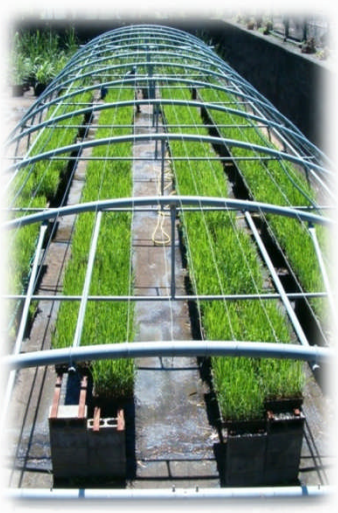





Optimization of sugarcane crossing and sowing processes in Réunion Island

Audrey Thong-Chane & al

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Facing challenges

- Improve the fertility of crosses
- Produce 80 000 seedlings for the 5 eRcane selection stations
- Supply the new eRcane selection station of Menciol with 15 000 seedlings
- Supply the fuzz for 7 emerging selection programmes in Africa. (56 000 seedlings)

TOTAL : 150 000 seedlings

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All steps from the panicle harvest to the seedlings rearing were analysed and optimised

I- Influence of the flowering location

II- Crossing process

III- Sowing process

IV- New tool for technical management

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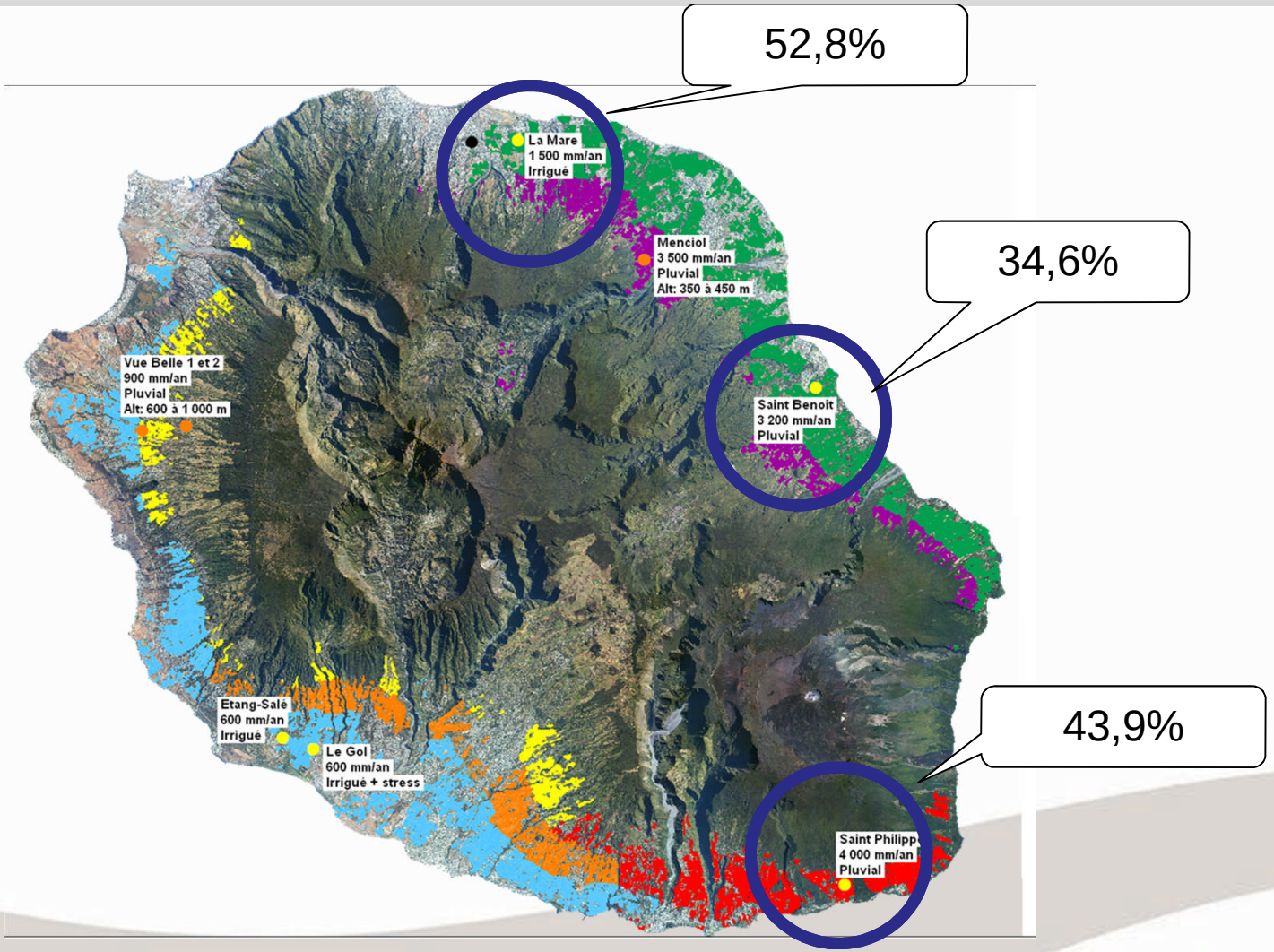
1- Influence of flowering location

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Comparison of panicle fertility between 3 eRcane stations

- From 2010-2015 (5 years)
- Panicles harvested in 3 eRcane stations : La Mare (LM), St Benoit (SB) and St Philippe (SP) in specific flowering fields.

3 locations and % flowering varieties in 2010



% of varieties flowering : LM>SP>SB

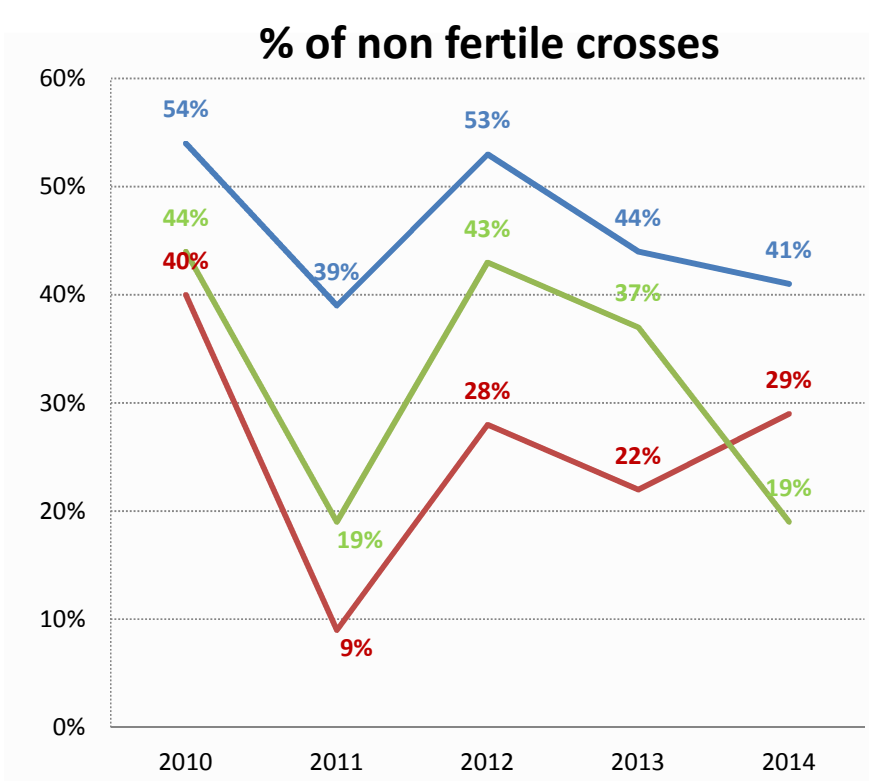
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Comparison of panicle fertility between 3 eRcane stations

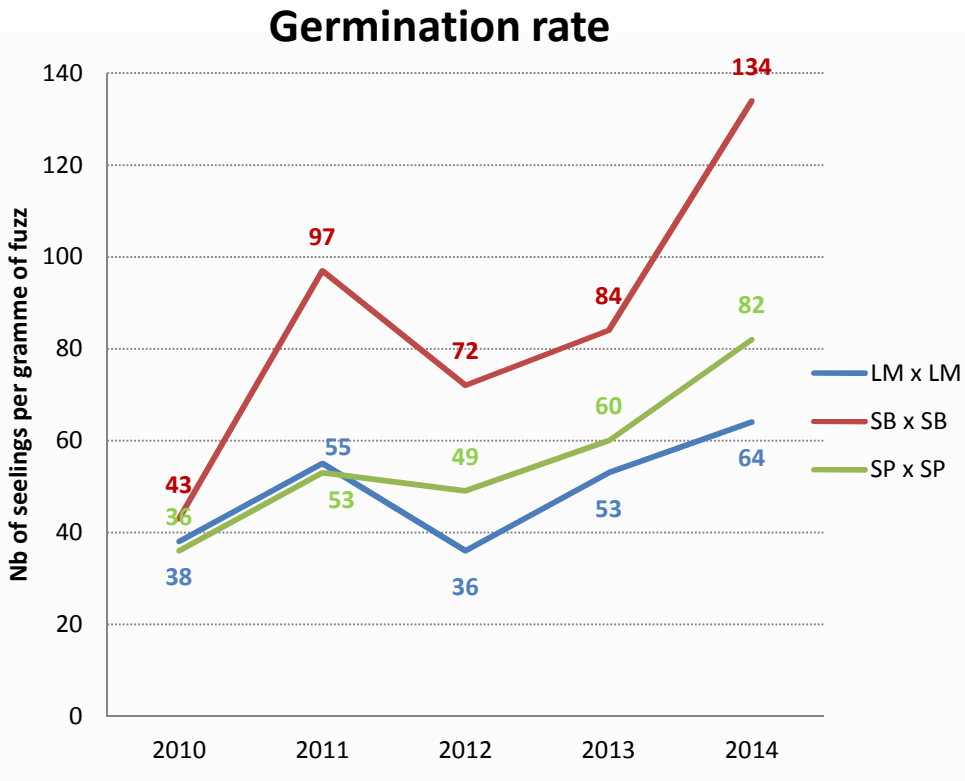
- From 2010-2015 (5 years)
- panicles harvested in 3 eRcane stations : La Mare (LM), St Benoit (SB) and St Philippe (SP) (specific flowering fields)

- More than 5000 crosses observed
- Measures : % of non fertiles crosses, mean germination rates/g of fuzz

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LM>SP>SB



SB>SP>LM

Planting specific flowering fields in SB and SP each year

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II- Crossing process

II-1 Refining the estimation of male fertility level

- Before : % of opened anthere to estimate the male fertility
- Add pollen staining (Alexander)

Step 1 : opened anthers

% of open anthers	Sex	score
95% à 100%	Male	9
85% à 94%	Male	8
70% à 84%	Male	7
55% à 69%	weak male	6
40% à 54%	weak male	5
20% à 39%	weak male	4
15% à 19%	Female	3
5% à 14%	Female	2
0% à 4%	Female	1

Step 2 : pollen staining

% of fertile pollen	Sexe
51% à 100%	Male
16 à 50%	Indefinite
0 à 15 %	Female

II-2 Concentration of the Hawaïan Solution

	Standard solution	solution x 1,5
Phosphoric Acid (ppm)	50	75
Nitric Acid (ppm)	25	38
Sulfuric Acid (ppm)	25	38
SO2 (ppm)	100	150





	Standard solution	solution x 1,5
Nb crosses	418	374
% of died female panicle during hybridization	27%	25%
% non fertile crosses	41%	36%
germination rate (nb seedlings/g of fuzz)	54	64

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II-3 A better control of the synchronisation during the hybridization

- After one week in lanterns, estimation of the percentage of opened flowers on the male and female panicles.
- If male arrows are completely opened whereas female are still developping; new males are introduced in the lantern

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II-4 Fuzz drying

- 2009 : drying the fuzz during 24H à 40°C
- Test of 48 H in 2010

drying time	Nb of crosses	% of non fertiles crosses	Mean nb of seedling/g of fuzz
24 hours	24	74%	26
48 hours	23	67%	58

⇨ Since 2010, drying time increased to 48 h.

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III-Sowing process

III-1 Test of new substrates

Standard substrate :

- 1/3 soil, 1/3 peat moss, 1/3 vulcano stones;
- Chemical disinfection (Dazomet=Basamid).

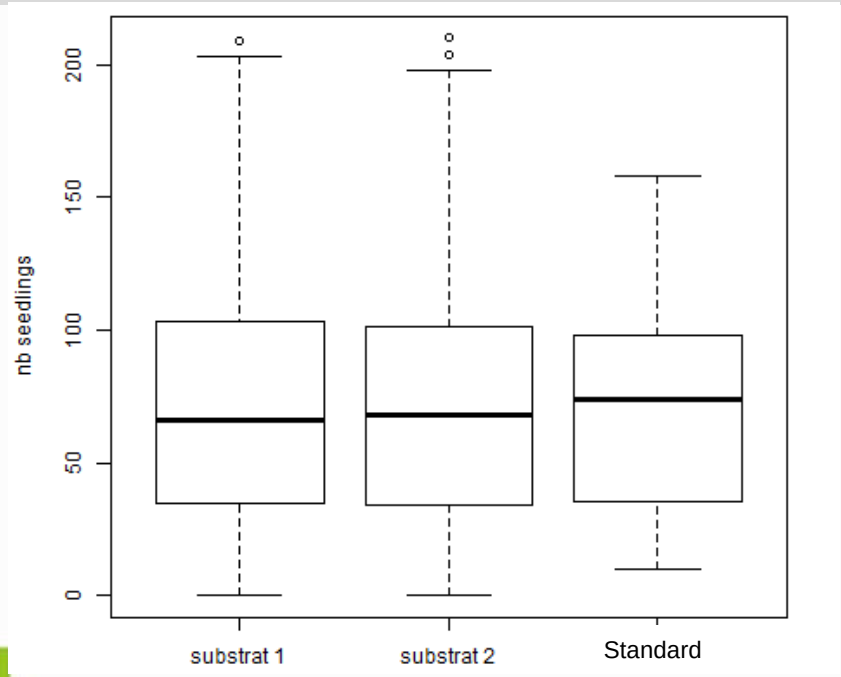
Test of 2 new substrates :

- Substrate 1 : 1/3 sand, 2/3 peat moss;
- Substrate 2 : 1/3 vulcanic stones, 2/3 peat moss.

Comparison of germination for 168 differents crosses.

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III-1 Test of new substrates





	Degree of freedom	F	Pr(>F)	
Substrate	2	0,3874	0,6791	
cross	167	10,911	<2 e -16	***
Substrate x cross	167	0,7776	0.9803	

Fischer test on likelyhood ratio

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III-2 Set up of a germination room

- Tests to compare the fuzz germination in the glasshouse vs in the germination room

- Germination Room:
- Constant temperature
- Lights 24h/24



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III-2 Set up of a germination room

- Temperatures tested : 30, 32 and 36 °C.
- Numerous crosses representative of the variability of germination rate in 2011
- 4 replicates (1g of fuzz) in each modality
- Seedling counting :14 days after sowing

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Results at 3 days

Cross	Class of germination	J+3 (36°C) p-value			Cross	J+3 (32°C) p-value			Cross	J+3 (30°C) p-value		
1084	0	0,1248			**553**	1			**917**	$1.429e^{-05}$	***	*GR*
1094	0	0,1248			**915**	0.2184			**2357**	0.0004445	***	*GR*
1498	1	0,06234			**1016**	$5.427e^{-06}$	***	*GR*	**756**	$6.093e^{-12}$	***	*GR*
1165	1	3,65e-06	***	*GR*	**1588**	$4.11e^{-10}$	***	*GR*	**1171**	0.002303	**	*GR*
1996	2	0,4999			**1583**	0.0004803	***	*GR*	**1715**	$2.354e^{-06}$	***	*GR*
918	2	1			**1974**	0.03113	*	*GR*	**2221**	0.000507	***	*GR*
2105	3	$2,24^{e}$-07	***	*GR*	**1622**	$1.273e^{-09}$	***	*GR*	**1177**	$7.11e^{-06}$	***	*GR*
1703	3	$<2,2e^{-16}$	***	*GR*	**1946**	0.0002137	***	*GR*	**1460**	$3.68e^{-15}$	***	*GR*
1235	4	4,11e-10	***	*GR*	**1261**	$2.768e^{-16}$	***	*GR*	**1027**	$2.509e^{-05}$	***	*GR*
185	4	$<2,2e^{-16}$	***	*GR*	**1676**	$<2.2e^{-16}$	***	*GR*	**2094**	0.294		
164	5	$<2,2e^{-16}$	***	*GR*	**1958**	0.2184			**951**	0.01311	*	*GR*
2051	5	$<2,2e^{-16}$	***	*GR*	**2222**	0.0001196	***	*GR*	**2378**	0.1247		
1019	6	$<2,2e^{-16}$	***	*GR*	**1188**	$<2.2e^{-16}$	***	*GR*	**898**	$<2.2e^{-16}$	***	*GR*
793	6	$<2,2e^{-16}$	***	*GR*	**1436**	$<2.2e^{-16}$	***	*GR*	**1591**	$<2.2e^{-16}$	***	*GR*

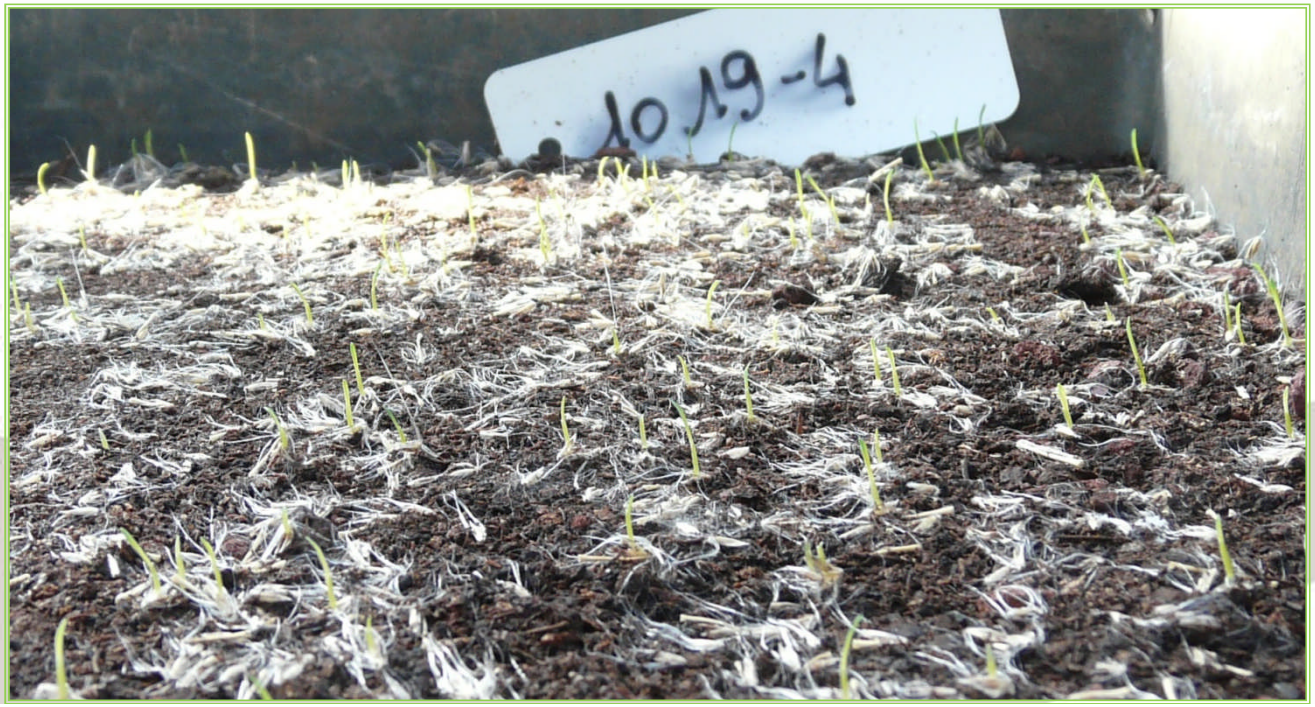
*** = 0,001 , ** = 0,01, * =0,05

GR : Germination room>Glasshousse,
GH : Glasshousse>Germination room

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J+3 Glasshouse



J+3 Germination room

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Results at 14 days

Class of germination	Cross	J+14 (36°C) p-value	Cross	J+14 (32°C) p-value	Cross	J+14 (30°C) p-value
0	**A1**	0,01889 * *GH*	**B1**	1	**C1**	0.3921
0	**A2**	0,03271 * *GH*	**B2**	1	**C2**	0.2458
1	**A3**	0,0177 * *GH*	**B3**	0.5806	**C3**	0.05307
1	**A4**	0,5411	**B4**	0.6919	**C4**	0.2792
2	**A5**	0,001191 ** *GH*	**B5**	0.1205	**C5**	0.1488
2	**A6**	0,3573	**B6**	0.03418 * *GR*	**C6**	0.8027
3	**A7**	0,000506 *** *GH*	**B7**	0.332	**C7**	0.1042
3	**A8**	0,004174 ** *GH*	**B8**	0.9315	**C8**	0.1009
4	**A9**	0,2832	**B9**	0.0003199 *** *GR*	**C9**	0.01722 * *GR*
4	**A10**	0,4015	**B10**	0.3568	**C10**	$<2.2e^{-16}$ *** *GH*
5	**A11**	0,1196	**B11**	0.738	**C11**	0.3455
5	**A12**	0,3986	**B12**	0.4726	**C12**	0.7907
6	**A13**	0,07428	**B13**	$7.897e^{-06}$ *** *GR*	**C13**	0.1953
6	**A14**	0,002978 ** *GH*	**B14**	0.4279	**C14**	0.8525

*** = 0,001 , ** = 0,01, * =0,05

GR : Germination room>Glasshousse,

GH : Glasshousse>Germination room

Fischer test on likelyhood ratio

➡ Germination room is settled at 32°C

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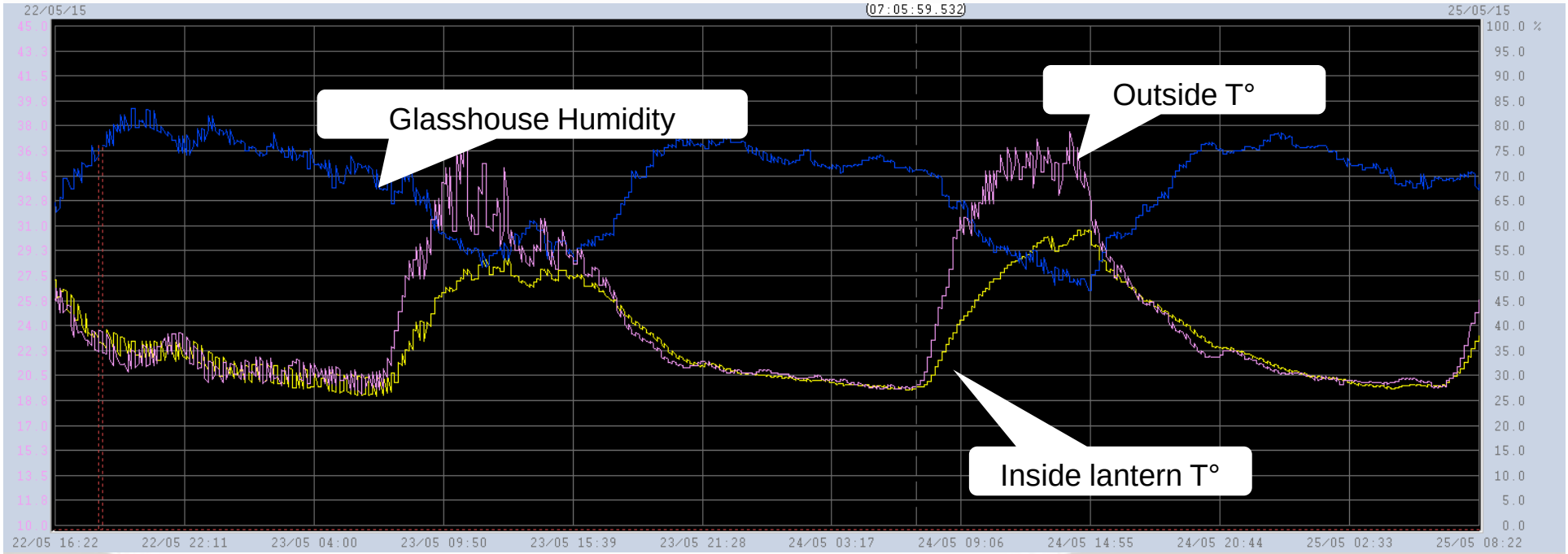
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IV- New tools to optimize the technical management

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4-1 Probes to pilote the climate in the glasshouse

- Temperature and humidity probes with automatic records on computer in real time.
- Used during the hybridization & seedlings rearing.



Modulation of humidity and Temperature :

- **Opening/closing windows**
- **Drop irrigation on the soil**

4-2 Improve of the traceability of the crosses

- Implementation of computerized data base that managed all the hybridization/sowing processes from panicle harvest to the dispatching of the seedlings in the different stations
- Bar codes on trays & on printed labels on the panicles/fuzz







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4-2 Improve of the traceability of the crosses

Use of a PDA in the glasshouse :

- To record tracking bar codes from cross to trays.
- To record the number of the seedlings per tray
- To build maps of the trays in the glasshouse/rearing area



g11	g12	g13	g14	g15	g16	g17	g18	g19	g20	g21	g22	g23	g24	sowing date
1584	1503	1547	1540	1420	1122	1041	1079	927	980	252	288	61	34	18/08/2014
1511	1585	1576	1566	1474	1196	1078	1018	998	962	280	293	56	21	
1583	1507	1548	1542	1455	1147	1071	1074	943	985	257	244	70	3	19/08/2014
1526	1504	1536	1512	1412	1165	1030	1026	965	960	851	251	57	45	
1595	1515	1592	1534	1496	1182	1017	1015	156		897	271		41	20/08/2014
1598	1501	1581	1590	1479	1110	1012	1006	984	988	816	240	48	76	
1579	1594	1519	1560	1449	1181	1085	1060	976	975	826	273	49	65	22/08/2014
1565	1525	1580	1529	1418	1176	1021	1049	967	952	872	285	6	23	
1558	1520	1508	1556	1461	1178	1059	1081	999	992	852	213	58	79	25/08/2014
1502	1531	1532	1516	1499	1067	1094	1091	961		857	268		83	
1596	1510	1521	1543	1421	1125	1068	1069	989	912	860	233	151	22	26/08/2014
1514	1569	1550	1530	1409	1156	1099	1011	903	940	820	265	159	43	
1500	1571	1518	1533	1495	1108	1037	1053	942	974	847	186	124	82	
1564	1586	1522	1597	1481	1157	1082	981	937	941	804	145	134	78	
1577	1517	1513	1593	1440	1117	1088	994	931		817	188		92	
1557	1554	1509	1567	1486	1111	1027	939	949	966	859	164	168	81	
1582	1570	1505	1527	1419	1190	1009	935	925	991	854	158	139	90	
1538	1537	1506	1528	1493	1160	1051	959	958	837	898	131	147	60	
345	1553	1539	1561	1435	1032	1022	944	901	877	878	133	171	96	
367	1568	1559	1572	1478	1077	1092	993	987		801	166		35	
369	1549	1563	1400	1441	1061	1045	924	953	849	880	112	140	67	
323	1562	1523	1483	1485	1089	1056	926	951	808	896	153	105	88	
304	1599	1545	1484	1488	1096	1065	900	982	867	873	144	104	9	
384	1524	1587	1405	1445	1007	1064	946	911	845	810	191	190	63	

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Global Results

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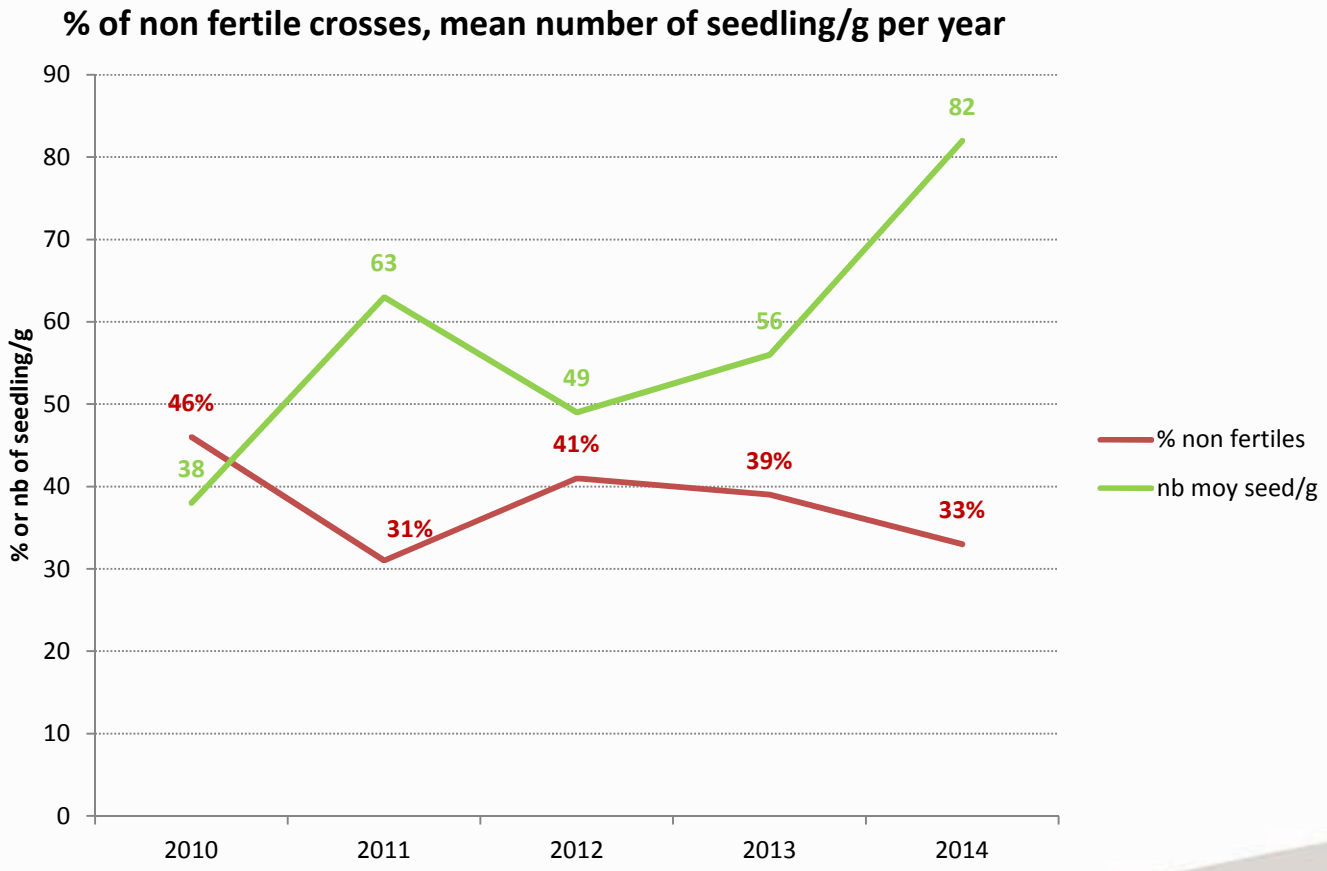
SINCE 2010

With the same team (1 engineer, 2,5 technicians, 2,5 workers) :

- nb of fertilised panicles increased from 2000 to 2500.
- Gain of time during the campaign production of seedlings thanks to the germination room.

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Improvement of the fertility of the crosses



- 13% of non fertile crosses
+ 44 seedlings/g of fuzz

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Thank You! Questions?



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