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Selection of potential multivoltine parental stocks for improving quality cross breed silk production through pre breeding

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Abstract

India, a country of tropical climatic conditions is mainly dependent on the crossbreed and multivoltine type of silk production. The total production of all types of raw silk in India is 26480 M.T, out of which mulberry silk is 19476.00 MT which includes 86.86 % multivoltine type and the rest is bivoltine silk. Therefore it becomes a priority to improve the quality and quantity of multivoltine silk. In this context, evolving superior multivoltine parental breeds for tropical conditions in the crossbreed dfls production is very much essential. The multivoltine silkworm genetic resources conserved at Central Sericultural Germplasm Resources Centre, Hosur comprising 69 indigenous and 10 exotic accessions would be handy to select the better multivoltine parent material. Therefore attempts were made through pre-breeding approaches using multivoltine genetic resources in shortlisting superior multivoltine breed combinations for further utilization by the breeders, for improvement of quality silk production.

Keywords: Silkworm genetic resources, Conservation, Multiple trait evaluation index

1. Introduction

India being the country of tropical climatic conditions is mainly dependent on the crossbreed and multivoltine type of silk production. In fact sericulture industry in India is multivoltine oriented and about 87 % of the mulberry silk production is of multivoltine origin and the rest is bivoltine ^[1]. Though there has been wide spread requirement for the superior and high quality bivoltine silk it is not possible to get bivoltine silk production from our farmers through out the year. At the same time it is quite possible to develop crossbreeds through selection of better multivoltine parent from our genetic wealth and produce high quality cross bred silk. Further the climatic conditions in the tropical countries like ours exhibit wide fluctuations in the weather parameters and this situation coupled with the inferior quality of mulberry leaves and poor management practices by the farmers requires hardy and flexible races.

The present production of silk, as per 2013-14 figures are 26480 M.T out of which Mulberry silk is 19476.00 MT, Tasar 2619 M.T, Eri 4237 M.T and Muga 148 M.T ^[1]. The demand for silk is 29,740 M.T. The production formed only 89.04 % of the requirement and the gap is met by imports. Even in the mulberry silk production statistics 16917 M.T (86.86 %) of the mulberry raw silk produced is of multivoltine variety and the remaining 2559 M.T (13.14 %) is BV silk it becomes a high priority to improve the quality of silk as also its production. In this context, evolving superior multivoltine parental breeds suiting to the tropical climatic conditions and better combiner in the crossbreed dfls production is very much essential. The selection of parents as resource material is a prerequisite contributing to the success of breeding potential breeds/ hybrids. Thorough and proper evaluation of the genetic resources and utilization of the native breeds of potential nature will help the breeder to select most effective genotypes before choosing the materials for breeding.

The multivoltine silkworm genetic resources wealth of the country conserved at Central Sericultural Germplasm Resources Centre, Hosur (CSGRC) numbering about 79 accessions (69 indigenous and 10 exotic) collected from the sericulturally advanced countries like China, Japan and India will come handy in order to select the better multivoltine parent material for inclusion in the breeding programme. These collections are conserved following standard germplasm maintenance protocols for more than 93 generations under the tropical conditions and have stabilized to the tropical conditions of the country ^[2]. Therefore these multivoltine genetic resources maintained would help in identifying and recommending superior multivoltine breeds for further utilization by the breeders in evolving new multivoltine

components for use in production of new cross breeds towards the improvement of quantum of silk production output of the country.

Selection of parents as resource material is a prerequisite contributing to the success of breeding potential breeds/ hybrids. Thorough and proper evaluation of the genetic resources and utilization of the native breeds of potential nature will help the breeder to select most effective genotypes before choosing the materials for breeding. Balancing and fixing the desirable traits for local environments being the challenge for the breeder, proper understanding on the range of reaction of the selected genotypes under variable environmental conditions for appropriate use in breeding programme is very essential [3, 4].

With this objective CSGRC, Hosur has initiated pre-breeding efforts to identify promising parental race from among the 77 multivoltine accessions, which has better combining ability when crossed with the popular male parent CSR-2. The data on the important rearing parameters and three post cocoon traits were also included for the evaluation and accessions performing better than the ruling popular cross breeds like PM X CSR-2 and Nistari X CSR-2 for the above traits were shortlisted for inclusion zonal evaluation and in crop improvement programmes. Though silkworm breeders have various statistical methods [5, 6, 7] and multiple trait evaluation Index method [8] is an ideal method in selecting the better parental races.

2. Materials and Methods

Seventy-seven multivoltine accessions conserved at CSGRC, Hosur from different geographical locations were taken for the study. These hybrid combinations of 77 multivoltine accessions after crossing with CSR-2 as male parents were reared in three replications with 250 larvae each per replication between March-April 2015. Rearing was conducted by following standard rearing procedures [9]. Data recorded for 10 rearing and three post cocoon traits for evaluating their superiority.

The rearing traits studied includes weight of 10 larvae (g.), larval duration (hrs.) cocoon yield/10000 larvae by no, cocoon yield/10,000 larvae by wt. (Kg), pupation rate (%), shell ratio (%), cocoon yield /100 dfls (kg.) and the post cocoon traits are

filament length (m), non broken filament length(m) and denier. In order to judge the superiority of silkworm accessions impartially the Evaluation Index (EI) developed [8] was utilized. The EI is an index of multiple traits or as a performance index, which is a single valued measure of the multiple trait performance of a population. EI was obtained as follows:

$$E.I. = \frac{A - B}{C} \times 10 + 50$$

Where A = mean of the particular trait
 B = overall mean of the particular trait
 C = standard deviation
 10= fixed value
 50 = constant

In silkworms when large numbers of hybrids are tested and promising ones are selected based on the Evaluation Index (EI). EI is the strategy, which can increase the precision of the selection of hybrids among the array of breeds and giving adequate weightage to all the yield component traits. As such it will be fair and precise to take the decision based on the multiple traits spanning the entire growth period, so that adequate weightage is given to various characters that are manifested in the entire growth phase of the silkworm.

3. Results and Discussion

The data on the top ten hybrid combinations for individual traits among the 77 MV hybrid combinations evaluated were presented in Table-1. The weight of 10 matured larvae was maximum 35.38 g (BMI-0073 x CSR2). The pupation rate was high in BMI-0025 x CSR-2 combinations with 96.32 %. The single cocoon weight was high in the cross breed between BMI-0025 x CSR-2 (1.468g.). The single shell weight was higher in the BMI-0062 x CSR-2 hybrid combination (0.301g). Hybrid combination of BMI-0027 x CSR-2 showed the maximum shell ratio of 22.82 % and the cocoon yield per 100 dfls was higher in the BMI-0027 x CSR2 combination. The average filament length was high in BMI-0077 x CSR2 (851.8 m) so is the Non-broken filament length (851.8 m). The hybrid combination of BMI-0022 x CSR2 showed less denier of 1.89 (Table-1).

Table 1: Top ten MV hybrid combinations for rearing and post cocoon traits

Trait	Values	Accessions number
Weight of 10 larvae (g)	35.4 - 32.4	BMI-0073,BMI-0017, BMI-0078, BMI-0036, BMI-0054, BMI-0026,BMI-0040, BMI-0025, BMI-0031, BMI-0077
Total Larval Duration (h)	576 - 576	BMI-0009, BMI-0020, BMI-0031, BMI-0011, BMI-0065, BMI-0008,BMI-0007, BMI-0070, BMI-0076,BMI-0077
Fifth Age Larval Duration (h)	132 - 132	BMI-0031, BMI-0078, BMI-0024, BMI-0025,BMI-0026,BMI-0027,BMI-0028, BMI-0022, BME-0030,BMI-0021
ERR (No.)	9900 - 9800	BMI-0026, BMI-0016, BMI-0021, BME-0005, BMI-0054, BMI-0025, BMI-0007, BMI-0060, BMI-0028, BMI-0009
ERR (Wt)	15.9 - 12.9	BMI-0027, BMI-0044, BMI-0041, BMI-0074, BMI-0025, BMI-0026, BMI-0042, BMI-0062, BMI-0001, BMI-0029
Pupation Rate %	96.3 - 94	BMI-0025, BMI-0021, BME-0005, BMI-0016, BMI-0007, BMI-0034, BMI-0026, BMI-0064, BMI-0001, BMI-0038
Single Cocoon weight (g)	1.46 - 1.37	BMI-0025, BMI-0054, BMI-0053, BMI-0031, BMI-0019, BMI-0027, BMI-0070, BMI-0024, BMI-0060, BME-0030
Single Shell Weight (g)	0.30 - 0.28	BMI-0062, BMI-0053, BMI-0070, BMI-0042, BME-0048, BMI-0064, BMI-0054, BMI-0074, BMI-0025, BMI-0014
Shell Ratio %	22.8 - 22.0	BME-0013, BMI-0073, BMI-0061, BMI-0042, BMI-0057, BME-0015, BMI-0074, BMI-0062, BMI-0018, BMI-0064
Cocoon Yield /100dfls(Kg.)	63.8 - 51.4	BMI-0027, BMI-0044, BMI-0041, BMI-0074, BMI-0025, BMI-0026, BMI-0042, BMI-0062, BMI-0001, BMI-0029
Average Filament Length (m)	852 - 779	BMI-0077, BMI-0028, BMI-0053, BMI-0021, BMI-0010, BMI-0031, BMI-0008, BME-0030, BMI-0016, BME-0015
Non BrokenFilament Length (m)	852 - 755	BMI-0077, BMI-0028, BMI-0053, BMI-0021, BMI-0010, BMI-0008, BME-0030, BMI-0022, BMI-0032, BMI-0002
Denier	1.89 -2.18	BMI-0022, BMI-0076, BME-0013, BMI-0020, BMI-0075,BME-0052, BMI-0077, BME-0050, BME-0015, BMI-0003

The better performing accessions based on the trait values though multiple traits ranking revealed that BMI-0025 x CSR2 hybrid combination ranked first with better trait values in 8 parameters out of the 13 parameters tested. BMI-0026 x CSR2 combination ranked with 6 traits followed by BMI-0077 x CSR-2, BMI-0021 x CSR-2, BMI-0031 x CSR-2 hybrid combinations with better values in five economic traits. Multivoltine accessions crossed with CSR2 of BME-0030,

BMI-0028, BMI-0042, BMI-0053, BMI-0054, BMI-0027, BMI-0062 and BMI-0074 showed higher values in four traits. The following hybrid combinations (8 Nos.) showed higher values in 3 traits BMI-0007 x CSR-2, BMI-0070 x CSR-2, BMI-0016 x CSR-2, BMI-0008 x CSR-2, BMI-0022 x CSR-2, BMI-0001 x CSR-2, BMI-0064 x CSR-2 and BME-0015 x CSR-2 (table-2).

Table 2: Better performing MV accession hybrid combinations for multiple traits

Accession No.	No. of traits	Trait No. and Values
BMI-0025 x CSR-2	8	1(32.787), 3(132), 4(9833.5), 5(14), 6(96.318), 7(1.468), 8(0.286), 10(56.133)
BMI-0026 x CSR-2	6	1(32.95), 3(132), 4(9900), 5(13.8), 6(94.666), 10(55.333)
BMI-0077 x CSR-2	5	1(32.36), 2(576), 11(851.775), 12(851.775), 13(2.10)
BMI-0021 x CSR-2	5	3(132), 4(9867), 6(95.666), 11(808.875), 12(808.875)
BMI-0031 x CSR-2	5	1(32.653), 2(576), 3(132), 7(1.386), 11(797.25)
BME-0030 x CSR-2	4	3(132), 7(1.369), 11(782.475), 12(782.47)
BMI-0028 x CSR-2	4	3(132), 4(9800), 11(844.575), 12(844.575)
BMI-0042 x CSR-2	4	5(13.45), 8(0.296), 9(22.358), 10(53.866)
BMI-0053 x CSR-2	4	7(1.396), 8(0.3), 11(816.3), 12(816.3)
BMI-0054 x CSR-2	4	1(33.608), 4(9833.5), 7(1.428), 8(0.29)
BMI-0027 x CSR-2	4	3(132), 5(15.95), 7(1.378), 10(63.866)
BMI-0062 x CSR-2	4	5(13.35), 8(0.301), 9(22.168), 10(53.334)
BMI-0074 x CSR-2	4	5(14.2), 8(0.29), 9(22.175), 10(56.8)
BMI-0007 x CSR-2	3	2(576), 4(9833.5), 6(95.334)
BMI-0070 x CSR-2	3	2(576), 7(1.378), 8(0.298)
BMI-0016 x CSR-2	3	4(9900), 6(95.666), 11(780.825)
BMI-0008 x CSR-2	3	2(576), 11(789.026785714286), 12(789.03)
BMI-0022 x CSR-2	3	3(132), 12(768.46875), 13(1.89)
BMI-0001 x CSR-2	3	5(13.05), 6(94.631), 10(52.266)
BMI-0064 x CSR-2	3	6(94.638), 8(0.294), 9(22.083)
BME-0015 x CSR-2	3	9(22.283), 11(779.475), 13(2.17)

Mano's evaluation index was scores for individual hybrid combinations alongwith cumulative evaluation index values and presented in Table-3. The ranking of the different multivoltine accessions hybrid combinations based on cumulative evaluation index values and also more number of parameters were shown in the table-4 which revealed BMI-0077 x CSR2 hybrid combination has out performed all the remaining combinations with a cumulative evaluation index

value of 54.13 and better values in all the 13 traits evaluated followed by BMI-0021 x CSR-2 and BMI-0028 x CSR-2 for 12 traits with EI values of 56.53 and 55.80 respectively (Table-3 and 4). The hybrids of 7 MV accessions have showed better CEI values and also ranked superior with higher trait values in 11 out of the 13 parameters studied. Similarly 5 multivoltine hybrid combinations have showed higher CEI values and ranked better in 10 parameters.

Table 3: Top ranking MV combinations based on Cumulative Evaluation Index (CEI) values

Accession No.	Lwt.	Tld	Vld	ERR (No.)	ERR (Wt)	PR	SCwt	SSwt	SR	CY/100 dfls	AFL	NBFL	Denier	CEI
BMI-0025 x CSR-2	60.23	53.61	46.73	59.84	65.54	61.91	68.62	61.27	43.22	65.69	57.25	60.18	63.57	59.05
BMI-0064 x CSR-2	55.75	53.61	46.73	56.97	57.94	60.03	58.35	63.95	65.08	57.77	57.02	54.36	53.51	57.01
BMI-0042 x CSR-2	57.73	53.61	46.73	49.79	62.93	48.53	56.68	64.62	67.46	63.00	50.73	54.31	64.79	56.99
BMI-0054 x CSR-2	62.47	53.61	46.73	59.84	59.60	55.93	65.28	62.61	49.96	59.51	53.01	56.36	55.05	56.92
BME-0030 x CSR-2	57.89	53.61	46.73	54.82	52.01	56.68	60.35	59.93	53.74	52.08	61.26	63.78	64.25	56.70
BMI-0021 x CSR-2	54.96	53.61	46.73	60.56	58.42	61.18	52.59	53.58	53.44	58.41	64.77	66.93	49.71	56.53
BMI-0028 x CSR-2	51.73	53.61	46.73	59.12	51.53	58.17	53.34	54.92	56.61	51.60	69.52	71.20	47.33	55.80
BMI-0044 x CSR-2	57.41	53.61	46.73	45.49	68.86	48.90	60.35	55.92	45.82	68.85	57.63	54.88	55.24	55.36
BMI-0027 x CSR-2	59.05	53.61	46.73	49.79	74.80	46.61	61.10	56.25	44.86	74.86	46.80	50.78	54.22	55.34
BMI-0041 x CSR-2	58.28	53.61	46.73	55.54	68.63	54.48	53.25	52.91	50.34	68.54	50.75	49.08	53.17	55.02
BMI-0032 x CSR-2	55.34	53.61	64.73	54.10	48.45	53.70	52.67	53.58	52.07	48.44	58.59	61.37	57.96	54.97
BMI-0053 x CSR-2	57.91	53.61	46.73	35.45	49.87	47.41	62.60	65.95	60.91	49.86	65.76	67.82	46.84	54.67
BMI-0061 x CSR-2	54.96	53.61	46.73	54.82	50.35	53.62	52.50	57.93	67.52	50.34	53.80	57.08	53.55	54.37
BMI-0073 x CSR-2	67.33	53.61	64.73	37.60	42.27	46.88	48.08	57.26	70.06	42.42	60.41	57.22	57.63	54.27
BMI-0029 x CSR-2	50.84	53.61	46.73	58.41	60.32	58.95	46.74	50.57	56.58	60.15	55.01	58.16	49.11	54.24
BMI-0074 x CSR-2	56.96	53.61	46.73	23.98	66.49	33.96	57.59	62.61	65.88	66.48	60.65	52.30	57.02	54.17
BMI-0077 x CSR-2	59.06	30.42	46.73	56.97	55.09	56.63	51.25	55.25	60.02	54.92	70.47	72.06	34.83	54.13

Lwt. -10 Larval weight (g), Tld -Total larval duration (h), Vld- fifth age larval duration (h), ERR(No.)-Effective rate of rearing (No.) ERR(Wt)- Effective rate of rearing (Kg.) PR- pupation Rate (%) SCwt - Single Cocoon weight (g) SSwt - Single Shell Weight (g) SR- Shell ratio (%) CY/100dfl-Cocoon yield/100 dfls (Kg) AFL-Average Filament Length (m) NBFL-Non broken filament length (m), Denier (d) CEI-Cumulative Evaluation Index

Table 4: Multivoltine hybrid combinations for ranked based on CEI and multiple traits.

Accession No.	Lwt.	Tld	Vld	ERR (No.)	ERR (Wt)	PR	SCwt	SSwt	SR	CY/100 dfls	AFL	NBFL	Denier	CEI	Para
BMI-0077 x CSR-2	59.06	30.42	46.73	56.97	55.09	56.63	51.25	55.25	60.02	54.92	70.47	72.06	34.83	54.13	13
BMI-0021 x CSR-2	54.96	53.61	46.73	60.56	58.42	61.18	52.59	53.58	53.44	58.41	64.77	66.93	49.71	56.53	12
BMI-0028 x CSR-2	51.73	53.61	46.73	59.12	51.53	58.17	53.34	54.92	56.61	51.60	69.52	71.20	47.33	55.80	12
BMI-0004 x CSR-2	54.78	53.61	46.73	54.11	50.11	51.46	56.76	52.24	43.63	50.02	52.83	50.84	49.31	51.26	11
BMI-0007 x CSR-2	54.36	30.42	46.73	59.84	53.43	60.81	53.17	49.90	44.98	53.50	60.30	57.13	44.00	51.43	11
BMI-0010 x CSR-2	51.64	30.42	46.73	27.55	52.01	27.69	52.75	52.91	51.24	52.08	64.77	66.93	49.71	48.19	11
BMI-0029 x CSR-2	50.84	53.61	46.73	58.41	60.32	58.95	46.74	50.57	56.58	60.15	55.01	58.16	49.11	54.24	11
BME-0030 x CSR-2	57.89	53.61	46.73	54.82	52.01	56.68	60.35	59.93	53.74	52.08	61.26	63.78	64.25	56.70	11
BMI-0031 x CSR-2	59.86	30.42	46.73	47.64	54.38	50.31	61.77	55.59	42.47	54.45	63.23	54.34	43.26	51.11	11
BMI-0061 x CSR-2	54.96	53.61	46.73	54.82	50.35	53.62	52.50	57.93	67.52	50.34	53.80	57.08	53.55	54.37	11

BMI-0064 x CSR-2	55.75	53.61	46.73	56.97	57.94	60.03	58.35	63.95	65.08	57.77	57.02	54.36	53.51	57.01	11
BME-0015 x CSR-2	51.99	53.61	46.73	56.25	53.67	59.32	44.15	51.91	66.81	53.66	60.86	47.90	38.39	52.71	10
BMI-0024 x CSR-2	54.64	53.61	46.73	52.66	52.48	51.51	60.93	56.59	44.85	52.39	55.04	52.69	61.65	53.52	10
BMI-0025 x CSR-2	60.23	53.61	46.73	59.84	65.54	61.91	68.62	61.27	43.22	65.69	57.25	60.18	63.57	59.05	10
BMI-0041 x CSR-2	58.28	53.61	46.73	55.54	68.63	54.48	53.25	52.91	50.34	68.54	50.75	49.08	53.17	55.02	10
BMI-0054 x CSR-2	62.47	53.61	46.73	59.84	59.60	55.93	65.28	62.61	49.96	59.51	53.01	56.36	55.05	56.92	10

Lwt. -10 Larval weight (g), Tld -Total larval duration (h), Vld- fifth age larval duration (h), ERR(No.)-Effective rate of rearing (No.) ERR(Wt)- Effective rate of rearing (Kg.) PR- pupation Rate (%) SCwt - Single Cocoon weight (g) SSwt - Single Shell Weight (g) SR- Shell ratio (%) CY/100dfl-Cocoon yield/100 dfls (Kg) AFL-Average Filament Length (m) NBFL-Non-broken filament length (m), Denier (d) CEI-Cumulative Evaluation Index, Para-No. of Parameters

The genetic resources are the basic materials for crop improvement and the success of breeding programme depends on the initial selection of parents, their effective utilization in desirable combinations. Further it also depends on the ability of breeds to assemble and recombine the genetic variability, to extract the potential gene combinations from the gene pool based on phenotypic expression leading to genetic fixation of the traits over generation [10]. The conventional methods of breeding needs a perfect selection procedure combined with the selection of desired traits for identifying the initial parental lines [11, 12]. Therefore silkworm genetic resources, needs to be properly evaluated and screened for identifying the potential and promising parental lines. Screening the promising pure breeds from the germplasm stocks is an important duty of the persons responsible for gene bank maintenance [13]. All the 77 MV accessions were evaluated for major rearing traits and post cocoon traits and they are compared with Pure Mysore and Nistari, which are the ruling multivoltine pure breeds utilized as female parents in the commercial hybrid /cross breed production to compare the potentiality of these accessions.

The values of the hybrid combinations pertaining to PM and Nistari were set as the cut off values and those combinations which have performed better than these ruling popular cross bred races for individual traits were short listed. Good numbers of potential multivoltine combinations are available for the various rearing an post cocoon traits studied (Table-5) which can serve as female parents in the multivoltine hybrids production for both South India (Pure Mysore) and Eastern India (Nistari). Some of the cross breed combinations involving the following MV accessions BMI-0025, BMI-0077, BMI-0021, BMI-0031, BMI-0030, BMI-0028, BMI-0042, BMI-0053, BMI-0074, BMI-0061 BMI-0032, BMI-0073 and BMI-0010 as female parent with CSR-2 as male parent are found to be potential. It includes the following cocoon breeds greenish yellow (5), chrome yellow (5) white (2) and off white (1) to cater to the needs of different zones. Further evaluation at the specific agro climatic zones as per the preference of the regions will help in identification of the better regional and season specific hybrids and cross breed silk production of the country.

Table 5: High performing multivoltine combinations better than popular hybrids.

Particulars	Better than Pure Mysore Hybrid		Better than Nistari Hybrid		
	Traits	Range	No. of combinations	Range	No. of combinations
Wt. of 10grown larvae (g)		35.3 - 27.4	54	35.3 - 35.3	1
Yield/10000 larvae (no.)		9900 - 9766	13	9900 - 9700	22
Yield/10000 larvae by wt. (Kg)		15.95 - 13.35	8	15.95 - 12.7	12
Pupation rate (%)		96.3 - 94.6	8	96.3 - 93	17
Single cocoon weight (g)		1.46 - 1.30	24	1.46 - 1.26	34
Single shell weight (g)		0.301 - 0.242	53	0.30 - 0.24	48
Shell ratio (%)		22.8 - 18.5	73	22.8 - 19.4	60
Average Filament Length (m)		851.7 - 590.3	72	851.8 - 647.1	61
Non Broken Filament Length (m)		851.7 - 543.0	71	851.8 - 542.5	71
Denier		1.89 - 2.51	56	1.89 - 2.76	75

During last four decades, several multivoltine crossbreeds were evolved [2, 14, 15]. Heterosis breeding in silkworm has substantially contributed to the increase in cocoon production and in improving the quality of raw silk [15]. This pre breeding study revealed the availability of some of the potential multivoltine silkworm accessions with high degree of variability, which when used as female parents can be superior to the ruling multivoltine silkworm cross breeds. These resources are well adapted and very consistent to the tropical conditions of the country and when used in combination with the bivoltine counter parts may contribute significantly to the heterobeltiosis in hybrid seed production programmes either by contributing to the development of parental lines, as pure breeds, or in development of the region/season specific parental breeds.

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