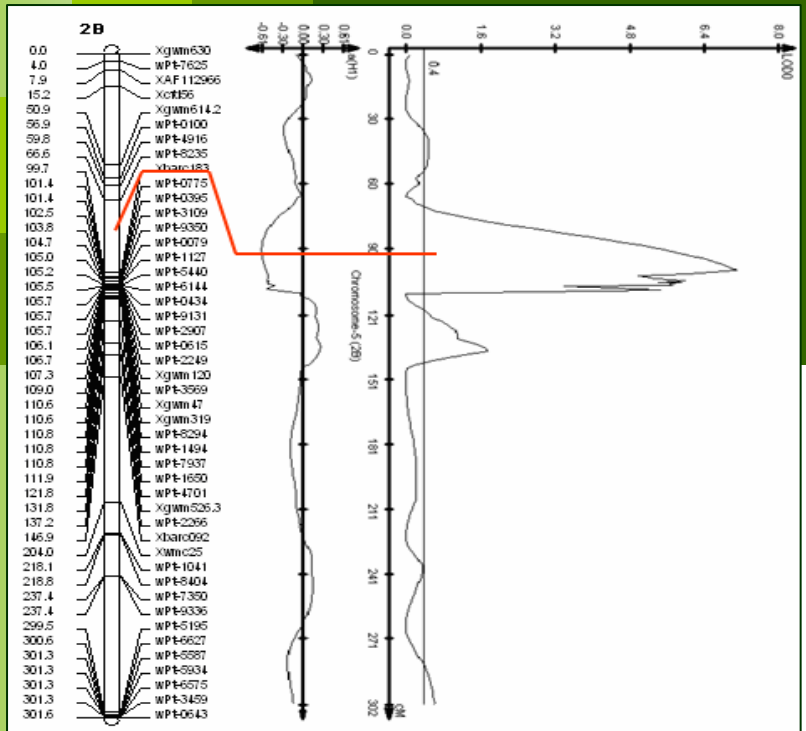
From Genes to Twinkies



Map of wheat chromosome 2B, and likely location of gene affecting flour water absorption

Mapping milling and flour traits in soft wheat

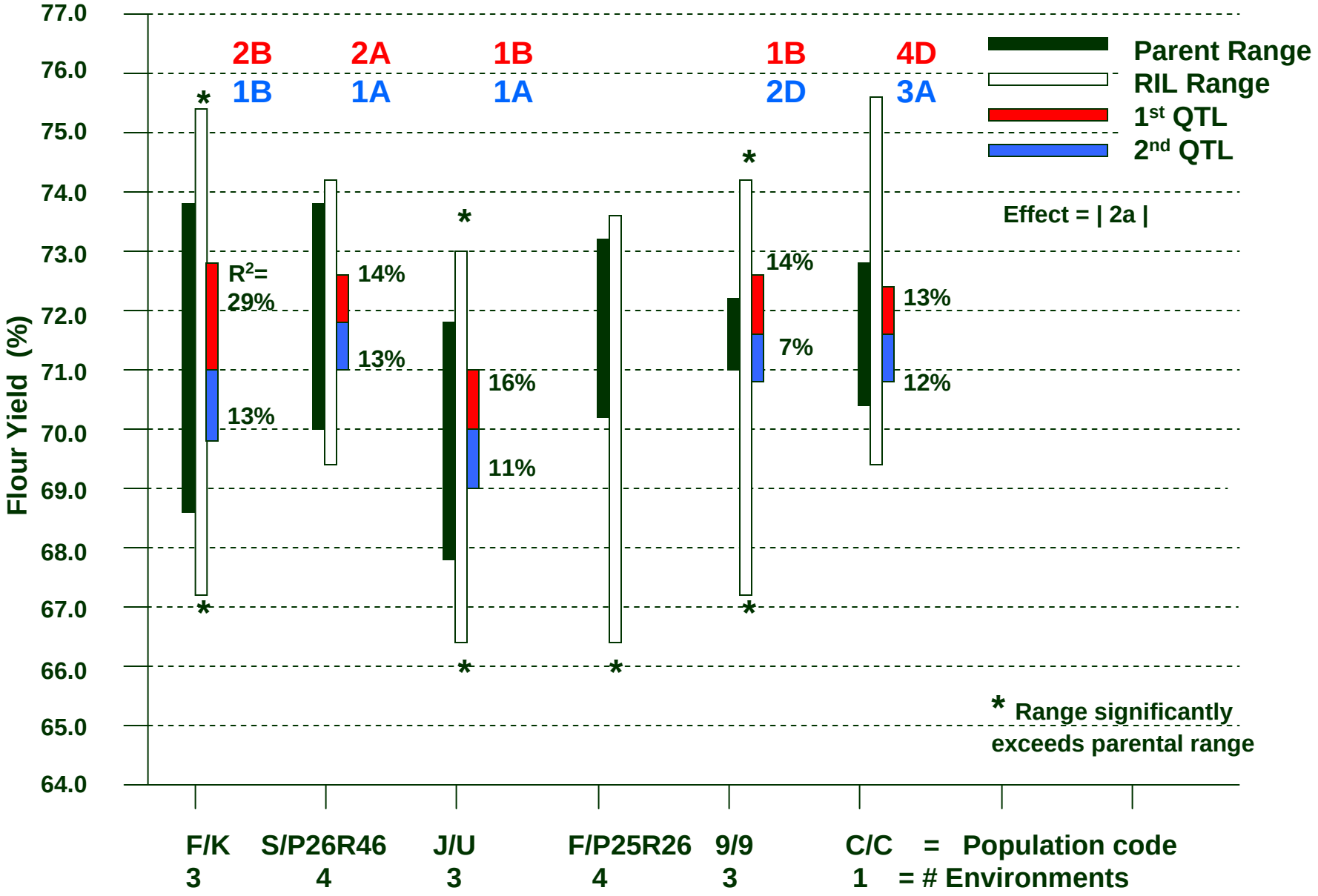
Edward Souza, USDA-ARS and Clay Sneller, Ohio State University

- Review of work with mapping populations
  - Funding: NRI Wheat CAP – Eastern US Wheat Researchers
  - Wooster – M. Guttieri, D. Hua
- Association mapping populations
  - Funding: OSU, USDA-ARS w/ VA Tech, Cornell, Purdue, U. Ky
  - Wooster – N. Smith, A. Sturbaum, M. Guttieri, D. Hua, A. Bugaj, S. Carson, S. Croskey
  - Collaborators – C. Griffey, M. Sorrells, H. Ohm, D. Van Sanford
- Genes of Interest – Arabinoxylan composition
  - Funding: USDA-ARS with Kraft Foods
  - Wooster – M. Guttieri, A. Bugaj

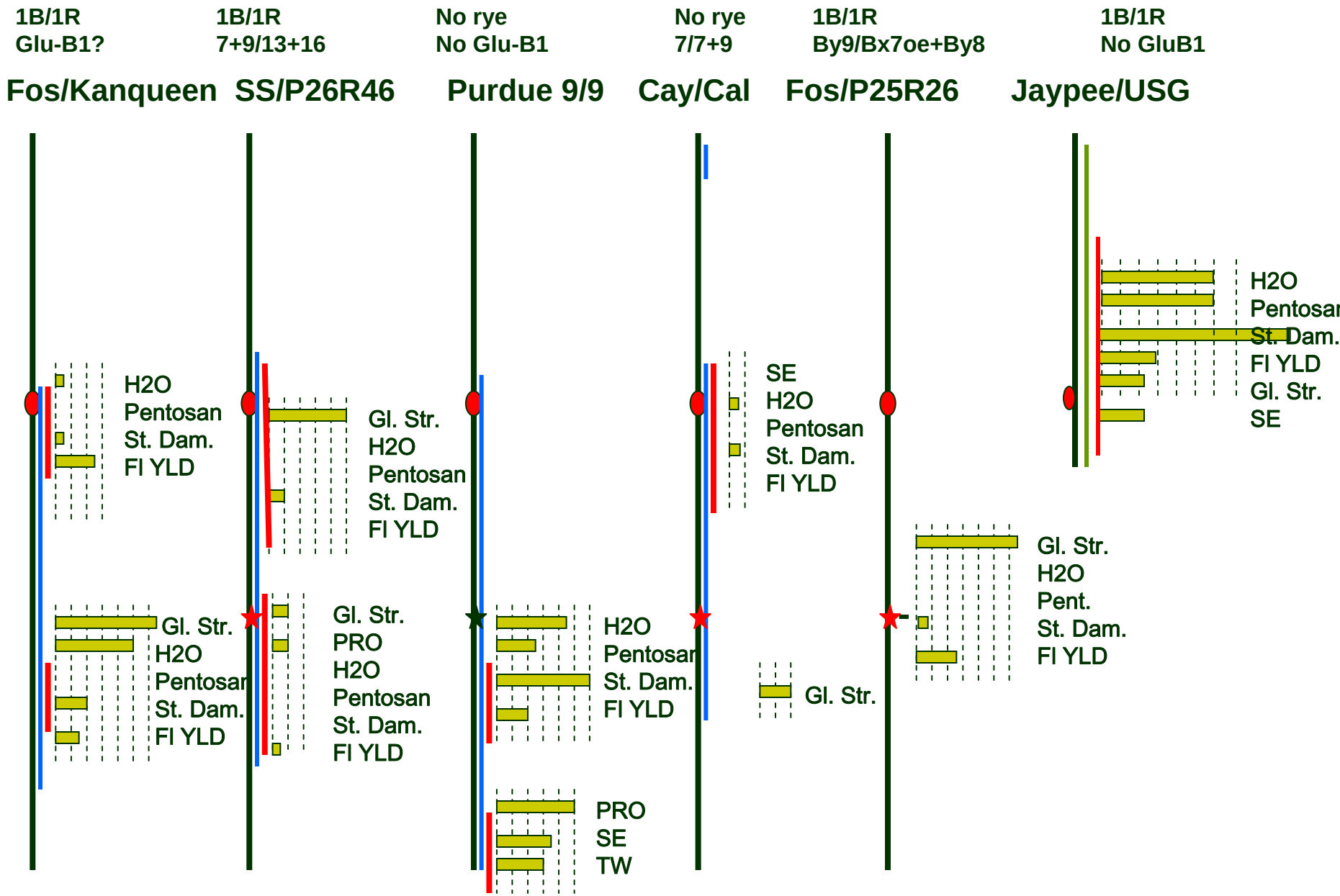
Populations & Phenotyping

	Pedigree	Envs done	Envs to do	Samples done	Samples to do	Total
1	**Foster/Kanqueen**	**NY05, NY06, OH06**	**NY07**	720	240	960
2	**SS 550/PIO 26R46**	**OH02, OH03, OH03, OH04**	**GA08, GA08**	600	300	900
3	**MO94-317/Ernie**	**None**	**None**			0
4	**Jaypee/USG 3209**	**@VA05, @NC05, VA06**	**VA07, KY07**	650	300	950
5	**Foster/PIO 25R26**	**OH05, OH06, NY06, OH07**	**OH08**	720	180	900
6	**92201/91193**	**NY05, IN05, OH05**	**IN07**	600	200	800
7	**Cayuga/Caledonia**	**NY05, NY06 *NY03, *NY04**	**NY08**	310	155	465

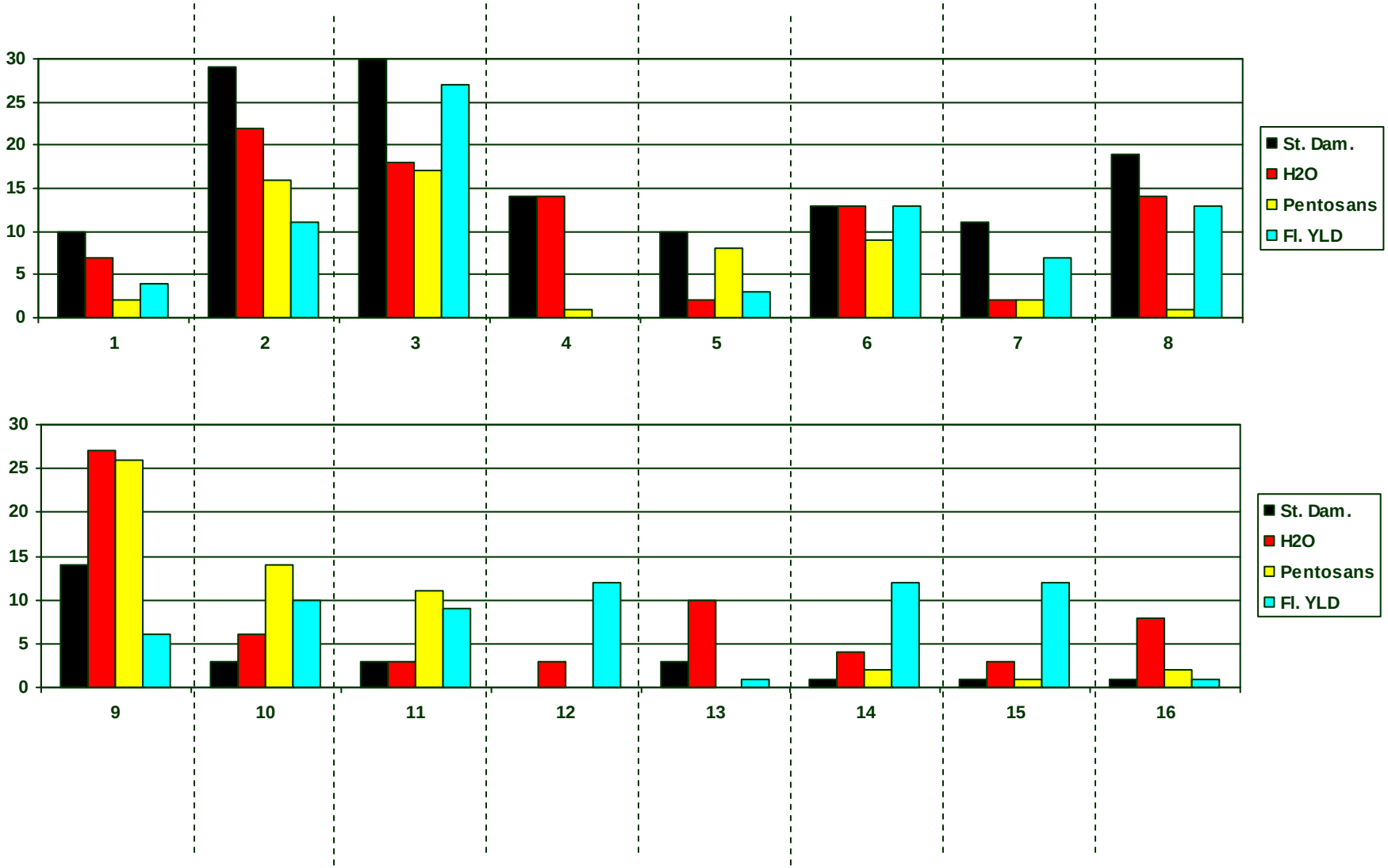
@, 109 of 130 lines in pop were phenotyped*, only did sodium carbonate on whole grain


Flour Yield (%)
77.0
76.0
75.0
74.0
73.0
72.0
71.0
70.0
69.0
68.0
67.0
66.0
65.0
64.0
2B
1B
2A
1A
1B
1A
1B
2D
4D
3A
Parent Range
RIL Range
1st QTL
2nd QTL
Effect = | 2a |
R2= 29%
13%
14%
13%
16%
11%
14%
7%
13%
12%
* Range significantly exceeds parental range
F/K
3
S/P26R46
4
J/U
3
F/P25R26
4
9/9
3
C/C = Population code
1 = # Environments

Chromosome 1B and known genes which segregate in cross



R^2 of QTLs affecting water absorption-flour yield complex



Conclusions

- Ample heritable variation in many soft wheat populations
- Transgressive segregants are common
- Complex inheritance despite some major genes
  - Transgressive segregants
  - Many QTLs
  - Much of progeny range is not accounted for by QTL analysis
- Some consistent effects: HMW glutenins, 2B, others
- Non-independence of some effects
  - HMW Glutenins
  - Starch damage and other
- Some genetic effects for SRC are large enough for MAS
  - Lactic acid: 10%, Water: 2%, Na_2CO_3: 5%, Flour Yield: 1.5%

Association Mapping of Quality in Eastern US Soft Winter Wheat

2007 Evaluation of 192 cultivars

Ohio, Indiana, Virginia, New York, Kentucky, and N. Carolina

Association Mapping (AM Study)

- Genes that are linked and enter the germplasm pool from the same source will remain linked through cycles of breeding
- Mapping across a large number cultivars can identify markers of value in a wide range of breeding programs
- Validation of bi-parental mapping populations of Wheat CAP

Composition of the AM Study

Source State	# Entries	Release date range	Region
ONT, MI, NY	35	1881 - 2001	North
IN, OH	72	1808 - 2005	Eastern Corn-belt
IL, KS, WI	11	1881 - 2001	Western Corn-belt
VA, NC, MD, PA	28	1837 - 2002	Mid-Atlantic
MO, KY, TN	9	1837 - 2002	Central
GA, SC, FL	23	1881 - 2001	South East
AR, LA, TX	15	1881 - 2003	Delta

Composition of the AM Study

Release period	# Entries
Before 1900	12
1900-1949	20
1950-1969	16
1970s	28
1980s	37
1990s	53
2000s	27

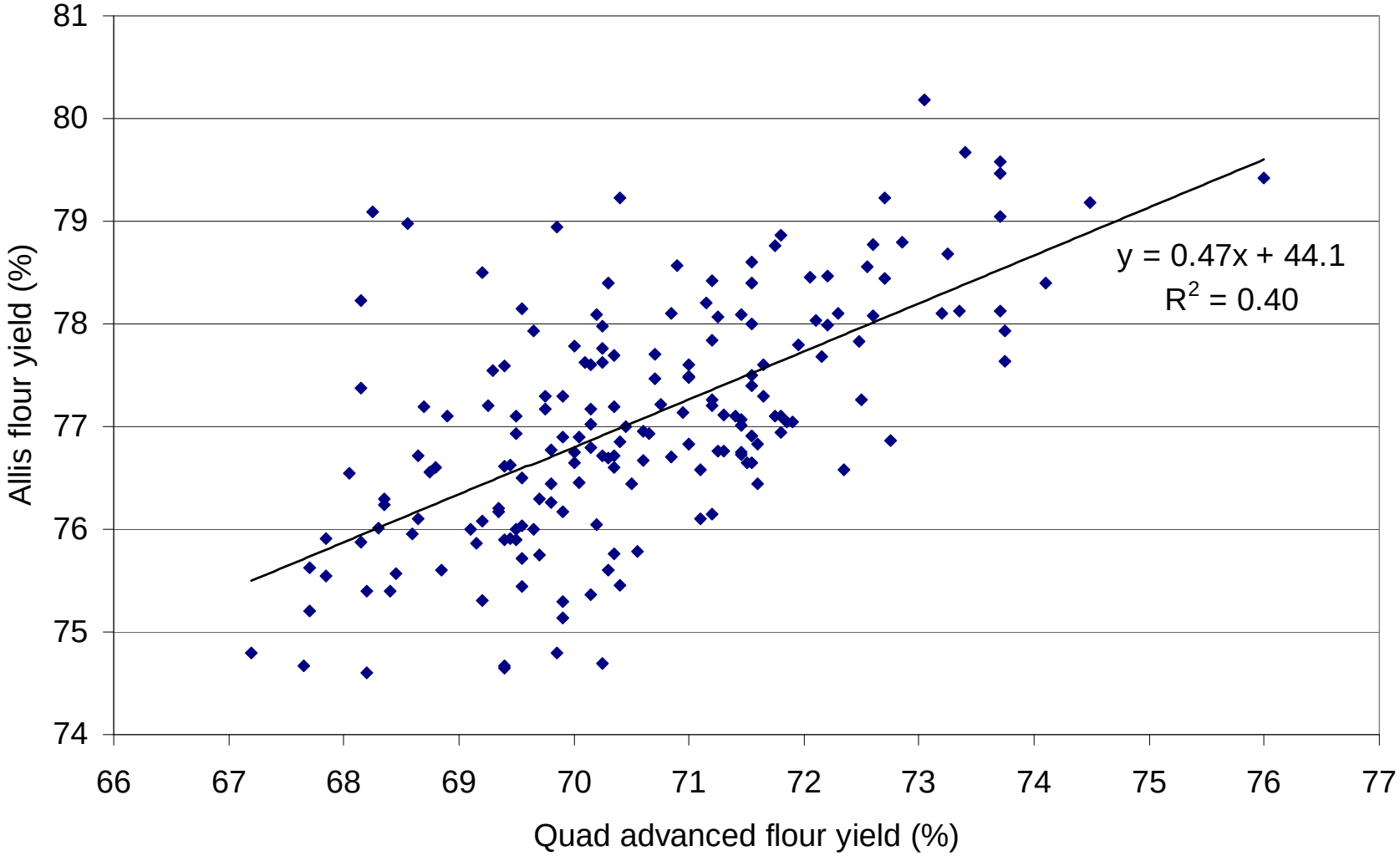
Planned Evaluation in 2007 and 2008

- Quad advanced milling
  - Four solvent SRC from all locations
  - Cookie bake from 3 locations in 2007 and 2008

- Quality Data at Core of New Database
  - Supplement Allis Database
  - Cover new cultivars more quickly
  - Include more intensive evaluation of samples from other regions – both breeding and extension trials

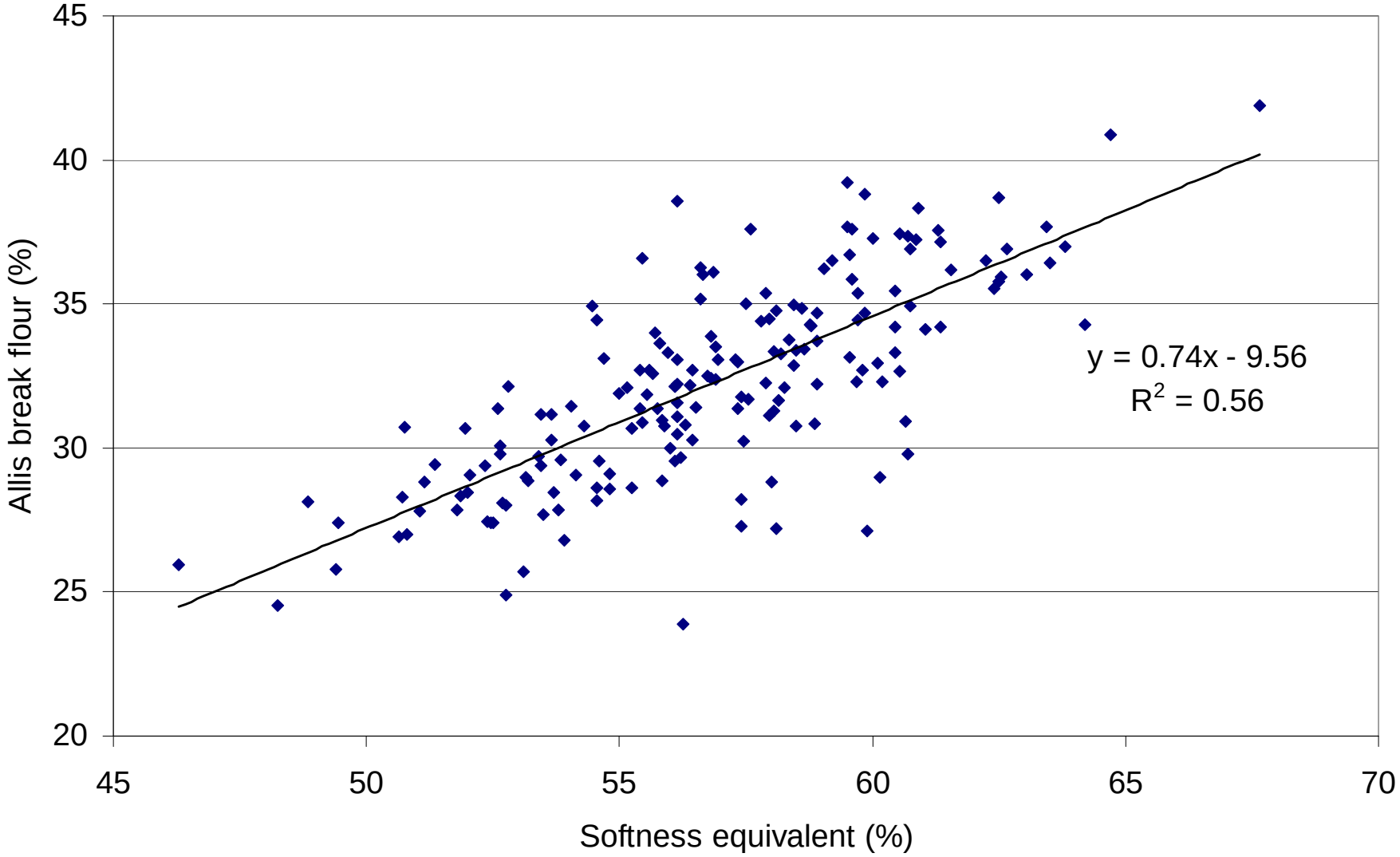
AM Study 2007

Quad Advanced Flour Yield with Allis Database Straight Grade Flour Yield



AM Study 2007

Quad Advanced Softness Equivalent with Allis Database Break Flour Yield



Planned Evaluation in 2007 and 2008

- Marker sets
  - DArT analysis for framework
  - QTL markers from bi-parental populations
  - Functional gene markers, such as glutenins, from USDA-ARS Wooster and Raleigh
  - Purified seed production for subsequent mapping will be available later this summer.

Association Mapping Study

Wooster and W. Lafayette, 2007 Mean Lactic Acid SRC (Gluten Strength)

	1B/1R Translocation	No Translocation
Glu-D1a (2+12)	100.0%	107.7%
Glu-D1d (5+10)	105.1%	117.1%

Association Mapping Study

Wooster and W. Lafayette, 2007 Mean Water, Na_2CO_3, Sucrose SRC 1B QTL

	Water SRC	Na_2CO_3 SRC	Sucrose SRC	Lactic acid SRC
By9	50.6%	65.4%	94.9%	107.9%
Non-By9	51.6%	66.7%	97.9%	110.7%
P-value significance	0.01	0.02	0.01	No difference

Completion of AM Study

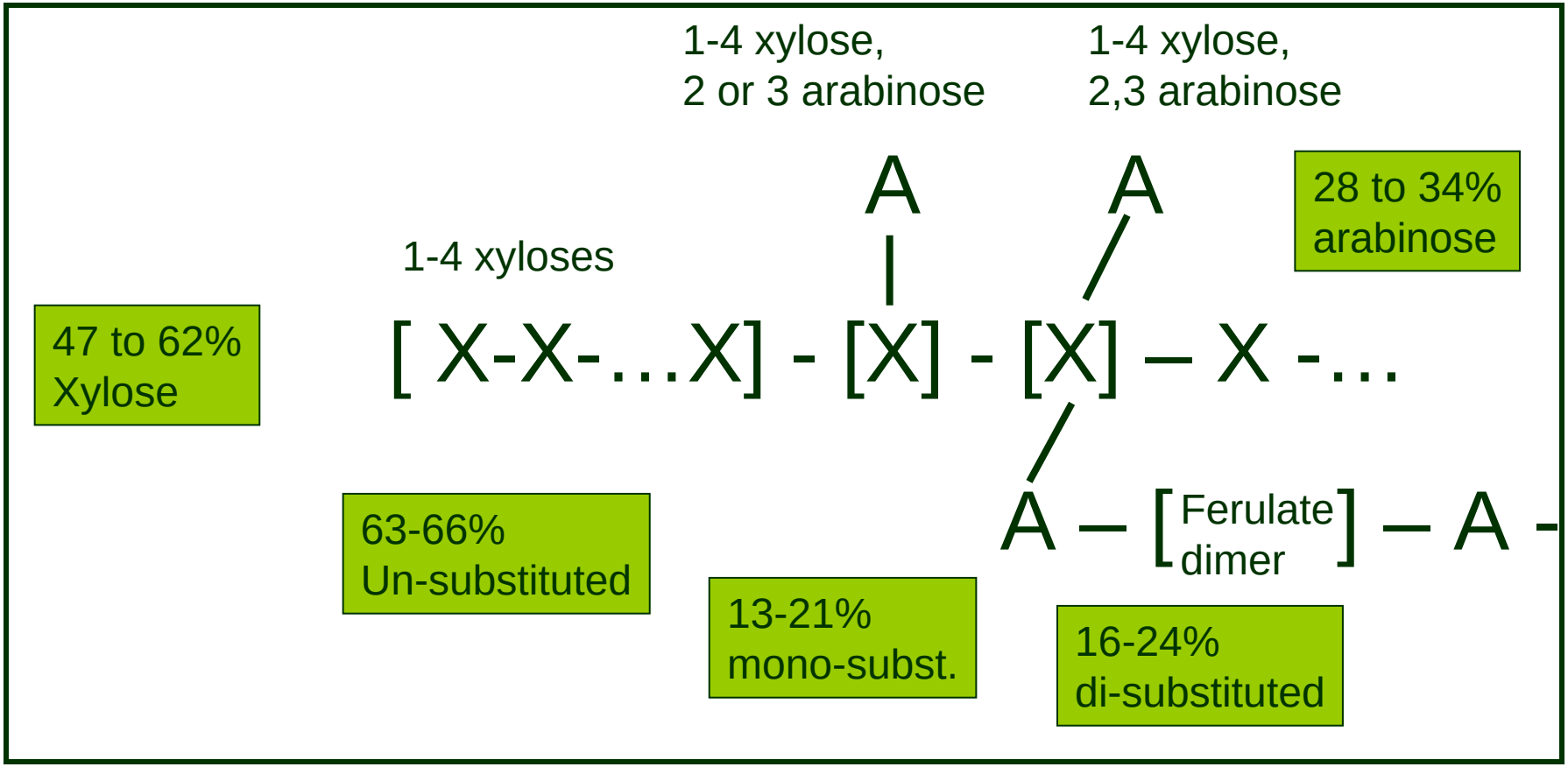
- Additional locations for 2007 and 2008
- Add Quality QTL markers from Wheat CAP study
- Use information for
  - Subsequent fine scale mapping
  - Compositional differences associated with mapped variation…

Arabinoxylans

Compositional Analysis

Arabinoxylans – Pentosans

J Delcour – Cereal Chem. 70:324 & J. Cereal Sci. 35:225



Enzyme extractable arabinoxylans have more arabinofuranosyl residues than water soluble arabinoxylans.

Increasing the degree of mono- and di-substituted xylopranosyl residues *decreases* flour water absorption.

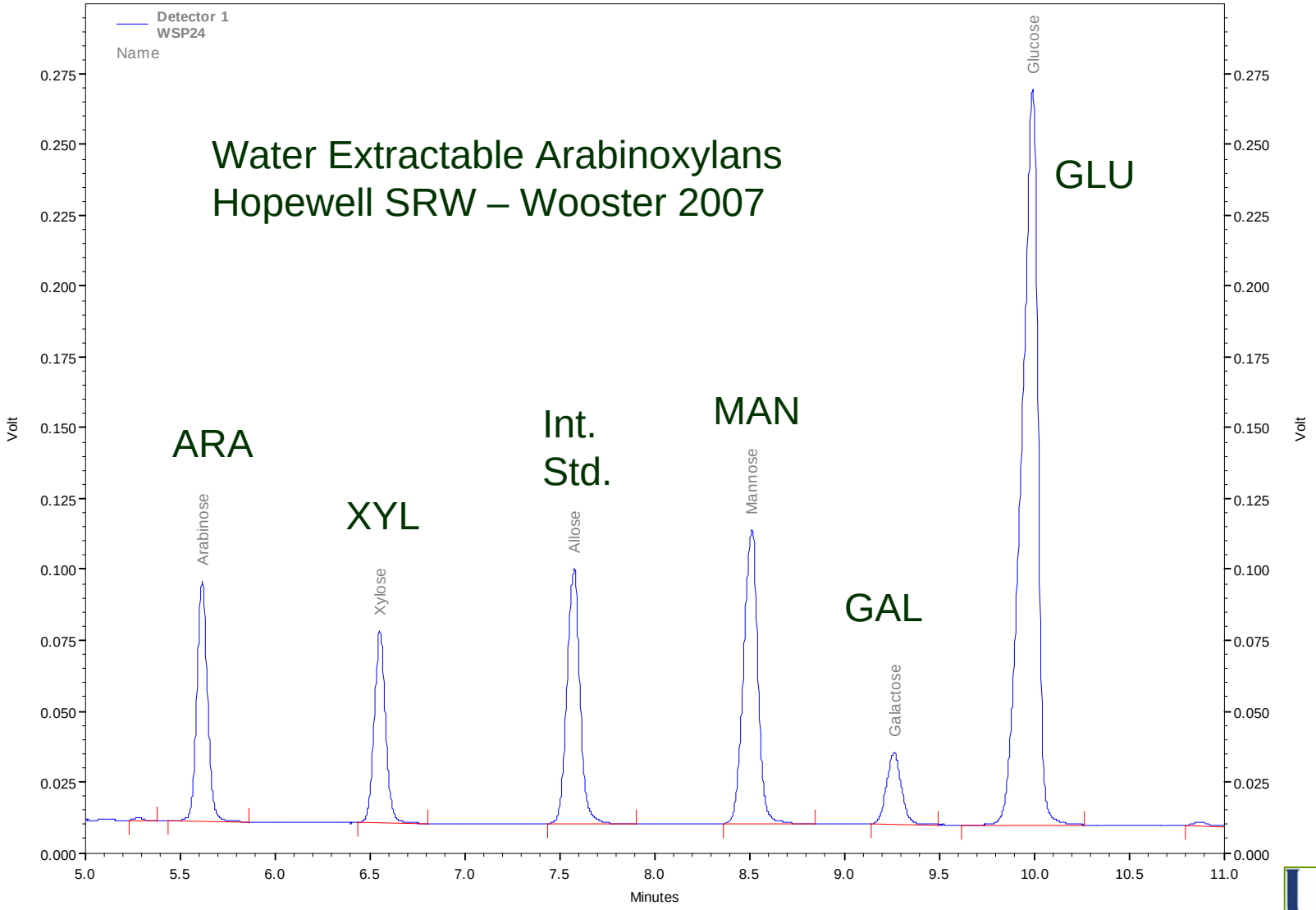
Colorimetric methods

- Based on xylose determination
- Methodological problem:
  - Background absorbance caused by glucose
- Assumption:

$$[\text{Arabinoxylan}] = C \cdot [\text{Xylose}]$$

Compositional method

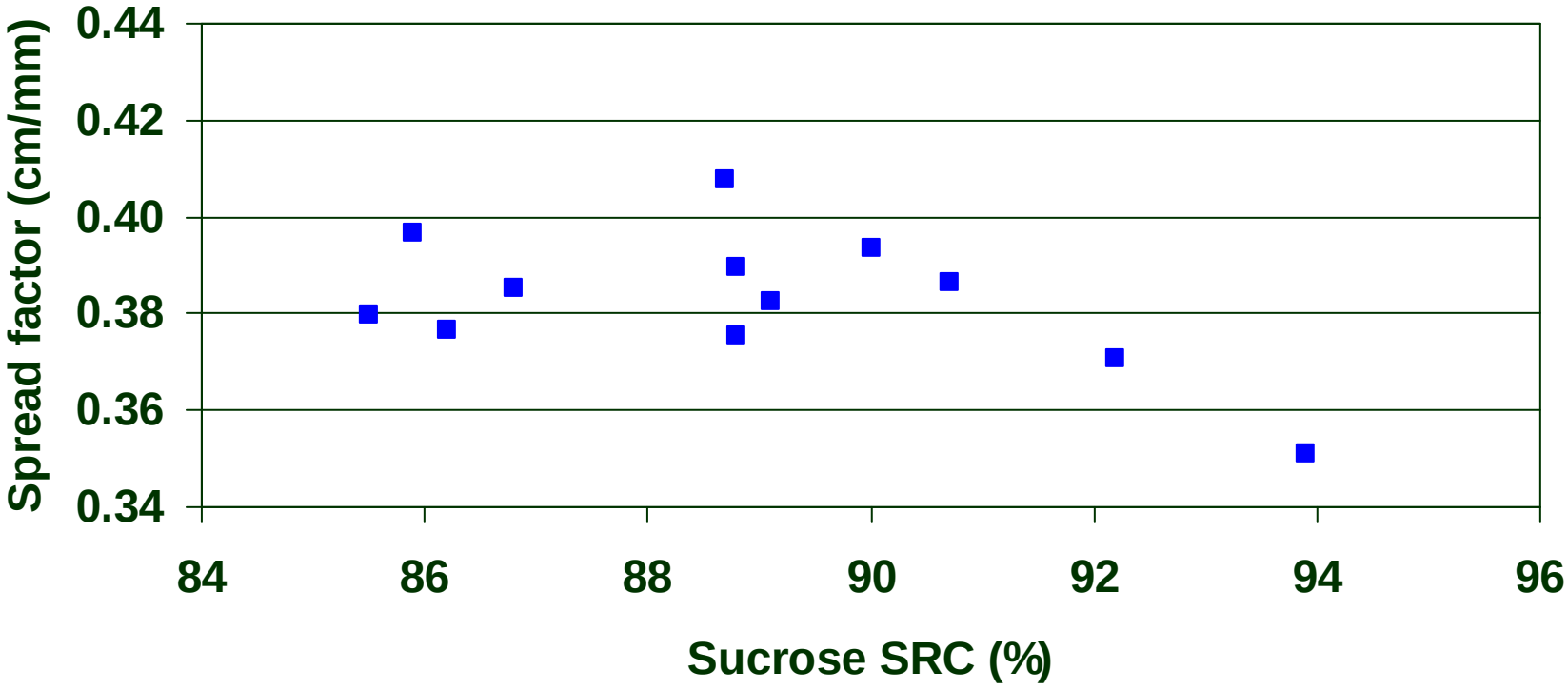
- Developed in laboratory of Jan Delcour, (Katholieke Universiteit Leuven, Belgium)
- Trifluoroacetic acid hydrolysis
- Alditol acetate derivitization
- Gas chromatographic quantification
  - Arabinose, Xylose, Mannose, Galactose, Glucose, Allose (Internal Std.)


Water Extractable Arabinoxylans
Hopewell SRW – Wooster 2007
Detector 1
WSP24
Name
ARA
XYL
Int.
Std.
MAN
GAL
GLU
Arabinose
Xylose
Allose
Mannose
Galactose
Glucose
Volt
Minutes
0.000
0.025
0.050
0.075
0.100
0.125
0.150
0.175
0.200
0.225
0.250
0.275
5.0
5.5
6.0
6.5
7.0
7.5
8.0
8.5
9.0
9.5
10.0
10.5
11.0

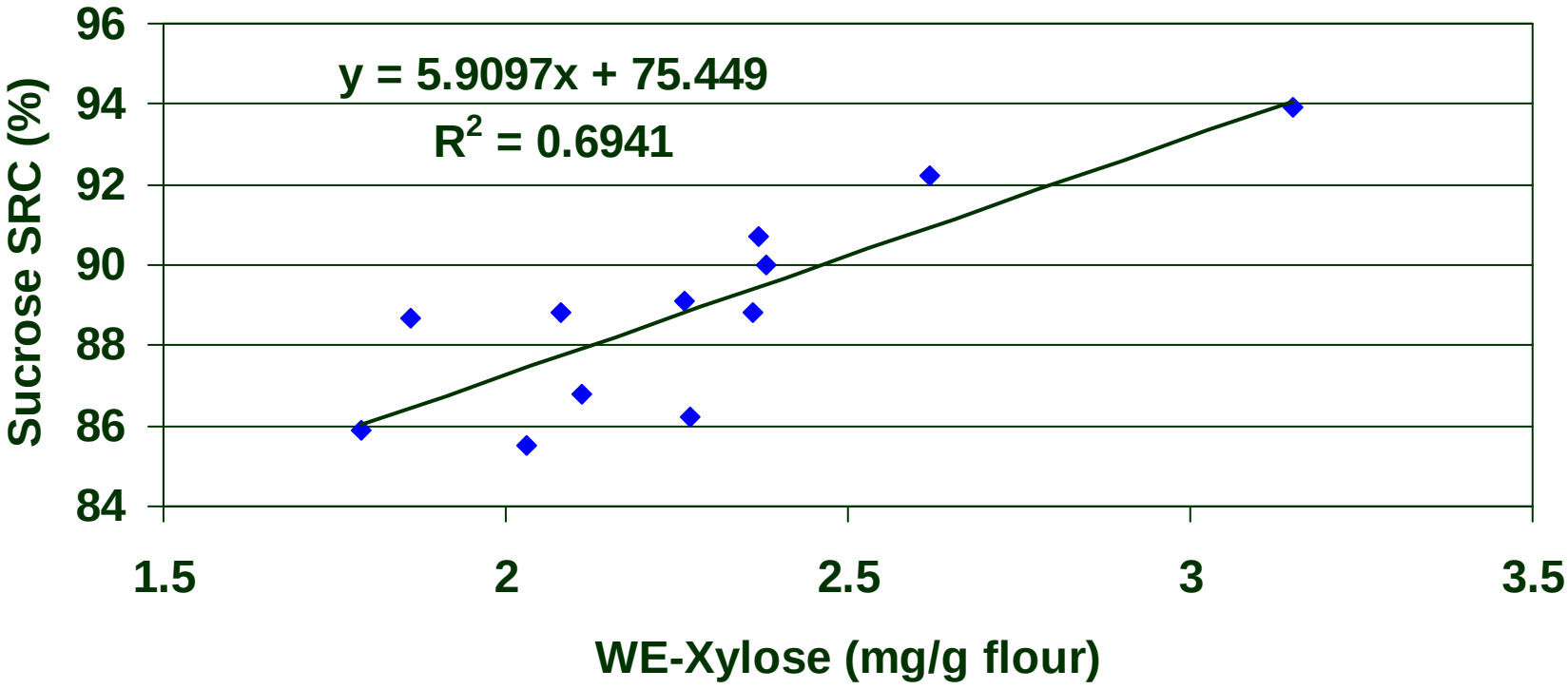
Experimental Design

- 6 Wheat genotypes:
  - Hopewell SRW, Caledonia Resel-L SWW, Richland SWW, E0028 SWW, E2017 SWW, NY92039-9065 SWW
- 2 Locations: Northwest Branch & Wooster
- Milled on Miag Multomat
- Duplicate bakes, chemical analyses

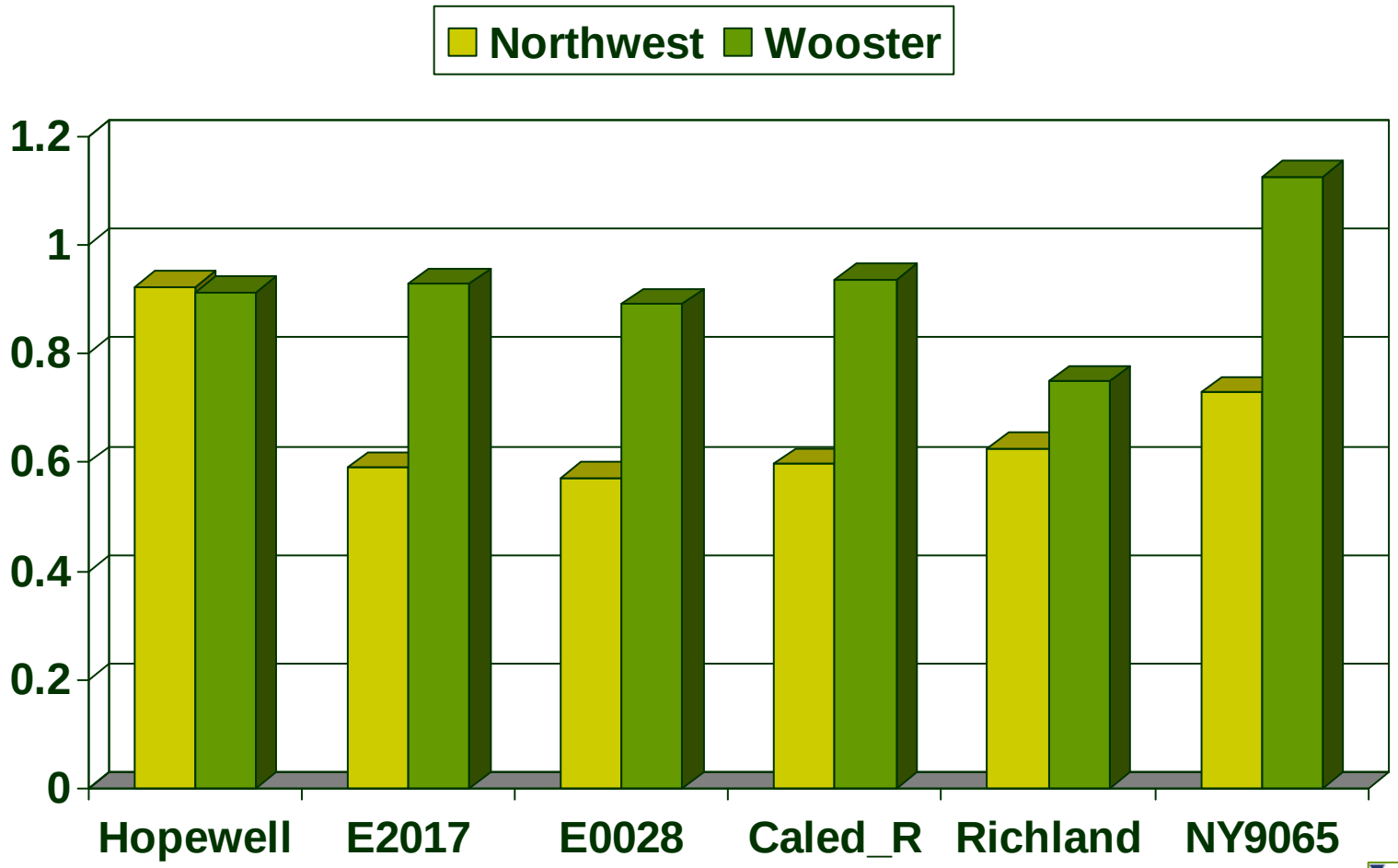
Wire Cut Cookie Spread Factor vs. Sucrose SRC



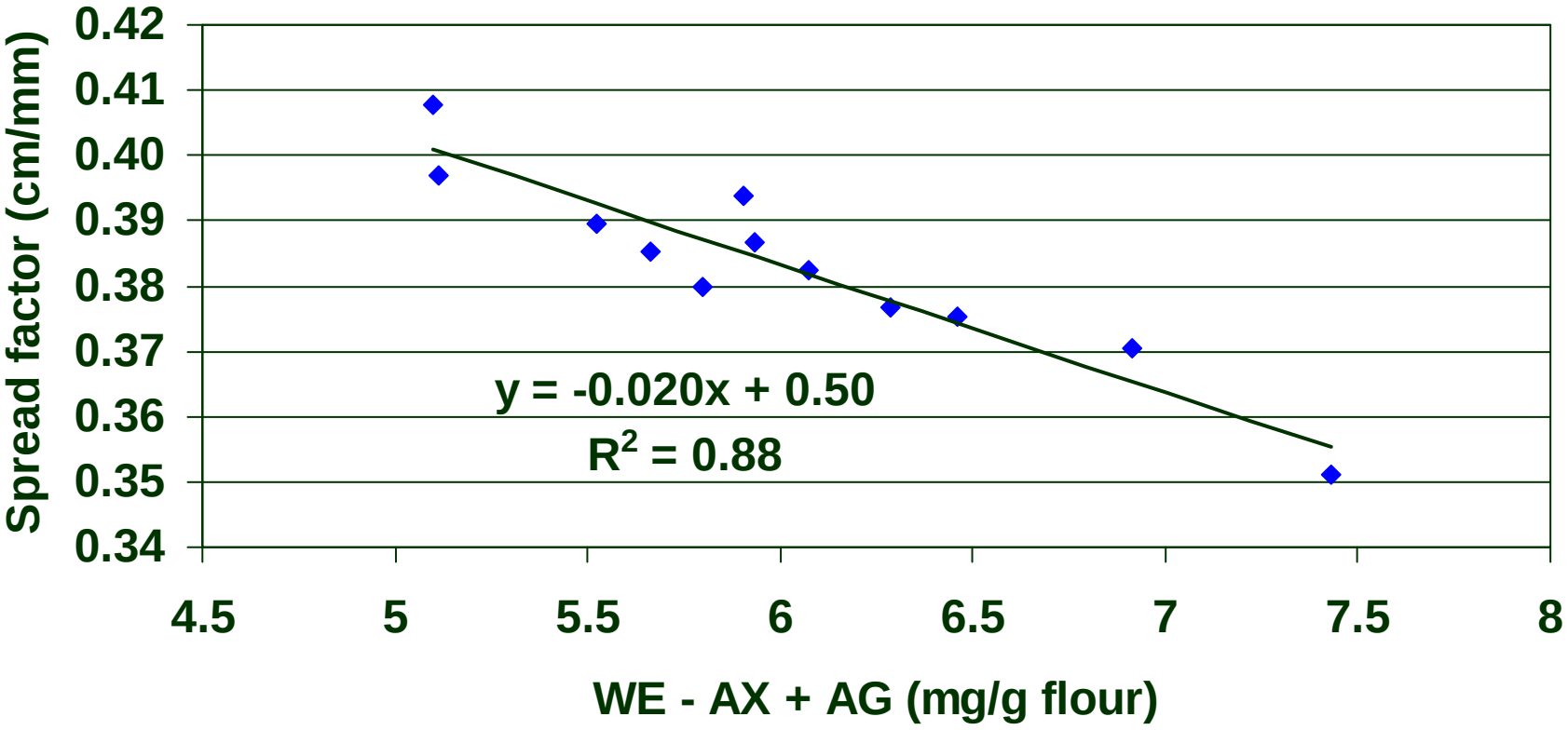
Sucrose SRC vs WE-Xylose



Arabinose:Xylose Ratio



Wire Cut Spread Factor vs Water-Extractable Arabinoxylan + Arabinogalactan



Relevant Questions for Mapping and Genetic Studies

- Milling loci often seem to be loci that also affect sucrose SRC and Na_2CO_3 SRC
  - Or….
- Arabinoxylan composition and degree of substitution affects sucrose SRC and milling yield, which in turn affects starch damage and Na_2CO_3 SRC?
- Genes coding for synthesis and regulation of arabinoxylan are unknown in wheat
  - UDP-arabinosyl transferase and UDP-xylosyl transferase are hypothesized but uncharacterized.

Relevant Questions for Mapping and Genetic Studies

- Association mapping population follow-up
  - Conduct AX composition studies
  - Experimental design based on phenotypic and genotypic information
- Positional cloning or gene expression studies may allow identification of genes controlling AX structure
- These are the contrasting approaches that are under discussion for Wheat CAPII