210-335) to 137.3 (RB-50) days under irrigated condition and from 129.7 (ELM 38) to 133.3 (MCN 09-40) under rainfed condition. Genotypic mean showed that JC 210-335 and MLM 41-13-2 (131.0) required lesser days to maturity and PBR 422 (135.3) showed more days to maturity (Table 3, Fig. 6). Days to maturity were lesser in crop grown under arid areas than under irrigated areas in rapeseed and mustard (Mustafa *et al* 2018). Similar findings were observed by Ihsan *et al* (2016) in wheat. However, days to maturity increased with increase in N level according to Gill (2018) in *B. napus*.

**Correlation of phenological traits**

Positive association existed between days to initiation of flowering and seed yield under irrigated (r= 0.562) and rainfed (r= 0.781**) conditions however, the association was

highly significant under rainfed condition. 50% flowering was positively and significantly associated with seed yield under irrigated (r= 0.700*) and rainfed (r= 0.697*) conditions. Flowering completion and seed yield were positively associated under irrigated (r= 0.440) and rainfed (r= 0.678*) conditions. Negative correlation existed between flowering duration and seed yield under irrigated (r= -0.083) and rainfed (r= -0.215) conditions. Under irrigated condition, siliqua initiation had non-significant positive correlation (r= 0.566) with seed yield while under rainfed condition highly significant correlation (r= 0.784**) existed. 50% siliquing was negatively but weakly associated (r= -0.034) with seed yield under irrigated condition but positively associated (r= 0.496) under rainfed condition. Weak positive association was observed between siliquing completion and seed yield under both irrigated (r= 0.245) and rainfed (r= 0.127) conditions. Siliquing duration was weakly and negatively correlated with seed yield under irrigated (r= -0.217) and rainfed (r= -0.486) conditions. Similar trend was found between reproductive phase and seed yield under irrigated (r= -0.328) and rainfed (r= -0.563) conditions. Moreover, days to maturity were positively associated with seed yield under irrigated (r= 0.530) and rainfed (r= 0.502) conditions. Initiation of flowering was strongly and positively correlated to 50% flowering under irrigated (r= 0.849**) as well as under rainfed (0.930**) conditions. Initiation of flowering showed positive association to flowering completion under rainfed condition (r= 0.843**) only. Moreover, 50% flowering had significant positive association with flowering completion under irrigated (r= 0.697*) and rainfed (r= 0.899**) conditions. Flowering initiation had significant positive correlation with siliqua initiation under irrigated (r= 0.977**) and rainfed (r= 0.994**) conditions. Similarly, 50% flowering was strongly and positively associated with siliqua initiation under irrigated (r= 0.854**) and rainfed (r= 0.935**) conditions. Under rainfed condition, initiation of flowering (r= 0.655*) and 50% flowering (r= 0.780**) was positively correlated with 50% siliquing. Flowering completion was positively and significantly correlated with siliqua initiation (r= 0.862**) and 50% siliquing (r= 0.920**) under rainfed condition whereas under irrigated condition, it was significantly correlated with 50% siliquing only (r= 0.705*). Under irrigated condition, 50% siliquing was strongly and positively correlated with siliquing completion (r= 0.758**). Moreover, siliquing duration had strong positive association with reproductive phase under irrigated (0.954**) and rainfed conditions (r= 0.988**) (Table 4).

Days to initiation of flowering ($R^2$= 0.6111) and siliqua initiation ($R^2$= 0.6144) had strong relationship with seed yield under rainfed condition but under irrigated condition flowering initiation ($R^2$= 0.3142) and siliquing initiation ($R^2$= 0.3215) had weak relationship with seed yield (Fig. 7). Sabaghnia *et al* (2010) also observed similar correlations between phenological traits and seed yield in canola under non-stressed and water-stressed environment. Sharma and Sardana (2016) reported that days to 50% flowering, 100% flowering and maturity were positively correlated with seed yield in Brassica cultivars.

Table 4: Correlation coefficients of phenological parameters under IR (irrigated) and RF (rainfed) conditions

| **Irrigated** | **FI** | **50% F** | **FC** | **FD** | **SI** | **50% S** | **SC** | **SD** | **RP** | **M** | **SY** |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **FI** | 1 | | | | | | | | | | |
| **50% F** | 0.849** | 1 | | | | | | | | | |
| **FC** | 0.453 | 0.697* | 1 | | | | | | | | |
| **FD** | -0.474 | -0.093 | 0.571 | 1 | | | | | | | |
| **SI** | 0.977** | 0.854** | 0.457 | -0.449 | 1 | | | | | | |
| **50% S** | 0.340 | 0.409 | 0.705* | 0.381 | 0.305 | 1 | | | | | |
| **SC** | 0.445 | 0.532 | 0.493 | 0.073 | 0.442 | 0.758** | 1 | | | | |
| **SD** | -0.347 | -0.156 | 0.131 | 0.444 | -0.369 | 0.532 | 0.671* | 1 | | | |
| **RP** | -0.575 | -0.347 | 0.007 | 0.533 | -0.556 | 0.361 | 0.477 | 0.954** | 1 | | |
| **M** | 0.485 | 0.490 | 0.142 | -0.304 | 0.500 | 0.067 | 0.304 | -0.100 | -0.197 | 1 | |
| **SY** | 0.562 | 0.700* | 0.440 | -0.083 | 0.566 | -0.034 | 0.245 | -0.217 | -0.328 | 0.530 | 1 |
| **Rainfed** | **FI** | **50% F** | **FC** | **FD** | **SI** | **50% S** | **SC** | **SD** | **RP** | **M** | **SY** |
| **FI** | 1 | | | | | | | | | | |
| **50% F** | 0.930** | 1 | | | | | | | | | |
| **FC** | 0.843** | 0.899** | 1 | | | | | | | | |
| **FD** | -0.318 | -0.094 | 0.242 | 1 | | | | | | | |
| **SI** | 0.994** | 0.935** | 0.862** | -0.274 | 1 | | | | | | |
| **50% S** | 0.655* | 0.780** | 0.920** | 0.439 | 0.661* | 1 | | | | | |
| **SC** | 0.308 | 0.410 | 0.426 | 0.189 | 0.276 | 0.506 | 1 | | | | |
| **SD** | -0.484 | -0.346 | -0.276 | 0.381 | -0.517 | -0.050 | 0.681* | 1 | | | |
| **RP** | -0.596* | -0.450 | -0.361 | 0.434 | -0.618* | -0.133 | 0.580* | 0.988** | 1 | | |
| **M** | 0.328 | 0.302 | 0.297 | -0.066 | 0.351 | 0.186 | -0.289 | -0.522 | -0.523 | 1 | |
| **SY** | 0.781** | 0.697* | 0.678* | -0.215 | 0.784** | 0.496 | 0.127 | -0.486 | -0.563 | 0.502 | 1 |

*Significant at 5%, **Significant at 1%, IF- Initiation of flowering, 50% F- 50% flowering, FC- Flowering completion, FD- Flowering duration, SI- Siliqua initiation, 50% S- 50% siliquing, SC- Siliquing completion, SD- Siliquing duration, RP- Reproductive phase, M- Maturity, SY- Seed yield

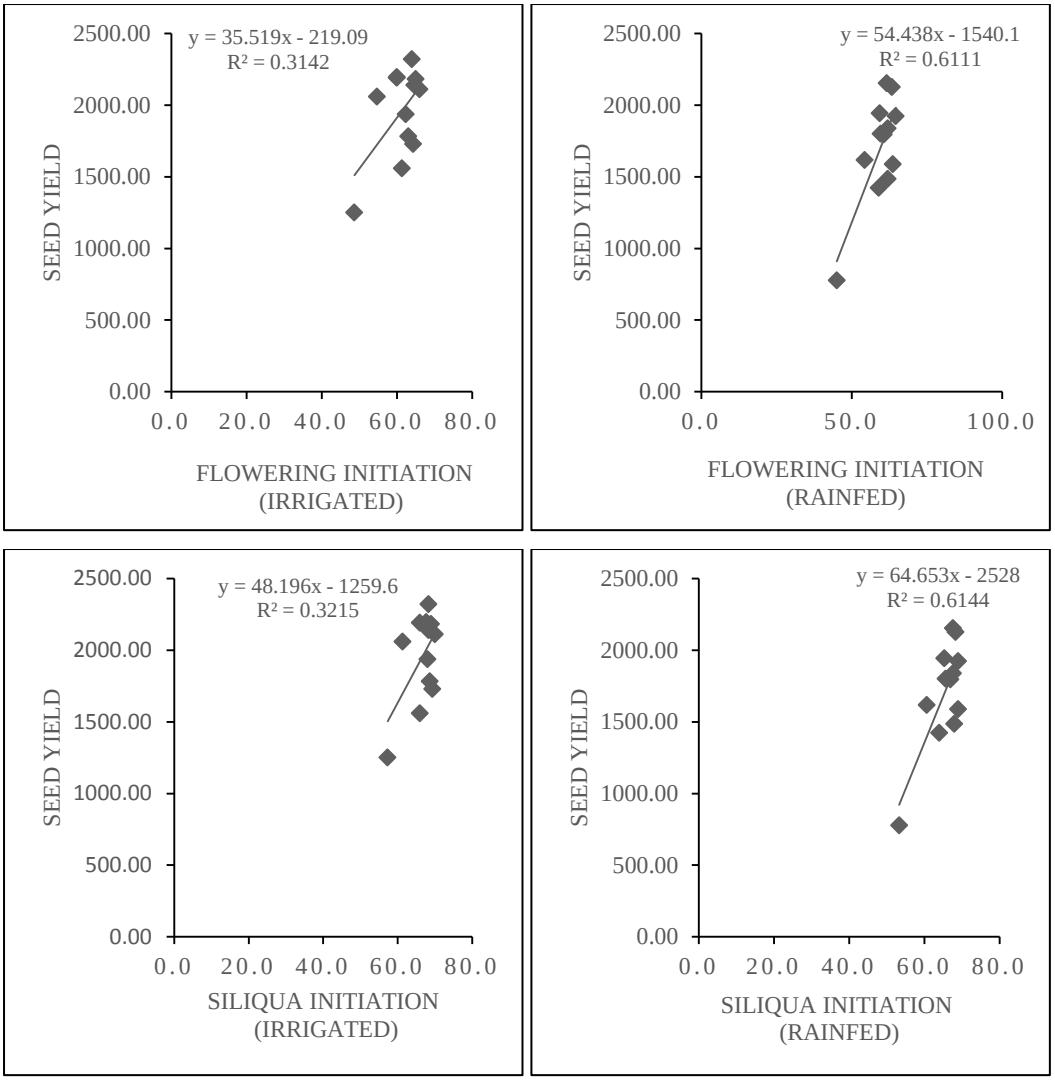



Fig. 7: Relationship of flowering and siliqua initiation with seed yield in *B. juncea*

**Physiological traits**

**Photosynthetic pigments**

Significant differences existed for photosynthetic pigments under stress (RF) and irrigated (IR) modules and their interactions (I×G) were significant. Pigments were reduced under rainfed condition. Moisture stress resulted in reduced photosynthetic pigments reduced by <20% in CJRD 1261, RB-50 and CSR 1163 (Table 5, Fig. 8).

**Chlorophyll a**

Chlorophyll a was reduced under moisture stress by 14.1% as compared to irrigated condition. Under irrigated condition, it ranged from 1.42 (MLM 41-13-2) to 1.77 mg/g FW (ELM 38) and from 1.18 (MLM 41-13-2) to 1.52 (ELM 38) mg/g FW under rainfed condition. Out of all genotypes, RH 406 faced least reduction in chlorophyll a (3.9%) and PBR 357 faced maximum reduction (20.4%).

**Chlorophyll b**

Average reduction of chlorophyll b due to moisture stress was 36.6% over irrigated

condition. CJRD 1261 had least chlorophyll b (0.25 mg/g FW) and PBR 422 had highest (0.66 mg/g FW) under irrigated condition whereas under rainfed condition ELM 38 had lowest chlorophyll b (0.20 mg/g FW) and the highest of 0.32 mg/g FW in two cultivars CSR 1163 and PBR 357. Genotypic mean indicated minimum reduction of chlorophyll b in CJRD 1261 (10.5%) and maximum reduction in MLM 41-13-2 (54.3%).

**Total chlorophyll**

Reduction in total chlorophyll content was 18.4% under rainfed over irrigated condition. Under irrigated condition, total chlorophyll content ranged from 1.72 in RB-50 to 2.27 mg/g FW in PBR 422 while from 1.40 in MLM 41-13-2 to 1.75 mg/g FW in CSR 1163 under rainfed condition. Reduction in total chlorophyll content was comparable in RB-50 and RH 406.

Dogra *et al* (2018) reported the decrease of total chlorophyll content in *B. juncea* under drought stress at all the crop growth stages i.e., 45, 60 and 90 DAS. Earlier, changes in chlorophyll content under different intensities of moisture stress in Brassica species has been reported by Majidi *et al* (2015). Bhuiyan *et al* (2019) observed the decline in chla (55%) and chlb (49%) and in total chl (50%) under drought stress in rapeseed seedlings. Chlorophyll content under drought stress gets reduced due to production of reactive oxygen species that damage the chloroplasts (Gill and Tuteja 2010). Similar trend was reported in cherry tomato which is considered as a characteristic indicator of oxidative stress and may be the outcome of pigment photo-oxidation and chlorophyll degradation (Hassan *et al* 2015).

**Carotenoids**

Carotenoid content decreased by 18.4% due to moisture stress and ranged from 0.41 (RB-50) to 0.56 mg/g FW (PBR 422) under irrigated condition and from 0.34 (RH 1518) to 0.43 mg/g FW (CJRD 1261 and RH 406) under rainfed condition. Carotenoids were reduced to 3.9% in RB-50 and to 29.9% in PBR 422 (Table 5). Our results are in agreement with Majidi *et al* (2015), according to their study carotenoids decreased under moderate drought stress over control in *B. juncea and B. napus*. In wheat carotenoid content was reduced more in severe stress than moderate, normal or watered plants (Abid *et al* 2018). Similarly, carotenoids also decreased under shaded condition over control/non-shaded in *B. juncea* genotypes (Kaur 2018).

**SPAD values**

Significant effect on SPAD was evident due to moisture stress in the studied genotypes and their interactions (I×G) were significant. SPAD values were reduced by 5.7% under rainfed over irrigated condition. Under irrigated condition, SPAD ranged from 45.1 (CSR 1163) to 49.6 (ELM 38) and from 42.7 (IAN) to 46.2 (PBR 357) under rainfed condition. SPAD was reduced to 2.9% in RH 1518 and 9.5% in MLM 41-13-2 (Fig. 9).

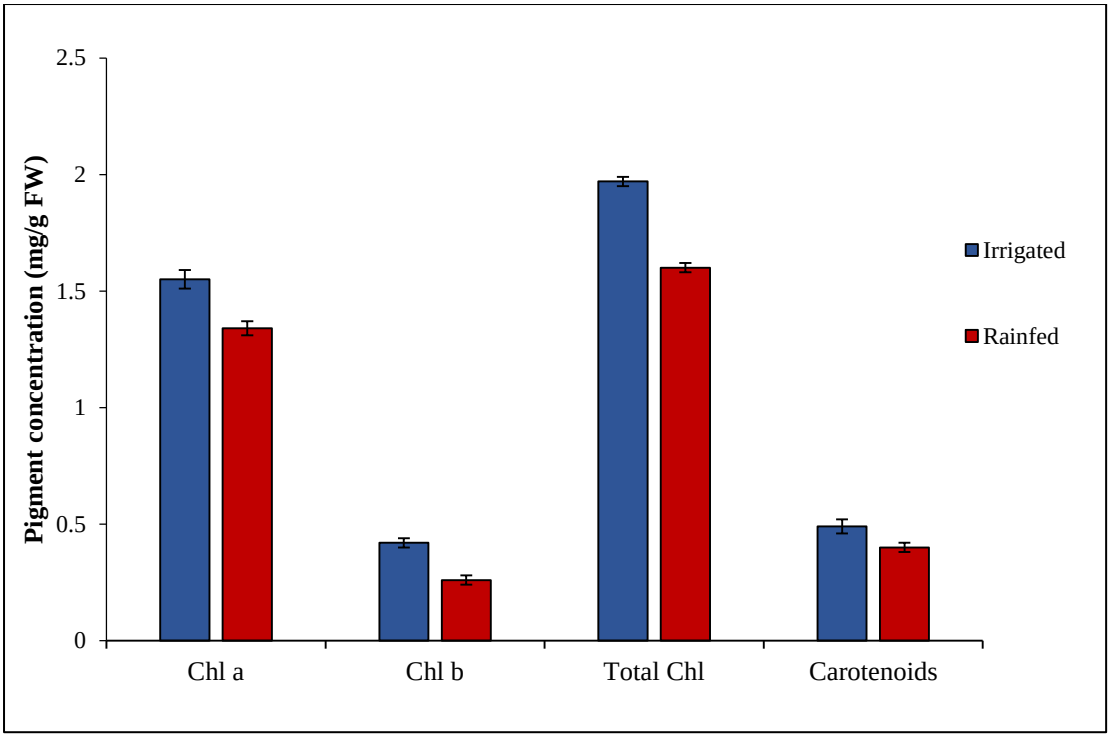



Fig. 8: Effect of moisture stress on mean photosynthetic pigments (mg/g FW)

Kumari *et al* (2019) reported decreased SPAD values under rainfed condition in 50 genotypes of *Brassica juncea*. Similar findings of Kaur and Sharma (2015c) in Indian mustard reported lowest SPAD values under moisture stress ($I_0$) as compared to restricted moisture ($I_1$) and normal moisture ($I_2$). However, the significant difference for SPAD values between moisture regimes was only at 90 DAS.

**Canopy temperature (CT)**

A significant increase of canopy temperature was observed under rainfed condition. Canopy temperature on an average increased by 1.5°C under rainfed condition as compared to irrigated condition (Fig. 9). Under irrigated condition, CT ranged from 21.6°C in CJRD 1261 to 23.3°C in PBR 422 from 23.6 in PBR 422 to 24.6°C in RH 1518 under rainfed condition. Genotypic mean indicated minimum CT in CJRD 1261 (22.8°C) and maximum in RH 1518 (23.6°C).

**Canopy air temperature differential (CATD)**

Moisture stress had significant effect on canopy air temperature differential of genotypes in present investigation. Average reduction of CATD (°C) under rainfed was 1.5°C as compared to irrigated condition. Under rainfed condition, CATD varied from -3.0 (PBR 422) to -2.0 (RH 1518) and from -5.0 (CJRD 1261) to -3.3 (PBR 422) with irrigations. Genotypic mean indicated variability of -2.9 (RH 1518) and -3.8 (CJRD 1261) (Fig. 9).

Table 5: Effect of moisture stress on photosynthetic pigments (mg/g FW)

| Genotypes/ Treatment | Chlorophyll a | | | Chlorophyll b | | | Total chlorophyll | | | Carotenoids | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** |
| RH 1518 | 1.47±0.02 | 1.28±0.03 | **12.8** | 0.31±0.04 | 0.22±0.01 | **29.7** | 1.78±0.02 | 1.50±0.05 | **15.8** | 0.42±0.01 | 0.34±0.02 | **19.6** |
| JC 210-335 | 1.68±0.07 | 1.41±0.03 | **16.4** | 0.35±0.03 | 0.27±0.02 | **22.7** | 2.03±0.02 | 1.68±0.02 | **17.5** | 0.52±0.04 | 0.42±0.03 | **17.7** |
| CJRD 1261 | 1.53±0.03 | 1.29±0.01 | **15.4** | 0.25±0.01 | 0.23±0.00 | **10.5** | 1.78±0.04 | 1.52±0.01 | **14.7** | 0.47±0.01 | 0.43±0.02 | **9.2** |
| RB-50 | 1.44±0.06 | 1.28±0.02 | **11.3** | 0.28±0.02 | 0.25±0.02 | **11.1** | 1.72±0.02 | 1.53±0.01 | **11.3** | 0.41±0.04 | 0.39±0.01 | **3.9** |
| RH 406 | 1.43±0.14 | 1.37±0.05 | **3.9** | 0.46±0.03 | 0.29±0.01 | **36.4** | 1.89±0.02 | 1.67±0.02 | **11.9** | 0.46±0.02 | 0.43±0.02 | **6.7** |
| PBR 422 | 1.61±0.04 | 1.31±0.03 | **18.7** | 0.66±0.02 | 0.31±0.05 | **52.7** | 2.27±0.01 | 1.60±0.01 | **29.3** | 0.56±0.02 | 0.39±0.01 | **29.9** |
| ELM 38 | 1.77±0.02 | 1.52±0.05 | **14.3** | 0.37±0.03 | 0.20±0.03 | **45.9** | 2.14±0.03 | 1.72±0.02 | **19.8** | 0.55±0.02 | 0.42±0.02 | **24.0** |
| CSR 1163 | 1.62±0.02 | 1.43±0.04 | **11.4** | 0.54±0.03 | 0.32±0.02 | **41.0** | 2.06±0.01 | 1.75±0.01 | **14.9** | 0.50±0.01 | 0.40±0.03 | **20.3** |
| IAN | 1.62±0.03 | 1.35±0.01 | **16.7** | 0.33±0.04 | 0.29±0.01 | **13.5** | 1.96±0.02 | 1.64±0.01 | **16.2** | 0.49±0.04 | 0.41±0.01 | **16.5** |
| MCN 09-40 | 1.43±0.04 | 1.35±0.01 | **5.5** | 0.51±0.02 | 0.25±0.03 | **50.5** | 1.94±0.02 | 1.60±0.01 | **17.4** | 0.48±0.04 | 0.41±0.02 | **14.5** |
| MLM 41-13-2 | 1.42±0.04 | 1.18±0.01 | **16.5** | 0.47±0.01 | 0.22±0.02 | **54.3** | 1.89±0.02 | 1.40±0.01 | **26.0** | 0.46±0.04 | 0.37±0.01 | **18.7** |
| PBR 357 | 1.65±0.01 | 1.32±0.04 | **20.3** | 0.52±0.01 | 0.32±0.06 | **38.0** | 2.17±0.20 | 1.64±0.02 | **24.6** | 0.53±0.01 | 0.40±0.02 | **25.1** |
| **Average** | **1.56±0.04** | **1.34±0.03** | | **0.41±0.02** | **0.26±0.02** | | **1.96±0.04** | **1.60±0.02** | | **0.49±0.03** | **0.40±0.02** | |
| CD (p=0.05) | I= 0.031, G= 0.075, I×G= 0.106 | | | I= 0.020, G= 0.050, I×G= 0.071 | | | I= 0.032, G= 0.079, I×G= 0.111 | | | I= 0.018, G= 0.043, I×G= 0.061 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

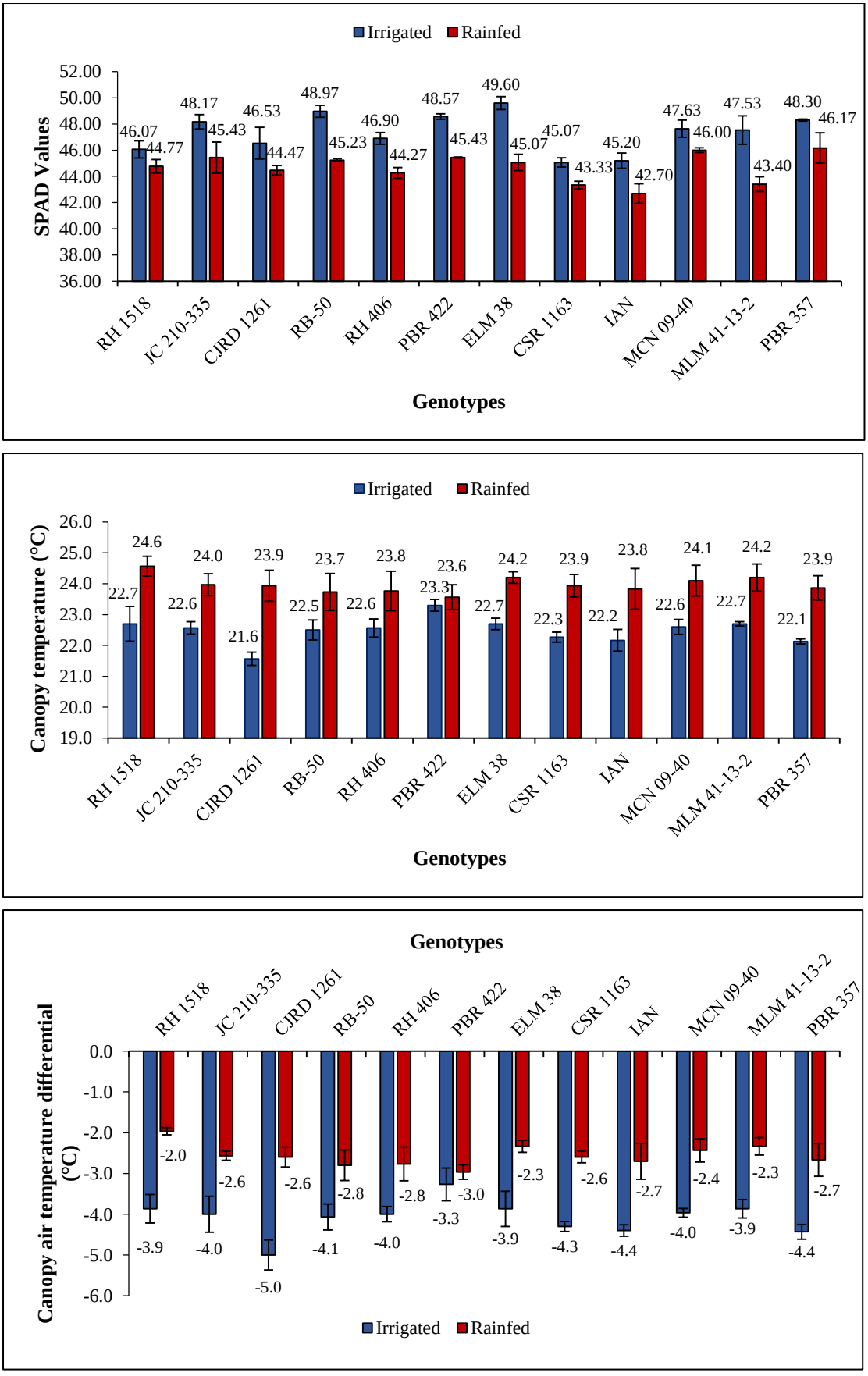



Fig. 9: Effect of moisture stress on SPAD, canopy temperature (CT) and canopy air temperature differential (CATD)

Drought stress increased the canopy temperature (CT) in rapeseed genotypes and CT was negatively correlated with stomatal conductance and relative water content (Eslam *et al* 2017). Similarly, the decrease in canopy temperature depression was under water deficit conditions in *B. juncea* genotypes. (Pandey *et al* 2017).

**Plant water status**

The plant water status measured by RWC (relative water content) and the computed traits varied significantly within the genotypes and significant effects of moisture stress were evident. The interaction between I×G for all the studied leaf water traits were also significant (Table 6, Fig. 10).

**Relative water content (RWC)**

Relative water content on an average decreased by 9.0% under moisture stress prevailed under rainfed condition. Under irrigated condition, RWC was more in PBR 357 (82.8%) trailed by MLM 41-13-2 (82.7%) and least in ELM 38 (73.9%). However, RWC was comparable in RH 1518 and RB-50. Under moisture stress, RWC was more in MLM 41-13-2 (81.2%) and least in ELM 38 and CSR 1163 (64.4%). Minimum reduction in RWC was 1.1% (JC 210-335) and maximum in RB-50 (16.6%).

Relative water content decreased significantly in all genotypes of *B. napus* and *B. carinata* under drought stress (Rana and Chaudhary 2013). Similar findings were also reported in wheat (Molnar *et al* 2002) and in *B. napus* (Eslam *et al* 2017). The decrease in RWC in wheat is the indicator of high leaf temperature in drought (Siddique *et al* 2001). The *B. juncea* genotypes showed increased RWC at restricted moisture condition as compared to that under moisture stress (Kaur 2012).

**Relative saturation deficit (RSD)**

Average increase of relative saturation deficit under rainfed condition was 21.6%. Under irrigated condition, RSD range was 14.7% (PBR 357) to 23.0% (ELM 38) and from 17.0% (MLM 41-13-2) to 31.7% (CSR 1163) under rainfed condition. Increase of RSD ranged from 3.3% in JC 210-335 ato 70.6% in RB-50 (Table 6, Fig. 10). The results are in accordance with the findings of Dedio (1975) indicating that RSD increased with the decrease in water content in wheat leaves. Similar trend was reported in shaded treatment as compared to control in *B. juncea* genotypes by Kaur (2018).

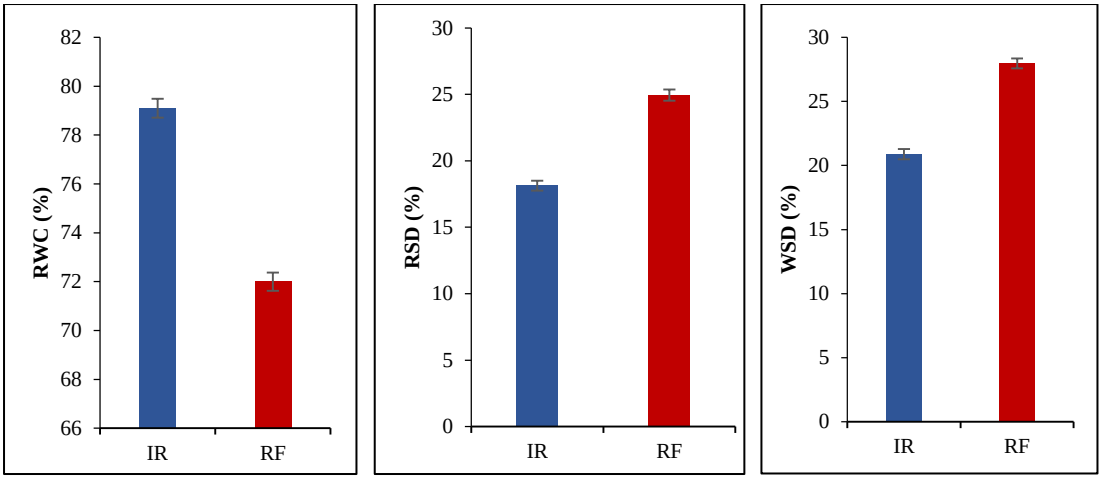
**Water saturation deficit (WSD)**

Water saturation deficit under rainfed condition increased due to water deficit in soil. As compared to irrigated condition, the increase in rainfed was 24.4%. Under irrigated condition, WSD was highest in ELM 38 (26.1%) and lowest in PBR 357 (17.2%) whereas under rainfed condition, was highest in two genotypes ELM 38 and CSR 1163 (35.6%) and lowest in MLM 41-13-2 (18.8%). The increase of WSD varied from 3.5% (JC 210-335) to 66.7% (RB-50) (Table 6, Fig. 12). In a study by Aldesuquy *et al* (2014), it was observed that

Table 6: Effect of moisture stress on plant water status in *B. juncea* genotypes

| Genotypes/ treatment | Relative water content (%) | | | Relative saturation deficit (%) | | | Water saturation deficit (%) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Inc (%) | Irrigated | Rainfed | Inc (%) |
| RH 1518 | 80.4±0.2 | 71.6±0.3 | **10.9** | 17.2±0.1 | 24.5±0.4 | **42.8** | 19.6±0.4 | 28.4±0.3 | **44.6** |
| JC 210-335 | 76.4±0.2 | 75.6±0.2 | **1.1** | 21.3±0.2 | 22.0±0.5 | **3.3** | 23.6±0.2 | 24.4±0.4 | **3.5** |
| CJRD 1261 | 77.4±0.3 | 72.8±0.3 | **6.0** | 19.7±0.3 | 24.6±0.4 | **25.0** | 22.6±0.3 | 27.2±0.4 | **20.4** |
| RB-50 | 80.0±0.5 | 66.7±0.5 | **16.6** | 17.1±0.3 | 29.2±0.0 | **70.6** | 20.0±0.5 | 33.3±0.5 | **66.7** |
| RH 406 | 81.5±0.3 | 80.6±0.3 | **1.2** | 16.0±0.7 | 17.5±0.2 | **15.1** | 18.5±0.3 | 19.4±0.4 | **5.4** |
| PBR 422 | 79.0±0.6 | 70.2±0.4 | **11.1** | 18.2±0.1 | 26.6±0.6 | **46.0** | 21.0±0.4 | 29.8±0.2 | **41.7** |
| ELM 38 | 73.9±0.7 | 64.4±0.1 | **12.9** | 23.0±0.3 | 31.5±0.2 | **36.6** | 26.1±0.5 | 35.6±0.4 | **36.5** |
| CSR 1163 | 74.7±0.3 | 64.4±1.0 | **13.8** | 21.7±0.6 | 31.7±0.4 | **45.8** | 25.3±0.8 | 35.6±0.4 | **40.8** |
| IAN | 81.0±0.2 | 69.3±0.6 | **14.4** | 16.6±0.3 | 27.1±1.0 | **63.2** | 19.0±0.3 | 30.7±0.7 | **61.4** |
| MCN 09-40 | 79.6±0.2 | 70.1±0.3 | **12.0** | 17.7±0.4 | 26.9±0.7 | **52.3** | 20.4±0.2 | 29.9±0.4 | **46.9** |
| MLM 41-13-2 | 82.7±0.7 | 81.2±0.1 | **1.8** | 15.1±0.6 | 17.0±0.2 | **13.0** | 17.3±0.4 | 18.8±0.1 | **8.5** |
| PBR 357 | 82.8±0.4 | 77.5±0.5 | **6.4** | 14.7±0.6 | 20.7±0.4 | **41.2** | 17.2±0.5 | 22.5±0.5 | **30.7** |
| **Average** | **79.1±0.4** | **72.0±0.4** | | **18.2±0.4** | **25.0±0.4** | | **20.9±0.4** | **28.0±0.4** | |
| CD (p=0.05) | I= 0.309, G= 0.756, I×G= 1.069 | | | I= 0.317, G= 0.777, I×G= 1.098 | | | I= 0.305, G= 0.746, I×G= 1.055 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype




IR= Irrigated and RF= Rainfed

Fig. 10: Effect of moisture stress on mean plant water status in *B. juncea* genotypes

water stress increased the WSD in flag leaf of wheat. Similar trend was observed in barley with regard to drought stress by Zhang *et al* (2015) indicating that tolerant genotypes faced lesser water loss. However, WSD increased with increased salinity in castor bean (Lima *et al* 2019).

**Leaf traits**

Genotypes differed significantly and moisture stress had profound effect on the leaf traits. Interactive effects (I×G) were also significant for these traits (Table 7).

**Number of leaves**

Number of leaves per plant decreased by 13.9% on average under rainfed condition. Under irrigated condition, leaf number was 27.8 in CJRD 1261 and 45.1 in CSR 1163 whereas the same genotypes had 23.3 leaves number and 43.6 in CSR 1163 under rainfed condition. Moisture stress reduced number of leaves in the genotypes which ranged from 3.3% (CSR 1163) to 21.5% (MLM 41-13-2) (Table 7).

A declining trend in leaf number in *B. napus* was recorded by Germchi *et al* (2010) under different irrigation treatments being minimum in 30% available water content condition which was later endorsed by Qaderi *et al* (2012).

**Leaf length**

Leaf length decreased by 11.4% under moisture stress and was 10.3 cm (PBR 357) and 15.4 cm in RB-50 while 13.1 cm in MLM-41-13-2 and 16.6 cm in RB-50 under irrigated condition. Reduction in leaf length varied from 1.1% (JC 210-335) and 32.2% (PBR 357).

**Leaf width**

Leaf width decreased by 8.9% under rainfed as compared to irrigated condition and it ranged from 6.8 (JC 210-335 and PBR 357) to 8.9 cm (ELM 38) under irrigated and from 4.8 (JC 210-335) to 8.4 cm (PBR 422 ad MLM 41-13-2) under rainfed condition. Reduction in leaf width was 1.2% (RB-50) and 29.3% (IAN) followed by JC 210-335 (29.2%).

**Leaf area**

Leaf area decreased by 13.7% under rainfed condition. Under irrigated condition, LA varied from 63.5 (JC 210-335) to 92.4 cm² (PBR 422) whereas from 42.9 (JC 210-335) to 82.7 $cm^2$ (PBR 422) under rainfed condition. The reduction of LA was 1.5% in MLM 41-13-2 followed by 1.6% in RH 406 and 32.4% in JC 210-335.

Decline in LA has been reported under saline and drought conditions in *B. napus* under water stress (Naderikharaji *et al* 2008). Moisture stress reduced LA in *B. juncea* (Kaur 2018), soybean (Amira and Qados 2014) and in groundnut (Madhusudan and Sudhakar 2014). The decrease in leaf surface is the defence mechanism against water stress as it caused reduction in water loss through transpiration (Fathi and Tari 2016). The significant reduction in length and width of the leaf and consequently in the leaf area contributed to the reduction of evaporation area under water scarce condition (Moaveni *et al* 2010).

Genotypes differed significantly for specific leaf area (SLA), specific leaf weight (SLW) and leaf water retention (LWR) and moisture stress reduced these traits significantly, however, interactive effects (I×G) were only significant for LWR (Table 8).

**Specific leaf area**

Specific leaf area (SLA) decreased under moisture stress by 8% which ranged from 0.20 (RH 1518) to 0.27 $cm^2$/mg (CJRD 1261) and from 0.22 (RH 1518) to 0.28 $cm^2$/mg (CJRD 1261) with irrigation. SLA declined from 1.0% (MLM 41-13-2) to 15.6% (PBR 422). Our results are corroborated with the earlier observations of Kaur and Sharma (2015b) in Indian mustard at different irrigation regimes.

**Specific leaf weight**

The decrease in SLW under rainfed condition was 5.6% on an average as compared to irrigated condition. Under irrigated condition, SLW varied from 3.74 mg/$cm^2$ in CJRD 1261 to 4.93 mg/$cm^2$ in RH 1518 while from 3.63 mg/$cm^2$ in CJRD 1261 to 4.58 mg/$cm^2$ in RH 1518 under rainfed condition. The reduction in SLW was 1.3% in MLM 41-13-2 and 15.7% in PBR 422. Chandra *et al* (2018) reported a similar declining trend in SLW in non-irrigated over irrigated *B. juncea* genotypes.

**Leaf water retention**

Leaf water retention (LWR) was reduced to 11.4% due to moisture stress. Under irrigated condition, LWR ranged from 49.3% in JC 210-335 to 84.5% in ELM 38 whereas under rainfed condition from 33.2% in JC 210-335 to 73.6% in ELM 38. The reduction in LWR was 4.8% (MLM 41-13-2) to 32.7% (JC 210-335). Decrease in water retention capacity of wheat leaves under drought stress at all growth stages have been reported by Tasmina *et al* (2016). In a similar study, leaf water retention capacity decreased under heat stress and was considered major physiological parameter for selecting high yielding Indian mustard genotypes (Ram *et al* 2016).

Table 7: Effect of moisture stress on leaf traits in *B. juncea* genotypes

| Genotypes/ treatment | Number of leaves | | | Length (cm) | | | Width (cm) | | | Area ($cm^2$) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** |
| RH 1518 | 30.2±0.2 | 24.6±0.4 | **18.5** | 14.3±0.5 | 12.4±0.3 | **13.0** | 7.9±0.1 | 7.6±0.2 | **3.2** | 69.7±0.4 | 65.7±1.4 | **5.7** |
| JC 210-335 | 37.0±0.4 | 34.1±0.7 | **7.8** | 13.4±0.2 | 13.2±0.6 | **1.1** | 6.8±0.2 | 4.8±0.3 | **29.2** | 63.5±1.8 | 42.9±1.3 | **32.4** |
| CJRD 1261 | 27.8±0.2 | 23.3±0.4 | **16.2** | 15.9±0.3 | 14.1±0.2 | **11.2** | 7.7±0.2 | 7.4±0.1 | **4.5** | 75.4±3.5 | 62.6±2.1 | **17.0** |
| RB-50 | 36.4±0.1 | 30.9±0.6 | **15.1** | 16.6±0.3 | 15.4±0.2 | **6.9** | 8.1±0.6 | 8.0±0.1 | **1.2** | 83.8±1.5 | 80.8±1.4 | **3.6** |
| RH 406 | 38.2±0.3 | 30.5±0.4 | **20.2** | 15.8±0.8 | 14.5±0.2 | **8.7** | 7.9±0.3 | 7.5±0.2 | **5.0** | 72.0±1.2 | 70.9±3.2 | **1.6** |
| PBR 422 | 36.2±0.3 | 31.2±0.7 | **13.8** | 16.0±0.2 | 14.0±0.4 | **12.0** | 8.8±0.2 | 8.4±0.5 | **5.3** | 92.4±3.0 | 82.7±0.7 | **10.5** |
| ELM 38 | 40.4±0.3 | 35.4±0.4 | **12.4** | 14.3±0.6 | 13.0±0.3 | **9.1** | 8.9±0.2 | 7.3±0.1 | **18.0** | 78.7±1.8 | 64.4±1.8 | **18.2** |
| CSR 1163 | 45.1±0.4 | 43.6±1.3 | **3.3** | 14.1±0.4 | 13.0±0.3 | **7.9** | 7.3±0.2 | 7.2±0.1 | **1.6** | 73.8±0.8 | 65.0±1.1 | **11.9** |
| IAN | 35.3±0.3 | 34.1±0.8 | **3.4** | 16.2±0.2 | 13.6±0.3 | **15.9** | 8.0±0.0 | 5.6±0.1 | **29.3** | 84.4±1.2 | 65.1±3.3 | **22.9** |
| MCN 09-40 | 34.3±0.3 | 28.7±0.5 | **16.3** | 13.8±0.3 | 12.3±0.4 | **11.2** | 7.9±0.2 | 7.8±0.2 | **2.0** | 70.8±1.0 | 63.0±1.6 | **11.1** |
| MLM 41-13-2 | 40.4±0.3 | 31.7±0.4 | **21.5** | 13.1±0.3 | 12.1±0.0 | **7.6** | 8.7±0.3 | 8.4±0.2 | **2.9** | 69.7±3.1 | 68.7±2.8 | **1.5** |
| PBR 357 | 38.6±0.2 | 31.2±0.7 | **19.2** | 15.2±0.2 | 10.3±0.4 | **32.2** | 6.8±0.1 | 6.2±0.2 | **7.9** | 65.6±1.3 | 45.0±2.3 | **31.5** |
| **Average** | **36.7±0.3** | **31.6±0.6** | | **14.9±0.4** | **13.2±0.3** | | **7.9±0.2** | **7.2±0.2** | | **75.0±1.7** | **64.7±1.9** | |
| CD (p= 0.05) | I= 0.580, G= 1.421, I×G= 2.009 | | | I= 0.257, G= 0.630, I×G= 0.891 | | | I= 0.173, G= 0.424, I×G= 0.600 | | | I= 1.385, G= 3.392, I×G= 4.798 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

Table 8: Effect of moisture stress on specific leaf area (SLA), specific leaf weight (SLW) and leaf water retention (LWR)

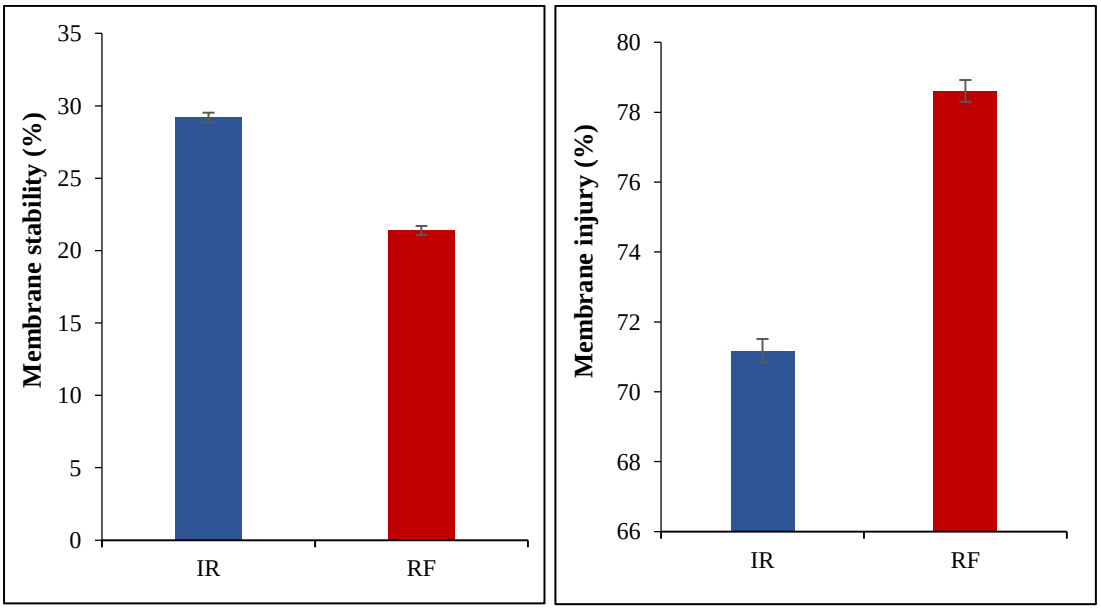
| Genotypes/ Treatment | Specific leaf area ($cm^2$/mg) | | | Specific leaf weight (mg/$cm^2$) | | | Leaf Water retention (%) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Red (%) |
| RH 1518 | 0.22±0.01 | 0.20±0.01 | **7.1** | 4.93±0.05 | 4.58±0.06 | **7.1** | 68.4±0.1 | 65.0±0.8 | **5.0** |
| JC 210-335 | 0.26±0.02 | 0.25±0.01 | **6.0** | 4.05±0.18 | 3.82±0.20 | **5.8** | 49.3±0.2 | 33.2±0.9 | **32.7** |
| CJRD 1261 | 0.28±0.01 | 0.27±0.02 | **3.0** | 3.74±0.14 | 3.63±0.13 | **3.0** | 71.8±0.7 | 65.9±0.3 | **8.3** |
| RB-50 | 0.26±0.01 | 0.25±0.01 | **3.8** | 4.00±0.02 | 3.85±0.08 | **3.7** | 73.1±1.0 | 60.9±0.3 | **16.7** |
| RH 406 | 0.24±0.02 | 0.23±0.01 | **1.2** | 4.32±0.24 | 4.24±0.06 | **1.9** | 64.4±0.3 | 60.7±0.5 | **5.7** |
| PBR 422 | 0.26±0.01 | 0.22±0.01 | **15.6** | 4.58±0.14 | 3.86±0.08 | **15.7** | 66.4±0.3 | 62.7±0.8 | **5.6** |
| ELM 38 | 0.24±0.01 | 0.23±0.02 | **3.5** | 4.27±0.05 | 4.12±0.05 | **3.5** | 84.5±0.7 | 73.6±0.5 | **12.9** |
| CSR 1163 | 0.24±0.02 | 0.23±0.01 | **3.5** | 4.29±0.06 | 4.15±0.13 | **3.3** | 76.7±0.6 | 64.9±0.3 | **15.4** |
| IAN | 0.24±0.01 | 0.22±0.02 | **6.4** | 4.49±0.18 | 4.20±0.13 | **6.6** | 69.1±0.4 | 61.7±0.5 | **7.3** |
| MCN 09-40 | 0.25±0.01 | 0.23±0.01 | **9.3** | 4.43±0.11 | 4.02±0.08 | **9.3** | 74.2±0.2 | 69.7±0.6 | **6.1** |
| MLM 41-13-2 | 0.25±0.01 | 0.25±0.01 | **1.0** | 4.07±0.16 | 4.02±0.09 | **1.3** | 74.2±0.7 | 70.6±0.6 | **4.8** |
| PBR 357 | 0.23±0.01 | 0.22±0.01 | **3.3** | 4.51±0.22 | 4.36±0.16 | **3.5** | 71.4±0.5 | 59.2±0.9 | **17.1** |
| **Average** | **0.25±0.01** | **0.23±0.01** | | **4.31±0.13** | **4.07±0.10** | | **70.3±0.5** | **62.3±0.6** | |
| CD (p=0.05) | I= 0.006, G= 0.014, I×G=NS | | | I= 0.092, G= 0.225, I×G= NS | | | I= 0.590, G= 1.444, I×G= 2.043 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

Table 9: Effect of moisture stress on membrane stability and membrane injury

| Genotypes/ Treatment | Membrane stability (%) | | | Membrane injury (%) | | |
|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Inc (%) |
| RH 1518 | 33.6±0.2 | 25.5±0.4 | **24.1** | 66.3±0.2 | 74.4±0.4 | **12.2** |
| JC 210-335 | 29.9±0.5 | 18.3±0.3 | **39.0** | 70.1±0.5 | 81.7±0.3 | **16.5** |
| CJRD 1261 | 26.9±0.2 | 18.4±0.2 | **31.7** | 73.0±0.2 | 81.6±0.2 | **11.7** |
| RB-50 | 28.8±0.5 | 21.4±0.1 | **25.7** | 71.2±0.5 | 78.6±0.1 | **10.4** |
| RH 406 | 25.0±0.1 | 19.4±0.3 | **22.0** | 75.2±0.1 | 80.7±0.3 | **7.3** |
| PBR 422 | 25.4±0.3 | 25.3±0.4 | **0.4** | 74.6±0.3 | 74.7±0.4 | **0.1** |
| ELM 38 | 29.9±0.4 | 20.6±0.1 | **31.4** | 70.0±0.4 | 79.4±0.1 | **13.4** |
| CSR 1163 | 31.3±0.6 | 24.4±0.3 | **21.9** | 68.8±0.6 | 75.6±0.3 | **9.9** |
| IAN | 26.3±0.2 | 19.3±0.1 | **26.5** | 73.7±0.2 | 80.7±0.1 | **9.4** |
| MCN 09-40 | 33.8±0.4 | 20.1±0.2 | **40.5** | 66.2±0.4 | 79.9±0.2 | **20.7** |
| MLM 41-13-2 | 30.2±0.2 | 23.5±0.3 | **22.1** | 69.8±0.2 | 76.5±0.3 | **9.6** |
| PBR 357 | 24.9±0.5 | 21.4±1.0 | **13.9** | 75.1±0.5 | 78.6±1.0 | **4.6** |
| **Average** | **28.8±0.3** | **21.5±0.3** | | **71.2±0.3** | **78.5±0.3** | |
| CD (p=0.05) | I= 0.256, G= 0.626, I×G= 0.886 | | | I= 0.255, G= 0.625, I×G= 0.883 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype




IR= Irrigated and RF= Rainfed

Fig. 11: Effect of moisture stress on membrane stability and membrane injury (mean)

**Membrane stability**

Moisture stress decreased the stability of membrane by increasing the cell membrane permeability. Membrane stability was reduced by 25.3% in rainfed over irrigated condition. Membrane stability was 33.8% in MCN 09-40 and 25.0% in RH 406 with irrigated module. With moisture stress, MS was 29.6% (RH 1518) and 22.1% (RH 406). PBR 422 maintained membrane stability under both irrigated (25.4%) and rainfed (25.3%) condition. The reduction of membrane stability was 0.4% (PBR 422) to 40.5% (MCN 09-40) (Table 9, Fig. 11). In *B. juncea* under non-irrigated condition leaf membrane stability decreased due to low water potential (Chandra *et al* 2018). Maintenance of integrity and stability of membranes under water deficit condition is a major component of drought tolerance in plants (Meena and Kaur 2019).

**Membrane injury**

Membrane injury is the extent of membrane leakage that increased under rainfed condition by 10.3% over irrigated module. Membrane injury was highest in RH 406 (75.2%) followed by PBR 357 (75.1%) and lowest but comparable in MCN 09-40 (66.2%) and RH 1518 (66.3%) under irrigated module. Injury under stressed condition was highest and comparable in JC 210-335 (81.7%) and CJRD 1261 (81.6%) also being highest whereas RH 1518 suffered least injury (74.4%). Membrane stability was comparable in PBR 422 under irrigated modules and subsequently in rainfed condition. Injury increased under moisture stress from 0.1% (PBR 422) to 20.7% (MCN 09-40) when compared over irrigated condition. Drought stress led to increase in membrane injury which was more pronounced at jointing stage in wheat (Abid *et al* 2018). Similar results were endorsed by Sehgal *et al* (2017) in lentil genotypes. However, drought stress damaged membranes more in sensitive genotypes than in tolerant genotypes.

**Correlation coefficients of physiological traits in *B. juncea***

Seed yield under irrigated condition was positively associated with SPAD (r= 0.141), total chlorophyll (r= 0.242) and carotenoid content (r= 0.157) under irrigated condition. Under rainfed condition, similar trend existed for SPAD (r= 0.240) and weak association for total chlorophyll (r= 0.017) but carotenoids were negatively associated (r= -0.237). Under irrigated condition, seed yield was weakly positively correlated with relative water content (RWC) (r= 0.044), canopy temperature (r= 0.197), canopy air temperature differential (CATD) (r= 0.199) and membrane injury (r= 0.036) but negatively with relative saturation deficit (RSD) (r= -0.105), water saturation deficit (WSD) (r= -0.054) and membrane stability (r= -0.036). However, seed yield showed reverse trend with RWC (r= -0.101), canopy temperature (r= -0.061), CATD (r= -0.062), membrane injury (r= -0.518), RSD (r= 0.106), WSD (r= 0.101) and membrane injury (r= -0.518) under rainfed condition.

Weak positive association of seed yield was with length (r= 0.178), width (r= 0.368) and leaf area (r= 0.201) under irrigated module whereas under rainfed, it was negatively correlated with length (r= -0.213), significantly and positively correlated with width (r= 0.591*) but weakly and positively associated with leaf area (r= 0.388). Number of leaves per plant were negatively correlated with seed yield under irrigated (r= -0.120) as well as under rainfed (r= -0.16) conditions. Specific leaf area (SLA) was negatively correlated with seed yield under both irrigated (r= -0.250) and rainfed (r= -0.545) conditions. However, specific leaf weight (SLW) had weak positive association with seed yield both under irrigated (r= 0.428) and rainfed (r= 0.432) conditions. Similarly, leaf water retention (LWR) had positive association with seed yield both under irrigated (r= 0.561) and rainfed (r= 0.638*) conditions (Table 10a-b). Chlorophyll content was positively correlated with carotenoids under both irrigated (r= 0.948**) and rainfed (r= 0.577*) conditions. RWC was significantly and negatively correlated with RSD under irrigated (r= -0.995**) as well as rainfed (r= -0.998**) conditions and also with WSD under irrigated (r= -1.000**) as well as rainfed (r= -1.000**) conditions. RSD was positively associated with WSD under irrigated (r= 0.995**) and rainfed (r= 0.998**) conditions. Membrane stability had negative correlation correlated with injury under both irrigated (r= -1.000**) and rainfed (r= -0.995**) conditions. Leaf length had significantly negative correlation with membrane stability (r= -0.679*) but positive with membrane injury (r= 0.679*) under irrigated module. Leaf area was positively correlatd with length (r= 0.662*) and width (r= 0.653*) with irrigation schedule and followed a similar trend with length (r= 0.643*) and width (r= 0.782**) under stress.

Relationship between membrane injury ($R^2$= 0.2686), leaf width ($R^2$= 0.3488) and SLA ($R^2$= 0.2839) with seed yield was relatively better under rainfed as compared to irrigated condition (Figure 12). Kaur and Sharma (2015c) also observed correlation between physiological traits and seed yield in Indian mustard under different moisture regimes. SLA was negatively correlated with SLW under moisture stress in *B. juncea* and further SLA had negative correlation with LWR and seed yield. Rana and Chaudhary (2013) revealed through correlation analysis that leaf area had weak positive association with seed yield. Eslam *et al* (2017) also observed negative correlation between RWC and seed yield under water deficit stress at rosette stage of rapeseed genotypes. Correlation coefficients of chlorophyll and carotenoids showed parallelism with findings of Majidi *et al* (2015) in Brassica species under both non-stress and severe stress conditions. Zarei *et al* (2016) also reported similar results in wheat under rainfed condition in wheat.

Table 10a: Correlation coefficients of physiological traits with seed yield under irrigated condition

| | SPAD | Chl. | Carotenoids | RWC | RSD | WSD | CT | CATD | Stability | Injury | Length | Width | Leaf area | Leaf no. | SLA | SLW | LWR | SY |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **SPAD** | 1 | | | | | | | | | | | | | | | | | |
| **Chl.** | 0.298 | 1 | | | | | | | | | | | | | | | | |
| **Carotenoids** | 0.321 | 0.948** | 1 | | | | | | | | | | | | | | | |
| **RWC** | -0.085 | -0.266 | -0.410 | 1 | | | | | | | | | | | | | | |
| **RSD** | 0.092 | 0.249 | 0.406 | -0.995** | 1 | | | | | | | | | | | | | |
| **WSD** | 0.077 | 0.253 | 0.398 | -1.000** | 0.995** | 1 | | | | | | | | | | | | |
| **CT** | 0.434 | 0.382 | 0.219 | 0.022 | -0.020 | -0.038 | 1 | | | | | | | | | | | |
| **CATD** | 0.436 | 0.386 | 0.222 | 0.025 | -0.023 | -0.041 | 1.000** | 1 | | | | | | | | | | |
| **Stability** | -0.128 | -0.298 | -0.318 | -0.322 | 0.339 | 0.332 | 0.159 | 0.156 | 1 | | | | | | | | | |
| **Injury** | 0.128 | 0.298 | 0.318 | 0.322 | -0.339 | -0.332 | -0.159 | -0.156 | -1.000** | 1 | | | | | | | | |
| **Length** | -0.015 | -0.124 | -0.120 | 0.217 | -0.259 | -0.224 | -0.217 | -0.217 | -0.679* | 0.679* | 1 | | | | | | | |
| **Width** | 0.277 | 0.020 | 0.016 | -0.038 | 0.017 | 0.026 | 0.527 | 0.525 | 0.020 | -0.020 | 0.121 | 1 | | | | | | |
| **Leaf area** | 0.102 | 0.164 | 0.150 | -0.122 | 0.073 | 0.105 | 0.290 | 0.288 | -0.340 | 0.340 | 0.662* | 0.653* | 1 | | | | | |
| **Leaf no.** | 0.110 | 0.499 | 0.367 | -0.199 | 0.173 | 0.201 | 0.271 | 0.272 | -0.048 | 0.048 | -0.313 | 0.013 | -0.060 | 1 | | | | |
| **SLA** | 0.229 | -0.154 | 0.051 | -0.258 | 0.255 | 0.250 | -0.209 | -0.211 | -0.227 | 0.227 | 0.215 | 0.115 | 0.281 | -0.293 | 1 | | | |
| **SLW** | -0.192 | 0.301 | 0.091 | 0.266 | -0.273 | -0.271 | 0.467 | 0.469 | 0.184 | -0.184 | -0.035 | 0.051 | 0.062 | -0.027 | -0.794** | 1 | | |
| **LWR** | 0.109 | 0.014 | -0.011 | -0.173 | 0.112 | 0.177 | -0.088 | -0.088 | 0.211 | -0.211 | 0.006 | 0.492 | 0.238 | 0.240 | -0.171 | -0.007 | 1 | |
| **SY** | 0.141 | 0.242 | 0.157 | 0.044 | -0.105 | -0.054 | 0.197 | 0.199 | -0.036 | 0.036 | 0.178 | 0.368 | 0.201 | -0.120 | -0.250 | 0.428 | 0.561 | 1 |

*Significant at 5%, **Significant at 1%

Chl- Chlorophyll, RWC- Relative water content, RSD- Relative saturation deficit, Stability- Membrane stability, Injury- Membrane injury, Leaf no.- Number of leaves per plant, SLA- Specific leaf area, SLW- Specific leaf weight, LWR- Leaf water retention, SY- Seed yield

Table 10b: Correlation coefficients of physiological traits with seed yield under rainfed condition.

| | SPAD | Chl | Carotenoids | RWC | RSD | WSD | CT | CATD | Stability | Injury | Length | Width | Leaf area | Leaf no. | SLA | SLW | LWR | SY |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **SPAD** | 1 | | | | | | | | | | | | | | | | | |
| **Chl** | 0.066 | 1 | | | | | | | | | | | | | | | | |
| **Carotenoids** | 0.056 | 0.577* | 1 | | | | | | | | | | | | | | | |
| **RWC** | 0.017 | -0.402 | -0.020 | 1 | | | | | | | | | | | | | | |
| **RSD** | 0.003 | 0.426 | 0.061 | -0.998** | 1 | | | | | | | | | | | | | |
| **WSD** | -0.017 | 0.402 | 0.019 | -1.000** | 0.998** | 1 | | | | | | | | | | | | |
| **CT** | -0.044 | -0.299 | -0.516 | 0.014 | -0.046 | -0.015 | 1 | | | | | | | | | | | |
| **CATD** | -0.051 | -0.302 | -0.515 | 0.005 | -0.037 | -0.005 | 1.001** | 1 | | | | | | | | | | |
| **Stability** | -0.090 | -0.276 | -0.845** | -0.163 | 0.139 | 0.163 | 0.288 | 0.289 | 1 | | | | | | | | | |
| **Injury** | 0.070 | 0.249 | 0.818** | 0.169 | -0.147 | -0.169 | -0.228 | -0.228 | -0.995** | 1 | | | | | | | | |
| **Length** | -0.229 | 0.003 | 0.278 | -0.280 | 0.260 | 0.280 | -0.468 | -0.462 | -0.241 | 0.202 | 1 | | | | | | | |
| **Width** | 0.017 | -0.497 | -0.383 | -0.058 | 0.048 | 0.058 | 0.062 | 0.067 | 0.560 | -0.555 | 0.202 | 1 | | | | | | |
| **Leaf area** | -0.242 | -0.301 | -0.253 | -0.289 | 0.264 | 0.289 | -0.267 | -0.259 | 0.411 | -0.434 | 0.643* | 0.782** | 1 | | | | | |
| **Leaf no.** | -0.342 | 0.665* | 0.179 | -0.373 | 0.389 | 0.373 | -0.236 | -0.234 | 0.132 | -0.137 | -0.051 | -0.256 | -0.078 | 1 | | | | |
| **SLA** | -0.061 | -0.238 | 0.477 | 0.152 | -0.126 | -0.152 | -0.247 | -0.242 | -0.526 | 0.524 | 0.370 | 0.007 | -0.055 | -0.094 | 1 | | | |
| **SLW** | -0.100 | 0.125 | -0.481 | 0.089 | -0.120 | -0.089 | 0.485 | 0.480 | 0.427 | -0.384 | -0.516 | -0.066 | -0.155 | 0.067 | -0.842** | 1 | | |
| **LWR** | -0.209 | -0.262 | -0.215 | -0.261 | 0.257 | 0.261 | 0.261 | 0.269 | 0.382 | -0.348 | -0.098 | 0.729** | 0.520 | -0.113 | -0.167 | 0.206 | 1 | |
| **SY** | 0.242 | 0.017 | -0.237 | -0.101 | 0.106 | 0.101 | -0.061 | -0.062 | 0.512 | -0.518 | -0.213 | 0.591* | 0.388 | -0.160 | -0.545 | 0.432 | 0.638* | 1 |

*Significant at 5%, **Significant at 1%

Chl- Chlorophyll, RWC- Relative water content, RSD- Relative saturation deficit, Stability- Membrane stability, Injury- Membrane injury, Leaf no.- Number of leaves per plant, SLA- Specific leaf area, SLW- Specific leaf weight, LWR- Leaf water retention, SY- Seed yield

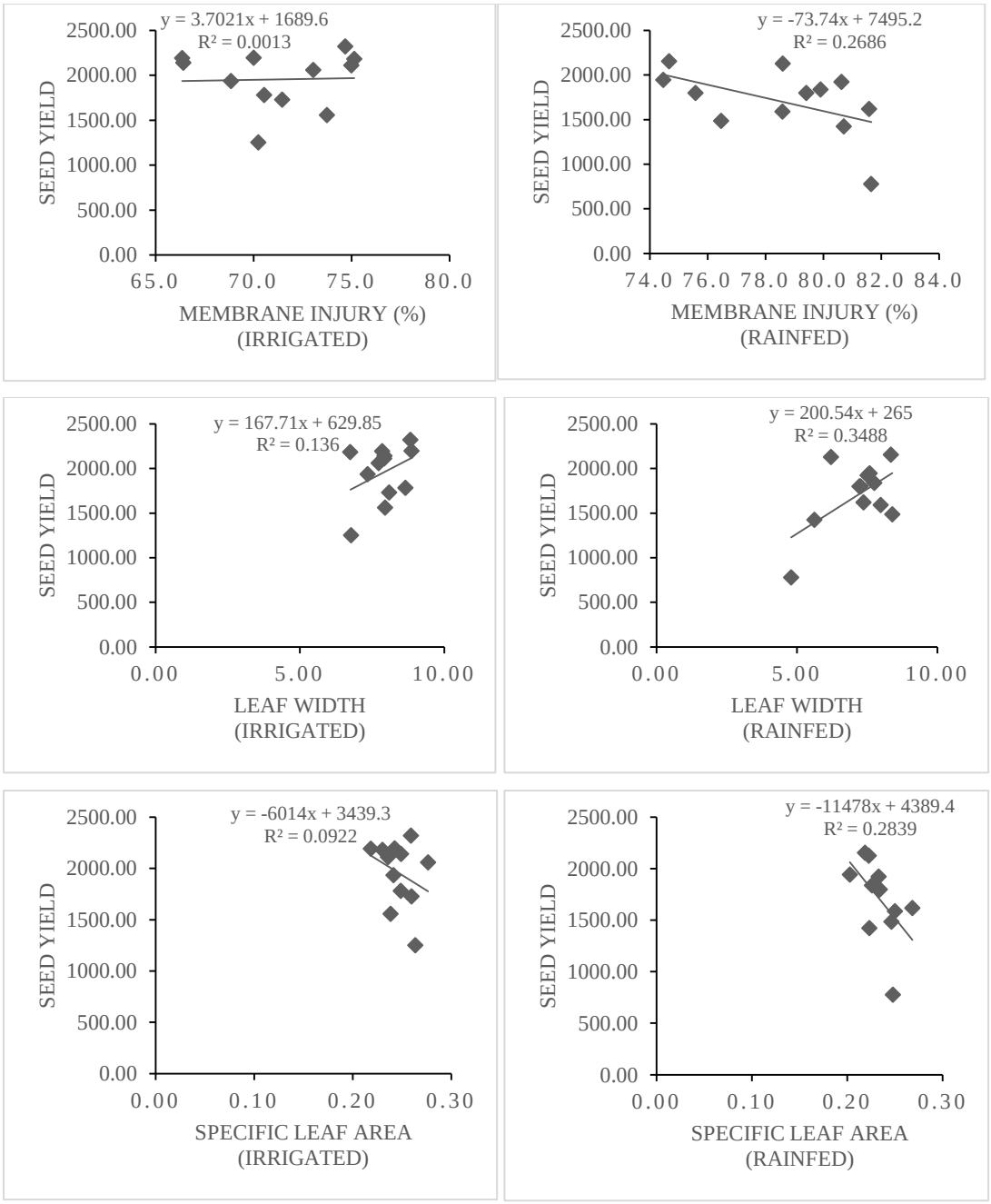



Fig. 12: Relationship of membrane injury, leaf width and SLA with seed yield

**Biochemical parameters**

Moisture stress had a profound effect on biochemical parameters of mustard plants. A significant difference for biomolecules was observed between different genotypes grown under rainfed and irrigated conditions at flowering stage. Moreover, interactions (I×G) were also significant for the estimated biomolecules.

**Sugars**

**Total soluble sugars**

Total soluble sugar (TSS) content under rainfed condition increased by 19.7 mg/g DW over irrigated condition. Under irrigated condition, TSS was minimum in PBR 422 (62.2

mg/g DW) maximum in IAN and ELM 38 (69.6 mg/g DW) and whereas under rainfed condition, it was minimum in PBR 422 (70.1 mg/g DW) and maximum in ELM 38 (90.1 mg/g DW). TSS increased from 12.7% (PBR 422) to 40.2% (RB-50) (Table 11, Fig. 13).

Kaur (2012) reported the increase in total sugars under moisture stress and restricted moisture conditions over normal moisture in *B. juncea* and *B. napus* genotypes. Similar increased trend of total soluble sugars was observed in wheat by Qayyum *et al* (2011). However, Kaur (2018) observed reduced total sugars under shading treatment. The higher soluble sugar content under drought condition may also be attributed to high amylase activity, which breaks down the starch under drought stress (Vaezi 2005).

**Reducing sugars**

Accumulation of RS was 9.8 mg/g DW higher than irrigated. Under irrigated, RS content was lower in MLM 41-13-2 (8.9 mg/g FW) and higher in MCN 09-40 (26.6 mg/g DW) while under rainfed it was lower in MLM 41-13-2 (17.2 mg/g DW) and higher in JC 210-335 (39.0 mg/g DW). Increase in reducing sugars under moisture stress varied from 19.7% (CSR 1163) to 116.6% (JC 210-335) (Table 11, Fig. 13).

**Non- reducing sugars**

Non- reducing sugars (NRS) were higher (9.9 mg/g DW) under rainfed than irrigated condition. Accumulation of NRS was lower in MCN 09-40 (41.8 mg/g DW) and higher in MLM 41-13-2 (60.4 mg/g DW) with irrigation module, whereas under rainfed, JC 210-335 (50.2 mg/g DW) had lesser while it was more in MLM 41-13-2 (65.3 mg/g DW). The increase of NRS ranged from 0.2% (JC 210-335) to 43.2% (CSR 1163) (Table 11, Fig. 13).

Kaur and Sharma (2015a) studied the effect of water stress on sugars under 3 irrigation regimes and were of the opinion that total sugars, reducing sugars and non-reducing sugars were highest under moisture stress and declined with the irrigation modules from one to two. Similarly, in lentil drought stress increased the amount of reducing sugars more (36-56%) than heat stress (11.3-24.4%) especially in tolerant genotypes as observed by Sehgal *et al* (2017). Increase in reducing sugars under non-irrigated condition was observed in *Triticum aestivum* (Khan and Naqvi 2012).

**Antioxidative enzymes**

The activity of antioxidative enzymes like catalase, superoxide dismutase and peroxidase increased under stress to tolerate oxidative damage caused by reactive oxygen species (ROS). Moisture stress had significant effect on enzyme activities and interactions (I×G) were also significant (Table 12, Fig. 13).

**Catalase (EC 1.11.1.6)**

Catalase activity increased by 41.6 mmol/min/g FW under rainfed over irrigated condition. Under irrigated condition, lowest enzyme activity was 35.8 mmol/min/g FW in RH 1518 and highest was 134.8 mmol/min/g FW in MLM 41-13-2.

Table 11: Effect of moisture stress on total soluble sugars, reducing sugars and non-reducing sugars (mg/g DW) in *B. juncea*

| Genotypes/ Treatment | Total soluble sugars | | | Reducing sugars | | | Non-reducing sugars | | |
|---|---|---|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Inc (%) | Irrigated | Rainfed | Inc (%) | Irrigated | Rainfed | Inc (%) |
| RH 1518 | 67.8±1.0 | 88.8±0.8 | **31.0** | 16.5±0.1 | 27.9±0.2 | **69.1** | 51.2±1.0 | 60.9±0.9 | **18.9** |
| JC 210-335 | 68.1±0.6 | 89.2±0.3 | **31.0** | 18.0±0.6 | 39.0±0.2 | **116.6** | 50.1±0.9 | 50.2±0.3 | **0.2** |
| CJRD 1261 | 64.8±0.6 | 85.9±0.3 | **32.6** | 11.4±0.3 | 22.4±0.5 | **96.5** | 53.3±0.4 | 63.5±0.5 | **19.1** |
| RB-50 | 64.0±0.5 | 89.7±0.2 | **40.2** | 13.2±0.5 | 26.6±0.4 | **101.5** | 50.8±0.2 | 63.1±0.1 | **24.2** |
| RH 406 | 66.1±0.2 | 89.3±0.4 | **35.1** | 20.3±0.1 | 27.0±0.2 | **33.0** | 45.8±0.3 | 62.3±0.4 | **36.0** |
| PBR 422 | 62.2±1.0 | 70.1±0.6 | **12.7** | 13.5±0.1 | 18.3±0.3 | **35.5** | 48.7±1.0 | 51.8±0.7 | **6.4** |
| ELM 38 | 69.6±0.4 | 90.1±0.3 | **29.4** | 24.9±0.2 | 32.7±0.3 | **31.3** | 44.7±0.6 | 57.5±0.3 | **28.6** |
| CSR 1163 | 66.2±0.4 | 89.4±0.4 | **35.0** | 22.9±0.2 | 27.4±0.3 | **19.7** | 43.3±0.3 | 62.0±0.5 | **43.2** |
| IAN | 69.6±0.4 | 89.5±0.3 | **28.6** | 26.2±0.2 | 37.8±0.4 | **44.3** | 43.4±0.5 | 51.7±0.6 | **19.1** |
| MCN 09-40 | 68.4±0.4 | 89.9±0.2 | **31.4** | 26.6±0.3 | 36.3±0.3 | **36.5** | 41.8±0.4 | 53.5±0.4 | **28.0** |
| MLM 41-13-2 | 69.3±0.7 | 82.4±0.3 | **18.9** | 8.9±0.4 | 17.2±0.4 | **93.3** | 60.4±0.9 | 65.3±0.7 | **8.1** |
| PBR 357 | 69.2±0.6 | 86.7±0.8 | **25.3** | 15.4±0.3 | 23.6±0.4 | **53.2** | 53.8±0.9 | 63.2±0.9 | **17.5** |
| **Average** | **67.1±0.6** | **86.8±0.4** | | **18.2±0.3** | **28.0±0.3** | | **48.9±0.6** | **58.8±0.5** | |
| CD (p=0.05) | I= 0.386, G= 0.946, I×G= 1.338 | | | I= 0.230, G= 0.563, I×G= 0.797 | | | I= 0.450, G= 1.103, I×G= 1.560 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype



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Table 12: Effect of moisture stress on antioxidative enzymes at flowering stage

| Genotypes/ Treatment | CAT (mmoles of $H_2O_2$ decomposed/min/g FW) | | | SOD (enzyme activity/min/g FW) | | | POD (mmoles of enzyme activity/min/g FW) | | |
|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Inc (%)** | **Irrigated** | **Rainfed** | **Inc (%)** | **Irrigated** | **Rainfed** | **Inc (%)** |
| RH 1518 | 35.8±0.3 | 49.5±0.2 | **38.2** | 97.7±0.4 | 119.5±0.4 | **22.3** | 1.06±0.05 | 1.10±0.04 | **3.8** |
| JC 210-335 | 131.5±0.2 | 358.9±0.4 | **172.9** | 82.4±0.4 | 147.4±0.4 | **78.9** | 0.35±0.01 | 1.05±0.05 | **200.0** |
| CJRD 1261 | 84.2±0.2 | 131.6±0.4 | **56.3** | 93.5±0.1 | 140.6±0.3 | **50.4** | 0.56±0.03 | 0.92±0.02 | **64.3** |
| RB-50 | 52.9±0.8 | 57.7±0.8 | **9.1** | 85.1±0.7 | 134.8±0.5 | **58.4** | 0.54±0.01 | 1.18±0.06 | **119.6** |
| RH 406 | 57.7±0.3 | 83.7±0.8 | **45.1** | 78.7±0.3 | 141.2±0.4 | **79.4** | 0.88±0.05 | 1.18±0.03 | **34.1** |
| PBR 422 | 94.5±0.6 | 107.0±0.6 | **13.2** | 82.0±0.2 | 144.3±0.4 | **76.0** | 0.94±0.04 | 1.01±0.04 | **7.4** |
| ELM 38 | 44.8±0.4 | 60.8±0.8 | **35.7** | 93.8±0.6 | 128.9±0.4 | **37.4** | 0.59±0.04 | 1.15±0.04 | **94.9** |
| CSR 1163 | 58.1±0.7 | 63.6±0.9 | **9.5** | 84.8±0.6 | 144.7±0.3 | **70.6** | 0.72±0.02 | 0.98±0.07 | **36.1** |
| IAN | 76.2±1.5 | 182.3±1.5 | **139.2** | 93.3±0.2 | 131.0±0.4 | **40.4** | 0.54±0.01 | 1.05±0.05 | **94.4** |
| MCN 09-40 | 49.9±0.7 | 53.8±1.3 | **7.8** | 98.8±0.4 | 147.7±0.3 | **49.5** | 0.55±0.04 | 0.79±0.05 | **43.6** |
| MLM 41-13-2 | 134.8±1.2 | 158.9±1.4 | **17.9** | 69.5±0.3 | 119.8±0.3 | **72.4** | 0.61±0.04 | 0.82±0.02 | **34.4** |
| PBR 357 | 81.6±1.0 | 93.9±0.8 | **15.1** | 82.6±0.4 | 125.6±0.5 | **52.1** | 0.52±0.03 | 1.00±0.07 | **92.3** |
| **Average** | **75.2±0.7** | **116.8±0.8** | | **86.8±0.4** | **135.5±0.6** | | **0.65±0.03** | **1.02±0.05** | |
| CD (p=0.05) | I= 0.583, G= 1.428, I×G= 2.019 | | | I= 0.279, G= 0.682, I×G= 0.965 | | | I= 0.030, G= 0.074, I×G= 0.104 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

Under rainfed condition, lowest activity was 49.5 mmol/min/g FW in RH 1518 and highest was 358.9 mmol/min/g FW found in JC 210-335. CAT activity ranged from 7.8% (MCN 09-40) to 172.9% (JC 210-335). Similar findings were observed in wheat where drought stress was imposed (Stoilova *et al* 2010). On the other hand, CAT activity decreased in rice seedlings under drought stress (Sharma and Dubey 2005). Moreover, catalase activity increased under shaded *B. juncea* as reported by Kaur (2018). Increase in catalase activity increased tolerance to oxidative stress by providing an energy efficient mechanism to remove hydrogen peroxide ($H_2O_2$) from the plant cell by decomposing hydrogen peroxide to water and molecular oxygen without consuming reductants (Ahmad *et al* 2010).

**Superoxide dismutase (EC 1.15.1.1)**

Superoxide dismutase (SOD) activity increased by 48.7 EA/min/g FW under rainfed condition over irrigated condition. Under irrigated condition, it was lowest in MLM 41-13-2 (69.5 EA/min/g FW) and highest in MCN 09-40 (98.8 EA/min/g FW) whereas under rainfed condition, it was lowest in RH 1518 (119.5 EA/min/g/FW) and highest in MCN 09-40 (147.7 EA/min/g FW). Increase in SOD activity varied from 22.3% in RH 1518 to 79.4% in RH 406. Kumari *et al* (2018b) demonstrated that more SOD activity was under drought stress than control in Indian mustard, with higher SOD activity in tolerant than sensitive variety. Similar trend was observed by Shafiq *et al* (2019) in maize. Similar increase in activity of Indian mustard was observed under shaded over control treatment (Kaur 2018). Increase in activity of SOD increased tolerance of variety by minimising the oxidative stress (Ahmad *et al* 2017).

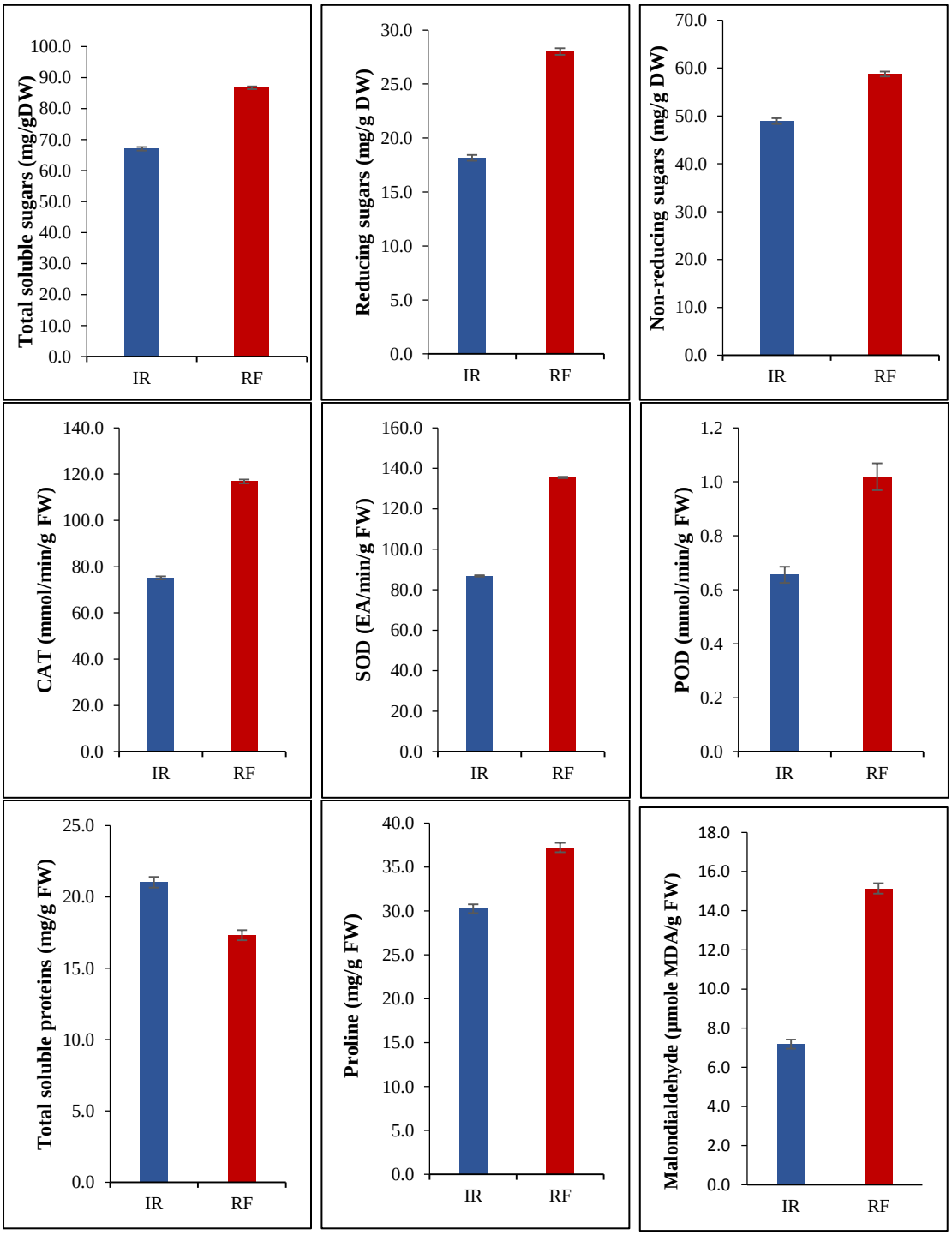
**Peroxidase (EC 1.11.1.7)**

Peroxidase activity (POD) was higher under rainfed condition by 0.37 mmol/min/g FW than under irrigated condition. POD activity was lowest in JC 210-335 (0.35 mmol/min/g FW) and highest in RH 1518 (1.06 mmol/min/g FW) under irrigated while it was lowest in MCN 09-40 (0.79 mmol/min/g FW) and highest in both RB-50 and RH 406 (1.18 mmol/min/g FW) under rainfed condition. POD activity increased from 3.8% (RH 1518) to 200.0% (JC 210-335) due to moisture stress (Table 13, Fig. 13). Drought stress increased peroxidase activity as observed by Abedi and Pakniyat (2010) in *Brassica napus* with minimum increase in cultivar Hyola 308 and maximum in cultivars Licord and Zarfam. A similar study in Indian mustard cultivars by Kumari *et al* (2018b) reported that under drought stress POD activity increased to more extent in RH 0406 than RH 0749. However, under shaded *B. juncea* genotypes, POD activity increased as well (Kaur 2018).

**Total soluble proteins**

Total soluble proteins (TSP) decreased under moisture stress and reduction was 17.6% over irrigated condition. Under irrigated condition, TSP concentration ranged from 16.4 (PBR 357) to 25.8 mg/g FW (RB-50) whereas under rainfed condition, from 14.5 (RH 1518) to 21.4 mg/g FW (IAN). Least reduction was 5.7% (JC 210-335) and highest 34.0%

(CJRD 1261) (Table 13, Fig. 13). Decreased protein content under drought stress has also been reported by Rezayian *et al* (2018) in *B. napus*. In rapeseed, water availability was also an indirect indicator of higher protein content. Kaur (2012) reported decrease in TSP under restricted moisture and moisture stress conditions. During water deficit, soluble proteins decreased due to severe reduction in rate of photosynthesis and non-availability of precursors needed for protein synthesis (Khanum *et al* 2019).




IR= Irrigated and RF= Rainfed

Fig. 13: Upregulation of metabolites and antioxidative enzymes under moisture stress

Table 13: Effect of moisture stress on total soluble proteins, proline and malondialdehyde at flowering stage

| Genotypes/ Treatment | Total soluble proteins (mg/g FW) | | | Proline (mg/g DW) | | | Malondialdehyde (µmoles MDA/g FW) | | |
|---|---|---|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Inc (%) | Irrigated | Rainfed | Inc (%) |
| RH 1518 | 18.8±0.6 | 14.5±0.2 | **22.9** | 32.1±1.0 | 39.2±0.8 | **22.1** | 8.6±0.2 | 14.8±0.2 | **72.1** |
| JC 210-335 | 17.4±0.3 | 16.4±0.2 | **5.7** | 22.6±0.1 | 24.2±0.7 | **7.1** | 5.1±0.2 | 15.1±0.2 | **196.1** |
| CJRD 1261 | 25.5±0.5 | 16.8±0.4 | **34.0** | 22.1±0.8 | 38.2±0.6 | **69.0** | 7.1±0.1 | 15.0±0.2 | **111.3** |
| RB-50 | 25.8±0.3 | 21.1±0.2 | **18.2** | 33.4±0.1 | 39.7±0.1 | **18.9** | 7.1±0.2 | 21.9±0.5 | **208.4** |
| RH 406 | 18.1±0.4 | 16.2±0.2 | **10.1** | 30.5±0.7 | 44.4±0.5 | **45.6** | 7.0±0.4 | 18.1±0.2 | **158.6** |
| PBR 422 | 25.4±0.5 | 18.0±0.1 | **29.2** | 28.9±0.4 | 33.1±0.3 | **14.5** | 5.6±0.3 | 13.0±0.3 | **132.1** |
| ELM 38 | 20.0±0.4 | 17.3±0.2 | **13.8** | 29.5±0.6 | 32.8±0.6 | **11.2** | 7.3±0.2 | 17.1±0.3 | **134.2** |
| CSR 1163 | 19.0±0.3 | 17.0±0.5 | **10.5** | 31.1±0.4 | 32.9±0.5 | **5.8** | 7.8±0.3 | 10.9±0.1 | **39.7** |
| IAN | 23.7±0.5 | 21.4±0.7 | **9.6** | 29.6±0.7 | 39.7±0.5 | **34.1** | 6.9±0.5 | 11.9±0.3 | **72.5** |
| MCN 09-40 | 17.6±0.3 | 16.0±0.6 | **9.4** | 34.7±0.2 | 45.7±0.5 | **31.7** | 7.4±0.1 | 15.5±0.2 | **109.5** |
| MLM 41-13-2 | 24.6±0.3 | 18.2±0.7 | **25.9** | 35.8±0.9 | 42.4±0.4 | **18.4** | 8.2±0.2 | 13.8±0.3 | **68.3** |
| PBR 357 | 16.4±0.1 | 14.9±0.3 | **9.2** | 32.7±0.2 | 34.7±1.0 | **6.1** | 8.3±0.1 | 14.7±0.3 | **77.1** |
| **Average** | **21.0±0.4** | **17.3±0.4** | | **30.2±0.5** | **37.2±0.5** | | **7.2±0.2** | **15.1±0.3** | |
| CD (p=0.05) | I= 0.286, G= 0.702, I×G= 0.992 | | | I= 0.398, G= 0.975, I×G= 1.379 | | | I= 0.197, G= 0.482, I×G= 0.681 | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

**Proline**

Proline content increased to provide osmosprotection in stressed plants. The increase in proline content was 7 mg/g DW under rainfed condition. Amount of proline was lesser in CJRD 1261 (22.1 mg/g DW) and more in MLM 41-13-2 (35.8 mg/g DW) under irrigated module whereas lesser proline was in JC 210-335 (24.2 mg/g DW) and more in MCN 09-40 (45.7 mg/g DW) under moisture stress. Increased proline content varied from 5.8% (CSR 1163) to 69.0% (CJRD 1261) (Table 13, Fig. 13).

Shafiq *et al* (2019) reported significant increase in proline content in maize under drought stress. Accumulation of higher proline content improved tolerance against drought stress (Bajji *et al* 2001). Proline content also increased in Indian mustard with increasing moisture stress (Kaur 2012) and increased salinity levels (Kannu Priya 2019). Proline increased tolerance by maintaining redox balance and protein integrity in plants (Szabo and Savoure 2010).

**Malondialdehyde**

Under rainfed condition, malondialdehyde (MDA) content increased to 7.9 mg/g FW as compared to irrigated condition. The amount of malondialdehyde was lesser in JC 210-335 (5.1 mg/g FW) and more in RH 1518 (8.6 mg/g FW) whereas under rainfed condition it was lesser in CSR 1163 (10.9 mg/g FW) and more in RB-50 (21.9 mg/g FW). MDA content increased from 39.7% (CSR 1163) to 208.4% (RB-50) (Table 13; Fig. 13).

Malondialdehyde (MDA) increased in drought stressed canola cultivars in both roots and shoots (Mirzaee *et al* 2013). Similar increased trend was observed in rapeseed seedlings by Bhuiyan *et al* (2019), and with shading in *B. juncea* (Kaur 2018). Increased MDA content indicated the increase in membrane injury as MDA is the ultimate product of membrane lipid peroxidation (Cunhua *et al* 2010).

**Correlation between biochemical parameters and seed yield**

Correlation of biochemical parameters with seed yield under both irrigated and rainfed conditions are depicted in Table 14. Under irrigated, catalase (CAT) was negatively associated with seed yield (r= -0.531) and highly significant negative correlation (r= -0.841**) under rainfed condition. Superoxide dismutase (SOD) had positive correlation with seed yield (r= 0.236) but negative correlation under irrigated and rainfed conditions respectively (r= -0.18). Peroxidase (POD) had significant positive correlation with seed yield (r= 0.632*) under irrigated module while weak positive correlation (r= 0.032) existed under moisture stress. Total soluble proteins had weak negative association with seed yield under irrigated (r= -0.079) and rainfed (r= -0.313) conditions. Proline content was weakly and positively associated with seed yield under both irrigated (r= 0.286) and rainfed (r= 0.377) conditions. Total soluble sugars (r= -0.239), reducing sugars (r= -0.016) and non-reducing sugars (r= -0.09) were negatively associated with seed yield under irrigated condition. Under

Table 14: Correlation coefficients of biochemical parameters under irrigated and rainfed conditions

| **Irrigated** | **CAT** | **SOD** | **POD** | **TSP** | **Proline** | **TSS** | **RS** | **NRS** | **MDA** | **SY** |
|---|---|---|---|---|---|---|---|---|---|---|
| **CAT** | 1 | | | | | | | | | |
| **SOD** | -0.669* | 1 | | | | | | | | |
| **POD** | -0.412 | 0.044 | 1 | | | | | | | |
| **TSP** | 0.224 | -0.148 | 0.032 | 1 | | | | | | |
| **Proline** | -0.29 | -0.165 | 0.238 | -0.081 | 1 | | | | | |
| **TSS** | 0.059 | 0.126 | -0.380 | -0.502 | 0.241 | 1 | | | | |
| **RS** | -0.509 | 0.550 | -0.104 | -0.498 | 0.060 | 0.426 | 1 | | | |
| **NRS** | 0.589* | -0.551 | -0.056 | 0.325 | 0.042 | -0.020 | -0.913** | 1 | | |
| **MDA** | -0.417 | 0.118 | 0.245 | -0.163 | 0.642* | 0.399 | -0.058 | 0.244 | 1 | |
| **SY** | -0.531 | 0.236 | 0.632* | -0.079 | 0.286 | -0.239 | -0.016 | -0.090 | 0.401 | 1 |
| **Rainfed** | **CAT** | **SOD** | **POD** | **TSP** | **Proline** | **TSS** | **RS** | **NRS** | **MDA** | **SY** |
| **CAT** | 1 | | | | | | | | | |
| **SOD** | 0.218 | 1 | | | | | | | | |
| **POD** | -0.088 | -0.106 | 1 | | | | | | | |
| **TSP** | 0.114 | 0.004 | 0.176 | 1 | | | | | | |
| **Proline** | -0.557 | -0.221 | -0.237 | 0.107 | 1 | | | | | |
| **TSS** | -0.033 | -0.072 | 0.241 | -0.061 | 0.134 | 1 | | | | |
| **RS** | 0.333 | 0.303 | 0.182 | 0.074 | -0.181 | 0.664* | 1 | | | |
| **NRS** | -0.474 | -0.474 | 0.011 | -0.161 | 0.378 | 0.167 | -0.627* | 1 | | |
| **MDA** | -0.226 | -0.030 | 0.505 | 0.140 | 0.235 | 0.302 | 0.033 | 0.270 | 1 | |
| **SY** | -0.841** | -0.180 | 0.032 | -0.313 | 0.377 | -0.335 | -0.502 | 0.314 | -0.022 | 1 |

*Significant at 5%, **Significant at 1%

CAT- Catalase, SOD- Superoxide dismutase, POD- Peroxidase, TSP- Total soluble proteins, TSS- Total soluble sugars, RS- Reducing sugars, NRS- Non-reducing sugars, MDA- Malondialdehyde, SY- Seed yield

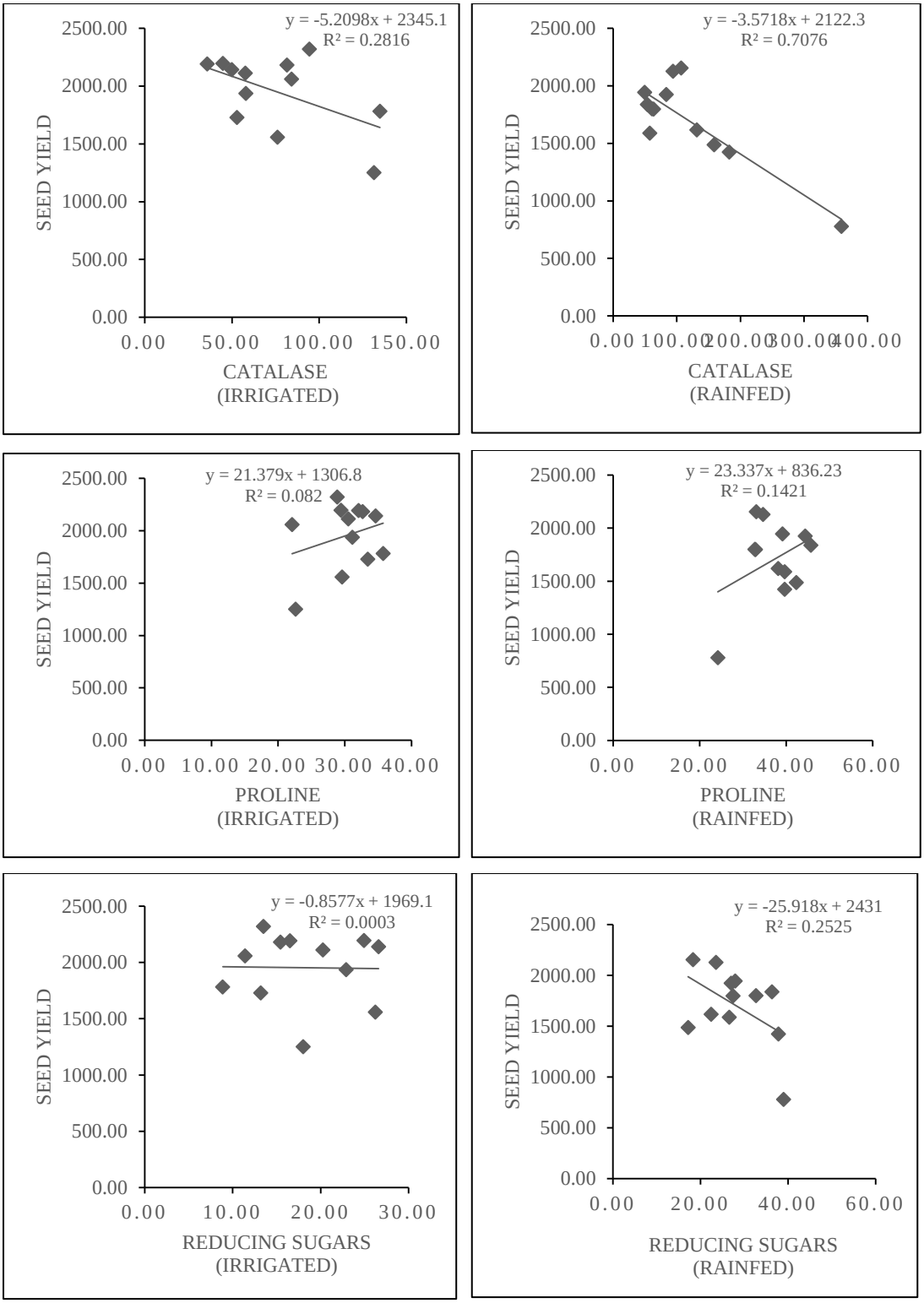



Fig. 14: Relationship between catalase, proline, reducing sugars with seed yield

rainfed condition, TSS (r= -0.335) and RS (r= -0.502) were negatively correlated while NRS had weak positive association with seed yield (r= 0.314). Malondialdehyde had positive correlation with seed yield (r= 0.401) under irrigated module but negative correlation with seed yield (r= -0.022) under rainfed. CAT was negatively correlated with SOD (r= -0.669*) and positively correlated with NRS (r= 0.589*) under irrigated condition. Proline was

positively correlated with MDA (r= 0.642*) under irrigated module. TSS was positively correlated with RS (r= 0.664*) under moisture stress. However, high negative association existed between RS and NRS under both irrigated (r= -0.913**) and rainfed (r= -0.627*) conditions.

Catalase had strong relationship with seed yield ($R^2$= 0.7076) under stress as compared to irrigated module. Relationship between proline ($R^2$= 0.1421) and reducing sugars ($R^2$= 0.2525) was better under stress relative to irrigated condition (Fig. 14). Bhardwaj (2017) also reported negative correlation between antioxidative enzymes and malondialdehyde with drought in *B. juncea*. It was observed because antioxidative enzymes play key role in eliminating $H_2O_2$ and MDA, thus protecting cell membrane integrity. In this study proline had a weak association with seed yield under both stressed and non-stressed conditions which was in agreement with the study of Majidi *et al* (2015) in *Brassica species*.

**Growth parameters**

Moisture stress reduced the growth parameters significantly in the studied genotypes, however differences existed for these traits in the studied genotypes. Interactive effects (I×G) were significant only for plant height (Table 15).

**Plant height**

Plant height decreased by 11.3% due to moisture stress. Under irrigated condition, it ranged from 149.7 (JC 210-335) to 227.8 cm (PBR 357) and from 142.5 (JC 210-335) to 191.0 cm (MCN 09-40) under rainfed condition. Amongst all genotypes, JC 210-335 suffered least reduction in plant height (4.8%) and PBR 357 (19.5%) maximum reduction. Decrease in plant height under water deficit was also recorded in canola by Moaveni *et al* (2010) and later by Ashraf *et al* (2013). Plant height was more under timely sown than under late sown Indian mustard genotypes as observed by Kumar *et al* (2017a) and earlier by Kaur (2016) in Ethiopian mustard.

**Main shoot length**

Reduction in main shoot length (MSL) due to moisture stress was 6.7%. MSL was 63.0 cm in IAN and 93.4 cm in CSR 1163 with irrigation whereas under rainfed again IAN had 55.9 cm and PBR 357 86.8 cm MSL. RH 406 had decline of 0.7% while 14.1% in CSR 1163 (Table 15). Our results are in agreement with the study of rapeseed where length of main inflorescence was significantly reduced with reduction in available water content in soil as reported by Germchi *et al* (2010). Similar decreasing trend was observed by Kaur (2012) with decreased number of irrigations in genotypes of *B. juncea* and *B. napus*.

**Primary branches**

Primary branches were reduced to 11.5% due to moisture stress and varied from 4.2 (RH 1518) to 6.6 (JC 210-335) under irrigated condition whereas from 3.9 (PBR 357) to 5.5 (CJRD 1261) under rainfed condition. Reduction in primary branch number was from 1.6% in

RH 1518 to 21.2% in JC 210-335. Chauhan *et al* (2007) reported reduced number of primary branches under drought stress. Decrease in number of primary branches/plant was observed with decrease in number of irrigations in Indian mustard by Jat *et al* (2018). Earlier similar decline was observed under late sown as compared to timely sown Indian mustard by Kumar *et al* (2017a).

**Secondary branches**

Secondary branches decreased by 17.5% with moisture stress as compared to irrigated condition. Under irrigated condition, secondary branches were 6.6 in JC 210-335 and 12.9 in IAN whereas 5.7 in RB-50 and 11.5 in CJRD 1261 under moisture stress. Reduction in secondary branch number ranged from 3.8 in PBR 422 to 26.8 in MCN 09-40 and MLM 41-13-2. Reduction in secondary branches was 21.05% in Indian mustard under drought stress as reported by Singh *et al* (2018). The reduced plant growth was an adaptive response to stress rather than as a secondary consequence of deficiency of resources (Rollins *et al* 2013).

**Yield and yield attributes**

Seed yield and yield attributes varied significantly in the genotypes and with irrigated module. However, differences existed for these traits under moisture stress. Interactions (I×G) were non- significant for siliquae on main raceme, siliqua length and oil content (Table 16, 17 and 18).

**Siliquae on main raceme**

Siliquae on main raceme (SMS) decreased by 7.1% due to moisture stress. It ranged from 26.8 in IAN and 58.9 in ELM 38 under irrigated condition whereas under rainfed condition variation existed in the same genotypes from 21.1 in IAN and 57.9 in ELM 38. Siliquae on main raceme were reduced by 0.5% in PBR 357 and 21.1% in IAN. A similar reduction in SMS was observed in Indian mustard by Singh *et al* (2018) and recently by Kumari *et al* (2019). SMS reduced significantly under restricted moisture and moisture stress as compared to normal moisture condition as per reports of Kaur (2012) in both the species of Brassica i.e., *B. juncea* and *B. napus*.

**Total siliquae/plant**

Total siliquae per plant (TS) decreased by 8.7% and were 176.0 in RB-50 and 331.0 in MLM 41-13-2 under irrigated condition while under rainfed condition siliquae/plant were 162.8 in RB-50 and 311.9 in ELM 38. Reduction in silquae/plant was 0.7% in ELM 38 and 25.4% in MLM 41-13-2. Khan *et al* (2010) reported that number of siliquae/plant decreased in canola with the decrease in number of irrigations. The decrease in pod number per plant was also observed in groundnut (Mabhaudhi and Modi 2013). TS reduced in Indian mustard genotypes under shading stress (Kaur 2018).

Table 15: Effect of moisture stress on growth parameters in *B. juncea* genotypes

| Genotypes/ Treatment | Plant height (cm) | | | Main shoot length (cm) | | | Primary branches/plant | | | Secondary branches/plant | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** |
| RH 1518 | 198.1±4.3 | 175.5±5.2 | **11.4** | 89.4±3.8 | 81.0±1.4 | **9.4** | 4.2±0.7 | 4.1±0.2 | **1.6** | 8.3±0.9 | 7.2±0.6 | **14.0** |
| JC 210-335 | 149.7±1.6 | 142.5±0.7 | **4.8** | 69.3±5.9 | 63.5±2.9 | **8.4** | 6.6±0.5 | 5.2±0.3 | **21.2** | 6.6±0.5 | 6.1±0.8 | **7.6** |
| CJRD 1261 | 191.3±4.5 | 170.5±4.7 | **10.9** | 66.2±0.9 | 57.7±1.8 | **12.9** | 6.2±0.5 | 5.5±0.4 | **10.8** | 12.7±0.8 | 11.5±0.8 | **9.5** |
| RB-50 | 192.2±4.0 | 176.9±3.1 | **8.0** | 88.7±1.8 | 86.5±2.9 | **2.5** | 4.6±0.6 | 4.0±0.4 | **13.0** | 6.9±0.1 | 5.7±0.7 | **17.3** |
| RH 406 | 195.7±0.9 | 170.7±2.4 | **12.8** | 80.3±0.7 | 79.7±0.7 | **0.7** | 5.4±0.4 | 5.1±0.2 | **4.9** | 12.2±0.7 | 9.7±0.8 | **20.2** |
| PBR 422 | 203.7±7.1 | 187.7±2.6 | **7.8** | 79.3±0.9 | 77.4±5.5 | **2.4** | 4.8±0.5 | 4.3±0.3 | **11.1** | 8.7±0.7 | 8.4±0.9 | **3.8** |
| ELM 38 | 206.9±8.3 | 180.3±6.2 | **12.9** | 82.3±4.5 | 78.0±2.7 | **5.1** | 5.2±0.2 | 4.8±0.1 | **7.7** | 12.1±0.3 | 9.5±0.6 | **21.0** |
| CSR 1163 | 202.0±6.0 | 187.2±6.8 | **7.3** | 93.4±3.6 | 80.2±4.1 | **14.1** | 5.1±0.7 | 4.3±0.4 | **16.9** | 11.1±0.8 | 8.8±0.6 | **21.0** |
| IAN | 182.0±6.5 | 152.5±8.8 | **16.2** | 63.0±3.0 | 55.9±6.4 | **11.2** | 5.1±0.4 | 4.5±0.3 | **11.8** | 12.9±0.8 | 11.2±0.3 | **13.4** |
| MCN 09-40 | 209.8±4.8 | 191.0±4.0 | **9.0** | 89.9±6.0 | 85.3±1.8 | **5.1** | 5.3±0.6 | 4.7±0.5 | **11.3** | 10.5±0.5 | 7.7±0.7 | **26.8** |
| MLM 41-13-2 | 211.7±5.9 | 185.5±1.9 | **12.4** | 92.6±4.2 | 86.0±4.3 | **7.2** | 5.6±0.2 | 5.1±0.3 | **8.3** | 12.3±0.6 | 9.0±0.3 | **26.8** |
| PBR 357 | 227.8±2.4 | 183.4±5.2 | **19.5** | 89.4±1.1 | 86.8±8.7 | **2.9** | 4.6±0.2 | 3.9±0.1 | **15.9** | 9.2±0.5 | 7.7±0.6 | **16.7** |
| **Average** | **197.6±4.7** | **175.3±4.3** | | **82.0±3.0** | **76.5±3.6** | | **5.2±0.5** | **4.6±0.3** | | **10.3±0.6** | **8.5±0.6** | |
| CD (p=0.05) | I= 3.429, G= 8.399, I×G= 11.879 | | | I= 2.658, G= 6.511, I×G= NS | | | I= 0.283, G= 0.692, I×G= NS | | | I= 0.442, G= 1.083, I×G= NS | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype interaction

**1000 seed weight**

Seed weight was reduced by 21.2% under rainfed condition. Seed size varied from 3.4 in ELM 38 to 6.7 g in RH 1518 under irrigated condition, however, JC 210-335 had 2.6 g of seed weight while 4.4 g was recorded in two genotypes (MCN 09-40 and PBR-357). Reduction in seed weight was 2.7% in PBR-422 and 40.6% in CSR 1163. Similar declining trend of 1000 seed weight under rainfed was also observed by Kumari *et al* (2019) in Indian mustard. Study of Moaveni *et al* (2010) revealed that 1000 seed weight was significantly reduced in *B. napus* under interrupted irrigation condition at different stages as compared to irrigated control, however decline was minimum when irrigation was interrupted at seed filling stage.

**Siliqua length**

Siliqua length decreased by 7.8% with moisture stress and was 3.8 cm in CJRD 1261 and 6.4 cm in RB-50 under irrigated condition whereas again in these two genotypes it was 3.7 cm in CJRD 1261 and 6.2 cm in RB-50 under rainfed condition. Siliqua length was reduced to 3.2% in RB-50 and 13.9% in JC 210-335. Our findings are in agreement with those of Jat *et al* (2018) in *B. juncea* where siliqua length was least in no irrigation treatment and subsequently increased with increasing number of irrigations. Similar results have been endorsed earlier by Naderikharaji *et al* (2008) in *B. napus*. Kaur (2012) also reported similar trend in *B. juncea* and *B. napus* with decreasing number of irrigations.

**Seed-filling**

Moisture stress had adverse impact on seed filling as number of seeds/siliqua and percentage of developed seeds decreased while shrivelled seeds percentage increased under rainfed condition. However, interactions (I×G) were also significant (Table 17, Fig. 15).

**Seeds per siliqua**

Seeds/siliqua decreased under rainfed by 14.2% and were 11.3 in CSR 1163 and 16.2 in RH 406 under irrigated condition whereas in the same genotype CSR 1163 seeds/siliqua were 8.2 and 14.2 in JC 210-335. Reduction in seeds/siliqua ranged from 3.7% (PBR 357) to 28.8% (RH 406). Reducing trend was reported for seed number per siliqua under drought condition in canola varieties (Nasri *et al* 2008) and also in Ethiopian mustard (Verma *et al* 2018).

**Developed seeds**

Developed seeds represented as percentage of total seeds/siliqua were reduced in all the genotypes under rainfed as compared to irrigated condition. Developed seeds were 94.1% (IAN) and 98.0% (CJRD 1261 and RB-50) under irrigated whereas 81.7% (PBR 357) and 96.8% (ELM 38) under rainfed condition. The percentage of developed seeds were comparable, 98.0% in CJRD 1261 and RB-50, 97.2% in ELM 38 and CSR 1163, 95.3% in MCN 09-40 and MLM 41-13-2, 96.5% in RH 406 and PBR 357 under irrigated. Similarly,

Table 16: Effect of moisture stress on siliquae on main raceme, total siliquae/plant, 1000 seed weight and siliqua length in *B. juncea*

| Genotypes/ Treatment | Siliquae on main raceme | | | Total siliquae/plant | | | 1000 seed weight (g) | | | Siliqua length (cm) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** | **Irrigated** | **Rainfed** | **Red (%)** |
| RH 1518 | 52.5±2.7 | 45.2±4.4 | **13.9** | 233.3±12.7 | 209.4±12.4 | **10.3** | 6.7±0.2 | 4.1±0.4 | **38.8** | 5.7±0.5 | 5.3±0.3 | **7.9** |
| JC 210-335 | 47.8±3.2 | 44.8±4.0 | **6.3** | 239.0±15.6 | 212.2±12.4 | **11.2** | 3.6±0.6 | 2.6±0.3 | **27.8** | 6.2±0.6 | 5.4±0.4 | **13.9** |
| CJRD 1261 | 51.3±2.4 | 48.3±2.4 | **6.0** | 291.4±16.4 | 271.9±9.2 | **6.7** | 4.1±0.5 | 2.7±0.4 | **34.3** | 3.8±0.1 | 3.7±0.1 | **4.0** |
| RB-50 | 44.6±1.2 | 41.4±2.5 | **7.1** | 176.0±15.6 | 162.8±11.8 | **7.5** | 4.4±0.4 | 3.8±0.3 | **13.1** | 6.4±0.2 | 6.2±0.5 | **3.2** |
| RH 406 | 45.4±1.6 | 43.9±2.6 | **3.2** | 286.1±11.2 | 272.2±9.4 | **4.9** | 5.8±0.8 | 4.3±0.6 | **25.3** | 5.3±0.2 | 4.8±0.3 | **10.1** |
| PBR 422 | 52.2±2.9 | 50.9±2.4 | **2.4** | 292.6±13.0 | 270.0±9.6 | **7.7** | 4.2±0.4 | 4.1±0.7 | **2.7** | 4.6±0.1 | 4.4±0.1 | **5.1** |
| ELM 38 | 58.9±3.1 | 57.9±3.3 | **1.8** | 314.1±18.8 | 311.9±15.8 | **0.7** | 3.4±0.7 | 3.3±0.4 | **3.1** | 4.3±0.3 | 3.9±0.1 | **9.8** |
| CSR 1163 | 56.8±3.5 | 51.3±4.3 | **9.6** | 306.7±7.8 | 287.7±9.2 | **6.2** | 6.0±0.6 | 3.6±0.4 | **40.6** | 4.6±0.4 | 4.1±0.3 | **11.7** |
| IAN | 26.8±1.8 | 21.1±2.8 | **21.1** | 239.9±13.7 | 232.9±8.2 | **2.9** | 5.2±0.5 | 4.2±0.4 | **19.6** | 6.3±0.3 | 5.8±0.1 | **8.5** |
| MCN 09-40 | 57.0±0.9 | 51.9±2.5 | **8.9** | 281.0±18.9 | 268.8±17.1 | **4.3** | 5.1±0.3 | 4.4±0.7 | **14.5** | 4.5±0.1 | 4.1±0.2 | **9.3** |
| MLM 41-13-2 | 58.1±3.0 | 52.7±4.3 | **9.4** | 331.0±7.9 | 247.1±12.0 | **25.4** | 3.7±0.4 | 3.6±0.2 | **3.0** | 4.4±0.2 | 4.2±0.1 | **4.2** |
| PBR 357 | 52.7±7.2 | 52.5±3.5 | **0.5** | 238.1±6.0 | 201.0±3.0 | **15.6** | 4.8±0.4 | 4.4±0.7 | **8.1** | 5.3±0.2 | 5.0±0.3 | **5.1** |
| **Average** | **50.4±2.8** | **46.8±3.2** | | **269.1±13.1** | **245.7±10.8** | | **4.7±0.5** | **3.7±0.5** | | **5.1±0.3** | **4.7±0.2** | |
| CD (p= 0.05) | I= 2.227, G= 5.455, I×G= NS | | | I= 8.373, G= 20.510, I×G= 29.005 | | | I= 0.339, G= 0.832, I×G= 1.176 | | | I= 0.195, G= 0.479, I×G= NS | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

developed seeds were at par in RH 1518 & RH 406 (95.7%), MLM 41-13-2 & JC 210-335 (94.9%), PBR 422 & CSR 1163 (95.4%) under moisture stress. Number of developed seeds were 11.0 in CSR 1163 and 15.6 in RH 406 under irrigated while 7.9 in CSR 1163 and 13.5 in JC 210-335 under moisture stress.

**Shrivelled seeds**

Shrivelled seeds represented as percentage of total seeds/siliqua were increased in all the genotypes under rainfed as compared to irrigated condition. Shrivelled seeds were 1.1% (ELM 38) and 4.9% (IAN) under irrigated whereas 3.1% (PBR 357) and 5.8% (IAN) under moisture stress. Number of shrivelled seeds were 0.16 in ELM 38 and 0.61 in IAN under irrigated while 0.37 in PBR 357 with comparable value 0.38 in CSR 1163 and 0.65 in RB-50 and IAN (0.65) under moisture stress. The impact of moisture stress on seed filling in the twelve *B. juncea* genotypes are vividly depicted in Figure 15.

Sehgal *et al* (2017) reported that high temperatures in drought stressed lentil plants at the time of seed filling caused a drastic reduction in seed quality and quantity which was due to the reduced supply of sucrose to the developing seeds leading to decrease in size and number of developed seeds whereas increase in number of shrivelled seeds. According to Ochatt (2015), moisture status greatly influenced the mechanism of synthesis and accumulation of various seed reserves and deficiency of water at this stage disrupts the seed filling. Sharma and Sardana (2013) reported that both fully developed seeds/siliqua and shrivelled seeds / siliqua were lesser in late sown than normal sown condition due to high temperature stress.

Moisture stress had adverse effect on yield. A significant effect of environment and genotypes was seen and their interactions were significant in biological yield, seed yield and harvest index but non-significant in oil content (Table 18).

**Biological yield**

Biological yield decreased by 6.2% due to moisture stress and varied from 7688.9 in JC 210-335 to 12711.1 kg/ha in PBR 422 under irrigated condition while again in the two genotypes 6218.9 (JC 210-335) to 12444.4 kg/ha (PBR 422) under rainfed condition. Biological yield was reduced to 0.4% in RB-50 and 19.1% in JC 210-335. Similar trend was recorded in biological yield where mean reduction was 13.5% in Indian mustard by Singh *et al* (2018). Biological yield was significantly lower under rainfed relative to irrigated condition in Bambara groundnut (Mabhaudhi and Modi 2013). Biological yield reduced under late sown as compared to timely sown *B. juncea* (Kumar *et al* 2017a). Moreover, BY increased with increase in nitrogen levels in canola genotypes (Gill 2018).

**Seed yield**

Seed yield was decreased by 12.7% due to moisture stress and was 1250.7 in JC 210-335 and 2319.1 kg/ha in PBR-422 under irrigated condition whereas same genotypes had

Table 17: Effect of moisture stress on seed filling in *B. juncea* genotypes

| Genotypes/ Treatment | Seeds/siliqua | | | Percentage of seeds | | | |
|---|---|---|---|---|---|---|---|
| | | | | Developed | | Shrivelled | |
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Irrigated | Rainfed |
| RH 1518 | 13.7±0.5 | 11.9±0.8 | **13.1** | 97.1±0.8 | 95.7±1.7 | 2.5±0.4 | 4.0±0.1 |
| JC 210-335 | 15.8±0.6 | 14.2±0.2 | **9.9** | 95.6±0.9 | 94.9±1.3 | 3.1±0.5 | 4.4±0.1 |
| CJRD 1261 | 12.3±0.7 | 10.2±0.4 | **17.0** | 98.0±0.2 | 96.0±1.9 | 2.2±0.1 | 3.9±0.6 |
| RB-50 | 13.0±0.6 | 11.5±0.2 | **11.3** | 98.0±0.9 | 94.0±2.6 | 3.2±0.8 | 5.7±0.3 |
| RH 406 | 16.2±0.3 | 11.5±0.9 | **28.8** | 96.3±1.3 | 95.7±0.7 | 1.9±0.5 | 5.5±0.7 |
| PBR 422 | 13.3±0.3 | 11.7±0.7 | **12.3** | 95.7±1.9 | 95.3±1.1 | 3.3±0.6 | 4.0±0.7 |
| ELM 38 | 14.6±0.6 | 10.9±0.2 | **24.9** | 97.1±3.0 | 96.8±11 | 1.1±0.3 | 4.8±0.1 |
| CSR 1163 | 11.3±0.7 | 8.2±0.4 | **27.5** | 97.2±0.8 | 95.4±1.4 | 2.0±0.3 | 4.6±0.6 |
| IAN | 12.4±0.1 | 11.3±0.7 | **9.1** | 94.1±1.3 | 93.6±1.7 | 4.9±0.6 | 5.8±0.7 |
| MCN 09-40 | 12.2±0.3 | 11.4±0.7 | **7.2** | 95.3±0.9 | 89.0±5.6 | 2.3±0.5 | 4.2±1.0 |
| MLM 41-13-2 | 12.1±0.5 | 11.5±0.4 | **5.1** | 95.3±2.7 | 94.8±2.3 | 2.8±0.5 | 3.7±0.6 |
| PBR 357 | 12.7 ±0.5 | 12.2±0.2 | **3.7** | 96.5±1.1 | 81.7±4.7 | 2.0±0.3 | 3.1±0.2 |
| **Average** | **13.3±0.5** | **11.4±0.5** | | **96.4±1.2** | **93.6±2.2** | **2.6±0.5** | **4.5±0.5** |
| CD (p= 0.05) | I= 0.318, G= 0.778, I×G= 1.100 | | | I= .416, G= 3.468, I×G= 4.904 | | I= 0.342, G= 0.837, I×G= 1.184 | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

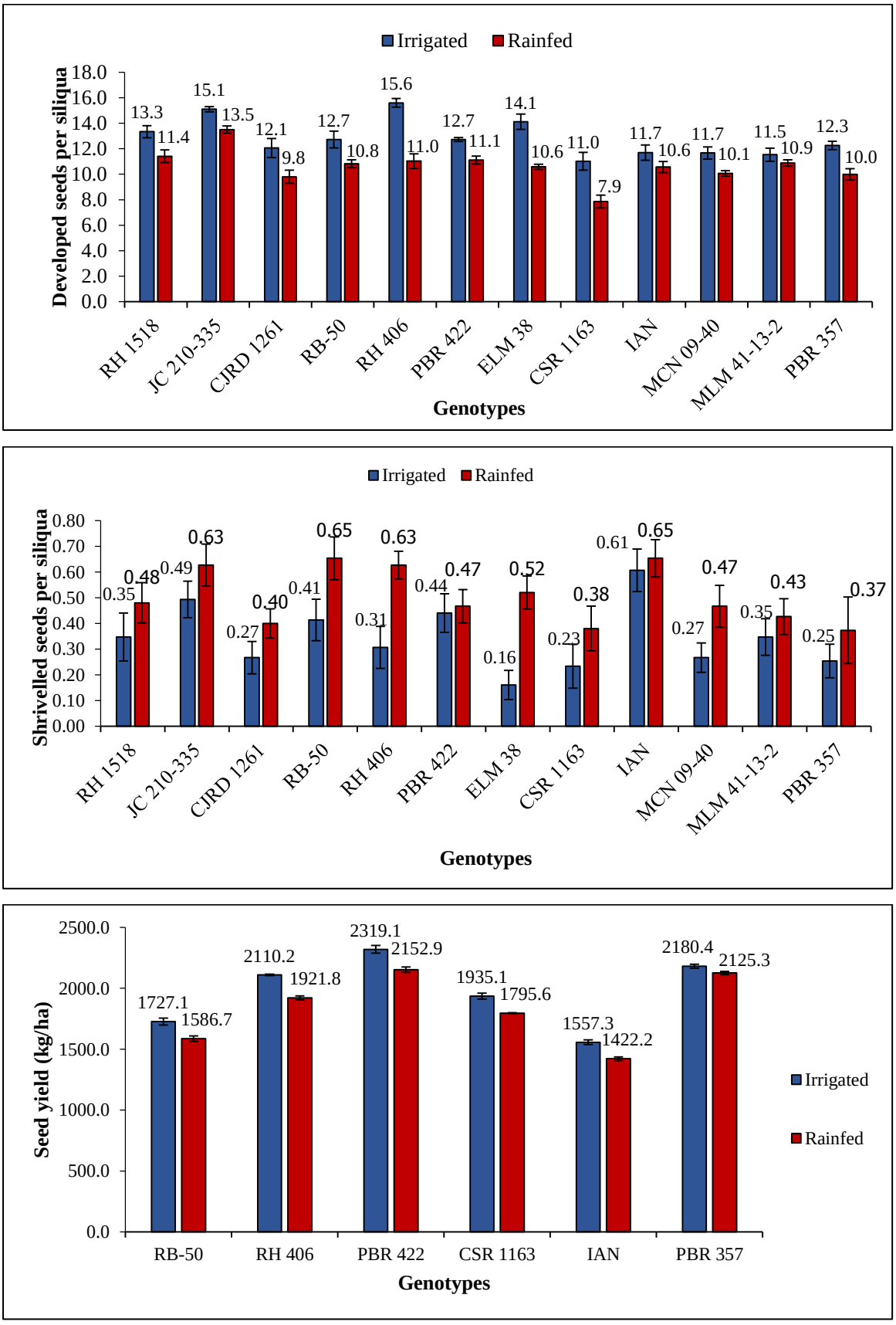



Fig. 15: Effect of moisture stress on seed filling and seed yield in *B. juncea* genotypes

reduced seed yield of 777.8 (JC 210-335) and 2152.9 kg/ha (PBR 422) under rainfed condition. Seed yield was reduced to 2.5% in PBR-357 and 37.8% in JC 210-335. Six genotypes had less than 10% reduction in seed yield under moisture stress (Fig. 15). Our results are in agreement with the findings of Chandra *et al* (2018) in *B. juncea*. The drought induced reduction in yield might be due to reduced photosynthetic rate and disturbed assimilate partitioning (Farooq *et al* 2009). Moreover, the decrease in seed yield under water deficit stress was due to disturbed growth and nutrient uptake of plants as reported by Raza *et al* (2015) in different oilseed rape varieties.

**Harvest index**

Harvest index (HI) decreased by 7.9% due to moisture stress and was 15.1 in IAN and 19.8% in RH 1518 under irrigated condition while 12.5 in JC 210-335 and 18.2% in RH 406 under rainfed condition. Reduction in HI was 3.2% in ELM 38 and 23.3% in JC 210-335. Similarly, decrease in harvest index was reported by Sodani *et al* (2017) in *B. juncea* and in *B. napus* genotypes with increasing water stress conditions by Germchi *et al* (2010).

**Oil content**

Oil content reduced by 1.5% under rainfed condition as compared to irrigated condition. It was 38.4% in MLM 41-13-2 and 40.7% in JC 210-335 under irrigated condition however, under rainfed condition oil content varied from 37.6% in ELM 38 to 40.4% in JC 210-335. Oil content was reduced to 0.2% in PBR 357 and 3.0% in ELM 38. Non-significant results were recorded by Deka *et al* (2018) in toria for oil content however, oil content was higher in irrigation regimes as compared to rainfed condition. Significant variations occurred among different irrigation scheduling of Indian mustard by Jat *et al* (2018) and earlier by Kaur (2012).

**Correlation between growth parameters, yield attributes and seed yield**

Plant height was significantly positively correlated with seed yield under irrigated (r= 0.772**) and rainfed (r= 0.772**) conditions. Main shoot length was positively associated with seed yield under irrigated (r= 0.405) and rainfed (r= 0.534) conditions. Plant height was significantly and positively correlated with main shoot length under irrigated (r= 0.663*) and rainfed (r= 0.759**) and also with biological yield under irrigated (r= 0.879**) and rainfed (r= 0.769**) conditions. Under rainfed condition, plant height was significantly and positively correlated with harvest index (r= 0.653*). Plant height had strong negative correlation with shrivelled seeds (r= -0.656*) under irrigated condition and significant negative correlation with developed (r= -0.633*) and shrivelled (r= -0.649*) seeds under rainfed condition. Main shoot length under irrigated module was positively correlated with siliquae on main raceme (r= 0.655*). Number of primary branches had negative association with seed yield under irrigated (r= -0.525) and rainfed (r= -0.505) conditions. However, number of secondary branches had weak but positive association under irrigated (r= 0.213)

Table 18: Effect of moisture stress on biological yield, seed yield, harvest index and oil content

| Genotypes/ Treatment | Biological yield (kg/ha) | | | Seed yield (kg/ha) | | | Harvest index (%) | | | Oil content (%) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Red (%) | Irrigated | Rainfed | Red (%) |
| RH 1518 | 11067±62 | 10978±27 | **0.8** | 2190.2±8.4 | 1942.2±11.9 | **11.3** | 19.8±0.1 | 17.7±0.1 | **10.6** | 39.9±0.2 | 39.0±0.3 | **2.3** |
| JC 210-335 | 7689±36 | 6219±42 | **19.1** | 1250.7±19.5 | 777.8±16.5 | **37.8** | 16.3±0.2 | 12.5±0.3 | **23.3** | 40.7±0.1 | 40.4±0.4 | **0.7** |
| CJRD 1261 | 11289±95 | 9333±47 | **17.3** | 2057.8±25.5 | 1616.0±14.8 | **21.5** | 18.2±0.3 | 17.3±0.1 | **5.0** | 39.9±0.4 | 39.0±0.2 | **2.2** |
| RB-50 | 10044±59 | 10000±52 | **0.4** | 1727.1±28.8 | 1586.7±22.5 | **8.1** | 17.2±0.3 | 15.9±0.3 | **7.7** | 39.2±0.4 | 38.8±0.2 | **1.0** |
| RH 406 | 10842±88 | 10533±63 | **2.8** | 2110.2±6.2 | 1921.8±16.1 | **8.9** | 19.5±0.2 | 18.2±0.2 | **6.3** | 39.7±0.9 | 38.9±0.1 | **2.1** |
| PBR 422 | 12711±95 | 12444±36 | **2.1** | 2319.1±31.4 | 2152.9±21.9 | **7.2** | 18.2±0.4 | 17.3±0.2 | **5.2** | 40.3±0.2 | 39.5±0.1 | **2.0** |
| ELM 38 | 12122±3 | 10264±22 | **15.3** | 2193.8±14.6 | 1798.2±32.5 | **18.0** | 18.1±0.1 | 17.5±0.3 | **3.2** | 38.8±0.2 | 37.6±0.3 | **3.0** |
| CSR 1163 | 11267±24 | 11200±82 | **0.6** | 1935.1±23.8 | 1795.6±4.9 | **7.2** | 17.2±0.2 | 16.0±0.1 | **6.7** | 39.4±0.4 | 38.9±0.4 | **1.2** |
| IAN | 10311±48 | 10089±55 | **2.2** | 1557.3±19.2 | 1422.2±15.2 | **8.7** | 15.1±0.1 | 14.1±0.1 | **6.7** | 39.0±0.1 | 38.2±0.3 | **2.0** |
| MCN 09-40 | 11733±62 | 11467±73 | **2.3** | 2139.6±9.8 | 1836.4±32.6 | **14.2** | 18.2±0.1 | 16.0±0.3 | **12.0** | 38.6±0.4 | 38.2±0.2 | **1.1** |
| MLM 41-13-2 | 10667±0.3 | 9422±36 | **11.7** | 1780.4±16.7 | 1485.3±16.8 | **16.6** | 16.7±0.2 | 15.8±0.1 | **5.6** | 38.4±0.6 | 38.1±0.5 | **0.8** |
| PBR 357 | 12678±54 | 12311±98 | **2.9** | 2180.4±16.2 | 2125.3±13.5 | **2.5** | 17.9±0.1 | 17.3±0.1 | **3.3** | 39.1±0.4 | 39.0±0.2 | **0.2** |
| **Average** | **11035±52** | **10355±53** | | **1953.5±18.3** | **1705.0±18.2** | | **17.7±0.2** | **16.3±0.2** | | **39.4±0.4** | **38.8±0.3** | |
| CD (p= 0.05) | I= 40.346,G=98.827, I×G=139.763 | | | I= 13.040, G= 31.941, I×G= 45.171 | | | I= 0.131, G= 0.322, I×G= 0.455 | | | I=0.236, G= 0.578, I×G= NS | | |

I= Irrigation, G= Genotypes, I×G= Irrigation × Genotype

and rainfed (r= 0.136) conditions with seed yield. Number of secondary branches were positively correlated with total siliquae per plant under irrigated (r= 0.672*) as well as rainfed (r= 0.626*) conditions. Siliquae on main raceme had positive association with seed yield under irrigated (r= 0.499) and rainfed (r= 0.357) conditions. Similarly, total siliquae/plant were positively associated under irrigated (r= 0.348) and rainfed (r= 0.252) conditions with seed yield.

Under irrigated condition, siliquae on main raceme had negative and significant correlation with siliqua length (r= -0.700*) and shrivelled seeds/siliqua (r= -0.693*) and again showed negative association with siliqua length (r= -0.699*) and shrivelled seeds/siliqua (r= -0.817**) under stress. Total siliquae per plant had highly significant negative correlation with siliqua length under both irrigated (r= -0.845**) and rainfed (r= -0.849**) conditions. Under irrigated condition, 1000 seed weight had weak positive association with seed yield (r= 0.27) while under rainfed condition the association was strong and positive with seed yield (r= 0.675*). 1000 seed weight had strong positive correlation with biological yield (r= 0.779**) under rainfed condition. Siliqua length had significant negative correlation with seed yield under both irrigated (r= -0.595*) and rainfed (r= -0.301) condition. Under irrigated condition, siliqua length had significantly negative correlation with biological yield (r= -0.600*). However, under both irrigated (r= 0.734**) and rainfed (r= 0.692*) conditions, siliqua length was positively correlated with shrivelled seeds/silique. Biological yield had highly significant positive correlation with seed yield under both irrigated (r= 0.913**) and rainfed (r= 0.951**) conditions. Similarly, HI had highly positive and significant association with seed yield under irrigated (r= 0.781**) and rainfed (r= 0.870**) conditions. Biological yield and harvest index were positively and strongly associated with each other under rainfed condition (r= 0.685*). Number of seeds/siliqua (r= -0.134), developed seeds/siliqua (r= -0.123) and shrivelled seeds/siliqua (r= -0.552) had negative correlation with seed yield under irrigated condition. Similar negative association was evident under rainfed condition for number of seeds/siliqua (r= -0.393), developed seeds/silique (r= -0.527) and shrivelled seeds/siliqua (r= -0.492) with yield. Total seeds/siliqua were positively correlated with developed seeds/siliqua under irrigated (r= 0.971**) and rainfed (r= 0.887**) conditions (Table 19a-19b).

Plant height ($R^2$= 0.596), biological yield ($R^2$= 0.9038) and harvest index ($R^2$= 0.7539) had relatively strong relationship with seed yield under rainfed condition as compared to irrigated module where relationship under latter were plant height ($R^2$= 0.5954), biological yield ($R^2$= 0.8333) and harvest index ($R^2$= 0.6134) with seed yield (Fig. 16). Similar findings were observed by Abbasian and Shirani-Rad (2011) in rapeseed cultivars under different moisture regimes. Singh *et al* (2018) reported a strong positive correlation

Table 19a: Correlation coefficients of growth and yield parameters under irrigated condition

| | PH | MSL | PB | SB | SMS | Total siliquae | 1000 SW | SL | BY | HI | Seeds/ siliqua | Developed seeds | Shrivelled seeds | SY |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **PH** | 1 | | | | | | | | | | | | | |
| **MSL** | 0.663* | 1 | | | | | | | | | | | | |
| **PB** | -0.586* | -0.541 | 1 | | | | | | | | | | | |
| **SB** | 0.299 | -0.197 | 0.201 | 1 | | | | | | | | | | |
| **SMS** | 0.490 | 0.655* | 0.016 | -0.054 | 1 | | | | | | | | | |
| **Total siliquae** | 0.326 | 0.110 | 0.333 | 0.672* | 0.542 | 1 | | | | | | | | |
| **1000 SW** | 0.154 | 0.280 | -0.536 | 0.020 | -0.174 | -0.201 | 1 | | | | | | | |
| **SL** | -0.511 | -0.182 | -0.219 | -0.550 | -0.700* | -0.845** | 0.230 | 1 | | | | | | |
| **BY** | 0.879** | 0.369 | -0.553 | 0.327 | 0.397 | 0.376 | 0.141 | -0.600* | 1 | | | | | |
| **HI** | 0.391 | 0.368 | -0.334 | -0.063 | 0.481 | 0.126 | 0.406 | -0.334 | 0.468 | 1 | | | | |
| **Seeds/siliqua** | -0.477 | -0.318 | 0.231 | -0.241 | -0.158 | -0.142 | -0.123 | 0.343 | -0.376 | 0.282 | 1 | | | |
| **Developed seeds** | -0.474 | -0.273 | 0.263 | -0.279 | -0.137 | -0.182 | -0.041 | 0.344 | -0.402 | 0.353 | 0.971** | 1 | | |
| **Shrivelled seeds** | -0.656* | -0.438 | 0.115 | -0.211 | -0.693* | -0.509 | -0.062 | 0.734** | -0.630* | -0.274 | 0.608* | 0.601* | 1 | |
| **SY** | 0.772** | 0.405 | -0.525 | 0.213 | 0.499 | 0.348 | 0.270 | -0.595* | 0.913** | 0.781** | -0.134 | -0.123 | -0.552 | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield

Table 19b: Correlation coefficients of growth and yield parameters under rainfed condition

| | PH | MSL | PB | SB | SMS | Total siliquae | 1000 SW | SL | BY | HI | Seeds/ siliqua | Developed seeds | Shrivelled seeds | SY |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **PH** | 1 | | | | | | | | | | | | | |
| **MSL** | 0.759** | 1 | | | | | | | | | | | | |
| **PB** | -0.359 | -0.499 | 1 | | | | | | | | | | | |
| **SB** | -0.048 | -0.538 | 0.462 | 1 | | | | | | | | | | |
| **SMS** | 0.658* | 0.570 | 0.082 | -0.216 | 1 | | | | | | | | | |
| **Total siliquae** | 0.311 | -0.127 | 0.462 | 0.626* | 0.400 | 1 | | | | | | | | |
| **1000 SW** | 0.440 | 0.515 | -0.622* | -0.062 | -0.178 | -0.133 | 1 | | | | | | | |
| **SL** | -0.525 | -0.057 | -0.469 | -0.475 | -0.699* | -0.849** | 0.236 | 1 | | | | | | |
| **BY** | 0.769** | 0.516 | -0.635* | 0.110 | 0.217 | 0.186 | 0.779** | -0.205 | 1 | | | | | |
| **HI** | 0.653* | 0.439 | -0.171 | 0.244 | 0.457 | 0.326 | 0.375 | -0.412 | 0.685* | 1 | | | | |
| **Seeds/siliqua** | -0.52 | -0.076 | 0.078 | -0.495 | -0.169 | -0.549 | -0.029 | 0.507 | -0.420 | -0.383 | 1 | | | |
| **Developed seeds** | -0.633* | -0.236 | 0.270 | -0.354 | -0.197 | -0.335 | -0.254 | 0.428 | -0.602* | -0.392 | 0.887** | 1 | | |
| **Shrivelled seeds** | -0.649* | -0.492 | -0.049 | -0.072 | -0.817** | -0.490 | 0.046 | 0.692* | -0.341 | -0.642* | 0.491 | 0.515 | 1 | |
| **SY** | 0.772** | 0.534 | -0.505 | 0.136 | 0.357 | 0.252 | 0.675* | -0.301 | 0.951** | 0.870** | -0.393 | -0.527 | -0.492 | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield

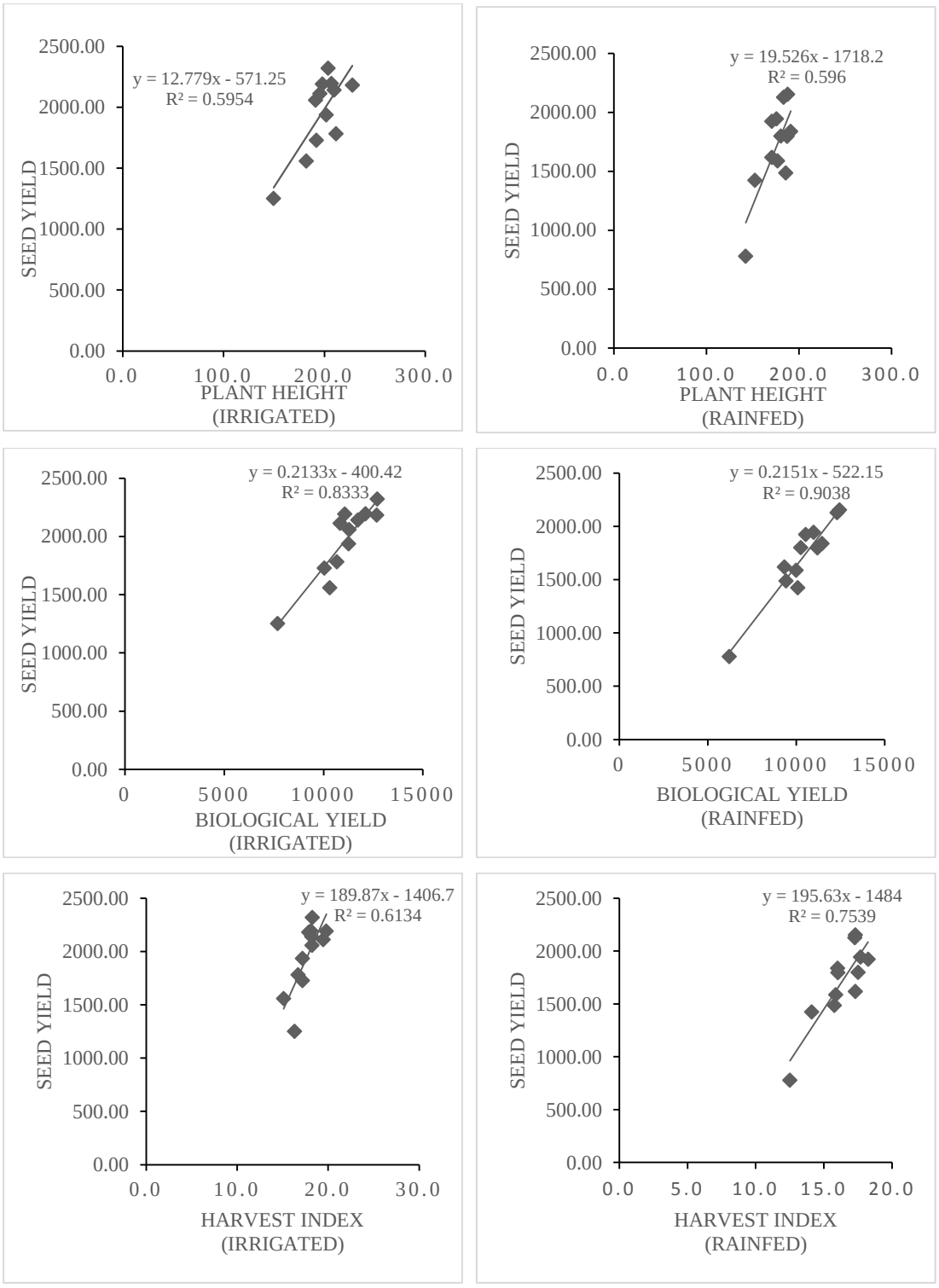



Fig. 16: Relationship of plant height, biomass and HI with seed yield in *B. juncea*

between biological yield and seed yield under irrigated and rainfed conditions in *B. juncea*. Our results are also in agreement with the correlation indices of growth and yield paramerers under irrigated and water stressed environments (Sabaghnia *et al* 2010). Seed size was positively and highly correlated with seed yield under drought stress in winter rapeseed cultivars (Moaveni *et al* 2010).

**Drought resistant parameters**

Susceptibility and tolerance indices alongwith the efficiency were calculated for growth parameters, yield components and seed yield.

**Drought susceptibility index**

Drought susceptibility index (DSI)≤0.5 for main shoot length (DSI= 0.43), primary branches (DSI= 0.07), 1000 seed weight (DSI= 0.38), seeds/siliqua (DSI= 0.22), biological yield (DSI= 0.47), seed yield (DSI= 0.48), harvest index (DSI= 0.41) and oil content (DSI= 0.09) which showed PBR 357 as highly tolerant genotype. This genotype however showed moderate tolerance for total siliquae (DSI= 0.95) and siliqua length (DSI= 0.63) and susceptible for plant height (DSI= 1.72), secondary branches (DSI= 1.79) and siliquae on main raceme (DSI= 1.37). Similarly, PBR 422 showed high tolerance for main shoot length (DSI= 0.36), primary branches (DSI= 0.35), total siliquae (DSI= 0.22), 1000 seed weight (DSI= 0.13), biological yield (DSI= 0.34) and moderate tolerance for plant height (DSI= 0.69), secondary branches (DSI= 0.89), siliquae on main raceme (DSI= 0.96), siliqua length (DSI= 0.63), seeds/siliqua (DSI= 0.85), seed yield (DSI= 0.56), harvest index (DSI= 0.65) and oil content (DSI= 0.99) and did not show susceptibility for any trait.

**Drought tolerance index**

Drought tolerance index (DTI) ≥ 1.0 for secondary branches (DTI= 1.08), siliquae on main raceme (DTI= 1.03), total siliquae (DTI= 1.12), 1000 seed weight (DTI= 1.14), seeds/siliqua (DTI= 1.06), seed yield (DTI= 1.06), harvest index (DTI= 1.13) and oil content (DTI= 1.00) showed RH 406 as highly tolerant genotype. However, this genotype showed moderate tolerance for plant height (DTI= 0.86), main shoot length (DTI= 0.95), primary branches (DTI= 0.79), siliqua length (DTI= 0.99) and biological yield (DTI= 0.94) and did not show susceptibility for any trait. Similarly, RH 1518 showed high tolerance for main shoot length (DTI= 1.08), 1000 seed weight (DTI= 1.26), siliqua length (DTI= 1.16), biological yield (DTI= 1.00), seed yield (DTI= 1.11), harvest index (DTI= 1.12), oil content (DTI= 1.00) and moderate tolerance for plant height (DTI= 0.89), primary branches (DTI= 0.93), secondary branches (DTI= 0.67), siliquae on main raceme (DTI= 0.64), total siliquae (DTI= 0.56) and seeds/siliqua (DTI= 0.93) and was not susceptible for any trait. Similarly MCN 09-40, showed high tolerance (DTI≥1.0) for plant height (DTI= 1.03), main shoot length (DTI= 1.14), primary branches (DTI= 1.17), secondary branches (DTI= 1.04), 1000 seed weight (DTI= 1.00), biological yield (DTI= 1.10), seed yield (DTI= 1.03) and showed moderate tolerance for siliquae on main raceme (DTI= 0.93), total siliquae (DTI= 0.76), siliqua length (DTI= 0.71), seeds/siliqua (DTI= 0.76), harvest index (DTI= 0.93) and oil content (DTI=0.95) and was not susceptible for any trait.

Table 20: Drought susceptibility index (DSI) of growth parameters in *B. juncea*

| Genotypes | Drought susceptibility indices | | | |
|---|---|---|---|---|
| | **Plant height** | **Main shoot length** | **Primary branches** | **Secondary branches** |
| RH 1518 | 1.01*±0.18 | 1.40*±0.41 | 1.98*±0.15 | 1.18*±0.12 |
| JC 210-335 | 0.43***±0.07 | 1.25*±0.22 | 0.90**±0.12 | 1.29*±0.14 |
| CJRD 1261 | 0.96**±0.35 | 1.92*±0.53 | 0.85**±0.24 | 0.77**±0.11 |
| RB-50 | 0.71**±0.03 | 0.38***±0.18 | 1.01*±0.15 | 0.86**±0.10 |
| RH 406 | 1.13*±0.14 | 0.10***±0.08 | 0.46***±0.11 | 0.56**±0.08 |
| PBR 422 | 0.69**±0.24 | 0.36***±0.17 | 0.35***±0.09 | 0.89**±0.07 |
| ELM 38 | 1.14*±0.08 | 0.77**±0.23 | 0.26***±0.04 | 0.08***±0.01 |
| CSR 1163 | 0.65**±0.27 | 2.10*±0.21 | 1.37*±0.21 | 0.71**±0.11 |
| IAN | 1.44*±0.62 | 1.67*±0.20 | 3.02*±0.36 | 0.34***±0.07 |
| MCN 09-40 | 0.79**±0.22 | 0.76**±0.29 | 1.27*±0.23 | 0.50***±0.09 |
| MLM 41-13-2 | 1.09*±0.22 | 1.07*±0.17 | 1.34*±0.27 | 2.91*±0.15 |
| PBR 357 | 1.72*±0.23 | 0.43***±0.26 | 0.07***±0.01 | 1.79*±0.13 |
| **Average** | **0.98±0.22** | **1.02±0.24** | **1.07±0.17** | **0.99±0.10** |

*Susceptible (≥1.0) **moderately tolerant (0.51-0.99) ***highly tolerant (≤0.5)

Table 21: Drought tolerance index (DTI) of growth parameters in *B. juncea*

| Genotypes | Drought tolerance indices | | | |
|---|---|---|---|---|
| | **Plant height** | **Main shoot length** | **Primary branches** | **Secondary branches** |
| RH 1518 | 0.89**±0.04 | 1.08***±0.06 | 0.93**±0.13 | 0.67**±0.01 |
| JC 210-335 | 0.55**±0.01 | 0.66**±0.06 | 0.84**±0.12 | 0.70**±0.08 |
| CJRD 1261 | 0.84**±0.02 | 0.57**±0.01 | 0.98**±0.03 | 1.09***±0.02 |
| RB-50 | 0.87**±0.03 | 1.14***±0.04 | 0.73**±0.02 | 0.40*±0.01 |
| RH 406 | 0.86**±0.01 | 0.95**±0.02 | 0.79**±0.05 | 1.08***±0.03 |
| PBR 422 | 0.98**±0.04 | 0.91**±0.06 | 1.05***±0.04 | 1.09***±0.09 |
| ELM 38 | 0.96**±0.07 | 0.95**±0.08 | 1.34***±0.13 | 1.35***±0.08 |
| CSR 1163 | 0.97**±0.05 | 1.11***±0.10 | 1.15***±0.15 | 1.22***±0.06 |
| IAN | 0.71**±0.03 | 0.52**±0.05 | 0.22*±0.04 | 0.77**±0.04 |
| MCN 09-40 | 1.03***±0.04 | 1.14***±0.06 | 1.17***±0.06 | 1.04***±0.13 |
| MLM 41-13-2 | 1.01***±0.03 | 1.18***±0.09 | 1.21***±0.04 | 1.13***±0.08 |
| PBR 357 | 1.07***±0.03 | 1.15***±0.10 | 1.09***±0.07 | 0.66**±0.01 |
| **Average** | **0.89±0.03** | **0.95±0.06** | **0.96±0.07** | **0.93±0.05** |

***Highly tolerant (≥1.0) **moderately tolerant (0.51-0.99) *susceptible (≤0.5)

Table 22: Drought susceptibility index (DSI) of yield attributes of *B. juncea*

| Genotypes | Drought susceptibility indices | | | | |
|---|---|---|---|---|---|
| | **Siliquae on main raceme** | **Total siliquae** | **1000 seed weight** | **Siliqua length** | **Seeds/siliqua** |
| RH 1518 | 0.14***±0.09 | 0.80**±0.38 | 1.82*±0.25 | 0.99**±0.15 | 0.99**±0.24 |
| JC 210-335 | 1.83*±0.49 | 0.44***±0.19 | 1.30*±0.29 | 1.74*±0.18 | 0.62**±0.21 |
| CJRD 1261 | 0.93**±0.12 | 0.54**±0.14 | 1.61*±0.34 | 0.50***±0.11 | 1.18*±0.32 |
| RB-50 | 1.12*±0.45 | 0.99**±0.16 | 0.61**±0.16 | 0.40***±0.08 | 0.79**±0.25 |
| RH 406 | 0.43***±0.13 | 1.16*±0.21 | 1.19*±0.15 | 1.26*±0.12 | 1.91*±0.21 |
| PBR 422 | 0.96**±0.34 | 0.22***±0.07 | 0.13***±0.27 | 0.63**±0.13 | 0.85**±0.14 |
| ELM 38 | 0.66**±0.37 | 1.20*±0.38 | 0.14***±0.15 | 1.22*±0.10 | 1.88*±0.22 |
| CSR 1163 | 1.46*±0.57 | 1.20*±0.36 | 1.91*±0.03 | 1.47*±0.14 | 1.91*±0.17 |
| IAN | 1.02*±0.28 | 0.77**±0.24 | 0.92**±0.13 | 1.06*±0.13 | 0.66**±0.12 |
| MCN 09-40 | 0.97**±0.21 | 1.53*±0.25 | 0.68**±0.12 | 1.16*±0.12 | 0.18***±0.06 |
| MLM 41-13-2 | 0.72**±0.32 | 1.53*±0.19 | 0.14***±0.04 | 0.53**±0.97 | 0.34***±0.08 |
| PBR 357 | 1.37*±0.22 | 0.95**±0.16 | 0.38***±0.09 | 0.63**±0.09 | 0.22***±0.05 |
| **Average** | **0.97±0.29** | **0.94±0.23** | **0.90±0.17** | **0.97±0.19** | **0.96±0.17** |

*Susceptible (≥1.0) **moderately tolerant (0.51-0.99) ***highly tolerant (≤0.5)

Table 23: Drought tolerance index (DTI) of yield attributes of *B. juncea*

| Genotypes | Drought tolerance indices | | | | |
|---|---|---|---|---|---|
| | **Siliquae on main raceme** | **Total siliquae** | **1000 seed weight** | **Siliqua length** | **Seeds/siliqua** |
| RH 1518 | 0.64**±0.10 | 0.56**±0.03 | 1.26***±0.16 | 1.16***±0.04 | 0.93**±0.08 |
| JC 210-335 | 1.27***±0.03 | 0.38*±0.03 | 0.43*±0.03 | 1.29***±0.02 | 1.28***±0.03 |
| CJRD 1261 | 1.27***±0.06 | 1.37***±0.14 | 0.49*±0.05 | 0.54**±0.01 | 0.72**±0.07 |
| RB-50 | 0.68**±0.08 | 0.37*±0.05 | 0.75**±0.09 | 1.53***±0.16 | 0.86**±0.03 |
| RH 406 | 1.03***±0.12 | 1.12***±0.15 | 1.14***±0.30 | 0.99**±0.04 | 1.06***±0.08 |
| PBR 422 | 0.76**±0.10 | 0.69**±0.13 | 0.77**±0.18 | 0.77**±0.01 | 0.89**±0.07 |
| ELM 38 | 0.92**±0.02 | 1.08***±0.05 | 0.50*±0.11 | 0.64**±0.05 | 0.89**±0.05 |
| CSR 1163 | 0.81**±0.19 | 0.92**±0.08 | 0.97**±0.14 | 0.72**±0.10 | 0.53**±0.05 |
| IAN | 0.84**±0.01 | 1.37***±0.11 | 1.00***±0.20 | 1.40***±0.06 | 0.82**±0.06 |
| MCN 09-40 | 0.93**±0.14 | 0.76**±0.06 | 1.00***±0.15 | 0.71**±0.04 | 0.76**±0.07 |
| MLM 41-13-2 | 1.06***±0.10 | 1.05***±0.08 | 0.59**±0.05 | 0.72**±0.06 | 0.78**±0.04 |
| PBR 357 | 0.66**±0.05 | 0.66**±0.01 | 0.97**±0.21 | 1.01***±0.05 | 0.89**±0.05 |
| **Average** | **0.91±0.09** | **0.86±0.08** | **0.82±0.14** | **0.96±0.05** | **0.87±0.06** |

***Highly tolerant (≥1.0) **moderately tolerant (0.51-0.99) *susceptible (≤0.5)

Table 24: Drought susceptibility index for biomass, HI, oil content and seed yield

| Genotypes | Drought susceptibility indices | | | |
|---|---|---|---|---|
| | **Biomass** | **Harvest index** | **Oil content** | **Seed yield** |
| RH 1518 | 0.13***±0.01 | 1.33*±0.05 | 1.17*±0.25 | 0.89**±0.07 |
| JC 210-335 | 3.08*±0.15 | 2.91*±0.12 | 0.36***±0.08 | 2.97*±0.09 |
| CJRD 1261 | 2.79*±0.18 | 0.63**±0.11 | 1.11*±0.23 | 1.69*±0.11 |
| RB-50 | 0.07***±0.01 | 0.96**±0.17 | 0.52**±0.15 | 0.64**±0.17 |
| RH 406 | 0.46***±0.10 | 0.78**±0.07 | 1.03*±0.25 | 0.70**±0.06 |
| PBR 422 | 0.34***±0.01 | 0.65**±0.05 | 0.99**±0.18 | 0.56**±0.09 |
| ELM 38 | 2.47*±0.03 | 0.40***±0.09 | 1.52*±0.24 | 1.42*±0.14 |
| CSR 1163 | 0.10***±0.01 | 0.83**±0.03 | 0.62**±0.16 | 0.57**±0.09 |
| IAN | 0.35***±0.01 | 0.83**±0.06 | 1.01*±0.13 | 0.68**±0.02 |
| MCN 09-40 | 0.37***±0.04 | 1.50*±0.12 | 0.57**±0.09 | 1.11*±0.12 |
| MLM 41-13-2 | 1.88*±0.05 | 0.69**±0.11 | 0.40***±0.06 | 1.30*±0.13 |
| PBR 357 | 0.47***±0.16 | 0.41***±0.05 | 0.09***±0.01 | 0.20***±0.10 |
| **Average** | **1.04±0.06** | **1.00±0.09** | **0.78±0.15** | **1.00±0.10** |

*Susceptible (≥1.0) **moderately tolerant (0.51-0.99) ***highly tolerant (≤0.5)

Table 25: Drought tolerance index for biomass, HI, oil content and seed yield

| Genotypes/ treatment | Drought tolerance indices | | | |
|---|---|---|---|---|
| | **Biomass** | **Harvest index** | **Oil content** | **Seed yield** |
| RH 1518 | 1.00***±0.01 | 1.12***±0.01 | 1.00***±0.01 | 1.11***±0.02 |
| JC 210-335 | 0.39*±0.01 | 0.65**±0.02 | 1.06***±0.01 | 0.25*±0.01 |
| CJRD 1261 | 0.87**±0.01 | 1.01***±0.02 | 1.00***±0.02 | 0.87**±0.01 |
| RB-50 | 0.82**±0.01 | 0.87**±0.01 | 0.98**±0.01 | 0.72**±0.01 |
| RH 406 | 0.94**±0.01 | 1.13***±0.02 | 1.00***±0.02 | 1.06***±0.01 |
| PBR 422 | 1.30***±0.02 | 1.01***±0.03 | 1.03***±0.01 | 1.31***±0.03 |
| ELM 38 | 1.02***±0.01 | 1.01***±0.02 | 0.94**±0.01 | 1.03***±0.02 |
| CSR 1163 | 1.04***±0.02 | 0.88**±0.02 | 0.99**±0.01 | 0.91**±0.01 |
| IAN | 0.85**±0.01 | 0.68**±0.01 | 0.96**±0.01 | 0.58**±0.01 |
| MCN 09-40 | 1.10***±0.01 | 0.93**±0.01 | 0.95**±0.01 | 1.03***±0.02 |
| MLM 41-13-2 | 0.83**±0.01 | 0.84**±0.01 | 0.94**±0.02 | 0.69**±0.01 |
| PBR 357 | 1.28***±0.02 | 0.98**±0.01 | 0.98**±0.01 | 1.21***±0.01 |
| **Average** | **0.95±0.01** | **0.93±0.02** | **0.99±0.01** | **0.87±0.02** |

***Highly tolerant (≥1.0) **moderately tolerant (0.51-0.99) *susceptible (≤0.5)

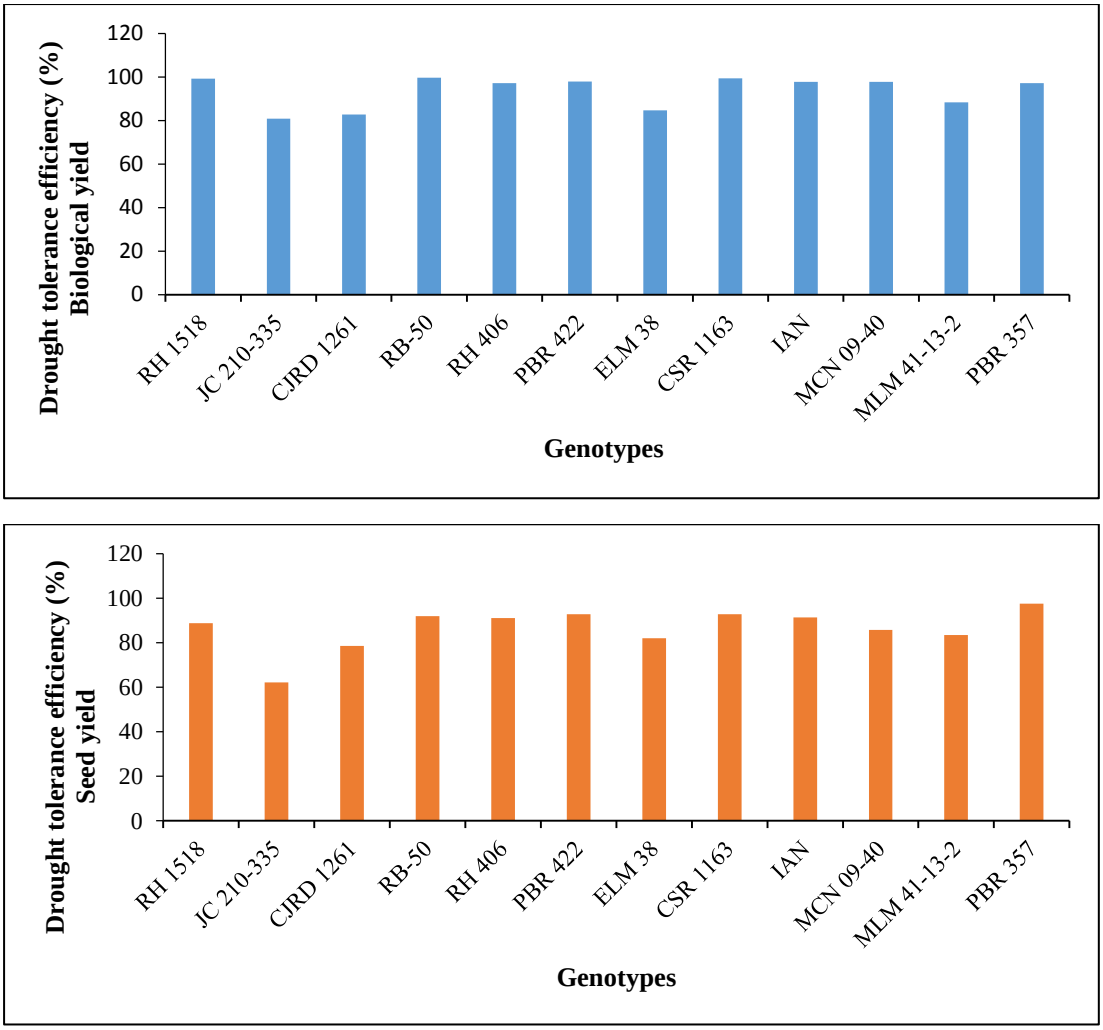



Fig. 17 Drought tolerance efficiency (%) of biomass and seed yield in *B. juncea*

**Drought tolerance efficiency**

Drought tolerance efficiency (DTE) of biological yield was greater in RB-50 (99.6%) and comparable in CSR 1163 (99.4%) while it was lower in JC 210-335 (80.9%). DTE of seed yield was greater in PBR 357 (97.5%), followed by CSR 1163 and PBR 422 (92.8%) and lower in JC 210-335 (62.2%).

**Correlation between drought resistant parameters**

Correlation coefficicent of drought susceptibility indices (DSI) of growth parameters, yield components and seed yield with drought tolerance efficiency are tabulated in Table 27. DSI of plant height was negatively correlated with harvest index (r= -0.603*). DSI of main shoot length was positively correlated to siliquae on main raceme (r= 0.592*) and 1000 seed weight (r= 0.656*). A negative correlation between drought susceptibility indices existed for primary branches (r=-0.645*), total siliquae (r= -0.629*) with oil content. DSI of siliqua length and harvest index (r= 0.593*), seeds per siliqua and oil content (r= 0.635*) were positively associated with each other. DSI of seed yield had highly significant positive

correlation with biological yield (r= 0857**) and harvest index (r= 0.725**). A highly significant and negative correlation was observed for DSI of biological yield (r= -0.857**), harvest index (r= -0.726**) and seed yield (r= -0.999**) with drought tolerance efficiency (Table 27).

Correlation coefficient of drought tolerance indices (DTI) of growth parameters, yield components and seed yield with drought tolerance efficiency are depicted in Table 28. DTI of plant height was positively correlated with DTI of main shoot length (r= 0.757**), siliquae on main raceme (r= 0.662*), biological yield (r= 0.839**) and seed yield (r= 0.781**) and also with drought tolerance efficiency (r= 0.650*). However, it was negatively correlated with DTI of seeds/siliqua (r= -0.590*). DTI of main shoot length and siliquae on main raceme (r= 0.580*), secondary branches and total siliquae (r= 0.603*) were positively associated with each other. DTI of primary branches was negatively correlated with 1000 seed weight (r= -0.631*) and biological yield (r= -0.638*) and with drought tolerance efficiency (r= -0.806**) too. Siliqua length had highly significant and negative correlation with siliquae on main raceme (r= -0.764**) and total siliquae (r= -0.873**). DTI of 1000 seed weight (r= 0.684*), biological yield (r= 0.791**) and seed yield (r= 0.679*) were positively correlated with drought tolerance efficiency. Seed yield was strongly and positively correlated with biological yield (r= 0.935**) and harvest index (r=0.847**) (Table 28).

Correlation coefficients of drought susceptibility indices (DSI) of growth parameters, yield components and seed yield are illustrated in Table 29. DSI of harvest index was negatively correlated with plant height (r= -0.603*) whereas positively correlated with siliqua length (r= 0.593*). DSI of main shoot length was positively associated with siliquae on main raceme (r= 0.592*) and 1000 seed weight (r= 0.656*). DSI of oil content was negatively correlated with primary branches (r= -0.645*) and total siliquae (r= -0.629*). DSI of number of seeds per siliqua was positively associated with oil content (r= 0.635*). DSI of biomass (r= 0.857**) and harvest index (r= 0.725**) were highly significantly and positively correlated with that of seed yield.

Correlation coefficients of drought tolerance indices (DTI) of growth parameters, yield components are tabulated in Table 30. DTI of plant height was significantly and positively correlated with main shoot length (r= 0.757**), siliquae on main raceme (r= 0.662*), biological yield (r= 0.839**) and seed yield (r= 0.781**). However, plant height and seeds/siliqua (r= -0.590*) were negatively correlated with each other. DTI of main shoot length and siliquae on main shoot (r= 0.580*), secondary branches and total siliquae (r= 0.603*) were positively associated with each other. DTI of primary branches was negatively correlated with 1000 seed weight (r= -0.631*) and biological yield (r= -0.638*). DTI of siliqua length was highly significantly and negatively correlated with that of siliquae

Table 27: Correlations between drought susceptibility indices with drought tolerance efficiency in *B. juncea*

| DSI | Drought susceptibility indices | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **PH** | **MSL** | **PB** | **SB** | **SMS** | **Total siliquae** | **1000 SW** | **SL** | **Seeds/siliqua** | **BY** | **HI** | **Oil content** | **SY** | **DTE** |
| **PH** | 1 | | | | | | | | | | | | | |
| **MSL** | -0.148 | 1 | | | | | | | | | | | | |
| **PB** | -0.295 | 0.194 | 1 | | | | | | | | | | | |
| **SB** | 0.212 | -0.105 | -0.245 | 1 | | | | | | | | | | |
| **SMS** | 0.055 | 0.592* | -0.129 | 0.062 | 1 | | | | | | | | | |
| **Total siliquae** | 0.130 | -0.047 | 0.072 | 0.157 | -0.070 | 1 | | | | | | | | |
| **1000 SW** | -0.282 | 0.656* | 0.022 | -0.178 | 0.375 | -0.227 | 1 | | | | | | | |
| **SL** | -0.318 | 0.242 | 0.266 | 0.057 | 0.123 | -0.370 | 0.417 | 1 | | | | | | |
| **Seeds/siliqua** | -0.146 | 0.140 | -0.261 | 0.021 | -0.187 | -0.533 | 0.376 | 0.367 | 1 | | | | | |
| **BY** | -0.172 | 0.244 | 0.213 | -0.177 | -0.264 | 0.145 | -0.044 | 0.161 | 0.050 | 1 | | | | |
| **HI** | -0.603* | 0.151 | 0.415 | -0.231 | 0.186 | 0.043 | 0.349 | 0.593* | -0.271 | 0.289 | 1 | | | |
| **Oil content** | 0.029 | 0.126 | -0.645* | -0.175 | 0.109 | -0.629* | 0.128 | 0.085 | 0.635* | 0.112 | -0.303 | 1 | | |
| **SY** | -0.494 | 0.284 | 0.303 | -0.220 | -0.039 | 0.076 | 0.176 | 0.430 | -0.059 | 0.857** | 0.725** | 0.011 | 1 | |
| **DTE** | 0.495 | -0.283 | -0.302 | 0.220 | 0.038 | -0.077 | -0.174 | -0.430 | 0.061 | -0.857** | -0.726** | -0.010 | -0.999** | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield, DTE- Drought tolerance efficiency

Table 28: Correlations between drought tolerance indices with drought tolerance efficiency in *B. juncea*

| DTI | Drought tolerance indices | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **PH** | **MSL** | **PB** | **SB** | **SMS** | **Total siliquae** | **1000 SW** | **SL** | **Seeds/ siliqua** | **BY** | **HI** | **Oil content** | **SY** | **DTE** |
| **PH** | 1 | | | | | | | | | | | | | |
| **MSL** | 0.757** | 1 | | | | | | | | | | | | |
| **PB** | -0.487 | -0.547 | 1 | | | | | | | | | | | |
| **SB** | 0.046 | -0.447 | 0.360 | 1 | | | | | | | | | | |
| **SMS** | 0.662* | 0.580* | 0.052 | -0.135 | 1 | | | | | | | | | |
| **Total siliquae** | 0.355 | -0.009 | 0.363 | 0.603* | 0.565 | 1 | | | | | | | | |
| **1000 SW** | 0.266 | 0.337 | -0.631* | -0.046 | -0.272 | -0.218 | 1 | | | | | | | |
| **SL** | -0.563 | -0.135 | -0.320 | -0.455 | -0.764** | -0.873** | 0.245 | 1 | | | | | | |
| **Seeds/siliqua** | -0.590* | -0.266 | 0.265 | -0.419 | -0.244 | -0.375 | -0.155 | 0.472 | 1 | | | | | |
| **BY** | 0.839** | 0.455 | -0.638* | 0.088 | 0.372 | 0.269 | 0.459 | -0.423 | -0.479 | 1 | | | | |
| **HI** | 0.566 | 0.377 | -0.276 | 0.080 | 0.420 | 0.275 | 0.395 | -0.426 | -0.115 | 0.619* | 1 | | | |
| **Oil content** | -0.550 | -0.368 | 0.222 | -0.442 | -0.200 | -0.281 | -0.099 | 0.248 | 0.546 | -0.294 | -0.063 | 1 | | |
| **SY** | 0.781** | 0.456 | -0.543 | 0.040 | 0.441 | 0.294 | 0.468 | -0.459 | -0.306 | 0.935** | 0.847** | -0.159 | 1 | |
| **DTE** | 0.650* | 0.467 | -0.806** | 0.100 | -0.077 | -0.065 | 0.684* | 0.038 | -0.509 | 0.791** | 0.425 | -0.367 | 0.679* | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield, DTE- Drought tolerance efficiency

Table 29: Correlations between drought susceptibility indices in *B. juncea*

| DSI | Drought susceptibility indices | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **PH** | **MSL** | **PB** | **SB** | **SMS** | **Total siliquae** | **1000 SW** | **SL** | **Seeds/ siliqua** | **BY** | **HI** | **Oil content** | **SY** |
| **PH** | 1 | | | | | | | | | | | | |
| **MSL** | -0.148 | 1 | | | | | | | | | | | |
| **PB** | -0.295 | 0.194 | 1 | | | | | | | | | | |
| **SB** | 0.212 | -0.105 | -0.245 | 1 | | | | | | | | | |
| **SMS** | 0.055 | 0.592* | -0.129 | 0.062 | 1 | | | | | | | | |
| **Total siliquae** | 0.13 | -0.047 | 0.072 | 0.157 | -0.070 | 1 | | | | | | | |
| **1000 SW** | -0.282 | 0.656* | 0.022 | -0.178 | 0.375 | -0.227 | 1 | | | | | | |
| **SL** | -0.318 | 0.242 | 0.266 | 0.057 | 0.123 | -0.370 | 0.417 | 1 | | | | | |
| **Seeds/siliqua** | -0.146 | 0.14 | -0.261 | 0.021 | -0.187 | -0.533 | 0.376 | 0.367 | 1 | | | | |
| **BY** | -0.172 | 0.244 | 0.213 | -0.177 | -0.264 | 0.145 | -0.044 | 0.161 | 0.05 | 1 | | | |
| **HI** | -0.603* | 0.151 | 0.415 | -0.231 | 0.186 | 0.043 | 0.349 | 0.593* | -0.271 | 0.289 | 1 | | |
| **Oil content** | 0.029 | 0.126 | -0.645* | -0.175 | 0.109 | -0.629* | 0.128 | 0.085 | 0.635* | 0.112 | -0.303 | 1 | |
| **SY** | -0.494 | 0.284 | 0.303 | -0.220 | -0.039 | 0.076 | 0.176 | 0.430 | -0.059 | 0.857** | 0.725** | 0.010 | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield

Table 30: Correlations between drought tolerance indices in *B. juncea*

| DTI | Drought tolerance indices | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **PH** | **MSL** | **PB** | **SB** | **SMS** | **Total siliquae** | **1000 SW** | **SL** | **Seeds/ siliqua** | **BY** | **HI** | **Oil content** | **SY** |
| **PH** | 1 | | | | | | | | | | | | |
| **MSL** | 0.757** | 1 | | | | | | | | | | | |
| **PB** | -0.487 | -0.547 | 1 | | | | | | | | | | |
| **SB** | 0.046 | -0.447 | 0.360 | 1 | | | | | | | | | |
| **SMS** | 0.662* | 0.580* | 0.052 | -0.135 | 1 | | | | | | | | |
| **Total siliquae** | 0.355 | -0.009 | 0.363 | 0.603* | 0.565 | 1 | | | | | | | |
| **1000 SW** | 0.266 | 0.337 | -0.631* | -0.046 | -0.272 | -0.218 | 1 | | | | | | |
| **SL** | -0.563 | -0.135 | -0.320 | -0.455 | -0.764** | -0.873** | 0.245 | 1 | | | | | |
| **Seeds/siliqua** | -0.590* | -0.266 | 0.265 | -0.419 | -0.244 | -0.375 | -0.155 | 0.472 | 1 | | | | |
| **BY** | 0.839** | 0.455 | -0.638* | 0.088 | 0.372 | 0.269 | 0.459 | -0.423 | -0.479 | 1 | | | |
| **HI** | 0.566 | 0.377 | -0.276 | 0.080 | 0.420 | 0.275 | 0.395 | -0.426 | -0.115 | 0.619* | 1 | | |
| **Oil content** | -0.550 | -0.368 | 0.222 | -0.442 | -0.200 | -0.281 | -0.099 | 0.248 | 0.546 | -0.294 | -0.063 | 1 | |
| **SY** | 0.781** | 0.456 | -0.543 | 0.040 | 0.441 | 0.294 | 0.468 | -0.459 | -0.306 | 0.935** | 0.847** | -0.159 | 1 |

*Significant at 5%, **Significant at 1%

PH- Plant height, MSL- Main shoot length, PB- Primary branches, SB- Secondary branches, SMS- Siliquae on main raceme, SW- Seed weight, SL Siliqua length, BY- Biological yield, HI- Harvest index, SY- Seed yield

on main raceme (r= -0.764**) and total siliquae (r= -0.873**). DTI of biological yield and harvest index (r= 0.619*) were positively associated with each other. DTI of biological yield (r= 0.935**) and harvest index (r= 0.847**) had highly significant and positive association with DTI of seed yield.

These results are in agreement with the findings of Kaur (2012) in which DSI of biomass and seed yield were negatively associated with DTE under one and two irrigations in *B. juncea* and *B. napus*. Stress tolerance index and stress susceptibility index had highly negative correlation under each saline level in *B. juncea* as observed by Kannu Priya (2019). Heat resistant parameters were associated with growth traits, heat tolerance efficiency by Sharma and Sardana (2013) and correlated with seed yield in Indian mustard. DSI for seed yield and other related traits were evaluated by Chauhan *et al* (2007) to find the relative tolerance of Indian mustard genotypes under watered and drought conditions.

**SDS PAGE-Protein profiling**

Protein profile of genotypes (1-9) are represented in plate 1 under rainfed and in plate 2 under irrigated condition while plate 3 represents comparative banding pattern of genotypes (10-12) under the two irrigation modules. Protein bands represented a molecular weight ranging from 15 kDa to 75 kDa under rainfed and irrigated situations however all genotypes showed a distinct band at 150 kDa with irrigation. The band density was lower under stressed module as compared to irrigated condition representing the lower protein content due to dehydration under stress condition. It is inferred that genotypes under rainfed condition were closely similar to each other with similar banding pattern except PBR 422 where banding pattern was similar to that under irrigated condition and this genotype showed a distinct band at 150 kDa. Yielding ability of PBR 422 was higher even under stress/rainfed. Previous studies of Indian mustard revealed that pro-cruciferin protein ranged from 54 to 71 kDa, cruciferin protein with alpha and beta structures ranged from 18.1 kDa and 31.2 kDa and napin protein ranged between 15 and 16 kDa in *B. juncea* (Mawlong *et al* 2017). The different banding pattern in the studied genotypes provided accurate and reliable results and hence it is useful technique for varietal identification (Chaudhary 2014). Under drought stress, several genes are induced which further synthesise the proteins like late embryogenesis abundant proteins (LEA), heat shock proteins (HSPs), lipid transfer proteins (LTPs) including protein phosphatases and protein kinases (Qazi *et al* 2019). Non-significant difference in the banding pattern of leaf proteins between different species of Brassica has been reported by Mukhlesur and Hirata (2004).

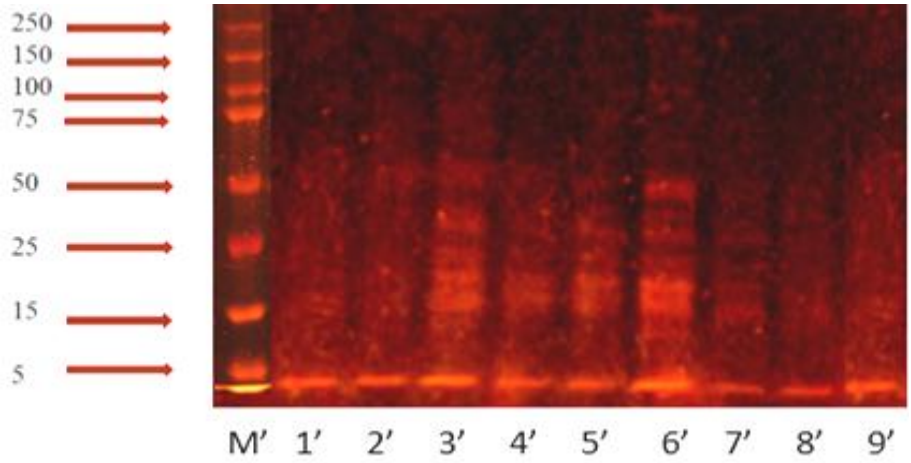



Plate 1: Banding pattern of *B. juncea* genotypes under rainfed condition
M': Marker, 1': RH 1518, 2': JC 210-335, 3': CJRD 1261, 4': RB-50, 5': RH 406, 6': PBR 422, 7': ELM 38, 8': CSR 1163, 9': IAN

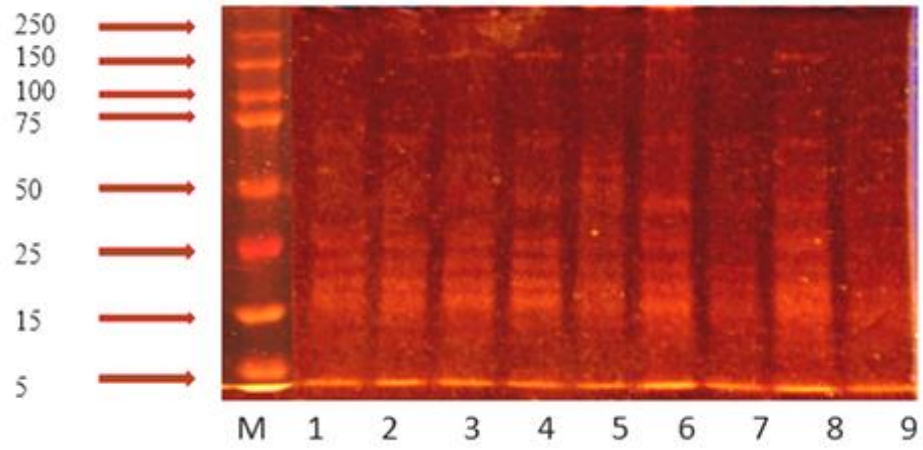



Plate 2: Banding pattern of *B. juncea* genotypes under irrigated condition
M: Marker, 1: RH 1518, 2: JC 210-335, 3: CJRD 1261, 4: RB-50, 5: RH 406, 6: PBR 422, 7: ELM 38, 8: CSR 1163, 9: IAN

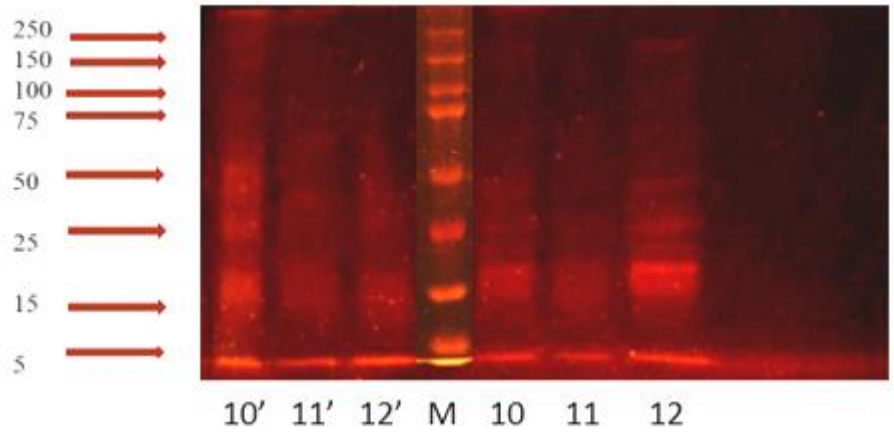



Plate 3: Comparison of *B. juncea* genotypes under rainfed (on the left) and irrigated (on the right) conditions
M: Marker
Rainfed- 10': MCN 09-40, 11': MLM 41-13-2, 12': PBR 357
Irrigated- 10: MCN 09-40, 11: MLM 41-13-2, 12: PBR 357

## CHAPTER-V

## SUMMARY

Rapeseed mustard is second most important oilseed crop of India after groundnut. More than 90% area under oilseed Brassica is occupied by the Indian mustard (*Brassica juncea*) due to its relative tolerance to abiotic and biotic stresses as compared to other oilseed Brassica species. Mustard seeds have 25-45% oil content and is used widely worldover for its tempting flavor and preservative value and also as moderating food. The mustard crop is grown mainly under the arid and semi-arid regions in India under conserved moisture. Due to low rainfall and increase in demand for other activities, irrigation water is becoming scarce. Oilseed Brassicas possess high sensitivity towards water depletion. In the present scenario existing water resources are fully exploited in Punjab and with the introduction of new varieties alongwith the climate change. The present investigation "Differential response of Indian mustard under rainfed and irrigated conditions (*Brassica juncea* L. Czern & Coss) genotypes" was planned and executed to compare the tolerance of different Indian mustard genotypes under moisture stress (RF) and irrigated (IR) modules. Field experiment was conducted at the research farm of oilseeds section and biochemical estimations were carried out in laboratories of Oilseeds, Department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana. Experiment was conducted in randomized block design with three replications. Twelve genotypes of *Brassica juncea* (JC 210-335, CJRD 1261, RB 50, RH 406, PBR 422, ELM 38, CSR 1163, IAN, MCN 09-40, MLM 41-13-2, PBR 357) were sown on 10$^{th}$ November, 2017 under two irrigation modules i) rainfed (only pre-sowing irrigation) and ii) irrigated (with 2 irrigations at 35 and 65 days after sowing) The salient findings of the study are summarized below:

- Flowering and siliquing behaviour suffered significantly under rainfed condition and genotypes registered variations in phenology. Flowering initiation and 50% flowering was early by 1.6 and 2.1 days respectively and flowering completion by 5 days under rainfed/moisture stress. Flowering duration declined by 3 days under rainfed over irrigated conditions. JC 210-335 took minimum days (45.0) for flower initiation, 50% flowering (58.0) and flowering completion (67.7) while flowering duration was of 16.0 days in RH 1518 under rainfed condition. Siliquing behaviour followed similar trend in the studied genotypes. Earliness was recorded for siliqua initiation (1.2 days), 50% siliquing (5.3 days), siliqua completion (4.6 days) and siliquing duration was shorter by 3.5 days under moisture stress over irrigated module. Differences existed for siliquing behaviour under moisture stress and again JC 210-335 took 53.3 days for siliqua initiation, 74.3 days for 50% siliquing and 104.3 days for its completion, siliquing

duration was 42.7 days for RH 406. Moisture stress reduced reproductive phase by 3.1 days and maturity by 2.9 days over irrigated conditions.

- Greenness of the leaves indicated by SPAD values decreased by 5.7% with minimal decline of 2.9% in RH 1518. Decline was also recorded for photosynthetic pigments along with carotenoids. The minimum reduction in chla was 3.9% in RH 406, chlb to 10.5% in CJRD 1261, total chlorophyll to 11.3% and carotenoids to 3.9% in RB-50.
- Canopy temperature (CT) significantly increased by 1.5°C under rainfed over irrigated conditions with minimum canopy temperature of 24.6°C in RH 1518. Canopy air temperature differential (CATD) was 1.5°C less under stressed condition with maximum of -3.0°C in PBR 422.
- RWC was reduced by 9% while RSD and WSD increased by 21.6% and 24.4% respectively due to moisture stress. Maximum RWC in MLM 41-13-2 (81.2%), while maximum RSD (31.7%) in CSR 1163 and WSD (35.6%) in ELM 38 and CSR 1163 under rainfed condition.
- Leaf traits i.e., length, width and leaf area of all genotypes were reduced by 11.4%, 8.9% and 13.7% respectively under rainfed condition. Minimum reduction of leaf length was in JC 210-335 (1.1%), width in RB-50 (1.2%) and leaf area in MLM 41-13-2 (1.5%). Moisture stress reduced SLW (8.0), SLA (5.6) and LWR (11.4) over irrigated module and minimum reduction for these traits was in MLM 41-13-2. Number of leaves per plant were significantly reduced to 13.9% but CSR 1163 had maximum leaves (43.6) under stressed condition.
- Membrane stability decreased while membrane injury increased significantly under moisture stress as compared to irrigated/non-stressed condition. Average membrane stability decreased by 25.3% while membrane injury increased by 10.3% under stressed condition. Moisture stress disrupted membrane to 24.6% and inflicted injury 12.2% in RH 1518 whereas reverse was in RH 406. Membrane stability was higher in RH 1518 and lesser in RH 406 whereas membrane injury followed a reverse trend in these genotypes. PBR 422 maintained comparable membrane stability under stressed and irrigated conditions and suffered a slight higher injury under rainfed condition.
- Under drought stress, cellular turgidity was maintained by increased total soluble sugars (TSS), reducing sugars (RS) and non-reducing sugars (NRS) which significantly increased under stressed condition over irrigated condition. Average increase was 29.3% in TSS, 53.8% in RS and 20.2% in NRS respectively. Under rainfed condition, ELM 38 had maximum TSS (90.1 mg/g DW), JC 210-335 reducing sugars (39.0 mg/g DW) and MLM 41-13-2 non-reducing sugars (65.3 mg/g DW) over irrigated module.

- Antioxidative enzymes increased significantly under rainfed condition. Maximum catalase activity was in JC 210-335 (358.9 mmol/min/g FW), SOD in MCN 09-40 (147.7 EA/min/g FW), peroxidase (POD) in RB-50 and RH 406 (1.18 mmol/min/g FW respectively) under rainfed condition.
- Proline, osmoprotectant increased under stress with highest content of 45.7 mg/g FW in MCN 09-40 under rainfed over irrigated condition. Moisture stress damaged the membranes as lipid peroxidation product (malondialdehyde) increased, maximum being in RB-50 (21.9 µmoles/g FW). Total soluble proteins decreased by 17.6% and minimum decline was 5.7% in JC 210-335 over irrigated conditions.
- Growth parameters, yield components and seed yield suffered significantly under moisture stress. However, all the studied parameters showed significant (I×G) interactions except main shoot length, primary branches, secondary branches, siliquae on main shoot, siliqua length and oil content. Plant height was reduced by 11.3%, main shoot length by 6.7%, primary branches by 11.5% and secondary branches by 17.5% under rainfed with respect to irrigated condition. Moisture stress affected the growth parameters to variable extent with minimal decline of plant height in JC 210-335 (4.8%), main shoot length (0.7%) in RH 406, primary branches in RH 1518 (1.6%) and secondary branches in PBR-422 (3.8%).
- Moisture stress significantly decreased number of siliquae on main shoot (SMS) by 7.1%, total siliquae/ plant by 8.7%, 1000 seed weight by 21.2% and siliqua length by 7.8% over irrigated condition. Significant effect of stress was witnessed on seed filling as total seeds/siliqua were reduced by 14.2% and developed seeds by 15.7% whereas shrivelled seeds enhanced by 50% over irrigated condition. Biomass was reduced by 6.2%, seed yield by 12.7%, HI by 7.9% and oil content by 1.5% over irrigated conditions. Least reduction in biomass was in RB-50 (0.4%) followed by CSR 1163 (0.6%), seed yield in PBR 357 (2.5%), HI in ELM 38 (3.2%) and oil content again in PBR 357 (0.2%).
- Initiation of flowering (r= 0.781*), flowering completion (0.678*) and siliqua initiation (0.784**), leaf width (r= 0.591*) and leaf water retention (r= 0.638*), plant height (r= 0.772**), 1000 seed weight (r= 0.675*), biological yield (r= 0.951**) and HI (r= 0.870**) had significant positive correlation with seed yield under rainfed condition.
- Seed yield had strong positive relationship with initiation of flowering ($R^2$= 0.611) and initiation of siliquing ($R^2$= 0.614), plant height ($R^2$= 0.596), biological yield ($R^2$= 0.903) and HI ($R^2$= 0.753). Physiological traits like membrane injury ($R^2$= 0.269), leaf width ($R^2$= 0.349), SLA ($R^2$= 0.284), LWR ($R^2$= 0.407), osmoprotectants like proline ($R^2$= 0.142), reducing sugars ($R^2$= 0.253) had weak positive relationship with seed yield.
- Correlation analysis of DSI and DTI of growth, seed yield and yield components revealed

that DSI and DTI of biological yield and harvest index showed strong positive association with seed yield. Moreover, DSI of biomass (r= -0.857**) and seed yield (r= -0.999**) were highly negatively correlated with DTE while DTI of respective parameters had strong positive correlation (r= 0.791** and r= 0.679*) with DTE.

- SDS-PAGE represented protein bands ranging 15kDa to 75kDa under stressed/rainfed and non-stressed/irrigated conditions. However, the band density was lower indicating lesser protein under rainfed as compared to irrigated condition. PBR 422 showed similar banding pattern under both the irrigation modules.

Moisture stress negatively affected the productivity via affecting the phenological, physiological and biochemical traits. Adverse impact was evident on growth, yield and yield components however variation existed within the genotypes. RB-50, RH 406, PBR 422, CSR 1163 and PBR 357 were promising under moisture stress with seed yield reduction of ≤20%, DSI≤0.5, DTI≥1 and DTE>90% with lesser decline in the morpho-physiological traits and increased antioxidative enzyme activities.

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## APPENDIX - I

**Weekly mean meteorological data recorded during the crop season (2017-18) at Meteorological Observatory, Department of Climate Change and Agricultural Meteorology, PAU Ludhiana**

| SMW No. | Dates | Temperature (°C) | | Mean | Relative Humidity (%) | | | Rainfall (mm) | No. of rainy days | Evaporation (mm) | Sunshine hours | Wind velocity (km/hr) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Max | Min | Mean | M* | E* | Mean | | | | | |
| 45 | Nov 5-11 | 26.1 | 14.1 | 20.1 | 96 | 57 | 76.5 | 0 | 0 | 7.6 | 1.7 | 0.8 |
| 46 | Nov 12 - 18 | 22.3 | 12.9 | 17.6 | 90 | 60 | 75 | 7 | 1 | 7.6 | 1.3 | 3.1 |
| 47 | Nov 19 - 25 | 23.9 | 7.4 | 15.7 | 94 | 29 | 61.5 | 0 | 0 | 16.2 | 7.9 | 2.1 |
| 48 | Nov 26 - Dec 2 | 25.4 | 7.9 | 16.7 | 94 | 31 | 62.5 | 0 | 0 | 14 | 7.4 | 1.2 |
| 49 | Dec 3 - 9 | 22.7 | 7.3 | 15.0 | 87 | 30 | 58.5 | 0 | 0 | 14 | 6.2 | 2.2 |
| 50 | Dec 10 - 16 | 17.1 | 9.3 | 13.2 | 90 | 70 | 80 | 24 | 1 | 10.2 | 3.4 | 2.8 |
| 51 | Dec 17 - 23 | 21.9 | 7.4 | 14.7 | 91 | 47 | 69 | 0 | 0 | 9 | 7.9 | 1.4 |
| 52 | Dec 24 - 31 | 20.7 | 6.3 | 13.5 | 96 | 49 | 72.5 | 0 | 0 | 13.8 | 4.9 | 1.5 |
| 1 | Jan 1 - Jan 7 | 15.9 | 5.4 | 10.7 | 96 | 66 | 81 | 0 | 0 | 6.2 | 2.1 | 2.4 |
| 2 | Jan 8 - 14 | 20.8 | 5.3 | 13.1 | 94 | 43 | 68.5 | 0 | 0 | 11.6 | 7.6 | 2.8 |
| 3 | Jan 15 - 21 | 22 | 6.1 | 14.1 | 92 | 40 | 66 | 0 | 0 | 14 | 7.7 | 3.5 |
| 4 | Jan 22 - 28 | 15.5 | 7.6 | 11.6 | 93 | 76 | 84.5 | 18.4 | 1 | 9.6 | 3.6 | 4 |
| 5 | Jan 29 - Feb 4 | 21.2 | 7.6 | 14.4 | 91 | 46 | 68.5 | 0 | 0 | 13.6 | 8.1 | 3.3 |
| 6 | Feb 5 - 11 | 21.1 | 5.6 | 13.4 | 89 | 38 | 63.5 | 2.4 | 0 | 15 | 8 | 2.9 |
| 7 | Feb 12 - 18 | 21.1 | 9.3 | 15.2 | 89 | 53 | 71 | 21.4 | 1 | 15.8 | 7.4 | 5 |
| 8 | Feb 19 - 25 | 25.5 | 11.7 | 18.6 | 88 | 48 | 68 | 3.2 | 0 | 17.4 | 7.5 | 3.1 |
| 9 | Feb 26- Mar 4 | 25.8 | 13.1 | 19.5 | 89 | 51 | 70 | 0 | 0 | 17.9 | 6.5 | 3 |
| 10 | Mar 5 -11 | 27.2 | 12.2 | 19.7 | 88 | 42 | 65 | 0 | 0 | 24.4 | 10.4 | 3.2 |
| 11 | Mar 12 - 18 | 29.9 | 14.1 | 22.0 | 85 | 30 | 57.5 | 0 | 0 | 29.8 | 10 | 3 |
| 12 | Mar 19 - 25 | 29.2 | 14.2 | 21.7 | 86 | 44 | 65 | 0 | 0 | 27.8 | 7.8 | 4.3 |
| 13 | Mar 26 - Apr 1 | 33.1 | 16.5 | 24.8 | 74 | 29 | 51.5 | 0 | 0 | 38.6 | 10.1 | 4.6 |
| 14 | Apr 2 - 8 | 34.8 | 20.3 | 27.6 | 66 | 33 | 49.5 | 0 | 0 | 42.2 | 5.9 | 5.1 |
| 15 | Apr 9-15 | 33.1 | 18 | 25.6 | 73 | 32 | 52.5 | 10 | 1 | 40.5 | 7.4 | 4.7 |

*M = Morning, E=Evening

## VITA

| | | |
|---|---|---|
| **Name** | **:** | Rhythm |
| **Father's name** | **:** | Sh. Ackshey Verma |
| **Mother's name** | **:** | Smt. Sureena Kumari |
| **Nationality** | **:** | Indian |
| **Date of birth** | **:** | 3rd May, 1996 |
| **Permanent address** | **:** | St. No. 2, Main Bazaar, Malout,<br>District Sri Muktsar Sahib, Punjab, India |

**EDUCATIONAL QUALIFICATION**

| | | |
|---|---|---|
| **Bachelor's degree** | **:** | B.Sc. (Medical) |
| **University** | **:** | Panjab University, Chandigarh |
| **Year of award** | **:** | 2017 |
| **%age of marks** | **:** | 76.25% |
| **Master's degree** | **:** | M.Sc. (Botany) |
| **University** | **:** | Punjab Agricultural University, Ludhiana |
| **Year of award** | **:** | 2020 |
| **Ocpa** | **:** | 7.44/10.00 |
| **Title of master's thesis** | **:** | Differential response of Indian mustard (*Brassica juncea* L. Czern & Coss) genotypes under rainfed and irrigated conditions |