

Combining ability analysis of yield and late blight [*Phytophthora infestans* (Mont.) de Bary] resistance of potato germplasm in Rwanda

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Abstract

Designed crosses among complementary parents and combining ability analysis are essential to recombine traits of economic importance and to select novel genotypes. The objective of this study was to estimate combining ability effects for yield and yield related traits and late blight resistance in potato. Crosses were performed using a 10 × 10 half diallel mating design to generate 45 F₁s. Only 28 families with sufficient individuals and eight parents were field evaluated in experiments laid out in a 6 × 6 lattice design with two replications across two sites (Kinigi and Nyamagabe) in Rwanda. Late blight resistance was estimated using the relative area under the disease progress curve (rAUDPC: 100 % max). Furthermore, data on total tuber yield, total tuber number, and average tuber weight were collected and subjected to combining ability analyses. Results showed that additive gene action and non-additive gene action were present affecting yield and late blight resistance in potato. Additive gene action was predominant over non-additive gene action for both traits. All the families selected for further evaluation showed improved levels of late blight resistance and high yields. The study identified ten top families with high tuber yield and resistance to late blight for further evaluation and release.

Keywords: Combining ability, gene action, late blight, potato, Rwanda.

Abbreviations: rAUDPC: relative area under the disease progress curve.

Introduction

Potato (*Solanum tuberosum* L.) is a staple food crop grown in 149 countries across the world (Hijmans, 2001). It is one of the four most important food crops along with rice (*Oryza sativa*), wheat (*Triticum aestivum* L.), and maize (*Zea mays* L.) (Lang, 2001). Potato breeding towards development of new cultivars involves sexual recombination to generate genetic variation and select novel recombinants using agromorphological traits (Acquaah, 2007). Segregating F₁ progeny from which to select superior clones contains suitable genetic constitution from its parents. It is important to understand the mode of gene action involved in the expression of important traits. Understanding gene action will help in selecting suitable parents and segregates in a breeding programme (Falconer and Mackay, 1996). Combining ability analysis is the basis for identification of the best parents and their crosses (Mondal and Hossain, 2006). The general combining ability (GCA) gives an indication on the average contribution of a parent to its progeny; it provides an estimation of the parental gametic contribution to its offspring by the mean performance of the progeny (Bradshaw and Mackay, 1994; Falconer and Mackay, 1996). The specific combining ability (SCA) is the deviation from the progeny mean from the expected on the basis of GCA (Bradshaw and Mackay, 1994). In this case, the performance

of the progeny is either superior or inferior to the parents. Yield and late blight disease field resistance are quantitatively inherited (Falconer and Mackay, 1996). In quantitative inheritance many genes are involved, each contributing a small effect to the phenotypic expression of the character concerned. In the literature, it has been reported that both GCA and SCA effects are significant for potato yield and late blight resistance with slight differences in magnitude across experiments (Bradshaw and Mackay, 1994; Gopal et al., 2008). Potato genotypes with relatively high yield level and resistance to the late blight disease are being developed by the International Potato Centre (CIP) (Landeo et al., 1997). These genotypes are frequently introduced to developing countries including Rwanda to strengthen the existing potato genetic resources to boost productivity and control potato late blight disease. Subsequently, the national potato programme of the Rwanda Agriculture Board (RAB) is continuously evaluating and screening the CIP genetic stocks and locally adapted genotypes under target growing environmental conditions to identify clones with high yield and late blight resistance. Consequently, a number of best genotypes were identified to be used as parents in subsequent crosses and selection (Muhinyuza et al., 2014). The objectives of this study were to estimate combining ability

effects for yield and yield related traits and late blight resistance. The selected best parental genotypes or families will be used for further breeding, evaluation and release in Rwanda.

Results

Analysis of variance

Table 2 summarizes the combined analysis of variance for the relative area under the disease progress curve (rAUDPC), total tuber weight (TTW), total tuber number (TTN) and average tuber weight (ATW) among the families. There were significant differences ($P \leq 0.05$) among families for rAUDPC, TTW and ATW. The environmental (site) effect was highly significant ($P \leq 0.001$) for TTW and TTN, and significant for ATW. The interaction between families x sites had high significant ($P \leq 0.001$) effects for rAUDPC and TTW.

Family means within and across locations

Means and ranking of rAUDPC, TTW, TTN and ATW across locations are presented in Table 3. The mean rAUDPC (= 100 max) across locations indicated that family Gikungu x CIP 391047.34 with disease severity of 8.1% and family Kigega x CIP 393036.201 (9.5%) were the most resistant, whereas families Kigega x Ngunda (41%) and Nderera x CIP 393280.82 (34.4%) were the most susceptible. Families Gikungu x CIP 391047.34 and CIP 393280.82 x CIP 391047.34 with TTW of 34.9 t/ha and 30.4 t/ha, respectively, were the best yielders, whereas family Gikungu x Nderera at 16.5 t/ha and Nderera x Ngunda (20.0 t/ha) were the lowest in TTW across locations. Families Gikungu x CIP 391047.34 with an average of 7.3 in TTN and Gikungu x Nderera (3.8) were the highest and lowest in TTN across locations, respectively. Families Kigega x Gikungu and Ngunda x CIP 393280.82 with ATW of 79.7 g and 77.9 g respectively were the highest in ATW, while families CIP 393036.201 x CIP 393385.39 (53.9 g) and Gikungu x CIP 393280.82 (55.7 g) were the lowest across locations in ATW. The ranking showed that the 10 most resistant potato families were Gikungu x CIP 391047.34 (rAUDPC of 8.1%), Kigega x CIP 393036.201 (9.5%), Kigega x CIP 393036.82 (10.3%), Gikungu x Ngunda (11.2%), Gikungu x CIP 393385.39 (13.9%), Gikungu x Nderera (14.2%), CIP 393036.201 x CIP 393385.39 (14.7%), CIP 393280.82 x CIP 391047.34 (16.4%), Nderera x CIP 393036.201 (20.4%), Ngunda x CIP 393280.82 (21.1%); while the 10 most high yielder potato families were Gikungu x CIP 391047.34 (34.9 t/ha), CIP 393280.82 x CIP 391047.34 (30.4 t/ha), Kigega x CIP 393280.82 (30 t/ha), Gikungu x Ngunda (29.2 t/ha), Ngunda x CIP 393280.82 (29.1 t/ha), Kigega x Gikungu (28.4 t/ha), Ngunda x CIP 391047.34 (28.4 t/ha), Kigega x CIP 393036.201 (26.8 t/ha) and Kigega x Ngunda (26.7 t/ha). The most resistant potato families were almost the high yielding ones.

General and specific combining ability variances of potato yield and yield related traits and late blight resistance at Kinigi

General and specific combining ability variances for the four traits at Kinigi are presented in Table 4. There were highly significant differences ($P \leq 0.001$) among families for rAUDPC and TTW. GCA was highly significantly ($P < 0.001$) different for TTW and significant ($P < 0.05$) for ATW. The SCA was highly significantly ($P < 0.01$) different for rAUDPC

and TTW. The ratio GCA/SCA provided an estimate of the relative importance of additive and non-additive gene effects in the expression of the traits assessed. GCA was more important than SCA in the expression of TTW and ATW; whereas the opposite was true for rAUDPC and TTN at Kinigi.

General combining ability effects of potato parents at Kinigi

General combining ability effects of parents at Kinigi are presented in Table 5. Parent Kigega had the highest GCA effects for TTW (4.2) and ATW (7.15) followed by parent 391047.34 with TTW (1.1) and ATW (1.42) while parents Nderera with TTW (-2.09) and Gikungu with ATW (-3.27) the lowest GCA effects for TTW and ATW respectively. Kigega had the lowest GCA effects for rAUDPC (-1.99) followed by parent 393385.39 (-1.93). Gikungu had the highest GCA effects for TTN while parent 393385.39 (-0.34) had the lowest.

Specific combining ability effects of potato families at Kinigi

Table 6 summarises specific combining ability effects of potato families at Kinigi. The best and positive SCA effects of 10.22 and 8.75 for TTW traits were recorded in families Gikungu x CIP 393036.201 and Kigega x Ngunda respectively. The lowest SCA effects of -13.32 and -10.70 for rAUDPC were observed in families Gikungu x CIP 393036.201 and Kigega x Nderera respectively.

General and specific combining ability variances of potato yield and yield related traits and late blight resistance at Nyamagabe

General and specific combining ability variances of potato tuber weight, relative area under the disease progress curve, total tuber number and average tuber weight at Nyamagabe are presented in Table 7. There were highly significant differences ($P \leq 0.001$) among families for all the selected traits. GCA and SCA were significantly different ($P < 0.05$) for rAUDPC, TTW, TTN and ATW. The GCA/SCA ratio provided an estimate of the relative importance of additive and non-additive gene effects in the expression of the traits assessed. GCA was more important than SCA in the expression of all traits at Nyamagabe.

General combining ability effects of potato parents at Nyamagabe

General combining ability effects of potato parents at Nyamagabe are presented in Table 8. Parent CIP 396036.201 had the highest GCA effect (2.25) for TTW followed by parent CIP 391047.34 (0.91) while parent Gikungu (-2.36) had the lowest GCA effects for the same trait. Nderera had the lowest GCA effects for rAUDPC (-2.55) followed by parents 393385.82 (-1.81) and Gikungu (1.79). Parent CIP 391047.34 had the highest GCA effects for TTN while parents Kigega (-0.21) and Gikungu (-0.21) had the lowest. Parents CIP 393280.82 (1.69) and CIP 396036.201 (1.59) had the highest GCA effects for ATW.

Specific combining ability effects of 28 potato families at Nyamagabe

Specific combining ability effects of 28 potato families at Nyamagabe are summarised in Table 9. The families

Table 1. Description of the 10 potato parents used in the crosses.

Parents	Source	Yield (t/ha)	Reaction to late blight	Flowering ability and pollen production
		Mean	rAUDPC (%)	
CIP 391047.34	CIP	37.4	24.1	Excellent
CIP 393371.58	CIP	50.9	21.6	Good
CIP 393385.39	CIP	27.2	30.1	Excellent
CIP 393280.82	CIP	34.2	12.5	Excellent
CIP 393036.201	CIP	30.1	33.5	Excellent
Gikungu	CIP	17.1	11.1	Excellent
Kigega	Rwanda	33.9	18.8	Excellent
Nderera	Rwanda	21.5	25.4	Excellent
Ngunda	Rwanda	33	19.3	Excellent
Kirundo	Rwanda	28.6	27.6	Good

CIP = International Potato Center; rAUDPC = relative area under the disease progress curve.
Source: Muhinyuza et al., 2015

Table 2. Combined analysis of variance of potato for late blight resistance, tuber yield and related traits at Kinigi and Nyamagabe in Rwanda.

Source of variation	DF	Mean squares			
		rAUDPC	TTW	TTN	ATW
Families	35	246.8***	61.9***	2.9 NS	133.3**
Sites	1	26 NS	313.9***	46.7***	735.8**
Rep (sites)	2	8.2 NS	5.1 NS	5 NS	9.9 NS
Families x sites	35	87.3***	32.9***	1.9 NS	82.8 NS
Error	70	27.9	9.6	2	53.9
Total	143				

Significance levels: * $p \leq 0.05$; ** $p \leq 0.01$; *** $p \leq 0.001$; NS: non-significant $p > 0.05$; DF: degrees of freedom; TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW = average tuber weight.

Gikungu x CIP 393036.201 (10.32) and Kigega x Nderera (5.58) had the best and positive SCA effects for the trait TTW while the families Kigega x CIP 393036.201 (-14.16), Kigega x Nderera (-12.92) and Gikungu x CIP 393036.201 (-10.82) had the lowest and negative SCA effects for the trait rAUDPC. Ten potato families with reduced SCA effects of RAUDPC were Kigega x CIP 393036.201 (-14.16), Kigega x Nderera, (-12.92) Gikungu x CIP 393036.201 (-10.82), Kigega x CIP 393385.39 (-10.66), CIP 393036.201 x CIP 391047.34 (-8.92), CIP 391047.34 x CIP 393385.39 (-8.67), Nderera x CIP 393385.39 (-8.47), CIP 393036.201 x CIP 393385.39 (7.15), Gikungu x CIP 391047.34 (-5.03) and Ngunda x CIP 393280.82 (-4.94), which is in a desirable direction for resistance breeding.

Correlation between traits

Correlations between the four traits are presented in Table 10. Correlation between TTW and rAUDPC was significant ($p \leq 0.05$) and negative (-0.39) across locations, and very significant ($p \leq 0.01$) and negative (-0.52) at Nyamagabe; whereas TTW and TTN had a highly significant ($p \leq 0.001$) positive (0.66; 0.59) correlation at Nyamagabe and across locations. Correlation between TTN and ATW was significant ($p \leq 0.05$) and negative (-0.37) at Kinigi; whereas correlation between rAUDPC and TTN was very significant ($p \leq 0.01$) and negative (-0.47) at Nyamagabe.

Discussion

The significant mean squares for families on rAUDPC and TTW indicated the presence of genetic variation among the parents and their crosses. This suggests that genotypes resistant to potato late blight with high yield may be selected. In addition, the significant environmental (site) effect on the traits observed justifies multi-locational testing of varieties prior to their release. The significant GCA and SCA mean squares of the traits observed shows that both additive and non-additive gene action were involved in the expression of

the traits. The GCA to SCA ratios ranged from 0.41-0.80 and 0.54-0.70, respectively at Kinigi and Nyamagabe for tuber yield and yield related traits, indicating that additive gene action was predominant. Contradictory results were reported in the literature. Some authors found both GCA and SCA to be significant for potato yield with GCA being less important in magnitude than SCA (Bradshaw and Mackay, 1994; Gopal et al., 2008; Haydar et al., 2009; Ortiz and Golmirzaie, 2004; Ruiz de Galarreta et al., 2006); whereas other authors (Plaisted et al. 1962; Tai, 1976; Killick, 1997; Gopal, 1998) reported GCA to be more important in magnitude than SCA in affecting potato yield. Some other authors reported GCA to be significantly greater than SCA for tuber yields and yield related traits in crosses between non-related parents (Neele et al., 1991; Ortiz and Golmirzaie, 2004). This would be the case found in this study since the clones evaluated were local landraces and CIP derived genetic materials. Muhinyuza et al. (2014) identified different and non-related parents used for crosses in the course of this study. The GCA/SCA ratios were 0.41 and 0.69 for rAUDPC at Kinigi and Nyamagabe respectively; indicating that both GCA and SCA are more or less equally important in the expression of late blight resistance in potatoes. This agrees with previous reports on the relative importance of GCA and SCA for potato late blight resistance: both GCA and SCA have been reported to be significant (Bradshaw and Mackay, 1994). This imply both additive and non-additive gene action are important in conditioning resistance to late blight in potato (Bradshaw and Mackay, 1994). Landeo et al. (2000) found both additive and non-additive gene action equally important for horizontal resistance. In general many reports supported the present findings with slight predominance of additive over the non-additive gene action in inheritance of quantitative resistance to late blight (Wastie et al., 1991; Stewart et al., 1992; Kumar et al., 2007). Studies on family means and the positive correlation between TTW and MTN indicated that TTN had a significant influence on tuber yield in this study. Mehdi (2008) also found similar results where total tuber yield was mainly attributed to higher number of tubers per plant and

Table 3. Family and parent means of tuber weight, relative area under the disease progress curve, total tuber number and average tuber weight of 28 potato families when evaluated at two locations in Rwanda.

Families	Traits																				
	rAUDPC				TTW (t/ha)						TTN						ATW (g)				
	Kinigi		Nyamagabe		Across locations		Kinigi		Nyamagabe		Across locations		Kinigi		Nyamagabe		Across locations		Kinigi	Nyamagabe	Across locations
	Mean	Mean	Mean	Rank	Mean	Mean	Mean	Rank	Mean	Mean	Mean	Rank	Mean	Mean	Mean	Rank	Mean	Mean	Mean	Rank	
Kigega x Gikungu	16.7	28.2	22.5	19	30.3	26.5	28.4	9	5	5	5	5.0	25	83.7	75.6	79.7	1				
Kigega x Ngunda	39.8	42.1	41.0	36	34.7	18.7	26.7	14	7.5	4.5	6.0	11	63.6	63.9	63.8	27					
Kigega x Nderera	18.7	43.2	31.0	31	40.2	16.6	28.4	10	5.5	3	4.3	32	72.8	67	69.9	9					
Kigega x CIP 393036.201	10.1	8.8	9.5	2	25.1	28.5	26.8	13	5.5	5	5.3	19	65.3	63.4	64.4	25					
Kigega x CIP 393280.82	11.7	8.8	10.3	3	33.4	26.5	30.0	4	7.5	5	6.3	6	73.9	66.6	70.3	7					
Kigega x CIP 3910147.34	23.6	19.2	21.4	16	26	17.9	22.0	28	5	3.5	4.3	33	70.6	65.3	68.0	13					
Kigega x CIP 393385.39	26.9	29.0	28.0	30	33.7	15.9	24.8	22	6	6	6.0	12	73.2	72.4	72.8	5					
Gikungu x Ngunda	9.2	13.2	11.2	4	32.8	25.6	29.2	6	5	5	5.0	26	63.5	73.4	68.5	11					
Gikungu x Nderera	12.3	16.1	14.2	8	16.9	16	16.5	36	4	3.5	3.8	36	63.3	66.3	64.8	22					
Gikungu x CIP 393036.201	32.2	30.2	31.2	32	22.8	17.8	20.3	34	7.5	3.5	5.5	17	60.5	62.7	61.6	31					
Gikungu x CIP 393280.82	39.4	15.1	27.3	27	21.7	20.9	21.3	31	5	5.5	5.3	20	52.4	58.9	55.7	35					
Gikungu x CIP 3910147.34	7.7	8.4	8.1	1	35.3	34.4	34.9	1	7.5	7	7.3	1	60	64.4	62.2	30					
Gikungu x CIP 393385.39	13.1	14.6	13.9	6	25.6	24.2	24.9	21	7	5.5	6.3	7	55.7	73.5	64.6	24					
Ngunda x Nderera	30.7	14.4	22.6	20	27	21.9	24.5	24	7	3.5	5.3	21	60.4	79.9	70.2	8					
Ngunda x CIP 393036.201	16.7	26.2	21.5	17	26.5	24.2	25.4	19	8	5	6.5	3	67.1	67.2	67.2	16					
Ngunda x CIP 393280.82	12.8	29.3	21.1	15	30.3	27.8	29.1	7	5	4.5	4.8	29	78.1	77.6	77.9	3					
Ngunda x CIP 3910147.34	24.6	22.8	23.7	23	29.9	26.9	28.4	11	8.5	5	6.8	2	53.5	74.4	64.0	26					
Ngunda x CIP 393385.39	20.8	32.9	26.9	26	21.3	21.6	21.5	30	6.5	4	5.3	22	60.6	73.7	67.2	17					
Nderera x CIP 393036.201	18.1	22.7	20.4	14	24.8	28.4	26.6	15	7.5	5	6.3	8	57.2	61.4	59.3	33					
Nderera x CIP 393280.82	39.3	29.4	34.4	35	25.5	26.5	26.0	17	6.5	5	5.8	13	57.9	73.1	65.5	21					
Nderera x CIP 3910147.34	35.2	32.6	33.9	34	20.1	19.8	20.0	35	5	4	4.5	31	62	64.5	63.3	29					
Nderera x CIP 393385.39	19.9	30.6	25.3	24	21.6	20	20.8	32	4	4	4.0	35	77.3	65.7	71.5	6					
CIP 393036.201 x CIP 393280.82	29.4	17.8	23.6	22	26.0	26	26.0	18	7	5.5	6.3	9	62.5	65.1	63.8	28					
CIP 393036.201 x CIP 391047.34	27.1	24.2	25.7	25	20.8	20.8	20.8	33	6.5	4	5.3	23	65.7	71.9	68.8	10					
CIP 393036.201 x CIP 393385.39	14.5	14.8	14.7	9	26.5	26.5	26.5	16	6	6.5	6.3	10	51.4	56.4	53.9	36					
CIP 393032.82 x CIP 391047.34	21.9	10.9	16.4	11	30.4	30.4	30.4	2	6	5.5	5.8	14	56.9	72.5	64.7	23					
CIP 393032.80 x CIP 393385.39	27.6	36.8	32.2	33	20.4	23.9	22.2	27	4	4.5	4.3	34	65.6	70	67.8	15					
CIP 391047.34 x CIP 393385.39	22.1	21.7	21.9	18	21.9	21.9	21.9	29	6	4	5.0	27	58.5	77.3	67.9	14					
Parents																					
Kigega	17.0	13.4	15.2	10	30.4	30.4	30.4	3	6.5	6.5	6.5	4	63.6	73.1	68.4	12					
Gikungu	13.9	11.0	12.5	5	27	21.9	24.5	25	6	5.5	5.8	15	57.5	75.3	66.4	18					
Ngunda	19.1	17.6	18.4	13	31.9	26.8	29.4	5	6	4.5	5.3	24	72.4	76.9	74.7	4					
Nderera	26.2	28.5	27.4	28	28.6	28.6	28.6	8	7	6	6.5	5	63	69	66.0	19					
CIP 393036.201	19.0	16.8	17.9	12	24.6	24.6	24.6	23	5.5	5.5	5.5	18	57	64.3	60.7	32					
CIP 393280.82	21.2	25.5	23.4	21	29.4	26.1	27.8	12	5	4.5	4.8	30	78.1	79.3	78.7	2					
CIP 390147.34	15.8	12.1	14.0	7	25	25	25.0	20	4.5	7	5.8	16	67.4	48.4	57.9	34					
CIP 393385.39	19.7	35.7	27.7	29	24.9	22.2	23.6	26	5.5	4.5	5.0	28	63.6	68.3	66.0	20					
Mean	21.5	22.4	21.9		27.2	24.2	25.7		6	4.9	5.5		64.3	68.9	66.6						
CV (%)	28.1	19.6	24.1		10.3	13.9	12.1		30.3	18	26.3		14	7.3	11						
S.E.D.	5.7	3.9	5.2		2.9	2.7	3.1		2	0.8	1.4		10	5.4	7.3						

TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight.

Table 4. General and specific combining ability variances of potato tuber weight, relative area under the disease progress curve, total tuber number and average tuber weight based on an 8x8 half diallel mating design at Kinigi.

Source of variance	Traits				
	DF	rAUDPC	TTW	TTN	ATW
Families	35	148.6***	52.7***	2.8 NS	122.9 NS
GCA	7	29.3 NS	38.8***	0.6 NS	103.4*
SCA	28	85.5***	23.2***	1.7NS	50.9 NS
Error	35	18.4	3.9	1.7	41.0
CV%		28.1	10.3	30.2	14.0
GCA/SCA		0.41	0.77	0.41	0.80

Significance levels: *p≤0.05; ** p≤0.01; ***p≤0.001; NS = non-significant p>0.05; DF: degrees of freedom; TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight.

Table 5. General combining ability effects of potato parents at Kinigi.

Parents	General combining ability effects			
	rAUDPC	TTW	TTN	ATW
Kigega	-1.99*	4.20**	-0.23	7.15**
Gikungu	1.06	-1.70	0.31	-3.27*
Ngunda	2.11**	0.06	0.26	0.18
Nderera	1.32	-2.09*	-0.18	-1.69
CIP 396036.201	-1.57	-0.36	0.06	-1.12
CIP 393280.82	-0.63	-0.84	0.21	-0.65
CIP 391047.34	1.63	1.1	-0.09	1.42
CIP 393385.39	-1.93*	-0.37	-0.34	-2.02

TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight; * and ** denote significant differences at p=0.05 and p=0.01, respectively.

Table 6. Specific combining ability effects of 28 potato families at Kinigi.

Families	Specific combining ability effects			
	rAUDPC	TTW	TTN	ATW
Kigega x Gikungu	19.26**	5.01*	1.38*	-4.63
Kigega x Ngunda	-2.89	8.75**	-0.56	1.12
Kigega x Nderera	-10.70**	-4.19*	-0.11	-4.47
Kigega x CIP 393036.201	-6.21*	2.41	1.63	3.52
Kigega x CIP 393280.82	4.70	-4.49*	-0.01	-0.25
Kigega x CIP 3910147.34	5.79*	1.20	0.28	0.33
Kigega x CIP 393385.39	-8.40*	1.78	-0.46	-5.83
Gikungu x Ngunda	7.50*	-2.74*	0.88	-0.72
Gikungu x Nderera	15.49**	-1.63	-1.16	-7.01
Gikungu x CIP 393036.201	-13.32**	10.22**	1.08	0.04
Gikungu x CIP 393280.82	-8.86*	0.96	0.43	-4.74
Gikungu x CIP 3910147.34	6.53*	0.46	0.73	-2.05
Gikungu x CIP 393385.39	-3.91	1.38	1.98*	8.04*
Ngunda x Nderera	-0.35	4.75*	2.38**	-9.36*
Ngunda x CIP 393036.201	-1.22	-5.59**	0.13	-2.76
Ngunda x CIP 393280.82	-4.90	-1.55	0.98	-6.64
Ngunda x CIP 3910147.34	14.09**	-2.80*	0.28	-8.00*
Ngunda x CIP 393385.39	13.54**	-6.78**	-0.96	-0.51
Nderera x CIP 393036.201	8.17*	1.26	1.08	0.95
Nderera x CIP 393280.82	4.89	-3.44*	0.43	3.67
Nderera x CIP 3910147.34	-9.97**	0.30	0.23	-12.64**
Nderera x CIP 393385.39	0.98	5.73**	0.48	-3.75
CIP 393036.201 x CIP 393280.82	2.77	-4.04*	-0.31	-4.08
CIP 393036.201 x CIP 391047.34	-7.69*	2.51*	0.48	-1.05
CIP 393036.201 x CIP 393385.39	-1.03	5.28**	0.23	-3.71
CIP 393032.82 x CIP 391047.34	3.68	1.15	0.83	-2.12
CIP 393032.80 x CIP 393385.39	0.03	-1.37	-0.41	-4.63
CIP 391047.34 x CIP 393385.39	-5.38	-2.92	-1.11	3.70

TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight; * and ** denote significant differences at p=0.05 and p=0.01, respectively.

Table 7. General and specific combining ability variances of potato tuber weight, relative area under the disease progress curve, total tuber number and average tuber weight based on an 8x8 half diallel mating design at Nyamagabe.

Source of variance	Traits				
	DF	rAUDPC	TTW	TTN	ATW
Families	35	185.5***	42***	2.1***	93.2 ***
GCA	7	101.2***	20.9**	1.2**	29.5*
SCA	28	90.6***	21***	1.0**	50.9 ***
Error	35	9.6	5.7	0.4	12.9
CV%		19.6	13.9	18	7.4
GCA/SCA		0.69	0.66	0.70	0.54

Significance levels: *p≤0.05; ** p≤0.01; ***p≤0.001; TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight.

Table 8. General combining ability effects of potato parents at Nyamagabe.

Parents	General combining ability effects			
	rAUDPC	TTW	TTN	ATW
Kigega	1.94*	-1.52*	-0.21	0.35
Gikungu	-1.79*	-2.36*	-0.21	-1.66*
Ngunda	7.13**	-0.47	-0.56*	1.23
Nderera	-2.55*	0.20	0.09	-2.56*
CIP 396036.201	-1.36	2.25*	0.29*	1.59*
CIP 393280.82	-1.81*	0.22	-0.16	1.69*
CIP 391047.34	-1.09	0.91	0.53*	1.11
CIP 393385.39	-0.46	0.77	0.23	-1.75*

TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight; * and ** denote significant differences at p=0.05 and p=0.01, respectively

Table 9. Specific combining ability effects of 28 potato families at Nyamagabe.

Families	Specific combining ability effects			
	rAUDPC	TTW	TTN	ATW
Kigega x Gikungu	19.58**	-1.60	0.02	-3.67
Kigega x Ngunda	11.75**	-5.59*	-1.12*	-3.41
Kigega x Nderera	-12.92**	5.58*	0.22	-3.22
Kigega x CIP 393036.201	-14.16**	1.53	0.02	-4.17
Kigega x CIP 393280.82	-3.26*	-4.98*	-1.02*	-5.57*
Kigega x CIP 3910147.34	5.78*	-7.67**	0.77	2.06
Kigega x CIP 393385.39	-10.66**	2.11	0.07	5.93*
Gikungu x Ngunda	2.49	-3.55*	-0.62	-5.75*
Gikungu x Nderera	-2.88	-1.12	0.72	-5.71*
Gikungu x CIP 393036.201	-10.82**	10.32**	2.52*	-4.41
Gikungu x CIP 393280.82	-4.12*	2.15	0.97*	4.59*
Gikungu x CIP 3910147.34	-5.03*	-0.83	-1.72*	11.57**
Gikungu x CIP 393385.39	6.13*	1.55	0.07	1.79
Ngunda x Nderera	-4.11*	2.97	0.57	6.91*
Ngunda x CIP 393036.201	4.76*	-4.41*	-0.62	2.01
Ngunda x CIP 393280.82	-4.94*	4.46*	0.82*	-10.35**
Ngunda x CIP 3910147.34	1.04	1.87	0.12	1.88
Ngunda x CIP 393385.39	3.56*	-4.73*	-0.57	-3.85
Nderera x CIP 393036.201	-0.62	-0.68	0.22	-2.80
Nderera x CIP 393280.82	6.18*	-3.85*	-0.82*	3.90
Nderera x CIP 3910147.34	-3.93*	1.15	0.97*	-11.03**
Nderera x CIP 393385.39	-8.47**	5.24*	0.27	7.95*
CIP 393036.201 x CIP 393280.82	2.50	-4.75*	-1.02*	5.20*
CIP 393036.201 x CIP 391047.34	-8.92**	3.05	0.77	1.57
CIP 393036.201 x CIP 393385.39	-7.15**	4.49*	0.07	6.60*
CIP 393032.82 x CIP 391047.34	9.08**	3.23*	0.72	-2.64*
CIP 393032.80 x CIP 393385.39	-3.25	-0.62	0.52	-4.51*
CIP 391047.34 x CIP 393385.39	-8.67**	-0.91	1.32*	-19.79**

TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight; * and ** denote significant differences at p=0.05 and p=0.01, respectively.

Table 10. Phenotypic correlation between four traits of 28 potato families tested across two locations in Rwanda.

Trait	rAUDPC	TTW	TTN	ATW
Kinigi				
rAUDPC	1			
TTW	- 0.23ns	1		
TTN	0.08ns	0.32ns	1	
ATW	-0.25ns	0.28ns	-0.37*	1
Nyamagabe				
rAUDPC	1			
TTW	- 0.52**	1		
TTN	-0.47**	0.66***	1	
ATW	0.08ns	-0.22 ns	-0.22 ns	1
Across locations				
rAUDPC	1			
TTW	- 0.39*	1		
TTN	-0.25ns	0.59 ***	1	
ATW	0.04 ns	0.12 ns	-0.37*	1

Significance levels: * $p \leq 0.05$; ** $p \leq 0.01$; *** $p \leq 0.001$; NS: Not significant; TTW = total tuber weight; rAUDPC = relative area under the disease progress curve; TTN = total tuber number; ATW= average tuber weight.

tuber size. There was a negative correlation between TTN and ATW in this research. Ruiz de Galarreta et al. (2006) found similar results in the first clonal generation. In addition, the negative correlation between TTW and rAUDPC observed in this study indicated that late blight impacts negatively on tuber yield through destruction of foliage and consequent reduction of photosynthetic capacity. Similar results were also reported (Dowley et al., 2008; Mantecón, 2009) and indicated that late blight negatively impacts on potato yield. The present study corroborated previous studies (Brazil et al., 2002; Kaushik et al., 2007; Muthoni et al., 2013) that genotypes with good levels of resistance to potato diseases had high tuber yields. Therefore almost all the families selected for further evaluation had high levels of late blight resistance and high yields.

Materials and Methods

Study sites

The study was conducted at two sites: Kinigi and Nyamagabe in Rwanda. Kinigi is located in the highlands of volcanic soils at an altitude of 2200 meters above sea level (masl), on longitude of 29° 38' East and latitude 1° 30' South (ISAR, 1987). Annual temperature and rainfall averages at 16°C and 1480 mm, respectively. Nyamagabe is also a RAB research station located in the highlands of Congo/Nile divide in the Southern agricultural zone at an altitude of 2300 masl on longitude of 29° 33' East and latitude of 1° 33' South (ISAR, 1987) with respective annual rainfall and temperature of 1600 mm and 19°C.

Parental materials and crosses

Ten genetically diverse parents were selected and crossed using a 10 × 10 half diallel mating design. Crossing was done in the field during the short rains season of 2012. Parents were selected based on their good flowering abilities, high yields and acceptable level of resistance to late blight. These parents were CIP 393371.58, CIP 391047.34, CIP 393385.39, CIP 393280.82, CIP 393036.201, Gikungu, Kigega, Nderera, Ngunda and Kirundo (Table 1). The unopened flowers of the female parent were emasculated one day before crossing to avoid self-contamination. A booster dose of fertilizer was applied in the form of N₁₇-P₁₇-K₁₇ at a rate of 200 kg ha⁻¹ at planting. Weeds were controlled by using hoeing and hand cultivation as required but no pesticide was applied.

Controlled hand pollination was performed following emasculation (Aquaah, 2007). At maturity, approximately 40 days after pollination, berries of the same cross were identified, harvested and bulked together, labeled and kept at room temperature for approximately two weeks to ripen. After softening, seeds were manually removed by pressing the berries in a cloth bag. Extracted seeds were washed thoroughly with water and soap, dried and packed in paper envelopes for storage until planting time.

F₁ seedlings evaluation

A total of 45 crosses were expected from a 10 × 10 half diallel. However, cross combinations involving two parents: Kirundo or clone CIP 393371.58, produced insufficient seeds (<50 seeds per cross) or dropping flowers after fertilization. This was below the required number of seed 100 to 250 seeds per family to account for potential germination and transplanting losses and expected genetic assessment. Therefore 17 families were left out and only 28 families included, constituting an 8 × 8 half diallel. A minimum of 100 F₁ seeds per cross was sown on a mixture of compost and sand on raised seed bed. All the seeds collected from each cross constituted F₁ seeds and the individual plants grown from these seeds are F₁ seedling plants. Sixty seedlings randomly selected from each cross were grown separately in the field to produce F₁ tubers. The selected seedlings were grown separately in the field during the long rain season of 2013, without replications (Hahn et al., 1979; Ceballos et al., 2004).

Clone I evaluation trial

Each of the selected seedling plants represented a clone. The 28 F₁ populations were grown along with parents in a 6 × 6 lattice design with two replications during the 2013 short rains season. A total of 40 plants per plot per family were planted in two rows of 20 plants each. The spacing was 0.9 m and 0.3 m between and within rows, respectively, providing a population density of 38 000 plants ha⁻¹. The plot area was 5.4 m² (0.9 x 0.3 x 20). For each evaluation, spreader rows of a susceptible cultivar (Victoria) were included in each plot and around each replication.

Pathogen preparation and inoculation

Potato leaflets with young late blight lesions were collected from a not previously fungicide sprayed plants and transferred to the laboratory for isolation of *Phytophthora infestans*. Newly infected leaflets were collected in a plastic envelope and immediately transferred in the laboratory and washed with distilled water. Each leaflet was washed in distilled water and placed in a sterile glass petri-dish with sterile filter paper moisten with distilled water. The petri-dishes were incubated at 20°C for 24 hours to allow the fungus on the leaflet to sporulate. Sporulation media were made of modified rye B agar (Caten and Jinks, 1968) consisting of the filtrate of pre-rinsed rye (*Secale cereale* L.) seeds (100.0 g/l) boiled for 1 hour, de-ionized (di) H₂O added to a final volume of 1.0 l, glucose (7.5 g/l), β-sitosterol (0.05 g/l) and agar (15.0 g/l). The mycelial/sporangial mat was rinsed in cold (4°C) sterile, distilled water and scraped from the agar plate surface with a rubber policeman. Sporangia were counted with a haemocytometer and the final concentration was adjusted to 1x10⁴ sporangia. ml. Sporangial cultures were incubated for 2-3 hours at 4°C to stimulate zoospore release. Plants and soil were thoroughly watered prior to inoculation. In all experiments, plots were inoculated late in the evening at 6.00 pm using a hand-held sprayer until run off. After inoculation, the trial was watered in the mornings and evenings with approximately eight litres of water per square meter each day using a watering-can in order to improve potato moisture and field humidity to promote sporulation and infection.

Data collection

In the seedling trial, data collected included pollinated fruits and number of seeds per cross, visual observation on vigour and late blight reaction. In the first clonal evaluation trial, data collected included late blight disease reaction, total tuber weight, average tuber weight and number of tubers per plant. Late blight symptoms were assessed after planting. Starting with the first appearance of the symptoms, plants within each plot were visually rated at 7 day intervals for percent leaf and stem area with late blight lesions. This was done visually by comparing the green and non-green leaf portions affected by the disease using the 1 to 9 scale devised by the International Potato Centre, that is, 1=0%, 2=2.5%, 3=10%, 4=25%, 5=50%, 6=75%, 7=90%, 8=97.5% and 9=100% leaf area showing disease symptoms (Henfling, 1987). The mean percent blighted foliar area per plot was calculated. Evaluations continued until the susceptible genotype reached 90-100% of leaf blight assessments. For all plots and assessment dates, the area under the disease progress curve AUDPC (Campell and Madden, 1990) was calculated (Bradshaw, 2007). The rAUDPC (%) was used in the analysis of variance. The rAUDPC was calculated using the following formula:

$$rAUDPC = \frac{\sum (T_{i+1} - T_i) * \left(\frac{D_{i+1} + D_i}{2} \right)}{T_{Total} * 100} \quad (1)$$

In equation 1, T_i is the ith day when an estimation of percent foliar late blight is made and D_i is the estimated percentage of area with blighted foliage at T_i. T_{total} is the number of days at which the final assessment was recorded. Total tuber weight (TTW) was measured and expressed in t/ha. This was

calculated as the total weight of all the tubers harvested in a plot and converted to t/ha. Total tuber number (TTN) was the total number of tubers harvested per plant. Average tuber weight (ATW) was calculated as the total tuber weight per plant divided by the total tuber number of tubers per plant.

Data analysis

The analysis of variance (ANOVA) for the traits was done using the GLM procedure of SAS (SAS Institute, 2004). Mean separation was performed using the least significant difference (LSD) procedure at a 5% probability level. Pearson's phenotypic correlation coefficients between the 28 families for each trait were calculated using PROC CORR of SAS (SAS Institute, 2004) to determine trait associations. Separate ANOVA were conducted per location with genotypes as the main effect and later combined ANOVA were calculated across locations after homogeneity of variance test. The diallel analysis was conducted using SAS-05 diallel programme (Zhang et al., 2005) in SAS 8th edition. Griffing's (1956) diallel method 2, model 1 for a fixed model was fitted to estimate the GCA and SCA effects as: Y_{ij} = μ + g_i + g_j + b_k + s_{ij} + e_{ijkl} Where: Y_{ij} = observed value of the cross between parent i and j; μ = overall mean; g_i = GCA effect of parent i; g_j = GCA effect of parent j; s_{ij} = SCA of the cross between parents i and j; b_k = effect of the kth block; e_{ijkl} = experimental error. The relative importance of GCA and SCA effects for each trait was determined according to the general predicted ratio (GPR) as follows: GCA/SCA = 2MS_{GCA}/(2MS_{GCA}+MS_{SCA}) (Baker, 1978). When the GCA/SCA ratio is greater than 0.5, additive effects are more important than non-additive effects in the inheritance of the concerned trait, whereas if the ratio is smaller than 0.5, dominance effects are more important in the inheritance of the concerned character (Baker, 1978).

Conclusion

The best cross combinations were achieved when involving parents Gikungu, Kigega, CIP 393036.201 and CIP 391047.34. These parents were selected for future breeding and evaluation processes. Overall, ten top families Gikungu x CIP 391047.34, Gikungu x CIP 393036.201, Kigega x CIP 393036.201, Kigega x CIP 393280.82, Gikungu x CIP 393385.39, CIP 393280.82 x CIP 391047.34, Nderera x CIP 393036.201, Ngunda x CIP 393280.82, Ngunda x CIP 391047.34, Gikungu x Ngunda expressing high tuber yield and resistance to late blight were selected for further evaluations.

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