

Maize Phenotyping under the Seeds of Discovery Initiative: *The Brute Force Method*



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Seeds of Discovery



Genome Wide Association Study (GWAS) in Maize

- Breeder's Core Collection from the CIMMYT Maize Germplasm Bank (4471 accessions).
- One plant per accession crossed with a CIMMYT hybrid to make a series of modified topcrosses.
 - ▶ The same plant was sampled for DNA extraction/GBS
- Accessions were crossed hybrids of their same environmental adaptation (where possible).

Tropical Accessions X Tropical Hybrid

Subtropical Accessions X Subtropical Hybrids

Highland Accessions X Highland Hybrid

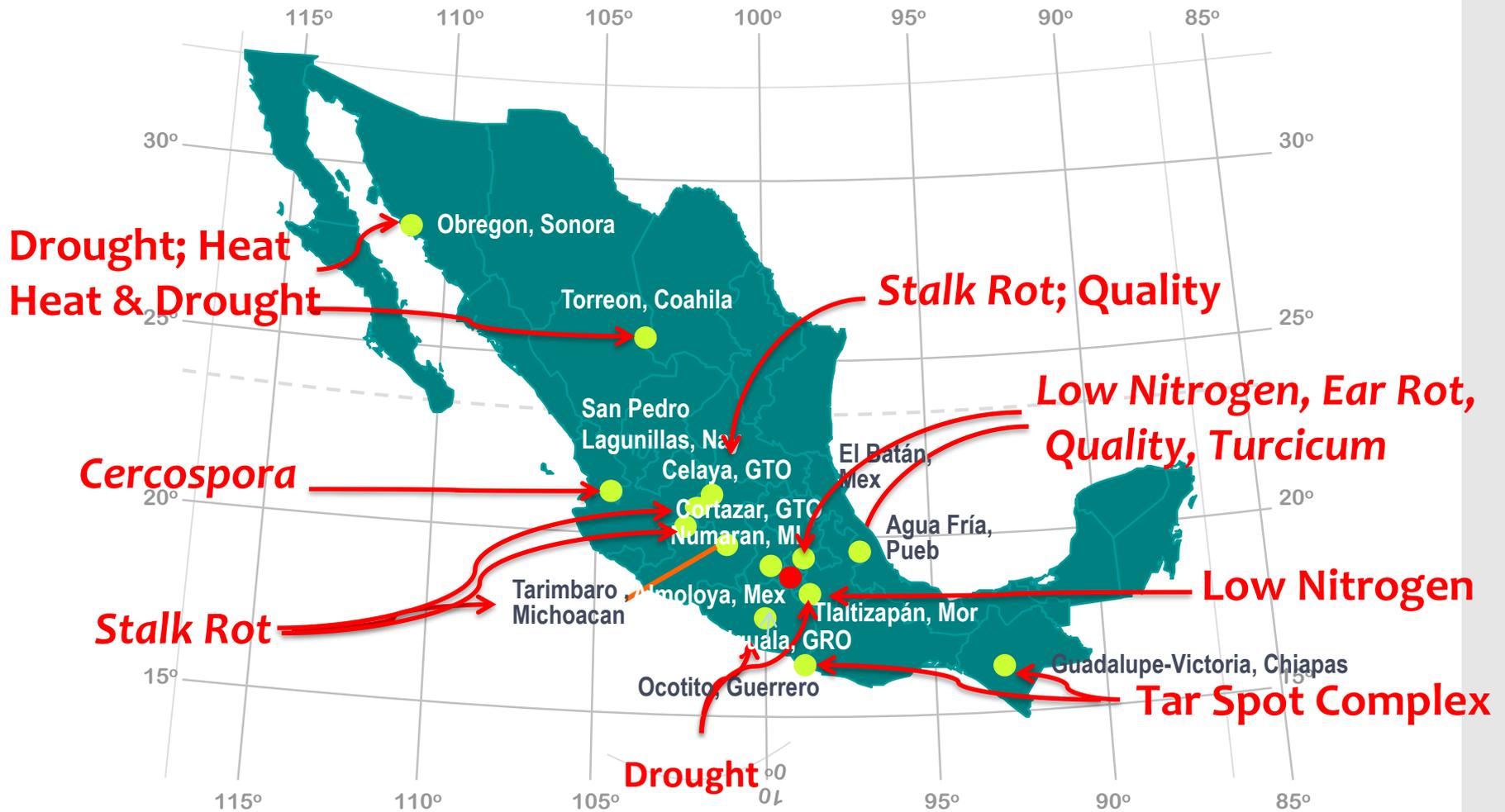


BA11-Seed-838
Entry: 838

StockID: BA11-Seed-838
Plot: 838 Entry: 838
IDACC: 10435 BANKACC: 10435 MEXI 58
Race1: CONICO 9 Race2:
Males (?)
Rows: 1 3 PLANTS
Origin: BA87 -2901- 68
Cross with Highland Single Cross



Trial Sites in Mexico: GWAS



Phenotypic Trials

- 36 trials planted - 34 harvested (34,606 rows and over 687,000 unique data points)
- 19 Trials for abiotic Stresses (Drought, Heat, Low Nitrogen)
- 11 Trials for diseases (Tar Spot Complex, Grey Leaf Spot, Turcicum Leaf Blight, Fusarium Ear Rot, Fusarium and Acremonium Stalk Rot)
- 3 Trials hand pollinated to produce grain for Quality Component Analysis.

Yield and Agronomic Data Taken : All Locations

- Yield (field weight, grain and cob weight, moisture, number of ears)
- Plant Height and Ear Height
- Male and Female Flowering (50% of row)
- Stalk and Root Lodging

Experimental Design

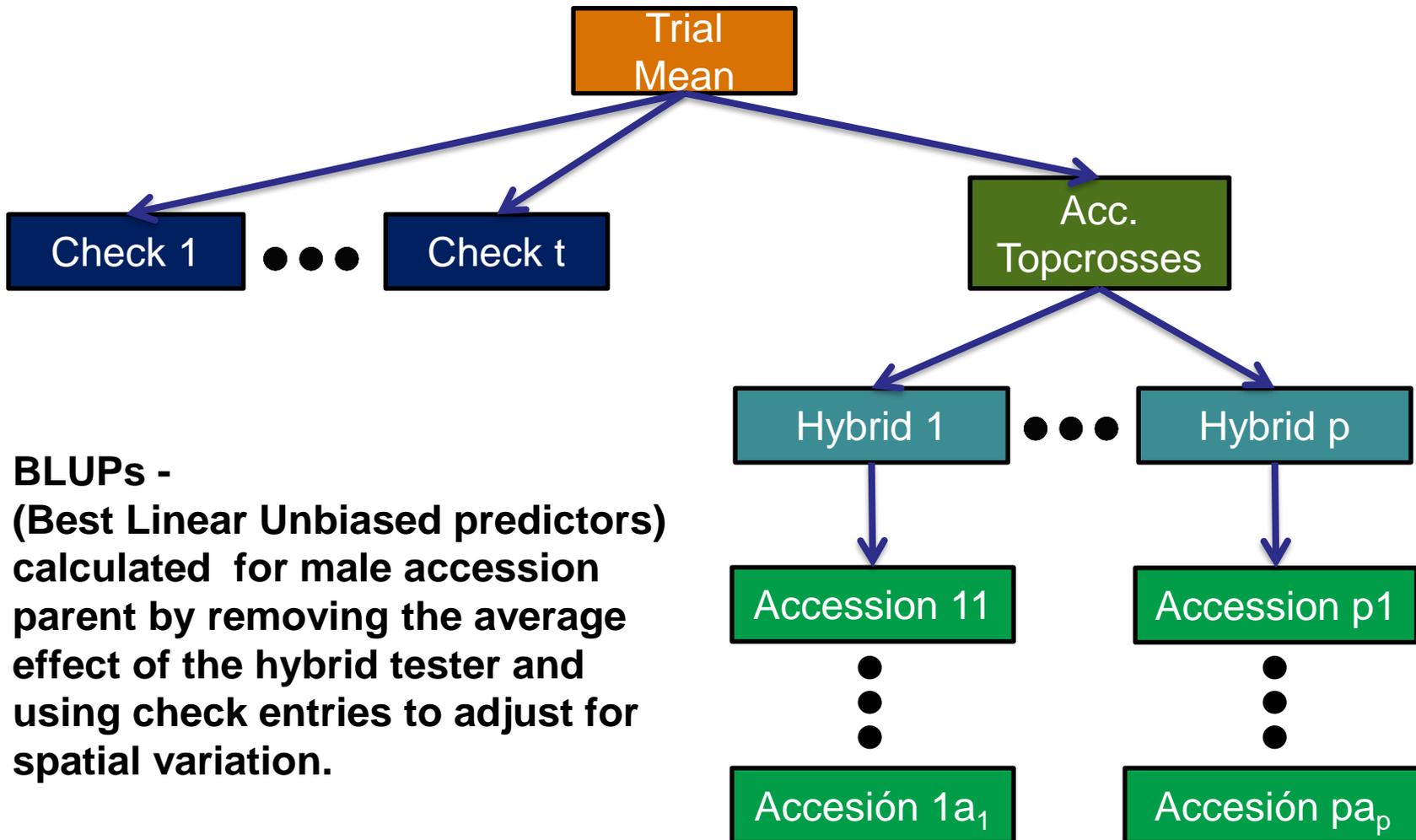
- ▶ **Unreplicated augmented row-column design**
- ▶ **For each trial location accessions are selected for adaptation zone and maturity.**
- ▶ **Overlapping sets of topcross entries**
- ▶ **Two widely adapted commercial checks and resistant and susceptible checks where appropriate**

Faja																				
20	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
19	40	39	38	37	36	35	34	33	32	31	30	29	28	27	26	25	24	23	22	21
18	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60
17	80	79	78	77	76	75	74	73	72	71	70	69	68	67	66	65	64	63	62	61
16	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100
15	120	119	118	117	116	115	114	113	112	111	110	109	108	107	106	105	104	103	102	101
14	121	122	123	124	125	126	127	128	129	130	131	132	133	134	135	136	137	138	139	140
13	160	159	158	157	156	155	154	153	152	151	150	149	148	147	146	145	144	143	142	141
12	161	162	163	164	165	166	167	168	169	170	171	172	173	174	175	176	177	178	179	180
11	200	199	198	197	196	195	194	193	192	191	190	189	188	187	186	185	184	183	182	181
10	201	202	203	204	205	206	207	208	209	210	211	212	213	214	215	216	217	218	219	220
9	240	239	238	237	236	235	234	233	232	231	230	229	228	227	226	225	224	223	222	221
8	241	242	243	244	245	246	247	248	249	250	251	252	253	254	255	256	257	258	259	260
7	280	279	278	277	276	275	274	273	272	271	270	269	268	267	266	265	264	263	262	261
6	281	282	283	284	285	286	287	288	289	290	291	292	293	294	295	296	297	298	299	300
5	320	319	318	317	316	315	314	313	312	311	310	309	308	307	306	305	304	303	302	301
4	321	322	323	324	325	326	327	328	329	330	331	332	333	334	335	336	337	338	339	340
3	360	359	358	357	356	355	354	353	352	351	350	349	348	347	346	345	344	343	342	341
2	361	362	363	364	365	366	367	368	369	370	371	372	373	374	375	376	377	378	379	380
1	400	399	398	397	396	395	394	393	392	391	390	389	388	387	386	385	384	383	382	381
Surco	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20

Example of Trial Design

Standard commercial checks ( and ) to adjust for spatial variance and link trials from multiple locations and trait specific, resistant () and susceptible () checks, to adjust for spatial variation for specific traits within the trial.

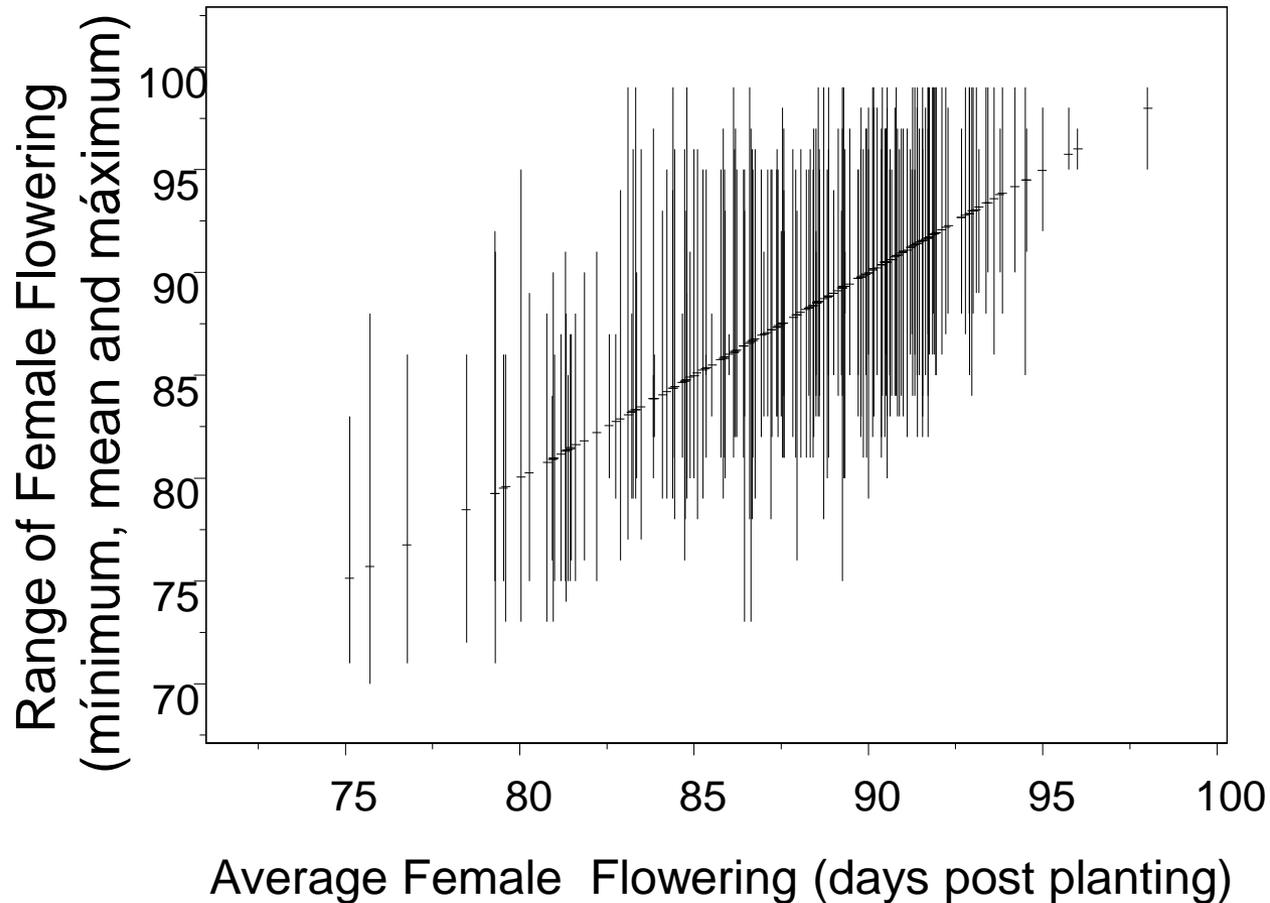
Model for Analysis



BLUPs -
(Best Linear Unbiased predictors)
calculated for male accession
parent by removing the average
effect of the hybrid tester and
using check entries to adjust for
spatial variation.

Challenges

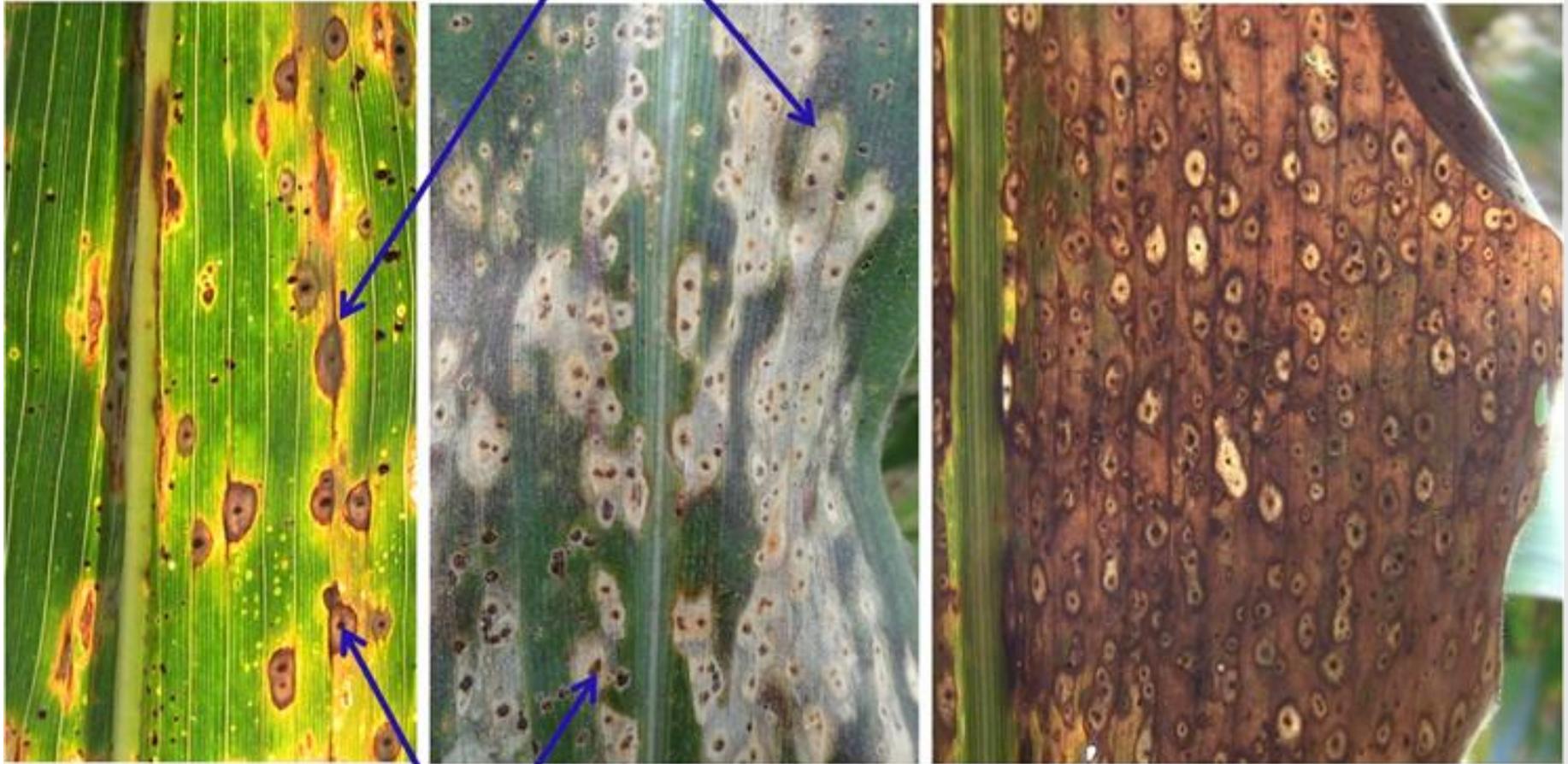
- Range of maturities (flowering extends for 5 weeks)
- Lodging
- Segregation
- Diversity of environments (both origin of accessions and evaluation sites).



Comparison of average female flowering date with range of female flowering for 191 accession topcrosses (per plant data)

Tar Spot Complex

The symptom caused by *Monographella maydis*



The symptom caused by *Phyllachora maydis*

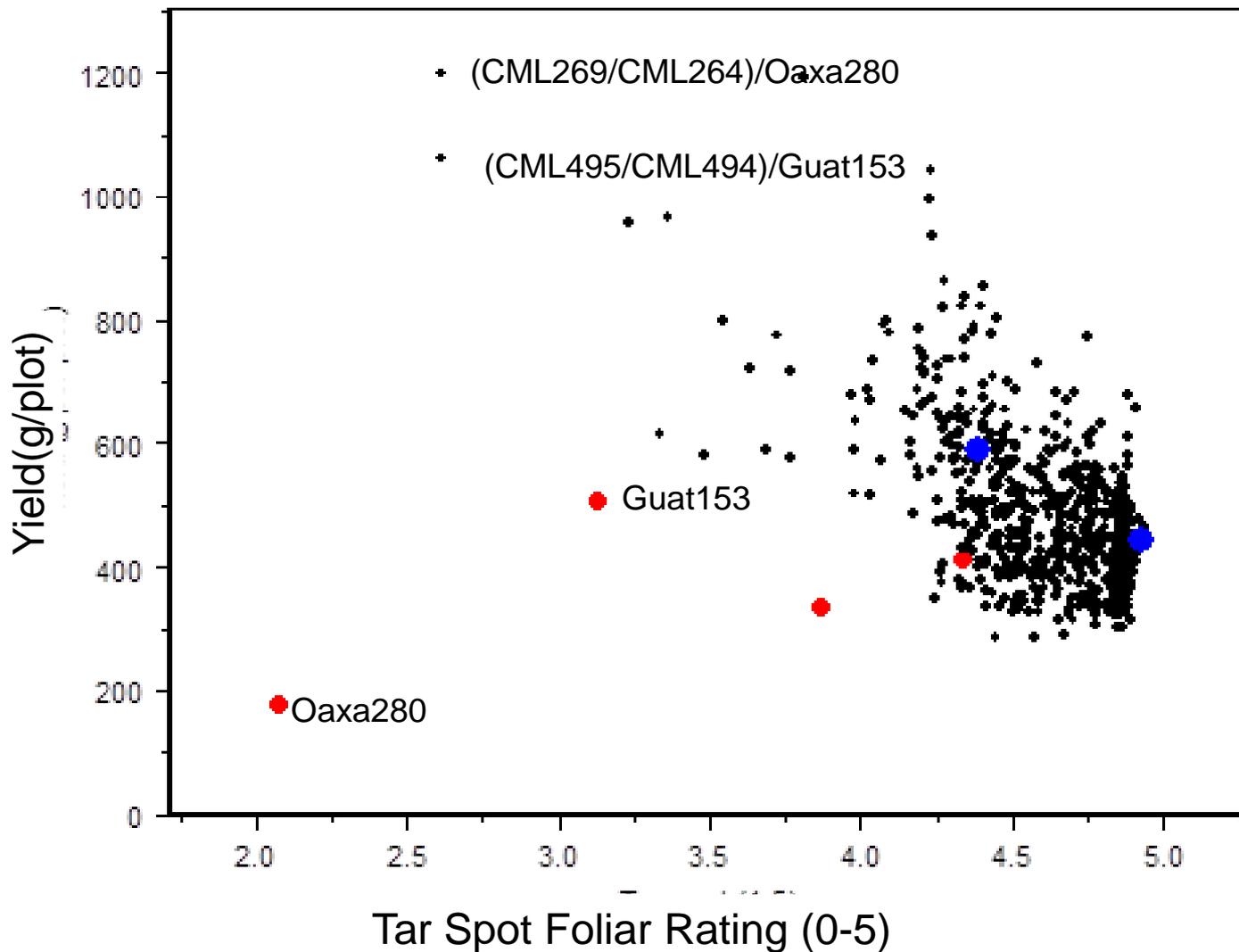
Tar Spot Complex



Photo: Rosemary Shrestha

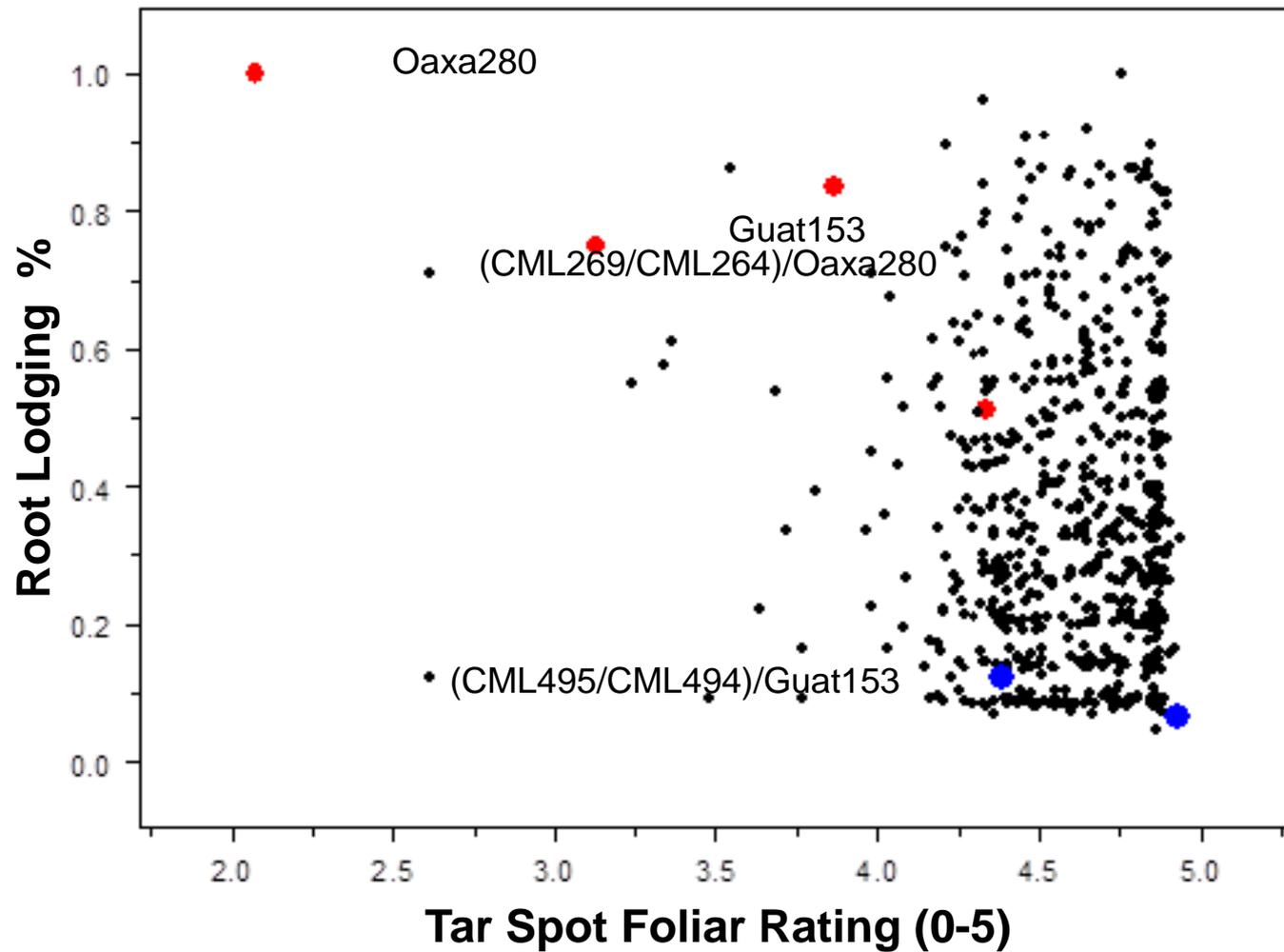
Tar Spot Trials

- 2011B Guadalupe-Victoria, Chiapas – 600 Accession/Topcross entries
- 2012B Guadalupe-Victoria, Chiapas – 810 accession/topcross entries (**including accessions per se**)
- 2 foliar ratings 0-5 scale (Ceballos and Deutsch) two weeks apart.
- Data taken both by row and as average of 6 plants per row



Relationship between Tar Spot rating and Yield (2nd foliar rating: scale 0-5; average of 6 plantas)

● = Accessions; ● = Topcrosses; ● = Commercial Checks



Relationship between Root Lodging and Tar Spot Rating (2nd foliar rating)

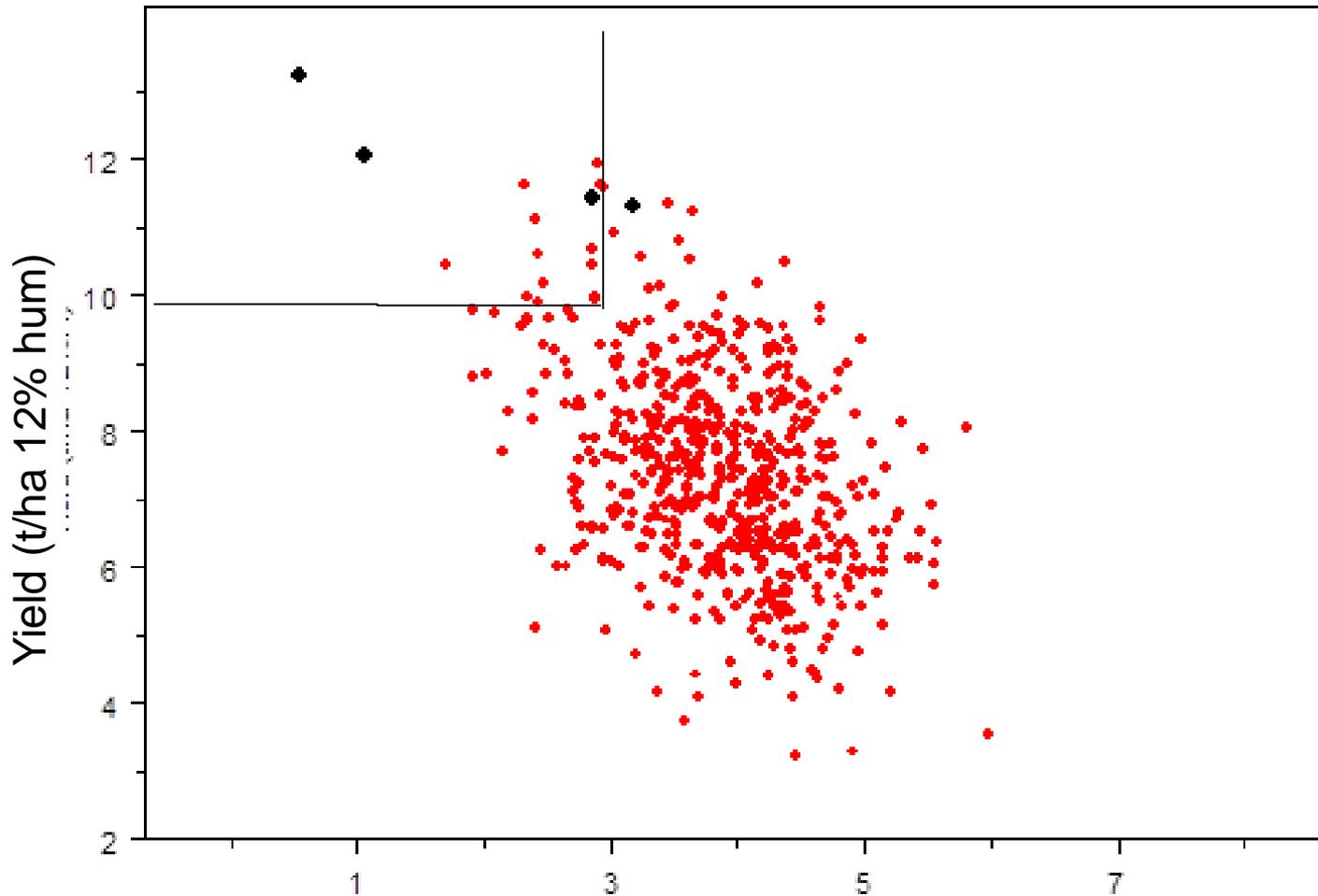
● = Accessions; ● = Topcrosses; ● = Commercial Checks

Characterization for Stalk Rot





Fusarium Stalk Rot: Artificial Inoculation



Stalk Rot Scale (proportion of stalks with *Acremonium strictum* times intensity 1-5)

Relationship between Yield and Stalk Rot Scale (percentage of stalks with disease x intensity of disease)

● = Topcrosses ; ● = Commercial Checks

Evaluation of Accessions PerSe for Acremonium Stalk Rot

Entry	Accession Perse (2013) Stalk Rot Scale Average	Accession Topcross (2012) Stalk Rot Scale Average
Commercial (resistant check)	0.43	0.5
PUEB45	1.16	1.7
MORE85	1.53	2.3
GUER125	1.59	2.4
NAYAGP6	2.13	2.5
CHIS128	2.56	2.9
MICH21	3.21	2.9
TC X URUG39 (susceptible)	4.5	5.97



Lots of data still to analyze...



Thank You



