

ESTIMATION OF PHENOTYPIC DIVERSITY OF CHILLI (*CAPSICUM* SPP.) GERMPLASM IN SRI LANKA

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ABSTRACT

Thirty eight chilli (*Capsicum* spp.) genotypes were evaluated in a field study to assess genetic diversity of *Capsicum* spp. in a randomized complete block design with two replications during the period of *yala* 2015 and *maha* 2015/16 at the field of Field Crops Research and Development Institute, Mahailuppallama. Fourteen morphological characters including days to 50 % flowering, plant height, plant breath, mature leaf length, mature leaf width, pod length, pod girth, pericarp thickness, number of primary branches per plant, number of secondary branches per plant, number of pods per plant, individual pod weight, number of seeds per pod, pod yield were scored and analysed using analysis of variance and multivariate methods. Analysis of variance revealed significant differences among genotypes for the most of tested traits. Pearson correlation coefficient showed significant positive correlation with most of morphological traits considered. Principal component analysis revealed that the four components explained more than 75% of total variation among genotypes. A total of six groups were defined through cluster analysis including each three main and minor groups. Distinct genetic variation was observed among these groups. Score plot of first two components score for genotypes confirmed the result of cluster analysis. Results of the study with respective to quantitative traits such as plant breath, leaf characters, pod length, pod width, pericarp thickness, pod weight, secondary branches per plant and yield will help for identifying materials and according to breeding objectives diversity of chilli genotypes can be utilised for future chilli improvement programs.

Keywords: Capsicum, Genotypes, Genetic diversity, Morphological characters, Yield

INTRODUCTION

Chilli (*Capsicum* Spp.) is an important cash crop valued for its taste, aroma and pungency. A wide range of variability reportedly exists in this crop (Nandi, 1992; Munshi and Behera, 2000). This variability can be utilized for crop improvement programs. Chilli peppers originated in South and Central America, where they were used by the native inhabitants for thousands of years. There is archaeological evidence at sites located in south-western Ecuador that chillies peppers were domesticated more than 6000 years ago. It is a diploid ($2n=24$) species and genetically self pollinated plant. However, 2 to 96% out-crossing was observed under open pollination (Hasanuzzaman, *et al.*, 2012). There are approximately 25 species within the genus *Capsicum*, five of which have been domesticated. The most widely grown and economically important species is *Capsicum annuum* which has Mexico as the major centre of genetic diversity. The four other domesticated species are *C. baccatum*, *C. pubescens*, *C. chinense* and *C. frutescence*. All originated in the tropical America (Andrew, 1995). Classification of *capsicums* below the species level is largely based on fruit types and uses. A wide diversity exists in fruit shapes like morphological traits in chilli both within and between species. Germplasm evaluation is done for identifying lines with desirable traits to use as parents if other characters are desirable (Dharmasena, *et al.*, 2003). Study of genetic diversity within available genetic resources is essential to understand the evolutionary and genetic relationship among accessions. Selection of breeding material depends on a number of morphological characters such as leaf characters, pod characters, number of branches and yield potential *etc.* Characterized and evaluated genotypes are essential for an effective breeding program to incorporate desired traits. If the variability in the population is largely due to genetic cause with less environmental effect the probability of isolating superior genotypes is high. Indeed knowledge of the nature and magnitude of relationship of the various characters is important in making selection in crops. Germplasm resource contains unique traits/genes that can be utilized for further crop improvement. Exploration, collection and evaluation of germplasm are the quickest and simplest method for acquiring the desired one for future breeding programmes. To realize this objective the estimation of phenotypic diversity was made based on quantitative traits of 38 germplams of *Capsicum* using multi variate analysis.

MATERIALS AND METHOD

This experiment was conducted in Field Crops Research and Development Institute, Mahailuppallama during *yala* 2015 and *maha* 2015/16. A total thirty eight germplasm of *Capsicum* species including twenty six of *Capsicum annuum*, nine of *Capsicum chinense* and three of *Capsicum frutescence* were evaluated in this study to understand the genetic variation among the genotypes. The experiment was laid out in Randomized Complete Block Design (RCBD) with two replicates.. The unit plot size was 6m x 1.8 m consisting of 3 rows with spacing 60 cm x 45 cm. There were 78 plants per plot, under irrigated conditions per Department of Agriculture management recommendations. Thirty days old seedlings were transplanted in the experimental plots.

Data were collected on five randomly selected plants from each treatment. Fourteen morphological characters were recorded of number of days to 50% flowering, plant height (cm), plant breadth (cm), mature leaf length (cm), mature leaf width (cm), pod length (cm), pod girth (cm), pericarp thickness (mm), number of pods per plant, individual pod weight (g), number of seeds per pod, pod yield (t/ha). Five fruits were selected randomly in each plot for recording observations on pod characters such as pod length, pod girth, pericarp thickness and individual pod weight. Morphological characters were recorded at second harvest except number of days to 50% flowering and pod yield.

Statistical analysis

Analysis of variance (ANOVA) was performed to test variations among genotypes accounting fourteen morphological traits using SAS 9.1 statistical package. Data for all tested morphological traits were checked as they fitted to normal distribution before performing ANOVA. Multivariate data analytical methods viz. principal component and cluster analysis were done to derive principal components and to construct phenotypic tree for the genotypes. So that similar genotypes can be classified into one group and dissimilar genotypes into distinct groups. Pearson correlation coefficient was also derived among quantitative traits of chilli germplasm. Statistical soft ware package MINITAB version 17 was used for the analysis of data.

78 **Table1. Sources and morphological characters of tested 38 capsicum genotypes**

No	Source	Origin	Characters
1	MICH3	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, dark green pods
2	MI2	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour lanceolate leaves, white pendent flower with exerted stigma, dark green pods with blunt end, large number of seeds per pod, higher pod weight, comparatively higher pungency
3	KA2	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
4	MI Green	Sri Lanka	Green colour stem, intermediate plant growth habit, nodal anthocyanin present, green colour ovate leaves, white pendent flower with exerted stigma, green pods
5	Arunalu	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, upward green pods
6	Galkiriyagama sel.	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour lanceolate leaves, white pendent flower with exerted stigma, green pods

7	985.3	AVRDC	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with same level stigma, dark green pods
8	Hene miris	Sri Lanka	Green colour stem, erect plant growth habit, green colour ovate leaves, nodal anthocyanin present, white pendent flower with exerted stigma, green pods
9	IR	Indonesia	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, dark green pods
10	Ruhunu miris	Sri Lanka	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, dark green pods, different pod shape
11	PC1	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, white pendent flower with exerted stigma, green pods, different conical pod shape
12	Waraniya Purple	Sri Lanka	Green colour stem, erect plant growth habit, purple dark green colour ovate leaves, whitish purple pendent flower with exerted stigma, dark purple wrinkle long pods,

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13	Jaffna Selection	Sri Lanka	Green colour stem, erect plant growth habit, green colour ovate leaves, nodal anthocyanin present, white pendent flower with exerted stigma, green pods
14	Waraniya Green	Sri Lanka	Green ovate colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
15	MI Waraniya 1	Sri Lanka	Green colour stem, erect plant growth habit, light green colour ovate leaves, white pendent flower with exerted stigma,, yellowish green wrinkle long pods, heavy plant growth, long plant duration
16	CAH36	ICPN	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
17	MI HOT	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour lanceolate leaves, white pendent flower with same level stigma, green pods
18	PBC380	Malaysia	Green colour stem, intermediate plant growth habit, green colour lanceolate leaves, white pendent flower with same level stigma, green pods

19	Hot Beauty	AVRDC	Green colour stem, erect plant growth habit, green colour lanceolate leaves, white pendent flower with exerted stigma, dark green long pods
20	Nut horse	Sri Lanka	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma green pods
21	ICPN 987.5	ICPN	Green colour stem, intermediate plant growth habit, green colour ovate leaves, white pendent flower with same level stigma, purple colour appear in flowers, yellow long smooth pods
22	Bhutan1	Bhutan	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
23	Bhutan 3	Bhutan	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
24	Bhutan 4	Bhutan	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods

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25	Bhutan 5	Bhutan	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
26	Bhutan 8	Bhutan	Green colour stem, erect plant growth habit, green colour ovate leaves, white pendent flower with exerted stigma, green pods
27	Kochchi 4	Sri Lanka	Green colour stem, erect plant growth habit, green colour leaves, greenish white pendent flower with exerted stigma, short conical shape dark green upward pods
28	Kochehi 5	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, greenish white pendent flower with exerted stigma, green colour intermediate size upward pods
29	Kochchi 6	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, yellow-green erect flower with exerted stigma, short conical shape butter white upward pods
30	Kochchi 7	Sri Lanka	Purple colour stem, dark purple nodal anthocyanin, erect plant growth habit, purplish dark green colour deltoid leaves, purple pendant flower with exerted stigma, purple colour calyx, dark purple irregular shape pods.
31	Kochehi 8	Sri Lanka	Green colour stem with purple strips, erect plant growth habit, green colour ovate leaves, light yellow to white pendent flower with same level stigma, green irregular shape pods

32	Kochchi 9	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, greenish white pendent flower with exerted stigma, green irregular shape pods
33	Kochchi10	Sri Lanka	Green colour stem, stems with nodal anthocyanin, erect plant growth habit, green colour deltoid leaves, yellow-green pendant flower with same level stigma, presence of calyx pigment, whitish purple conical shape intermediate pods
34	Kochchi11	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, greenish white pendent flower with exerted stigma, light and dark green colour conical shape intermediate pods
35	Kochchi12	Sri Lanka	Green colour stem with purple strips, stems with nodal anthocyanin, erect plant growth habit, green Green colour deltoid leaves, yellow erect flower with exerted stigma, green colour irregular shape pods, pods turn to orange to red when ripening

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36	Kochchi13	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, greenish white pendent flower with exerted stigma, small green colour upward pods
37	Kochchi14	Sri Lanka	Green colour stem, erect plant growth habit, green colour deltoid leaves, greenish white pendent flower with exerted stigma, irregular shape light green colour pods
38	Kochchi15	Sri Lanka	Green colour stem with purple strips, some stems with nodal anthocyanin erect plant growth habit, green colour deltoid leaves, yellowish white pendant flower with exerted stigma, green colour rounded pods with deep furrows

RESULTS AND DISCUSSION

Thirty eight genotypes with fourteen morphological traits, the highest variation was observed for pods per plant followed by seeds per pod and days to 50% flowering. Moderate variation was observed for plant height and plant breadth. Lowest variation was observed for pericarp thickness of the pods (Table 2).

Table 2. Mean, standard error (SE) range, minimum, maximum values, F value and CV% of tested traits of 38 capsicum genotypes.

Traits	Mean		SE	Minimum	Maximum	F value	CV (%)
DTF	81.1	±	1.2	68	114	9.39*	0.92
LL	7.8	±	0.3	4.46	14.9	2.34	2.89
LW	3.8	±	0.2	1.56	9	0.88	6.14
PLH	36.4	±	0.9	20.6	53.2	8.8*	5.4
PLB	28.9	±	0.8	15.6	48	5.22*	4.23
PODL	6.8	±	0.3	2.36	13.8	0.09	1.26
POD W	1.4	±	0.1	0.59	3.1	0.01	6.35
P.THIC	1.3	±	0.03	0.9	2.1	0.14	2.35
P.BR	5.7	±	0.2	3	12	.	0
S.BR	5.6	±	0.2	4	10	1	4.05
POD/P	49.1	±	3.1	22	140	30.77*	1.47
PWT	2.1	±	0.1	0.5	3.4	19.13*	2.24
S/POD	35.6	±	1.6	17	50	31.19*	1.22
YD	4.2	±	0.2	0.6	8.5	31.19*	1.22

Note: * significant difference at 0.05 % probability level

Note: DTF = Days to 50 % flowering, LL = Mature leaf length (cm), LW= Mature leaf width (cm), PLH = Plant height at second pick (cm), PLB= Plant breadth at second pick (cm), PODL= Pod length (cm), POD W= Pod width(cm), P.THIC= Pericarp thickness (mm), P.BR= number of primary branches per plant, S.BR= number of secondary branches per plant, POD/Pod = Number of pods per plant, PWT= Pod weight, S/POD = number of seeds per pod, pod yield (t/ha)

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Behaviour of each morphological character among *Capsicum*

genotypes

Analysis of variance revealed significant differences at 0.05% probability level among different genotypes for days to 50 % flowering, plant height, plant breadth, number of pods per plant, individual pod weight, seeds per pod and yield scored (Table 2).

Identifying grouping of variables

Principal component analysis quantifies each trait to calculate the Principal Components (PCs) which help in describing grouping of variables. The Table 3 shows that principal components, related factors and eigen values. The first principal component (PC1) is related to mature leaf length, leaf width, pod length, pod weight and yield (Table 3). It explained 39% of total variability. The second principal component (PC2) is related to pod width, pericarp thickness and pods per plant explained 15% of total variability. The third principal component (PC3) is related to morphological characters such as number of primary branches, number of pods per plant and yield. These four components explained more than 75% of total variability among the chilli genotypes evaluated.

Leaf characters (length and width), pod length, pod width and yield were recorded higher magnitudes (above 0.35) for the PC1. Leaf characters were positively correlated with PC1. Furthermore, pod width, pericarp thickness were negatively correlated with high magnitudes for PC2 while, pods per plant were recorded high positive magnitude for PC2.

Table 3. Principal Components (PCs) for 14 morphological traits in capsicum genotypes

Traits	PC1	PC2	PC3	PC4	PC5
DTF	0.299	0.105	0.035	-0.045	-0.253
LL	0.372	-0.211	-0.065	0.072	-0.161
LW	0.392	-0.183	-0.038	0.029	-0.191
PLH	0.156	0.189	0.116	-0.603	0.395
PLB	0.238	0.07	0.069	-0.567	0.099
POD L	-0.353	-0.171	-0.204	-0.208	-0.057
POD W	0.184	-0.545	0.002	0.046	0.027
P.THIK	0.07	-0.503	-0.271	-0.302	-0.032
P.BR	0.066	-0.168	-0.523	0.163	0.446
S.BR	0.269	0.137	-0.253	0.131	0.421
POD/P	0.279	0.322	-0.362	0.075	-0.077
PWT	-0.341	-0.234	-0.095	-0.25	-0.089
S/POD	0.007	0.249	-0.503	-0.24	-0.541
YD	-0.321	0.154	-0.358	0.02	0.141
Eigen value	5.503	2.131	1.591	1.328	0.982
Proportion	0.393	0.152	0.114	0.095	0.07
Cumulative	0.393	0.545	0.659	0.754	0.824

Genetic diversity of Capsicum genotypes

The dendrogram derived based on Pearson distance in Figure 1 displays the relative positions of chilli genotypes scored on morphological traits. At 40 similarity level there were six clusters with each three main clusters and minor clusters. Germplasm of *Capsicum annuum* which having local or foreign origin was grouped in I, II and III clusters. In first cluster variety (2) MI2 stayed separately. In second cluster (19) Hot Beauty having foreign origin stayed distantly to others. At this similarity level, (15) MI Waraniya1 and (21) ICPN 987.5 lines were grouped together which has same morphological characters like long pod character. *Capsicum chinense* and *Capsicum frutescence* lines were clustered separately at cluster IV and V. The line of kochchi 13 which belongs to *Capsicum frutescence* was stayed separately at cluster VI. It has shown unique characteristic and genetically distance to others.

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The dendrogram showed that genetic distinctness of parents. Genetic diversity within closely located genotypes is lower than that of distantly located genotypes. The distance parents would be assisted to different genetic constitution that can be utilized for future breeding programmes. In intra-specific crosses of *Capsicum annuum*, germplasm of cluster I (MICH 3, Galkiriyagama selection and MI2) could be used with germplasm of cluster II (KA2, Arunalu, MI Hot etc.) and cluster III (MI Waraniya1 and ICPN 987.5 lines). For inter-specific crosses the germplasm of cluster I, II and III which belong to *Capsicum annuum* could be combined with germplasm of cluster IV, V and VI which belong to *C. chinense* and *C. frutescence* to have new genetic makeup.

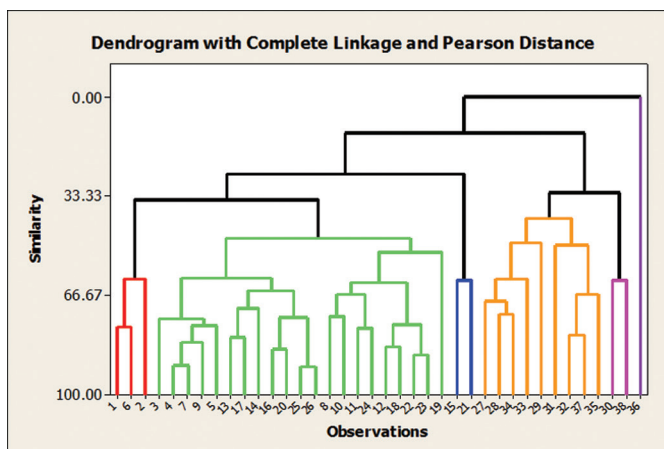


Figure1. Dendrogram based on 14 morphological characters in 38 chilli genotypes

Table 4. Trait mean of six clusters in 38 chilli genotypes

Group	No of geno	DTF	LL	LW	PLH	PLB	PODL	PODW	P.THICK	P.BR	S.BR	POD/P	PWT	S/ YD
														(t/ ha)
1	3	74	6	2.1	32.1	27.6	6.7	0.9	1.1	7	7	75	1.8	46
2	21	76	6.2	2.5	35.7	26.9	7.9	1.2	1.2	5	5	32	2.5	33
3	2	78	6.7	3	34.5	25.1	13.1	1.6	1.9	6	5	47	4.32	48
4	9	94	10.8	6.5	40.8	32.8	3.6	1.8	1.4	6	8	74	0.91	27
5	2	76	13.4	8.1	24.8	28.7	4.5	2.8	1.6	8	4	45	1.52	41
6	1	106	10.6	6.6	49.5	48.2	2.1	0.6	0.9	5	6	111	0.45	97
														2.03

Note: No of geno.=No of genotype, DTF=Days to 50 % flowering, LL= Mature leaf length (cm), LW= Mature leaf width(cm), PLH=Plant height at second pick (cm), PLB= Plant breath at second pick (cm),PODL= Pod length (cm), POD W= Pod width (cm), P.THICK= Pericarp thickness (mm), P.BR = number of primary branches per plant,S.BR= number of secondary branches per plant, POD/Pod = Number of pods per plant, PWT= Pod weight, S/POD= number of seeds per pod, pod yield (t/ha)

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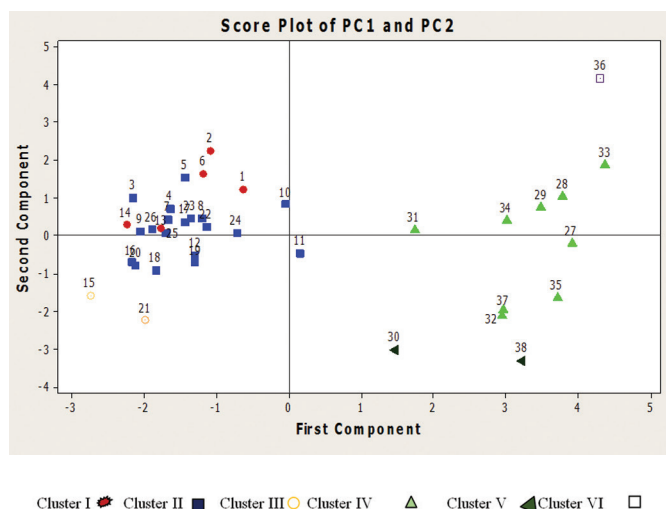


Figure 2. Score plot of PC1 and PC2 for 38 chilli genotypes

Score Plot Analysis

The major principal components that are first two accounting 54% of the variance were plotted to observe the relationship between clusters (Fig. 2). All genotypes in cluster I were grouped together in score plot. Germplasm of cluster II were closely stayed in the score plot except variety PC 1(11). Genotypes of cluster IV showed close relationship. Genotypes of minor cluster of III *i.e* MI Waraniya 1 (15) and ICPN 985.7 (21) were arranged closely. As same minor clusters of V *i.e* Kochchi 7 (30) and Kochchi 15 (38) were also arranged closely. Results of score plot confirm the genotype pattern that is found in cluster analysis. This also implies the potential for utilizing such variability in crop improvement programmes. Germplasm resource contains unique traits/genes that can be utilized for further crop improvement. Exploration for collection of germplasm is the quickest and simplest method for acquiring the desired one.

Relationship between variables

In Table 5, Pearson correlation coefficient among the measured traits of capsicum genotypes are presented. There were significant positive correlations shown between leaf related characters, plant breath, pod width, pericarp thickness, secondary branches

per plant, weight per pod and yield with most of morphological traits. Yield is complex character influenced by a number of other component characters (Jose and Khader, 2002). Weight per fruit and number of fruits per plant are the most important components of yield. Thus, during selection there should be high consideration on those characters. Other yield components to improve yield are plant width, fruit length, fruit diameter, days to fruit set and fruit density. (Ahamed and Hurra, 2000). Higher phenotypic and genotypic coefficients of variation were observed for leaf area, fruits per plant, fruit weight, fruit length, fruit girth and yield per plant. High heritability coupled with high genetic advance observed for these characters imply the potential for crop improvement through selection.

CONCLUSIONS

Principal Component analysis and cluster analysis given comparable results for thirty eight chilli germplasm and plotting of score plot assisted to confirm the results. Therefore, Estimation diversity with respective to quantitative traits such as plant breath, leaf characters, pod length, pod width, pericarp thickness, pod weight, secondary branches per plant and yield will help for identifying parental materials. High genetic variation is created with more genetic distance parents in order to achieve maximum heterosis. Therefore, according to breeding objectives diversity of available chilli genotypes can be utilised for future improvement program.

92 Table 5. Correlation matrix among the measured traits in capsicum genotypes

	DTF	LL	LW	PLH	PLB	PODL	PODW	P.THIK	P.BR	S.BR	POD/P	PWT	S/ POD
DTF													
LL	0.49 *												
LW	0.56 *	0.97 *											
PLH	0.26	0.16	0.19										
PLB	0.36 *	0.34 *	0.42 *	0.59 *									
PODL	-0.58 *	-0.6 *	-0.66 *	-0.24	-0.43 *								
PODW	0.21	0.58 *	0.57 *	0.11	0.21	-0.22							
P.THIK	0.03	0.32 *	0.36 *	0.01	0.15	0.25	0.53 *						
P.BR	-0.01	0.21	0.14	-0.03	-0.04	-0.03	0.29	0.22					
S.BR	0.34 *	0.44 *	0.47 *	0.25	0.24	-0.48 *	0.09	0.09	0.27				
POD/P	0.54 *	0.47 *	0.52 *	0.21	0.28	-0.51 *	-0.09	-0.04	0.17	0.67 *			
PWT	0.48 *	0.59 *	0.64 *	-0.22	0.36 *	0.86 *	-0.89	0.23	-0.07	0.54 *	0.63 *		
S/POD	0.09	0.04	0.05	0.01	0.15	0.1	0.28	-0.01	0.11	-0.01	0.4 *	0.04	
YD	0.52	0.72	0.78	0.28	0.36 *	0.63 *	0.39 *	0.15	0.14	-0.25	0.15	0.53 *	0.24

Note: * significant difference at 0.05 % probability level.

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