



# FL24D, a Non-dormant Red Clover Selected for Herbicide Resistance to Improve Broadleaf Weed Control

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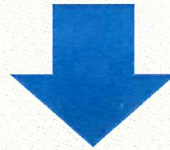
# Background

## Red Clover (*Trifolium pratense* L.)

High N-Fixation (150 kg ha)

High quality and versatile (graze or hay)

Red clover as “baleage” N. FL 2014





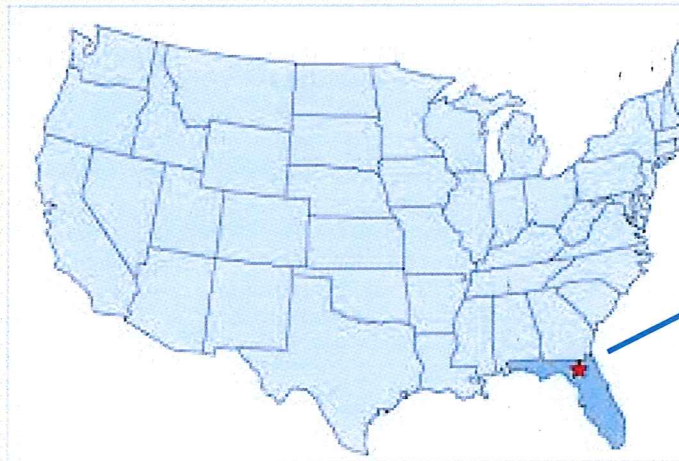
# Background

**University of Florida Forage Breeding Program:**

**High yield non-dormant – Cherokee**

**Resistant to Root-Knot Nematodes (RKN) – Southern Belle**

**Mid-dormant resistant to RKN – Barduro**



**Establishment of red clover is still a challenge!**



# Outline

**Herbicides - specifically 2,4-D**

**Population development**

**Forage yield and 2,4-D evaluation**

**Molecular markers and plans to move forward**





# Herbicides

**First introduced in 1940; important tool to manage weeds**

**2,4-D the fifth most used in the world and third in the U.S.**

**Major use in natural and cultivated grasslands**

**Causes uncontrolled cell division in vascular tissue**

**Resistance is not well understood:**

- Detoxification**
- Absorption barrier (surface wax)**
- Controlled by a single gene**
- Detriment to growth**



# Cultivar Development

Initial research by S.G. Taylor in 1985

Base population was a mixture of **Kenstar** (mid dormancy), **Nolins** (low dormancy), and **Cherokee** (low dormancy).

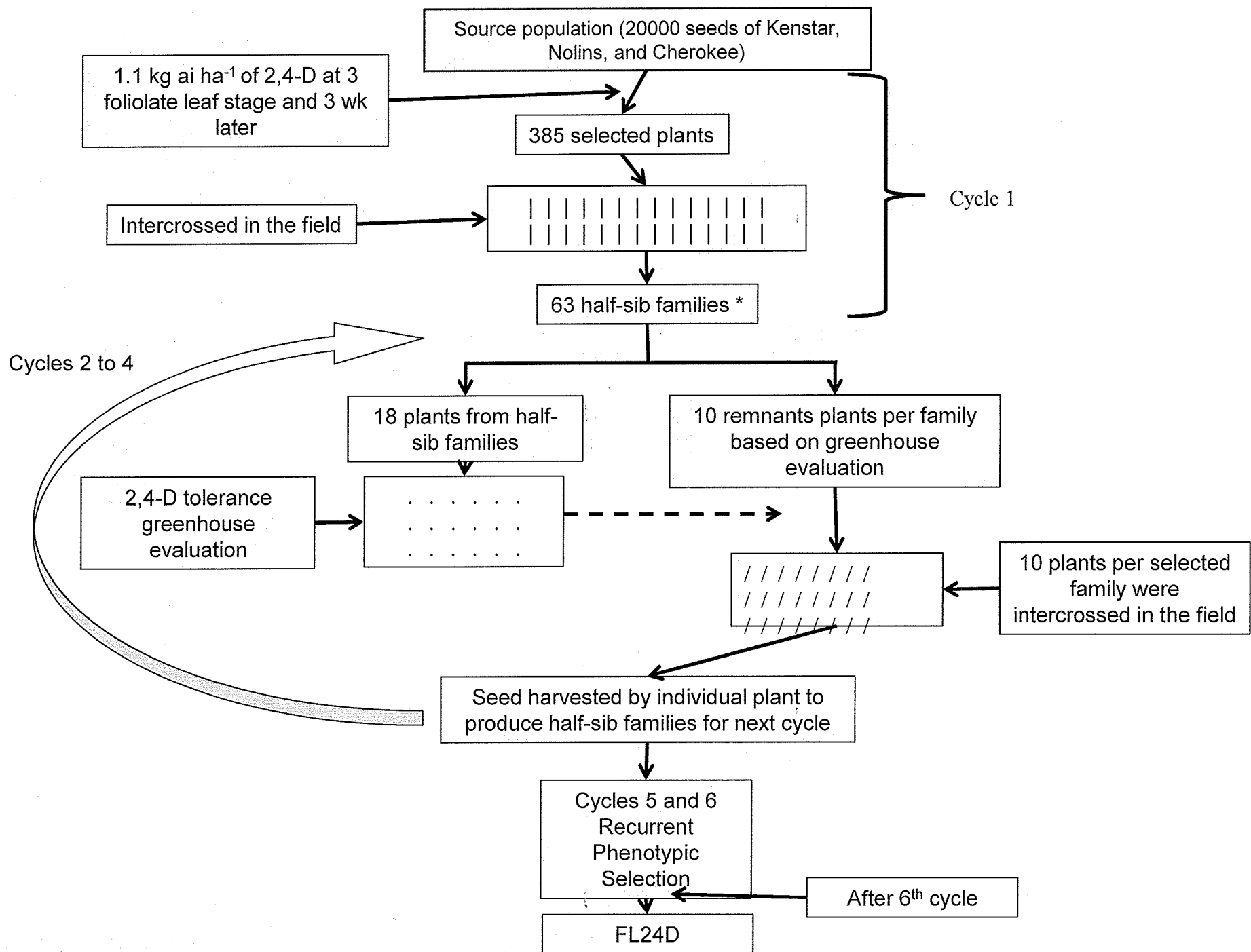
Approximately 20,000 plants were screened initially, spraying with 2,4-D at the trifoliate leaf stage.



385 plant were selected but only 63 produced sufficient seed for further testing.

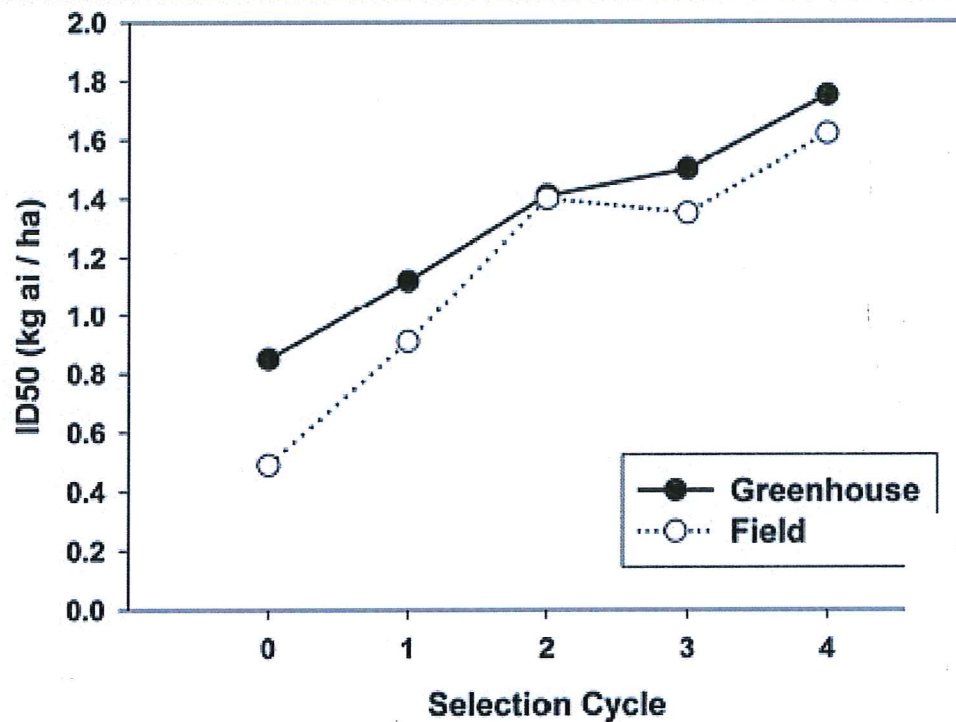
Four cycles of half sib family selection were completed by Taylor. We later completed two additional RRPS cycles.





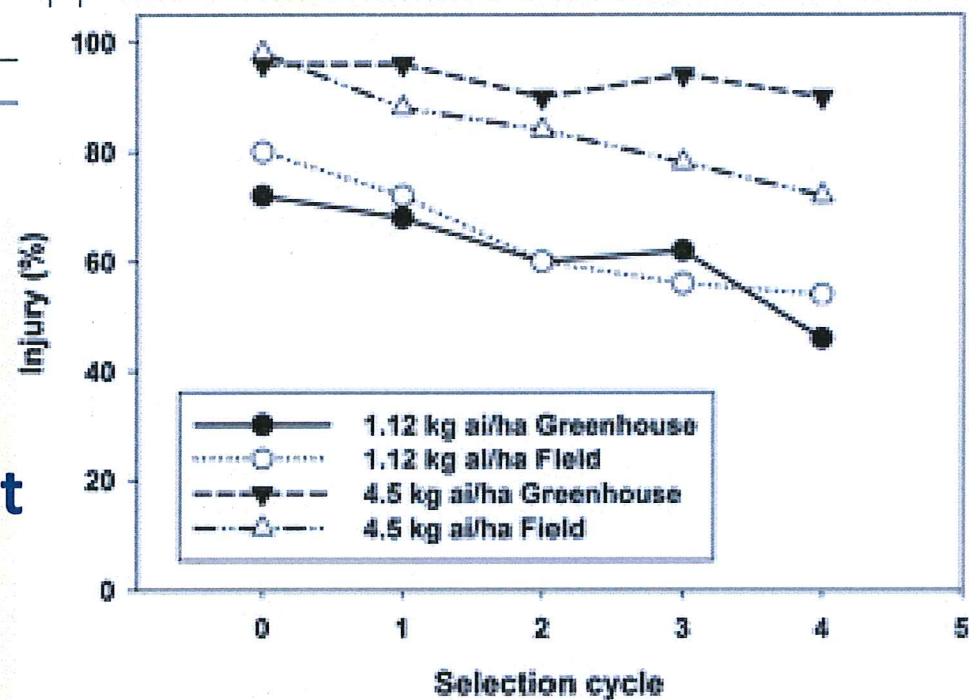


## 2,4-D Evaluation



**ID50= Rate required to cause 50% injury in the plant**

**% Injury in the plant**



Taylor et al 1989



## Greenhouse (GH) and Field (FD) Response of FL24D and Southern Belle (SB) to Rates of 24-D in 2012

Rate	GH 2012 10 day		GH 2012 17 day		FD 2013 14 day		FD 2013 28 day	
	FL24D	SB	FL24D	SB	FL24D	SB	FL24D	SB
<b>2x</b>	4.b	1.4b	4.9c	1.1b	5.5c	3.3c	6.2c	1.3c
<b>1x</b>	6.5b	1.9b	7.0b	1.2b	7.3b	4.9b	6.6c	1.9c
<b>1/2x</b>	6.4b	2.4b	6.7b	1.5b	7.7b	5.4b	7.4b	4.1b
<b>0x**</b>	8.8a	8.3a	9.0a	8.7a	9.0a	9.0a	9.0a	9.0a

Means within a column followed by the same letter are not significantly different, Duncan's Multiple Range Test,  $\alpha=0.05$

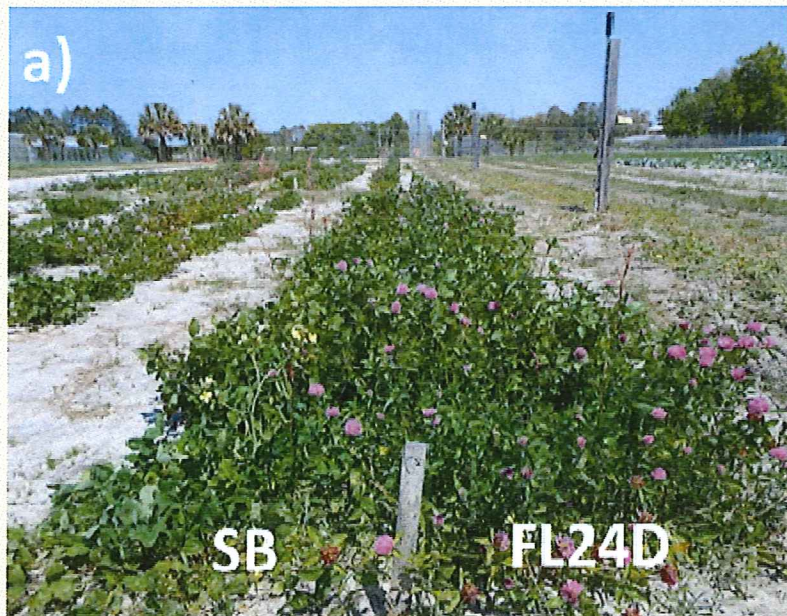
\*\* damage due to herbicide drift occurred on the GH experiment and explain values below 9 on the 0x application rate. Herbicide rates were: 1/2x=0.53, 1x=1.06 and 2x=2.12 Kg ha<sup>-1</sup>.



## 2,4-D Evaluation

After six cycles of selection;  
Southern Belle (SB) and the 2,4-D resistant cultivar (FL24D)  
2012-2013

Not Sprayed



Sprayed with 2,4-D (1X)

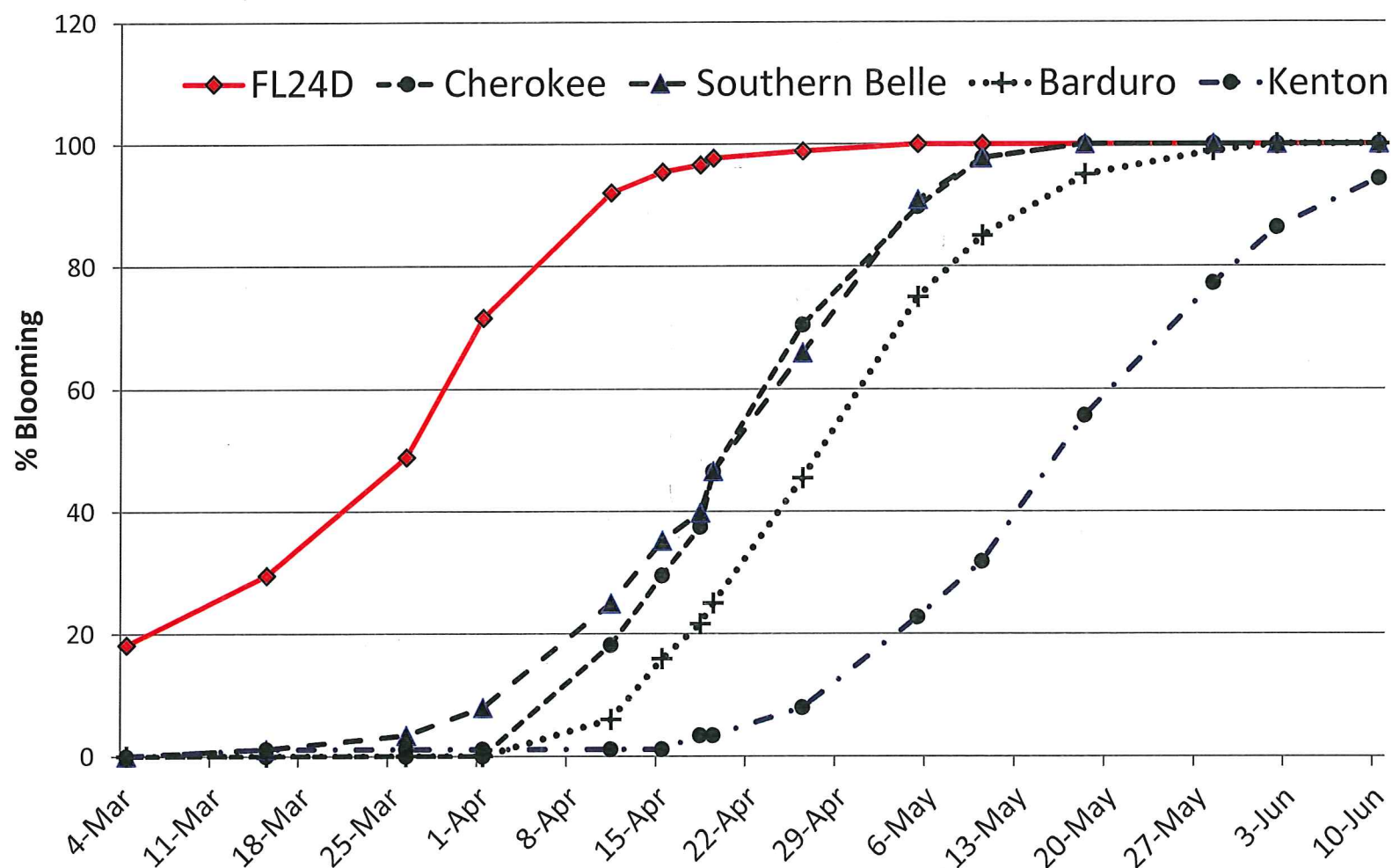




## Early Flowering

## No selection for Earliness

FL24D is the earliest red clover that we have tested





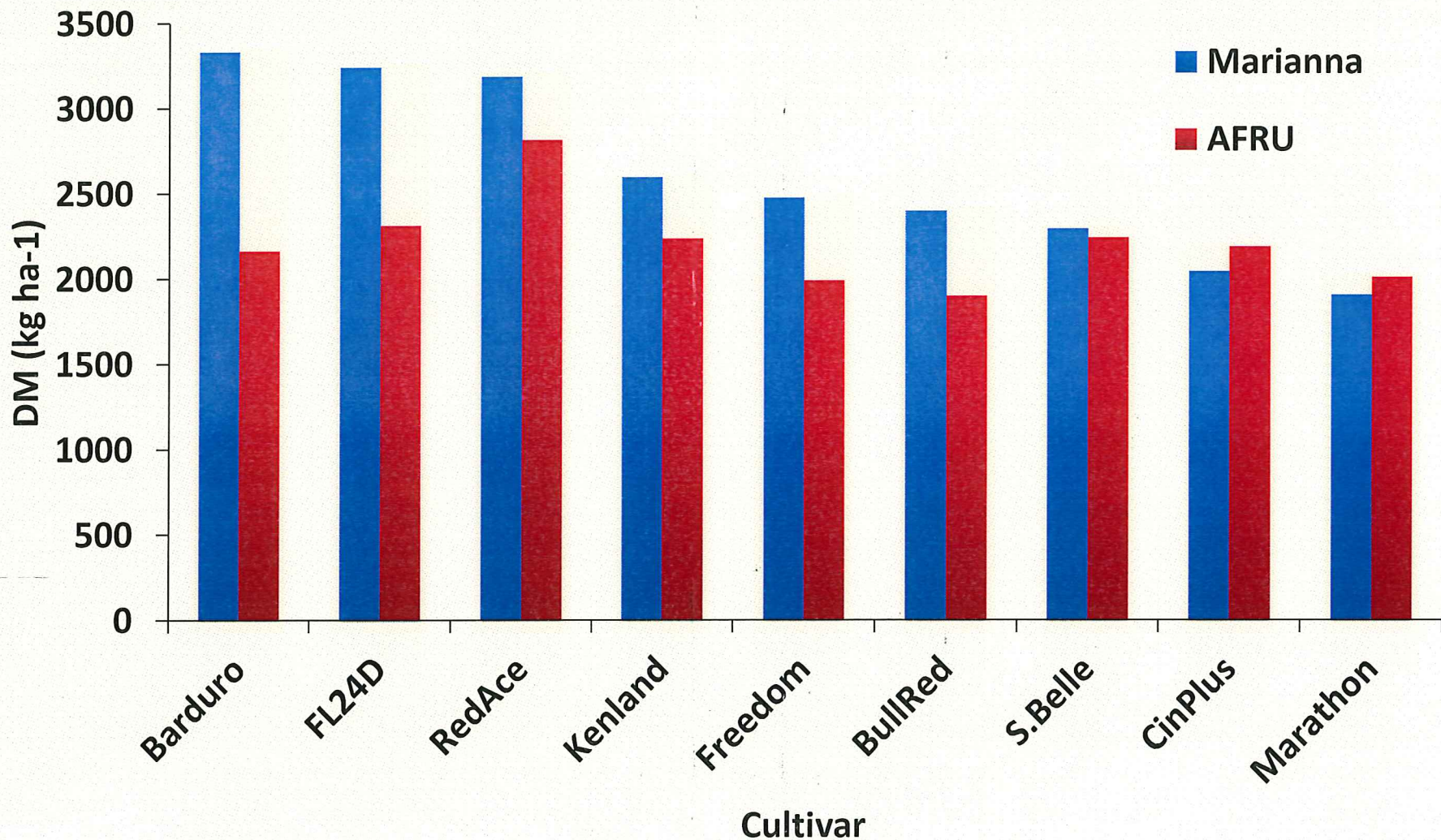
# Red Clover Cultivar Evaluation Experiment Gainesville, 2014

(Photo 29 April)





# Harvest 1 Dry Matter Yield for Nine Red Clover Cultivars at Two Locations, 2014





# Forage Evaluation

## Variety trial not sprayed 2009-2010

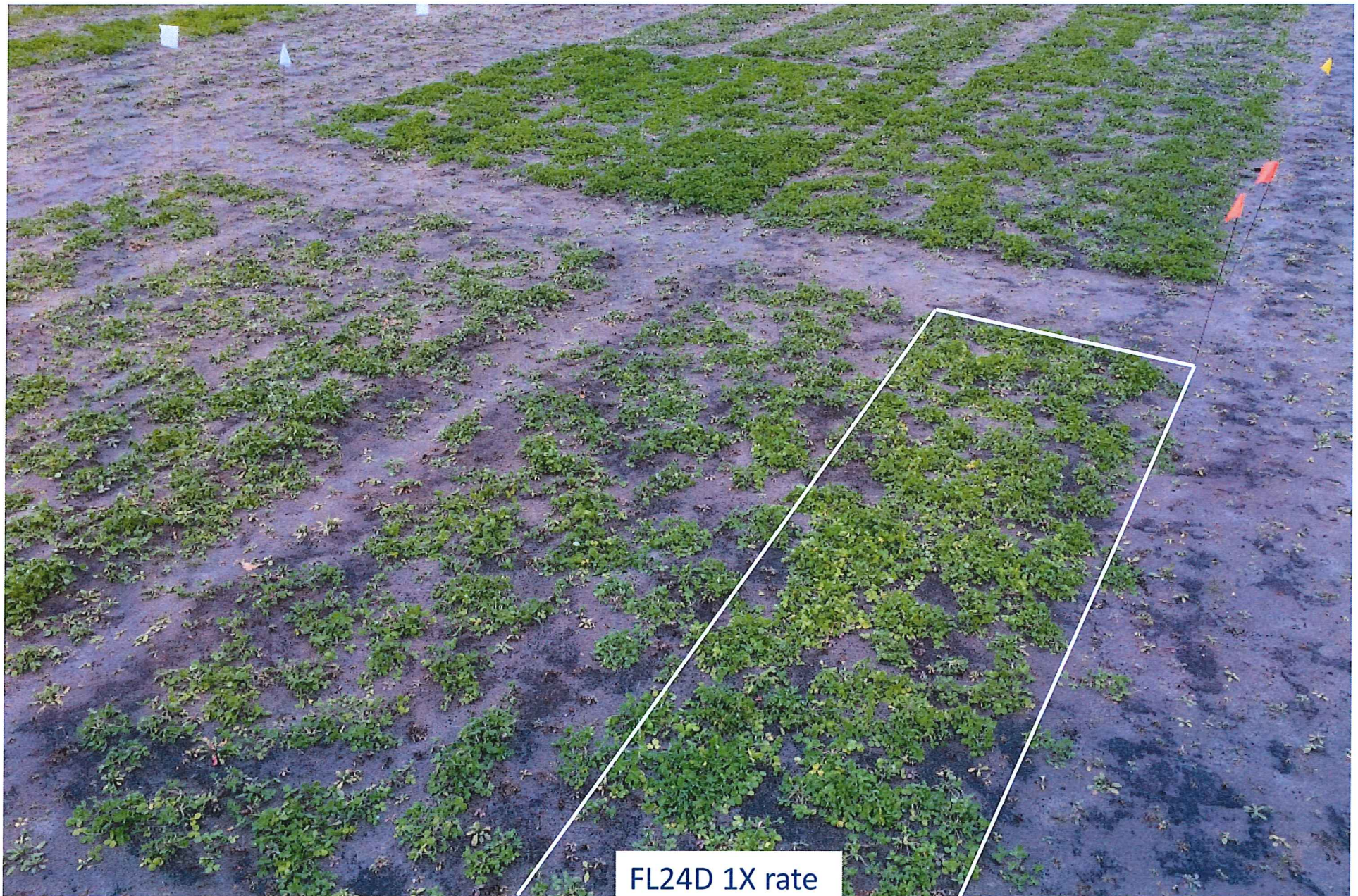
Entry	Harvest1 04/15/2010	Harvest2 05/20/2010	Total Harvest
Barduro	3790 a	1117 bc	4800 a
S. Belle	3680 a	1065 bc	4750 ab
Cherokee	3530 ab	1040 bc	4570 abc
<b>FL24D</b>	<b>3460 ab</b>	<b>1320 bc</b>	<b>4780 a</b>
Morningstar	3060 bc	820 cd	3880 cd
Freedom	2560 cd	1058 bc	3615 d
Kenton	2530 d	1430 b	3960 bcd
Redland Max	2300 d	1980 a	4271 abcd
Marathon	2240 d	1390 b	3630 d
Mean	3040	1157	4195

Means within a column followed by the same letter are not significantly different,  
Duncan's Multiple Range Test,  $\alpha=0.05$



# 2,4-D Herbicide Rate Experiment, Gainesville, 2014

(Photo 7 February)





# 2,4-D Herbicide Rate Experiment, Gainesville, 2014

(Photo 29 April)



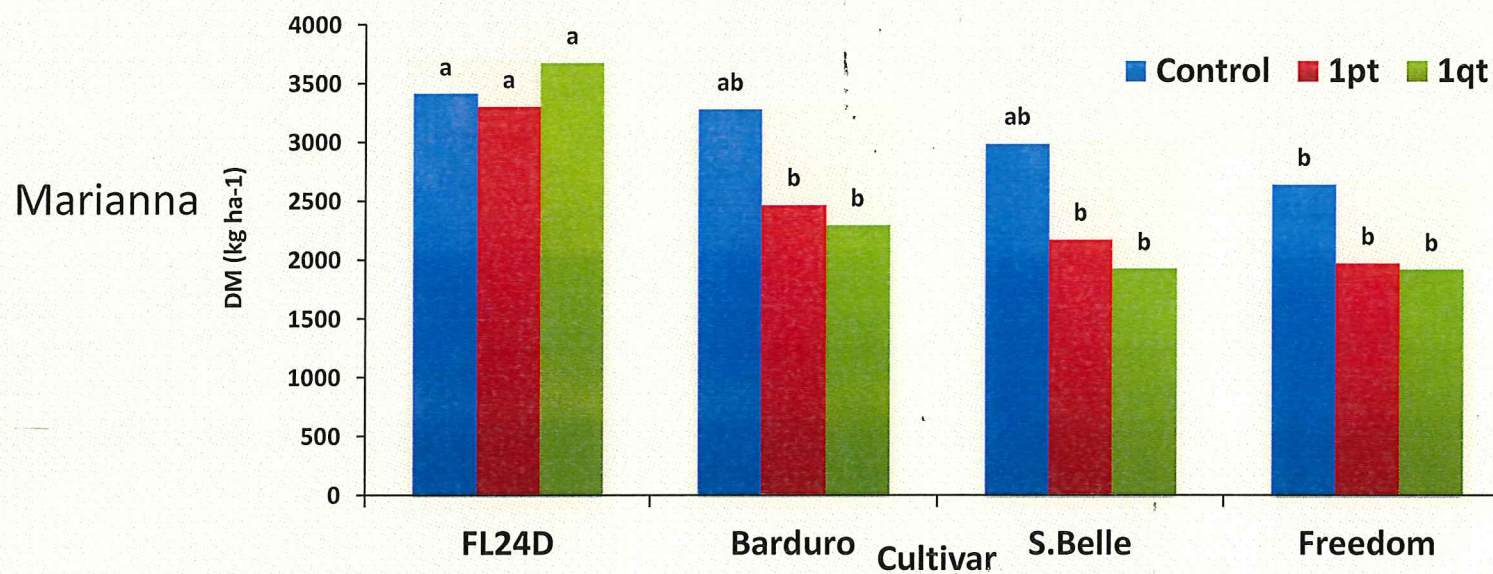
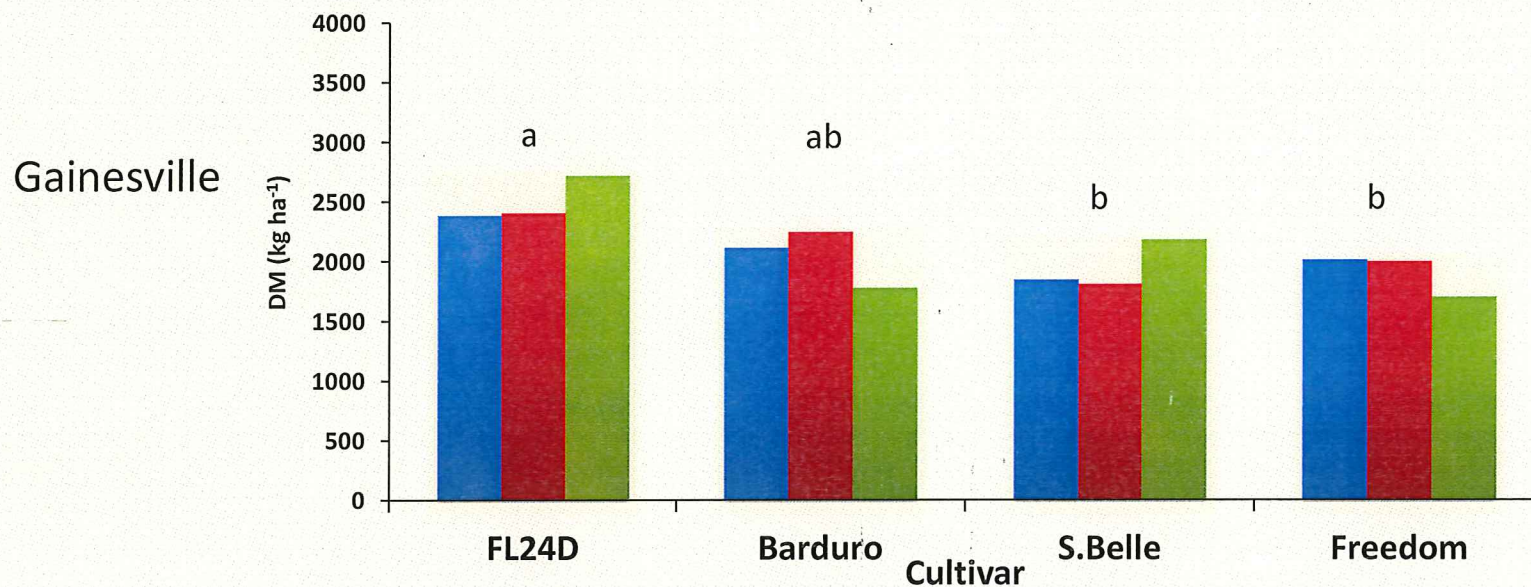






# Forage Evaluation

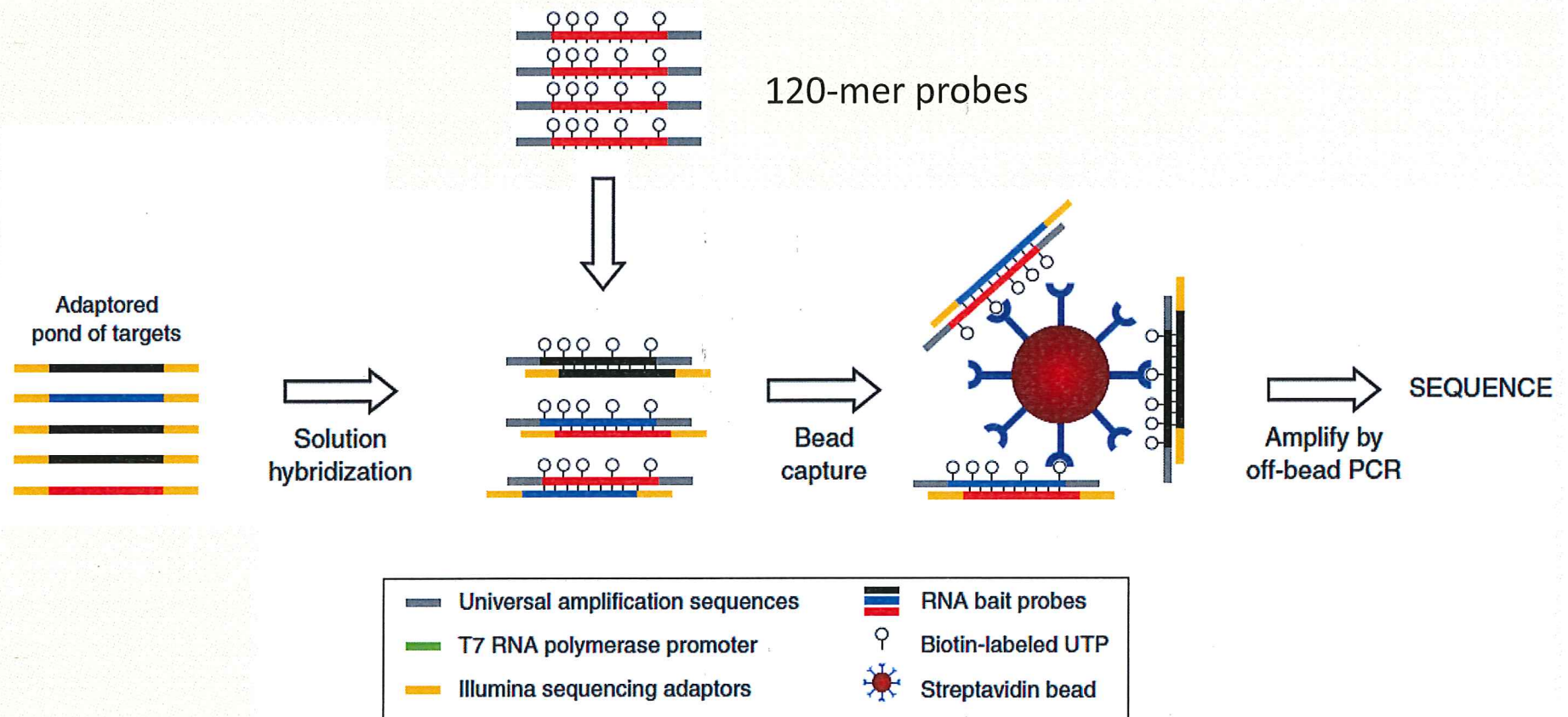
Variety trial sprayed 2013-2014





# Markers Plan

## Sequence/Exome Capture on ~47000 predicted genes (Istvánék et al 2014)



Fisher *et al.* **Genome Biology.** 12:1. 2011





BL	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120	121	122	123	124	125	126	127	128	129	130	131	132	133	134	135	136	137	138	139	140	141	142	143	144	145	146	147	148	149	150	151	152	153	154	155	156	157	158	159	160	161	162	163	164	165	166	167	168	169	170	171	172	173	174	175	176	177	178	179	180	181	182	183	184	185	186	187	188	189	190	191	192	193	194	195	196	197	198	199	200	201	202	203	204	205	206	207	208	209	210	211	212	213	214	215	216	217	218	219	220	221	222	223	224	225	226	227	228	229	230	231	232	233	234	235	236	237	238	239	240	241	242	243	244	245	246	247	248	249	250	251	252	253	254	255	256	257	258	259	260	261	262	263	264	265	266	267	268	269	270	271	272	273	274	275	276	277	278	279	280	281	282	283	284	285	286	287	288	289	290	291	292	293	294	295	296	297	298	299	300	301	302	303	304	305	306	307	308	309	310	311	312	313	314	315	316	317	318	319	320	321	322	323	324	325	326	327	328	329	330	331	332	333	334	335	336	337	338	339	340	341	342	343	344	345	346	347	348	349	350	351	352	353	354	355	356	357	358	359	360	361	362	363	364	365	366	367	368	369	370	371	372	373	374	375	376	377	378	379	380	381	382	383	384	385	386	387	388	389	390	391	392	393	394	395	396	397	398	399	400	401	402	403	404	405	406	407	408	409	410	411	412	413	414	415	416	417	418	419	420	421	422	423	424	425	426	427	428	429	430	431	432	433	434	435	436	437	438	439	440	441	442	443	444	445	446	447	448	449	450	451	452	453	454	455	456	457	458	459	460	461	462	463	464	465
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