TOWARDS A VIABLE PERENNIAL GRAIN SORGHUM IN NORTH GEORGIA THROUGH INTEGRATED GENETIC, ECONOMIC, AND SPATIAL ANALYSIS

by

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(Under the Direction of Andrew H. Paterson)

ABSTRACT

The replacement of annual cereal crops with perennial varieties has great potential for increased ecosystem resilience and improved food security. Successful establishment of such cropping systems requires development of both system “hardware” (perennial grain varieties) and “software” (agronomic and ecological systems facilitated by the perennial germplasm). Thus, a multi-pronged research approach to perennial grain sorghum (*S. bicolor*) establishment in north Georgia was undertaken. Quantitative trait locus (QTL) analysis of a novel $F_{2:3}$ population derived from an *S. bicolor* x *S. halepense* cross illuminated genomic regions pertinent to perenniality and can contribute to marker-assisted selection in breeding. Simultaneous investigations into Georgia sorghum farm budgets highlighted varying yield targets contingent on perennial cropping scenario as well as other breeding targets to further improve viability for Georgia farmers. A RUSLE-based GIS model of soil erosion in two north Georgia watersheds validated breeding efforts for a dual use grain production and soil conservation system.

INDEX WORDS: Perennial grains; Rhizomes; Quantitative trait loci; RUSLE; Farmer enterprise budgets; Interdisciplinarity

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Perennial Grains

Global food security hinges on humanity's capacity to continuously supply sufficient food for itself and future generations. While conventional row crop agriculture is effective at maximizing food output for a given area of land, it also results in significant environmental degradation, including but not limited to: soil erosion, decreased soil organic matter, water pollution, and increased water scarcity (Pimentel et al. 1995; Cox et al. 2006). This degradation compromises the capacity to supply food for future generations. Thus, it is imperative to consider viable agricultural alternatives that do not degrade the earth's limited stock of agricultural land. One such alternative option is perennial agriculture.

Compared to their annual counterparts, perennial plants provide a suite of ecosystem services, including: protecting soil from erosion, sequestering carbon, aiding in nutrient retention, and increasing water infiltration (Glover et al. 2007). Notably, perennial cover is more than 50 times as effective than annual crops in maintaining topsoil (Gantzer et al. 1990) and 30-50 times more effective than annuals at preventing nitrogen losses (Randall and Mulla 2001). Interestingly, perennial grasses may prove to be a more reliable carbon sink than trees under increasingly volatile climate realities, as belowground mechanisms of carbon storage are more resilient to fires (Dass et al. 2018). Additionally, perennial plant's continuous growth cycle reduces the need for annual tillage and herbicide application, which in turn reduces labor inputs while contributing to overall soil and water health. The lower inputs required in a perennial cropping scenario can facilitate a greater degree of autonomy for farmers.

Although a handful of agricultural commodities are perennial, such as fruit and nut trees, the vast majority of farmland is devoted to annual planting systems. Of particular importance are annual grains and oilseeds, which make up roughly 70% of the world's agricultural land and a comparable amount of humanity's caloric intake (Monfreda et al. 2008; Pimm 2001). Consequently, shifts in farming practices within this particular group of crops holds great potential for increased ecosystem resilience worldwide, making them an ideal target for perennial cropping systems.

Essentially all major grain crops are currently grown as annuals, with some progress in production of perennial varieties of rice and intermediate wheatgrass (Huang et al. 2018; DeHaan et al. 2018). Perennial grain cultivars can be developed using two basic methodologies, either direct domestication of a wild perennial or the introgression of perenniality into a current annual grain cultivar through wide hybridization (Cox et al. 2002). While conventional breeding methods of recurrent selection for perennial traits of interest have been underway for decades, marker-assisted selection in breeding can greatly increase the efficiency of perennial grain germplasm development.

The concept of perennial grains agriculture is attractive in its simplicity. By positing that an economically viable crop could simultaneously provide necessary ecosystem services, it is able to sidestep the common sustainability question of balancing production with conservation. In practical execution, it is more complicated, as perennial grains tend to yield less than their annual counterparts and experience diminishing yields over years of regrowth (DeHaan and Ismail 2017; Smaje 2015). Consequently, the viability of a perennial grain crop becomes dependent on the evaluation of economic returns in relation to potential ecosystem services, which can vary across a physical or political landscape. Marginal lands that are more vulnerable to environmental degradation have more to gain from a perennial cropping system, while policies that provide financial incentives for environmentally conscious land management practices can further enhance the feasibility of such systems.

Thus, it is clear that the plant breeding barrier is not the only one to contend with in the establishment of perennial grain cropping systems. Agriculture is a realm in which the natural and social sciences are inextricably linked and consequently, agricultural progress requires an integrated effort from multiple disciplines. Fischer et al. (2015) highlights conceptual and methodological pluralism as key to progress in sustainability science, stating that the use of multiple conceptual frameworks is critical to effectively engaging with complex socio-ecological problems. Likewise, a shift towards perennial grain cropping systems requires a multi-pronged approach that integrates the development of both system “hardware” (perennial grain varieties) and “software” (agronomic and ecological systems facilitated by the perennial germplasm), as delineated by Cox et al. (2013).

Sorghum

Grain sorghum (*Sorghum bicolor*) is the world’s fifth most important cereal crop, following maize, rice, wheat, and barley (http://www.fao.org). It is the primary grain crop for over 750 million people residing in semi-arid tropical regions of Africa, Asia, and Latin America. While the United States is currently the top sorghum producer worldwide, the crop was first domesticated in Africa and arrived in the U.S. with the slave trade between 200-300 years ago (Smith and Frederiksen 2000). U.S. grown sorghum is not primarily consumed as a grain crop, but rather exported or used domestically for livestock feed and biofuel production (Cox et al. 2018). Sorghum’s importance is bolstered by its comparative heat and drought tolerance amongst the primary grain crops (Paterson et al. 2013). In addition, while many crops are expected to have decreased nutrient content under climatic conditions with elevated CO_2 levels, C4 photosynthesis crops such as sorghum have exhibited resilience in maintaining their nutrient content (Myers et al. 2014; Zhu et al. 2018). Hence, in the context of a changing environment, improved sorghum cultivars may prove key to global food security.

In addition to sorghum's general importance as a primary grain crop, it is a particularly appealing target for perenniality. The Sorghum genus contains interfertile species that act as both perennials and annuals, meaning that two perennial grasses *Sorghum halepense* and *S. propinquum* can function as genetic sources of perenniality when intercrossed with the grain crop *S. bicolor* (Washburn et al. 2013). Additionally, sorghum has the capacity to grow across highly diverse landscapes, which when paired with its drought and heat tolerance make this crop theoretically capable of productivity on lands deemed marginal for other crops. FAO (2000) estimated that roughly 910 M ha of arable land are either too marginal or degraded for crop productivity. Thus, any crop with the capacity to transition some of this land into productivity has great potential for increased global food production.

Sorghum also has the ability to regrow or 'ratoon' after harvest, while its relatives such as sugarcane are routinely ratooned (McCormick et al. 1995). The capacity of sorghum to ratoon varies across cultivars and is only feasible in tropical or semitropical environments (Duncan et al. 1980; Duncan and Moss 1987). Harvests from the secondary ratooned crop tend to be significantly lower-yielding than the primary stand (Duncan and Moss 1987). Thus, there is the potential for a short-term goal of an improved ratooning sorghum for warmer climates as the longer-term goal of a true perennial sorghum is pursued.

Finally, sorghum is a remarkably versatile crop, with varieties for food, forage, fiber (Berenji et al. 2011), sugar (Winberry 1980), and potential for biofuel (Han et al. 2012).

Theoretically, this allows for a wide array of perennial sorghum production systems to meet a wide range of human needs.

Perennial sorghum "hardware" development

There has been increasing interest in the development of perennial cereals in recent years due to their purported ecological benefits relative to annuals (DeHaan and Ismail 2017; Glover et al. 2010; Smaje 2015). These benefits can be distilled into four key advantages: a longer growing season, improved access to water and nutrients, more efficient use of nutrients, and better adaptation to marginal growing environments (Cox et al. 2006). Particularly salient to subsistence or smallholder agriculture systems is the added benefit of perenniality functioning as a form of insurance against uncertainties in future income and capital availability. This is due to perennial crops facilitating: reduced expenditure on inputs such as seed and fertilizer; reduced labor in weed control; increased reliability in stand establishment; and reduced transplanting and other stoop labor (Cox et al. 2013).

However, there are some major barriers to the perennial vision, the first being the perennial "hardware", or germplasm. While all major cereal crops are grown as annuals, many are thought to have been domesticated from perennial ancestors and/or have perennial relatives. Accordingly, the development of perennial grain sorghum has been undertaken by hybridization of the annual *S. bicolor* with one of two wild perennial grasses, *S. halepense* or *S. propinquum*. Both *S. bicolor* and *S. propinquum* are diploid (2n=20) species, estimated to have diverged about 1-2 million years prior to domestication (Paterson 2008). *Sorghum halepense* is a tetraploid resulting from natural hybridization of *S. bicolor* and *S. propinquum* followed by a doubling of chromosome number to 40 (Sezen et al. 2016). *Sorghum propinquum* is native to southeast Asia and while it holds significant potential for the development of a perennial grain sorghum in the tropics and subtropics, it has limited capacity to overwinter and is therefore not viable parent material for mid-latitude or northern temperate regions. Alternatively, *S. halepense* is a noxious invasive weed, present on six continents, and can serve as a source of parent material for temperate regions in which overwintering is necessary (Paterson et al. 2013).

Vegetative regrowth in perennial plants can be initiated from rhizomes and/or the crown of the plant, thus the perennial phenotype is associated with both tillering and rhizomatousness (Paterson et al. 1995). Both tillers and rhizomes develop from axillary buds at the lowermost nodes of the erect leafy shoot of a plant. Rhizomes are subterranean stems which grow diageotropically and sprout to form the next season's crop and are considered key to winter survival (Gizmawy et al. 1985). Both the perennial *S. propinquum* and *S. halepense* produce rhizomes, while the annual *S. bicolor* does not. Unlike the reduced yields observed in secondary ratoon crops in sorghum, secondary yields from rhizomatous sorghum have been found to be comparable to first-season grain yields, thereby reinforcing the idea that rhizomes are necessary for a truly perennial grain sorghum (Nabukalu and Cox 2016).

Sorghum has served as a useful model to determine the genetic bases of perenniality, with *S. bicolor* x *S. propinquum* derived populations investigated first and in most detail to date (Paterson et al. 1995; Kong 2013b). Additionally, it's relatively small genome size of 730 Mb has allowed it to become a model organism for C4 photosynthesis crops more generally and function as a useful complement to the C3 model crop, rice (Paterson et al. 2009). In an RFLP mapping study of an *S. bicolor* x *S. propinquum* cross, Paterson et al. (1995) found genomic regions associated with rhizomatousness and perenniality dispersed over at least nine of sorghum's ten linkage groups. Studies have indicated QTL correspondence between tillering and rhizomatousness, suggesting pleiotropic effects for these genomic regions (Paterson et al. 1995;

Hu et al. 2003; Westerbergh and Doebley 2004; Kong 2013a). Prior studies have indicated that the lack of rhizomes in *S. bicolor* has minimal effect on the corresponding genes that show rhizome enriched expression in *S. propinquum*, and that the more extensive rhizomes in *S. halepense* compared to its progenitors are accompanied by "recruitment" of *S. bicolor* genes (Jang et al. 2009). Comparative QTL studies have indicated non-random correspondence in genomic regions conferring rhizomatousness in rice and sorghum (Hu et al. 2003) and some corresponding genomic regions in maize and sorghum (Westerbergh and Doebley 2004), which suggests that further elucidation of rhizome QTLs in sorghum may be generalized to other species.

While F1 hybrids derived from crosses between the diploid *S. bicolor* and tetraploid *S. halepense* are typically tetraploid, it has recently been shown that certain crosses result in a high proportion of diploid progeny (Cox et al. 2018). Viable diploid germplasm resulting from such crosses can greatly increase the expediency of perennial sorghum development, allowing for homozygosity to be reached more quickly, simplifying genetic analysis, and permitting a broader base of parent material in having the same chromosome number as conventional sorghums.

Broadening the scope of perenniality research in sorghum to include *S. bicolor* x *S. halepense* derived populations has great potential to accelerate the perennial sorghum germplasm development. One particularly appealing theory is the potential for *S. halepense* to confer an array of potentially beneficial alleles due to its capacity to naturalize in a wide range of environments, perhaps facilitating lines that are cold tolerant, drought tolerant, and disease resistant (Paterson et al. 2013). Kong's (2017) investigations of a novel BC_1F_2 population derived from a *S. bicolor* x *S. halepense* cross, with *S. bicolor* as the recurrent parent, highlighted the correspondence of rhizome QTLs to *S. halepense*'s progenitors, *S. bicolor* and *S.*

propinquum. This research also assisted in further narrowing of candidate genes for rhizomatousness.

Proposed here is an exploration of a novel F2:3 *S. bicolor* x *S. halepense* derived population, which can further elucidate genomic regions pertinent to perenniality and contribute to germplasm development.

Perennial sorghum "software" development

A task that is arguably equally as daunting as breeding perennial germplasm is development of perennial "software", or agronomic and ecological systems facilitated by the perennial germplasm. While germplasm is obviously the primary limiting factor to perennial grain systems, adoption of such germplasm is not guaranteed. Adebyi et al. (2015) categorized perennial grains as a "transformative technology" in that they fundamentally differ from their conventional annual counterparts. Motivations for adoptions of such agroecological technologies are poorly understood and often exist in tension with tendencies towards risk aversion by farmers (Duru et al. 2015). Thus, any efforts towards "derisking" the novel technology of perennial germplasm by developing robust, viable perennial "software" is of critical importance. With great modern advancements in plant breeding, the timeline of variety development is greatly reduced, allowing for concurrent investigations into both "hardware" and "software" development (Crews et al. 2018)

However, much of the attempted "software" development of functional perennial grain agricultural systems suffers from a "Catch-22" due to the novelty of the varieties. Breeders would like input regarding the agronomic system a perennial grain will be grown in to inform their selections, yet field-level assessments cannot be performed because these crops are not yet fully developed (Cox et al. 2013). Thus, attempting to ascertain minimum yield and ecosystem service thresholds required for a viable perennial grain sorghum becomes a challenge, but one worth attempting to overcome. Participatory plant breeding (PPB), in which both breeders and farmers participate in the selection and development of crop varieties, has been suggested to result in varietal release 7-10 years faster than conventional breeding methods (Witcombe 2005). This highlights the efficacy of breeding efforts that concurrently integrate agronomic assessments. These field-level assessments will likely prove to be even more critical to the timeliness of perennial variety development, as they necessitate novel growing systems (Adebyi et al. 2015). The successful implementation of PPB methods in organic variety development further supports the notion that alternative modes of low-input farming that are more closely tied to local ecologies have much to gain from PPB (Campanelli et al. 2015; Ghaouti et al. 2008; Shelton and William 2016).

In the absence of a truly perennial grain sorghum, there are a few key avenues in which some ecosystem services of these future crops can be deduced: assessments of intermediate germplasm; assessments from other perennial grain crops that may be further along the germplasm development pipeline, namely intermediate wheatgrass (*Thinopyrum intermedium*, also referred to as Kernza) and rice; modelling or simulations of the anticipated germplasm; and interviews conducted with farmers who are either early adopters of perennial grains or considering adoption of such varieties.

Research to-date

In a 2009 perennial sorghum study, the highest-yielding experimental lines derived from an *S. bicolor* x *S. halepense* cross had mean yields that were 54% of the commercial grain sorghum check. Additionally, it was found that the perennial lines and commercial check produced similar number of seeds per unit of land area, highlighting that the difference in yield was entirely due to difference in seed weight (Cox et al. 2018). Strength of perenniality among the hybrid progeny was less than that of *S. halepense* but did not decline over recurrent selection cycles for increased yield and remained sufficient.

In regard to improved nutrient access, of particular concern to the Georgia Piedmont region is an accumulation of phosphorus in the soils due to repeated broiler litter application. This phosphorus is vulnerable to transport to surface waters, and thus a significant pollution concern (Butler et al. 2010). Strategic sorghum planting could potentially be a useful tool in phosphorus management for the region, as the genus has shown a propensity to associate with arbuscular mycorrhizal fungi that increases its phosphorus uptake in some conditions (Neumann and Eckhard 2004). Additionally, *S. halepense* contains nitrogen-fixing endophytic bacteria in its rhizomes which make phosphorus more readily available (Weiman 2012).

While field-level research has been, until quite recently, inhibited by the lack of perennial grain varieties, this is beginning to shift with progress in Kernza breeding efforts. Initial research has indicated that dual-use management (forage and grain) of Kernza results in greater grain yields, heightened short-term nutrient availability, and greater root biomass with no negative effects on soil health (Pugliese 2017). When compared to its annual counterpart, Kernza has been found to have higher water-use efficiency, higher evapotranspiration rates, and function as a stronger carbon sink (de Oliveira et al. 2018). Intermediate wheatgrass has also been found to reduce nitrate leaching by 98% when compared to annual wheat (Culman et al. 2013).

Additionally, interviews with initial Kernza adopters indicates that it is primarily valued as a multiple-use crop and tends to be grown on marginal land within a grower’s farm (Lanker 2018). Weed suppression and labor reduction were often cited as the crop’s biggest draws. Comparative surveys between French and U.S. farmers indicate that U.S. farmers are particularly interested in dual-use varieties for grain and forage (Wayman et al. 2019). Adebyi et al. (2015)

investigated motivations for adoptions of the novel perennial wheat through a series of farmer interviews in Michigan and Ohio. Very few farmers were found to be concerned with perennial wheat's ability to compete with annual wheat on the basis of yield, but many farmers were interested in this crop utilizing a niche that was currently unaccounted for, particularly marginal or under-used areas of their respective farms.

Building on the idea of perennial grains being utilized for reasons other than their primary grain yield, economic analysis of perennial wheat systems in Australia indicate that dual-use grain and forage systems are the most viable (Bell 2008; Bell 2013). However, Pimentel et al.'s 2012 production cost comparison between annual and perennial wheat cropping systems under two literature-derived scenarios (Bell et al. 2008; Glover et al. 2010), indicated that a grain-only perennial wheat crop could still be an economically viable alternative. Ryan et al. (2018) emphasize that multifunctionality is key to the success of a perennial grain system, as it is the simplest way to offset the relatively lower grain yields. In particular they suggest dual-use grain and forage systems intercropped with legumes to maximize benefits. More diverse perennial polycultures may maximize potential ecosystem services, but the high level of complexity in such systems may inhibit their adoption. This all suggests that the development of a multi-use perennial sorghum should be a priority for breeders.

Multifunctional perennial grain sorghum

In light of increasing evidence that a multi-use perennial sorghum is the most viable cropping scenario, it becomes important to investigate the realities of such a scenario. For sorghum, there are a number of multi-use options available to complement the primary grain harvest, primarily: forage, soil conservation, and biofuel. While there has been growing interest in sorghum as a biofuel source (Murray et al. 2008), as of yet this has not been realized (Mathur et al. 2017). Thus, dual-use grain and forage, and dual-use grain and soil conservation, appear to be the most likely scenarios to date.

Economic viability is often of utmost importance to farmers, who frequently rely on enterprise budgets for profitability analysis (Doye 2016). UGA's Department of Agricultural and Applied Economics provides such budgets for Georgia grain sorghum, both dryland and irrigated (agecon.uga.edu/extension/budgets.html).). These farm budgets can serve as the basis for comparison between an annual and ratooning/perennial grain sorghum under different dual-use scenarios to determine the viability and potential adoption rates of these alternative cropping systems. These farm budgets can also help determine break-even yield targets for breeders. While it's estimated that up to 50% of farmer costs can be reduced under perennial cropping schemes (Crews et al. 2018), research in this space has been relatively lacking.

One limitation to a dual-use grain and forage system is the propensity for prussic acid to accumulate in sorghum, which is poisonous to livestock and ruminants in particular (Robson 2007; Vough 1978). This is prohibitive to the direct grazing model Bell investigated in perennial wheat (2013). Toxicity concerns are abated in harvested and cured silage, so there is still the potential for an economic model in which forage is consumed in this manner (Vough 1978). This type of system will necessitate the additional cost of biomass harvest and will thus theoretically be less profitable than a direct grazing scenario.

Critical to the analysis of a dual use grain and soil conservation scenario is the provision of Natural Resources Conservation Service (NRCS) Environmental Quality Incentives Program (EQIP) payments. NRCS provides financial and technical support to farmers and ranchers in the voluntary employment of conservation practices, with EQIP being the agency's most frequently utilized conservation program (nrcs.usda.gov). EQIP payments can supplement the primary revenue stream, helping to facilitate shifts to perennial sorghum cropping systems. Price incentives, such as subsidies or price premiums have been shown to be powerful tools in facilitating shifts to organic production models (Serra et al. 2008). Of additional interest is the eligibility of farmers in the north Georgia region for such payments. Investigating erosion levels in the region under current cropping scenarios can potentially highlight such eligibility. Similar work involving RUSLE-based simulations of shifts to perennial planting schemes in Tuscany, Italy have indicated significant reduction in soil erosion potential (Vallebona et al. 2016).

Ultimately, the burgeoning literature in this field can be synthesized in attempts to quantify thresholds of ecosystem services and minimum yields that would maximize the adoptability potential of perennial grains and inform the ongoing breeding process.

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CHAPTER 2

QUANTITATIVE TRAIT MAPPING OF RHIZOMATOUSNESS AND OVERWINTERING IN A $F_{2:3}$ POPULATION OF *SORGHUM BICOLOR* X *S. HALEPENSE*[1]

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Abstract

Perennial grains hold great potential for meeting future food needs in an ecologically sustainable manner. Sorghum (*S. bicolor*), the world's fifth most important cereal crop, is a particularly appealing target for conversion to perenniality due to its close relationship to two wild perennial grasses, *S. halepense* and *S. propinquum*, and its potential to grow across highly diverse landscapes. Arguably the most direct route to perenniality lies in the development of rhizomes, subterranean stems that sprout to form the next season's crop. A novel $F_{2:3}$ population derived from an *S. bicolor* x *S. halepense* cross was grown in two environments: Watkinsville, GA and Salina, KS. Quantitative trait locus (QTL) analysis was employed to identify genomic regions pertinent to rhizome development and winter survival, which can contribute to marker-assisted selection in breeding of a perennial sorghum variety. QTLs pertaining to perenniality were found scattered across eight of sorghum's ten chromosomes, mostly corresponding to regions identified in prior *S. bicolor* derived populations. Results contributed to the validation of previously identified QTLs relating to rhizomatousness in the *Sorghum* genus and identified the first QTLs relating to winter survival in a *S. bicolor* x *S. halepense* derived population. Results also provided support for a previous finding that later flowering is associated with fewer rhizomes as well as the potential for *S. halepense* alleles to improve conventional sorghum varieties for traits such a flowering, plant height, and grain yield.

Introduction

In recent years, there has been increasing interest in perennial grains due to their hypothesized benefits relative to their annual counterparts (DeHaan and Ismail 2017; Glover et al. 2010; Smaje 2015). Such benefits can be delineated as either ecological, encompassing a longer growing season, improved access to water and nutrients, more efficient use of nutrients, and better adaptation to marginal growing environments; or social, including reduced input costs, amortized seed cost over the lifespan of the plant stand, reduced weed control, increased reliability in stand establishment, and reduced stoop labor such as transplanting (Cox et al. 2006; Cox et al. 2013). However, essentially all the major grain crops are currently grown as annuals and significant breeding efforts are a prerequisite to establishment of perennial alternatives.

One prime candidate for perennialization is grain sorghum (*Sorghum bicolor*). Sorghum is the fifth most important cereal crop, globally, and has high heat and drought tolerance paired with the capacity to grow across highly diverse landscapes (Paterson et al. 2013). These qualities inherent to the crop can potentially bolster the ecological advantages of perennialization, resulting in cultivars theoretically capable of productivity on lands deemed marginal for other crops. Hence, sorghum has great potential for increased food security and ecological resilience.

In addition to *S. bicolor*'s general importance as a food crop, the species has particular advantages from a breeding perspective. The Sorghum genus contains interfertile species that act as both annuals and perennials, with the perennial grasses *Sorghum halepense* and *S. propinquum* potentially serving as genetic sources of perenniality when intercrossed with the grain crop *S. bicolor* (Washburn et al. 2013). Sorghum also has the capacity to regrow or 'ratoon' after harvest, albeit varying across cultivars and only feasible in tropical or semi-tropical environments (Duncan et al. 1980; Duncan and Moss 1987). Secondary 'ratoon crop' harvests tend to be markedly lower yielding than the primary crop but offer the potential of an improved ratooning cultivar as a segue toward a perennial cultivar and associated production system (Duncan and Moss 1987).

Perennial habit is associated with both tillering and rhizomatousness, with vegetative regrowth in perennial plants initiated from rhizomes and/or the crown of a plant (Paterson et al. 1995). While both tillers and rhizomes develop from axillary buds at the lowermost nodes of the erect leafy shoot of a plant, rhizomes, subterranean stems, are considered critical to winter survival (Gizmawy et al. 1985). Both the perennial *S. propinquum* and *S. halepense* produce rhizomes, while the annual *S. bicolor* does not. Unlike the reduced yields observed in secondary ratoon crops in sorghum, secondary yields from rhizomatous sorghum have been found to be comparable to first-season grain yields, thereby reinforcing the idea that rhizomes are necessary for a truly perennial grain sorghum (Nabukalu and Cox 2016). Diploid *S. propinquum* (2n=2x=20) has a limited capacity to overwinter in mid-latitude and temperate regions, while tetraploid *S. halepense* (2n=4x=40) is an invasive and noxious weed present on six continents and can serve as parent material for temperate regions in which overwintering is required (Paterson et al. 2013).

Sorghum has served as an excellent model in dissecting the genetic basis of perenniality, with the first and most detailed investigations to date taking place in *S. bicolor* x *S. propinquum* derived populations (Paterson et al. 1995; Kong 2013; Kong et al. 2015). Previous studies have indicated that the lack of rhizomes in *S. bicolor* has minimal effect on the corresponding genes that show rhizome enriched expression in *S. propinquum* and that the more extensive rhizomes in *S. halepense* are accompanied by "recruitment" of *S. bicolor* genes (Jang et al. 2009). Genomic regions containing QTLs pertaining to rhizomatousness show some enrichment of genes associated with rhizome expression (Jang et al. 2006). Further elucidation of rhizome QTLs in sorghum can potentially be generalized to other species, as prior studies have indicated non-random correspondence between genomic regions conferring rhizomatousness in rice and sorghum (Hu et al. 2003) and some correspondence between maize and sorghum (Westerbergh and Doebley 2004).

In this project, we conducted a quantitative trait locus (QTL) analysis of traits pertaining to perenniality in a novel $F_{2:3}$ *S. bicolor* x *S. halepense* population, derived from a tetraploid *S.* bicolor BTx623 and the *S. halepense* accession Gypsum 9E. QTLs pertaining to perenniality can be compared to rhizome-conferring chromosomal regions identified in *S. bicolor* x *S. propinquum* populations (Paterson et al. 1995; Kong et al. 2015) and a *S. halepense* x *S. bicolor* BC_1F_2 (Kong 2017), thereby distinguishing "consensus" QTLs from those potentially novel to the *S. halepense* cross. Potential linkages between the perennial phenotype and other traits pertinent to a viable grain sorghum, such as inflorescence weight and plant height will also be investigated. Of additional interest is an investigation into the relationship between flowering time and fewer rhizomes, which was an unexpected finding in a prior study (Kong 2017). Additionally, *S. halepense*'s high degree of genetic diversity may elucidate significant QTL differences across environments. The methods used will capitalize on methodology established in prior studies (Paterson et al. 1988; Kong 2017) and utilize the previously constructed *S. bicolor* reference genome (Paterson et al. 2009).

Methods

Genetic Stocks

A tetraploid F_1 hybrid, H6, was derived from a cross between two morphologically different parents, *S. bicolor* BTx623 and *S. halepense* Gypsum 9E. The F_1 hybrid was selfed to generate 300 F_2 individuals. These individuals were planted in a completely randomized design with 2 plants per plot at 12-inch intervals in Watkinsville, GA in May 2011. Ten plots were randomly assigned as controls in which were planted one of each parent plant, *S. bicolor* and *S. halepense*. F_2 plants were selfed to generate the $F_{2:3}$ population. F_3 plants (3 m rows of about 10 plants) were grown in a completely randomized design in 2012 in two environments:

Watkinsville, GA (three replications, planted May 17) and Salina, KS (3+ replications, planted June 5). As in the F_2 population, 10 plots were randomly assigned as controls in which were planted one of each parent, *S. bicolor* and *S. halepense.* Plants were phenotyped for traits relevant to perennial germplasm development as outlined in Kong (2017) and elaborated on below.

Genotyping

Leaf samples of the F_2 population were frozen at -80 C and lyophilized for 48 hours. Genomic DNA was extracted from the leaf samples according to Al-Janabi et al. (1999). Genotyping by sequencing (GBS) was performed using a slightly modified version of Multiplex Shotgun Genotyping (Andolfatto et al. 2011). Sequencing was performed with an Illumina HiSeq 2500 Rapid V2 kit, which generated roughly 150 million reads of 100 base pair fragments per run using single-end sequencing. Single nucleotide polymorphisms (SNPs) were called using a modified TASSEL pipeline utilizing the previously constructed reference genome *S. bicolor* BTx623 *v1.4* (Paterson et al. 2009). Burrows-Wheeler Aligner was used to map reads to the reference genome (Li and Durbin 2009), Samtools was used to sort the data (Li et al. 2009), an index was built using Picard Tool's build bam index function (Wysoker et al. 2013) and GATK's Haplotype Caller was used to call SNPs (Poplin et al. 2017). SNPs were then filtered based on GATK's recommended filtration. Markers with coverage less than 10x were considered as missing data. Markers were removed if more than 50% of genotypes were missing.

Phenotyping

Varying climatic survival rates resulted in phenotyping of 238 F_3 genotypes in the GA population and 170 F_3 genotypes in the KS population, along with the 300 F_2 parents for the following traits:

Rhizome number (RN) was measured by counting the number of rhizome-derived shoots emerging aboveground at the end of the growing season.

Rhizome distance (RD, not recorded in KS F_3 population) was measured as the distance of the rhizome-derived shoot (cm) from the middle of the source crown.

Rhizomatousness (RZ) was calculated for each genotype as the number of plants with rhizomes as a proportion of the total number of plants. For the single plants in the F_2 population, only scores of 0, 1, or .5 (intermediate, for cases in which above-ground shoots had some characteristics of rhizomatous origin but inadequate separation from the crown to be certain and digging would have risked damage) were possible.

Winter Survival (WS) was measured in the spring following the initial growing season by calculating the percentage of plants that displayed spring regrowth for a given genotype.

Plant height, (HT) a component of aboveground plant productivity, was measured at the plant's physiological maturity, from the base of the plant to the tip of the primary flower head.

Inflorescence weight (INFL, GA) and grain yield (YLD, KS) was taken after harvesting as components of crop yield.

Flowering time (FL) was measured by recording the number of days until flower heads emerged from the leaf whorl ('boot') of a plant from planting date.

Shattering, (SH) or seed dispersal by disarticulation, was assessed at seed maturity.

Averages of the F_2-derived F_3 progeny phenotypes in each environment were taken. In order to facilitate regression modelling, skewed data was transformed to best approximate a normal distribution using either log or square root functions. Flowering time and inflorescence/grain weight were transformed with the square root function, while rhizomatousness and rhizome count were log (n+1) transformed. Data was reviewed for clear outliers, which were omitted when appropriate.

Narrow sense heritability was calculated using parent-progeny regression. Parent-progeny regression is most accurately used to estimate narrow sense heritability when environment is not a covariate among the parent and progeny populations (Vogel et al. 1980, Ward 1994). For this reason, comparing the F_2 Georgia grown population to the F_3 Kansas grown progeny presumably best approximates narrow sense heritability for the traits in question. For instances in which a given phenotype was not recorded in the KS population (e.g. rhizome distance, shattering), parent-offspring regression was performed on the GA F3 population, although such estimates may overstate the true narrow-sense heritability. This is due in part to the violation of a core linear regression assumption of no multicollinearity. A pooled parent-offspring analysis was also performed as a reference point. For the pooled analysis, environment was accounted for as an additional factor in the regression model.

Correlation analyses were performed for all traits in each population using the statistical package R (R Core Team 2013), while graphs were constructed with the program Corrplot (Wei and Simko 2017).

Genetic Mapping

A QTL is a locus deemed statistically significant in its association with quantitative variation in a phenotype. It is defined by boundaries that are demarcated by linked genetic markers. QTL mapping is a method of linking a phenotype of interest to chromosomal regions harboring causal genes. While the simplest method of QTL mapping, single marker analysis, does not require the construction of a genetic map, more advanced methods, interval mapping (Lander and Botstein 1989) and composite interval mapping (Zeng 1994), utilize flanking marker information to achieve higher statistical power, and thus require a genetic map. This research utilizes interval mapping, requiring construction of a genetic map as well as single marker analysis.

Genetic map construction in polyploids, as in the tetraploid progeny investigated here, is inherently more complicated, primarily because of the greater number of possible genotypes (Ferreira et al. 2019; Ripol et al. 1999). One method of dealing with the added complexity of polyploids is reliance on simplex markers, where single dosage of the allele approximates diploid inheritance patterns (Wu et al. 1992). Thus, after SNPs were filtered using the methodology outlined above, markers were further culled to consist of only simplex markers. This imposes the constraint that only DNA markers that were heterozygous in the SH parent are used in the analysis.

Markers were then reviewed and further filtered in R/qtl, with the removal of markers with over 60% missing data, removal of individuals exceeding 80% of missing marker data, and the removal of markers with segregation distortion explicable by chance with likelihood of less than 1E-10. The dropSimilarMarkers function in qtlTools was used to remove redundant, less informative markers on each chromosome (Delaneau et al. 2016). JoinMap was employed in final map construction, with linkage groups formed with a minimum LOD score of 10, and a *de novo* marker order established for each linkage group (Stam 1993). The genetic map was constructed using the Kosambi mapping function and an error probability of 0.01.

Single marker analysis

Informative markers were culled to a suitable subset based on the following augmentation of the procedure outlined in Kong (2017):

1. Select simplex markers based on the genotype of the F_1 parent. Single marker analysis was also tested in duplex and triplex markers, but these markers had less power in QTL detection that was assumed to stem from inaccuracies in identifying number of allele copies. Simplex markers had comparable genome coverage to other marker types.
2. Remove markers with missing data in excess of 60%.
3. Remove markers with minor/homozygous allele frequency less than .02
4. Remove markers that fail to show linkage to any other marker at a 20% recombination rate.
5. Select markers that are at least 100 bp apart, as SNP markers closer than this distance provide little additional info in populations displaying strong linkage disequilibrium.

Once a suitable subset of markers was formed, a linear regression was run for each phenotypic trait, with phenotype as the response variable for each marker genotype in the population. For instances of categorical response variables, such as the presence or absence of rhizomes, logistical regression was used for analysis. Subsequent p-values were transformed by -log10 and then had a significance threshold of applied according to permutation test results for the trait in question. Statistical analyses were performed in R (R Core Team 2016). A SNP was only considered significant if it was flanked by at least four markers also crossing the significance threshold, with peak SNPs chosen by the highest value within a grouping.

QTL mapping

QTL mapping was carried out with the interval mapping method on each trait of interest in each population x environment (F_2, GAF_3, KSF_3) (Lander and Botstein 1989). R/qtl was used to perform the QTL analysis (Broman et al. 2003). Percentage of variance explained for each QTL was determined by constructing an additive model with significant QTLs using the ‘fitqtl’ function. A standard LOD threshold of 3.0 was applied. While permutation tests were performed for the traits in question, suggested LOD thresholds for an alpha value of .05 were prohibitive to the detection of any QTLs pertaining to perenniality, thought to be a result of the high environmental effect in traits pertaining to rhizome development and capacity to overwinter. A LOD interval of 1 was used to determine the QTL range, with the flanking left and right borders corresponding to 1-LOD interval drops in likelihood.

Trait RD contains many zero data points for individuals which did not exhibit rhizome development. Thus, a two-part QTL model was employed in evaluating rhizome distance (RD), which allows for the disentanglement of QTLs pertaining to the presence of rhizomes and distance of the same rhizomes (Kong 2017). First a binary model is run for the presence/absence of rhizomes and then a subsequent analysis is run on the rhizomatous individuals to evaluate the genomic regions associated with distance of the rhizomes present.

Comparison with prior studies

Correspondence between QTLs detected in this investigation and prior *S. bicolor* derived populations was determined using the physical locations reported in the Sorghum QTL Atlas, which provides a basis for comparison of QTL intervals across different studies in sorghum (Mace et al. 2019). In the case of Kong (2017), whose findings were not reported there, the physical locations of QTL intervals reported in the research were used as reference. A QTL was determined as previously undetected if its likelihood interval did not overlap with the likelihood interval of any prior QTL. A shortcoming of the single marker analysis approach employed here is that it prohibits the ability to detect a likelihood interval for a QTL. In such cases it is noted if a SMA QTL peak does not overlap with a previously identified QTL interval.

Results

Summary statistics and heritability

The GAF_3 population displayed the highest prevalence of rhizomes, with an average RZ value of 0.46 (Table 2.3). The F_2 and KSF_3 population were lower, with average values of 0.38 and 0.32. The lack of replication in the F_2 population limits the number of possible values for this trait to 0, 1, or occasionally .5 in instances deemed intermediate, which contributes to the larger standard deviation for this trait in that population. Mean rhizome count was markedly similar among all three populations, ranging from 0.73-0.75 (Table 2.3). Among the F_3 populations, the KS environment showed a greater standard deviation for this trait. All populations had significantly less rhizomes than the perennial *S. halepense* G9E parent, but the large standard deviation relative to mean values suggests sufficient variability for selection on this trait. Mean rhizome distance ranged from 11.97 cm in the F_2 population to 13.32 cm in the GAF_3 population (Table 2.3) but was not recorded in the KSF_3 population. GAF_3 values were comparable to the G9E parent in 2012.

Winter survival rates starkly varied between the two growing environments in the F_3 population, at 61% for the GA grown population compared to 8% of the KS population (Table 2.4). Both environments' standard deviation rates suggest significant variability and the potential for trait improvement. Survival rates fell far below those of the perennial parent, G9E.

Average flowering date was 57 days after planting in the F_2 population, 61 days after planting in the GAF_3 population, and 80.3 days after planting in the KSF_3 population (Table 2.5). All mean $F_{2:3}$ population values fell between the parent values for that environment and standard deviations were comparable among all three populations. Plant height values were similar between the two GA grown populations, but considerably lower than in the KSF_3 population

(2.5). Average plant heights in the GAF_2 and KSF_3 populations exceeded both parental values, suggesting potential transgressive segregation for this trait. Average dried inflorescence weight was higher in the F_2 population than the GAF_3 population (Table 2.5). KSF_3 population data was for grain yield rather than dried inflorescence and had lower average values. Mean inflorescence and grain yield values more closely resembled the perennial G9E parent but did exhibit significant variation and the potential for increased yields.

Flowering time, plant height, and shattering all proved to be highly heritable traits (Table 2.6). Inflorescence weight and rhizome count were intermediate, showing significant heritability in the GA and pooled analysis, but not in the KS parent-offspring regression. The suitability of a parent-offspring regression may have been impacted for trait dry weight inflorescence in KS, due to the KS population having a slightly different trait (grain yield) measured. The $F_{2:3}$ population was segregating for floret sterility, which may have introduced substantial noise to the relationship between head and grain weight. Rhizome distance had a low heritability that was not statistically significant.

The F_2 population displayed few statistically significant correlations amongst the various phenotypic variables (Figure 2.1). Inflorescence dry weight was positively associated with both plant height and rhizome distance, while flowering time had a negative association with inflorescence dry weight and number of rhizomes.

In the Georgia F_3 population, flowering time was positively associated with plant height and negatively associated with both number of rhizomes and winter survival rates (Figure 2.2). Inflorescence dry weight was positively associated with plant height and negatively associated with seed shattering. Shattering also had a slight positive association with winter survival.

Rhizomatousness, number of rhizomes, and rhizome distance were all positively associated with winter survival.

In the Kansas F_3 population, flowering time was positively associated with plant height and negatively associated with grain yield, rhizomatousness, and number of rhizomes (Figure 2.3). Grain yield dry weight was positively associated with plant height, rhizomatousness, and number of rhizomes. Rhizomatousness and number of rhizomes intuitively has a strong positive association with one another and with winter survival.

Genetic map

Initial SNP calling resulted in 135,977 markers. Subsequent thinning and filtering steps culled this number down to 49,375 markers. Of these, 10,795 were simplex markers suitable for map construction. We formed 2161 markers into linkage groups in JoinMap using stringent thresholds of a minimum LOD score of 10. The final genetic map consisted of 240 individuals and 816 markers, spanning a total of 2498.2 cM. Three of the genotyped individuals were removed due to large amounts of missing data. The genetic map had 27 linkage groups, with each Sorghum chromosome corresponding to 1-5 homologous groups (Table 2.1). Average marker spacing on linkage groups ranged from 1.93-17.42 cM, with average spacing for linkage groups falling at 4.27 cM.

Chromosomes 3 and 9 exhibited particularly high levels of segregation distortion, leaving fewer markers suitable for map construction. Excessively high levels of segregation distortion can prove prohibitive to accurate genetic map construction, as unlinked markers can appear linked. This was apparent in an initial analysis of simplex markers after the SNP calling and filtering steps, where Chromosome 3 displayed disproportionately high levels of genotype AAAA and Chromosome 9 displayed disproportionately high levels of genotype ABBB (Table

2.2). Because this is a subset of simplex markers, genotypes ABBB and BBBB should not be present assuming Mendelian segregation, however occasional inference of these genotypes is expected due to the limitations of GBS in accurately identifying the number of allele copies at a given marker location or due to double reduction (Clark et al. 2019). Despite this expectation, the 14% of ABBB genotypes on chromosome 9, is markedly higher than 7% average found across the entire genome. Interestingly, while one might expect an increase of genotype ABBB to correspond with a paucity of genotypes AABB or AAAB, indicating inaccuracy in identifying the number of alternative allele copies, in the case of chromosome 9 there is instead a paucity of genotype AAAA. Markers were less concentrated in the central chromosomal regions, as compared to the distal regions across all chromosomes, corresponding to observations in prior *S. bicolor* x *S. halepense* derived populations (Kong 2017).

QTL Analysis

Flowering time (FL)

In the F_2 population, three QTLs were detected for flowering time with interval mapping (Table 2.7). Two of these QTLs accounted for 21.2% of the variation. An additional 5 QTLs were identified in the F_3 populations, with two QTLs on chromosome 6 detected in both the GA and KS environment in the F3 population, accounting for 26.5% and 25.2% of variation, respectively. Seven of the eight QTLs detected by interval mapping had a positive allele effect, suggesting that *S. halepense* alleles tend to contribute to later flowering, despite our observation that *S. halepense* flowered earlier than *S. bicolor* in all three populations (Table 2.5). For populations grown in the GA environment, flowering time was transformed with square root function to approximate normal distribution. This contributed to the different scales of additive allele effect among the GA and KS grown populations.

Single marker analysis supported the findings of interval mapping and identified a few additional putative QTLs (Figure 2.4). Two QTLs were detected on chromosome 6 and one on chromosome 10 for all three populations (Table 2.14). Chromosome 6 contained the QTL with the strongest effect in all three populations, with qFL6.F2.2 at 43.32 Mb having an effect of 11.54 days in the F_2 population, qFL6.F3.2 at 45.03 Mb having a 9.94 day effect in the GAF_3 population, and qFL6.F3.4 at 43.2 Mb having a 12.4 day effect in the KSF_3 population. A QTL was detected on chromosome 1 in the F_2 population with a negative effect of 6.65 days. One additional QTLs with a negative effect on chromosome 1 was detected in the $GAF_{2,}$ suggesting that *S. halepense* also harbors alleles that can reduce flowering time (Table 2.14).

Inflorescence weight (INFL, GA) and grain yield (YLD, KS)

Interval mapping revealed one QTL in the F_2 population, accounting for 7.2% of the phenotypic variance in inflorescence dry weight (Table 2.8). A QTL in a comparable region of linkage group 1A was detected in the KSF_3 population with an additive allele effect of -0.49, suggesting that *S. bicolor* alleles contribute to increased inflorescence/grain weight. All populations were transformed with square root function, making effect size correspond to square root of dry weight (g). Additional QTL was detected on linkage group 1C and 4B in the KS population, with additive allele effects ranging from 0.69-0.74, suggesting that *S. halepense* alleles can also contribute to increased inflorescence/grain weight.

Single marker analysis resulted in 2, 2, and 3 QTLs in the F_2, GAF_3, and KSF_3 populations (Figure 2.5). The consensus QTL detected on chromosome 1 by interval mapping was supported by single marker analysis (Table 2.15). Both QTLs in the F_2, one out of two QTLs in the GAF_3, and two out of the three QTLS in the KSF_3 displayed a negative allele effect, further suggesting that both *S. bicolor* and *S. halepense* alleles can contribute to increased inflorescence/grain weight.

Plant height (HT)

Interval mapping resulted in one QTL for plant height, qHT.1A.F3.1, in the F_3 population, with an additive effect of -9.4 cm and 6.3% explanatory power (Table 2.9). Two QTLs were detected in the KSF_3 population, accounting for approximately 20% of the variation. The QTL on chromosome 6 accounted for the strongest effect in the F3 population. Two of the three QTLs detected by interval mapping had positive allele effects, suggesting that *S. halepense* alleles generally contribute to increased plant height, but that it also harbors alleles that can reduce height.

Single marker analysis revealed 3 and 5 QTLs in the GAF_3, and KSF_3 populations (Figure 2.6). QTLs with negative allele effect were found on chromosome 1 in both populations (Table 2.16). Five of the eight QTLs detected by SMA had positive allele effects, further suggesting that *S. halepense* harbors alleles for both increased and reduced plant height.

Rhizomatousness, the presence or absence of rhizomes (RZ)

Interval mapping in the F_2 population employed a binary model for presence/absence of rhizomes and a normal model for the F_3 populations where replications permitted a greater degree of intermediate values. Effect size in the F_3 populations is for log(n+1) of rhizome presence. Three QTLs were detected in the F_2 population, accounting for 12.8% of the variation in rhizomatousness (Table 2.10). Effects were positive for two of the three QTLs, suggesting that both *S. halepense* and *S. bicolor* alleles contribute to increased rhizome presence. One QTL was detected in the GAF_3 population, accounting for 5.5% of the variation.

Single marker analysis identified 4, 2, and 2 QTLs in the F_2, GAF_3, and KSF_3 populations (Figure 2.7). Four QTLs on chromosome 6 were detected in total across the three populations, all of which had a negative allele effect, indicating that *S. bicolor* alleles in this region contribute to increased rhizomatousness (Table 2.17). A potential QTL, qRZ7.F3.1, was detected on chromosome 7 in the GAF_3 population, which had the highest allele effect among QTLs detected in that population.

Rhizome number (RN)

Interval mapping detected three QTLs in the F_2 and two in the GAF_3 population. No QTLs were detected in the KSF_3 population for number of rhizomes (Table 2.11). The three F_2 QTLs accounted for 18.8% of the variance in rhizome number, all with a positive allele effect. The two GAF_3 QTLs explained 9.6% of rhizome number variation, both having a positive additive effect. The one QTL detected in KS, qRN.10A.F3.1, accounted for 6.8% of the variance and displayed a negative additive allele effect of -0.11.

Single marker analysis detected 3, 4, and 3 QTLs in the F_2, GAF_3, and KSF_3 populations, which was roughly twice the number of QTLs detected by interval mapping (Figure 2.8). A consensus QTLs on chromosome 1 was found across all three populations, all of which displayed a positive allele effect. All three QTLs detected on chromosome 6 displayed a negative allele effect, corroborating RZ findings (Table 2.18). QTLs were only detected on chromosome 3 in the F_2 population, as in the prior interval mapping. An additional potential QTL, qRN8.F3.1, was detected in the F3 population, which had the strongest effect among the associations found in that population. Similar to RZ, findings here suggest that both *S. bicolor* and *S. halepense* alleles contribute to increased number of rhizomes.

Rhizome distance (RD)

A two-part model was employed by interval mapping for the rhizome distance trait, which allows for a delineation between QTLs effecting rhizome presence/absence and QTLs effecting the physical spread of the rhizomes (Kong 2017). Two QTLs were detected in the F_2 population and three in the GAF_3 population (Table 2.12). This trait was not measured in the KSF_3 population. Normal effect size corresponds to log(n+1) (cm) of rhizome distance, while binary effect corresponds to rhizome presence/absence. In the GAF_3 population, qRD.7B.F3.1 had the highest explanatory power at 8.2% and qRD.1D.F3.1 had the greatest allele effect (2.8 normal, 0.7 binary). Three of the five QTLs displayed a positive allele effect, indicating that *S. halepense* alleles tend to contribute to increased rhizome distance at these locations.

Attempts to run single marker analysis for RD on only non-zero data points, which would detect QTLs that only effect the actual spread of rhizomes, did not return many results (Figure 2.9). Instead, results were reported for single marker analysis on both zero and non-zero data points, however the presence/absence of rhizomes is conflated with RD in this case (Figure 2.10). This analysis supported the identification of QTLs on chromosome 1 and 3 by interval mapping as well as a potential QTLs on chromosome 6, which had a negative allele effect (Table 2.19). QTL locations identified here closely mirrored those of RZ and RN QTLs and similarly suggested that both *S. bicolor* and *S. halepense* alleles contribute to rhizome presence and spread.

Winter Survival (WS)

Interval mapping detected two QTLs for winter survival in GAF_3 and one in KSF_3 (Table 2.13). QTL qWS.3A.F3.1 was accepted in the KS population at a lowered LOD threshold of 2.5 because of correspondence to the QTL detected in the GA environment as well as through SMA.

A binary model was used for the KS population due to the lower line survival rates. The single QTL detected in KS, qWS.3A.F3.1, had a 6.9% explanatory power and a positive additive allele effect of 0.54. Total variance explained by the QTLs detected in GA was 14%. QTL qWS.6B.F3.1 was the only QTL with a negative effect of -0.06, corresponding to findings for RZ, RN, and RD that *S. bicolor* alleles in this region contribute to increased rhizomatousness and capacity to overwinter, while other QTLs indicate that *S. halepense* contributes to increased winter survival.

Single marker analysis detected two QTLs in GA and two in KS (Figure 2.11). Only the QTLs identified on chromosome 3 corresponded to those detected by interval mapping, with the other two QTLs differing (Table 2.20). An additional QTL on chromosome 1 was detected in the GA environment, exhibiting a positive allele effect. A putative QTL on chromosome 5 was also detected in the KS environment, displaying a positive allele effect. None of the four QTLs detected had a negative allele effect, indicating that *S. halepense* alleles contribute to increased capacity to overwinter.

Comparison to prior *S. bicolor* perenniality studies

Two of the four RZ QTLs detected by interval mapping overlap with rhizome QTLs detected in prior *S. bicolor* x *S. propinquum* F_2 (PQ- F_2), *S. bicolor* x *S. propinquum* RIL (PQ-RIL) and *S. bicolor* x *S. halepense* BC_1F_2 (SBSH- BC_1F_2) populations (Paterson et al. 1995; Kong et al. 2015; Kong 2017). Similarly, all RZ QTLs detected by single marker analysis overlap with rhizome QTLs detected in these same prior populations. Two previously undetected QTLs, qRZ.7A.F2.1 and qRZ.7B.F3.1 were detected by interval mapping in the F_2 and GAF_3 population, all of which did not overlap with likelihood ranges determined in prior studies. In determining consensus between prior studies and findings here, all QTLs pertaining to rhizomatousness were considered, including: presence/absence of rhizomes, rhizome number, and rhizome distance.

Four of the five RN QTLs detected by interval mapping overlap with rhizome QTLs detected in previous PQ-F_2, PQ-RIL, and SBSH- BC_1F_2 populations (Paterson et al. 1995; Kong et al. 2015; Kong 2017). The one non-consensus QTL, qRN.4A.F2.1, was detected only in the F_2 population and was not found in single marker analysis.

Four of the five RD QTLs detected by interval mapping overlap with rhizome QTLs detected in prior PQ-F_2, PQ-RIL, and SBSH- BC_1F_2 populations (Paterson et al. 1995; Kong et al. 2015; Kong 2017). The one non-consensus QTL, qRD.7A.F2.1, aligns with the other novel RZ QTL qRZ.7A.F2.1 detected in this study.

While detection of winter survival QTLs was inhibited in the previous SBSH- BC_1F_2 study due to insufficient survival rates, line survival in the SBSH- $F_{2:3}$ population here permitted detection of a few WS QTLs (Kong 2017). All three QTLs detected by interval mapping overlapped with rhizome QTLs detected in prior PQ-RIL and SBSH- BC_1F_2 populations (Kong et al. 2015; Kong 2017). Three of the four QTLs detected by single marker analysis also corresponded to previously detected rhizome QTLs. Single marker analysis also detected one potential WS QTLs on chromosome 5, qWS5.F3, which only overlaps with one QTL relating to regrowth/ratooning in a previous PQ- F_2 population (Paterson et al. 1995). Single marker analysis does not allow for a precise detection of QTL likelihood interval, but for this QTL in question, SNPs exceeding a -log10(pval) threshold of 2.5 range from .43 Mb-18.10 Mb. This putative range for qWS5.F3.1 does overlap with another rhizomatous QTL detected in the prior PQ- F_2 population (Paterson et al. 1995). The one potential genomic region corresponding to winter survival detected by single marker analysis of the SBSH- BC_1F_2 (Kong 2017) was validated, corresponding to QTLs: qWS.3C.F3.1, qWS.3A.F3.1, qWS3.F3.1, and qWS3.F3.2.

Comparison of rhizomatousness/winter survival and flowering time

Prior investigations into an SBSH- BC_1F_2 population indicated a negative relationship between later flowering time and rhizome development, which warranted further investigation (Kong 2017). There was compelling evidence for a genetic basis for this negative relationship in the SBSH-$F_{2:3}$ population here. Interval mapping revealed four major QTLs for flowering time among these populations and 16 QTLs related to rhizomatousness (RZ, RN, RD) and winter survival that overlapped with the FL QTLs (Table 2.21). All 16 of these overlapping rhizome and winter survival QTLs had opposing allele effects to what was observed in the corresponding flowering QTL. QTLs qFL.6A.F3.2, qFL.6B.F2.1, and qFL.10A.F2.1 all displayed a positive allele effect for flowering time, while all RZ, RD, and RN QTLs in that region had a negative allele effect for rhizome traits. Similarly, the negative effect of QTL qFL.1D.F3.1 overlapped with seven RZ, RD, RN, and WS QTLs that all displayed a positive allele effect. This all suggests that later flowering is associated with reduced rhizome presence and capacity to overwinter.

Comparison of rhizomatousness/winter survival and plant height

SBSH progeny show transgressive segregation for plant height, with offspring height often exceeding the height of both parents (Kong 2017) (Table 2.5). Some regions prefer tall sorghums – in Africa, for example, sorghum stalks are important as a building material. There are also advantages to increased plant height in breeding improved cultivars for biofuel (Fernandez et al. 2009) – however, excessive height may also prove prohibitive to grain sorghum suitable for machine harvest, as harvesting equipment is tailored to the height of current *S.*

bicolor varieties. Investigations into tradeoffs between plant height and rhizome development yielded mixed results (Table 2.22). QTL qHT.6B.F3.1 displayed opposing allele substitution effects with its overlapping RZ, RN, RD, and WS QTLs, indicating that decreased plant height corresponds to increased rhizome presence and vice versa. However other major plant height QTLs did not overlap with any rhizome and winter survival QTLs. These findings indicate no clear relationship between plant height and rhizome development or capacity to overwinter.

Comparison of rhizomatousness/winter survival and inflorescence weight/grain yield

There has been an oft debated theory that plants must allocate their limited resources to either aboveground or belowground biomass development, thereby making a high-yielding perennial variety infeasible (Crews and DeHaan 2015; Smaje 2015). Without adequate yield a perennial variety will not be viable for farmers, and thus investigating this tradeoff is critical. Investigations into a genetic basis for tradeoffs between inflorescence weight and rhizomatousness returned mixed results (Table 2.23). QTL qYLD.1C.F3.1 shared a positive allele effect with its overlapping RZ and RN QTLs, indicating that increased inflorescence weight corresponds to greater winter survival. The remaining INFL/YLD QTLs had no overlapping rhizome or winter survival QTLS. Overall, INFL and YLD QTLs had notably fewer overlapping rhizome and overwintering QTLs as compared to FL and HT, providing little support for any significant relationship between the two traits.

Discussion

The final genetic map had 27 linkage groups, which is slightly less than the 36 and 38 linkage groups observed in two previously mapped SBSH-BC_1F_2 populations (Kong 2017). It is possible that the larger population size in the F_2 population allowed for greater statistical power in linkage group detection. The aberrant chromosomal behavior on chromosome 9 detected here was also observed in the BC_1F_2 population. Results from that study suggested that inaccuracies in detecting allele dosage might compromise the statistical power of linkage mapping in this population (Kim 2015; Kong 2017). Accordingly, single marker analysis proved to be a useful complement to interval mapping in the detection of QTLs, often providing further validation for regions detected by interval mapping while identifying potential additional QTLs.

Investigations into the relationship between flowering time and rhizome development and/or capacity to overwinter built on findings from a previous SBSH-BC_1F_2, which found four QTL regions associated with later flowering that overlapped with QTL regions associated with reduced numbers of rhizomes (Kong 2017). Phenotypic analysis in the SBSH-$F_{2:3}$ indicated a statistically significant negative correlation between later flowering and reduced rhizome number in all three populations, as well as a negative relationship between flowering time and winter survival in the GAF_3 population and flowering time and rhizomatousness in the KSF_3 population (Figure 2.1, 2.2, 2.3). Comparison of FL and RZ, RN, RD, and WS QTLs provide evidence for a genetic basis for this relationship (Table 2.21). The four FL QTLs detected by interval mapping overlapped with 16 RZ, RN, RD, and WS QTLs, all of which displayed opposing allele effects to those of the corresponding FL QTL. This indicates that later flowering is consistently associated with decreased rhizomatousness and capacity to overwinter, providing support for the intriguing hypothesis that rhizomes have a relative advantage as propagules rather than storage organs, resulting in their positive correlation with reproductive growth and negative relationship with vegetative growth (Kong 2017). It has also been hypothesized that the release of apical dominance by flowering may promote rhizome development, indicating that the ‘signals’ that control flowering have been appropriated to also control rhizome development (Kong 2017).

Phenotypic analysis indicated no relationship between plant height and rhizomatousness or capacity to overwinter (Figure 2.1, 2.2, 2.3). Further investigations into a genetic relationship between these two traits provided mixed results, with one HT QTL overlapping with QTLs corresponding to decreased rhizomatousness and overwintering, and other HT QTLs corresponding to no rhizome or winter survival QTLs (Table 2.22). These results suggest the capacity to develop both short and tall perennial sorghum varieties.

The absence of or slight positive correlation between below-ground rhizome prevalence and above-ground seed production in both environments (Figure 2.1, 2.2, 2.3) contributes to the abatement of concerns regarding a theoretical evolutionary tradeoff between seed production and vegetative propagation (Nabukalu and Cox 2016). This is supported by the mixed results from investigations into a genetic basis for such a relationship (Table 2.23). While one YLD QTL did overlap with rhizome QTLs with the same allele effect, other INFL/YLD QTLs had no overlapping RZ, RN, RD, or WS QTLs. Additionally, only 3 of the RZ, RN, RS, and WS QTLs detected in this study overlapped with INFL QTL regions, suggesting varying genetic bases for these traits. Ultimately, both rhizomatousness and yield prove to be low heritability traits with high degrees of environmental effect, further suggesting that improved cultivars for both perenniality and yield are possible given favorable conditions (Table 2.6). Future investigations should aim to parse apart how genotype, environment, and genotype x environment intersect with traits pertaining to perenniality and yield to develop suitable cultivars for varying conditions, as has been done in perennial rice (Zhang et al. 2017).

Two previously undetected QTLs, qRZ.7A.F2.1, and qRZ.7B.F3.1 were found by interval mapping. Additional previously undetected RD and RN QTLs, qRN.4A.F2.1 and qRD.7A.F2.1, corresponded to the locations of the novel RZ QTLs. Three of these four QTLs were detected solely in the F_2 population. Power for detection of rhizome presence is reduced in the single plant F_2 population and resulting QTLs that are not corroborated in other populations warrant further validation.

The significantly different climates of GA and KS suggest variable pressure for winter survival among the two environments. This was made evident in the comparative survival rates, with 92.9% of lines exhibiting some level of winter survival in GA as compared to 28.8% in KS (Table 2.4). This study is the first SBSH derived population with sufficient winter survival to elucidate QTLs (Kong 2017). The single KS QTL detected by interval mapping corresponded to the QTL with the highest explanatory power detected in the GA environment (Table 2.13). However, single marker analysis discovered a putative novel QTL on chromosome 5 that was unique to the KS environment (Table 2.20). The more severe winter climate in KS may have elucidated a genomic region pertinent to winter survival that is disparate from rhizome development. However, this warrants future investigation and validation because the lower winter survival rates in KS also means a much smaller sample size of the surviving group and decreased confidence in results.

In addition to contributions of the findings here to perennial sorghum development, they also suggest the potential for *S. halepense* to improve conventional sorghum varieties. QTL mapping results for this SBSH-$F_{2:3}$ revealed that *S. halepense* harbors alleles that can contribute to earlier flowering, increased inflorescence/grain weight, and shorter or taller plant height. The canonical model for plant height in sorghum suggests control by four general loci, *DW1-4* (Quinby and Karper 1945). Findings here detected *DW2* in all populations (Quinby and Karper 1945). However, many additional QTLs for plant height were detected outside of the *DW* regions, potentially indicating divergent control of plant height in *S. halepense* and *S. bicolor* and/or providing further support for a more recent model of plant height being controlled by many genes with small effect sizes (Zhang et al. 2015). Six genes, *Ma1-6*, are thought to control flowering time in sorghum (Quinby and Karper 1945a; Quinby 1966; Brady 2006). Findings here suggest two QTLs on chromosome 6 which likely correspond to *Ma1* and *Ma6* (Brady 2006; Murphy et al. 2011; Cuevas et al. 2016), as well as additional QTLs where *S. halepense* alleles can accelerate flowering: qFL.1D.F3.1 and qFL10B.F3.1.

Investigations here contribute significantly to the validation of previously identified QTLs relating to rhizomatousness in the *Sorghum* genus and is the first to identify QTLs relating to winter survival in a SBSH derived population (Paterson et al. 1995; Kong et al. 2015; Kong 2017). Although most QTLs identified here appear to have some consensus with genomic regions identified in prior *S. propinquum* derived populations, there is perhaps some evidence for a heightened presence of rhizome SBSH QTLs identified from the 40-60 Mb region of chromosome 6 which warrants further investigation. Identification of novel QTLs and the narrowing of QTL regions can contribute to future perennial germplasm development. As previously touched on, these genomic regions may not be confined in their applicability to the Sorghum genus, but extrapolatable to other species such as rice or maize (Hu et al. 2013; Westerbergh and Doebley 2004).

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Table 2.1. Summary of the genetic map from the $F_{2:3}$ population of *S. bicolor* x *S. halepense*.

LG	# Markers	Length (cM)	Avg spacing (cM)	Max spacing (cM)
1A	47	112.2	2.44	6.57
1B	17	45.3	2.83	7.26
1C	25	60.0	2.50	7.18
1D	22	70.5	3.36	7.78
1E	18	46.7	2.75	10.81
2A	56	106.3	1.93	6.04
2B	6	37.8	7.55	11.98
2C	61	161.3	2.69	10.46
3A	37	144.6	4.02	9.41
3B	21	109.7	5.49	15.89
3C	3	64.1	17.42	25.64
4A	79	158.8	2.04	8.87
4B	31	144.5	4.82	13.83
5	47	117.3	2.55	9.80
6A	39	87.0	2.29	6.55
6B	52	109.0	2.14	7.47
6C	9	19.1	2.38	3.82
7A	52	135.0	2.65	17.43
7B	33	137.4	4.30	14.77
8A	22	146.3	6.97	18.54
8B	24	140.1	6.09	30.46
9A	10	36.5	4.05	5.97
9B	5	19.9	5.43	7.95
9C	7	62.2	5.84	8.30
9D	14	68.8	5.29	13.95
10A	48	136.0	2.89	18.04
10B	31	78.7	2.62	7.85

LG: Linkage Group

Table 2.2: Genotype frequencies of an F2 population from a *S. bicolor* x *S. halepense* derived cross, in which parental F_1 marker genotype was simplex AAAB.

Chrom	% AAAA	% AAAB	% AABB	% ABBB	% BBBB	% missing
1	0.25	0.33	0.21	0.06	0.01	0.14
2	0.26	0.33	0.20	0.06	0.01	0.13
3	0.36	0.27	0.15	0.06	0.02	0.14
4	0.26	0.32	0.21	0.06	0.02	0.13
5	0.29	0.32	0.18	0.05	0.01	0.15
6	0.24	0.32	0.20	0.08	0.02	0.15
7	0.25	0.31	0.21	0.06	0.02	0.15
8	0.27	0.30	0.19	0.07	0.02	0.15
9	0.16	0.28	0.26	0.14	0.01	0.14
10	0.22	0.30	0.22	0.09	0.01	0.16
Total	0.26	0.31	0.20	0.07	0.02	0.14

Table 2.3: Rhizomatousness, rhizome count, and rhizome distance among F_2 and F_3 populations in the *S. bicolor* x *S. halepense* derived cross.

Trait	Env	Mean	Median	SD	Min	Max	N	BTx mean	G9E mean
RZ	GAF2	0.38	0.00	.48	0.0	1	235	0	1.00
RZ	GAF3	0.46	0.33	.35	0.0	1	222	0	.92
RZ	KSF3	0.32	0.25	.33	0.0	1	165	0	1.00
RN	GAF2	0.75	0.00	1.30	0.0	6	235	0	7.00
RN	GAF3	0.74	0.67	0.74	0.0	4	222	0	1.50
RN	KSF3	0.73	0.40	1.11	0.0	6	165	0	38.00
RD	GAF2	11.97	11.75	4.05	5.7	25	83	-	28.29
RD	GAF3	13.32	12.25	5.55	6.0	42	163	-	13.96

Table 2.4: Winter survival among GA and KS $F_{2:3}$ populations in the *S. bicolor* x *S. halepense* derived cross (0-1 scale).

Env	Mean	Median	SD	N (total)	N (survived)	BTx mean	G9E mean
GAF3	0.61	0.67	0.29	228	212	0	.96
KSF3	0.08	0.00	0.17	170	49	0	1.00

Table 2.5: Flowering date, plant height (cm), and dried inflorescence weight (g) (GA) or grain yield (KS) among F_2 and F_3 populations in the *S. bicolor* x *S. halepense* derived cross.

Trait	Env	Mean	Median	SD	Min	Max	N	BTx mean	G9E mean
FL	GAF2	57.2	55.0	13.7	28	118	244	58.7	44.0
FL	GAF3	61.3	59.0	12.8	41	99	229	65.4	59.3
FL	KSF3	80.3	82.6	14.4	45	107	170	89.0	65.5
HT	GAF2	162.0	158.5	41.2	41	270	220	98.7	157.0
HT	GAF3	166.6	165.7	30.4	86	247	229	101.6	194.0
HT	KSF3	239.1	240.0	38.1	140	330	170	180.0	215.0
INFL	GAF2	75.4	55.0	69.4	0	357	242	-	-
INFL	GAF3	66.5	61.7	32.9	4	175	229	121.9	54.5
YLD	KSF3	43.5	36.1	34.0	0	242	168	187.0	19.0

Table 2.6: Narrow sense heritability estimates derived from parent-offspring regression on $F_{2:3}$ populations in a *S. bicolor* x *S. halepense* derived cross.

	GA		KS		Pooled	
Trait	h^2	p-val	h^2	p-val	h^2	p-val
flowering	62.3%	9.41E-21	51.1%	2.03E-09	57.5%	1.27E-27
inflor	16.2%	1.85E-05	11.2%	0.051	14.1%	1.54E-05
height	50.5%	5.03E-10	38.3%	1.85E-06	46.3%	2.04E-14
rhiz count	13.2%	4.60E-03	10.1%	0.113	11.8%	1.96E-03
rhiz dist	7.8%	0.352	-	-	-	-
shattering	38.2%	2.60E-13	-	-	-	-

Table 2.7 QTL Interval mapping results for flowering time (FL) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qFL.6A.F2.1	GA	6A	17.5	38.8-45.4	8.9	14.79	0.45	3.6	46.0
qFL.6B.F2.1	GA	6B	7.5	37.9-39.4	10.9	18.65	0.53	5.7	45.2
qFL.10A.F2.1	GA	10A	90.0	42.4-55.1	3.1	2.56	0.18	1.9	58.2
qFL.1D.F3.1	GA	1D	19.9	63.6	4.1	5.80	-0.27	60.3	74.2
qFL.6A.F3.1	GA	6A	2.5	.2-2.4	6.2	9.19	0.34	2.4	45.5
qFL.6B.F3.1	GA	6B	0.0	4.5	10.0	15.03	0.50	4.5	46.0
qFL.6A.F3.2	KS*	6A	2.5	.24-2.4	8.7	15.68	7.03	2.4	45.5
qFL.6B.F3.2	KS	6B	14.1	45.3	6.6	9.51	6.11	4.5	46.1

*KS flowering data approximated normal distribution and was not transformed with square root function, unlike both GA populations, resulting in allele effect discrepancy

Table 2.8 QTL Interval mapping results for inflorescence dry weight (INFL) (GA) and grain yield (YLD) (KS**)** in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qINFL.1A.F2.1	GA	1A	63.5	13.1	3.88	7.20	-0.88	6.3	13.8
qYLD.1A.F3.2	KS	1A	44.2	7.5	2.5	2.96	-0.49	3.8	10.2
qYLD.1C.F3.1	KS	1C	43.7	7.2	3.1	4.65	0.74	1.2	7.1
qYLD.4B.F3.1	KS	4B	21.9	1.4	2.5	4.13	0.69	0.1	62.4

Table 2.9 QTL Interval mapping results for plant height (HT) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qHT.1A.F3.1	GA	1A	55	10.2-10.4	3.6	6.29	-9.39	5.1	17.2
qHT.6B.F3.1	KS	6B	2.5	5.7-37.9	4.0	11.38	18.29	4.5	46.0
qHT.9C.F3.1	KS	9C	20	1.8-2.2	3.4	8.64	1.67	1.8	3.3

Table 2.10 QTL Interval mapping results for rhizomatousness (RZ) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qRZ.1C.F2.1	GA*	1C	59.9	7.1	3.0	4.31	0.26	6.5	7.1
qRZ.3A.F2.1	GA	3A	40.3	53.2	3.4	4.83	0.68	38.2	59.3
qRZ.7A.F2.1	GA	7A	110	63.5-64	3.0	3.66	-0.31	62.8	64.2
qRZ.7B.F3.1	GA	7B	10.7	2.0	3.6	5.46	0.08	1.4	2.0

*A binary model was used in the F_2 population, contributing to the different scales of effect in the F_2 and F_3 populations

Table 2.11 QTL Interval mapping results for number of rhizomes (RN) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qRN.1C.F2.1	GA	1C	59.9	7.1	3.3	7.08	0.11	7.1	8.0
qRN.3A.F2.1	GA	3A	72.5	61.3-62.9	3.2	5.91	0.12	12.3	66.2
qRN.4A.F2.1	GA	4A	152.2	68.4	3.0	5.78	0.13	67.5	68.4
qRN.1D.F3.1	GA	1D	48.9	73.0	3.4	5.42	0.11	61.8	77.0
qRN.7B.F3.1	GA	7B	10	1.4-2	3.2	4.15	0.09	0.9	58.2

Table 2.12 QTL Interval mapping results for rhizome distance (RD) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	LOD normal	LOD binary	% variance norm	% variance binary	Effect normal	Effect binary	Left SNP (Mb)	Right SNP (Mb)
qRD.3A.F2.1	GA	3A	62.5	56.8-59.3	3.6	2.5	1.8	4.18	2.86	1.64	0.49	54.8	62.9
qRD.7A.F2.1	GA	7A	119.1	64.1	3.7	1.2	2.7	1.98	4.33	-0.68	-0.32	62.8	65.1
qRD.1D.F3.1	GA	1D	60.5	75.7	4.8	4.1	2.7	6.77	4.61	2.84	0.74	67.2	77.0
qRD.7B.F3.1	GA	7B	12.5	1.4-2	6.1	2.0	3.4	8.18	3.35	1.24	0.67	0.9	6.9
qRD.10A.F3.1	GA	10A	33.6	1.9	4.0	1.7	1.3	2.75	1.32	-1.74	-0.47	1.2	22.7

Table 2.13 QTL Interval mapping results for winter survival (WS) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

QTL	Env	Chr	Position (cM)	Position (Mb)	LOD	% variance explained	Additive effect	Left SNP (Mb)	Right SNP (Mb)
qWS.3C.F3.1	GA	3C	41.1	67.4	4.52	8.01	0.12	59.9	65.1
qWS.6B.F3.1	GA	6B	52.3	55.6	3.04	5.99	-0.06	5.3	56.1
qWS.3A.F3.1	KS*	3A	47.7	55.9	2.56	6.91	0.54	52.3	56.8

*A binary model was used in the KS population, contributing to the different scales of effect size

Table 2.14: Flowering time (FL) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qFL1.F2.1	GA	S1_15724632	1	15.72	3.8	-6.65
qFL1.F2.2	GA	S1_54283292	1	54.28	4.7	6.21
qFL6.F2.1	GA	S6_509950	6	0.51	10.3	8.71
qFL6.F2.2	GA	S6_43321755	6	43.32	17.3	11.54
qFL10.F2.1	GA	S10_12333400	10	12.33	4.8	5.74
qFL10.F2.2	GA	S10_42433087	10	42.43	5.6	5.45
qFL1.F3.1	GA	S1_64766571	1	64.77	5.7	-5.87
qFL6.F3.1	GA	S6_5574112	6	5.57	9.7	8.19
qFL6.F3.2	GA	S6_45034834	6	45.03	11.5	9.94
qFL10.F3.1	GA	S10_10761993	10	10.76	3.5	4.61
qFL6.F3.3	KS	S6_340782	6	0.34	11.7	11.11
qFL6.F3.4	KS	S6_43187131	6	43.19	14.2	12.40
qFL10.F3.2	KS	S10_52395866	10	52.40	3.8	7.33

Table 2.15: Inflorescence dry weight (INFL) (GA) and grain yield (YLD) (KS) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qINFL1.F2.1	GA	S1_10203393	1	10.20	3.8	-1.34
qINFL1.F2.2	GA	S1_78430617	1	78.43	3.6	-1.58
qINFL1.F3.1	GA	S1_18047346	1	18.05	3.6	-0.72
qINFL6.F3.1	GA	S6_53942723	6	53.94	3.6	1.57
qYLD1.F3.1	KS	S1_5706141	1	5.71	4.6	1.22
qYLD2.F3.1	KS	S2_2606039	2	2.61	3.5	-0.98
qYLD6.F3.1	KS	S6_51416652	6	51.42	4.7	-1.05

Table 2.16: Plant height (HT) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qHT1.F3.1	GA	S1_12682195	1	12.68	4.4	-29.06
qHT1.F3.2	GA	S1_54113822	1	54.11	3.6	-17.17
qHT6.F3.1	GA	S6_47197650	6	47.20	4.0	16.85
qHT1.F3.3	KS	S1_5873345	1	5.87	3.4	-19.31
qHT3.F3.1	KS	S3_52149227	3	52.15	3.5	17.61
qHT5.F3.1	KS	S5_4111066	5	4.11	4.2	24.06
qHT6.F3.2	KS	S6_340782	6	0.34	5.0	20.20
qHT6.F3.3	KS	S6_43496639	6	43.50	5.8	22.94

Table 2.17: Rhizomatousness (RZ) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qRZ1.F2.1	GA*	S1_18659013	1	18.66	4.6	0.22
qRZ1.F2.2	GA	S1_64515694	1	64.52	4.0	0.17
qRZ3.F2.1	GA	S3_55948105	3	55.95	4.3	0.18
qRZ6.F2.1	GA	S6_42721568	6	42.72	3.6	-0.23
qRZ6.F3.1	GA	S6_3914995	6	3.91	3.5	-0.09
qRZ7.F3.1	GA	S7_1962888	7	1.96	5.5	0.12
qRZ6.F3.2	KS	S6_509950	6	0.51	3.6	-0.10
qRZ6.F3.3	KS	S6_42708531	6	42.71	3.5	-0.11

*GAF$_2$ population analysis was performed with logistical regression rather than linear, accounting for some of the varying effect differences between the F$_2$ and F$_3$ populations

Table 2.18: Rhizome number (RN) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qRN1.F2.1	GA	S1_18659013	1	18.66	4.2	0.17
qRN1.F2.2	GA	S1_64080786	1	64.08	3.6	0.16
qRN3.F2.1	GA	S3_55917930	3	55.92	3.5	0.13
qRN1.F3.1	GA	S1_74588048	1	74.59	4.6	0.14
qRN6.F3.1	GA	S6_9441786	6	9.44	4.2	-0.14
qRN7.F3.1	GA	S7_5884461	7	5.88	3.5	0.11
qRN8.F3.1	GA	S8_61652234	8	61.65	3.6	-0.16
qRN1.F3.2	KS	S1_1205077	1	1.21	3.7	0.22
qRN6.F3.2	KS	S6_810226	6	0.81	4.2	-0.17
qRN6.F3.3	KS	S6_46007590	6	46.01	3.6	-0.20

Table 2.19: Rhizome distance (RD) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qRD1.F2.1	GA	S1_21838827	1	21.84	5.2	3.21
qRD1.F2.2	GA	S1_64080786	1	64.08	4.7	2.66
qRD3.F2.1	GA	S3_59302319	3	59.30	4.8	3.00
qRD6.F2.1	GA	S6_42721568	6	42.72	3.7	-2.76
qRD1.F3.1	GA	S1_75731211	1	75.73	5.7	3.22

Table 2.20: Winter survival (WS) QTLs identified in single marker analysis.

QTL	Env	Peak SNP	Chr	Position	-log10(pval)	Effect
qWS1.F3.1	GA	S1_71172308	1	71.17	4.3	0.13
qWS3.F3.1	GA	S3_68375380	3	68.38	3.8	0.10
qWS3.F3.2	KS	S3_62858499	3	62.86	4.0	0.18
qWS5.F3.1	KS	S5_15100336	5	15.10	3.3	0.13

Table 2.21: Comparison of rhizome and winter survival QTLs with flowering time QTLs in a SBSH- $F_{2:3}$ population.

FL QTL	RZ QTL	RN QTL	RD QTL	WS QTL
qFL.1D.F3.1 (-)	qRZ1.F2.2 (+)	qRN.1D.F3.1 (+) qRN1.F2.2 (+) qRN1.F3.1 (+)	qRD.1D.F3.1 (+) qRD1.F2.2 (+)	qWS1.F3.1(+)
qFL.6A.F3.2 (+)	qRZ6.F2.1 (-) qRZ6.F3.1 (-)	qRN6.F3.1 (-)	qRD6.F2.1 (-)	
qFL.6B.F2.1 (+) qFL.10A.F2.1 (+)	qRZ6.F3.3 (-)	qRN6.F3.3 (-)	qRD6.F2.1 (-) qRD.10A.F3.1 (-)	qWS.6B.F3.1 (-)

(-)(+) indicates negative or positive allele effect for the trait in question

Table 2.22: Comparison of rhizome and winter survival QTLs with plant height QTLs in a SBSH-$F_{2:3}$ population.

HT QTL	RZ QTL	RN QTL	RD QTL	WS QTL
qHT.1A.F3.1 (-)	-	-	-	-
qHT.6B.F3.1 (+)	qRZ6.F2.1(-) qRZ6.F3.1(-) qRZ6.F3.2(-)	qRN6.F3.1(-) qRN6.F3.3(-)	qRD6.F2.1(-)	qWS.6B.F3.1(-)
qHT.9C.F3.1 (+)	-	-	-	-

(-)(+) indicates negative or positive allele effect for the trait in question

Table 2.23: Comparison of rhizome and winter survival QTLs with inflorescence weight/grain yield QTLs in a SBSH-$F_{2:3}$ population.

INFL QTL	RZ QTL	RN QTL	RD QTL	WS QTL
qINFL.1A.F2.1(-)				
qYLD.1C.F3.1(+)	qRZ.1C.F2.1(+)	qRN.1C.F2.1(+) qRN1.F3.2(+)		
qYLD.4B.F3.2(+)				

(-)(+) indicates negative or positive allele effect for the trait in question

fl				***	
-0.07	infl	***			*
0.21	0.39	height			
0.13	-0.14	-0.14	shttr		
-0.19	-0.02	0.19	0.01	rhizno	
-0.15	0.28	0.03	0.08	-0.05	rhizdist

Figure 2.1 Correlation coefficients among days to flowering (FL), dry weight inflorescence (INFL), plant height (HT), seed shattering (SHTTR), number of rhizomes (RN), and rhizome distance (RD) in the F_2 population derived from a *S. bicolor* x *S. halepense* derived cross.

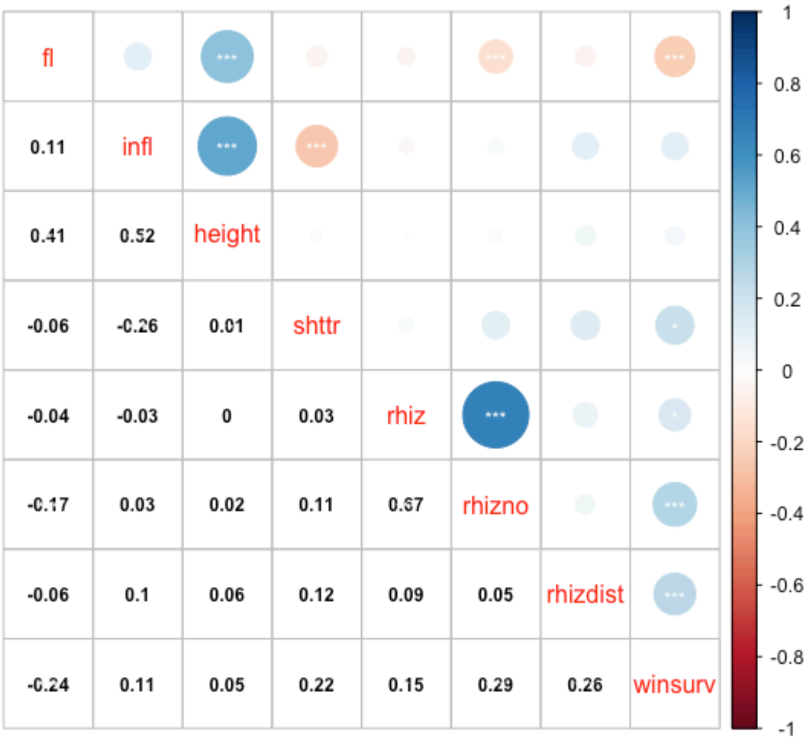

Figure 2.2 Correlation coefficients among days to flowering (FL), dry weight inflorescence (INFL), plant height (HT), seed shattering (SHTTR), rhizomatousness (RZ), number of rhizomes (RN), rhizome distance (RD), and winter survival rates (WS) in the F_3 GA population derived from a *S. bicolor* x *S. halepense* derived cross.

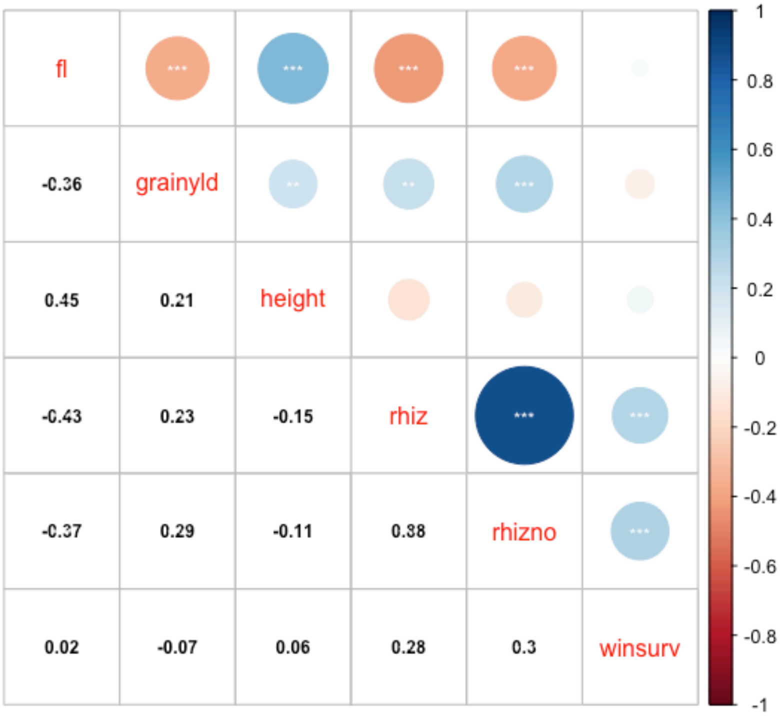

Figure 2.3 Correlation coefficients among days to flowering (FL), grain yield (YLD), plant height (HT), rhizomatousness (RZ), number of rhizomes (RN), and winter survival rates (WS) in the F_3 KS population derived from a *S. bicolor* x *S. halepense* derived cross.

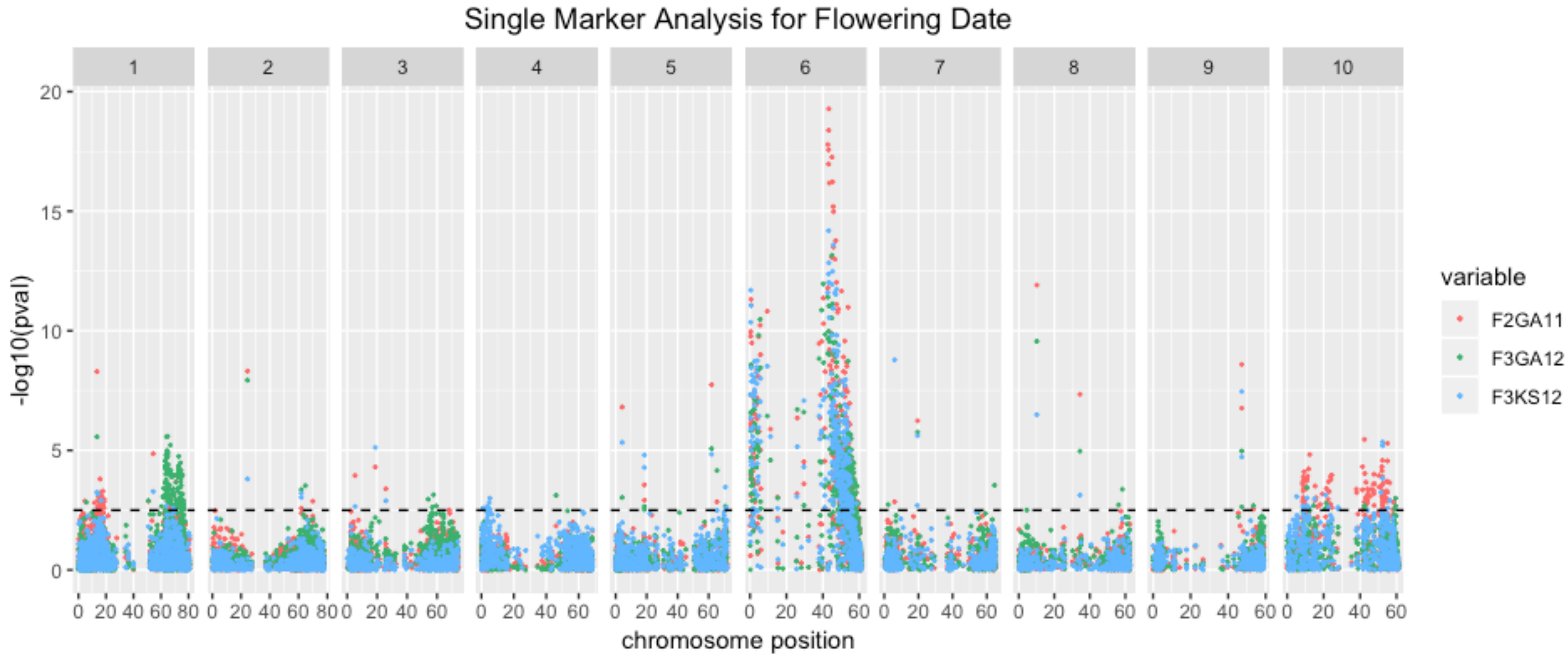

Figure 2.4 Single marker analysis of days to flowering (FL) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

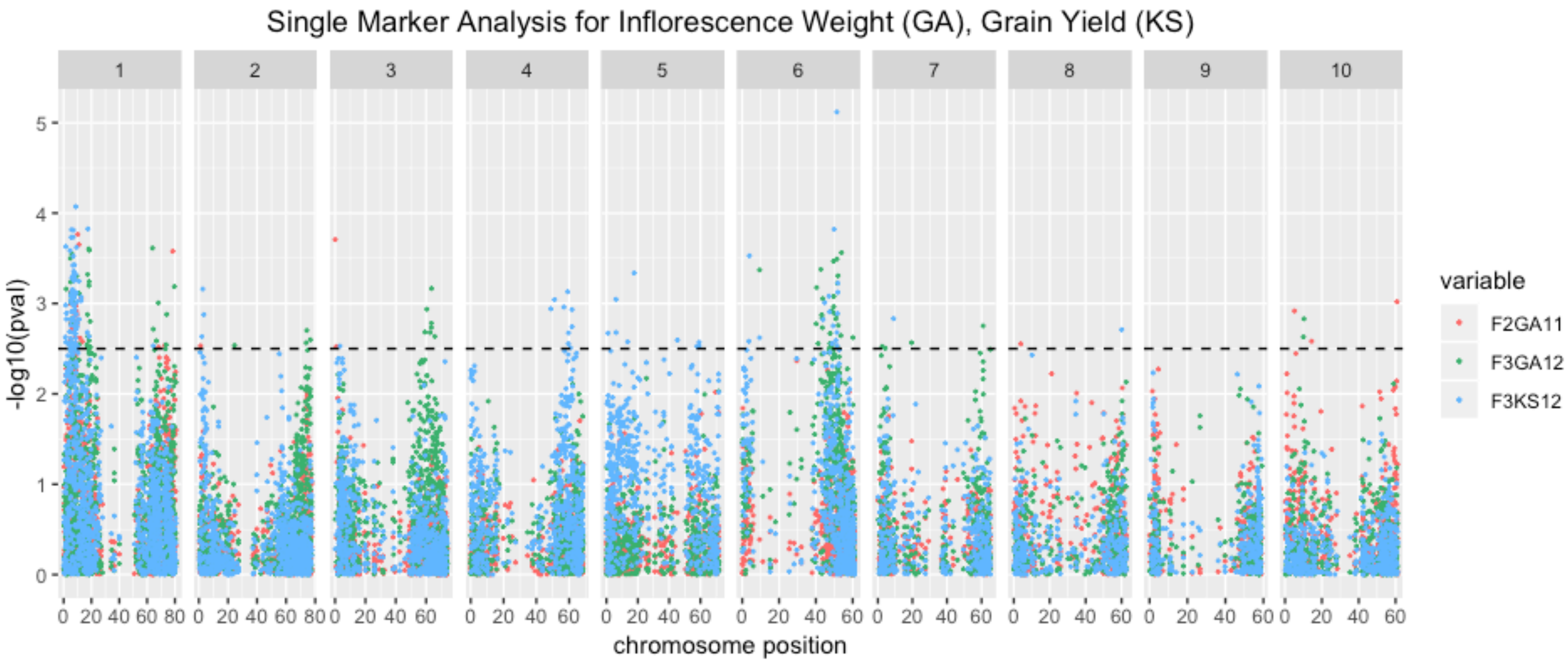

Figure 2.5 Single marker analysis of inflorescence dry weight (INFL) (GA) and grain yield (YLD) (KS) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

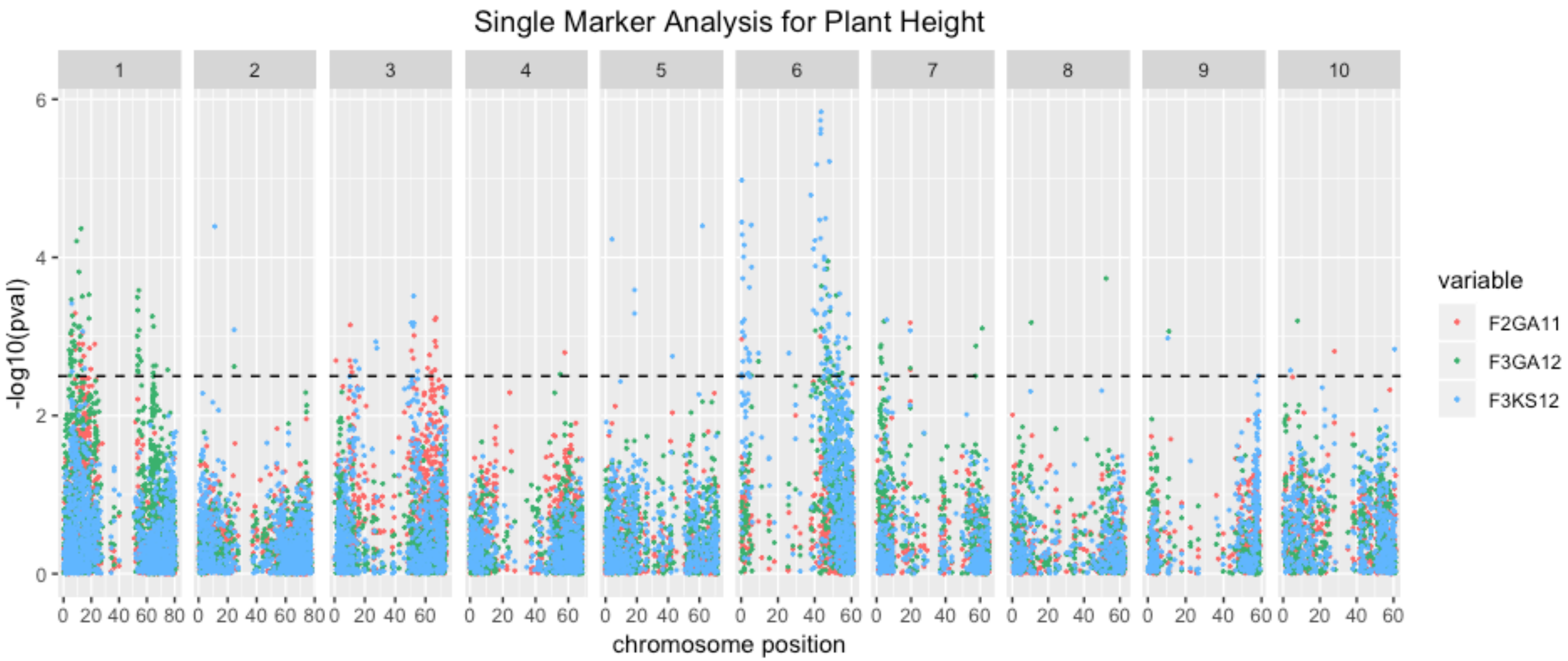

Figure 2.6 Single marker analysis of plant height (HT) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

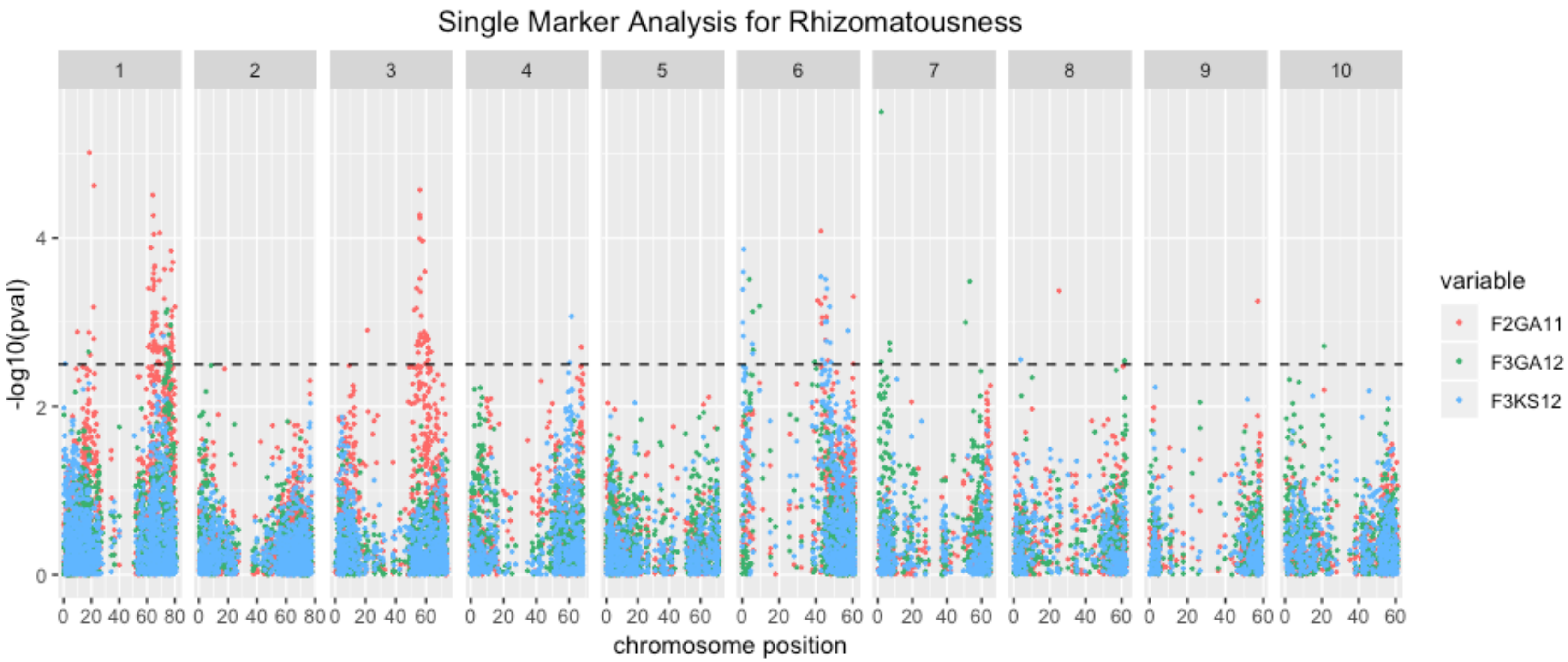

Figure 2.7 Single marker analysis of rhizomatousness (RZ) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

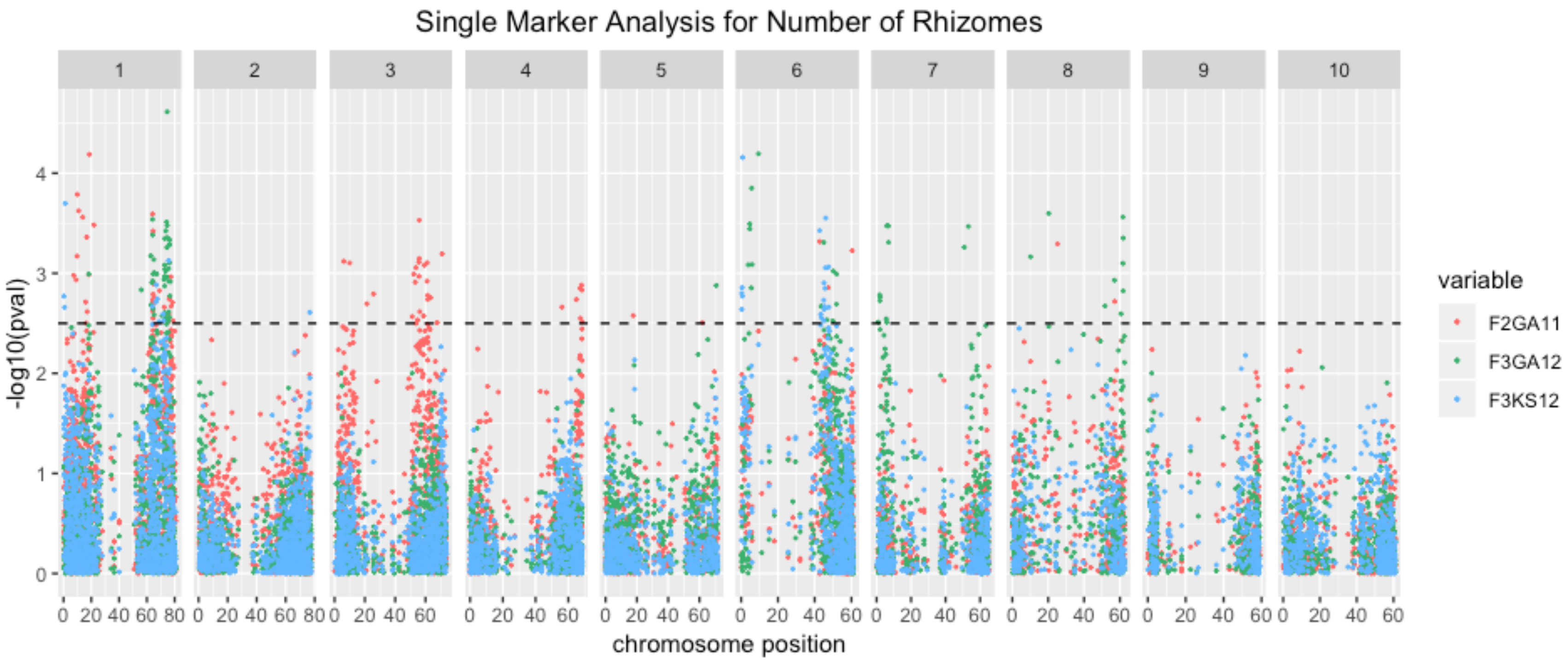

Figure 2.8 Single marker analysis of number of rhizomes (RN) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

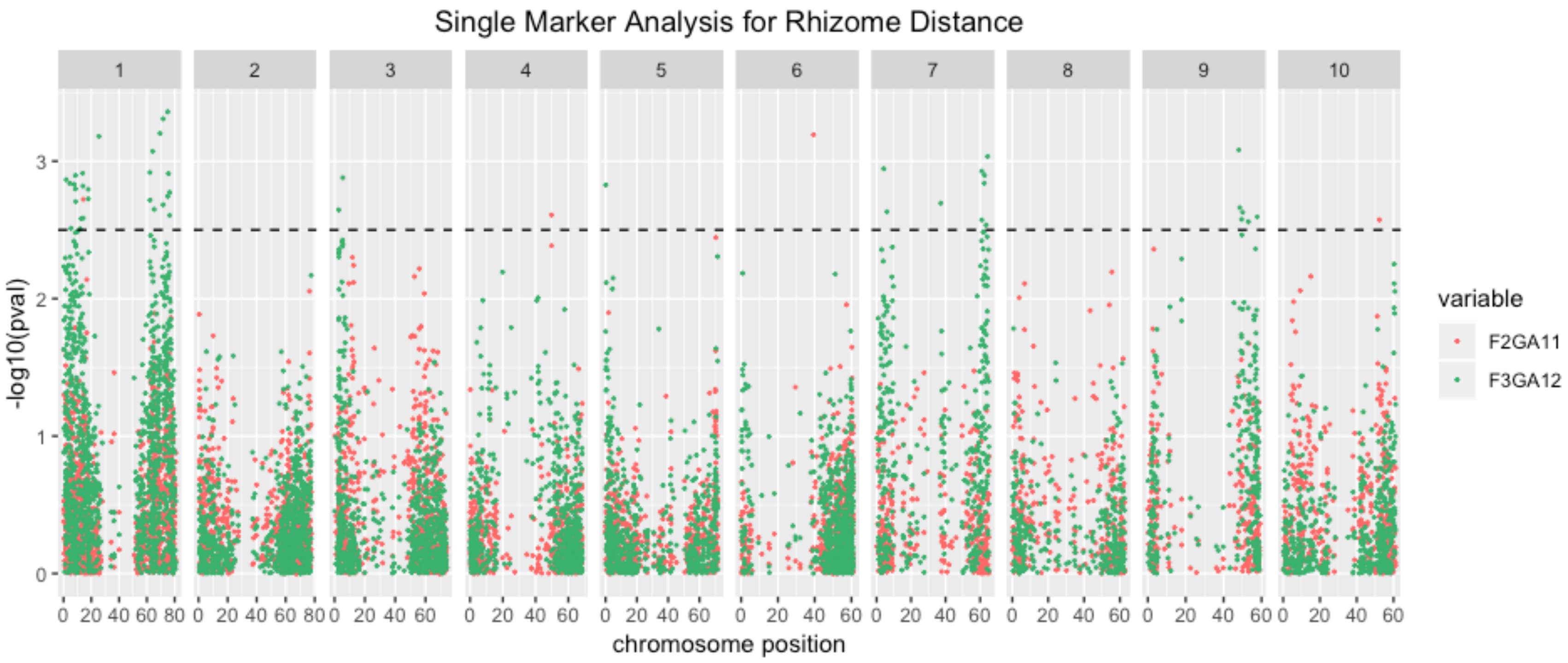

Figure 2.9 Single marker analysis of rhizome distance (RD) in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

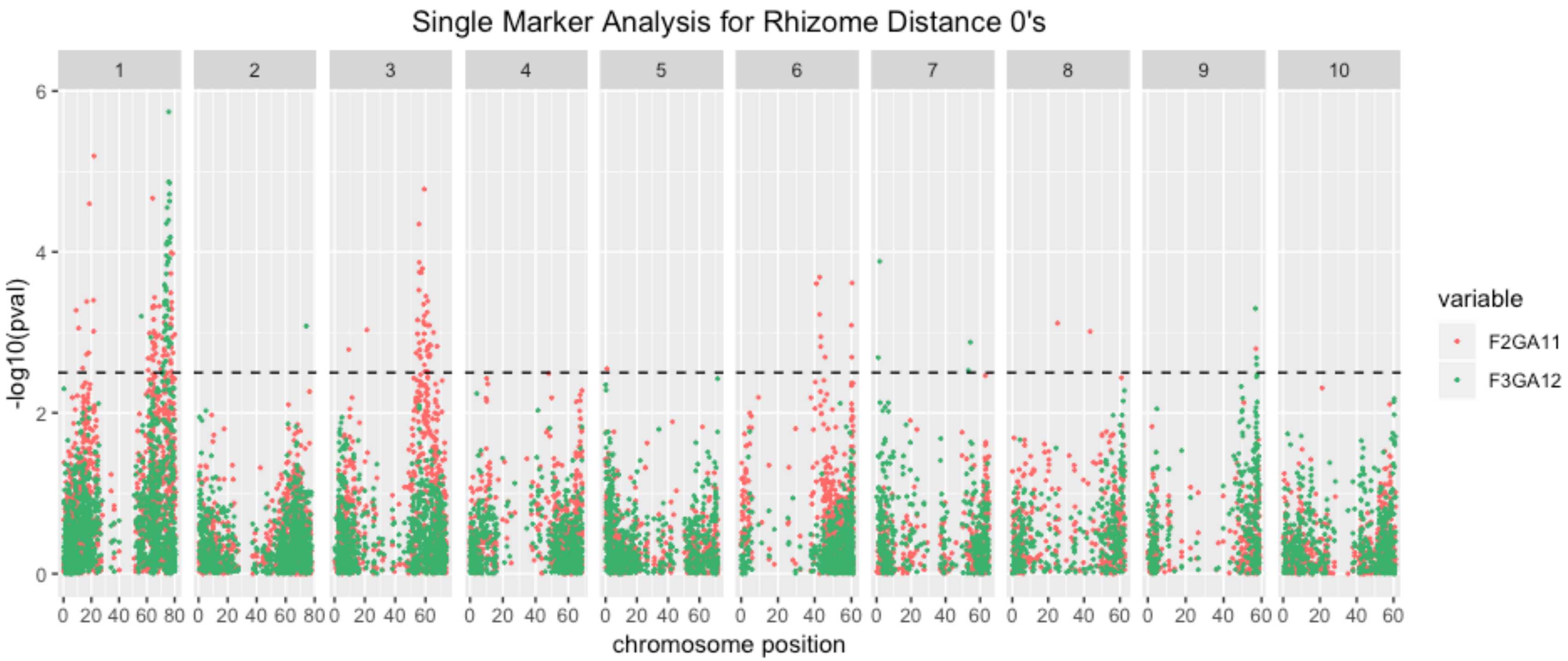

Figure 2.10 Single marker analysis of rhizome distance (RD) including zero data points in the F_2 and F_3 populations derived from a *S. bicolor* x *S. halepense* derived cross.

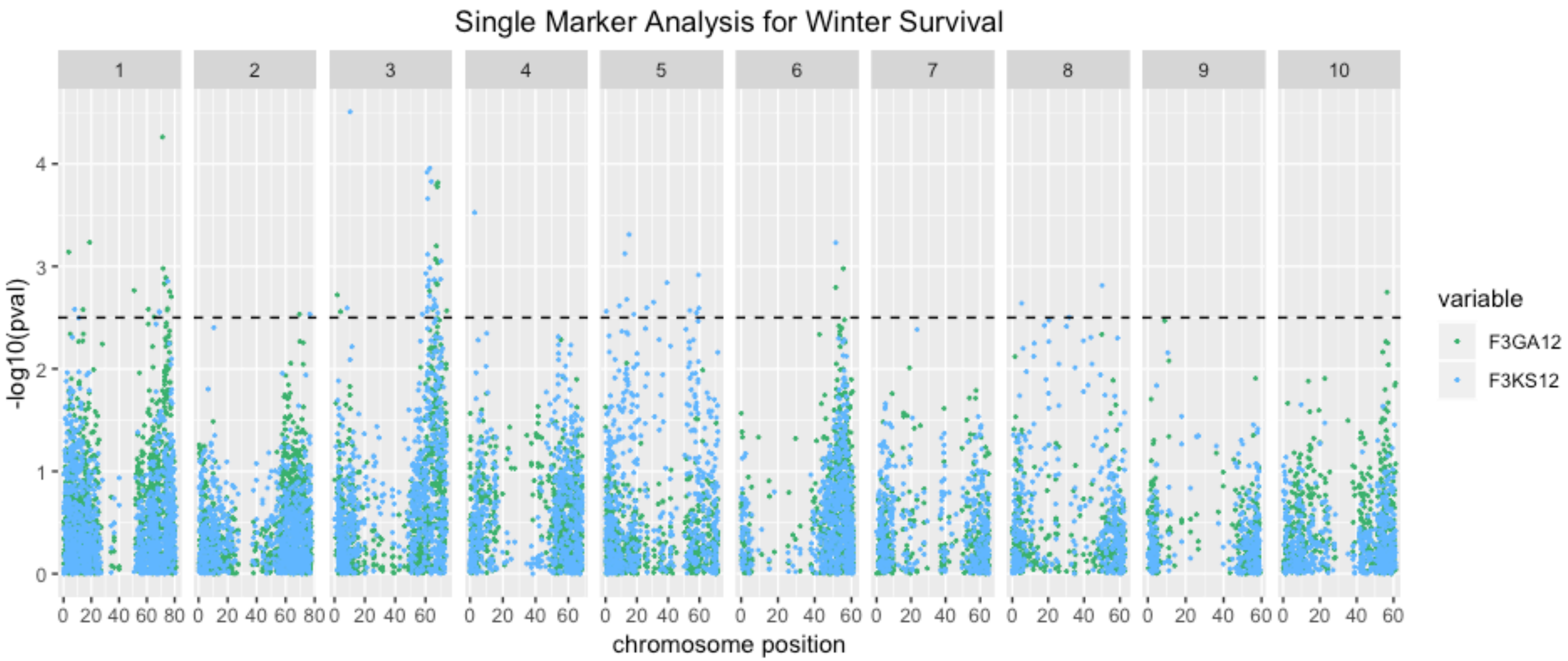

Figure 2.11 Single marker analysis of winter survival (WS) in the $F_{2:3}$ population derived from a *S. bicolor* x *S. halepense* derived cross.

CHAPTER 3

ECONOMIC ANALYSIS OF PERENNIAL GRAIN SORGHUM SYSTEM VIABILITY IN NORTH GEORGIA[2]

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[2] Conway, TM, Mullen, JD, and Paterson, AH. To be submitted to Agriculture, Ecosystems, and Environment.

Abstract

The burgeoning field of perennial grain development has had scant research into the economic viability of these novel growing systems, a component critical to their adoption potential. Perennial grain viability will depend on the evaluation of economic returns in relation to ecosystem services, which can vary across physical and political landscapes, requiring regional analyses to inform appropriate variety development. Investigations into break-even yields of three perennial/ratooning sorghum systems in north Georgia revealed the comparative advantage of the multi-use “Grains for Grass” and “Grains for Conservation” systems over grain-only. The supplementary income streams from either NRCS EQIP payments or silage production increase the resilience of such systems, allowing them to remain profitable under lower yields and/or diminishing yields in subsequent years of the plant stand. Findings suggest a perennial sorghum could be viable at yields as low as 62% of current annual yields and that all three perennial systems can operate on par with current annual varieties at substantially lower yields. Ratooning annual sorghum does not appear viable without NRCS EQIP payments at current ratooning yields, but improved ratooning varieties could make both a grain-only and “Grains for Grass” scenario viable for ratooning. Results indicate little economic benefit for a perennial stand exceeding three years. Findings support a segmented mode of adoption for perennial sorghum in the region, starting with a “Grains for Conservation” scenario on marginal land and culminating in a grain-only scenario when higher yields are achieved.

Introduction

There has been growing interest in perennial grain varieties as an alternative to annual ones due to their potential for ecologically resilient food production. Establishment of perennial grain systems requires the development of both “hardware” (perennial grain varieties) and “software” (the agronomic and ecological systems facilitated by the perennial germplasm) (Cox et al. 2013). The plant-breeding barrier is not the only one to contend with in perennial grain system establishment, as these novel varieties are considered “transformative technology” and motivations for adoption of such agroecological technologies are poorly understood (Adebiyi et al. 2015). Thus, while germplasm development has been the primary focus to-date, certain perennial grain breeding efforts in rice and intermediate wheatgrass (*Thinopyrum intermedium*, trademark name Kernza) are now reaching the point of commercialization, highlighting the need for concurrent “software” development (Huang et al. 2018; DeHaan et al. 2018). One particularly salient question is the economic viability of perennial varieties, which is sure to prove critical to their adoption.

A primary concern in perennial grain profitability is the prospect of reduced yields, which may eventually be counteracted through time-intensive breeding efforts (Crews et al. 2018). In light of this current reality, one common argument for ensuring the economic viability of perennial grains is dual usage of these crops (Ryan et al. 2018). While lower input costs can offset some decrease in yield, additional revenue streams through forage, biofuel, or environmental conservation payments can further enhance viability of such systems. Prior economic analyses of perennial wheat systems in Australia indicate that dual-use grain and forage systems are the most viable (Bell 2008; Bell 2013), but there has been some indication that a grain-only perennial wheat crop could still be a profitable alternative (Pimentel et al. 2012).

Ultimately, while there has been much speculation as to the financial viability of perennial grain crops through reduced input costs and secondary revenue streams, research in this space has been relatively lacking (Crews et al. 2018; Ryan et al. 2018). System viability will be dependent on the evaluation of economic returns in relation to potential ecosystem services, which can vary across both a political and physical landscape. Marginal lands that are more vulnerable to environmental degradation have more to gain from a perennial cropping system, while policies that provide financial incentives for environmentally conscious land management practices can further enhance the feasibility of such systems. Thus, economic analyses should be performed for a given crop x environment, which may provide insight to necessary crop characteristics and inform varietal development for a particular growing region.

Research here aims to investigate one such scenario: perennial and annual ratooning grain sorghum viability in north Georgia. The perennial variety will be the result of a cross between the annual grain sorghum, *Sorghum bicolor,* and the perennial *S. halepense* as described in Conway et al. (2020). Grain sorghum is the crop most advanced along the perennial breeding pipeline after rice and Kernza, while the annual *S. bicolor* also has the ability to 'ratoon' or regrow after harvest (McCormick et al. 1995; Cox et al. 2018). This allows for the potential for a short-term goal of an improved ratooning cultivar in Georgia's relatively warm climate, while the longer-term goal of a perennial variety is pursued. A 2011-2013 field study of early progress in perennial sorghum breeding efforts showed the highest yielding *S. bicolor* x *S. halepense* lines achieving 54% of the commercial check yield with significant improvement in seed weight and grain yield across breeding cycles (Nabukalu et al. 2016). A recent discovery of viable diploid germplasm derived from a *S. bicolor* x *S. halepense* cross is expected to greatly increase the expediency of perennial sorghum development, further necessitating timely investigations into the economic viability of perennial varieties (Cox et al. 2018).

Farmers often rely on enterprise budgets for profitability analysis (Doye 2016). UGA's Department of Agricultural and Applied Economics provides such budgets for Georgia grain sorghum, both dryland and irrigated (agecon.uga.edu/extension/budgets.html). These farm budgets can serve as the basis for comparison between an annual and ratooning or perennial grain sorghum to determine the viability and potential adoption rates of these alternative cropping systems.

In efforts to better understand what traits of perennial crops would best incentivize farmers to switch from annual to ratooning/perennial cropping system, this project will simulate three perennial and ratooning sorghum cropping system scenarios in the Georgia Piedmont region to explore costs under each scenario, using current Georgia sorghum enterprise budgets as a benchmark. The three cropping systems investigated will be grain-only (GO), “Grains for Grass” (GFG), and “Grains for Conservation” (GFC). The grain-only scenario is one in which perennial/ratooning grain sorghum is adopted solely for grain production. “Grains for Grass” describes a scenario under which perennial grains are adopted for dual-use grain and forage production, while “Grains for Conservations” describes a scenario in which perennial grains are adopted for dual-use grain and soil conservation purposes. Both a ratooning annual sorghum and perennial sorghum will be investigated in all three scenarios. Through a calibration of literature values, the analysis will develop farm enterprise budgets for each perennial/ratooning scenario. Comparative analysis between the ratooning/perennial enterprise budgets and the budget of the current annual systems will highlight necessary thresholds for ecosystem services, yield, and/or cost reductions for a perennial system to appear viable.

Methods

Perennial Analysis

Three perennial systems were considered and compared to their conventional annual counterpart: a grain-only system, a ‘Grains for Grass’ system, and a ‘Grains for Conservation’ system. Analysis was performed through the modification of 2020 GA dryland sorghum enterprise budgets as provided by UGA's Department of Agricultural and Applied Economics (agecon.uga.edu/extension/budgets.html). It was assumed that year one costs were consistent across all perennial and annual systems, except for an additional silage harvest cost in the GFG system, which will be elaborated on later. For the conventional system, it was assumed that there would be consistent costs across all years. In all perennial systems it was assumed that in year two there would be no seed cost, weed control cost, or preharvest field preparation costs, which included fuel, labor, and machinery repair.

For most analyses, barring the yield reduction analysis, it was assumed that yields remained constant across years. Prior investigations into early breeding efforts for perennial sorghum varieties derived from a *S. bicolor* x *S. halepense* cross found no discernable change in yield from year one to year two (Nabukalu and Cox 2016), although it is possible to see increasing yields up to year two as seen in perennial grain Kernza (Culman et al. 2013), or a gradual decline in yield. Future long-term field trials in the region are needed to clarify how age of plant stand impacts yields. For most analyses, a three-year perennial cropping system was assumed, due to Bell et al.'s findings that additional years beyond three did not markedly change the economics of a perennial wheat system (2013).

In the GO and GFC system, it was assumed that there would be a halved fertilizer cost in years 2+. Bell et al. assumed halved fertilizer inputs across all years in an economic analysis of a perennial wheat system (2008), while Glover et al. suggest the potential for no fertilizer inputs in a perennial grain system (2010). Thus, our assumption of full fertilizer cost in year one and halved fertilizer cost in year 2+ is deemed conservative. However, it was assumed that fertilizer cost remained constant across all years in the 'Grains for Grass' system. This is because the removal of both grain and biomass from the system in the GFG scenario is considered costly from a nutrient perspective, requiring a greater degree of nutrient inputs. Estimates of annual fertilizer replacement for an annual grain sorghum system in which both grain and biomass is harvested are 103 kg/ha for N and 27.2 kg/aha for P (Williams et al. 2016). Similarly, investigations into biomass yields of perennial *S. bicolor* x *S. halepense* lines used 180 N kg per hectare to receive their observed yields. It is possible to meet the heightened nutrient requirements of a GFG system by other means, such as legume intercropping or the integration of livestock into the system, but those scenarios are not investigated here.

Livestock integration may be inhibited in perennial sorghum systems because of the species' propensity to accumulate prussic acid, which can be poisonous to ruminants (Slade 1903). Sorghum contains a precursor to prussic acid called dhurrin, which is not in itself toxic, but may be an indicator of the toxicity potential of sorghum (Gorz et al. 1987).

Critical to the GFC scenario is the assumption of a Natural Resource Conservation Service (NRCS) Environmental Quality Incentives Program (EQIP) payment in year one of the plant stand establishment. NRCS provides financial and technical support to farmers and ranchers in the voluntary employment of conservation practices, with EQIP being the agency's most frequently utilized conservation program (nrcs.usda.gov). Conversations with a regional NRCS representative established that EQIP Code# 512 (Forage and Biomass Planting) was most appropriate for the introduction of a perennial grain sorghum system, as sorghum was already listed as a forage option for that practice code. As of 2019, EQIP Code #512 for the introduction of perennial grasses amounted to an average state cost of $208.72/acre for perennial grain planting, available to the farmer in year one. Since EQIP is a cost sharing program, the government share can vary from 50-90%, with "HU" or historically underserved farmers eligible for the higher cost sharing rates, but this amount of $208.72 represents the state average. Aside from this additional revenue stream, the GO and GFC scenarios were the same.

In the GFG scenario it was assumed that silage yield was equal to average sorghum silage yields for the region, based on statewide variety yields conducted from 2017-2019 (swvt.uga.edu). Results across the north Georgia region were averaged to obtain dry yield of 5.13 tons/acre. This was also converted to a green yield of 14.57 tons/acre based on average percent dry matter for the region. Prior research in perennial *S. bicolor* x *S. halepense* lines found that the top ten percent of lines for biomass production outperformed the commercial forage sorghum hybrid, suggesting that a perennial sorghum could outperform current annual varieties for biomass production (Habyarimana et al. 2018). Similarly, Conway et al.'s investigations in a *S. bicolor* x *S. halepense* $F_{2:3}$ population found transgressive segregation in the progeny for traits pertaining to plant height and biomass, providing support for the idea of perennial sorghum varieties' potential to exceed biomass production of their annual counterparts (2020). The assumption in this analysis was based on average commercial forage hybrid performance across all varieties, not the best performing, and was thus considered conservative.

Prior investigations revealed a significant positive correlation between dry biomass yield and dry grain yield in perennial *S. bicolor* x *S. halepense* lines (Habyarimana et al. 2018). Based on this association, it was assumed that a percent change in grain yield equivocated to the same percent change in biomass yield. While this assumption is simplistic, it leads to quite conservative estimates for the instance of low yielding systems. Since silage markets are less formalized, more complex, and highly variable compared to commodity grain ones (Guyer and Duey, 2974; Berger 2013; Lauer and Sterry 2013), it was deemed preferable to remain conservative in estimates of their economic viability. It was assumed that .715 tons/acre of biomass were left on the field per soil conservation requirements (Gallagher and Baumes 2012; Williams et al. 2016). Literature on appropriate sorghum silage pricing is limited due to the complexities outlined above, but a price of $37.57/ton was used based on 2019 reports for sorghum silage pricing from The Farm Financial Management Database (finbin.umn.edu). This was less than the $43/ton reported in the Northern Texas High Plains in 2016 (Mensah 2016) and the $42/ton reported in the California San Joaquin Valley in 2016 (Wright et al. 2016). This pricing was applied to the silage green yield, as indicated in the literature.

Additional silage harvest cost was based on GA corn silage harvest costs and corn silage labor requirements as outlined by UGA extension (agecon.uga.edu/extension/budgets.html). It was assumed that silage yields remained consistent in all years of the plant stand, except for the instances in which reduced secondary yields are explicitly investigated. This assumption was based on prior investigations into other perennial C4 grasses, which indicated that year two biomass yields typically exceed establishment year yields (Propheter et al. 2010). Additionally, initial research into the perennial grain crop Kernza has indicated that the harvesting of forage significantly increased aboveground biomass production (Pugliese et al. 2019).

Most analyses in the perennial system are displayed relative to break-even yield for both the net return above variable cost (NRAV) and net return above both variable and fixed cost (NRATC). Because the perennial sorghum breeding effort is still ongoing, this metric seemed the most useful, as it can serve as a means of informing this ongoing breeding process. Alternative analyses in which yield is assumed would be speculative and not particularly informative to growers in GA, as yield trials have not yet been performed in the state.

Investigations across the perennial systems include: break-even yield under standard assumptions outlined here, break-even yield comparison between three and four-year system, break-even yields under variable fertilizer and sorghum pricing based on twenty-year historical fertilizer data (https://www.ers.usda.gov) and available regional dryland sorghum pricing data since 2011 (https://agecon.uga.edu/extension/budgets/historical-budgets.html), tolerance for yield reduction in years 2+, break-even yields in a GFG system under a variable relationship between grain yield and biomass yield, and a crop profitability comparison for crops frequently put into rotation with grain sorghum in GA.

Crops for profitability comparison were determined by reviewing land use changes under ten-year historical Cropscape NASS CDL data for the region. Cropscape data indicated that grain sorghum is often put into rotation with corn, cotton, soy, and wheat in north Georgia. Thus, economic analysis of a perennial sorghum must also consider the opportunity cost of displacing these crops with a consistent perennial sorghum stand. UGA's 2020 farmer enterprise crop comparison tool was used to determine financial viability for dryland grain sorghum relative to other dryland crops for the region (agecon.uga.edu/extension/budgets.html).

Ratooning Analysis

As in the perennial analysis, the economic viability of a ratooning cultivar was investigated with the sorghum enterprise budgets provided by UGA's Department of Agricultural and Applied Economics (agecon.uga.edu/extension/budgets.html). The ratooning analysis differs from the perennial one in that expected yield of both the primary and secondary harvest is known. Therefore, investigated here was profitability under average primary and ratoon harvest, as well as under a theoretical improved ratooning cultivar based on observations of high-yielding ratooning lines in the literature. It was assumed that in year two of a ratooned sorghum, there would be no seed cost, weed cost, and a reduced preharvest cost because no planting occurred. Fertilizer was assumed to be consistent across years, unlike in the perennial investigations. While ratooned sorghums are not typically harvested beyond year two, analyses here were carried out to year three to serve as a basis of comparison for the perennial analysis. Additionally, it was assumed that a ratooning annual sorghum could also be eligible for the NRCS EQIP payment in year one, as the regional NRCS rep confirmed that sorghum was already listed as a forage for practice code #512.

To investigate a ratooning GFG scenario, it was assumed that annual grain sorghum could produce roughly 70% as much silage as a silage hybrid, according to relative yields observed in a KS sorghum biomass study (Williams et al. 2016). As in the perennial scenario, it was assumed that an additional .715 tons/acre were left in the field to meet soil conservation standards. This left an assumed silage green yield of 9.54 tons/acre or 3.35 tons/acre dry yield. Pricing was consistent with what was used in the perennial scenario.

Vinutha et al.'s evaluation of 36 *S. bicolor* lines for primary and ratooned biomass yield for forage revealed an average ratoon yield that was 37.2% of the primary yield. The line with the highest ratooning yield relative to the primary yield, forage variety COS 28, had a secondary biomass yield of 67.25% of the primary (2017). McCormick et al's investigations into ratooned *S. bicolor* forage yields also found significantly lower yields in the ratoon cut when compared to the first cut (1995). This was similar to what was observed in Duncan and Moss's evaluation of grain yield in ratooned sorghum, where temperate hybrid Savanna 5 had the highest yielding ratoon crop relative to primary crop at 66% of the primary yield (1987). On average, secondary ratooned grain yield came in at 46.1% of the primary yield. Accordingly, the ratooning analysis investigated viability under average ratoon yields for both grain and biomass, as well as "improved yields" as represented by the highest yielding ratooning lines for both grain and forage. It was assumed that yields of these lines relative to the primary yield represented reasonable breeding targets for improved ratooning cultivars.

Results

Perennial system

Break-even

UGA's 2020 farmer enterprise crop comparison tool indicated that dryland grain sorghum is expected to receive a net return of $25/acre above variable costs (NRAV) and a net loss of -$60/acre relative to total costs (NRATC) (agecon.uga.edu/extension/budgets.html). Perennial system findings are presented as break-even yields relative to NRAV and NRATC. Additionally these yield values are presented as % of current expected annual grain sorghum yields (65 bushels/acre) and % of break-even yields for annual grain sorghum, since current GA yields indicate a net loss relative to NRATC. In the instance of the GFC scenario, where it is likely that marginal lands eligible for soil conservation payments produce lower yields than regional averages, a range of % of current expected annual sorghum yields is provided, spanning from the lower end of the yield distribution (49 bushels/acre) to the expected average (65 bushels/acre). A range of values here reflects the non-specific nature of marginal land as a classification, which can be determined by a number of factors ranging from adverse morphology to inadequate transportation infrastructure (Pancaldi et al. 2020). Hopefully, this collection of metrics permits a more comprehensive picture of both necessary breeding yield targets and the viability of a perennial variety relative to its annual counterpart.

At current pricing, a conventional annual dryland sorghum system can break even with variable costs at 60.7 bushels/acre/year but requires a yield of 89.2 bushels/acre/year to break even according to total costs, which is 137% of current expected yield for the region (Table 3.1).

A perennial GO system is economically viable at a yield of 35.4 and 61.1 bushels/acre according to NRAV and NRATC, respectively (Figure 3.1). A GFG system performs slightly poorer according to NRAC, with break-even yields at 36.3 bushels/acre and better according to NRATC, at 46.1 bushels/acre (Figure 3.2). The most viable system is the GFC one, which can break even at yields of 15.1 bushels/acre/year according to NRAV and 40.1 relative to NRATC (Figure 3.3). These findings suggest that a perennial system could be profitable at yields 23.2%-54.5% of current standard dryland sorghum yields according to NRAV, and at 61.6%-94% of current yields according to NRATC (Table 3.1). Because current annual yields are not profitable relative to total cost, it is also useful to consider that the perennial systems are profitable at 44.9%-68.5% of the yields required for an annual dryland system to be profitable.

Length of perennial system

While there may be ecological benefits to a perennial system that extends beyond three years, there are not substantial economic benefits (Table 3.2). A four-year plant stand, as compared to a three-year plant stand, does permit profitability at a slightly lower yield in a GO and GFG system, but this only amounts to a difference of a maximum of 2 bushels/acre/year. Conversely, a GFC system actually needs higher minimum yields to break even in a cropping scenario that extends beyond three years. This is because the ability of the NRCS EQIP payment in year one to compensate for losses in subsequent years diminishes as the age of the plant stand increases.

Historical Fertilizer and Sorghum Pricing

Investigations into variable fertilizer pricing reveal greater sensitivity to nitrogen than phosphate (Table 3.3, Figure 3.4- 3.7). Both nitrogen and phosphate price indexes were based on twenty-year historical data for these fertilizer inputs (https://www.ers.usda.gov). Results indicate that the perennial systems become relatively more viable with fertilizer input price increases, due to the smaller slope of the perennial break-even yield in response to fertilizer pricing (Table 3.3). The GFG system, despite having higher fertilizer inputs relative to both the GO and GFC system, has the lowest sensitivity to fertilizer pricing, due to this system's multiple revenue streams which increase revenue per unit fertilizer relative to the other systems. Results also indicate greater NRATC break-even sensitivity relative to the NRAV metric, with this difference being greater in the annual compared to the perennial system. While this analysis was intended for pricing of fertilizer, the same findings would also apply to relative rate of fertilizer application, due to the linear relationship between both metrics and total cost.

Yield Loss

While the bulk of the analyses here assumed consistent yields across age of the plant stand, it is also possible that yields decrease across years or perhaps improve up to year two and then decrease. Analysis into yield loss tolerance across the perennial systems revealed the comparative advantage of a GFC system in lower primary yield scenarios (Table 3.3, Figure 3.8). This is due to the supplementary NRCS income stream, which can compensate for future yield reductions. Additionally, because our model assumes that both grain yield and biomass yield are shifting in tandem in a GFC scenario, once a certain primary yield threshold is hit (around 48 bushels/acre) those two revenue streams build on each other and the GFC system quickly become able to tolerate high rates of yield loss in subsequent years. As observed in the break-even analysis, a GO system cannot tolerate yield reductions if the primary yield falls below 61.1 bushels/acre/year, while a GFG and GFC systems have yield floors of 46.1 and 40.1 bushels/acre/year, respectively, relative to NRATC.

‘Grains for Grass’ variable yield relationships

The majority of analyses into break-even yield here assumed that a percent change in grain yield was matched with an equivalent percent change in biomass yield, due to the observed positive relationship between biomass and grain yields in lines of early stage perennial sorghum breeding efforts (Habyarimana et al. 2018). However, the strength of this positive relationship has not yet been determined in GA, and it is therefore of interest to see how break-even yields shift as the relationship between grain yield and biomass yield fluctuates. Results indicate that when biomass yields are relatively stable compared to the degree of change in grain yields, break-even yields drop precipitously (Figure 3.9). However, when biomass yields are highly sensitive to changes in grain yield, the gap between NRAV and NRATC yields tightens and both break-even yields approach current standard dryland sorghum yields. This indicates that the stability of biomass yields is critical to the economic viability of a GFG scenario.

Comparison to other dryland Crops

Historical Cropscape data indicated that grain sorghum is often put into rotation with corn, cotton, soy, and wheat in north Georgia. Dryland corn, soy, and wheat are all projected to have greater returns than dryland conventional sorghum according to both NRAV and NRATC (Figure 3.10, Figure 3.11). Cotton is less viable than conventional dryland grain sorghum. The perennial sorghum systems prove to be more competitive with these crop alternatives, with a GFC system being the most competitive except when compared to wheat. Strikingly, a GO perennial sorghum would need to achieve yields higher than current annual yields to be price competitive with wheat relative to both NRAV and NRATC. The GFG system is the most competitive with wheat, where the secondary silage revenue allows for lower break-even yields. Perennial sorghum systems could be on par with both corn and soy rotations with yields significantly lower than current annual sorghum yields. These mixed results indicate that there may be difficulty in converting land currently in mixed wheat/sorghum systems to grain-only perennial system in this region, but GFG and GFC might be possible with increased yields. It is noteworthy that the perennial systems are consistently more competitive than current annual sorghum.

Ratooning Annual System

Investigations into ratooning cultivars at average yields revealed that a system in which only grain is harvested or which both grain and forage is harvested is not viable after year one (Table 3.5, Figure 3.12). A GFC system would still operate at a net profit if harvested at year two, but this would represent a total decrease in income compared to only harvesting in year one. There is not strong evidence for the viability of ratooning systems in GA with yields as they currently stand without the supplementary NRCS EQIP payment in year one.

An annual system operating with improved ratooning cultivars achieving 66% of primary yield had better results. Under this scenario, results suggest that a GO system might be harvested in subsequent years, because the system is at least operating above variable costs and operating at least as well as year one, which is the standard performance of dryland grain sorghum for the region (Table 3.6, Figure 3.13). A GFC system could potentially be viable enough for a farmer to harvest out to year three, although this would represent a slight net loss relative to total costs. While the GFG system is profitable in year two relative to variable costs, there is a loss relative to total cost, which increases in year three, suggesting a farmer would not harvest in the third year. Analyses here assumed that the GFG system was able to achieve secondary biomass yields of 67%, as observed in best performing lines in Vinutha et al. (2017).

Discussion

Results here highlighted the comparative advantage of dual-use perennial grain sorghum systems, where the grain revenue stream is supplemented with either NRCS EQIP payment in year one or additional silage sales. These findings indicate that yield targets as low as 61.6% of current dryland sorghum yields could prove viable in GA for the GFC scenario, which is encouraging for breeders who have already observed perennial yields amounting to 54% of their annual counterparts. However, it is also important to consider farmer eligibility for such NRCS payments in the region, which could either support or negate breeding efforts for such yield targets.

The relative immediacy of a viable GFC scenario emphasizes the need for breeding efforts that are tailored particularly to marginal lands. The stated yield target of 61.6% of current annual yields is relative to expected regional yields of 65 bushels/acre, while marginal lands eligible for NRCS EQIP payments are likely to yield less than that average. While it is anticipated that perennial varieties will be relatively more productive than annual ones on marginal land, such assumptions need to be validated through field trials. It is important to breed cultivars specifically for productivity on marginal land, by making selections in such growing environments rather than optimal ones. In theory, the genetic diversity inherent to the perennial parent *S. halepense* can be capitalized on in making such selections.

While the GFG scenario did not prove to be quite as attractive as the GFC scenario, future breeding efforts could greatly improve the viability of this option. Sorghum varieties as they currently stand cannot be used in a direct grazing model, due to the propensity of this species to produce prussic acid, which can be fatally poisonous to ruminants. Consequently, the economic analysis here only considered a GFG scenario in which sorghum biomass is harvested for silage, which eliminates the risk of prussic acid poisoning. At roughly $265/acre, silage harvest is a significant expense, which compromises the profitability of GFG systems. Additionally, there is the potential for integrated livestock to redeposit nutrients back into sorghum system, allowing for a reduction in external fertilizer inputs. Thus, the development of grain sorghum lines without prussic acid could facilitate direct grazing GFG systems that could be both immensely more profitable and ecologically sustainable. Even in a silage harvest scenario, GFG proved to be a fairly viable option, particularly if biomass yields remain relatively stable and close to average yields even under decreased grain yield scenarios.

It is noteworthy that all perennial sorghum systems appeared quite competitive with current annual dryland systems. For instance, while a GO system would need yields at 94% of current average dryland yields to break-even with total cost, a GO system reaches break-even at 68.5% of the yield that an annual variety breaks-even at. This is because current annual sorghum systems operate at a net loss above total cost. Similarly, all perennial systems were more competitive than annual systems when compared to other crops frequently put into multi-year rotation with sorghum. This suggests that perennial sorghum might facilitate greater adoption of grain sorghum in the region, generally.

Projections here did not assume any form of price premium for a perennial variety compared to a conventional annual grain sorghum. This may be considered conservative, as preliminary pricing data suggests that the perennial grain Kernza does receive a premium over its annual wheat counterpart (Muckey 2019). However, grain sorghum also differs from wheat and wheat alternatives in that it is primarily used as animal feed in the U.S. (Cox et al. 2018). There may be less potential for a price premium when the crop in question is not intended for human consumption, inhibiting its access to a market which may exhibit increased willingness to pay for ecologically sustainable products. Establishing a robust U.S. consumer market for grain sorghum may be a complementary mode of ensuring financial viability of these perennial varieties. However, there are limits to relying on price premiums in dissemination of novel crop varieties. There is tension in balancing economic viability for farmers with downstream affordability for consumers, as a limited consumer base ultimately equivocates to limited adoption of perennial grain varieties and limited environmental gains.

The goal is not to get perennial grains in the ground via whatever means possible, as there would be extreme implications for global food security if all annual grain crops were replaced with perennial varieties with yields as they currently stand. However, economic viability can inform a segmented mode of adoption as grain yields are improved. Based on findings here, this could play out as follows in GA:

1. Grains for Conservation: Those eligible for soil conservation payments from NRCS for perennial planting schemes can adopt a perennial grain, thereby increasing soil quality while deriving extra income from the harvestable grain. This will ultimately increase food security through the production of food on marginal lands previously deemed unfit for agriculture.
2. Grains for Grass: Farmers growing forage for livestock transition to growing perennial grain varieties, adding another revenue stream through grain production.
3. Grain-only: As yields improve, perennial grain varieties are eventually primed to be planted with the primary intent of grain production.

Many assumptions were made in this analysis due to the uncertainty of the performance of yet undeveloped cultivars, which may differ from what is observed in the final product. The flexibility of this analysis in some ways emulates the phenotypic variance observed in the early stage perennial sorghum breeding lines and associated potential for improvement. Hopefully the observations and trends can be informative to breeders in their formulation of a perennial grain sorghum and inform an iterative approach to breeding as new information becomes available.

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Table 3.1. Break-even yields (bushels/acre/year) in a conventional, perennial grain-only, perennial ‘Grains for Grass’, and perennial ‘Grains for Conservation’ system according to both NRAV and NRATC. Break-even yields are also compared to standard dryland grain sorghum yields for the region (65 bushels/acre) and in the perennial systems, relative to the break-even yields of a conventional annual grain sorghum system.

	NRAV			NRATC		
	Break-even yield	% of standard yield	% of conventional break-even	Break-even yield	% of standard yield	% of conventional break-even
Conventional	60.7	93.4%	-	89.2	137.2%	-
Grain-only	35.4	54.5%	58.3%	61.1	94.0%	68.5%
Grains for Grass	36.3	55.8%	59.8%	46.1	70.9%	51.7%
Grains for Conservation	15.1	23.2%-30.8%*	24.9%	40.1	61.6%-81.2%*	44.9%

*in the instance of a Grains for Conservation scenario, it is likely that marginal land eligible for NRCS conservation payments would produce lower yields that is considered standard for the region, thus provided here is a range of % standard yield spanning from the lower end of the expected yield distribution (49 bushels/acre/year) to standard expected yields (65 bushels/acre/year)

Table 3.2. Break-even yields (bushels/acre/year) in a perennial grain-only, perennial 'Grains for Grass', and perennial 'Grains for Conservation' dryland grain sorghum system according to both NRAV and NRATC. Break-even yields are presented for both a three-year and four-year plant stand system.

	Three-year system		Four-year system	
	NRAV	NRATC	NRAV	NRATC
Grain-only	35.4	61.1	33.7	59.2
Grains for Grass	36.3	46.1	35.8	45.6
Grains for Conservation*	15.1	40.1	18.4	43.8

*in GFC, year 2+ yields below 28.5 (NRAV) and 53.4 (NRATC) bushels/acre represent a net loss, however the year one NRCS payment compensates for these losses

Table 3.3. Slope of dryland grain sorghum system break-even yields in response to variable fertilizer pricing. Fertilizer price index is relative to 2018 USD. Analysis is for a conventional and perennial system. One unit is equivalent to one additional bushel/acre/year in grain sorghum yield.

	Nitrogen		Phosphate	
	NRAV	NRATC	NRAV	NRATC
Conventional	9.5	10.5	3.8	4.2
Perennial GO	4.8	5.3	1.9	2.1
Perennial GFC	4.8	5.3	1.9	2.1
Perennial GFG	3.2	3.5	1.3	1.4

Table 3.4. Yield loss tolerance in a perennial grain-only, perennial ‘Grains for Grass’, and perennial ‘Grains for Conservation’ dryland grain sorghum system according to both NRAV and NRATC. First column represents primary yield (bushels/acre/year) and subsequent columns represent required yield in years 2-3 (as % of primary yield) in order to achieve break-even.

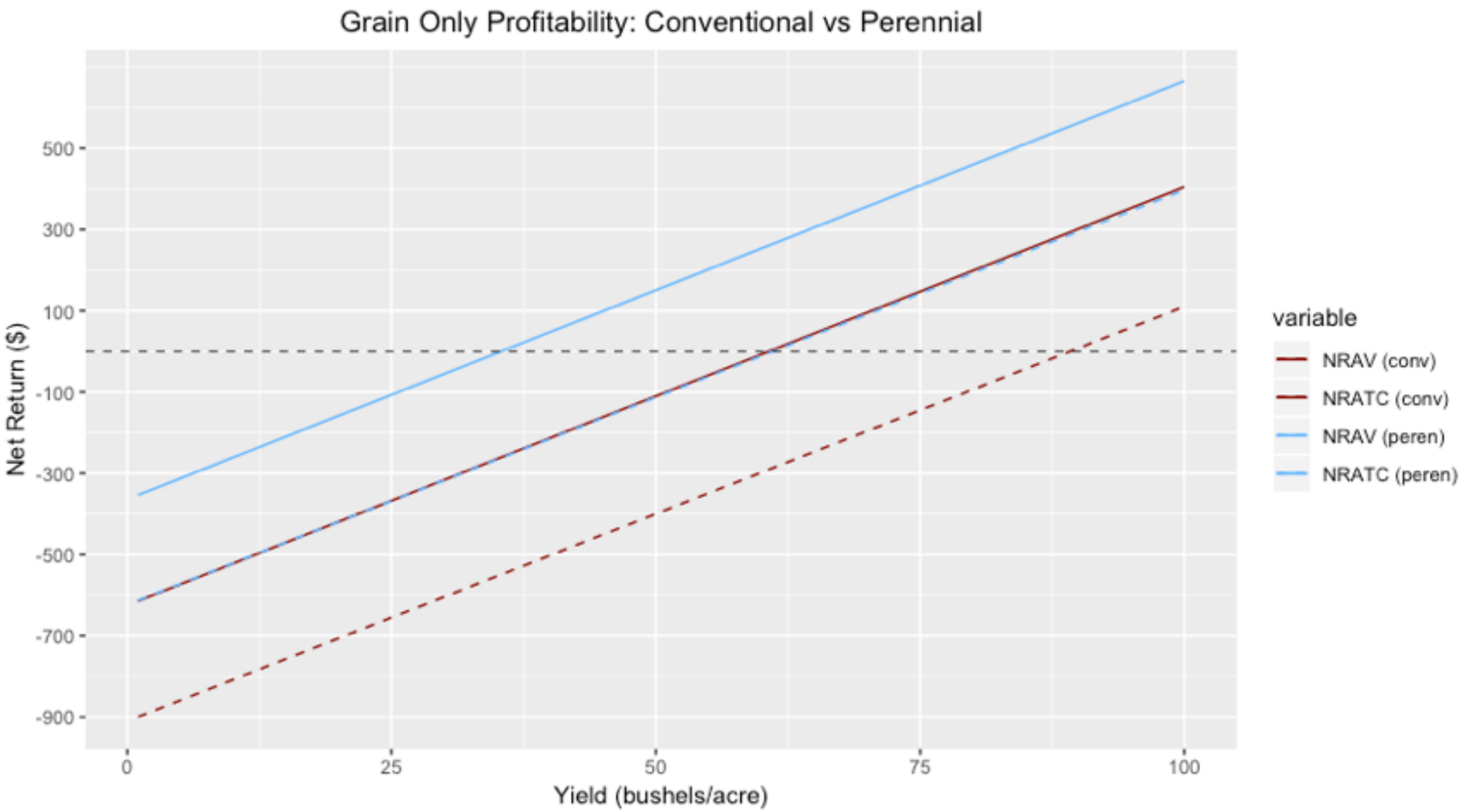
	Grain-only		Grains for Grass		Grains for Conservation	
Primary yield	NRAV	NRATC	NRAV	NRATC	NRAV	NRATC
40	83%	180%	53%	181%	7%	103%
45	68%	155%	2%	114%	0%	86%
50	57%	135%	0%	62%	0%	72%
55	47%	118%	0%	17%	0%	61%
60	39%	104%	0%	0%	0%	52%
65	32%	91%	0%	0%	0%	44%
70	26%	81%	0%	0%	0%	38%
75	21%	73%	0%	0%	0%	32%
80	16%	65%	0%	0%	0%	27%

Table 3.5. Total net return of a ratooning annual system across three years of a plant stand for an average ratooning cultivar (ratooned grain yield 46% of primary yield, ratooned biomass yield 37% of primary yield). Analysis is for both NRAV and NRATC for three ratooning annual systems: Grain-only, ‘Grains for Grass’, and ‘Grains for Conservation’.

		Year 1	Year 2	Year 3
Grain-only	NRAV	$14.76	-$15.82	-$46.41
	NRATC	-$82.29	-$201.36	-$320.43
Grains for Grass	NRAV	$130.09	-$3.19	-$136.46
	NRATC	$8.73	-$236.61	-$481.95
Grains for Conservation	NRAV	$223.48	$192.90	$162.31
	NRATC	$126.43	$7.36	-$111.71

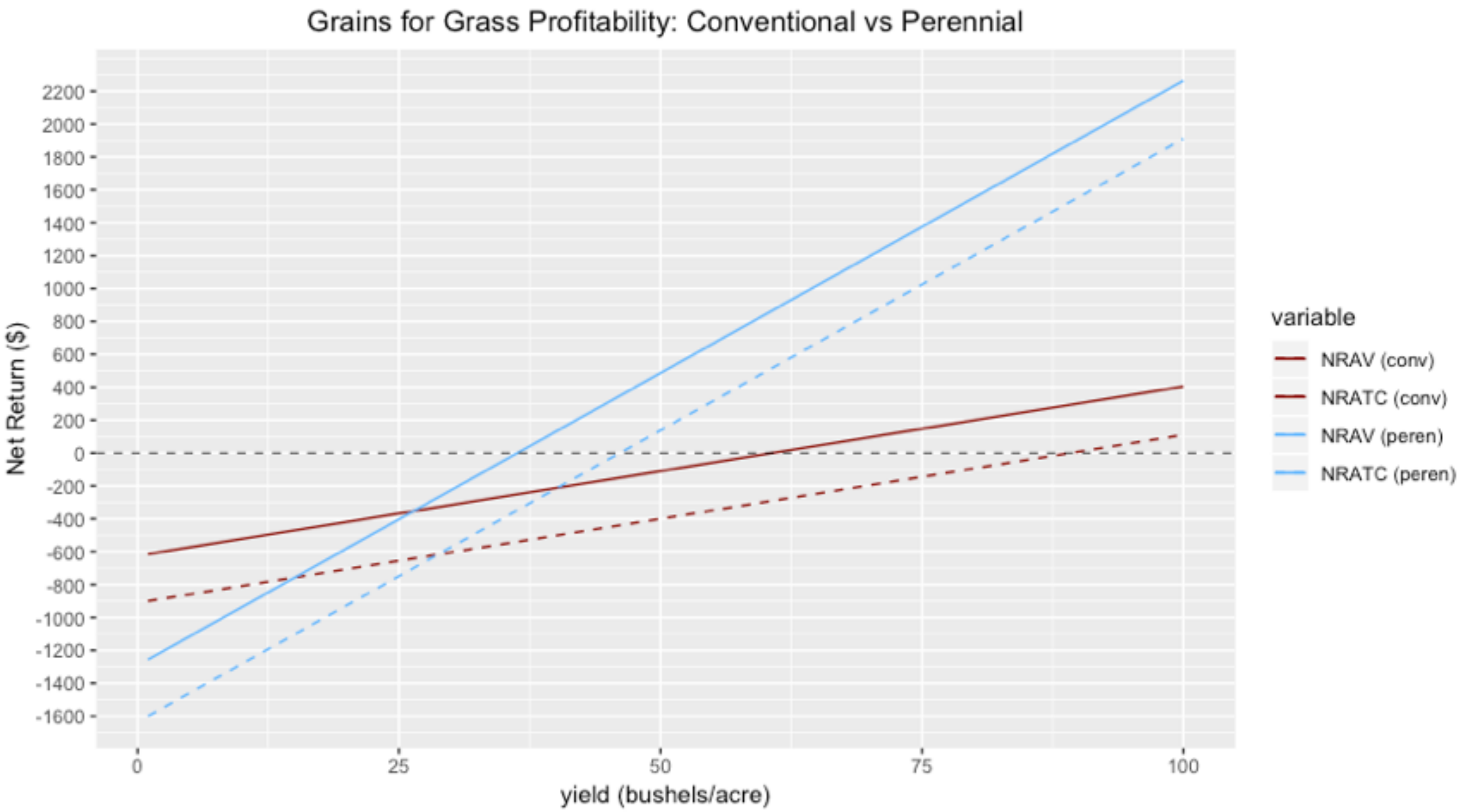
Table 3.6. Total net return of a ratooning annual system across three years of a plant stand for an improved ratooning cultivar (ratooned yield 66% of primary yield). Analysis is for both NRAV and NRATC for three ratooning annual systems: Grain-only, 'Grains for Grass', and 'Grains for Conservation'.

		Year 1	Year 2	Year 3
Grain-only	NRAV	$14.76	$28.57	$42.38
	NRATC	-$82.29	-$157.36	-$232.44
Grains for Grass	NRAV	$130.09	$148.24	$166.39
	NRATC	$8.73	-$85.58	-$179.90
Grains for Conservation	NRAV	$223.48	$237.29	$251.10
	NRATC	$126.43	$51.36	-$23.72



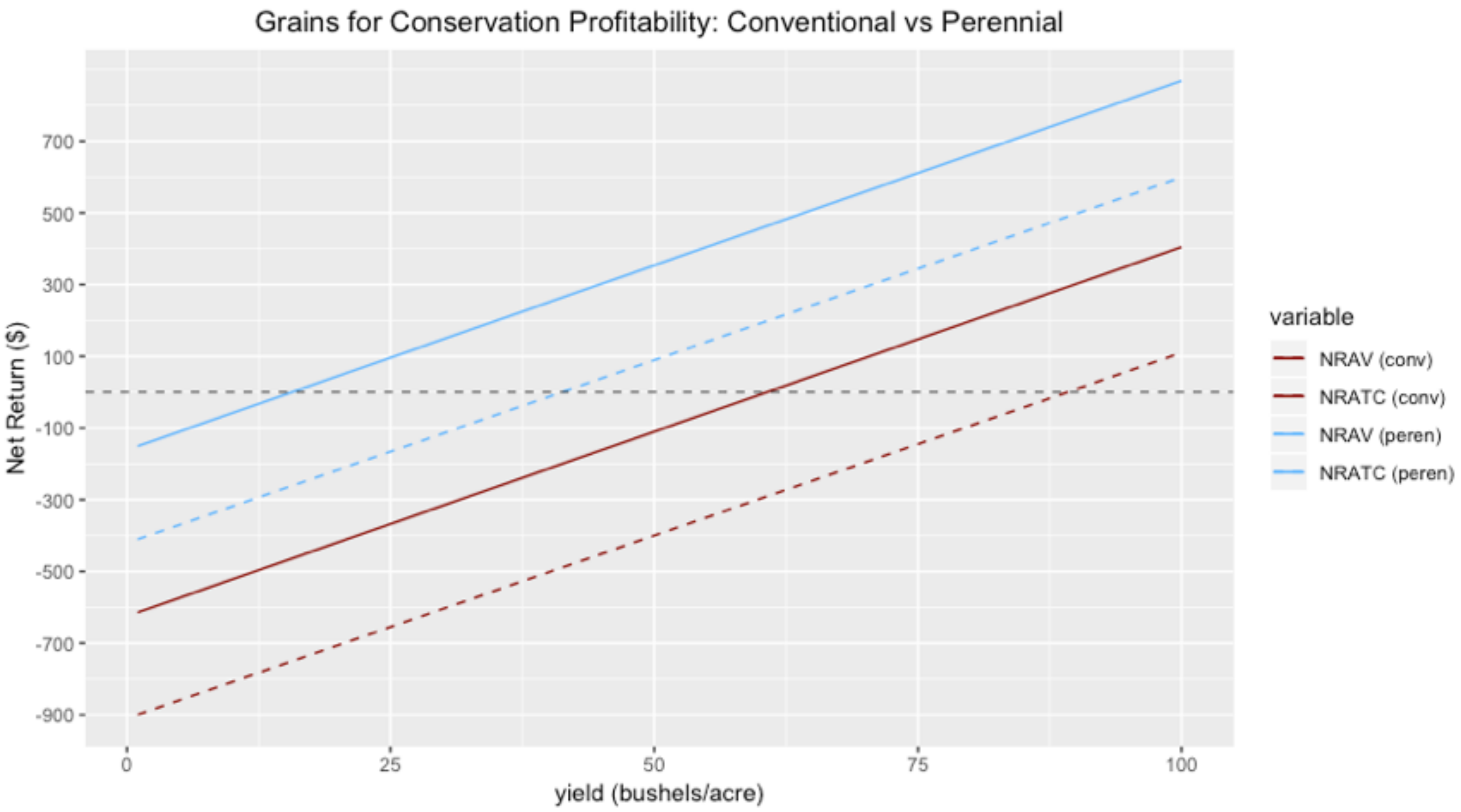
*dashed lines represent NRATC

Figure 3.1 Comparison of profitability in two dryland grain sorghum systems in GA: conventional annual and perennial grain-only. Analysis was conducted based on both NRAV and NRATC assuming 2020 pricing of $3.74/bushel for grain sorghum, $0.50/lb for nitrogen, and $.40/lb for phosphate.



*dashed lines represent NRATC

Figure 3.2 Comparison of profitability in two dryland grain sorghum systems in GA: conventional annual and perennial "Grains for Grass". Analysis was conducted based on both NRAV and NRATC assuming 2020 pricing of $3.74/bushel for grain sorghum, $0.50/lb for nitrogen, and $.40/lb for phosphate.



*dashed lines represent NRATC

Figure 3.3 Comparison of profitability in two dryland grain sorghum systems in GA: conventional annual and perennial “Grains for Conservation”. Analysis was conducted based on both NRAV and NRATC assuming 2020 pricing of $3.74/bushel for grain sorghum, $0.50/lb for nitrogen, and $.40/lb for phosphate.

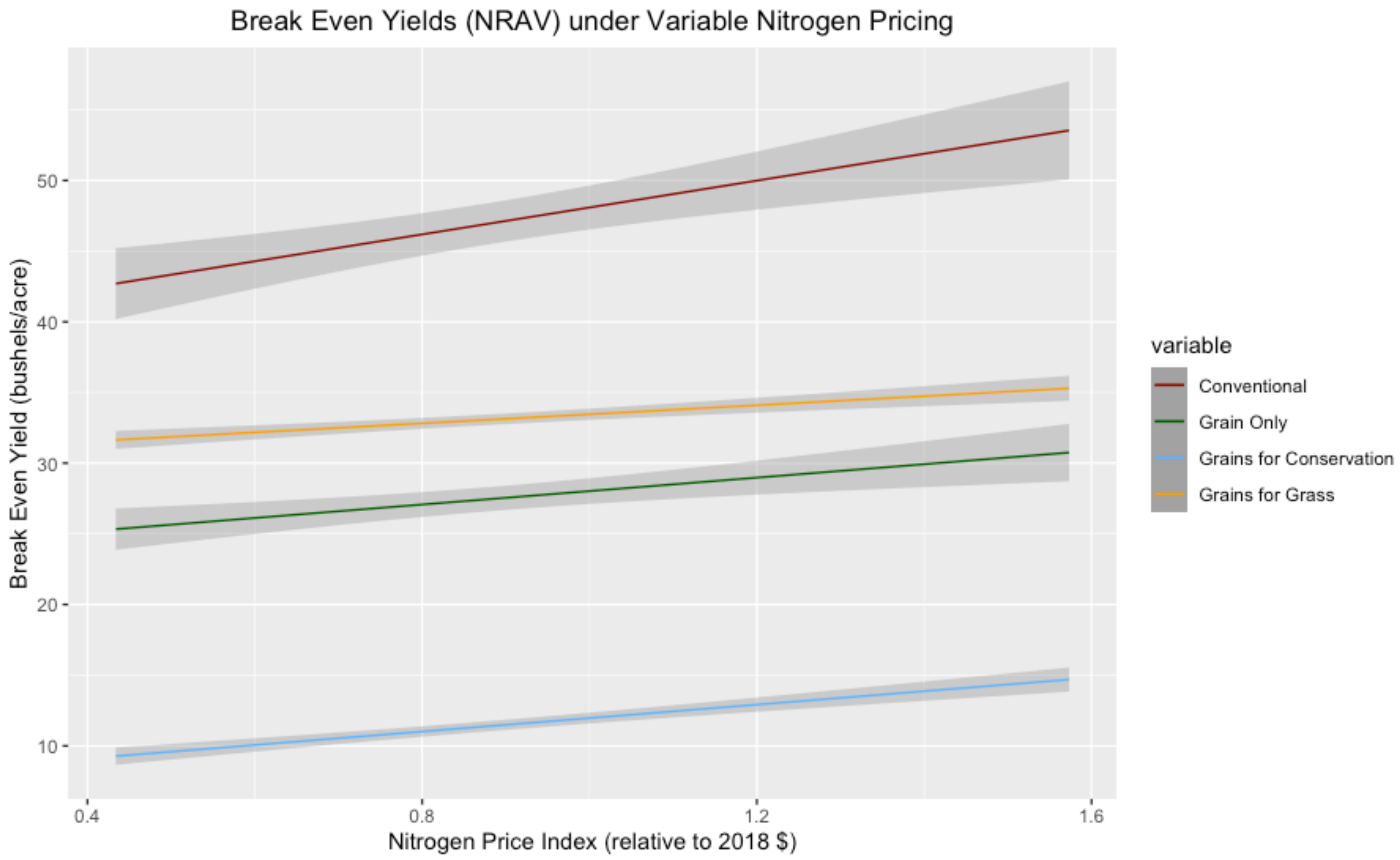


Figure 3.4 Break even yields (NRAV) for dryland grain sorghum across variable nitrogen costs (based on twenty-year historical data of nitrogen pricing). Shaded area represents 95% confidence interval based on 9-year historical sorghum prices for the region. Analysis was performed for a conventional annual, perennial grain-only, perennial 'Grains for Conservation', and perennial 'Grains for Grass' system.

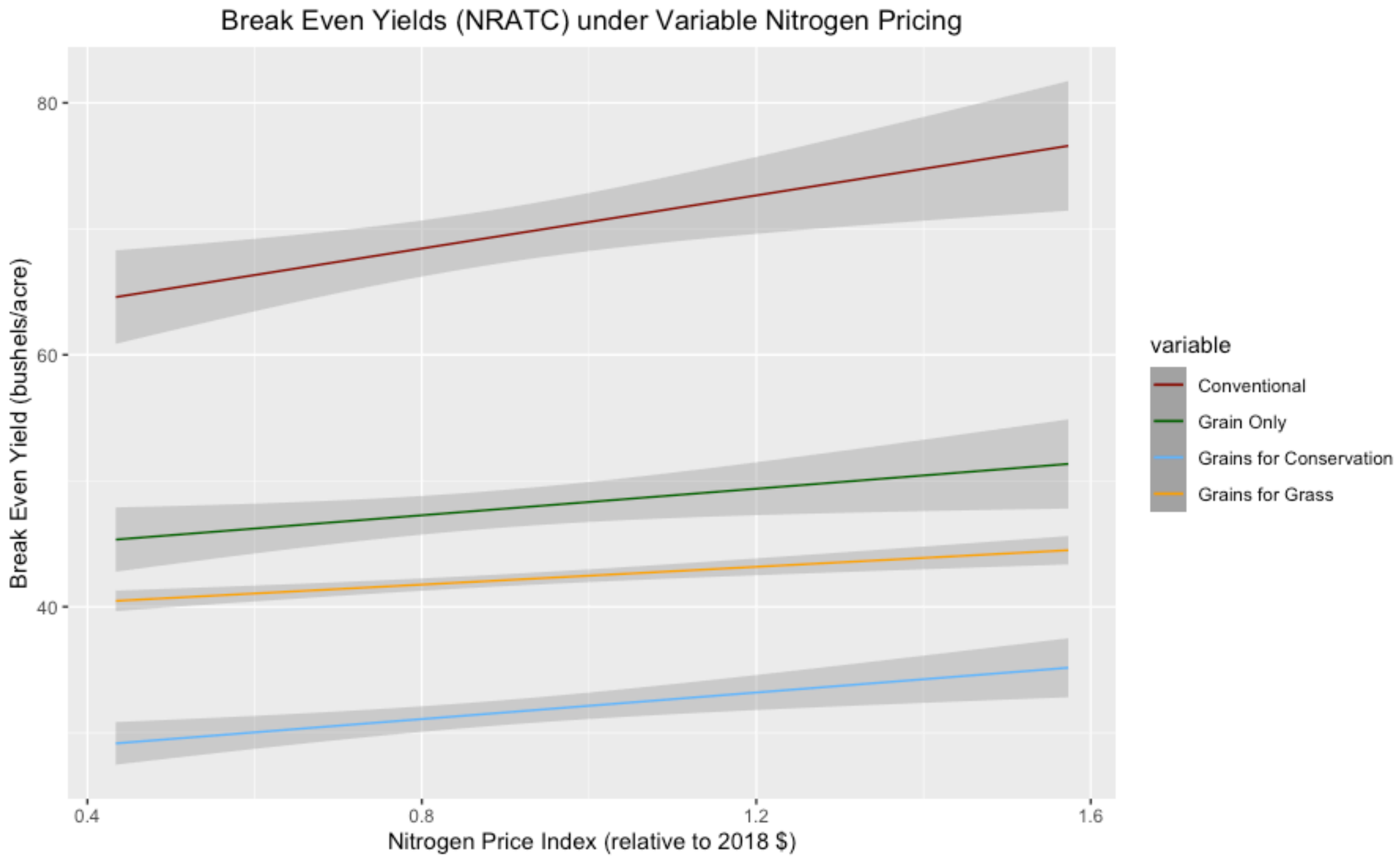


Figure 3.5 Break even yields (NRATC) for dryland grain sorghum across variable nitrogen costs (based on twenty-year historical data of nitrogen pricing). Shaded area represents 95% confidence interval based on 9-year historical grain sorghum prices for the region. Analysis was performed for a conventional annual, perennial grain-only, perennial 'Grains for Conservation', and perennial 'Grains for Grass' system.

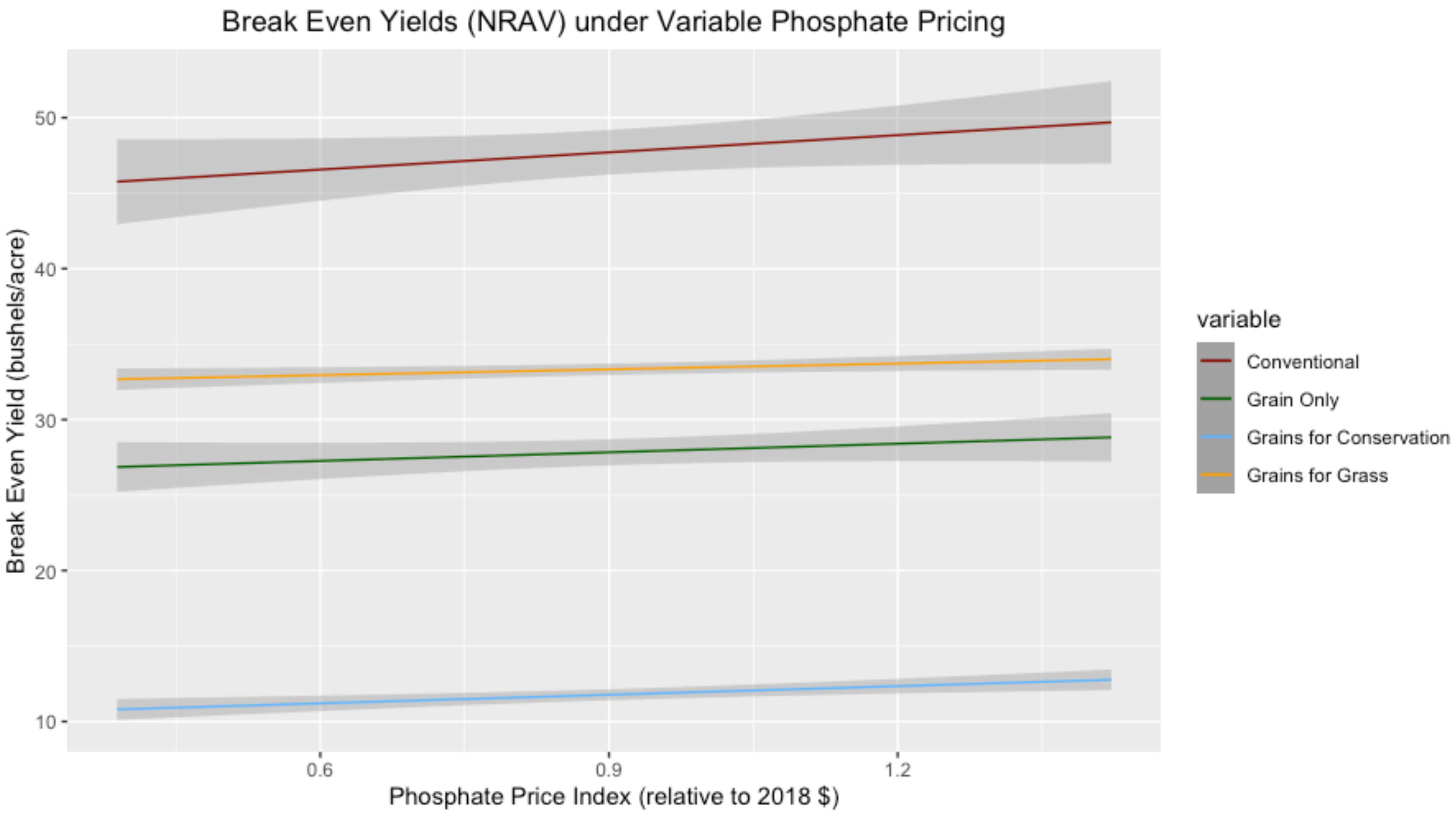


Figure 3.6 Break even yields (NRAV) for dryland grain sorghum across variable phosphate costs (based on twenty-year historical data of phosphate pricing). Shaded area represents 95% confidence interval based on 9-year historical sorghum prices for the region. Analysis was performed for a conventional annual, perennial grain-only, perennial ‘Grains for Conservation’, and perennial ‘Grains for Grass’ system.

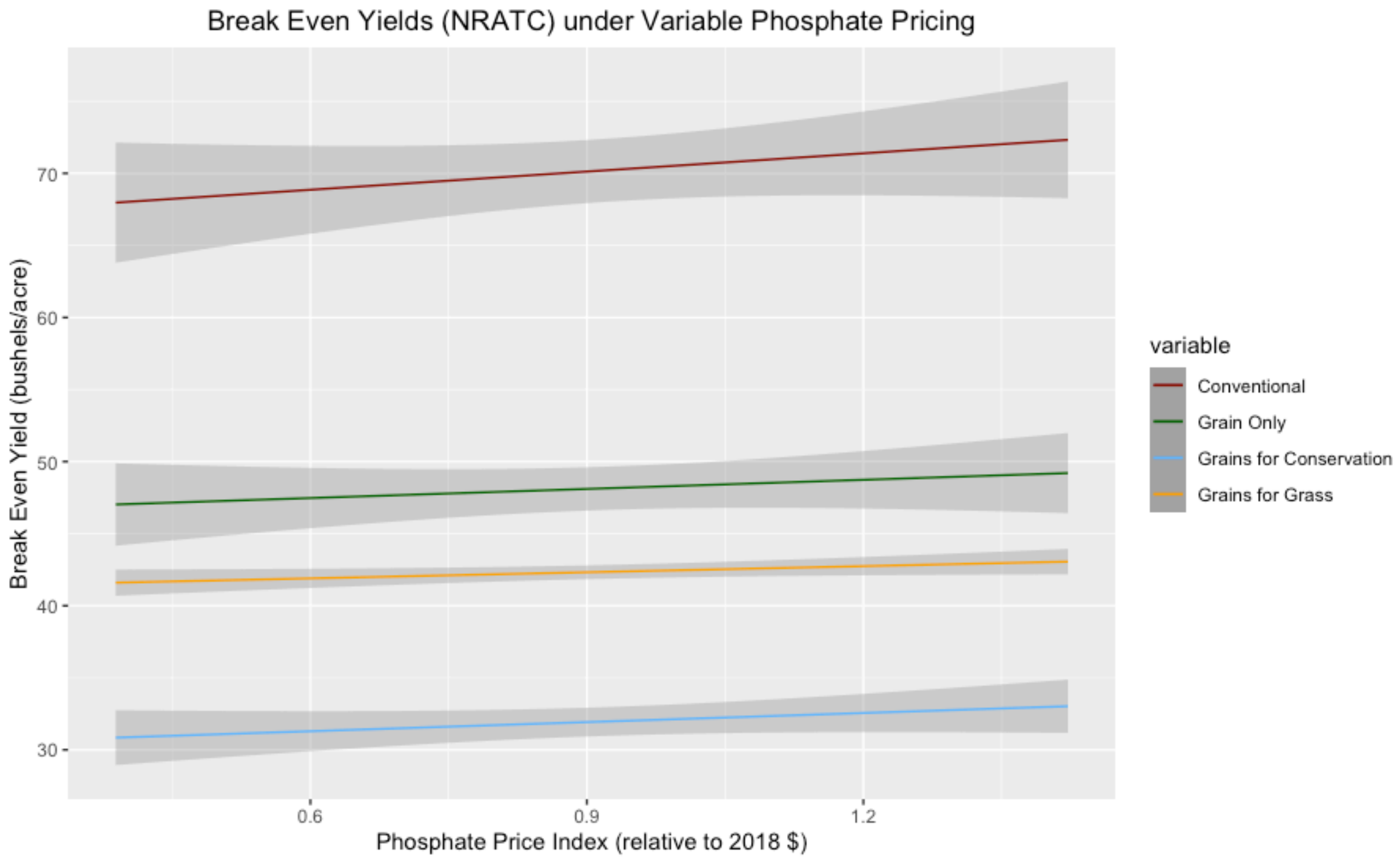


Figure 3.7 Break even yields (NRAV) for dryland grain sorghum across variable phosphate costs (based on twenty-year historical data of phosphate pricing). Shaded area represents 95% confidence interval based on 9-year historical sorghum prices for the region. Analysis was performed for a conventional annual, perennial grain-only, perennial 'Grains for Conservation', and perennial 'Grains for Grass' system.

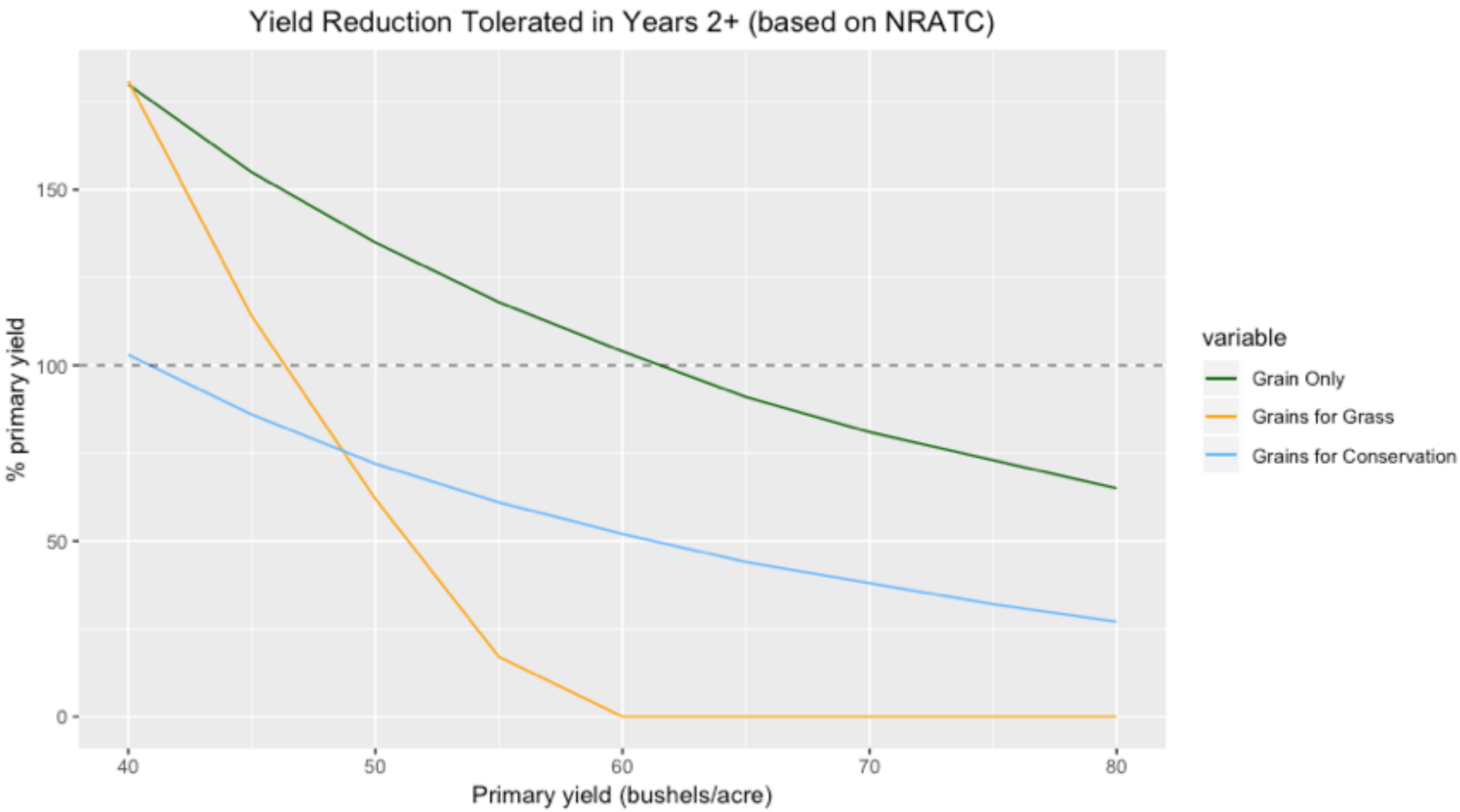


Figure 3.8 Comparison of primary yield and acceptable levels of yield reduction in subsequent years in order to maintain profitability based on NRATC. Analysis was performed on three perennial dryland grain sorghum systems in GA: grain-only, “Grains for Conservation”, and “Grains for Grass”.

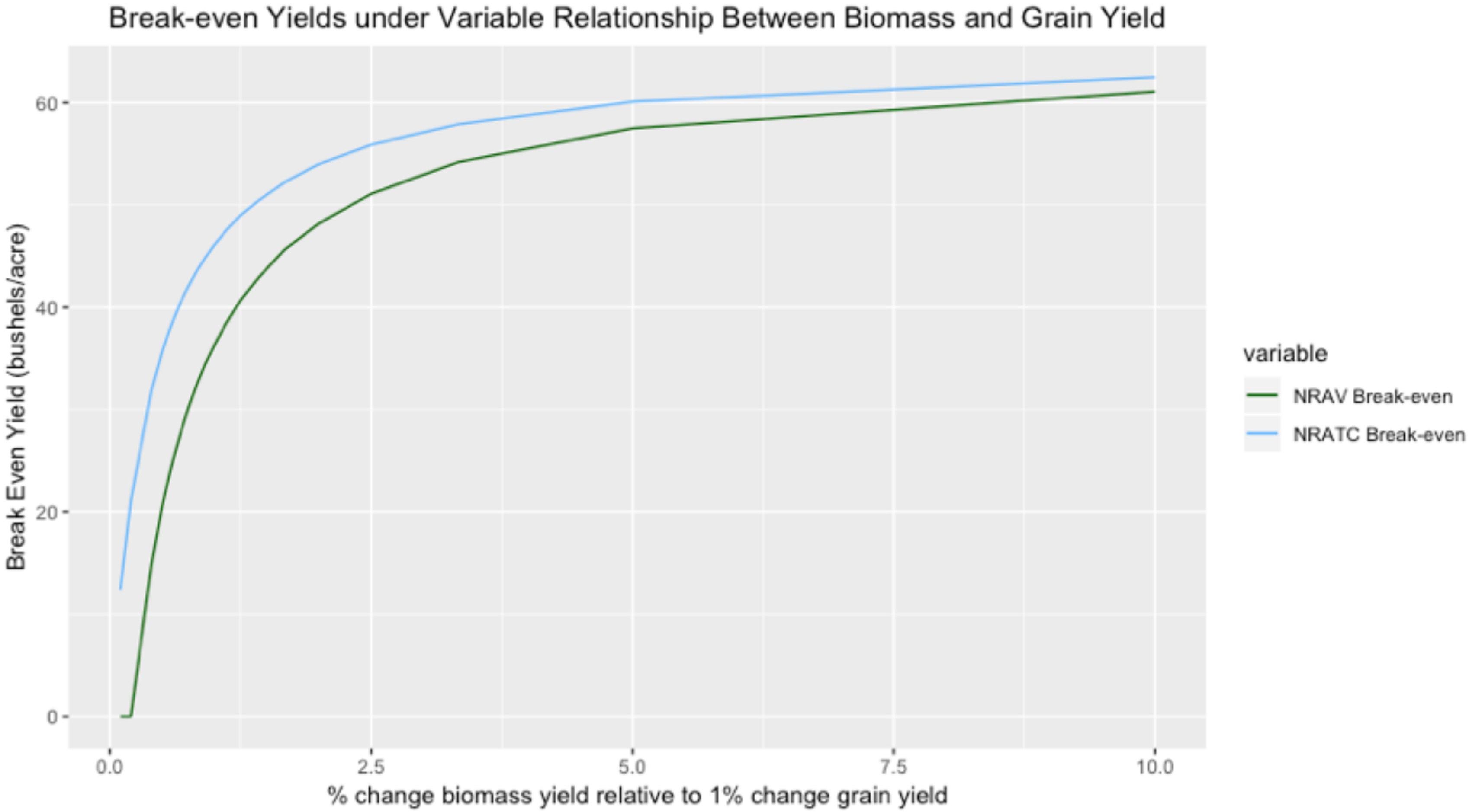
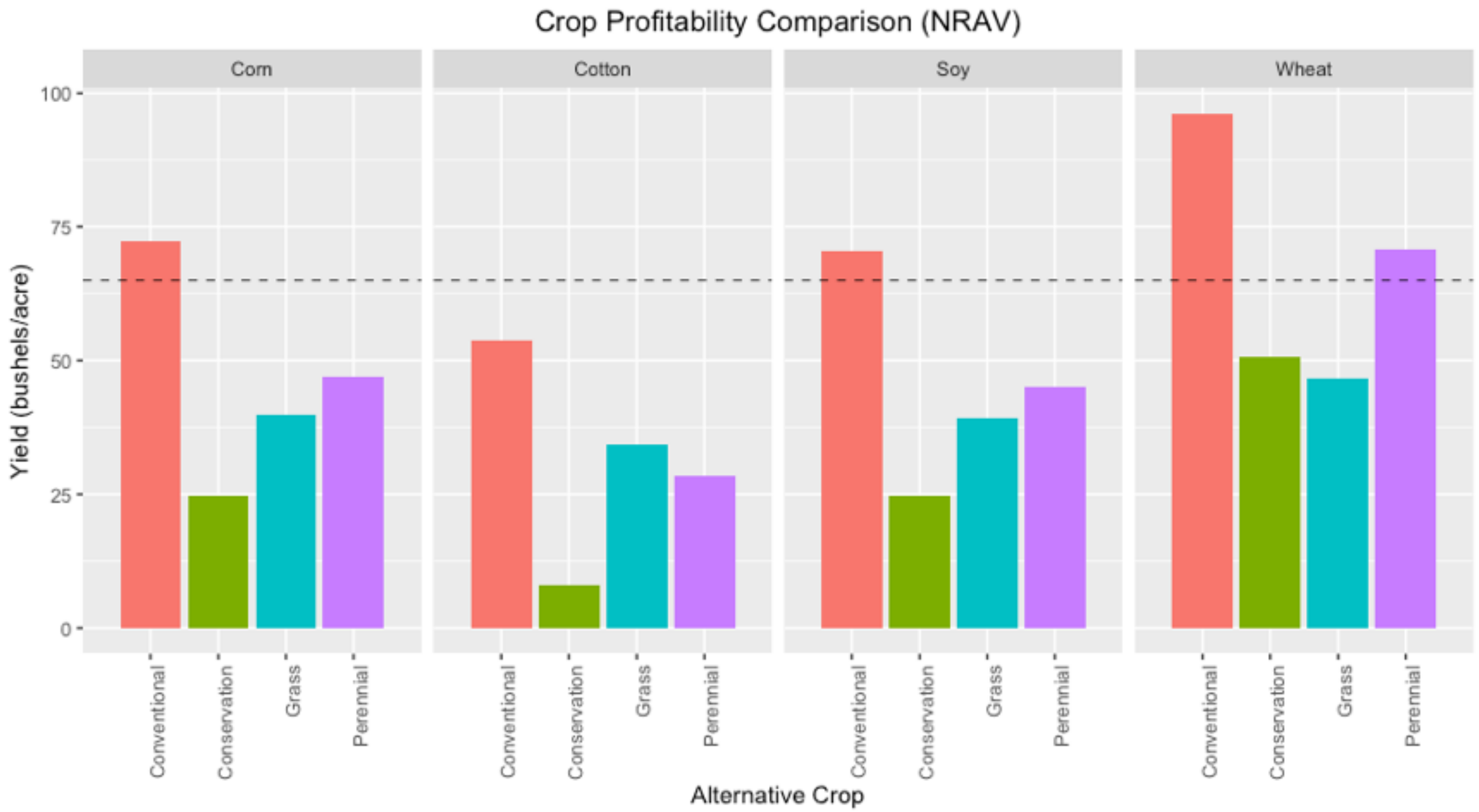
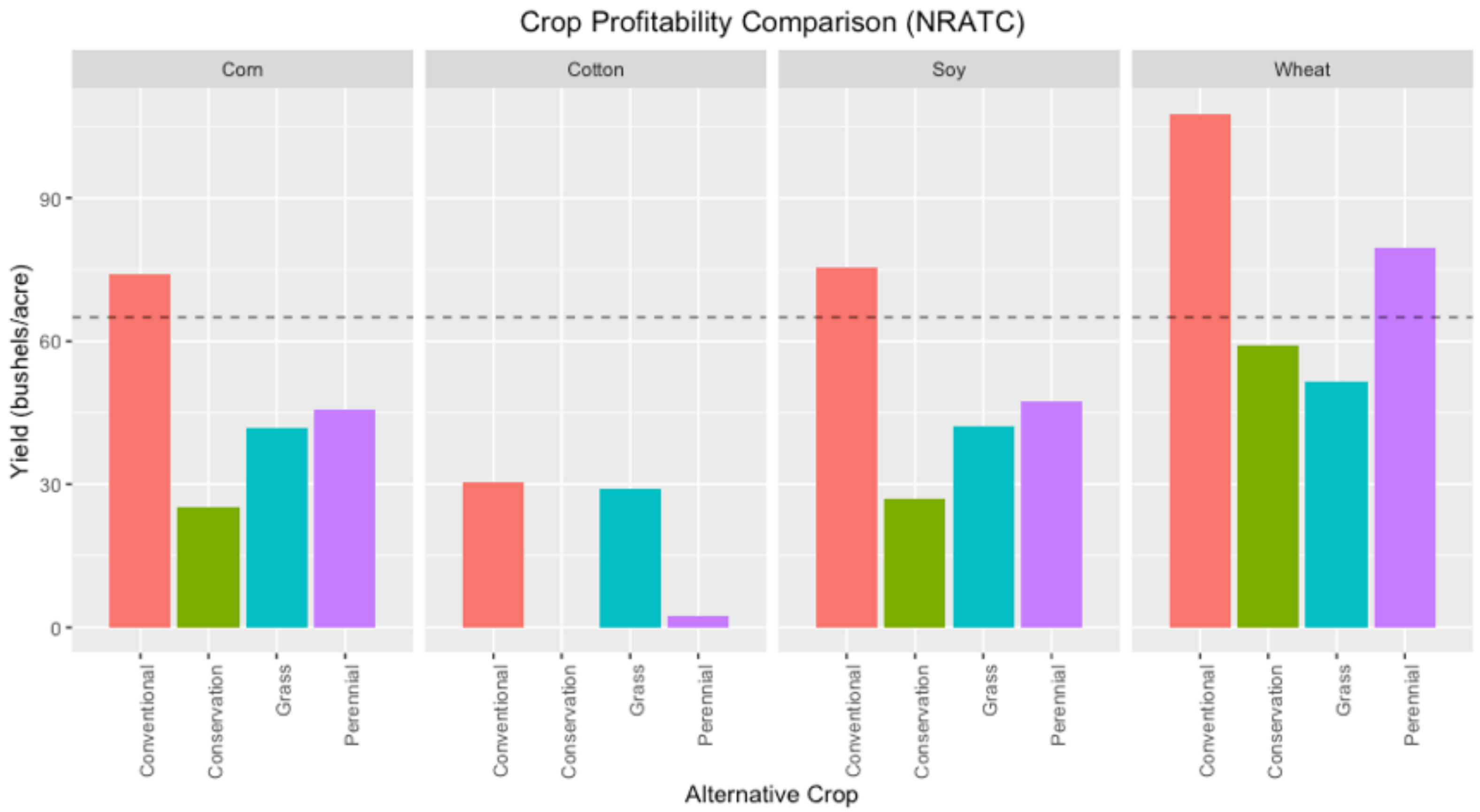


Figure 3.9 Break even yields in a ‘Grains for Grass’ system under a variable relationship between biomass yield and grain yield.



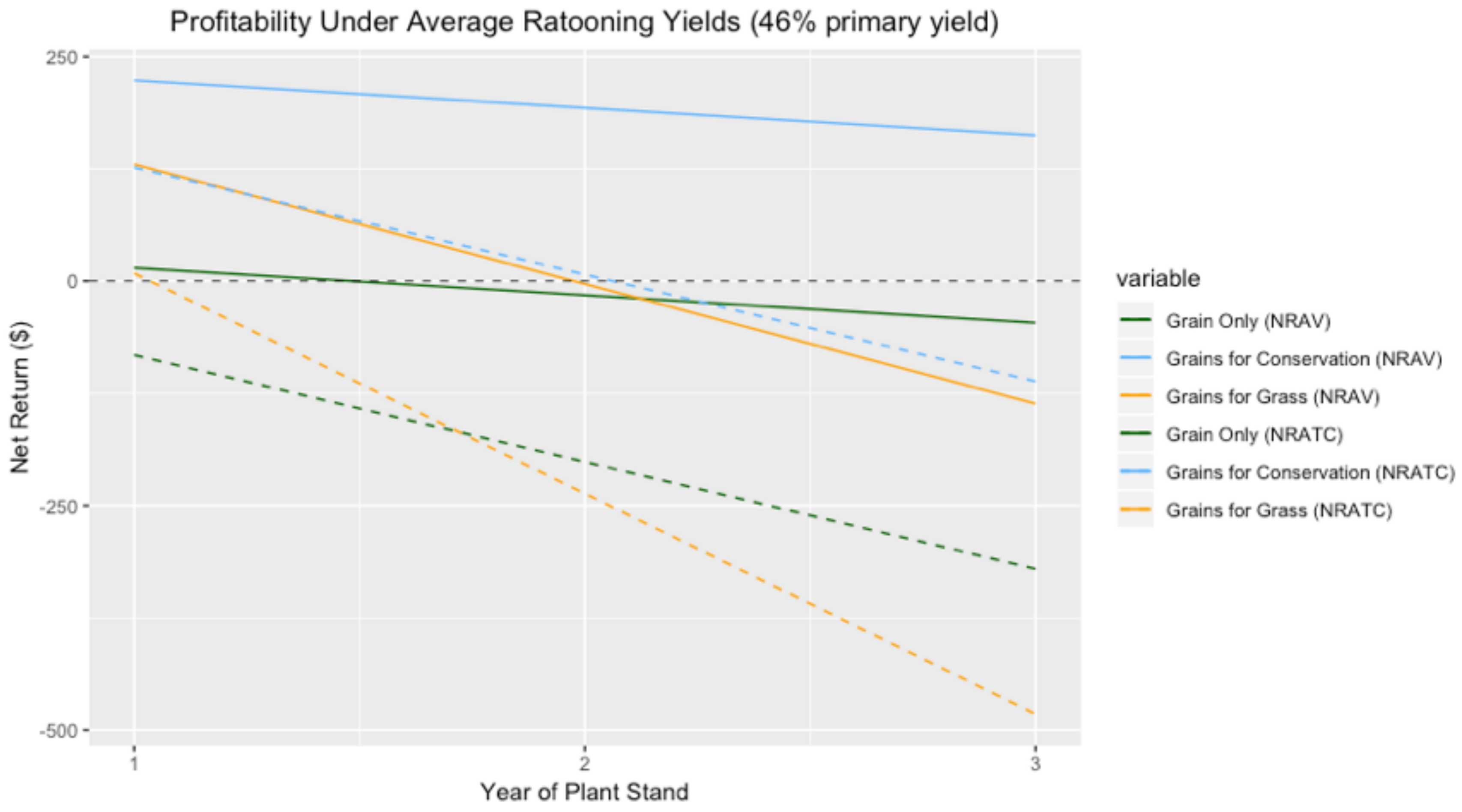
*dashed line represents current expected dryland sorghum yields in GA: 65 bushels/acre

Figure 3.10 Comparison of grain sorghum profitability relative to other dryland crops frequently put into a multi-year rotation with sorghum in north GA, based on NRAV values for each crop. Comparison was done for a conventional annual, perennial grain-only, perennial ‘Grains for Conservation’, and perennial ‘Grains for Grass’ system. Values displayed are required sorghum yields in each system to match the profitability of the alternative crop in question.



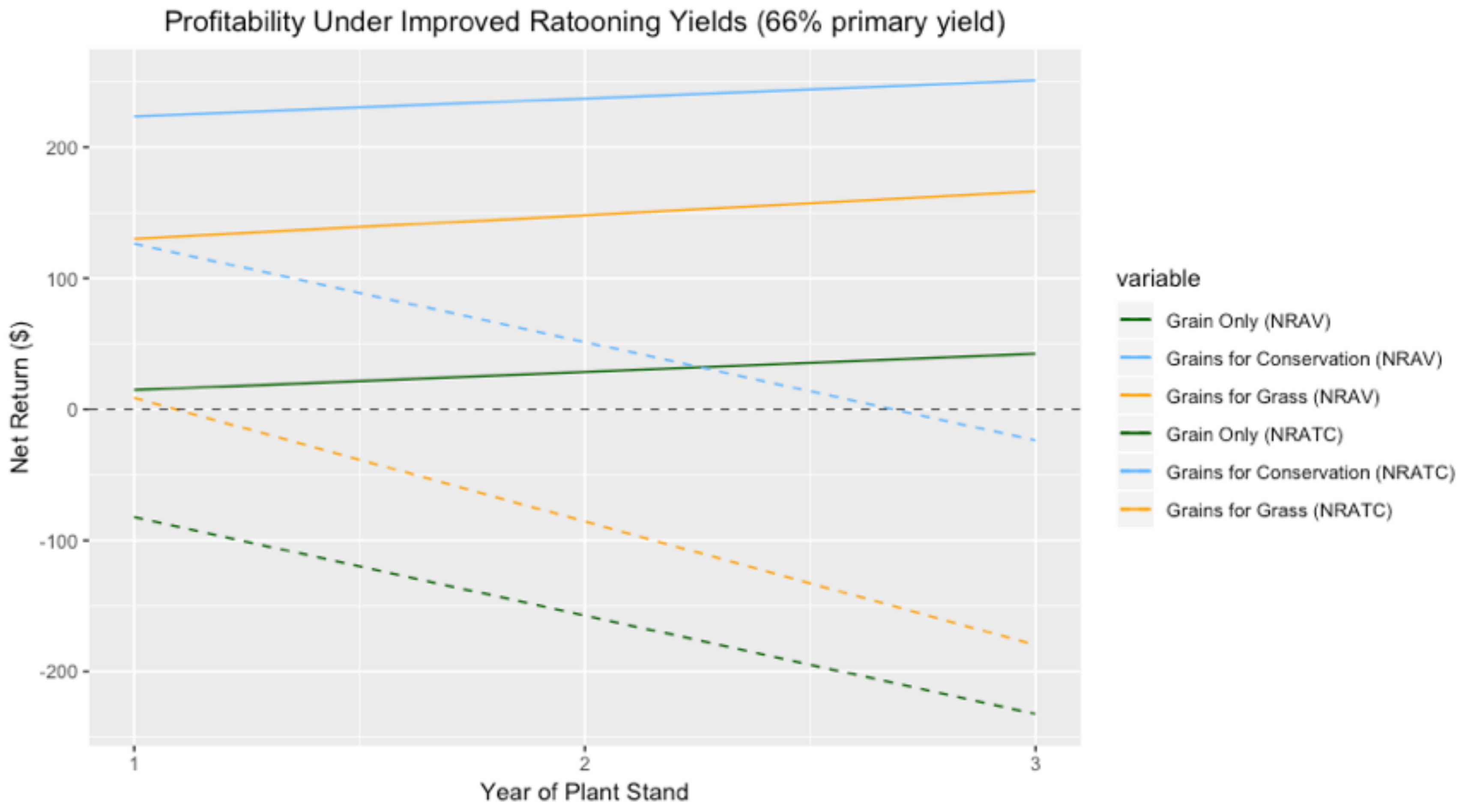
*dashed line represents current expected dryland sorghum yields in GA: 65 bushels/acre

Figure 3.11 Comparison of grain sorghum profitability relative to other dryland crops frequently put into a multi-year rotation with sorghum in north GA, based on NRATC values for each crop. Comparison was done for a conventional annual, perennial grain-only, perennial 'Grains for Conservation', and perennial 'Grains for Grass' system. Values displayed are required dryland sorghum yields in each system to match the profitability of the alternative crop in question.



*dashed lines represent NRATC

Figure 3.12 Comparison of total net return ($) of three ratooning dryland grain sorghum systems under average ratooning yields of 46% of primary grain yield and 37% primary biomass yield. Analysis spans 1-3 years of the system in question.



*dashed lines represent NRATC

Figure 3.13 Comparison of total net return ($) of three ratooning dryland grain sorghum systems under improved ratooning yields of 66% of primary grain yield and 67% primary biomass yield. Analysis spans 1-3 years of the system in question.

CHAPTER 4

RUSLE-BASED GIS MODEL TO ESTIMATE SOIL EROSION REDUCTION POTENTIAL IN NORTH GEORGIA WATERSHEDS[3]

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[3] Conway, TM and Paterson, AH. To be submitted to Agriculture, Ecosystems, and Environment.

Abstract

The replacement of annual cereal crops with perennial varieties has great potential for increased ecosystem resilience and improved food security. One particularly salient benefit to a perennial cropping system is the potential for reduction of soil erosion. The Natural Resources Conservation Service (NRCS) provides Environmental Quality Incentives Program (EQIP) payments to farmers employing conservation practices on land that meets the need for "substantial reduction in erosion". Such payments can help incentivize farmers to adopt perennial grain varieties and allow for financial viability of perennial grain systems at yields lower than what is currently required for their annual grain counterparts. Thus, investigations into farmer eligibility for NRCS EQIP payments can help inform the viability of certain yield targets for breeders. A RUSLE-based GIS model was implemented in two north Georgia watersheds to highlight such eligibility, with 2xT (soil loss tolerance) used as a metric for EQIP eligibility. Results highlighted a relatively high presence of soil erosion occurring at a rate greater than or equal to 2xT, with approximately 41.66% and 40.98% of land currently under crop cover in the two watersheds experiencing such levels of erosion. Such results validate breeding efforts targeted for a scenario in which farmers adopt perennial grains in a system receiving two revenue streams of both grain yield and EQIP payments, in this region.

Introduction

A foundational issue to the global food system is the unsustainability of current modes of food production, namely annual row crop agriculture. Arguably the most immediately concerning consequence of modern agriculture is soil erosion, with roughly one-third of the earth's arable land lost to erosion since the mid-20th century (Pimentel et al. 1995). Estimates of median rates of soil erosion are at 1.537 mm/year under conventional, tilled agriculture and .082

mm/year under no-till agriculture (Montgomery 2007). While this difference is the driving impetus behind the push for conservation tillage, both of these rates still far exceed median rates of soil formation at .017 mm/year. Thus, even no-till methods of farming are unsustainable and insufficient in ensuring continued food security. More radical shifts in modes of production are required to ensure agricultural resilience. There is particular room for improvement in Georgia, where from 1982-2012, the average annual rate of soil loss on cultivated cropland slightly increased from 5.6 to 5.7 tons/acre/year (USDA 2016).

One alternative option is perennial agriculture, particularly perennial grain agriculture. Perennial grain crops have the potential to both reduce soil erosion and regenerate soil health in soils degraded by tillage, along with a suite of other ecological benefits (Ryan et al. 2018). Establishment of such perennial grain systems requires development of both system "hardware" (perennial grain varieties) and "software" (agronomic and ecological systems facilitated by the perennial germplasm) (Cox et al. 2013). This development is often hampered by a chicken-or-egg dilemma, in which breeders desire input from field-level assessments, but such field level assessments cannot be performed in the absence of the perennial grain germplasm. Thus, in the absence of such germplasm to perform field level assessments, modelling can be an important tool in simulating the agronomic potential of these novel varieties.

Conway et al.'s efforts in identifying perennial sorghum breeding yield targets through an economic analysis of Georgia sorghum farm budgets highlighted a potential lower breeding target for a dual-use in system in which perennial grain sorghum is adopted for both grain and soil conservation purposes (2020). This scenario is contingent on a farmer receiving Natural Resources Conservation Service (NRCS) Environmental Quality Incentives Program (EQIP) payments in year one of plant stand establishment. NRCS provides financial and technical support to farmers and ranchers in the voluntary employment of conservation practices, with EQIP being the agency's most frequently utilized conservation program (nrcs.usda.gov). Eligibility for payments like EQIP can inform the viability of breeding targets for a scenario in which perennial sorghum is adopted for dual-use grain and soil conservation purposes, hence referred to as a "Grains for Conservation" scenario. Such ecosystem service payouts to farmers may prove critical to perennial grain adoption, as incentive payments have previously been shown to encourage farmer employment of conservation practices (Cooper and Keim 1996).

While determining eligibility for such payments is a fairly complicated process involving age of farm, best conservation practice for a given piece of land, socioeconomic factors, and more (nrcs.usda.gov), the metric of "2xT" is frequently used as shorthand in classifying land as meeting the need for "substantial reduction in erosion" (NSAC, 2018). This statutory language of "substantial reduction" equivocates to potentially suitable for EQIP, while the "2xT" represents twice the amount of soil loss tolerance, or T-value, for a specified soil (Skidmore 1982). Essentially, if a piece of land is undergoing erosion at twice the rate of the soils inherent capacity to regenerate itself under current management practices, it can roughly be considered eligible for EQIP payments. Therefore, methods of analysis that highlight current soil erosion levels relative to that same soil's T-value can be used to indicate NRCS EQIP eligibility.

One such tool is the Revised Universal Soil Loss Equation (RUSLE), an internationally accepted standard for measuring soil erosion caused by rainfall and the associated overland flow. RUSLE is a frequently implemented model in GIS to estimate soil erosion (Fu et al. 2005; Lu et al. 2004; Prasannakumar et al. 2012; Shi et al. 2004). Particularly pertinent to research here, RUSLE-based simulations of shifts to perennial planting schemes have indicated significant reduction in soil erosion potential in Southern Tuscany (Vallebona et al. 2016). The growth in publicly available GIS data in recent years has made implementation of RUSLE-based analysis in GIS increasingly accessible. Ultimately, RUSLE-based GIS models can indicate farmer eligibility for conservation payments in north Georgia, which then informs the viability of devising breeding targets around such a scenario.

While there has been much conjecture as to the potential of subsidies for agro-ecological production practices to facilitate conversion from annual to perennial cropping systems (Crews et al. 2018; Duchene et al. 2019), there has been little research in this space. Attempts to deduce applicability of current conservation payments for a specific region can be enlightening when deciphering which areas may be ripe for perennial grain adoption. Additionally, while research here is particularly concerned with perennial grain sorghum, such investigations would be applicable to any perennial grain or oilseed variety. Although it is appealing to attempt to deduce the regional soil erosion reduction potential given perennial grain adoption, such research would be predicated on inexact assumptions of erosion under a perennial grain variety on a particular soil. Further agronomic work can facilitate future research in this area, perhaps elucidating how year of stand establishment, intercropping, or prescribed lifespan of a perennial grain system may impact levels of erosion. In the meantime, it is possible to use erosion levels under current management practices as an indicator of which land has the most to gain from perennial grain adoption.

Methods

This research aimed to elucidate soil erosion levels relative to their respective T-values in two north Georgia watersheds. Bartow County (Figure 4.1) was chosen due to its incidence of development, high quality farmland, and proximity to Lake Allatoona, a body of water threatened by excessive algal growth partly due to agricultural runoff (Lin et al. 2009). This confluence of factors suggested that the region might be particularly well suited for water-quality improving soil conservation practices such as perennial cropping schemes.

The research area was further pared down to two watersheds within Bartow County (Figure 4.2). These two watersheds were chosen due to their relatively high incidence of agricultural land use. This was deduced through examining land cover data from Cropscape NASS (National Agricultural Statistics Service) CDL (cropland data layer). Other watersheds in the county did not have enough of an agricultural presence to warrant investigation. Watershed data was obtained from U.S. Geological Survey's (USGS) National Hydrography Dataset (NHD) at the ten-digit hydrologic unit (HU) scale. This scale is generally referred to as the "watershed" scale, which falls below the "subbasin" classification and above the "subwatershed" classification, and is well suited for this analysis as it closely mirrors the maximum spatial extent of much available data needed for RUSLE.

RUSLE

The calculation of soil erosion under the two cropping scenarios was carried out in ArcMap using the Revised Universal Soil Loss Equation (RUSLE), which measures long term average annual soil erosion caused by rainfall (Renard et al. 1991; Renard et al. 1997). Versions of this model have been used to estimate erosion for decades and it is widely utilized by government agencies throughout the world.

The RUSLE model rests on the equation of:

$$A = R \times K \times LS \times C \times P$$

Where A= mean annual soil loss (tons/acre/year), R=rainfall and runoff erosivity factor, K=soil erodibility factor, LS= slope length factor, C= cropping-management factor, and P = support practice factor (Renard et al. 1991).

R-factor

R, the rainfall erosivity factor, measures the erosive potential of rain for a given place. This value is calculated as the average annual summation of a location's EI values, with E being the kinetic energy of the storm and I representing the maximum 30-minute intensity of the storm (USDA 2001). R-factor values for the watershed were estimated using the EPA's R-factor evaluation tool. Numerous locations within the county returned the same R-factor value of 304, thus the R-factor was assumed to be a constant value of 304 for the entire watershed.

K-factor

K, the soil erodibility factor, is a measure of the susceptibility of a given soil to erosion and its rate of runoff (USDA 2001). This is informed by the soil type, organic matter content, and former management practices. K-factor values were taken from the USGS web soil survey and data was courtesy of USDA NRCS, September 2019. Soil survey data was in a 30m resolution, vector format, and WGS 1984 projection. These values were transformed to a UTM 16N projection and data was converted to a raster format in order to facilitate future data manipulations. K-factor values for the watershed ranged from .05 to .49, with higher values indicating a higher susceptibility to erosion (Figure 4.3).

LS-factor

LS, the slope length factor, is a ratio of soil loss from a given slope length relative to a 72.6 foot slope length on the same soil type and gradient (USDA 2001). The slope length factor is a function of both flow accumulation and slope. First a digital elevation model (DEM) was constructed for the watershed. Elevation data was procured from ASTER, which dated to October 2011. Elevation data was in 30m resolution and WGS 1984 projection, which was transformed into a UTM 16N projection (Figure 4.4).

Next, a depressionless DEM had to be constructed to enable future watershed flow calculations (Pelton et al. 2012). This involved creating a flow direction raster from the DEM map layer and then applying the “sink” tool to create a raster of sinks in the elevation model. The “watershed” tool was then implemented on the sink raster to identify the contributing points for each sink. Then the “zonal fill” tool was used to identify the watershed’s maximum and minimum elevations, the minimum values were then subtracted from the maximum values to identify the watershed’s sink depth or z-limit. This z-limit can then be entered into ArcMap’s “fill” tool, which is applied to the original DEM, the resulting map layer is a depressionless DEM. This DEM was then utilized in the creation of a flow direction map (Figure 4.5) and subsequent flow accumulation map (Figure 4.6). This DEM was also used as the basis for slope calculations, using the slope tool in ArcMap, which resulted in a slope raster for the watershed in degrees (Figure 4.7).

Map algebra was then used to calculate slope length factor using the following equation based on a method described in Kim (2014):

Power(facc * cell resolution / 22.1, 0.4) * Power(Sin(slp * 0.01745) / 0.09, 1.4) * 1.4

Here, “facc” represents the flow accumulation map layer and “slp” represents the slope map layer. Cell resolution in this case was 30, due to the 30m projection of the elevation data. The resulting slope length factor (LS) map layer can be seen in Figure 4.8.

C-factor

C, cover-management factor, reflects the effect of a cropping system and management on soil erosion. C-values are relative to a standard of a clean-tilled, continuously fallow land management practice, which higher values equivocating to more erosion. First, land cover for the watershed was determined using Cropscape NASS CDL data. This data is from 2017, in a raster format, 30m resolution, and was projected in UTM 16N (Figure 4.9). Once land cover for the watershed was established, this data was summarized and exported into a .csv file. From there, crop C-values were taken from NRCS for the region (CMZ 66) for the various land cover features, added to the .csv file, uploaded to ArcMap, and joined to the landcover dataset. C-values were established for crops assuming single-cropping systems with tillage and averaged over various planting times (early and late). Any values not available from NRCS were taken from the literature (Kim 2014; Panagos et al. 2015). This tended to be for non-crop land cover such as developed land or forest cover. While literature-derived values may be less precise for this location as compared to the NRCS data, non-crop land was not considered in the final analysis of potential suitability for NRCS EQIP payments. Figure 4.10 displays the C-factor values under the current land cover.

P-factor

P, support practice factor, measures the ratio of soil loss with contouring and/or strip cropping relative to row farming up and down a slope (USDA 2001). There was not sufficient data to accurately map P-factor values for this region, thus they were assumed to be equal to 1, which represents straight-row farming. Setting the P-value to a constant of 1 is a common practice across the literature (Kim 2014; Vallebona et al. 2016).

T-factor

T, soil loss tolerance, is the average annual soil loss that can occur with minimal or no long-term degradation expressed in tons/acre/year. T-values can range from 1-5, with typical values occurring in the 3-5 range. T-values were obtained from the USGS web soil survey and data was courtesy of USDA NRCS, September 2019. Soil survey data was in a 30m resolution, vector format, and WGS 1984 projection. These values were transformed to a UTM 16N projection and data was converted to a raster format in order to facilitate future data manipulations (Figure 4.11). While T-values are not a factor in the RUSLE equation, it is a useful basis of comparison to the resulting value obtained from RUSLE. The output of RUSLE is an A-value, which represents a given soil's annual erosion in tons/acre/year. Thus A-values in excess of a soil's inherent T-value represents unsustainable erosion levels, whereas an A-value greater than or equal to twice the amount of a soil's T-value might indicate eligibility for NRCS EQIP payments.

A-factor

Map algebra was used to multiply the R, K, LS, C, and P factors together to obtain an A value map layer of soil loss in tons/acre/year. This was done for each of the two watersheds. Extreme upper outliers accounting for less than .1% of the data were removed from both A-value data sets. These data points were drastically outside three standard deviations from the mean value, and significantly skewed average A-value results for the watershed. It is assumed that these outliers stemmed from artifacts or insufficient resolution in the ASTER elevation data, as this was the map layer that contained the widest spread of variability.

The T-factor map layer was doubled to obtain a 2xT map layer for each watershed. These two map layers were then subtracted from their respective watershed's A-value map layer to produce an erosion exceeding soil loss tolerance (2xT) map layer (Figure 4.13). Next, these map layers were delineated into a binary yes/no layer, with yes being attributed to values of zero or greater and no being attributed to any negative value. In this case, "yes" meaning land with erosion levels equal than or greater to 2xT and "no" meaning land with erosion levels less than 2xT. These map layers were converted from raster to polygon to facilitate future analyses.

The land cover (C-factor) map data layer was duplicated and converted into a binary cropland/non-cropland cover for each of the two watersheds (Figure 4.12). This layer was also then converted from raster to polygon. The intersect tool was deployed to calculate the intersection of cropland and soil erosion greater than or equal to 2xT for each watershed (Figure 4.14, Figure 4.15). The tabulated outcome could then be used to calculate the current area of land under crop cover that is meeting or exceeding erosion levels of 2xT.

Results

K-factor

K-values (soil erodibility factor) for the two Bartow watersheds ranged from 0.05-0.49 (Figure 4.3). Higher values indicate greater susceptibility to erosion, which is indicated on the map with the darker shades of blue. Watershed three, the eastern watershed, shows greater variability in K-value as compared to watershed four, as well as a consistent section of high k-values surrounding the Etowah River, that cuts east-west across the county. K-values exceeding .40 are generally considered to be high (Michigan State University 2002).

LS-factor

LS-values (slope length factor) ranged from 0-55 in Bartow County (Figure 4.8). Some of the starkest changes in elevation for the county (Figure 4.4) occur outside the subset of two watersheds investigated here. LS-value map does not highlight particular areas for concern within the study area.

C-factor

Land cover types, acreage, and associated C-values (cover-management factor) for Bartow County can be found in Table 4.1. C-values for the study area spanned the whole potential range of 0.0-1.0, with higher values indicating higher susceptibility to erosion. Land cover for the two watersheds investigated here shows a concentration of agriculture cover surrounding the Etowah River in watershed three (Figure 4.9), which corresponds to relatively higher C-values (Figure 4.10). As previously mentioned, this area also has relatively higher K-values, suggesting that this region may be more prone to higher erosion rates.

T-factor

T-values (soil loss tolerance) for the study region spanned across all potential values, from 1-5 (Figure 4.11). T-values are the one metric in which higher values are better, as they indicate a higher erosion tolerance. Most of the study area, and particularly the regions with a higher incidence of agricultural land use, had relatively higher T-values of 4-5, indicating greater tolerance for the more highly erosive activities occurring there.

A-factor

A-values (soil loss in tons/acre/year) equal to or exceeding 2xT (Figure 4.13) closely mirror regions in which land is currently under crop cover (Figure 4.12). In watershed three (Figure 4.15), 40.98% of land under crop cover had erosion levels meeting or exceeding 2xT, while only 3.34% of land not under crop cover had such erosion levels (Table 4.2). Similarly, in watershed four (Figure 4.14), 41.66% of land under crop cover had erosion levels meeting or exceeding 2xT, while 4.31% of land not under crop cover met such erosion levels (Table 4.3). Missing data in Tables 4.2 and 4.3 is attributed to lack of K-value data (Figure 4.3) for highly developed areas of the watershed. This missing factor prohibits the calculation of A-values for that area.

Discussion

The results here highlight a relatively high presence of soil erosion occurring at a rate greater than or equal to 2xT, with approximately 41.66% and 40.98% of land currently under crop cover in the two watersheds experiencing such levels of erosion. While this does not precisely equivocate to NRCS EQIP payment eligibility, it can provide a rough estimation of such eligibility. Thus, the high incidence of erosivity illuminated in these two watersheds supports the practicality of breeding efforts targeted for a “Grains for Conservation” scenario. Such a scenario allows for a lower yield of perennial sorghum to be economically viable, due to the supplemental income source of NRCS EQIP payments in year one of the establishment of the plant stand. It appears that there would be a sufficient market of farmers to take on perennial grain crops in such a manner, warranting the breeding yield targets for such a scenario.

In this way, RUSLE can function as a useful tool in identifying areas susceptible to erosion that would most benefit from a perennial grain cropping system. This could potentially be a very useful resource to prospective farmers considering perennial grains. It would be effective to add an additional layer to this equation which represents potential soil conservation payments. Farmers could then use those numbers to help further inform their decision-making process, as a switch to perennial crops will most likely involve some trade-off in yield.

Future avenues of investigation include evaluating the potential erosion reduction when implementing a perennial cropping system, as in Vallebona et al. (2016). Critical to this analysis would be a precise C-value for the perennial grain variety in question. This might vary across the crop, length of plant stand, and year of plant stand establishment. The analysis would also rest on assumptions of modes of perennial grain adoption. While a model assuming full perennial cover would be simple to implement, this is not likely to be the way in which perennial grains are adopted. It is much more likely that they will be a portion of a mixed system, and further analyses under various mixed cropping schemes would be a very useful complementation to future work.

RUSLE is a widely-used model in part because of its relative simplicity, but this means that the quality of the input data is critical. For precise assessment of potential erosion savings at an individual farm scale, higher resolution soil and elevation data would be required. Additional considerations such as support practice factor, P, and precise C-factor values for the exact management type (influenced by tillage practice employed, planting time, harvest time, and crop rotation), could shift the numbers away from what is modeled here, making a perennial system more or less attractive to a given farmer

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Table 4.1: Bartow County land cover and associated C-values (cover management factor).

Land Cover	Acreage	% land cover	C-val
Deciduous Forest	106641.1	35.45%	0.003
Evergreen Forest	54345.9	18.07%	0.003
Developed/Open Space	30968.7	10.29%	0.003
Grass/Pasture	26420	8.78%	0.013
Shrubland	25087.5	8.34%	0.009
Other Hay/Non Alfalfa	15920.8	5.29%	0.34
Developed/Low Intensity	11847.6	3.94%	0.013
Open Water	7295.7	2.43%	0
Mixed Forest	6479.2	2.15%	0.003
Developed/Medium Intensity	3563.2	1.18%	0.2
Soybeans	2981.9	0.99%	0.23
Corn	2936.5	0.98%	0.24
Cotton	1959.3	0.65%	0.37
Developed/High Intensity	1797.6	0.60%	0.45
Barren	877.3	0.29%	1
Sod/Grass Seed	775.5	0.26%	0.013
Dbl Crop WinWht/Corn	413.9	0.14%	0.1
Woody Wetlands	208.4	0.07%	0.001
Sunflowers	90.1	0.03%	0.3
Winter Wheat	52.9	0.02%	0.25
Dbl Crop WinWht/Soybeans	65.4	0.02%	0.1
Fallow/Idle Cropland	57.4	0.02%	0.013
Herbaceous Wetlands	20.5	0.01%	0.003
Sorghum	2	0.00%	0.3
Peanuts	0.7	0.00%	0.3
Rye	0.4	0.00%	0.14
Millet	0.4	0.00%	0.14
Canola	1.1	0.00%	0.14
Peaches	13.1	0.00%	0.13
Apples	0.4	0.00%	0.13
Pecans	5.8	0.00%	0.13
Dbl Crop WinWht/Cotton	2.2	0.00%	0.1
Dbl Crop Soybeans/Oats	0.4	0.00%	0.1
Total	**300833**	**100%**	

Table 4.2: Bartow County watershed three, % of land cover type experiencing soil erosion at levels greater than or equal to 2xT (soil loss tolerance).

Land Cover	Erosion >= 2T	% Land Cover
Cropland	No	56.27
Cropland	Yes	40.98
Cropland	No data	2.75
Non-cropland	No	81.46
Non-cropland	Yes	3.34
Non-cropland	No data	15.20

Table 4.3: Bartow County watershed four, % of land cover type experiencing soil erosion at levels greater than or equal to 2xT (soil loss tolerance).

Land Cover	Erosion >= 2T	% Land Cover
Cropland	No	57.70
Cropland	Yes	41.66
Cropland	No data	0.64
Non-cropland	No	90.82
Non-cropland	Yes	4.31
Non-cropland	No data	4.87

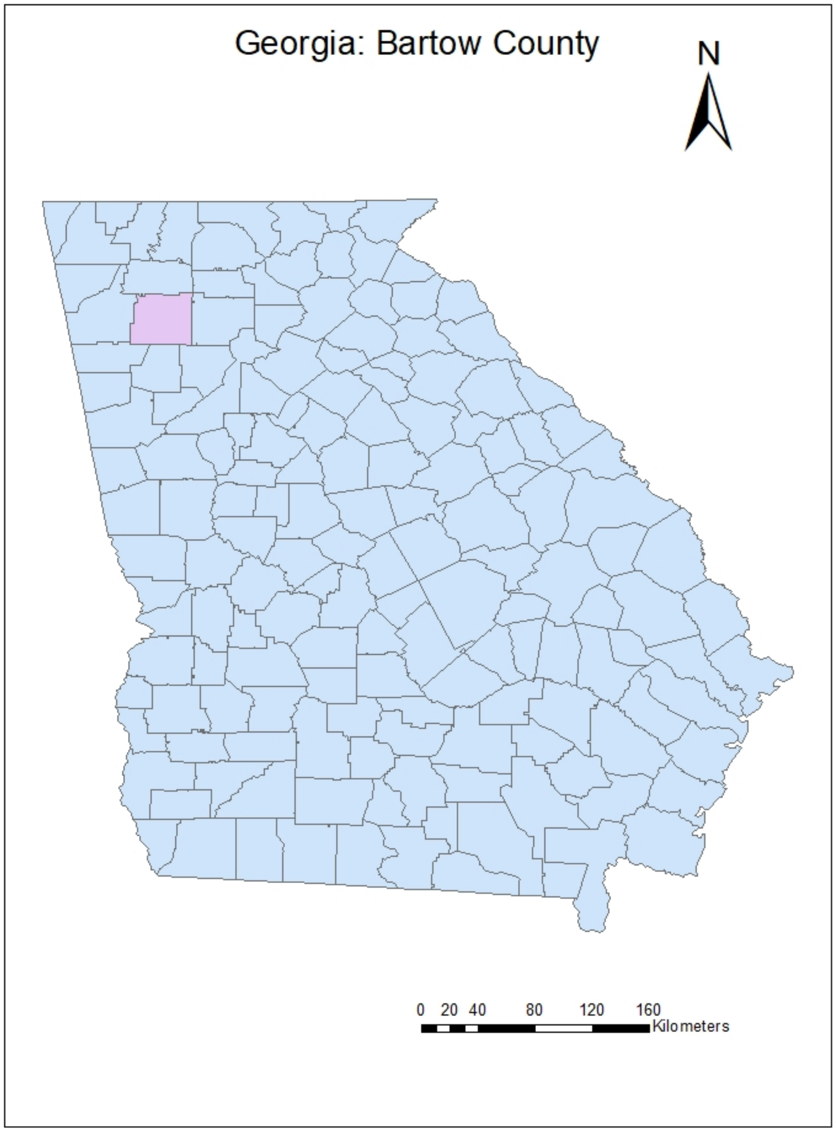

Figure 4.1. Bartow County, GA. Source: Atlanta Regional Commission (ARC), Tara M Conway, created 10/19/2019, data circa 2012, UTM 16N.

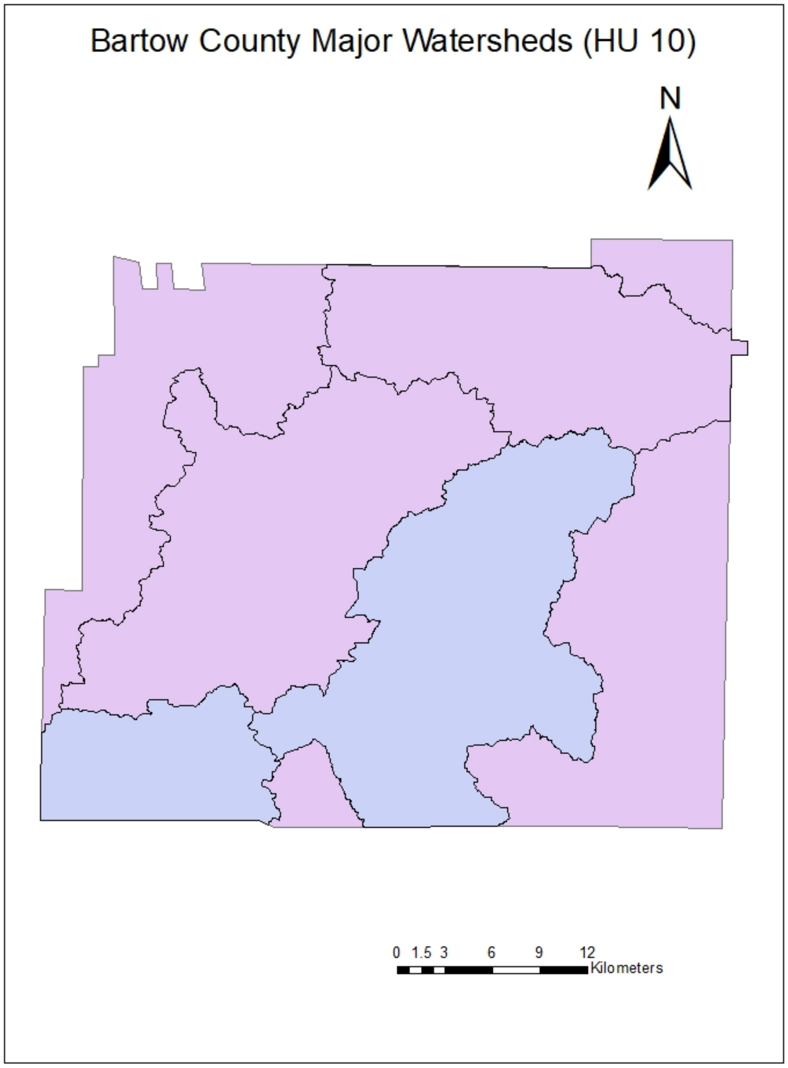

Figure 4.2. Watershed map (HU10) of Bartow county, GA. Research focus area of two watersheds highlighted in blue (watershed 3 and watershed 4). Source: USGS NHD, Tara M Conway, created 10/19/2019, data circa 11/2018, UTM 16N.

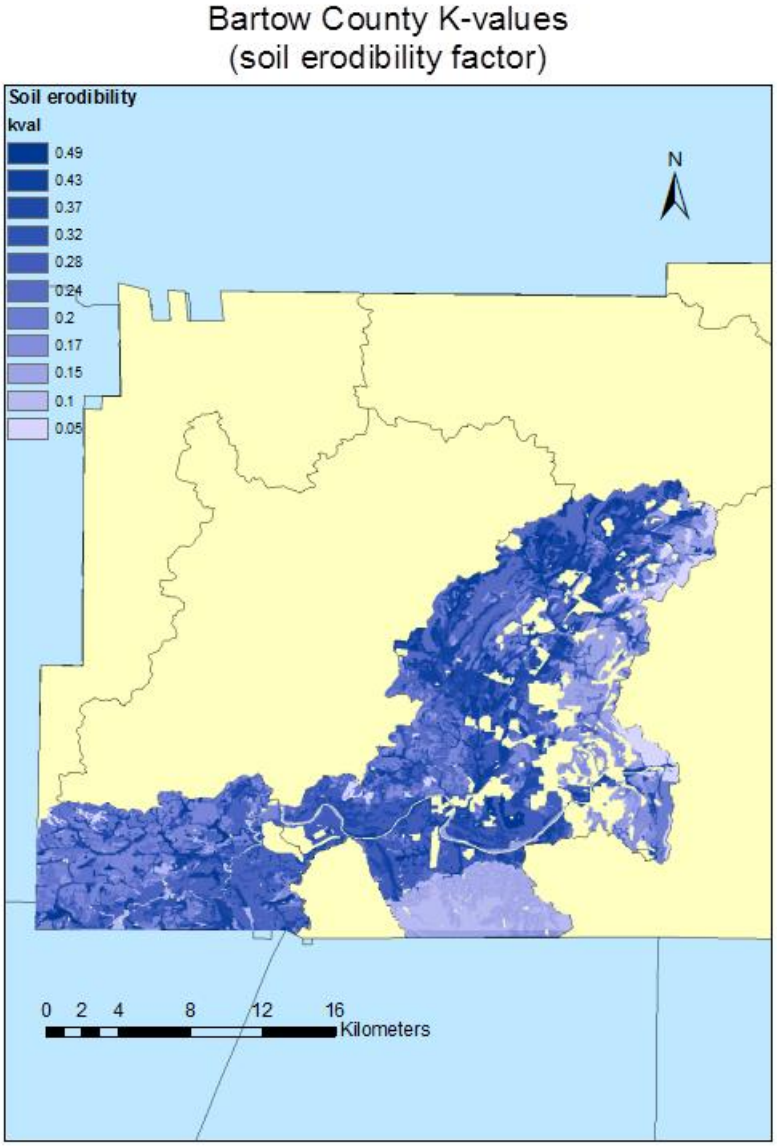

Figure 4.3. Soil K-factor values in two Bartow county, GA watersheds. Source: USDA NRCS, Tara M Conway, created 10/19/2019, data circa 09/2018, UTM 16N.

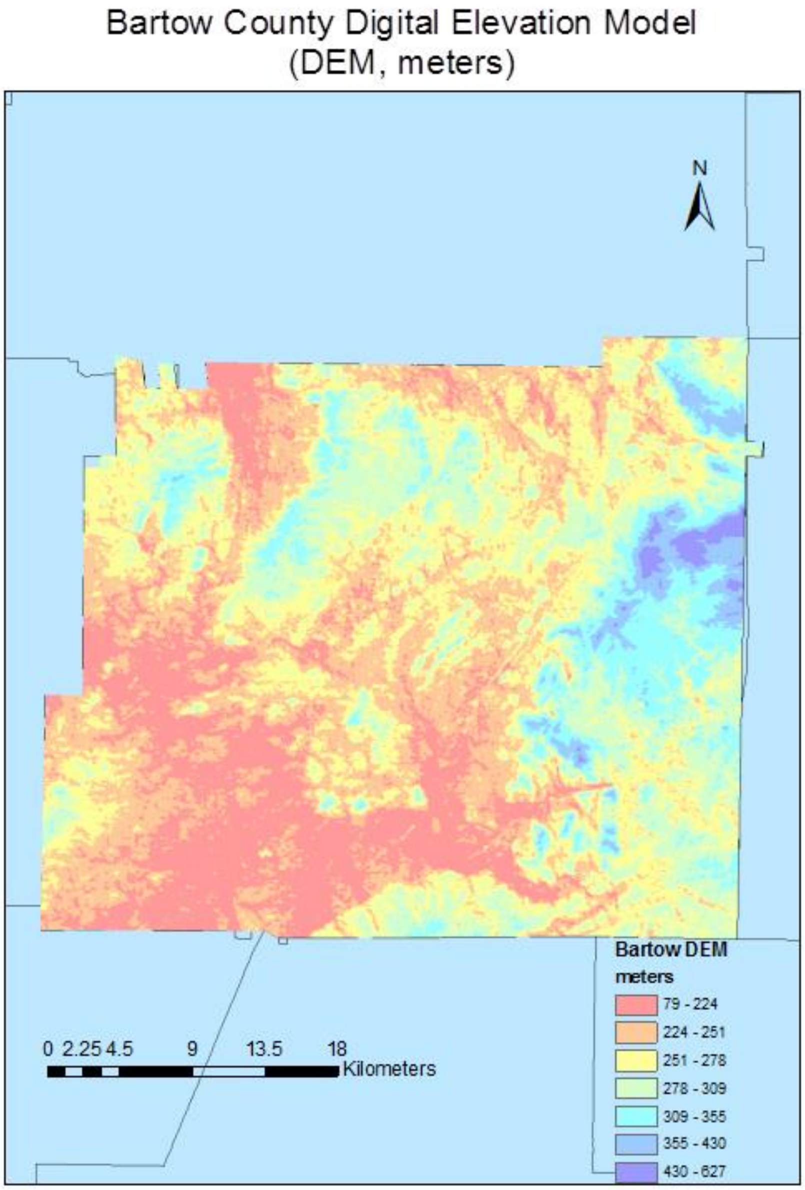

Figure 4.4. DEM of Bartow County, GA (meters). Source: USGS ASTER, Tara M Conway, created 10/19/2019, data circa 10/2011, UTM 16N.

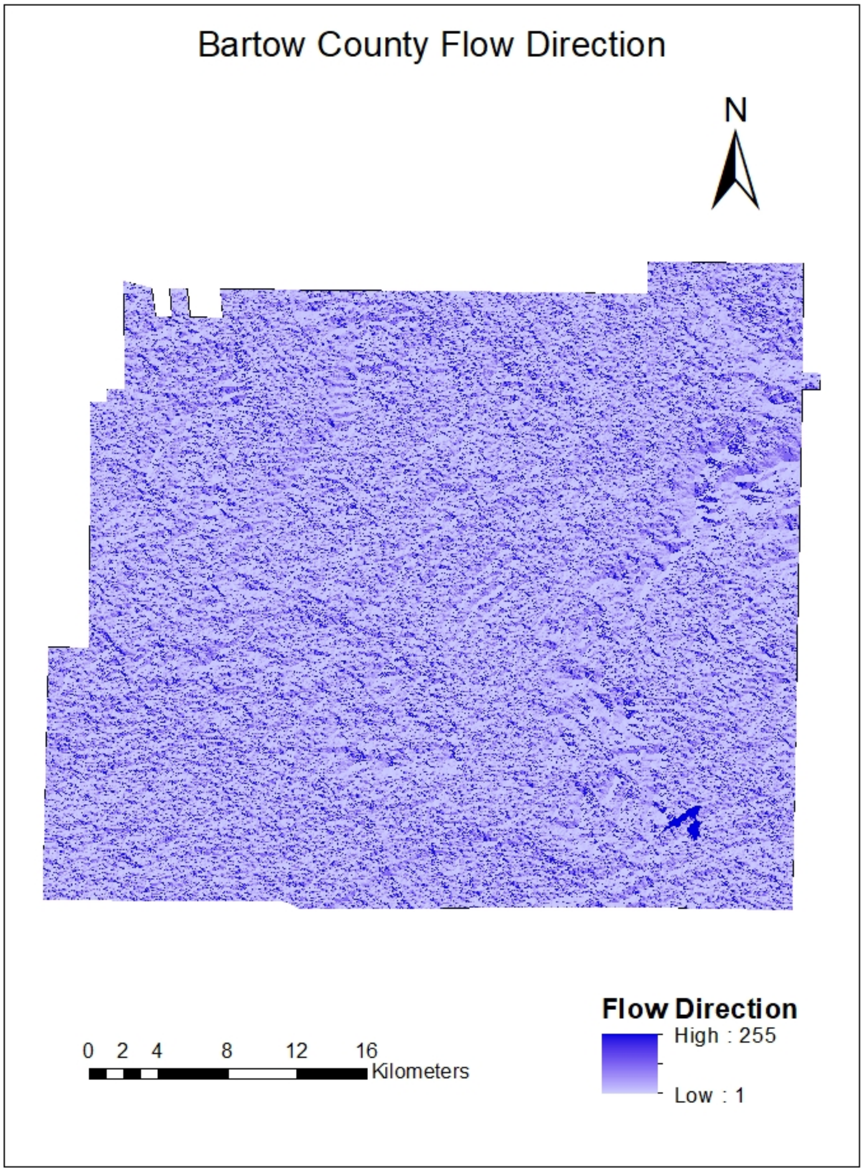

Figure 4.5. Flow direction of Bartow County, GA. Source: USGS ASTER, Tara M Conway, created 10/19/2019, data circa 10/2011, UTM 16N.

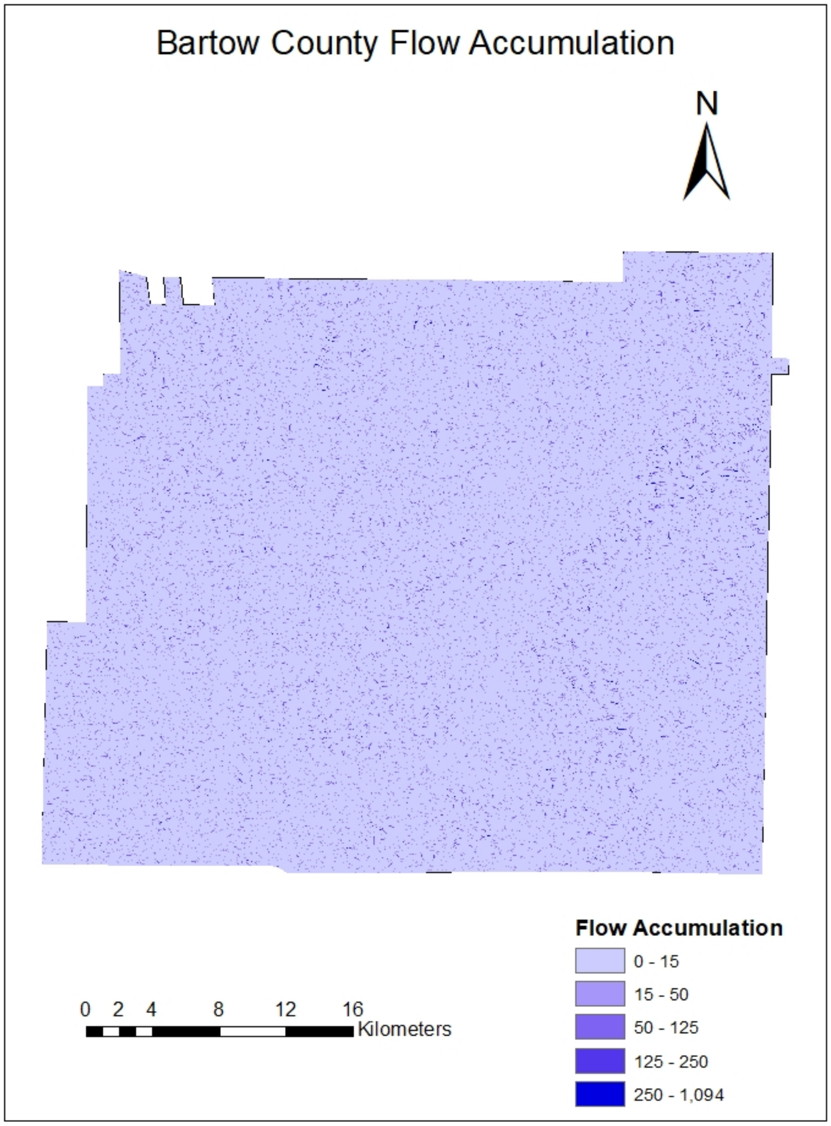

Figure 4.6. Flow accumulation of Bartow County, GA. Source: USGS ASTER, Tara M Conway, created 10/19/2019, data circa 10/2011, UTM 16N.

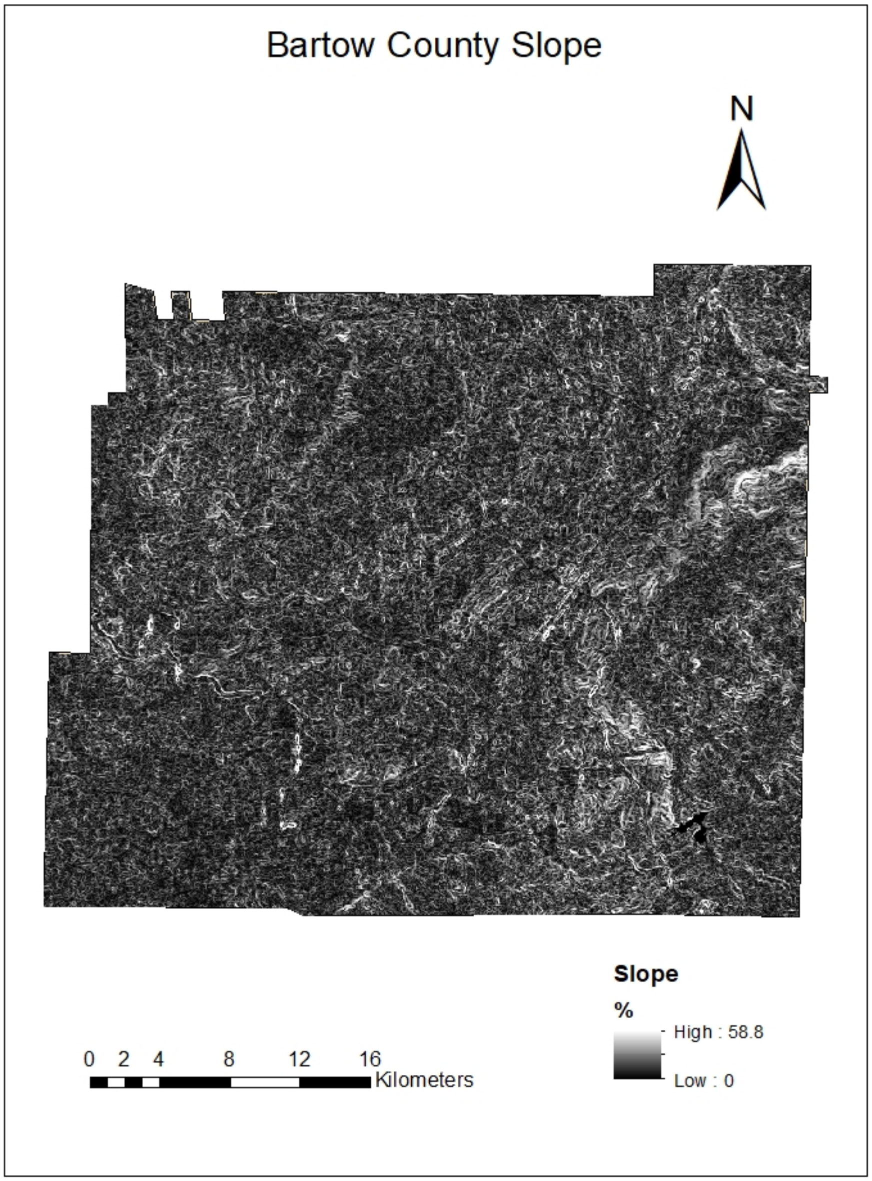

Figure 4.7 Slope (degrees) of Bartow County, GA. Source: USGS ASTER, Tara M Conway, created 10/19/2019, data circa 10/2011, UTM 16N.

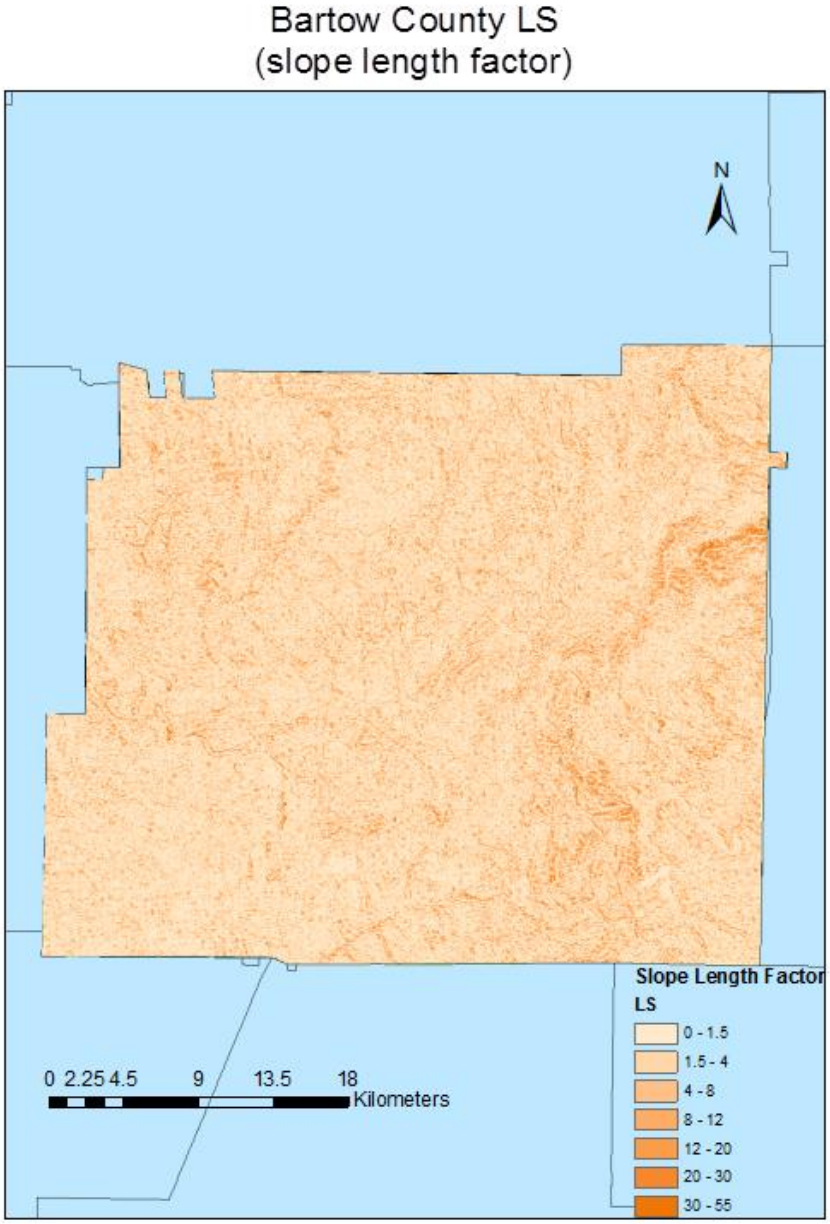

Figure 4.8. LS (slope length factor) of Bartow County, GA. Source: USGS ASTER, Tara M Conway, created 10/19/2019, data circa 10/2011, UTM 16N.

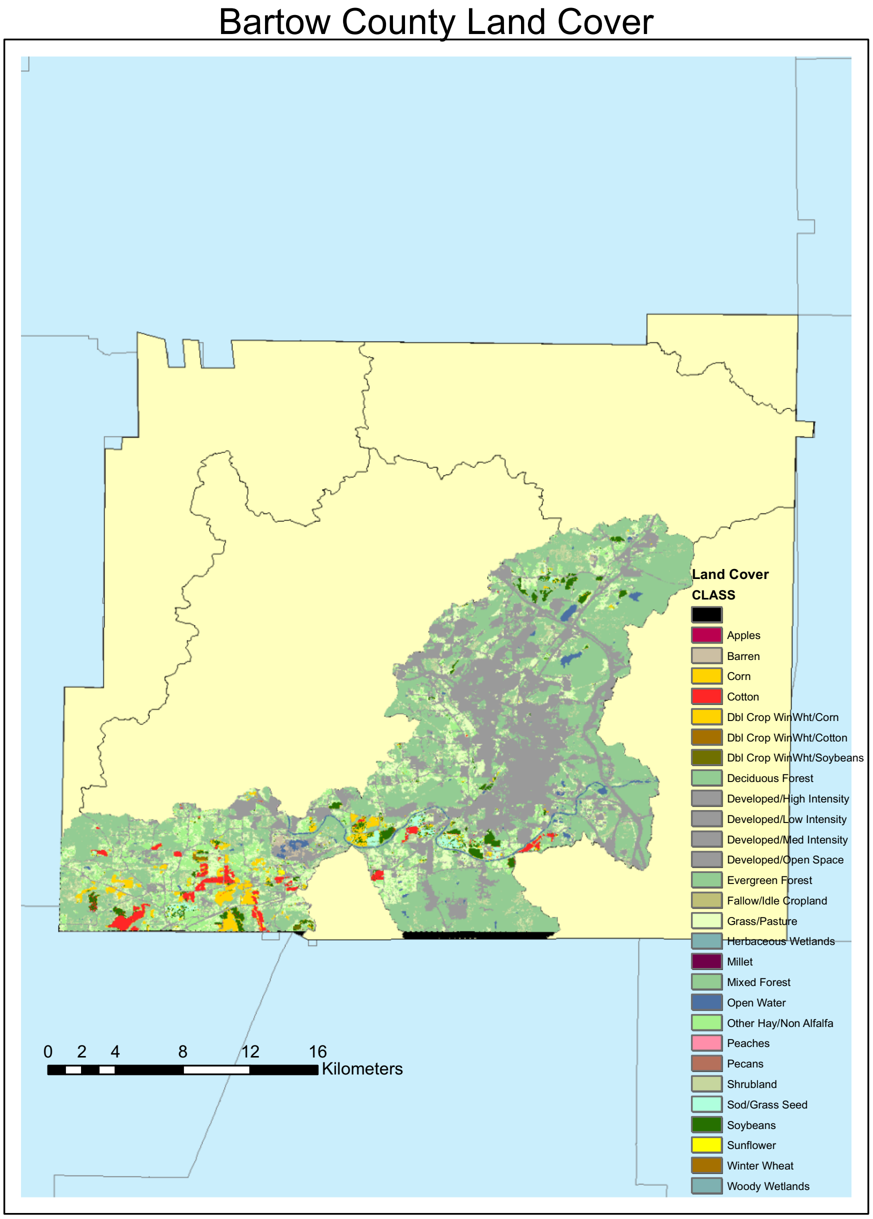

Figure 4.9. Current land cover of two Bartow County, GA watersheds. Source: NASS CDL, Tara M Conway, created 10/19/2019, data circa 2017, UTM 16N.

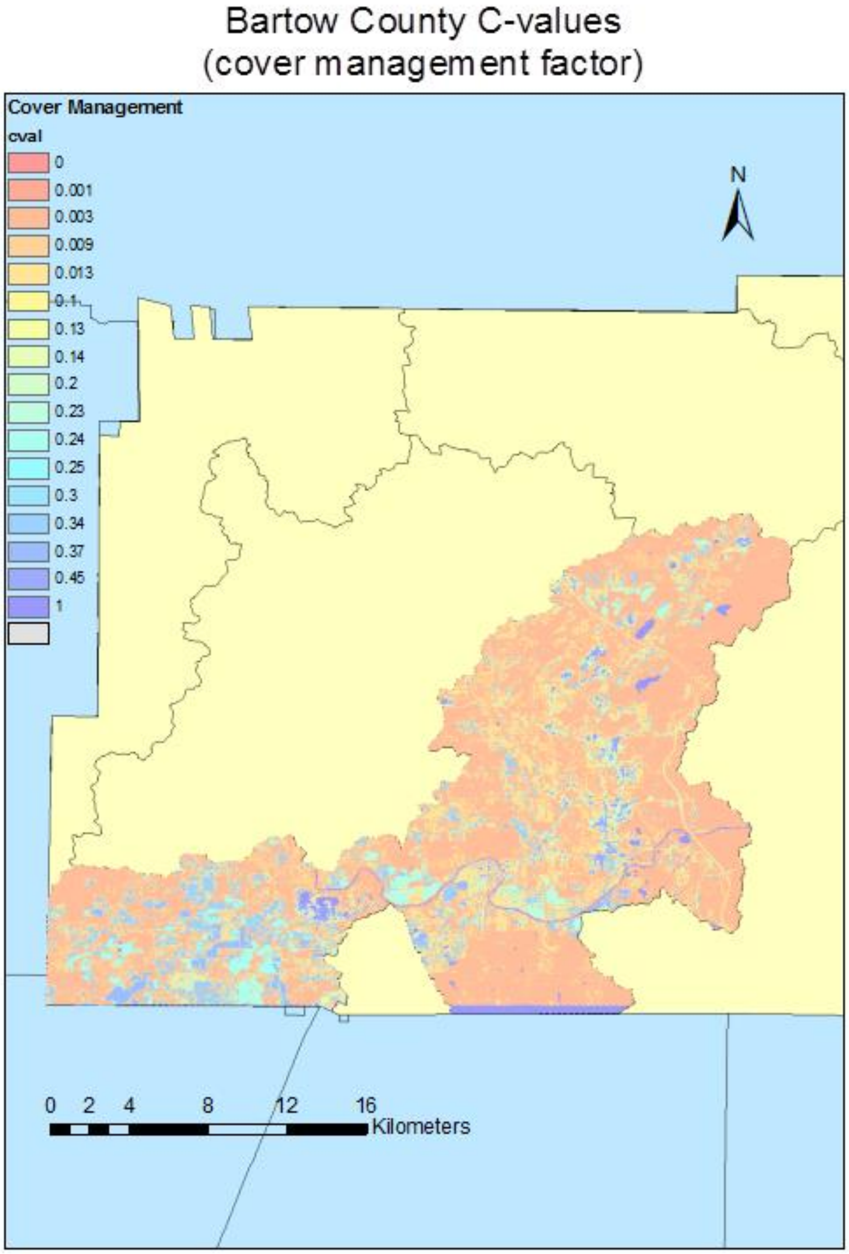

Figure 4.10. C-factor (cover-management) values of current land cover for two Madison County, GA watersheds. Source: NASS CDL, Tara M Conway, created 10/19/2019, data circa 2017, UTM 16N.

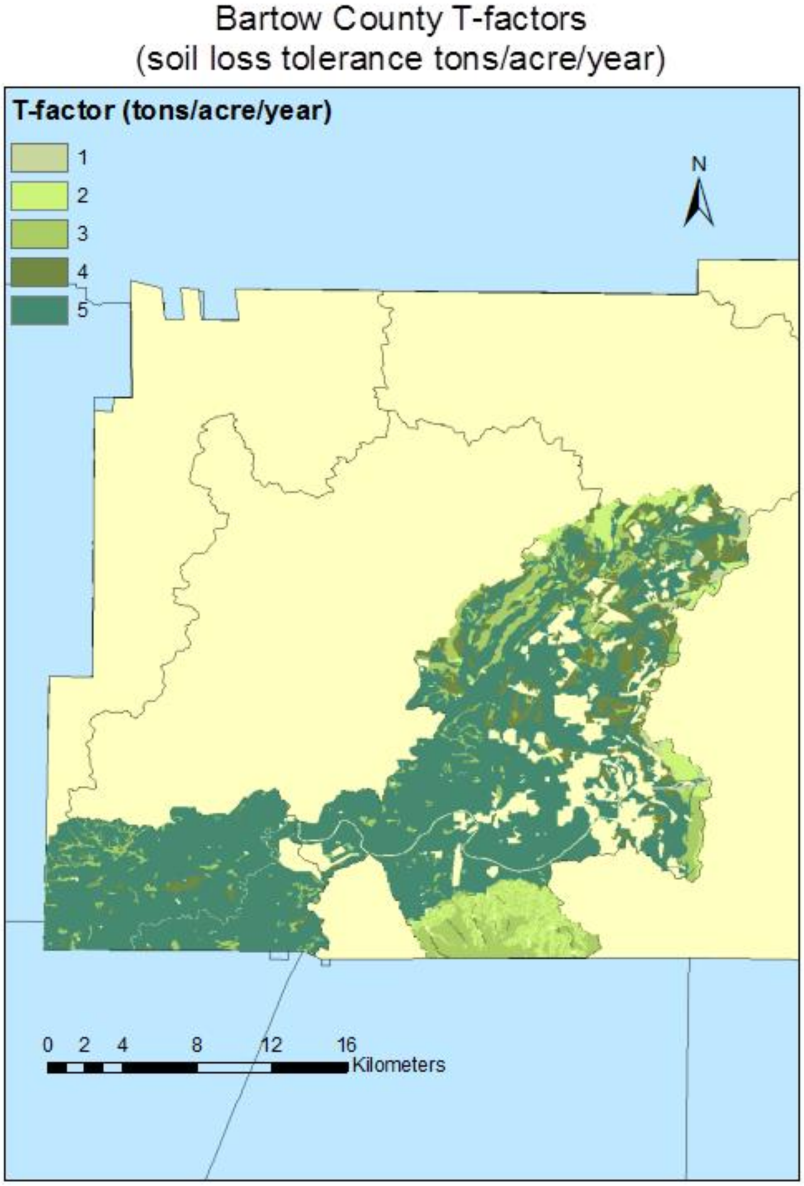

Figure 4.11. Soil T-factor values (soil loss tolerance) in two Bartow county, GA watersheds.

Source: USDA NRCS, Tara M Conway, created 10/19/2019, data circa 09/2018, UTM 16N.

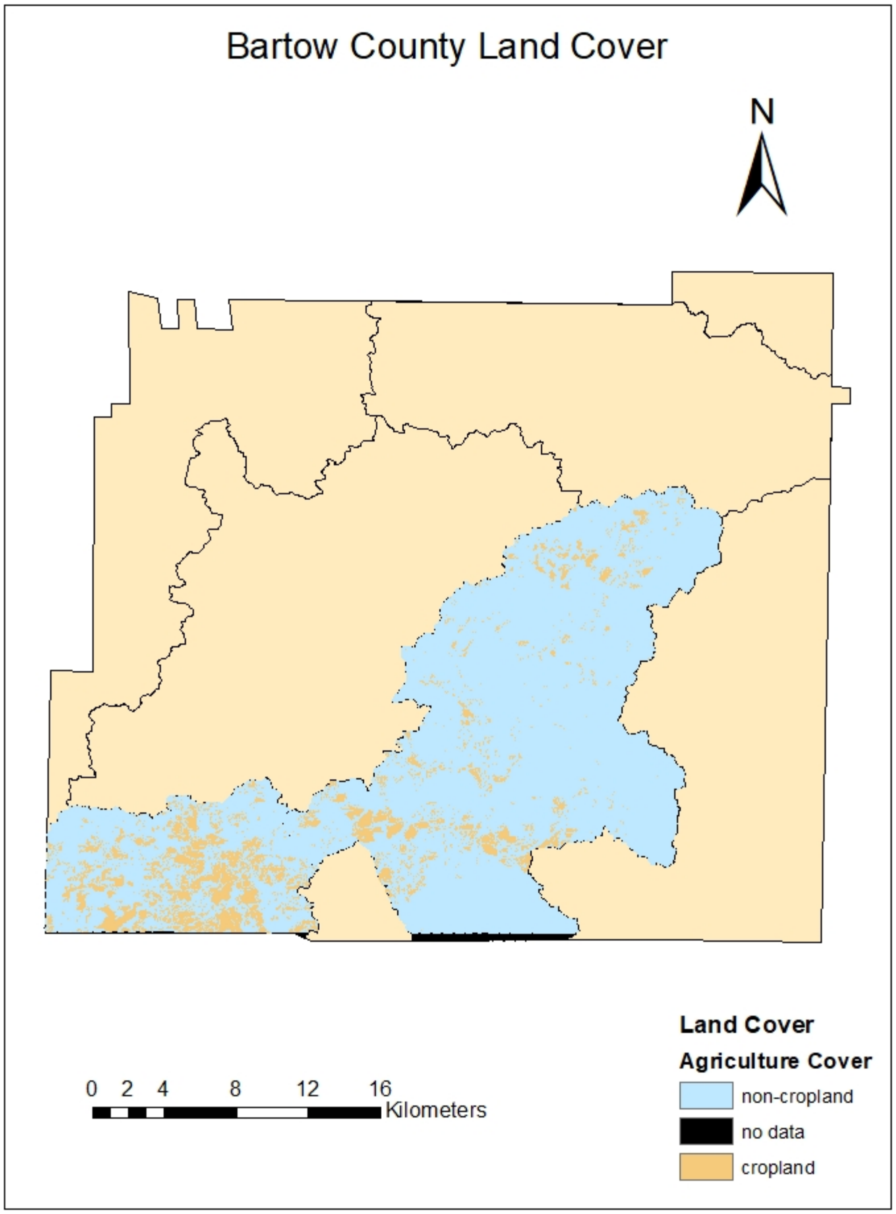

Figure 4.12. Land cover type (cropland vs non-cropland) in two Bartow county, GA watersheds. Tara M Conway, created 10/19/2019, UTM 16N.

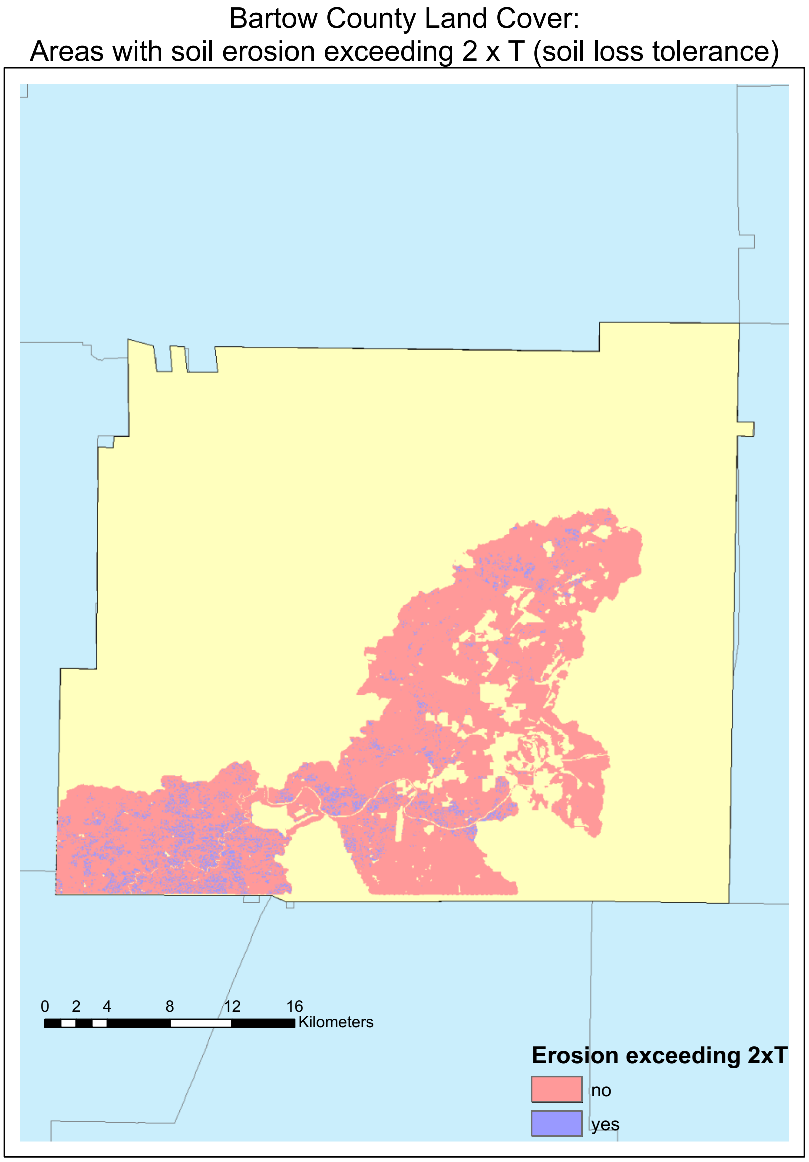

Figure 4.13. Areas in two Bartow county, GA watersheds with soil erosion greater than or equal to 2xT (soil loss tolerance). Tara M Conway, created 10/19/2019, UTM 16N.

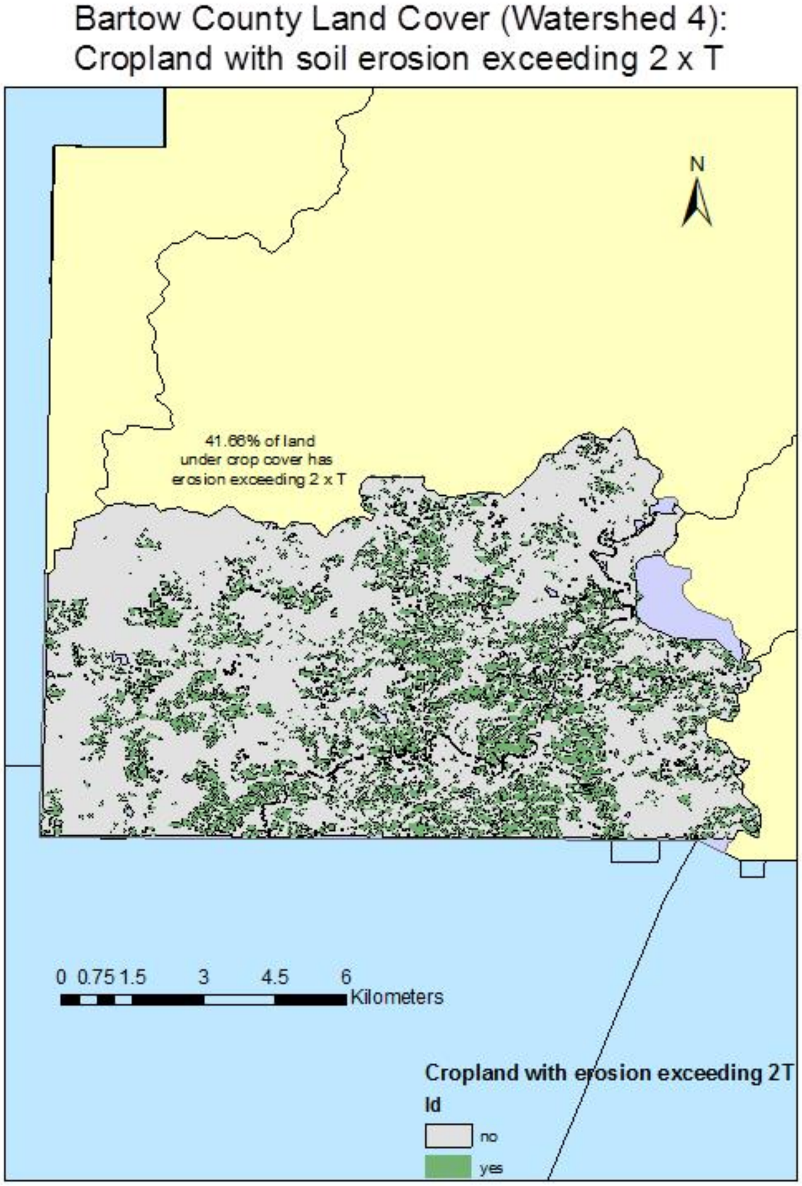

Figure 4.14. Cropland in Bartow county, GA watershed 4 experiencing soil erosion greater than or equal to 2xT (soil loss tolerance). Tara M Conway, created 10/19/2019, UTM 16N.

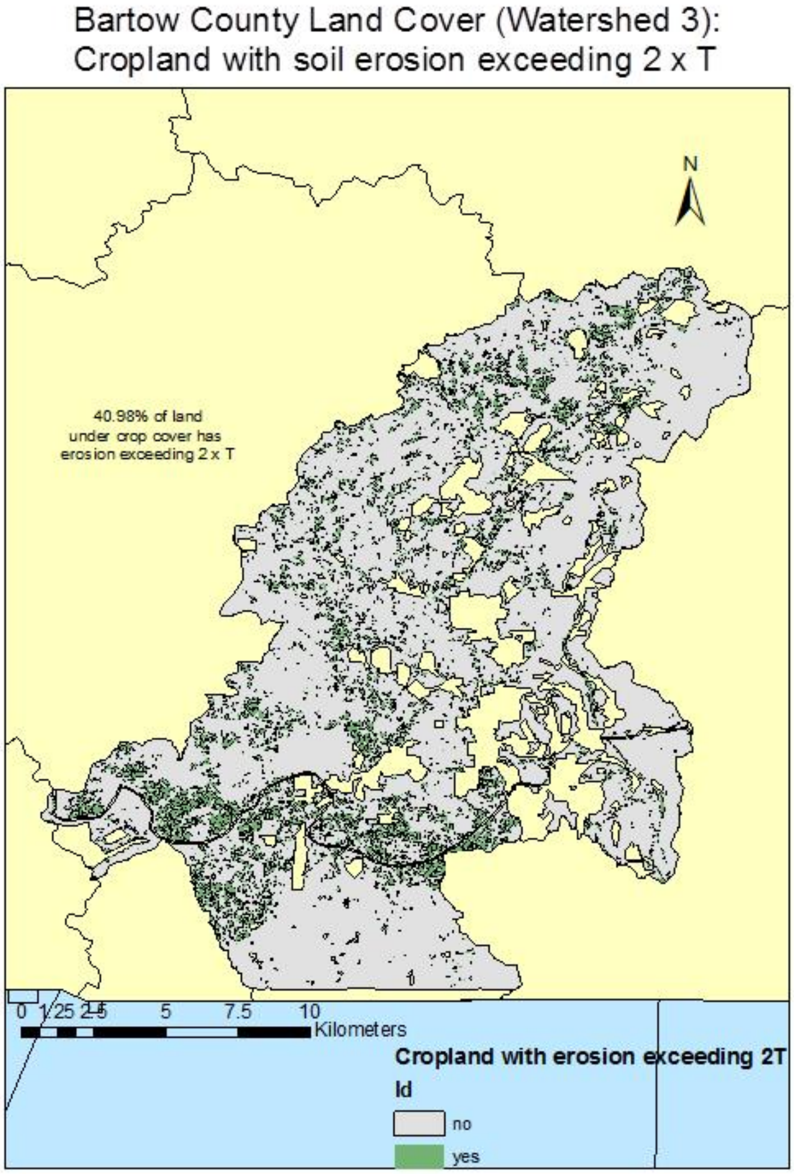

Figure 4.15. Cropland in Bartow county, GA watershed 3 experiencing soil erosion greater than or equal to 2xT (soil loss tolerance). Tara M Conway, created 10/19/2019, UTM 16N.

CHAPTER 5

CONCLUSION

Research here investigated components of both perennial grain sorghum "hardware" and "software" development. The process highlighted the permeability of such designations, with "hardware" results informing "software" development and vice versa. While perennial germplasm is critical to perennial grain system establishment, its availability does not beget farmer adoption and is thus of little practical value without consideration of the germplasm's ecological, economic, and social context. Investigations here brought valuable information to the fore, including: genomic regions pertinent to rhizome development and overwintering in sorghum, the capacity of *S. halepense* to improve *S. bicolor* varieties, the potential to breed perennial sorghum varieties with both increased yield and capacity to overwinter, economic viability of dual-use perennial sorghum systems in Georgia at yields significantly lower than current annual yields, the importance of breeding perennial sorghum for marginal environments, increased adoption potential of perennial sorghum varieties without prussic acid, and sufficient NRCS EQIP eligibility in north Georgia to warrant breeding targets for dual-use grain and soil conservation systems. These tangible findings perhaps obscure an equally important result, which is that an interdisciplinary approach to perennial sorghum system establishment in Georgia permits better, more complex questions to be asked. Approaching the problem from an economic perspective informs broader investigations of the sorghum genome, while genetic results inform the ability to perform certain system economic analyses that were perhaps previously inconceivable. Similarly, investigations into payments for environmental services highlight critical ecological functions for a plant breeder as well as the spatial extent to which such varieties may be adopted. Going forward, the findings of plant breeders and agronomists may inform new policies that offer incentives for the environmental services provided by novel varieties. Thus, the development of novel growing systems benefits from a degree of systems thinking, with only a small portion of the innumerable agroecological questions addressed here. Findings here provide support for the efficacy of a transdisciplinary approach in perennial system establishment, especially pertinent when timeliness is critical to realizing environmental benefits.