CONSEQUENCE OF LAC CULTIVATION ON PIGEONPEA (*CAJANUS CAJAN*)SEEDS AND SEEDLINGESTABLISHMENT

Authors: J. Ghosh[1*], Akash Jyoti[2], V. D. Lohot[1], N. K. Sinha[1], S. Ghosal[1], Thamilarasi K[1], V. V. Thakur[1]
[1.] ICAR-Indian Institute of Natural Resins and Gums, Namkum, Ranchi
[2.] Department of Biotechnology, VinobaBhave University, Hazaribhag
Lac Production Division, ICAR-Indian Institute of Natural Resins and Gums, Namkum, Ranchi 834010, Jharkhand
e-mail:*jghoshranchi@gmail.com

ABSTRACT

Six promising germplasm of pigeonpea (Birsa Arhar 1, Bahar, Assam local 1, IPA 8-2, RCMP2 and RCMP5) identified from earlier experiment was taken to know whether seeds and its seedlings obtained from lac infected plant vis-à-vis control differed significantly with respect to morphology, physiology and biochemistry.The seedlings which were raised from lac infected plants reduced germination percent by 10.91 % at 21 days as compared to control. However, there was no significant change in fresh and dry weight of seedlings,root length, shoot length, leaf lengthand width length and even leaf area noted after 21 days. As far as biological traits are concern there was no change in total sugar in leaves, but soluble protein content decreased and phenol increased in seedlings which was raised from seeds harvested from lac infected plants as compared to control.In our study there was slight decrease in root activity andslight increase in membrane permeability in seedlings raised from the seedsof lac infested plants. Though rearing of lac insect on pigeonpea reduces grain component significantly but most of them are not hindering the establishment of the new plant. Thus farmers can cultivate pigeonpea for grain and lac resin to enhance the economy.

Kay words: Pigeonpea, Cajanuscajan, lac cultivation, Kerria lacca Kerr., effect, seedling vigour

Introduction

Pigeonpea[*Cajanus cajan* (L.) Mills.], a legume plant belongs to the family Fabaceae and subfamily Papilionaceae.India contributes over 90% of the pigeonpea production in the world where it is mostly consumed as dehusked splits or dal. The crop travelled from India to Malaysia, then to East Africa, then up the Nile Valley to West Africa, then travelled to Zaire or Angola prior to the main slave trade. Small-scale farmers of rainfed agriculture in the arid and semi-arid tropics (SAT) cultivate this legume as backyard plant, grow them on the field bunds or use them as intercrop or mixed crop. This crop is unique as it is both a legume anda woody shrub. It has an inherent ability to withstand environmental stress, specifically short periods of drought. Considered as a traditional food crop, it is a major income generator in many households (Areke 2004).These days, pigeonpea is an important food grain legume in Asia, South America and in southern and eastern Africa. In these parts of the world, pigeonpea are grown either for canning or split pulse (dal). The pigeonpea is well balanced nutritionally and an excellent source of protein whether eaten as a green pea or as dried grain (Faris and Singh1990). In addition to protein, pigeonpea provides carbohydrates and 5-fold higher levels of Vitamin A and C (Faris*et. al.* 1987).

Glover in 1937 reported that pigeonpea is an important lac host-plant in Assam, North-East India which prefers *Kerriachinensischinensis*species of lac insect(Sharma, Bhattacharya, Sushil 1999).Since then trials begins at ICAR-Indian Institute of Natural Resins and Gums (erstwhile Indian Lac Research Institute) Ranchi, India to explore its potential for lac production and found that *rangeeni* strain of *K. lacca* (Kerr) was most suitable for lac production on pigeonpea. It is a popular lac insect host-plant in Yunnan province of China also (Li *et al*., 2001).On-farm lac production with pigeonpea has recently emerged as a result of increasing demand of lac from various parts of world. Our earlier studies though showed that lac cultivation on pigeonpea seed quality decreases seed yield butincome obtained through combination of seed as well as lac culture rather from seed as sole crop will compensate the loss (Sharma and Ramani 2013 and Lohot *et al*., 2018). Ghosh *et. al.,* 2014 reported non-significant reduction in crude protein in seeds after lac culture.

In India there is practice prevalent among the farmers to use seeds of previous years for current season crop. Therefore, it is necessary to study whether lac cultivation has an impact on seed germination and seedling establishment.With this view present work has been framed to know the effect of lac culture on pigeonpea seed germination and seedling establishment.

Materials and Methods

Six germplasms(3 germplasm *viz*., Assam local 1, RCMP2, RCMP5 are from North East region, IPA 8-2 is of high grain yielding advance line and two checks *viz*., Bahar and Birsa Arhar 1, released for this region)weretaken for study. Seeds harvested from lac inoculated plants and control plants (without lac inoculation) in each germplasm were sown in RBD with six replications to know the after harvest effect of lac cultivation on seed quality, germination ability of seeds, seedling vigour, physiology and leaf biochemical constituents etc. Data were recorded for morph-physico traits *viz*., 100 seed weight (in gram), germination of seeds after 7, 14 and 21 days of soaking in water (in percent), fresh and dry weight of seedlings, dry weight of shoot and roots (in gram), shoot and root length (in cm) of seedlings, leaf length and width (in cm) and area (cm^2). The leaf biochemical constituents'*viz*., Total Sugar was estimated by phenol method as described by Dubois *et al*., (1951) and Buysse and Merck, (1993). Estimation of total soluble proteinwas done as per method described by Lowry *et al*., (1951) and Total (Free) Phenols by Folin-Ciocalteau method of Bray and Thorpe (1954).

Root activity in terms of its dehydrogenation capacity and maintaining a status of redox potential to carry out the normal respiratory cycle is the greatest limitation from the point of view of the healthy growth of the crop. The methodology develop for fast screening of genotypes on the basis of the calibration of dehydrogenage activity in roots using 2-3, 5 triphenyletetrazolium chloride and measuring the intensity of the reduced product formazan formed giving pink colour, stood the test of varietal screening best suited for lac cultivation.Twenty one days pigeonpea plant was used for the study of root activity and root membrane activity. Five mm length of root apices were excised for the determination of root activity (Sinha et. al. 20014) and root membrane permeability was measured from root diffusate which was collected containing 20 ml of distilled water in beaker (Sinha et. al., 2006). Data were analysed from standard statistical package to draw conclusive result.

Results and Discussion

- ***Post harvest effect of lac culture on morpho-physiological traits***

The traits in table 1 was average of six germplasm evaluated for this study. Weight of 100 seeds was measured from harvest in lac inoculated plants and no lac inoculated plants (i.e., control)

separately. Only 3.94% decrease in 100 seed weight was observes as effect of lac culture (Table 1).Sharma and Ramani 2013 and Ghosh *et. al.* 2014 (Lohot *et al*., 2018) also observed reduction in grain yield after cultivation of summer rainy season lac. In this experiment germination percent observed at 21 days on the seedlings which was raised from lac infected plants reduced by 10.91 % as compared to control.The range of germination of seeds varied from 70.42 (seeds harvested from with lac cultured plants *i.e*.,infected) to 79.04 (seeds harvested from without lac cultured plants *i.e*., control). Seedling grew smoothly from both types of seeds and there was no significant difference in fresh weight and dry weight of seedlings, and dry weight of shoots and roots. The differences in length of shoots, roots and leaves of seedling raised from both type of seeds were also non significant. Even leaf area measured was found non significant. Observations for morphological trails revealed minor non significant differences in almost all traits except slight reduction in germination.

Table 1. Post harvested effect of lac culture on morphological traits in pigeonpea seedlings

Morphological traits	Seeds harvested from without lac culture	Seeds harvested from withlac culture	% Increase/ decrease	CD at 5 % interaction
100 seed weight (g)	9.38	9.01	3.94	0.12
Germination of seeds	79.04	70.42	10.91	2.02
Fresh weight (g) of seedlings	4.80	4.77	0.63	ns
Dry weight (g) of seedlings	1.14	1.09	4.10	ns
Dry weight (g) of roots	0.133	0.130	2.50	ns
Dry weight (g) of shoots	1.01	0.96	4.94	ns
Shoot length (cm) of seedlings	16.76	16.48	1.68	ns
Root length (cm) of seedlings	4.58	4.48	2.11	ns
Leaf length (cm) of seedlings	4.78	4.78	0.10	ns
Leaf width (cm) of seedlings	1.49	1.53	2.58	ns
Leaf area (cm^2) of seedlings	4.70	4.75	1.06	ns

- ***Post-harvest effect of lac culture on biochemical traitsin pigeonpea seedlings***

Sugar in leaves of seedlings of pigeonpea

Sugar is important to plants because it is a source of energy that can be converted into other components like starch or cellulose. These components are necessary for cell structures such as the cell walls and for the plant's development. Glucose is also necessary for respiration in plants.There was significant variation in sugar content in leaves of seedlings of pigeonpea germplasm which was raised from harvest of lac inoculated plants (infested) and without lac inoculated plants (control). The interaction among germplasm and infestation condition was also significant indicating that germplasm behaved differently on infestation.The seedling raised from seeds of lac inoculated plants had elevated sugar in three germplasm (BisraArhar 1, Bahar, Assam local 1). It means, lac insect exceeded more sugar accumulation in these germplasm. It employs that photosynthetic rate augmented in Birsa Arhar 1, Bahar and Assam local 1 to feed plant in case of seedlings raised from lac cultured plants. Remaining three (IPA 8-2 and RCMP2 and RCMP 5)had its reduced level as compared to control. In other word lac insectsrestrict these germplasm to produce more photosynthate. Among six genotypes evaluated, Bahar and Assam local 1 has relatively less deviation in sugar content in leaves. Thus it is supposed to have better genetic composition for sustaining lac culture. Overall there was no significant post-harvest effect of lac culture on level of sugar in leaves of seedlings.

Protein in leaves of seedlings of pigeonpea

Proteins, the building material, are made up of smaller building blocks called amino acids. Plants require about 20 amino acids in order to start protein synthesis and for plant growth to occur. Germplasm had significantly different quantity of protein in their leaves (Table 2). Highest protein content of 68.5 mg/g frwt was recorded in leaves of RCMP 2, followed by 53.6 and 56.6 mg/g frwt in RCMP 5(both from Manipur) in control condition. There was no distinct trend of deviation in protein content in leaves of seedlings raised from harvest of lac infested plants as compared to control.It increased in Birsa Arhar 1, Assam local 1 and RCMP5 and decreased in Bahar and IPA 8-2 and RCMP2 in leaves.There is reallocation of synthates for their self defense. In spite of increase or decrease in protein content little deviation was observed in Assam local 1 which conserve protein content in leaves. Ghosh *et. al.* 2014 reported non-significant decrease in protein content in matured pods after lac culture. After infestation of chewing and sap sucking insect, peroxidase activity in the sap and total soluble protein (TSP) enhanced (Singh et. al. 2013). Proteins are often reallocated away from pathogen-infected tissues, while the same infection sites may draw carbohydrates to them. There is a tug of war in which the plant withdraws critical resources to block microbial growth while the microbes attempt to acquire more resources. Insects can increase local sink strength, drawing carbohydrates that support defense production (Schultz et. al. 2013).

Polyphenolin leaves of seedling of pigeonpea

Polyphenols are phytochemicals, meaning compounds found abundantly in natural plant food sources that have antioxidant properties. These are secondary metabolites of plants and are generally involved in defense against ultraviolet radiation or aggression by pathogens (Beckman 2000). They play key roles in the growth, regulation and structure of plants and vary widely within different plants. Plants need phenolic compounds for pigmentation, growth, reproduction, resistance to pathogens and for many other functions (Lattanzioet. al. 2006).

Germplasm,lac infested conditions andinteraction between them was significant in leaves of seedling of pigeonpea for poly phenol (Table 2). Polyphenol level in leaves of seedlings which was raised from harvest of lac cultured plants increase in general. Bahar felt least stress in seedlings raised from lac cultivated plantsas compared to remaining germplasm as the differencesofpolyphenol in two situations was recorded low. It means phenol shifted to inoculation area which results in increase of its level in leaves.

Table 2. Post harvested effect of lac culture on biochemical traits in pigeonpea seedlings

Germplasm	Sugar in leaves of seedlings raised from seeds of two conditions (mg/g fresh weight)		Protein leaves of seedlings raised from seeds of two conditions (mg/g fresh weight)		Polyphenol leaves of seedlings raised from seeds of two conditions (mg/g fresh weight)	
	Inocula	Contr	Inocula	Contr	Inocula	Contr

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Birsa Arhar 1	29.7	22.4	33.3	28.3	19.9	15.8
Bahar	33.2	28.2	29.3	40.3	21.7	20.5
IPA 8-2	21.3	26.6	35.6	41.0	19.3	17.6
Assam local 1	22.1	18.0	59.3	52.0	16.4	15.1
RCMP2	19. 5	22.2	60.3	68.5	16.4	16.5
RCMP5	28.6	37.0	66.3	53.6	15.6	9.2
Mean	**25.7**	**25.7**	**47.3**	**57.3**	**18.2**	**15.8**
CD (germplasm)	3.5		9.6		2.46	
CD Inoculation	N/A		5.6		1.42	
CD interaction	4.9		13.6		3.48	

Katarzyna*et. al*. (2014) noted significant increase of polyphenol (peroxidase) activity and proline content after 7 days of insects feeding.Shafiquea*et. al.* 2014 observed that defensive biochemical's of cotton i.e. phenolics and terpenoids were also significantly increased (up to 7 times) at mealy bug injury. Plants respond to herbivore through various morphological, biochemicals and molecular mechanisms to counter/offset the effects of herbivore attack.The defensive compounds are either produced constitutively or in response to plant damage, and affect feeding, growth, and survival of herbivores. In addition, plants also release volatile organic compounds that attract the natural enemies of the herbivores. These strategies either act independently or in conjunction with each other (War *et. al.* 2012). Singh *et. al.* 2013 reported that insect infestation by *Rhizoperthadominica* causes weight loss in wheat yield and significant reduction in sugars, proteins whereas phenol content spiral up.

- **Post-harvest effect of lac culture onphysiological state of seedling of pigeonpea**

The data recorded in table 3, clearly demonstrates that root activity increased in seedling grown from seeds harvested from lac inoculated plants of pigeonpea in general with exception in Birsa Arhar 1. RCMP 2 had showed significant deviation in root activity. Seeds harvested from lac cultured plants observed to be enhanced root activity which trigger plant growth. The decrease in root activity refers to the possibility of accumulation of end product in a toxic concentration which inhibit root function under the situation of biotic and abiotic stress. Sinha et al. (2004) have also reported rapid accumulation of toxic products which lead to the inhibition of the root activity in terms of total dehydrogenase activity under stress caused by herbicides (Sinha et. al., 2004) and acidity (Sinha et. al., 2016). Chirkova (1978) has reported rapid accumulation of fermentation product which leads to the inhibition of the dehydrogenage activity. In our experiment, root activity in Birsa Arhar-1 and IPA8-2 found higher even in arhar germplasm produced from the lac inoculated plants.

Likewise, the loss of membrane permeability increases in the susceptible plants, as evident from the data presented with regard to soluble sugar concentration in the diffusate around rhizosphere (table 3). The use of higher concentration herbicide anilophos affecting the membrane integrity by increasing permeability was also observed by Sinha et al. (2006) in maize plant and in rice grown under acidic situation (Sinha et. al., 2016) which might corroborate with finding obtained in Table-3. This observation very much corresponds with the level of dehydrogenase in plant which fluctuates with the intensity of biotic stress. Zhang et. al.(1992) have reported that water submergence increased with the permeability of root in waterlogged sensitive cultivar of beet root this increased the passive transport which may occur either through the cell membrane or via an extracellular pathway (Greenway, 1962). Further, maize during early growth stages, under low temperature stress situation loosed the membrane permeability of the maize plants (Sinha et al., 2001). In the present study, in pigeon pea germplasm Bahar, RCMP5, RCMP2 observed stable in its root membrane even produced from the lac inoculated plants.

Table 3. Post harvested effect of lac culture on root activity in pigeonpea seedlings

Germplasm	Root activity (mg TTC reduced hr^{-1} per plant)				Root permeability (µg sugar diffused hr^{-1} per plant)			
	With lac	Without lac	Mean	% Change	With lac	Without lac	Mean	% Change
Birsa Arhar 1	45	57.7	**51.35**	-22.01	3.21	1.95	**2.58**	64.62
Bahar	32.5	28.3	**30.40**	14.84	4.16	5.25	**4.71**	-20.76
IPA 8-2	35.7	30.9	**33.30**	15.53	5.7	4.08	**4.89**	39.71
RCMP 2	46.2	46.4	**46.30**	-0.43	0.95	1.91	**1.43**	-50.26
RCMP 5	47.4	44	**45.70**	7.73	4.21	8.26	**6.24**	-49.03
Assam local 1	62.5	45.6	**54.05**	37.06	5.55	1.03	**3.29**	438.83
Mean	44.9	44.2	**43.5**	1.58	3.96	3.75	**3.86**	5.78
CD germplasm			**2.32**				**0.26**	
CD inoculation			**1.34**				**0.15**	
CD interaction			**3.29**				**0.37**	

Conclusion

The pigeon pea germplasm viz., Assam local 1, RCMP2, RCMP5, IPA 8-2, Bahar and Birsa Arhar 1) raised from the lac inoculated and non-innoculated (control) plant showed stable and no significant difference observed for morph-physiology and biochemistry of seedlings. Thus farmers can cultivate pigeonpea for grain and lac resin to enhance the economy. Hence, these germplasm open a new window for intensive lac cultivation.

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