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90 Years of Red Clover Breeding at Wisconsin USDA-ARS

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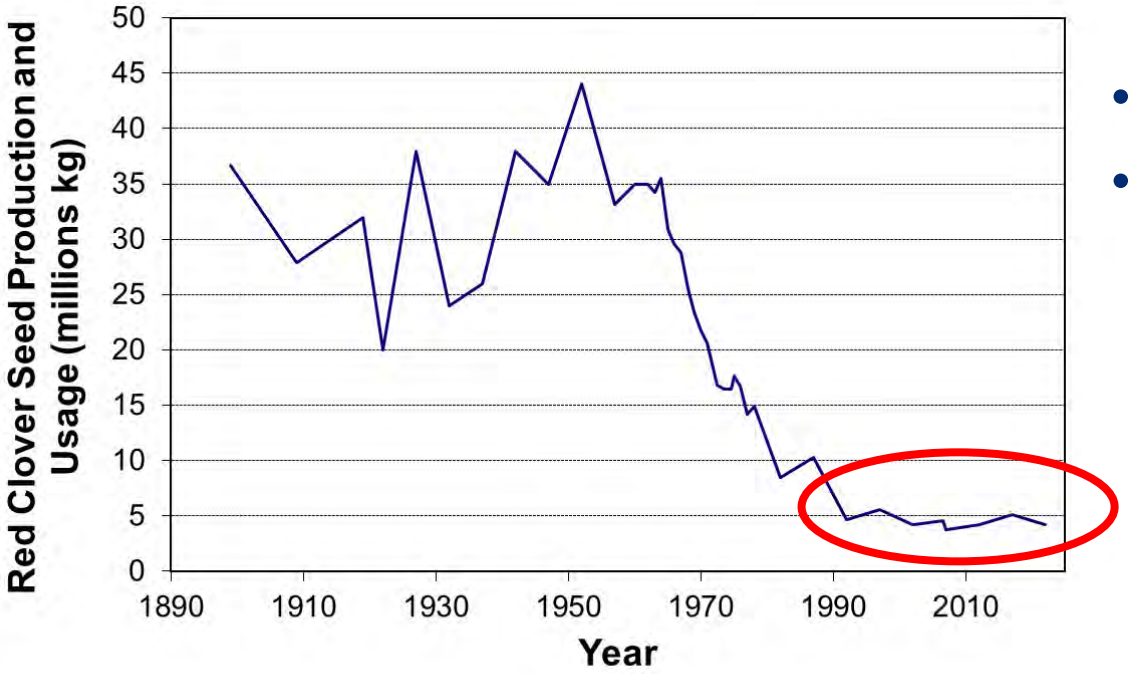
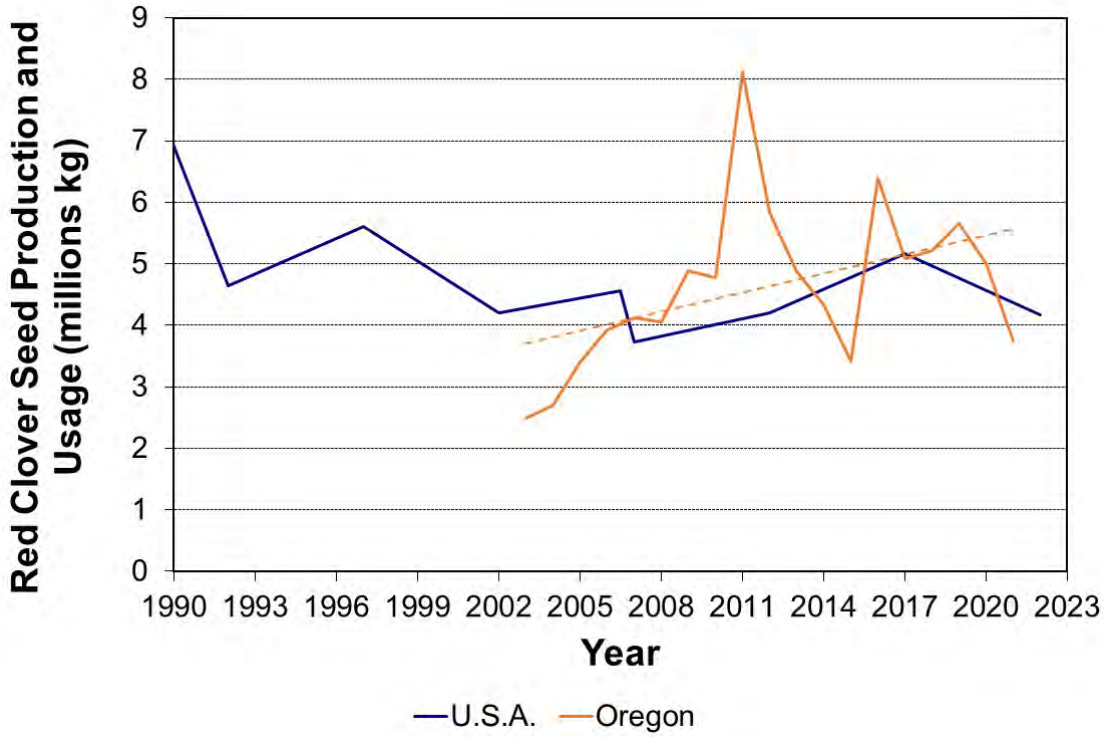
U.S. Dairy Forage Research Center

Background – Red Clover Usage



(Taylor and Quesenberry, 1996; NASS, 1899-2022)

- Large 20th century red clover usage decline
- Usage stabilization in 1990s; in U.S. around 5 million kg yr^{-1} seed



(Anderson et al. 2014-2022; NASS, 1899-2022; Young et al., 2005-2013)

- Evidence of seed production increase of 0.1 million kg yr^{-1} in Oregon?

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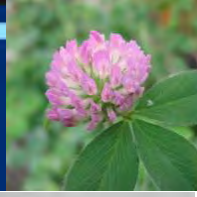
USDA Wisconsin Red Clover Breeding Program



W. K. Smith 1937 – 1965: Agricultural geneticist whose work on sweet clover led to the development of a non-toxic variety of great importance to dairly farmers. Along with R.A. Brink, Smith discovered that the toxic substance in sweet clover was coumarin. Karl Paul Link and Mark Stahman capitalized on this discovery when they developed the rat poison, Warfarin, and the medical anti-coagulant, dicoumarol.

R.R. Smith USDA-ARS: 1966 – 1999: Worked extensively on breeding for disease resistance

M. Casler USDA-ARS: 2001-2004

H. Riday USDA-ARS: 2004 – present

- 1953/1959 – Lakeland red clover released by W.K. Smith (Hollowell, 1961) bred for powdery mildew and northern anthracnose resistance.
- 1973 – Arlington red clover released by R.R. Smith (Smith et al., 1973) bred for powdery mildew and northern anthracnose resistance
- 1987 – Marathon red clover released by R.R. Smith (Smith, 1994) bred for northern anthracnose resistance
- 2014 – FF 9615 red clover released by H. Riday and R. Schmidt (Riday and Schmidt, 2019) bred for increased biomass yield and persistence.
- 2019 – Resolution red clover released by H. Riday and R. Schmidt bred for increased biomass yield and persistence

Breeding Progress - Persistence

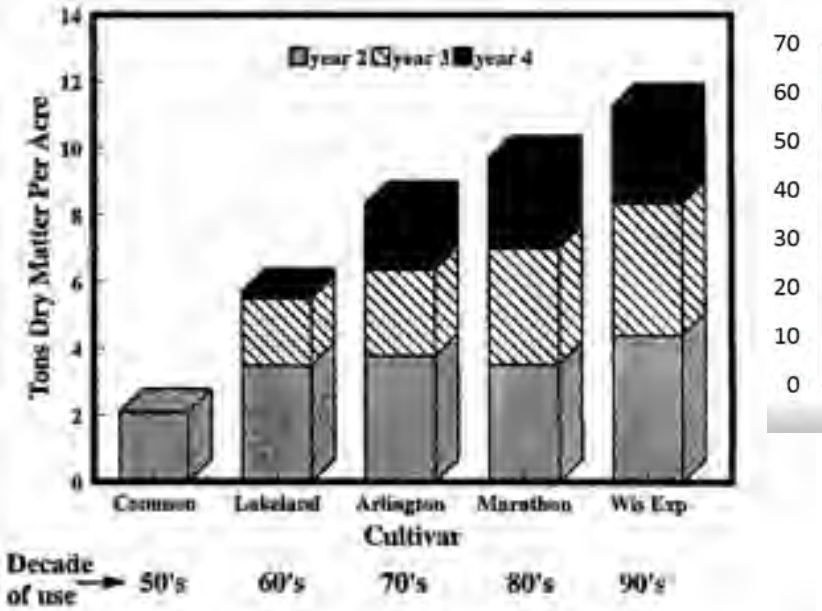
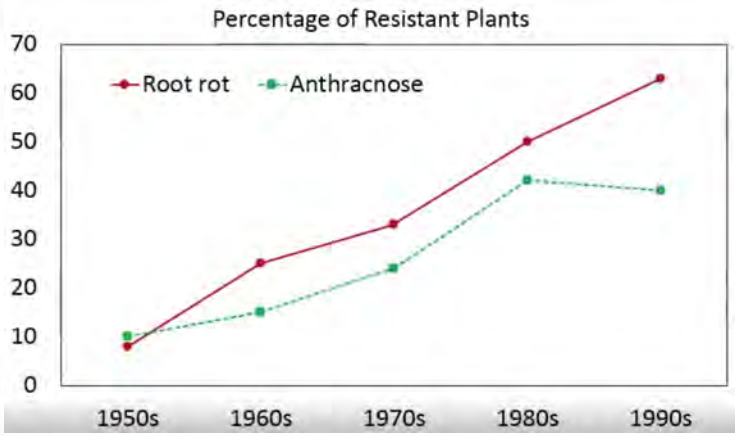


Figure 2. Forage production of red clover resulting from five decades of breeding for persistence and disease resistance.

(Smith, 2000)



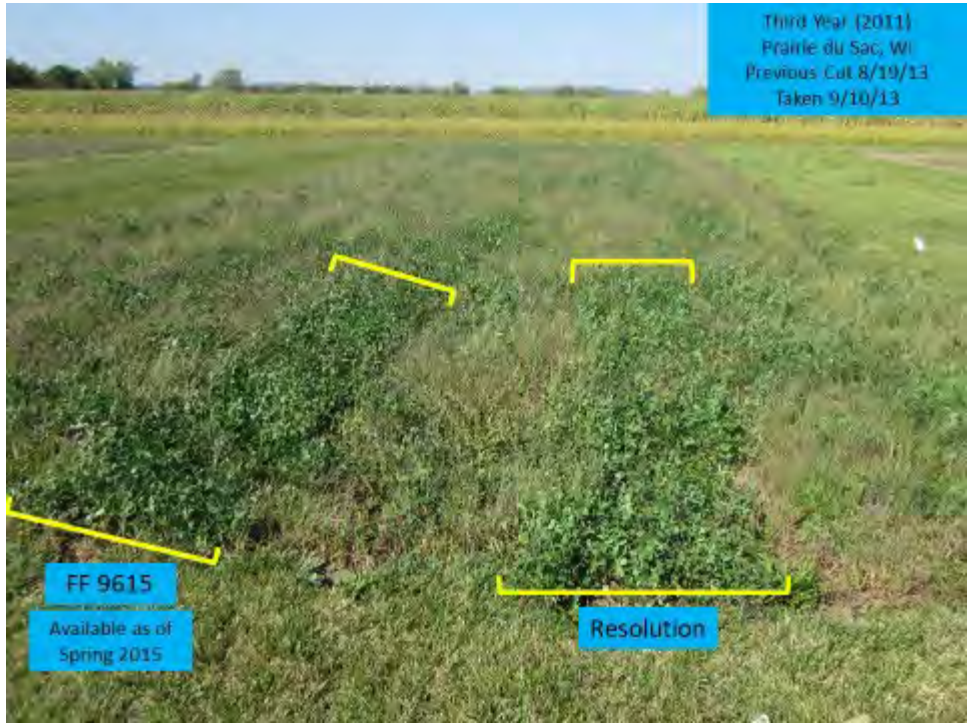
Resistance to Northern Anthracnose

Resistance to Root Rot

Cultivar	Year of release	1 tap root	2	3	4	5 fibrous root	Mean root type	3-yr Stand
		%	%	%	%	%		%
Lakeland	1961	1	60	36	3	0	2.40	32
Arlington	1973	2	52	40	6	0	2.51*	48
Marathon	1987	3	25	59	14	0	2.53**	67

(Casler et al., 2019)

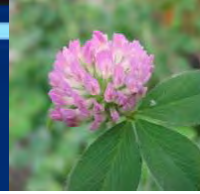
(Casler et al., 2019)



"For optimum forage yield, quality and persistence, we found that current red clover varieties should be harvested three times during the year when plant are between bud stage and 20 percent bloom" (Two cuts prior to Sept. 1st and one after 15 Oct or frost.) D. Wiersma

"Making three cuts during the summer can put to much stress on red clover plants . . . That's especially true if the weather is dry during late summer or fall" R. Smith

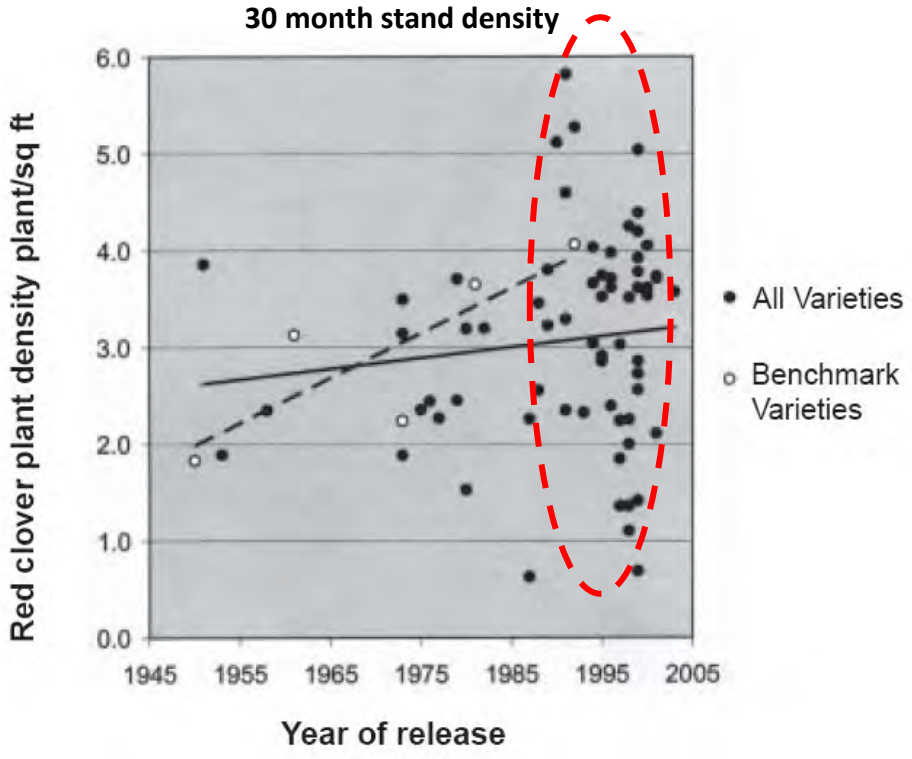
Breeding for Grazing Systems

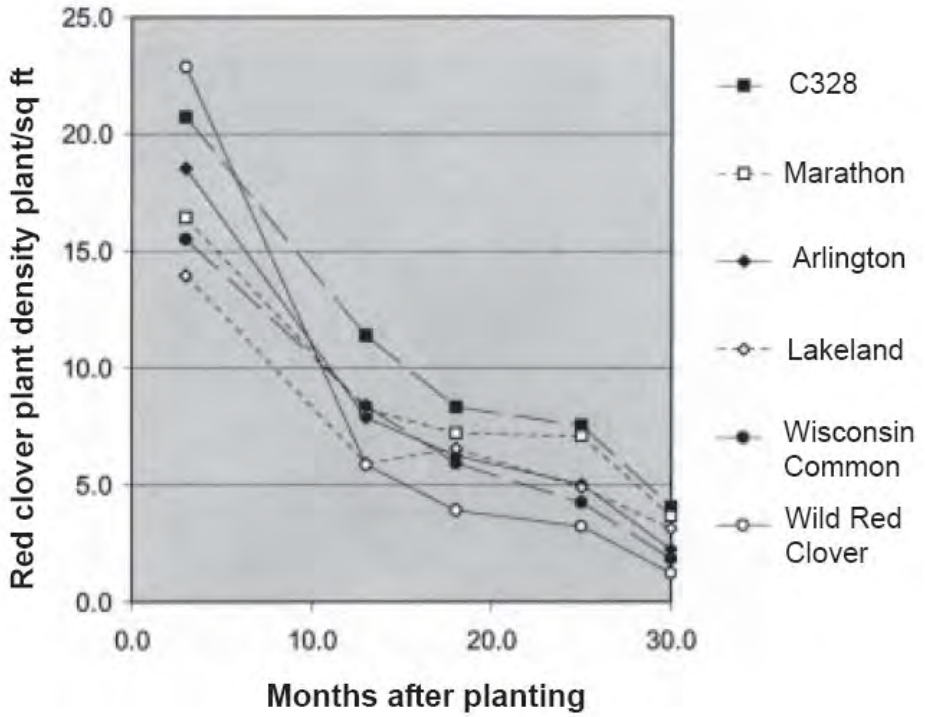


- Initial focus on breeding for grazing systems
  - More frequent cutting with 28 to 30 day cutting interval (4 to 5 cuts per year)
  - Put red clover in mixture with grasses (pasture systems).
    - Competition (light, moisture, and soil)
  - Evaluate biomass performance during the establishment year.
  - More even biomass distribution across the year

Breeding Progress Grazed Mixture with Grass

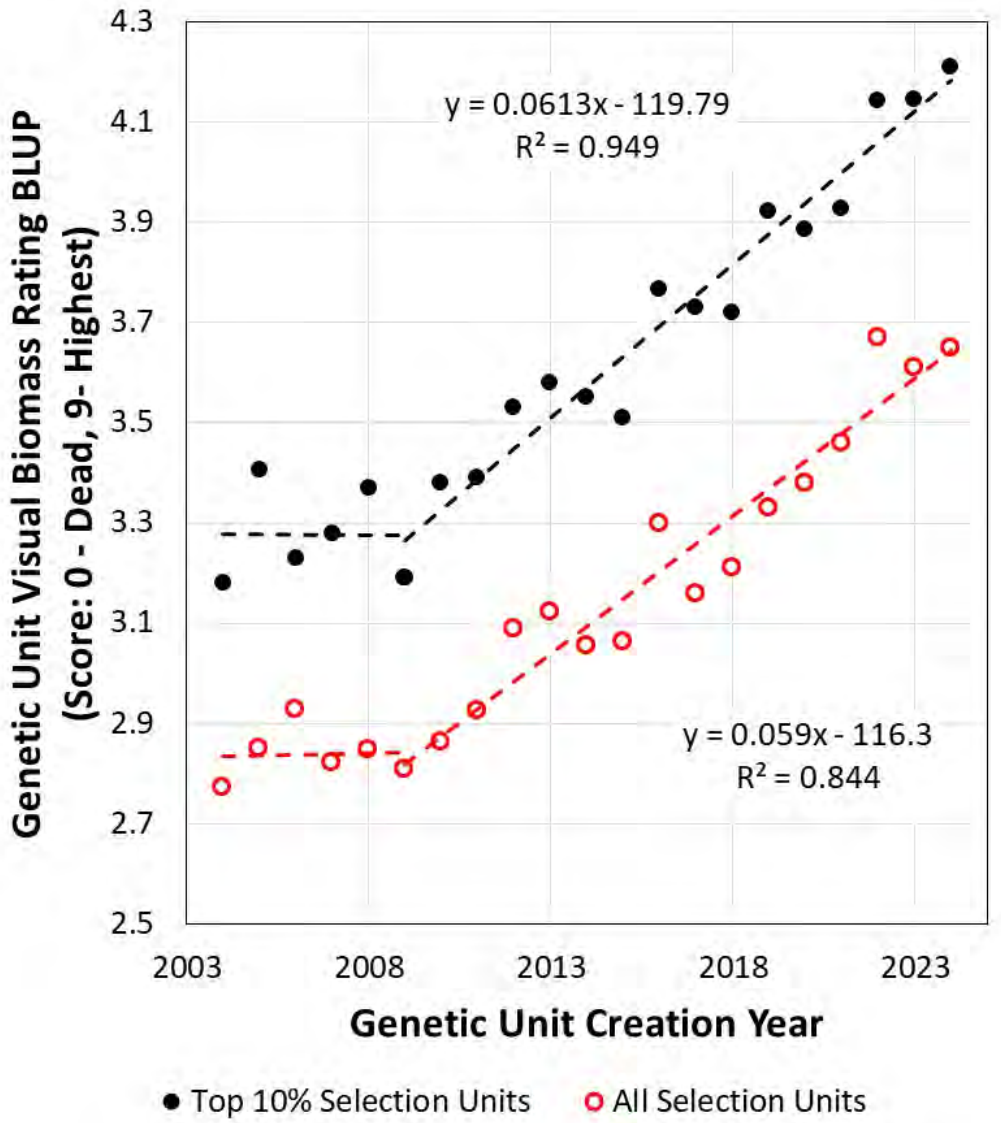
- Riday et al., 2007
  - 100% stand at 9 plants sq. ft^{-1}
  - Large persistence variation in new varieties
  - All varieties increasing 0.011 plants sq. ft^{-1} yr^{-1} (0.12% stand yr^{-1} increase)
  - Benchmark varieties increasing at 0.047 plants sq. ft^{-1} yr^{-1} (0.52% stand yr^{-1} increase)



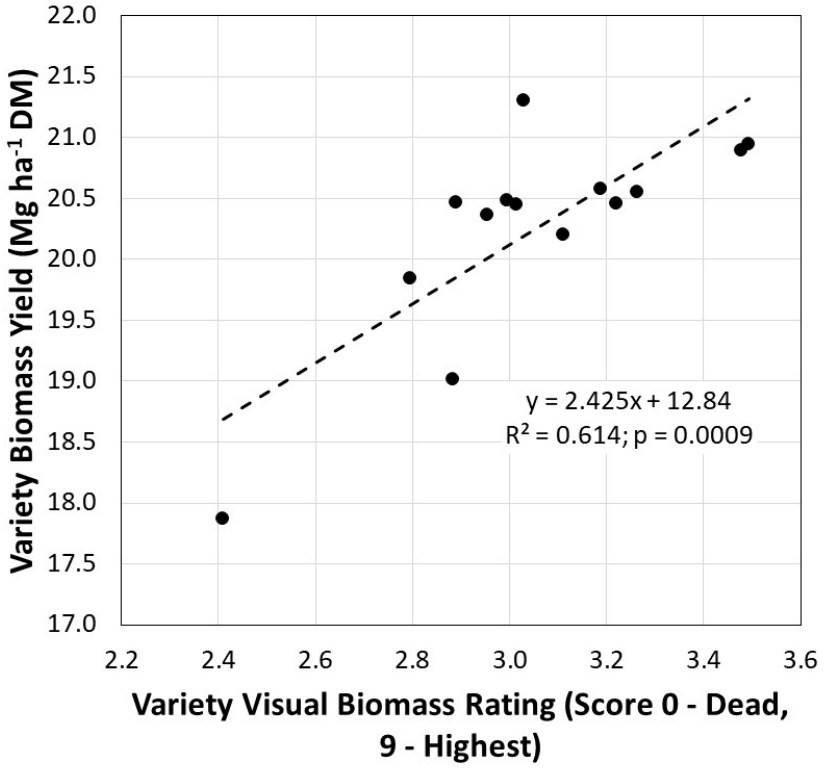


Breeding Progress Persistence – Biomass Yield

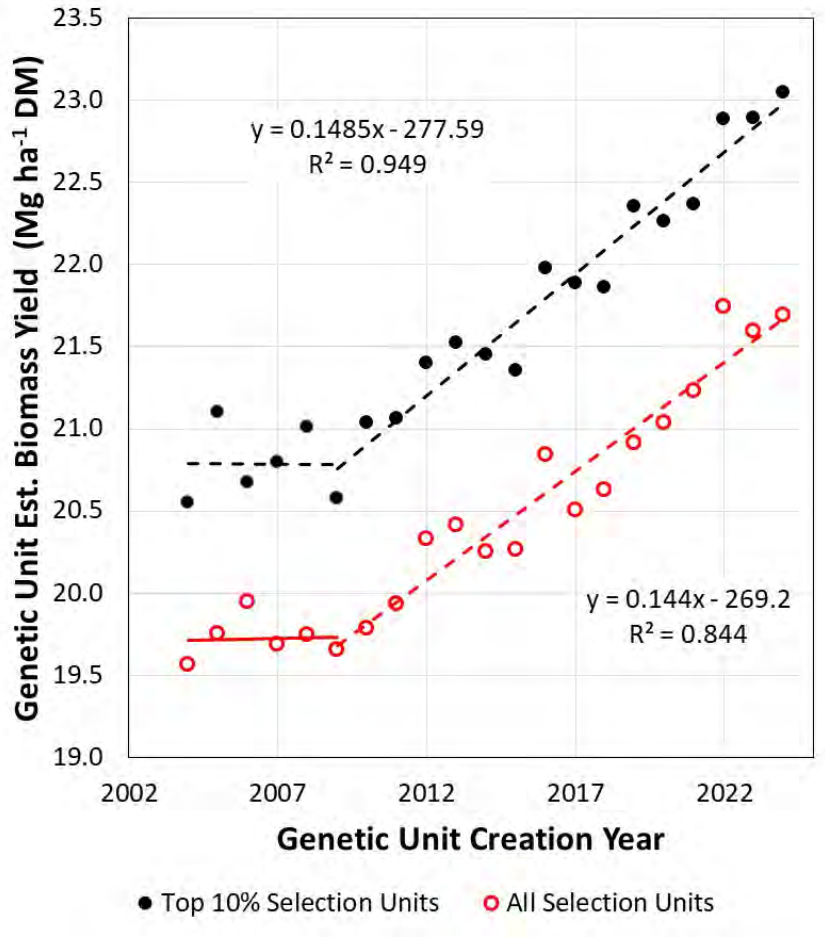
- 22 years of breeding in Wisconsin
  - Nursery visual vigor evaluations
    - 2,387,677 obs. (12.7 obs./plant)
    - 187,868 plants (1,287 plants/nursery)
    - 146 nurseries (6.6 nurseries/year)
    - 5,898 halfsib families, populations, or varieties (31.8 plants/unit)
    - ~150-200 new halfsibs, populations, or varieties evaluated/year
  - Low density genotyping
    - 29,123 plants (15.5% genotyping rate)
  - Genetic gain 1.4% per year



Vigor Ratings versus Sward Biomass Yield

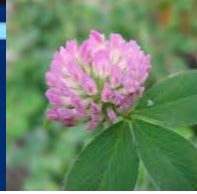


- Regression: visual space plant biomass rating vs. sward trials biomass yield
  - 160 Sward trials (1992-2022); 123 Space plant nurseries (2006-2022)
  - Minimum 10 sward trials per variety (y)
  - Minimum 80 individual space plants per variety (x)

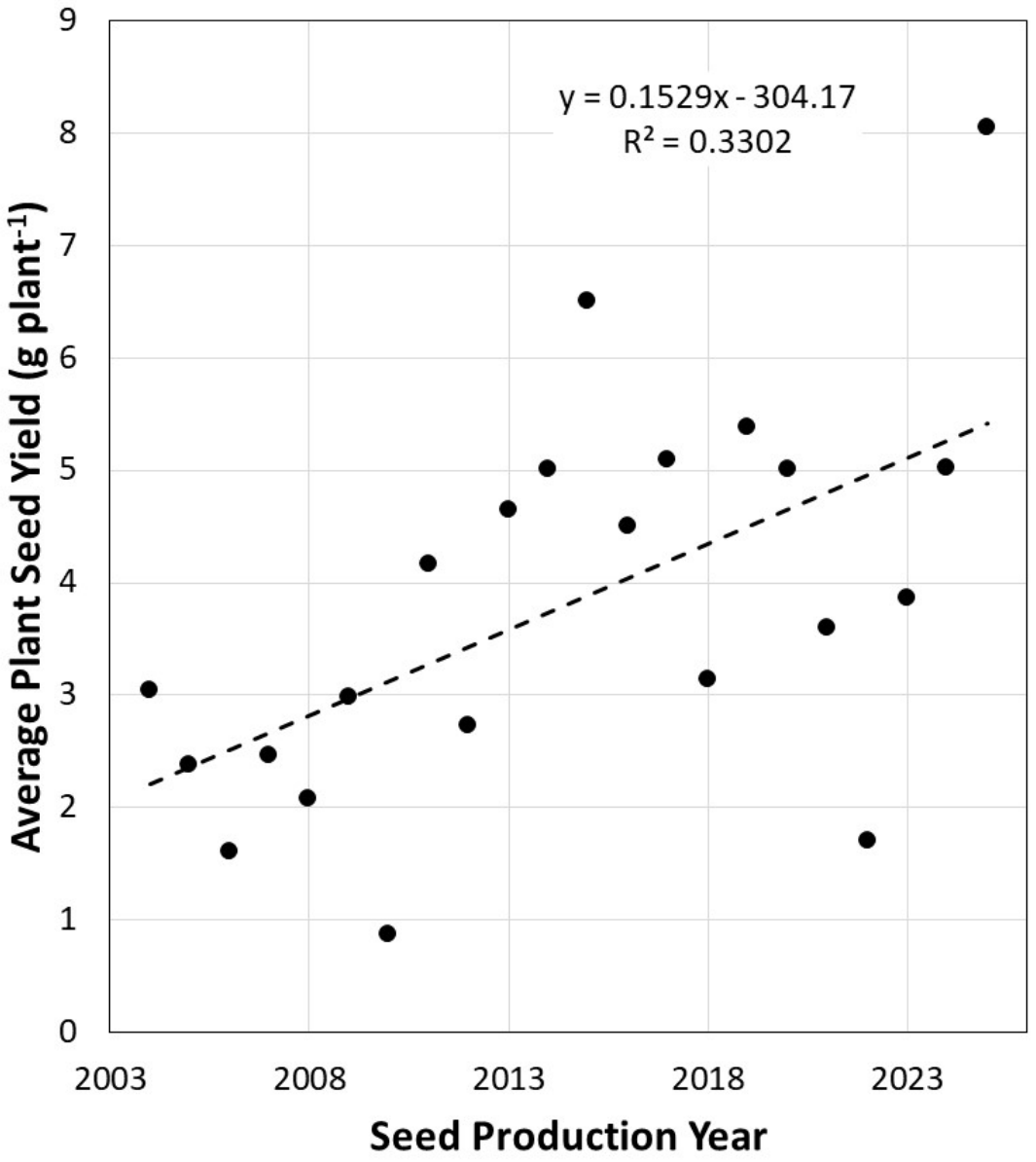


- Putative 0.15 Mg ha^{-1} biomass DM gain per year
  - 0.54% gain yr^{-1}
    - (1.4% gain yr^{-1} when based on visual score)

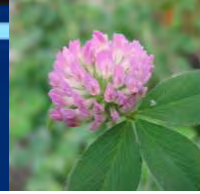
Breeding Progress Seed Yield



- 22 years of breeding in Wisconsin
  - Polycross individuals for halfsib seed production
    - 20,1817 parents polycroseed
    - 375 polycrosses (55.5 parents/polycross)
      (17.0 polycrosses/year)
  - Low Density Genotyping
    - 14,997 parents (72% genotyping rate)
  - Genetic gain 6.8% per year (0.15 g plant^{-1} year^{-1})

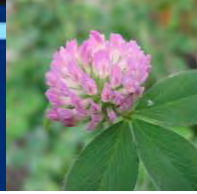


Breeding Intensification



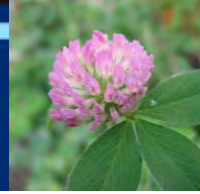
- Selection strategies:
  - Minimum seed yield threshold to advance halfsib seed (weaker selection for seed yield)
    - Halfsib advancement decision out of polycrosses based on seed yield only (2004)
  - Phenotypic selection (dig individual surviving plants from nurseries or variety trials) **(5 years cycle)** (2008)
  - Genotypic selection – Maternal-line selection (produce new halfsib families from polycrossing indviduals from remnant seed of the best maternal halfsib families) **(3 year minimum cycle)** (2009)
  - Low density DNA markers (~16 SSR markers) ($1/genotype) (2010)
    - Program quality control (use DNA for QC by confirming selected plant identity and pedigree linkages)
    - Realize full pedigree information in the program using paternity testing (pollen parent determination)
  - Intensive phenotypic selection (polycross best individuals from each location together based on life-time phenotypic values) (high selection intensity [0.5%]) (2016)
  - Polycross plant positioning based on a plants phenotypic value. (Exploit pollinator movement) (2017)
  - Phenotyping individual plant heights (before 1st and 2nd cut) (2021)

Breeding Intensification

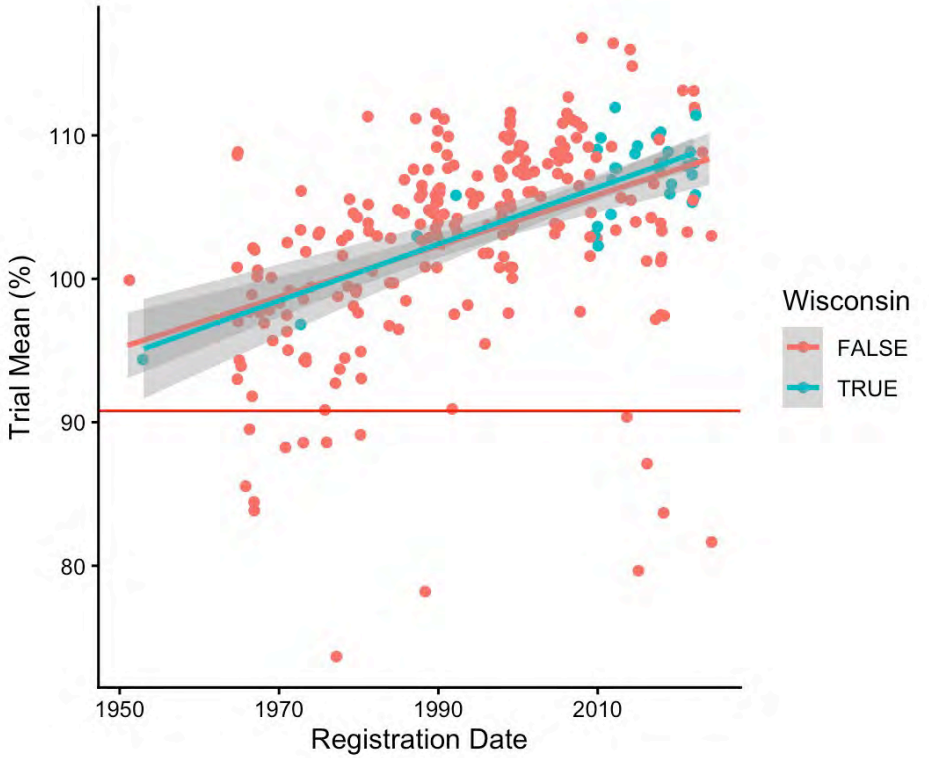


- Selection strategies cont.
  - Estimating plant breeding values (or BLUPs) using full pedigree information (i.e., like in animal breeding) (2024)
    - More information (higher heritabilities) for both phenotypic and genotypic selection
    - Halfsib advancement out of polycrosses incorporating vigor BLUPs
    - Forward breeding using BLUPs (predicting an individuals breeding value without phenotyping it or any progeny testing) **(1 year minimum cycle $\frac{1}{2}\sigma^2_A$)**
  - High density DNA markers systems (2025)
    - Red clover genome assembly (Bickhart et. al, 2022)
    - DArT protocol (~2,000 SNPs @ $14 per genotype)
    - **?!** DFRC Bajgain (~20,000 SNPs @ $3 per genotype)
  - Genomic prediction modeling and genomic selection (GS) (2026 – Future)
    - 1 year minimum cycle
    - Is GS better than simple BLUP pedigree selection? (how much more?)
    - GS allows selection within fullsibs **(1 year minimum cycle σ^2_A)**
    - Pooled DNA and creating genomic prediction models for sward-based traits!

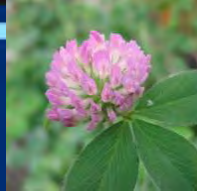
Developing Priorities and Other Traits



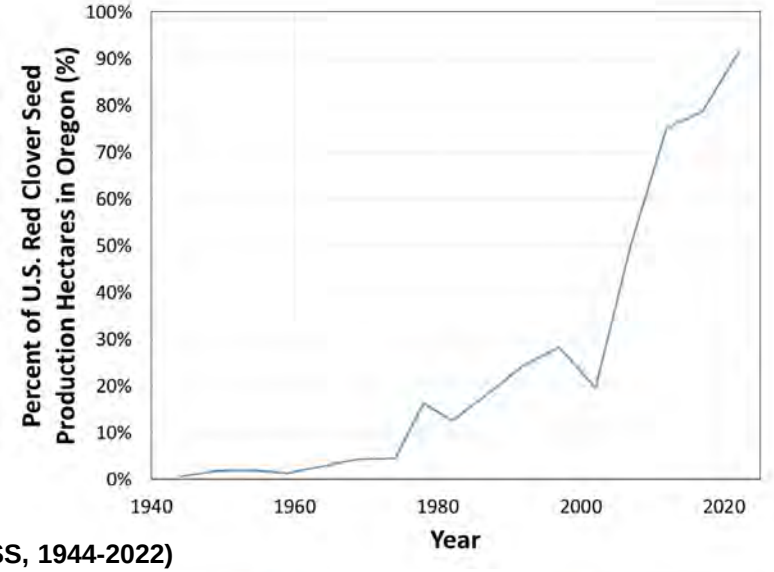
- Grazing variety trialing
- National forage variety trialing
- Red clover as a winter annual cover crop
- 2,4D resistance
- Phytoestrogens



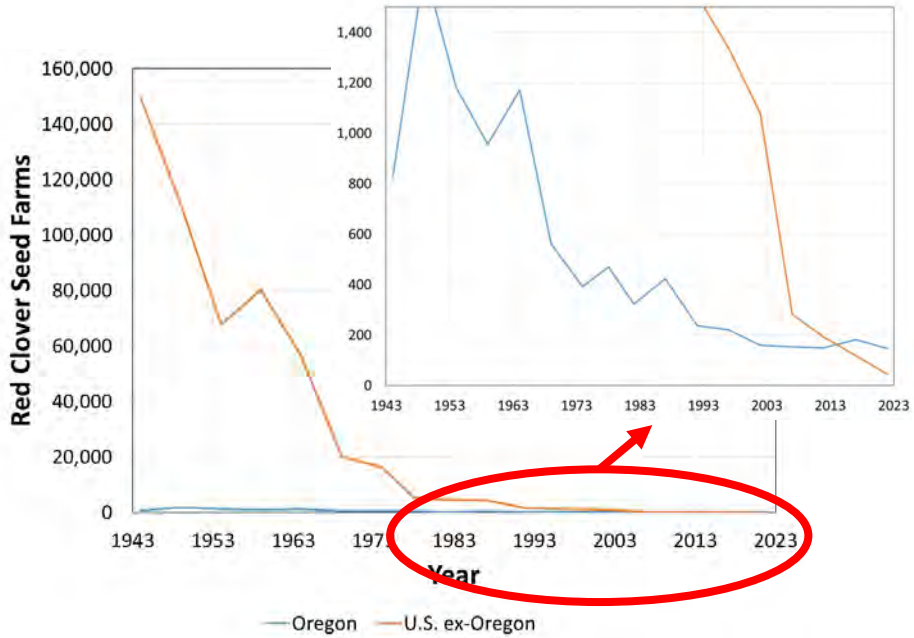
Developing Priorities and Other Traits



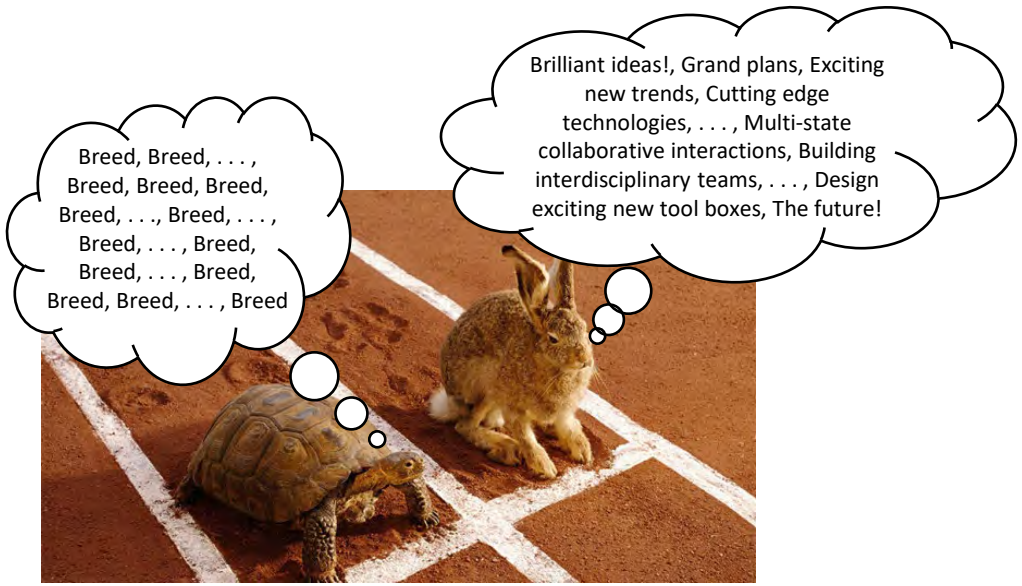
- Seed production and adaptation in Oregon
  - 91%+ of U.S. seed production
  - < 200 farms produce 35% of global red clover seed
  - Different diseases
  - Seed yield variety trials



(NASS, 1944-2022)



Questions?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Brilliant ideas!, Grand plans, Exciting new trends, Cutting edge technologies, . . . , Multi-state collaborative interactions, Building interdisciplinary teams, . . . , Design exciting new tool boxes, The future!