APPSA 2nd SCIENTIFIC CONFERENCE

2 - 4 April 2025 | Manthabiseng Convention Centre
Maseru, Kingdom of Lesotho

Population structure and genetic diversity analyses of common bean germplasm collections of Southern Africa using morphological traits and high-density SNP markers

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Centre for Coordination of Agricultural Research and Development for Southern Africa

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Bean production

- Common bean (*Phaseolus vulgaris* L) -field crops that suffers from low yield due to environmental constrains.
- Global average yield of 2,000 kg ha^{-1} (Margaret et al. 2014).
- In southern Africa -drought and poor soils reduce bean yield by at least <200 kg ha^{-1}
- Hence, there is a need to develop high yielding and environmentally adaptable varieties to improve productivity.
- Genetic diversity in plant germplasm is vital for crop improvement

Bean genotypes characterization

- Characterizing bean genetic diversity;
  - Morphological and agronomic traits, influenced by the environment
  - Biochemical markers such as isoenzymes, proline
  - and molecular markers; e.g. RAPD, SSR, AFLP, SNP (Gutierrez et al. 2023; Raatz et al. 2019; Josia et al 2021)

- Single Nucleotide Polymorphism (SNP) markers are used to assess genetic diversity in common bean.
- SNP markers identify genetic variations, aiding in selection for traits like drought tolerance, disease resistance and yield.
- Recent advances in the next-generation sequencing platform has made it possible to discover over a million SNP markers in common beans.

Objectives

- (i) Analyze genotype environment interaction (GEI) for common beans grain yield by the additive main effects and multiplicative interaction (AMMI) model.
- (ii)) Assess genetic diversity and population structure using 3127 SNP markers produced using the diversity array technology (DArT)

Descriptions of the 22 accessions tested in this study;

11 breeding lines developed at Mokhotlong by Thabo Sekhonyana - yield and taste

3 -CIAT (Mozambique)

8 - landraces and commercial cultivar/varieties grown widely in Lesotho

Code	Sample Name	Source	Cultivation status	Seed colour	Growth habit
LRB L1	Pinto Sierra	Lesotho	Variety	Mottled creamy bean with brownish speckles	Determinate
LBR L2	Kufuna	Mozambique	Breeding line	Red mottled	Determinate
LRB L3	Basotho Green	Lesotho	landraces	Green	Determinate
LBR L4	Matina	Mozambique	Breeding line	Whole red	Determinate
LRB L5	Senqu	Lesotho	Breeding line	Light brown with deep brown zebra stripe	Indeterminate
LRB L6	Khaki	Lesotho	Breeding line	Brownish green	Determinate
LRB L7	Basotho brown	Lesotho	Breeding line	Light brown	Determinate
LRB L8	Small white	Lesotho	Landrace	White	Determinate
LRB L9	Rwamballi	Tanzania	Landrace	Dull Purple	
LRB L10	Pinto Nodak	South Africa	Variety	Creamy mottled bean with brown speckles	Indeterminate
LRB L11	Khoalints'o	Lesotho	Breeding line	Mottled bean with black pinto speckles	Indeterminate
LRB L12	Mets'ets'e	Lesotho	Breeding line	Light brown	Indeterminate
LRB L13	Lebete NUA 45	Lesotho	Cultivar	Red mottled	Determinate
LRB L14	Sugar bean PAN 148	South Africa	Cultivar	Creamy mottled bean with red speckled	Determinate
LBR L15	Tiyela	Mozambique	Breeding line	Creamy mottled bean with red speckled	Determinate
LRB L16	Borotho	Lesotho	Breeding line	White	Indeterminate
LRB L17	Carolina	Lesotho	Breeding line	White	Determinate
LBR L18	Thaba-bosiu	Lesotho	Breeding line	Golden yellow	Indeterminate
LRB L19	Lesotho sugar bean	Lesotho	Breeding line	Pink mottled bean with purple speckles	Determinate
LRB L20	Lebabalasi	Lesotho	Breeding line	Creamy light brown	Indeterminate
LBR L21	Majuba	Lesotho	landraces	Golden yellow	Indeterminate
LBR L22	Maloti	Lesotho	Breeding line	Creamy light brown	Indeterminate

Lebete NUA 45 Thaba-Bosiu Borotho Khoalints'o

Basotho brown Pinto Sierra Sengu Small white

Sugar bean Lesotho Sugar Pindo Nodak Mets'ets'e Lebabalasi

Breeding lines collected from the International Centre for Tropical Agriculture (CIAT) earlier tested in Mozambique as phosphorus efficient cultivars

Phosphorus Efficient Varieties (Mozambique)

Common bean *(Phaseolus vulgaris L.)*

Matina (AP 82)

Kafuna (AP 89)

Tyela (LPA 31)

Experimental trials laid in three agro-ecological zones and characteristic of the locations

Agro-ecological zones	Location	Annual Rainfall	Average Temperature	Geographical coordinates		Altitude (asl)	Soils	Agric
				Latitude	Longitude			Potential
Highlands	Thabang Agric research station (Mokhotlong District)	1200 mm	17.3°C	-29.3243° S	28.9892° E	3,100m	Clay loam	Short growing season
Northern lowlands	Lesotho Agricultural College (LAC) Farm (Leribe District)	800 mm	26.8°C	-28.8623° S	28.0529° E	2,000m	Loam	High yielding
Southern lowlands	Siloe Agricultural Research Station (Mohale's hoek District)	400 mm	27.6°C	-29.6408° S	27.2220° E	1,500m	Sandy Clay	Dry poor yields

Trial layout

- ✓ The trials ran for seasons (2022/2023 and 2023/2024) - (November and April).
- ✓ RCBD with three replications.
- ✓ Phenotypic data was collected twice in a season from each location.

Agro-morphological traits used to screen the genotype

Character	Class /unit
Internode length	cm
Flower colour	1 = white, 2 = purple
Leaf area index (length and width)	mm
Days to flowering and maturity	days
Yield determination	
✓ plant population/final crop stand	no.
✓ number of pods per plant	no.
✓ number of seeds per pod	no.
✓ 100 seed weight	grams

AMMI (Additive Main effects and Multiplicative Interaction) analysis - to assess genotype and environment interactions (GEI) used to identify stable genotypes across different environments.

It combines Analysis of Variance and principal component analysis (PCA)

ANOVA for genotype–environmental interaction and first and second main component (IPCA) of bean genotypes in Lesotho

Significant effects of GEI indicated that calculated grain yield of genotypes varied across the environments and seasons

Source	df	SS	MS	F_prob	%SS	%SS of GEI
Total	299	60618565	202738			
Genotypes	19	7477488	393552	0.0001***	12,34	
Environments	4	9329442	2332360	0.0001***	15,39	
Block	10	3243952	324395	0.05*		
GxE	76	15102990	198724	0.05*	24,91	
IPCA1	22	5461982	248272	0.05*		36,16
IPCA2	20	4106775	205339	0.07ns		27,19
Residuals	34	5534233	162772	0.20797		
Error	190	25464693	134025			

Genotypic and environmental yield means, IPC1, IPC2–stability

Thus, high yielding genotypes such as G7, G12 and G20 have large IPCA1 scores and are unstable

While genotypes G5 -Senqu, G15- Thaba-Bosiu, G18-Lesotho Sugar and G19-Khoali-Khube are stable

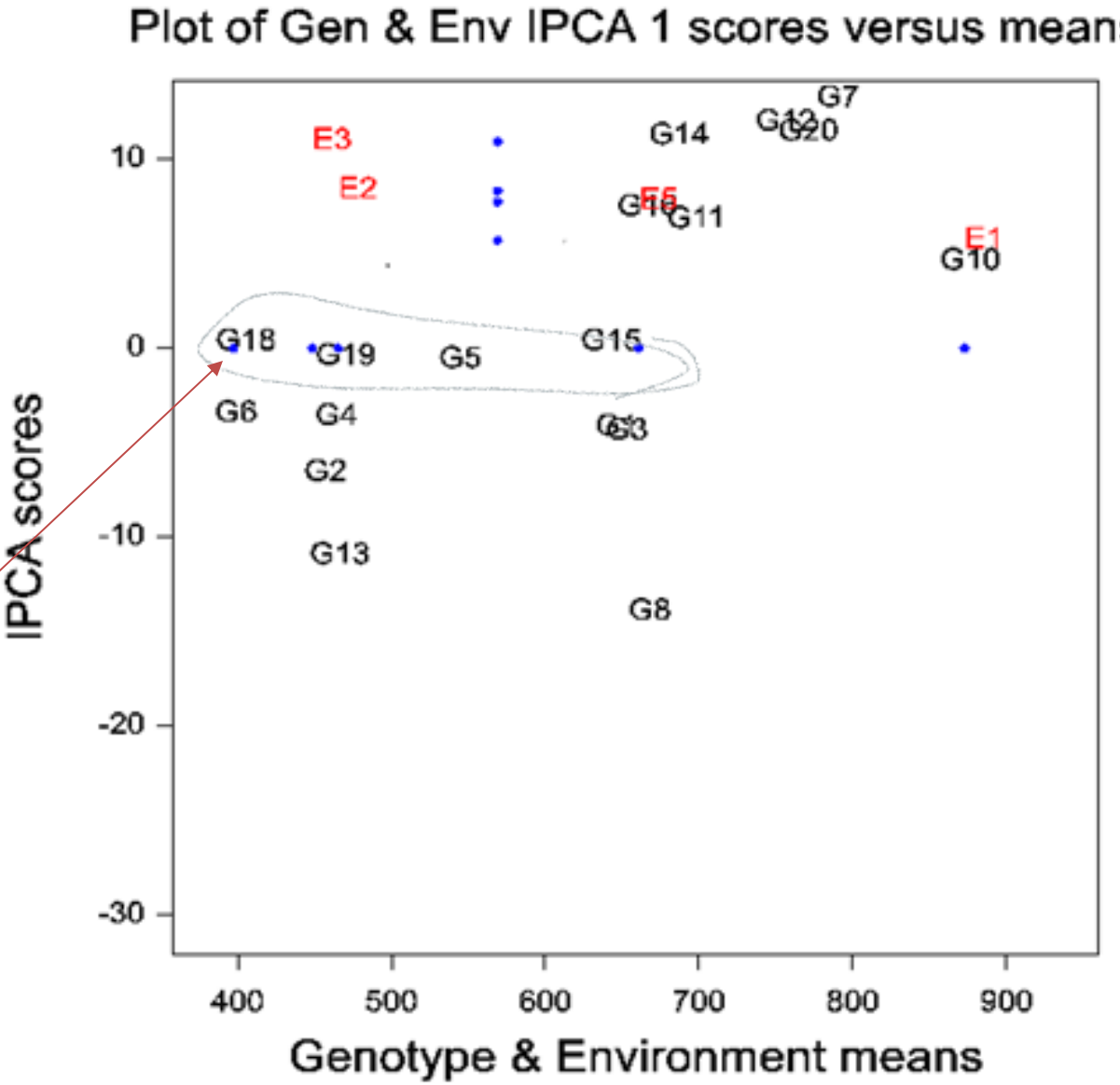
Genotypes	Identification	Means (Kg/ha)	IPCA1	IPCA2
G1	Pinto Seirra	632,60	-4,21	9,59
G2	Majuba	443,10	-6,62	1,55
G3	Basotho Green	639,10	-4,43	-10,24
G4	Small white	450,30	-3,66	3,79
G5	Senqu	530,80	-0,64	-0,14
G6	Khaki	385,00	-3,50	-2,13
G7	Basotho Brown	776,50	13,18	1,64
G8	Sugar bean PAN 148	654,20	-13,97	-4,81
G9	Rwambali	266,50	-3,73	-1,16
G10	Pinto Nodak	**856,80**	4,56	-6,16
G11	Khoalintso	679,10	6,77	-0,62
G12	Mets'ets'e	736,70	11,91	2,16
G13	Lebete NUA 45	446,70	-11,01	5,94
G14	Maloti	667,30	11,20	2,42
G15	Thaba Bosiu	623,20	0,30	8,19
G16	Borotho	646,90	7,41	-16,58
G17	Carolina	357,10	-14,81	-0,81
G18	Lesotho Sugar	385,60	0,30	-14,88
G19	Khoalikhube	450,60	-0,48	14,91
G20	Lebabalasi	751,30	11,39	7,33

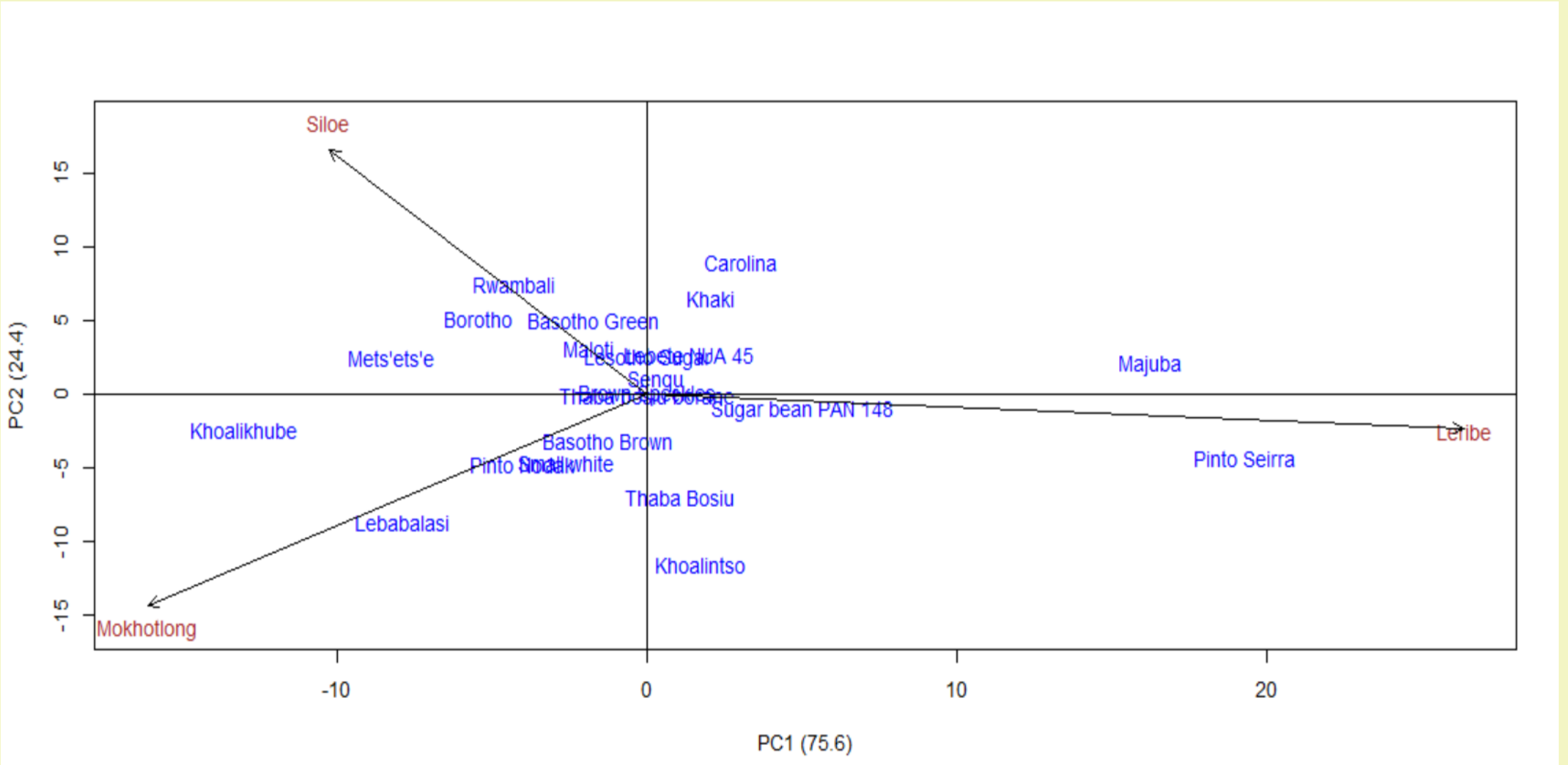
GGE Biplot 1 for the genotype–environmental interaction

- *E1; Leribe/ northern lowlands 2023,*
- *E2; Mokhotlong mountains 2023,*
- *E3; Siloe/southern lowlands 2023;*
- *E5; Mokhotlong/mountains 2024 –*

- G10 - Pinto Nodak is best adapted to Leribe

- G11 - Khoalintso, G16 - Borotho, G14 - Maloti are best suited for Mokhotlong (All breeding lines from Mokhotlong)
- Genotypes located in the near distance from the origin are stable such G18, G19, G5 and G15





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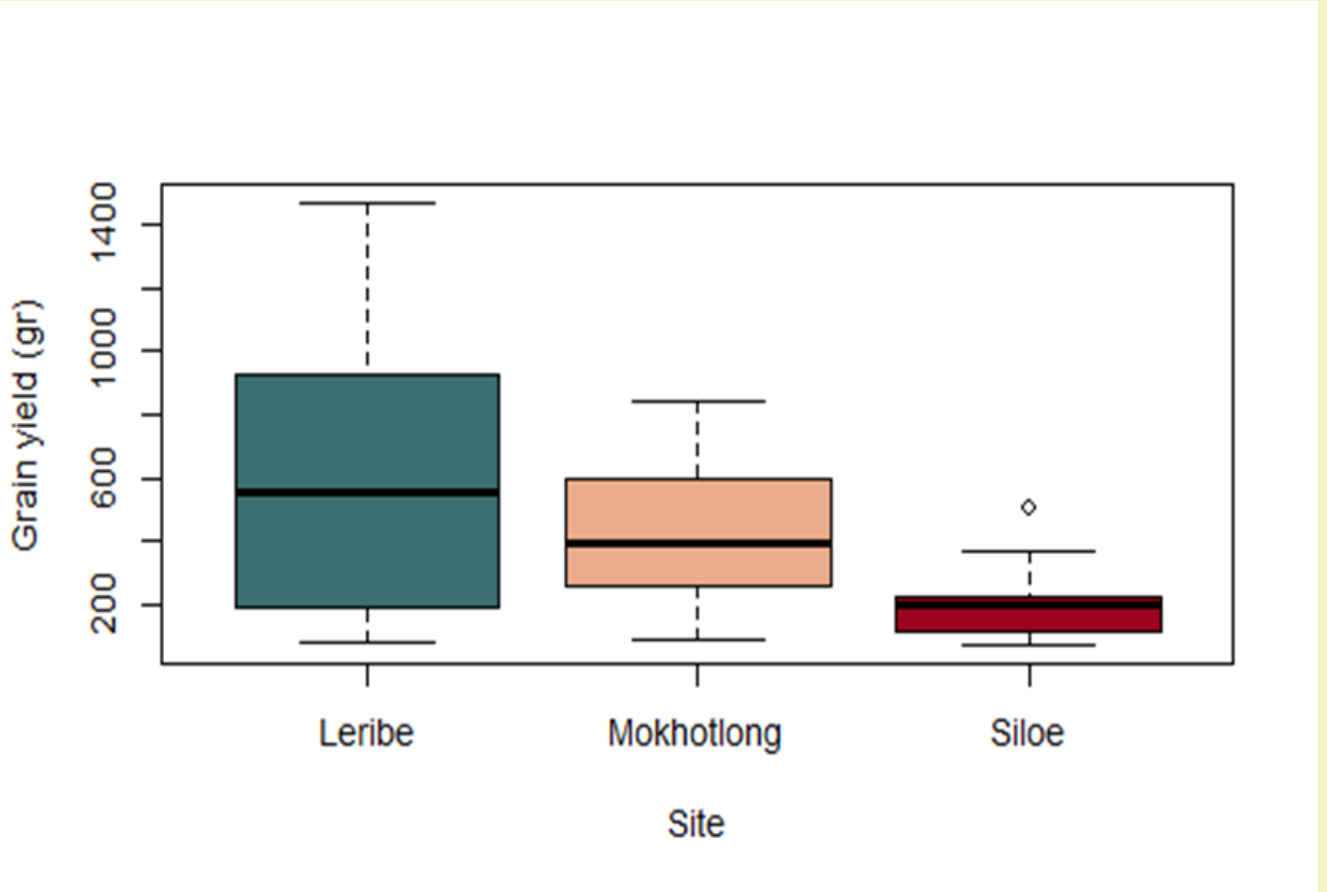
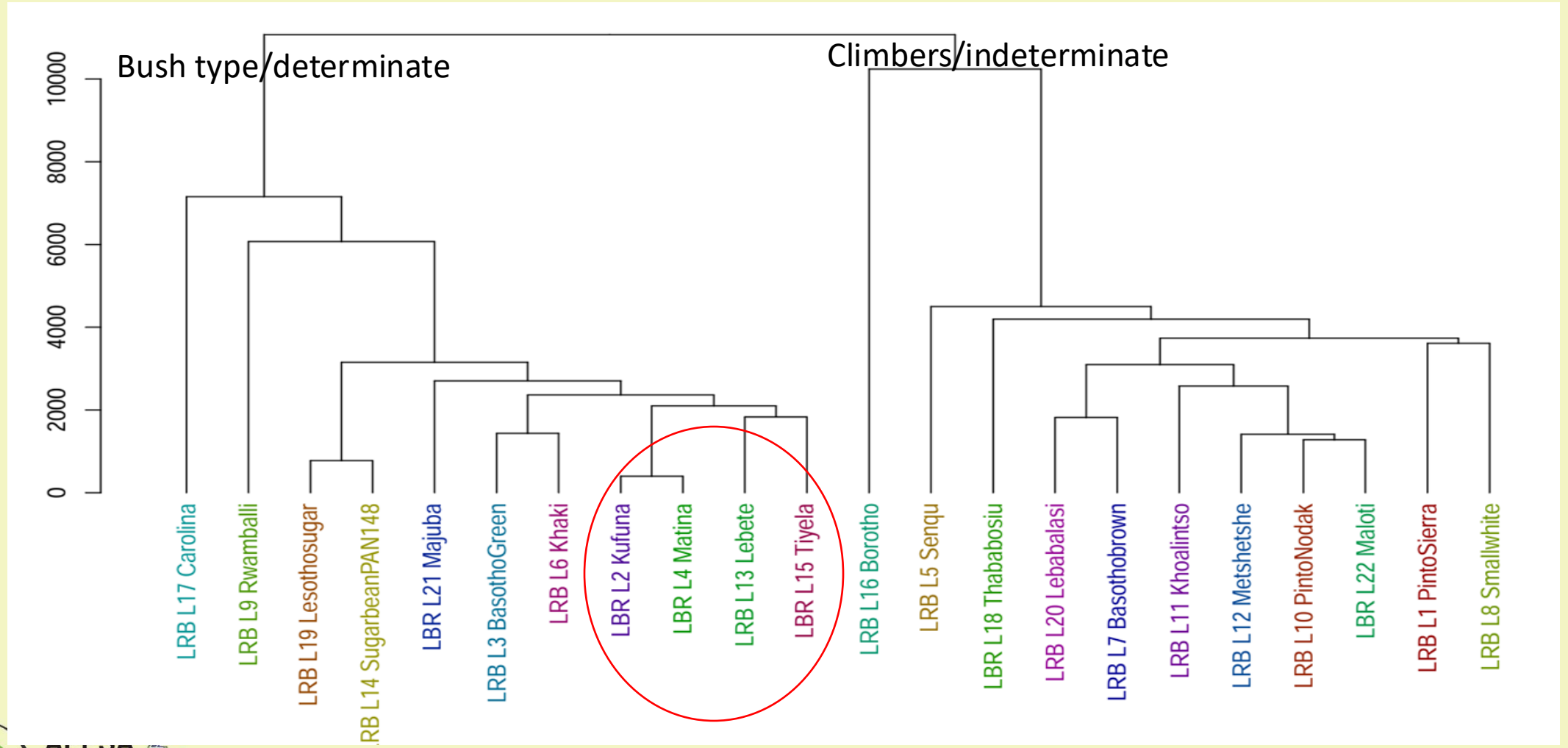
Leribe on average of seasons had highest yields



Genotypic Analysis

- Cluster analysis divided the 22 common bean genotypes into two major genetic groupings based on SNPs markers
  - i) Climbers/indeterminate
  - ii) Bush types/determinate genotypes.
- The climbers subsets consist of
  - LRBL16 - Borotho (white seeds)
  - Genotypes that showed to be stable AMMMI - Senqu and ThabaBosiu
  - Breeding lines from Mokhotlong.
- The determinate clusters have three sub-clusters, namely
  - LRBL17 Carolina (white seeds),
  - LRBL9 Rwamballi (purple seeds from Tanzania), and
  - 3 common commercial cultivars in Lesotho and 3 cultivars from CIAT.
- The genotypes from CIAT, are clustered together with LRBL13-Lebete.

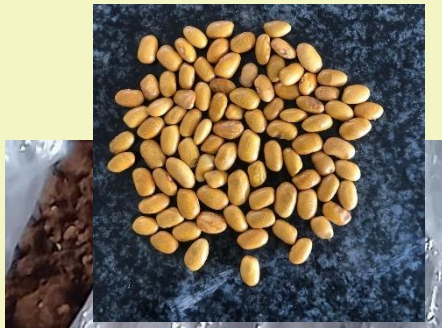
A dendrogram based on the dissimilarity matrix between the 22 genotypes



Dissimilarity matrix also showed LBRL16 and LBRL17 are dissimilar from all other genotypes

*	LBR L15	LBR L18	LBR L2	LBR L21	LBR L22	LBR L4	LRB L1	LRB L10	LRB L11	LRB L12	LRB L13	LRB L14	LRB L16	LRB L17	LRB L19	LRB L20	LRB L3	LRB L5	LRB L6	LRB L7	LRB L8	LRB L9
LBR L15	0	0.57698	0.10801	0.14923	0.6295	0.10688	0.62791	0.63259	0.62061	0.62956	0.09456	0.16443	0.40451	0.27289	0.15502	0.63112	0.12419	0.55873	0.11322	0.63281	0.6291	0.22079
LBR L18	0.57698	0	0.60041	0.54982	0.16496	0.59799	0.20774	0.16887	0.18791	0.14848	0.58142	0.53724	0.29361	0.47558	0.54493	0.19975	0.57476	0.22814	0.5965	0.20464	0.25113	0.48874
LBR L2	0.10801	0.60041	0	0.13788	0.65577	0.02001	0.65021	0.65383	0.6442	0.65436	0.09221	0.15683	0.41055	0.28731	0.15143	0.65569	0.11176	0.5808	0.09403	0.65329	0.66114	0.21288
LBR L21	0.14923	0.54982	0.13788	0	0.60939	0.13459	0.60142	0.6075	0.59596	0.61229	0.139	0.15187	0.38225	0.27029	0.14827	0.61347	0.10534	0.54926	0.09894	0.6092	0.602	0.21908
LBR L22	0.6295	0.16496	0.65577	0.60939	0	0.65553	0.16847	0.05752	0.12958	0.06437	0.63732	0.58931	0.27475	0.49221	0.59649	0.15992	0.64039	0.19199	0.66129	0.15044	0.2104	0.53885
LBR L4	0.10688	0.59799	0.02001	0.13459	0.65553	0	0.64867	0.65379	0.64335	0.65372	0.08984	0.15586	0.40512	0.28646	0.15195	0.65432	0.10703	0.57948	0.09174	0.65336	0.65991	0.20981
LRB L1	0.62791	0.20774	0.65021	0.60142	0.16847	0.64867	0	0.17782	0.18088	0.17071	0.63419	0.57861	0.37228	0.4852	0.5862	0.20911	0.63188	0.21397	0.65647	0.21115	0.1756	0.53967
LRB L10	0.63259	0.16887	0.65383	0.6075	0.05752	0.65379	0.17782	0	0.10541	0.08451	0.63698	0.58596	0.29598	0.49028	0.59504	0.16721	0.63728	0.19676	0.65889	0.12015	0.21743	0.53014
LRB L11	0.62061	0.18791	0.6442	0.59596	0.12958	0.64335	0.18088	0.10541	0	0.1437	0.62756	0.57302	0.34005	0.48592	0.58452	0.1886	0.62587	0.21675	0.64599	0.17013	0.22198	0.51931
LRB L12	0.62956	0.14848	0.65436	0.61229	0.06437	0.65372	0.17071	0.08451	0.1437	0	0.63693	0.58721	0.27867	0.49609	0.59715	0.13203	0.63659	0.19613	0.65919	0.12145	0.21703	0.52856
LRB L13	0.09456	0.58142	0.09221	0.139	0.63732	0.08984	0.63419	0.63698	0.62756	0.63693	0	0.16384	0.40696	0.27686	0.15499	0.63788	0.12095	0.5649	0.11061	0.63922	0.63965	0.22203
LRB L14	0.16443	0.53724	0.15683	0.15187	0.58931	0.15586	0.57861	0.58596	0.57302	0.58721	0.16384	0	0.38609	0.28505	0.03373	0.58972	0.10661	0.53664	0.13466	0.58859	0.59321	0.2417
LRB L16	0.40451	0.29361	0.41055	0.38225	0.27475	0.40512	0.37228	0.29598	0.34005	0.27867	0.40696	0.38609	0	0.39298	0.38946	0.35567	0.3554	0.29166	0.36482	0.3583	0.40523	0.2925
LRB L17	0.27289	0.47558	0.28731	0.27029	0.49221	0.28646	0.4852	0.49028	0.48592	0.49609	0.27686	0.28505	0.39298	0	0.28246	0.50974	0.26869	0.44912	0.2744	0.50013	0.47808	0.33351
LRB L19	0.15502	0.54493	0.15143	0.14827	0.59649	0.15195	0.5862	0.59504	0.58452	0.59715	0.15499	0.03373	0.38946	0.28246	0	0.59808	0.09653	0.53996	0.12698	0.5988	0.59942	0.2359
LRB L20	0.63112	0.19975	0.65569	0.61347	0.15992	0.65432	0.20911	0.16721	0.1886	0.13203	0.63788	0.58972	0.35567	0.50974	0.59808	0	0.64043	0.24211	0.66263	0.08924	0.20665	0.54717
LRB L3	0.12419	0.57476	0.11176	0.10534	0.64039	0.10703	0.63188	0.63728	0.62587	0.63659	0.12095	0.10661	0.3554	0.26869	0.09653	0.64043	0	0.55754	0.04441	0.63989	0.64128	0.19771
LRB L5	0.55873	0.22814	0.5808	0.54926	0.19199	0.57948	0.21397	0.19676	0.21675	0.19613	0.5649	0.53664	0.29166	0.44912	0.53996	0.24211	0.55754	0	0.57931	0.24309	0.23448	0.47069
LRB L6	0.11322	0.5965	0.09403	0.09894	0.66129	0.09174	0.65647	0.65889	0.64599	0.65919	0.11061	0.13466	0.36482	0.2744	0.12698	0.66263	0.04441	0.57931	0	0.65941	0.66792	0.18523
LRB L7	0.63281	0.20464	0.65329	0.6092	0.15044	0.65336	0.21115	0.12015	0.17013	0.12145	0.63922	0.58859	0.3583	0.50013	0.5988	0.08924	0.63989	0.24309	0.65941	0	0.20985	0.54769
LRB L8	0.6291	0.25113	0.66114	0.602	0.2104	0.65991	0.1756	0.21743	0.22198	0.21703	0.63965	0.59321	0.40523	0.47808	0.59942	0.20665	0.64128	0.23448	0.66792	0.20985	0	0.57124
LRB L9	0.22079	0.48874	0.21288	0.21908	0.53885	0.20981	0.53967	0.53014	0.51931	0.52856	0.22203	0.2417	0.2925	0.33351	0.2359	0.54717	0.19771	0.47069	0.18523	0.54769	0.57124	0

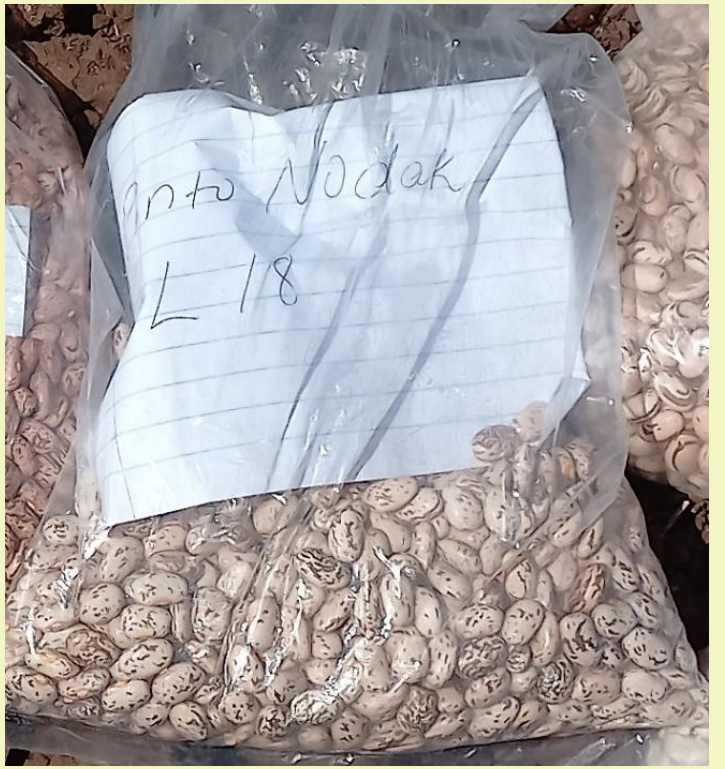
Based on IPCA1 scores Senqu, Thaba-Bosiu, Lesotho sugar and Khoalikhube are 4 most stable genotypes for the tested environments



G7 - Basotho Brown, G12 - Mets'ets'e and G20- Lebabalasi ranked the highest in all environments for grain yield, however were the most unstable genotype

Highest yields for the 2 seasons were obtained in Leribe

- Most yielding variety and adapted to Leribe - Pindo Nodak (mahe a likoekoe)
- However, it is unstable in tested environments



Conclusions and Recommendation

- The three genotypes with low IPCA1 score and the high yielding genotypes are grouped under one genetic cluster based on SNP markers - intermediate.
- The relationship between genetic markers and phenotypic expression is essential to determine the genetic variability and appropriate breeding strategies in Lesotho in order to select genotypes that can adapt to changing environmental conditions, including climate change, pests, and diseases.

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