RESPONSE OF BRASSICA VEGETABLES TO CLUBROOT DISEASE: SEEDLING ASSAY AND ON-FARM EVALUATION

Ram Devi Timila Plant Pathology Division, Khumaltar, Lalitpur

ABSTRACT

Cultivation of Brassica vegetables specially cauliflower and cabbage is one of the important income generating livelihood options to the farmers. Clubroot disease (Plasmodiophora brassicae Woronin) is causing huge losses in cabbage and cauliflower production. Use of resistant varieties is the easiest, safest and cheapest method to combat with disease. No resistant varieties against this disease have been identified yet in Nepal. During 2005 to 2007, seedling screening of different Brassica vegetables collected from local market and Agriculture Research Stations was conducted under screenhouse at Khumaltar. Likewise, during 2008-2010 a set of ten resistant cauliflower and cabbage hybrids from Syngenta Seeds were verified for seedling and adult plant resistance in screenhouse and in infested farmers' field at Bhaktapur. A scale of 1-6 was used for assessing the clubroot severity. Of the Brassicas evaluated, radish genotypes and broad leaf mustard cv. Marpha Chaudapat were found resistant. Cauliflower hybrids: Clapton A, Clapton B, Clarify A, Clarify B and cabbage hybrids: Tekila, Kilazol, Kilaherb, Kilaton and Kilaxy were found highly resistant (score 1) while susceptible cauliflower check Milky Way scored more than 3. Cauliflower hybrid, Tetris was found moderately resistant. Marketable curd yield of those resistant hybrids of cauliflower and cabbage ranged from 22 to 39 Kg and 20 to 27 Kg per 5 m2 respectively. The use of those resistant hybrids could be the non chemical tool for the management of clubroot disease. However, seed should be made available in the market through concerned agencies.

Key Words: Brassica vegetables, Clubroot, Resistant, Screening,

INTRODUCTION

Brassica vegetables are the most important vegetables in Nepalese diet and play an important role in the economy of the farmers. These are highly preferred vegetables. Cultivation of cauliflower and cabbage is one of the most potential income generating sources for the farmers' livelihood especially to them who have small land holdings. Due to introduction of hybrid varieties suitable for off-season production, intensive cultivation, at least three crops of cauliflower alone or cauliflower and cabbage have been exploited. Those are highly profitable crops compared to cereal crops. The outbreak of clubroot disease caused by *Plasmodiophora brassicae* Woronin is making harassment to the cauliflower/cabbagegrowers of Kathmandu, Bhaktapur, Lalitpur, Makawanpur, Kavrepalanchowk and Ilam districts. This disease is one of the most serious problems in the cultivation of Brassica crops worldwide. At Makawanpur, the production of cauliflower went down to 300 kg from 5-6 metric tons in one of the years (2002/2003) due to this disease (Timila *et al.*, 2008)

So far in Nepal, the disease has been appeared in cauliflower, cabbage, broadleaf mustard, knol-khol, broccoli, radish (rare), garden cress, mustard and turnip. Intensive cropping of cauliflower and cabbage have been the most favoring factors of rapid building up of inoculum density in soil. Because of soilborne nature of the pathogen and its resting spores able to exist for more than 18 years in absence of Brassica hosts ((Toit, 1990 and Mitani et al., 2003), it is very difficult to control once established in the field.

Although, several management tactics are available for integrated management such as managing good drainage of cultivated field, liming of infested field to correct pH of the soil to 7.2, proper disposal of clubbed roots and the use of healthy seedlings (grown in disease free area), it is difficult to achieve satisfactory control. It could be due to the presence of highly virulent populations of *P. brassicae* and its higher density of resting spores. In relation to its management, recently Nebijin (flusulphamide) found effective (Timila and Neupane, 2009) and registered in Nepal. It is now available in the Nepalese market.

But due to health concerns and environmental pollution, today's demand is the non-chemical tool for disease management.

Use of resistant varieties is the easiest, safest, economical and efficient method for the control of plant diseases. Attempts have been made to assess Brassica vegetables for resistance to clubroot disease under screenhouse conditions (seedling assay) and to verify exotic resistant cauliflower and cabbage hybrids for their resistance to clubroot disease in field conditions of Nepal.

MATERIALS AND METHODS

Seedling assay under screenhouse conditions:

The experiments were conducted in artificially inoculated conditions under screenhouse during April – June/ July – September. There were two sets of brassica vegetables.

Different varieties and hybrids from different sources: The sources of seed were Research Stations, local market, Agroenterprinuers, NGOs, and local farmers. During 2005-2006, twenty one different brassicas were screened. Likewise, in 2008 twelve cauliflower and seven cabbage hybrids available in the market were included in the test.

Resistant hybrids from Syngenta Seeds: The Seed source was from Syngenta Seeds Vegetable (S&G). It was received from Prof. Dr. Jim Correll, Arkansas University, USA. There were five cauliflower hybrids: Clapton A, Clapton B, Clarify A, Clarify B, and Tetris, and five cabbage hybrids: Tekila, Kilaton, Kilaherb, Kilaxy and Kilazol.

For both sets of seed, seedlings were grown in steam sterilized soil on plastic trays. At 5-week-old stage, resting spore suspension (10⁵ /ml) at the ratio of 1:30 (inoculum and soil) was drenched in soil. Clubroot severity was assessed 7 weeks after inoculation by uprooting each and every seedling. Depending on the seedlings established, 12-24 seedlings per genotype were assessed.

For resistant hybrids, seedlings were transplanted in plastic pots and inoculated with the same inoculum and dose. Each hybrid was replicated three times with 12 seedlings each. Rami hybrid of cauliflower was included as susceptible check.

On-farm verification of resistant hybrids.

The experiments were conducted in naturally infested field at Bhaktapur. In 2009 only cauliflower hybrids were included. The number of plants evaluated ranged from 11 to 17 (based on seedlings available). In 2010, both cauliflower and cabbage hybrids (mentioned in the Table 3) were included. Cauliflower hybrid, Milky Way included as susceptible check. Disease-free seedlings were raised at Plant Pathology Division, Khumaltar. Plot size for each hybrid was 5.25 Sq.m. and replicated three times with 16 plants per plot. The experimental design was randomized complete block. Transplanting was done in last week of August. Horticultural practices were applied as per need. Chemical fertilizers and compost applied were 200:160:60 NPK Kg and 20 mt per hecter. Clubroot severity as well as yield parameters such as above ground plant biomass and marketable curd yield were recorded at the time of harvest.

Clubroot severity was assessed with 1-6 scale (as described below) modified from 0-4 scale of Mitani et al., 2003.

1= no disease or club

2= few clubs on lateral roots only

3= clubs in lateral or tap root slightly clubbed

4= well developed clubs in lateral or tap root but with enough roots

5=> 50% root converted to club with poor lateral roots, tap root clubs decayed or not.

6= plant died with entire clubs decayed.

Based on the clubroot severity score, the genotypes were categorized as: Clubroot scored less than 2 = resistant, 2 - 2.5 = moderately resistant and more than 3 = susceptible.

RESULTS AND DISCUSSION

Intensive farming of Brassica vegetables, limited knowledge of farmers on clubroot disease and its pathogen have favored the rapid building up of inoculum density in the soil. The disease has been spread rapidly through the movement of symptomless infected seedlings grown in an infested field and through farm implements. Roots of infected plants left in the field after harvest observed to be one of the most important limiting factors for the management of this destructive disease.

In seedling assay, among the 21 Brassica vegetables, radishes, cress and Marpha chaudapat of broadleaf mustard showed resistant reaction to clubroot disease (clubroot severity score less than 2). Kathmandu local variety of cauliflower was found moderately resistant (Table 1). It was observed that in the farmers' field visit, this variety was less affected by clubroot disease compared to other hybrids.

Table 1: Reaction of Different Brassicaceous vegetable genotypes to Clubroot disease under inoculated conditions (under Screen house). Khumaltar, 2005-2006.

S. No. Genotypes		2005		2006		
		Clubroot severity index (1-6)	Disease reaction	Clubroot severity index (1-6)	Disease	
1.	Brocooli, green sprouting	4.5	S	2.49	MR	
2.	Broad leaf mustard (BLM), local	2.22	MR	2.98	MR	
3.	BLM, Tankuwa local	3.8	S	3.11	S	
4.	BLM, Marpha chaudapat	1.8.	R	1.79	R	
5.	BLM, Ktm Local	3.11	S	2.77	S	
6.	Cauliflower, Ktm local	2.06	MR	2.24	MR	
7.	Cauliflower, Kibo giant	2.8	S	-	-	
8.	Cauliflower, Jyapu local (3 months)	3.17	S	-		
9.	Cabbage, Golden Acre	1.2	R	-	_	
10.	Cabbage, Pride of India	2.58	S	3.35	S	
11.	Chinese Cahbage, IV1116 (CR2)	1.3	R	-	-	
12.	Chinese cabbage, Meikyo (CR2)	1.0	R	-	-	
13.	Cress, Kath. Local	1.00	R		-	
14.	Cress, Indian	1.00	R			
15.	Radish, Minoearly	1.54	R	2.5	MR	
16.	Radish, 40 days	1.25	R	1.0	R	
17.	Radish, Tokinase	1.0	R	-		
18.	Radish, Pyuthane Rato	1.00	R	1.00	R	
19.	Radish, Lumle Rato	1.25	R			
20.	Radish, Local Red	2.58	S	1.64	R	
21.	Turnip, Purple top	3.00	S	-	-	

Among seven cabbage hybrids, T-621 and Green Rise were resistant, Super Green, Green Coronate, and Uji No.1were moderately resistant. Sky Ace and Golden Ball were susceptible in seedling assay (Figure 1). Similarly, of the twelve cauliflower hybrids collected from the market, four showed moderately resistant and six were susceptible (Figure 2). Hybrids Snow Mastique and Laxmi were almost susceptible. Among the four cultivars of broadleaf mustard generally cultivated in Nepal, Marpha chauda was found moderately resistant to resistant. Tankhuwa Rayo as well as Khumal Ratopat were susceptible (Figure 3).

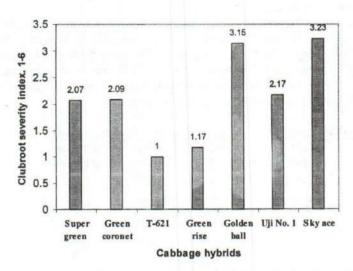


Figure 1. Reaction of different cabbage hybrids to clubroot disease in seedling assay (2008).

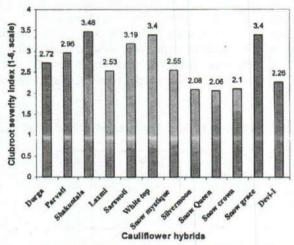


Figure 2. Reaction of different cauliflower hybrids to clubroot disease in seedling assay (2008).

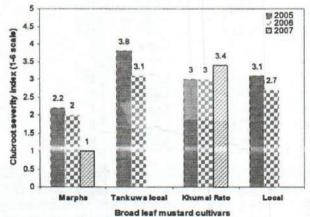


Figure 3. Reaction of broad leaf mustards to clubroot disease in seedling assay

Verification of resistant cabbage and cauliflower hybrids:

The seedling assay of resistant cauliflower and cabbage from Syngenta Seeds & Vegetable, showed that except Tetris all were found free from clubroot showing highly resistant reaction. However Tetris also showed resistant reaction (1.86 clubroot index with 29% disease incidence). The susceptible check, Rami showed 2.73 clubroot severity index with 53% incidence. On-farm test during 2009 showed similar reactions to clubroot disease by cauliflower hybrids. Tetris was found to be moderately resistant with 59% clubroot incidence whereas, the susceptible check Milky Way showed 99% incidence (Table 2).

Table. 2. Clubroot reaction of resistant cauliflower hybrids in the infested field at Bhaktapur, 2009

S.N.	Cauliflower hybrids	No. of tested plants	Severity index (1-6)	Mean Incidence %	Total marketable curd yield (kg/5	Mean weight per curd	Disease. Reaction
1.	Clarify A	15	1	0	13.9	0.92	R
2.	Clarify B	17	1	0	25.5	1.50	R
3.	Clapton A	11	1	0	14.4	1.34	R
4.	Clapton B	13	1	0	23.4	1.80	R
5.	Tetris	12	2.42	58.33	14.4	0.95	MR
6.	Milky Way	15	3.5	99.98	6.59	1.25	S

Among cauliflower hybrids tested in 2010, Clapton A, Clapton B, Clarify A and Clarify B were free of clubroot reacting resistant and Tetris showed susceptible reaction (clubroot severity index of 2.57) with disease incidence of 53% (Table 3). Though, it was supposed to be resistant. The susceptible check, Milky Way showed 3.2 clubroot severity index and 61% incidence. The marketable curd yield of those resistant hybrids of cauliflower ranged from 22 to 39 Kg per 5 m². The curds of cauliflower hybrids were solid with dense florets and strong against splitting. The weight of individual marketable curd ranged from 1.8 to 2.5 kg, which showed good yield potential. Farmer preferred the cauliflower curds, however, they were not smooth enough.

Table 3. Clubroot reaction of resistant cauliflower and cabbage hybrids in the infested field at Bhaktapur, 2010 (2067/68)

S, No.	Hybrids	Mean Severit y index (1-6)	Mean Incidence, %	Mean Weight of top of the plants, 5 m ² (Kg)	Mean Weight of marketable curd, 5 m ² (Kg)	Average weight per curd/head	Disease Reaction
	Cauliflower						
1.	Clarify A	1	0	56.86	35.92	2.24	R
2.	Clarify B	1	0	61.56	39.37	2.46	R
3.	Clapton A	1	0	46.81	29.29	1.83	R
4.	Clapton B	1	0	59,08	38.44	2.40	R
5.	Tetris	2.57	52.22	32.73	22.09	2.20	S
6.	MilkyWay Cabbage	3.19	60.59	19.70	11.25	1.12	S
1.	Tekila	1	0	35.24	20.44	1.27	R
2.	Kilaton	1	0	42.15	22.52	1.49	R
3.	Kilaherb	1	0	38.80	26.64	1.66	R
4.	Kilaxy	1	0	34.40	20.00	1.25	R
5.	Kilazol	1	0	31.44	20.58	1.29	R

Likewise, all cabbage hybrids, Tekila, Kilaton, Kilaherb, Kilazol and Kilaxy were free from clubroot, which showed highly resistant reaction. The marketable cabbage head yield of those hybrids ranged from 20 to 27 Kg per 5 m². Individual marketable head ranged from 1.2 to 1.7 Kg. The farmer preferred Kilaherb among cabbage hybrids. It was round in shape with 1.7 Kg per head. Cabbage heads of other hybrids were flexible in shape and size. All those cauliflower and cabbage hybrids mentioned to be good in shape and size (S&G, 2007). The use of those resistant varieties will also help to lower the inoculum build up in the field.

It was found that the resistant hybrids from Syngenta Seeds were resistant to our indigenous local isolate of *P. brassicae* of Bhaktapur. It is necessary to test the genotypes with local isolate of the pathogen to confirm their resistance in the given location or place before introducing for cultivation. Sinceresistance of the host plant may be affected by the virulence of the pathogens, their density (inoculum pressure) and the local environment. Until 2004, resistant varieties in Brassica crops were only available in Chinese cabbage, oriental radish, turnip and oil seeds (S&G, 2007). In evaluation of 126 cauliflower, 38 cabbage and 240 kale French landraces, except two kale landraces, which were resistant, all other Brassica landraces tested were susceptible to test isolate of the pathogen (Manzanares-Dauleux *et al.* 2000). Similarly, no resistance was found in 109 germplasms of Brassica accessions screened against clubroot disease (Crete and Chiang, 1980).

In the experiment, KathmanduLlocal variety of cauliflower showed moderately resistant reaction, but farmers are not interested to use that variety any more because of its longer cropping period. In another seedling assay of 36 Brassica varieties grown in Nepal, one turnip and one broadleaf mustard (Marpha Chaudapat) were moderately resistant and resistant, respectively ((PPD, 2009). It showed that resistance to clubroot disease in the Brassicas is rare.

CONCLUSIONS

It is concluded that radishes are mostly resistant. Hybrids of cauliflower or cabbages available in the market are mostly susceptible to clubroot disease. However, results of seedling assay needs to be verified in the field conditions. But resistant hybrids of cauliflower (Clapton A and B, Clarify A and B) and cabbage hybrids (Tekila, Kilaton, Kilazol, Kilaherb and Kilaxy) from Syngenta Seeds could be used in managing clubroot disease in Nepal also. Cultivation of those resistant hybrids might help reduce the inlculum building in the field. However, the seed should be made available in the market through concerned agencies.

ACKNOWLEDGEMENTS

Sincere thanks are expressed to the former Chief Plant Pathologist, Mrs. S. Sharman and Dr. HK Manandhar Present Chief Plant Pathologist, Plant Pathology Division, NARC for their valuable suggestions and supports. Special thanks are extended to Prof. Dr. Jim Correll, Arkansas University, USA for making seeds available. The help of farmer, Mr. Sri Bhakta Sujakhu is highly acknowledged for availing field for the experiment.

REFERENCES

- Crete R. and M. S. Chang. 1980. Screening brasicas for resistance to clubroot, Plasmodiophora brassicae Wor. Can. Plant Dis. Surv. 60(1)17-19.
- Manzanares-Dauleux M.Z., I. Divaret,, F. Baron and G. Thomas. 2000. Evaluation of French Brassica oleracea landraces for resistance to Plasmodiophora Brassicae Euphytica 103.211-218. (Abs.)
- Mitani, S., K. Sugimoto, H. Hayashi, Y. Takii, T. Ohshima and N. Matsuo. 2003. effects of Cyazofamid against Plasmodiophora brassiocae Woronin on Chinese cabbage. Pest Manag Sci. 59:787-293.
- PPD. 2009. Annual report. Plant Pathology Division. Nepal Agriculture Research Institute, NARC.
- S and G. 2007. Syngenta Catalogue.
 - http://www.syngenta.com/country./se/sv/gronsaksfro/documents/clubroot.pdf
- Timila, R.D. and J.D. Neupane. 2009. Nebijin (flusulfamide) on the Management of Clubroot Disease of Cauliflower. 274-279. In the Proceedings of 5th National Seminar on Hoticulture held at Khumaltar, 9-10 June, 2008.
- Timila, R. D., J. C. Correll, and V. R. Duwadi. 2008. Severe and idespreadClubroot Epidemics in Nepal. Plant Disease. 92(2):317 DOI: 10.1094/PDIS-92-2-0317B (Disease Note)
- Toit, Lindsey, J. du. 1990. Clubroot of cabbage and other crucifers. In revised Plant Disease Bulletin (2004). Washington State University. Extension Subject Codes 270,356 A