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# COBERTURA VEGETAL DEL SUELO Y RIEGO DEFICITARIO PARA MAXIMIZAR EL CONTROL DE LA DISPONIBILIDAD HÍDRICA DEL VIÑEDO EN CLIMA HÚMEDO

**por**

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RESUMEN

El estudio se orienta a la mejorar de la calidad y regularidad del producto vino Tannat, a través de un manejo del suelo orientado a aumentar el control potencial de la disponibilidad hídrica del viñedo. Se plantea un cambio en la "lógica productiva actual", donde en una viticultura en secano, se busca preservar el agua del suelo mediante la eliminación de competencia a través de la aplicación de herbicidas bajo la fila de plantación (H), aceptando asi los impredecibles periodos de déficit y exceso hídrico. Se propone una nueva “lógica productiva” sustentada en la hipótesis de que es posible mediante la completa cobertura vegetal (UTCC), restringir la elevada disponibilidad de agua del suelo y mediante irrigación evitar los períodos ocasionales de excesivo déficit hídrico. Dado que la interacion con otras prácticas culturales es determinante del equilibrio entre los beneficios y perjuicios eco-sistémicos de los cultivos de cobertura en viñedos, esta estrategia fue evaluada en combinación con otras prácticas de manejo (densidad de plantación, sistema de conducción, fertilización nitrogenada y estrategias de riego) desde el año 2011 al 2015 en el sur de Uruguay. UTCC fue una herramienta efectiva para restringir el excesivo crecimiento vegetativo de la vid, siendo la disponibilidad de agua en el periodo post-floración el factor determinante del crecimiento vegetativo. UTCC aumentó la concentración de Brix y antocianinas en las uvas, así como también el perfil aromático y sensorial de los vinos Tannat. Dependiendo del periodo donde ocurra el déficit, UTCC podría promover una excesiva restricción del crecimiento vegetativo y productividad del viñedo, siendo una práctica no recomendable de aplicar en secano. Contrariamente, bajo riego suplementario, se trata de una alternativa económica y ambientalmente sostenible para producir vinos de alta calidad consistentemente en ambientes húmedos. El resultado más novedoso obtenido es la significativa reducción observada en la incidencia de podredumbres de racimos observada en el tratamiento UTCC respecto al H, independientemente del crecimiento vegetativo.

**Palabras clave:** *Botrytis*, Tannat, balance de la vid, potencial hídrico, sensorial del vino.

**COVER CROP AND DEFICIT IRRIGATION TO MAXIMIZE CONTROL OF VINEYARD WATER AVAILABILITY IN HUMID CLIMATE**

ABSTRACT

The objective of the study is to improve the quality and regularity of Tannat wine, through soil management aimed at increasing the potential control of vine water availability. We propose a change in the current grape growing paradigm, where the strategy in non-irrigated vineyards is to avoid competition with the cover crops through herbicide applications and accept unpredictable periods of water deficit or excess. A "new logic" instead, is based on the assumption that it is posible with under trellis cover crops (UTCC) limits water availability during periods of excess and avoid excesive vine water stress, during deficit periods with irrigation. Due to the interaction with other cultural practices will determine the balance between eco-systemic services and desservices of cover crops in vineyards, this strategy was evaluated in combination with other management practices (plantation density, trellis systems, nitrogen fertilization and irrigation strategies) from 2011 to 2015 in the south of Uruguay. UTCC was an effective tool to restrict the excessive vine vegetative growth, being water availability during post-bloom period the key driver for vegetative growth. UTCC increased grapes Brix and anthocyanins concentration, as well as enhance the aromatic and sensory profile of Tannat wines. Depending on the level and period in which water deficit occurs, UTCC could excessive restrict vine vegetative growth and yield, being a practice not recomended to apply in dry land. Conversely, with supplemental irrigation, it is an economically and environmentally sustainable alternative to consistently produce high quality wines in humid environments. The novelty of our results is the significant reduction of bunch rot incidence observed in UTCC compered to H treatment, independently of vegetative growth.

**Keywords:** Botrytis bunch rot, Tannat, vine balance, water potencial, wine sensory attributes.

# 1. INTRODUCCIÓN

El balance de la vid y en particular el de su canopia, desempeña un papel determinante en la calidad del producto final (Smart y Robinson 1991). El concepto de balance de la vid hasta los años setenta se encontraba fuertemente asociado al equilibrio fuente/fosa de la planta, siendo orientados los esfuerzos principalmente a la búsqueda de una adecuada relación hoja/fruto como forma de maximizar la calidad de la uva y el vino (Ravaz 1911, Partridge 1925, Winkler et al. 1974, Bravdo et al. 1985, Kliewer y Dokoozlian 2005). Posteriormente, comienza a prestarse más atención al estudio del microclima de la canopia, entendiéndose por balance de la vid no solo la adecuada relación hoja/fruto, sino también, como la adecuada relación hoja expuesta/sombreada y el adecuado "vigor vegetativo (Smart y Robinson 1991). Si bien relaciones altas hoja/fruta son asociados en general a una buena calidad de uva, dependiendo de la variedad, tipos de canopia y capacidad de carga de la planta, podría también estar asociado a brotes vigorosos, hojas grandes, entrenudos largos y múltiples brotaciones laterales (feminelas). Esto resultaría en una canopia densa con gran proporción de hojas y racimos excesivamente sombreados microclima favorable tanto para la maduración como para el desarrollo de podredumbres de racimo (Guilpart et al., 2017, Smart y Robinson, 1991, English et al., 1990).

Un mayor tamaño de bayas (Ojeda et al., 2002), una reducida iluminación de los racimos, o la competencia vegetativo/reproductiva (Champagnol, 1984; Keller et al., 1998; Dry et al., 2001), contribuye a un retraso de la maduración y a una pobre calidad de la uva, hecho que ha sido reportado asociado a viñedos excesivamente vigorosos. Plantas vigorosas, producirán uvas de pobre contenido de sólidos solubles, concentración de antocianos, intensidad colorante, relación tartárico/málico y composición aromática (Keller et al., 1998; Dry et al., 2001), así como también una excesiva acumulación de potasio, (Rojas-Lara y Morrison 1989; Smart et al., 1985; Archer y Strauss 1989; Coniberti et al., 2012) e incidencia de podredumbres de racimo (Emmett et al., 1994).

Ristic (2007), quien comparó vinos de la variedad Shiraz producidos en viñas de canopias excesivamente sombreadas versus los producidos en viñas con buen grado de exposición, demostró como los vinos provenientes de viñedos sombreados, presentaban menores caracteres “licorosos” deseados y un mayor carácter “herbáceo”, además de una mayor astringencia, que aquellos producidos con uvas provenientes de viñedos de canopias expuestas.

El vigor de la planta juega también un rol determinante en el desarrollo de podredumbres de racimo. En plantas vigorosas los racimos serán más grandes y compactos, los brotes vigorosos con largos entrenudos, hojas grandes, y múltiples brotaciones laterales (feminelas), lo que resultaría en una canopia densa con gran proporción de hojas y racimos excesivamente sombreados resultando en un ambiente mas propicio para el desarrollo de podredumbres (Emmett et al., 1994, Jackson y Lombard, 1993; Intrieri et al., 2001; Valdés-Gómez et al., 2008). Valdés-Gómez et al. 2008, con el objetivo de identificar las principales variables involucradas en la mayor incidencia de podredumbres de racimos en plantas vigorosas, define a la compacidad de racimo, como la característica morfológica clave para la infección por *B. cinerea*. Adicionalmente, en canopias excesivamente compactas, la efectividad de control de los productos funguicidas se ha visto sumamente disminuida, debido a la reducida capacidad de penetración (Gubler et al., 1987).

Debido al hábito de crecimiento indeterminado de la vid (Reynolds, 2000), en regiones como la zona sur de Uruguay, donde la temperatura en la estación de crecimiento, la fertilidad y capacidad de retener agua de los suelos predominantes en la región (Brunosoles y Vertisoles) y el régimen de precipitaciones, inducen altas tasas de crecimiento vegetativo, es común observar canopias densas y desequilibradas. Es así que el excesivo vigor es un problema común a muchos de los viñedos orientados a la producción de vinos finos en el Departamento de Canelones y Montevideo Rural; zonas tradicionales de producción vitivinícola uruguaya.

La necesidad de mantener plantas, en viñedos comúnmente no irrigados, con un buen sistema radicular capaces de tolerar los impredecibles periodos de sequía estival, ha determinado que la mayoría de los productores adopte una estrategia definida. Esta consiste en combinar el empleo de portainjertos de vigor medio a alto (SO4, 3309C, 1103P) con la aplicación de herbicidas durante toda la estación de crecimiento, eliminando la competencia establecida por malezas en una franja de aproximadamente 1,0 m bajo la fila. Esta estrategia es sustentada tanto en el conocimiento empírico, como en resultados de investigación nacional, donde portainjertos poco vigorosos como *Vitis riparia* han mostrado una limitada capacidad de reducción de vigor en temporadas lluviosa y una elevada sensibilidad a períodos de déficit hídrico (Disegna et al., 2001). Ello concuerda también con lo reportado por Pouget (1987), quien concluye que cualquier portainjerto se comportará como vigoroso en situaciones de alta disponibilidad de agua y nutrientes, observandose diferencias significativas entre portainjertos únicamente en condiciones restrictivas.

Muchos de los estudios internacionales donde se han utilizado cultivos de cobertura con el objetivo principal de restringir el excesivo crecimiento vegetativo de la vid, reportan que la utilización de gramíneas como cultivo de cobertura en la entrefila y en particular la *Festuca arundinacea*, producen una reducción del crecimiento vegetativo de la vid (Tesic et al., 2007; Morlat y Jacquet, 2003; David et al., 2001) incluso en condiciones de suelo profundo y abundante régimen de precipitación (>ETc) (Maigre y Aerny, 2001a). Sin embargo, aunque la exploración radicular de la vid en los suelos predominantes de nuestra área tradicional de producción, sea limitada (De Lucca, 1983), la reducción del vigor conseguida mediante la utilización de coberturas vegetales altamente competitivas en la entrefila (*Festuca arundinacea*), ha sido limitada e inconsistente (Calero, 2003; Filgueira, 2005).

Más allá del manejo de entrefila, existen otras herramientas disponibles para promover la reducción del vigor en condiciones de clima y suelo favorables al desarrollo vegetativo. Entre ellas, los sistemas de conducción tendrán una influencia dominante sobre la cantidad de hojas potencialmente expuestas (Kliewer y Dokoozlian, 2005). Muchos estudios han reportado los beneficios que presentan los sistemas de canopia dividida para la conducción de viñedos excesivamente vigorosos (Jackson y Lombard, 1993). Dichos estudios concuerdan en que la mejora de calidad obtenida en los sistemas de canopia dividida frente a los de canopia simple, se explica por un mayor porcentaje de hojas por sobre el punto de compensación (200 $\mu$ E/m$^2$/s) en los primeros, (Carbonneau et al., 1978; Kliewer y Dokoozlian, 2005; Schultz, 1995; Shaulis et al., 1966; Shaulis y May, 1971; Smart y Smith, 1988; Smart, 1980; Smart et al., 1985; Smart et al., 1982). Esta mejor intercepción de luz en plantas excesivamente vigorosas, se lograría a través de la reducción del vigor de brotes individuales (asociado a un mayor número de brotes por planta) (Shaulis et al., 1966; Partridge, 1925 – citado por Howel et al., 1991) y por un mejor posicionamiento de los mismos (Shaulis et al., 1966; Smart, 1980; Smart et al., 1982), lo que impacta directamente sobre la radiación recibida por racimos y hojas del interior de la canopia (Shaulis et al., 1966; Smart, 1980; Smart et al., 1985; Smart et al., 1982), aumentando la fotosíntesis neta (Smart et al., 1982), la inducción floral (Shaulis et al., 1966; Smart et al., 1985; Smart et al., 1982), afectando la fisiología de la planta en términos de estrés hídrico (Reynolds et al., 1995) y maduración (Smart y Smith, 1988), reduciendo el pH de uvas y vinos (Howell et al., 1991) e incrementando la concentración de sólidos solubles, monoterpenos y antocianinas (Jackson y Lombard, 1993; Iacono et al., 1995 – citados por Tardaguila y Martinez de Toda, 2005; Reynolds et al., 1995). Los estudios nacionales (Disegna et al., 2005a) realizados sobre la variedad Tannat, también reportan una consistente reducción del vigor de brotes individuales en sistemas de canopia dividida (Lira) frente a aquellas conducidas en Espaldera (VSP). Ello se tradujo en una reducción de la compacidad de la canopia (igual numero de brotes por metro de cordón), una mayor intercepción de luz y de producción de fruta tanto por planta como por hectárea. Sin embargo, contrariamente a lo reportado (Carbonneau et al. 1978) para otras variedades y condiciones, esta mayor captación de luz al interior de la canopia, no se tradujo en una reducción de la incidencia de podredumbres de racimos, ni tampoco en un incremento de la inducción floral o composición de uvas y vinos (Disegna et al., 2005a).

Por otro lado, muchos investigadores han señalado que un adecuado espaciamiento entre plantas, favorecería el balance de la vid, minimizando la necesidad de acciones correctivas (poda en verde, raleo de racimos, manejo de enfermedades) (Davidson, 1992; Archer, 1991). La mayoría de estos trabajos señalan a la distancia en la fila como la principal determinante tanto del vigor de las plantas como de la calidad de la uva (Jackson y Lombard, 1993; Davidson, 1992; Archer, 1991; Shaulis et al., 1989; Intrieri, 1987; Kliewer, 1980). A mediados de los 80, el Dr. Richard Smart planteaba la teoría de "la vid grande", donde proponía como método para la reducción del vigor, ampliar la distancia entre plantas, consiguiendo la buscada reducción del vigor a través de un mayor número de yemas por planta (vigor entendido como tasa de crecimiento de brotes individuales) (Smart et al., 1989). En contraposición, la teoría europea insiste en que con altas densidades, el vigor de la vid se ve controlado a través de una mayor competencia radicular. Champagnol (1984) postula que una alta densidad de plantas producirá plantas pequeñas, lo cual es indispensable para minimizar la gestión del viñedo y obtener vino de calidad. Tales ideas son tan dominantes en la viticultura francesa que normas estrictas ligadas a denominaciones de origen, controlan la densidad de plantación a densidades que pueden alcanzar las 10.000 plantas por hectárea (Jackson y Lombard, 1993). Por otro lado, trabajos sudafricanos demostraron como una elevada densidad promueve el uso más eficaz de los nutrientes y el agua pero causa un excesivo estrés hídrico frente a prolongados períodos de sequía (Archer y Strauss, 1989). Sin embargo, mas allá que la interacción especifica clima-suelo-planta es la que en definitiva determinará la distancia óptima de plantación, no todos los intentos de establecer competencia a través de la densidad de plantación han sido exitosos. Jackson y Lombard (1993) a partir de resultados publicados en un amplio rango de climas y regiones vitícolas concluyen: 1) La alta densidad permitirá reducir el vigor de plantas y proporcionará un microclima ideal, en suelos y climas donde el potencial de crecimiento no sea elevado. 2) Una alta densidad de plantación, será en cambio, incapaz de controlar vigor en suelos fértiles, promoviendo un microclima desfavorable, recomendando para estas situaciones la utilización de sistemas de conducción de canopia dividida (Scott Henry, Lira, GDC).

La poda invernal es otra herramienta que permite regular la expresión vegetativa y vigor de las plantas. El principal objetivo de la poda invernal de la vid es la de regular la carga del viñedo a fin de equilibrar la superficie foliar con la capacidad de la vid para madurar una determinada cantidad de uva. En general, dado que el número de inflorescencias está definido al momento de la poda, cuanto más yemas sean dejadas en pulgares o cañas, mayor será el rendimiento del viñedo. (Jackson et al., 1984; Shaulis, 1971; Champagnol, 1984). El incremento de rendimiento será lineal hasta un nivel a partir del cual la respuesta disminuirá en relación a la disponibilidad de foto-asimilados, incidiendo en el porcentaje de brotación (Jackson et al., 1984) y/o peso final de los racimos (Main y Morris, 2008) Sin embargo, el numero de yemas no solo influirá en la productividad del viñedo, sino también en el vigor individual de los brotes, viéndose este reducido a medida que el numero de yemas es incrementado (Main y Morris, 2008; Clingeleffer, 199; Reynolds y Wardle, 2001). Sin embargo, aumentando el número de yemas sin modificar la distribución espacial de las mismas, se reducirá la distancia entre los futuros brotes, resultando en una mayor densidad de la canopia, fundamentalmente en la zona de los racimos (Ferreira y Marais, 1987), impactando negativamente sobre la maduración y sanidad de la uva (fundamentalmente en climas húmedos) (Smart et al., 1989).

Es así que, a razón de la limitada capacidad de establecer factores de estrés bajo la forma tradicional de nuestra producción (en secano), la mayoría de los trabajos de investigación nacionales han enfocado la resolución del problema a través de la aplicación de prácticas correctivas como el despuntado de ápices vegetativos (rognage) (Coniberti et al., 2007a), los deshojados parciales (Coniberti et al., 2007a; Disegna et al., 2005b) y deshojados a nivel de los racimos (Coniberti et al., 2007b; Disegna et al., 2005b; Ferrer y Gonzalez, 2000). Si bien, estas prácticas han mostrado tener un impacto substancial sobre la calidad y sanidad de la uva y el vino resultante, ofrecen sólo soluciones parciales (Kurtural, 2011).

El presente trabajo pretende contribuir con el posicionamiento de la Categoría "Vinos de Uruguay". Por lo que se orienta con un enfoque agroecológico aplicado, a la mejora de la calidad de uvas y vinos, vía desarrollo de un paquete tecnológico enmarcado en nuestras "limitantes" edafo-climáticas. En este sentido, se plantea un cambio de la "lógica productiva actual", donde la estrategia mencionada anteriormente para combatir los impredecibles períodos de déficit hídrico, se basa en utilización de portainjertos vigorosos, la fertilización y la constante eliminación de competencia mediante la aplicación de herbicidas. La "nueva lógica" planteada en cambio, reconoce en el manejo controlado del estrés, el establecimiento de la competencia y la incorporación del riego, herramientas con gran potencial de aplicación a nuestro sistema de producción vitivinícola. El enfoque se sustenta en la hipótesis de que es posible mediante la completa cobertura vegetal, promover una competencia suficientemente agresiva como para restringir la elevada disponibilidad de agua y nutrientes de nuestros suelos, favorecer el balance de la planta y mejorar la calidad de la uva; al tiempo que mediante la irrigación es posible hacer frente a eventos climáticos extremos, como sequías estivales severas. El trabajo parte de la premisa que el conocimiento de la interacción entre cobertura vegetal, fertilidad del suelo y crecimiento del viñedo es compleja y dinámica, por lo que la comprensión de estas interacciones es clave y debe ser estudiada en detalle si pretendemos hacer de las coberturas vegetales, una herramienta efectiva para el control del crecimiento vegetativo y disponibilidad hídrica del viñedo.

## 1.1 OBJETIVOS

### 1.1.1 Objetivo general

Ajustar prácticas de manejo que permitan maximizar el control de los principales factores de crecimiento de la vid (agua y nitrógeno) en las condiciones edafo-climáticas del sur de Uruguay.

### 1.1.2 Objetivos específicos

Evaluar para las condiciones edafo-climáticas del sur de Uruguay, el potencial de la cobertura vegetal permanente (parcial y/o total) como herramienta para restringir el crecimiento vegetativo de viñedos excesivamente vigorosos.

Probar la hipótesis que la competencia establecida con la vid por agua y nitrógeno durante el periodo de máximo crecimiento vegetativo, es el factor determinante de la reducción de crecimiento vegetativo y productividad observada en viñedos bajo cobertura vegetal permanente.

Evaluar el potencial de la cobertura vegetal permanente como herramienta para el control del crecimiento vegetativo y mejora de los atributos de calidad de las uvas Tannat, frente a otras prácticas de manejo alternativas (densidad de plantación, sistema de conducción).

Analizar el impacto de la reducción del vigor sobre la productividad, composición de la uva y atributos de calidad del vino Tannat.

Analizar el impacto del vigor de la vid sobre la susceptibilidad a podredumbres de racimos de la variedad Tannat.

## 2. MARCO TEÓRICO

Aparte de su función principal en la estabilización del suelo, son varios los objetivos perseguidos al definir un cultivo de cobertura y/o estrategia de gestión del suelo del viñedo. Los cultivos de cobertura son ampliamente utilizados en viñedos de Estados Unidos y de Europa templada, donde la práctica convencional es el cultivo de coberturas vegetales en la entrefila, combinado con una faja libre de competencia de ancho variable debajo de las plantas, habitualmente mantenida mediante la aplicación de herbicidas (Wolf, 2008).

Los principales beneficios reportados como ventajas de la utilización de cubiertas vegetales permanentes incluyen controlar la erosión, la estructura, drenaje interno y la capacidad de infiltración del suelo (Champagnol, 1984; Wheaton et al., 2008; Gaudin et al., 2010), incrementar su contenido de materia orgánica (Merwin et al., 1994; McGourty y Reganold 2005) facilitar la gestión de las malezas (Teasdale y Mohler, 2000), favorecer la actividad y diversidad microbiana del suelo (Baumgartner et al., 2005, Ingels et al., 2005; Steenwerth y Belina, 2008a), como el establecimiento de insectos benéficos (Sullivan, 2003), así como también influir sobre la disponibilidad de agua y nutrientes controlando el vigor de la vid (Maigre y Aerny, 2001a, Morlat y Jacquet 2003, Hatch et al., 2011; Tesic et al., 2007). Además de estos beneficios ésta permite reducir los costos anuales de manejo (Giese et al., 2015; Yeh et al., 2014), la susceptibilidad a enfermedades (Morlat y Jacquet, 2003; Nazrala, 2008; English et al, 1990) y potenciar los aspectos deseables en la calidad de la uva y el vino Giese y Wolf, 2009; Lopes et al., 2008; Monteiro y Lopes, 2007; Tesic et al., 2007).

Los cultivos de cobertura se han utilizado con variado éxito para regular el crecimiento vegetativo de la vid (Hatch et al., 2011). Las principales consideraciones para la elección de los cultivos de cobertura en viñedo, incluyen el grado de competencia por agua y nutrientes que poseen, el ciclo de crecimiento (perenne vs. anual), su adaptación y persistencia en el medio ambiente vitivinícola y su propensión a albergar organismos que puedan afectar positiva o negativamente a la vid (Giese et al., 2015). En varias regiones se ha estudiado el impacto de diferentes especies de cobertura vegetal de entrefila, sobre los parámetros de crecimiento vegetativo, rendimiento y composición de la uva (Guerra y Steenwerth, 2012). Una extensa lista de especies y mezclas de especies (generalmente integradas por gramíneas y leguminosas) han sido estudiados y recomendadas para su uso como cultivos de cobertura en viñedos (Guerra y Steenwerth, 2012). A modo de ejemplo más de 50 especies y combinaciones de plantas se usan comúnmente como cultivos de cobertura en viñedos de California (Ingels et al., 1998). El impacto del cultivo de cobertura y su sostenibilidad a largo plazo dependerá además en gran medida de las de las condiciones edáficas, fundamentalmente en lo que respecta a su capacidad de retención de agua y del régimen de precipitación estacional de la región (Monteiro y Lopes, 2007; Tesic et al., 2007). Si bien existen modelos que aumentan la comprensión o predicción de los sistemas de cultivos anuales y sus efectos a largo plazo de las prácticas de gestión del suelo, en aspectos como, la composición de especies de malezas, el control de la erosión y la salud del suelo (Holst et al., 2007), todavía no se ha avanzado de igual forma en el desarrollado de modelos adaptados a sistemas de vitivinícolas. Las posibles combinaciones de suelo, régimen pluviométrico, variedad, objetivo productivo, que sean numerosas las posibles especies a seleccionar como cultivo de cobertura, sin embargo es posible restringir la decisión al clasificar a los principales grupos botánicos de acuerdo a sus funciones y características dominantes (Guerra y Steenwerth, 2012). La amplia mayoría de las gramíneas presentan raíces fibrosas que penetran y agregan efectivamente el suelo (Colugnati et al., 2004). Su alta relación C/N se asocia con tasas de descomposición más lentas que las de las leguminosas (McGourty y Reganold, 2005; Olmstead, 2006). Las gramíneas son capaces de producir una gran cantidad de biomasa que puede ayudar a aumentar la materia orgánica del viñedo además de reducir la erosión y compactación de suelo (Colugnati et al, 2004; McGourty y Reganold, 2005; Olmstead, 2006). Las leguminosas por su parte, presentan a menudo una menor persistencia que las gramíneas, y su capacidad de fijar N atmosférico incrementaría la disponibilidad de nitrógeno en el suelo (Patrick et al., 2004). Si bien la capacidad de las leguminosas de fijar N atmosférico, depende de la especie, la eficiencia de nodulación, la humedad y temperatura del suelo (Madge, 2005), la mayoría de las estimaciones oscilan en un aporte de entre 50 y 330 kg/ha (Ingels et al., 1998) lo que representa un aporte de N significativo al sistema. Esto se ha visto generalmente reflejado en un mayor contenido de nitrógeno en hojas y tejidos, además del nitrógeno fácilmente asimilable del mosto (YAN) (Fourie et al., 2006;

Sweet y Schreiner, 2010), en contraposición a la reducción generalmente reportada en viñedos empastados con gramíneas perennes (Van Leeuwen et al., 2000; Wheeler et al., 2005; Filgueira, 2005). Si bien en algunos casos se podría incrementar la disponibilidad de nitrógeno más allá de lo deseado en un viñedo excesivamente vigoroso, también dada a la relevancia de este nutriente durante los procesos de vinificación, su acumulación en bajas concentraciones puede ser una característica deseable (Canoura et al., 2018)

A su vez, los aspectos espaciales y temporales de la captación de N y agua, tanto del cultivo de cobertura como de la vid, son de extrema relevancia (Celette et al., 2009). Tanto cultivos perennes como anuales han logrado suprimir el excesivo crecimiento vegetativo de la vid, a través de la competencia por agua y nitrógeno (Hatch et al., 2011, Guerra y Steenwerth, 2012). Sin embargo, los cambios temporales encontrados en el contenido de N, tanto en el cultivo de cobertura como en órganos de almacenamiento de la vid, evidencian la mayor capacidad de competencia establecida por los cultivos de gramíneas perennes respecto a las especies anuales (Celette et al., 2009; Giese et al., 2015). Las especies perennes producen importante biomasa radicular en el período otoño-invierno y consecuentemente tienen mayor potencial de secuestro de agua temprano en la temporada y nitrógeno que las plantas anuales (Celette et al., 2009; Stork y Jerie, 2003). La vid presenta crecimiento indeterminado, pero su mayor crecimiento vegetativo ocurre generalmente desde brotación a envero, siendo el pico de tasa de elongación de brotes coincidente con el fin de la floración (Giese et al., 2015). Las gramíneas perennes presentan un patrón de crecimiento acompasado al de la vid, estableciendo una competencia significativa por la humedad y nutrientes durante este periodo crítico, a diferencia de las invernales y de las estivales que se encuentran al final de su ciclo y en periodo de establecimiento respectivamente (Guerra y Steenwerth, 2012). Por tanto, en regiones donde la reserva de humedad del suelo y las precipitaciones estacionales contribuyen al excesivo crecimiento vegetativo de la vid (Monteiro y Lopes, 2007), parece más lógica la utilización de gramíneas perennes que establezcan la competencia durante la fase inicial del crecimiento vegetativo (Giese et al., 2015).

Desde los primeros estudios realizados en los años ochenta hasta el presente, la mayoría ellos reportan haber corregido con éxito las situaciones de excesivo vigor mediante cultivos de cobertura de entrefila y aumentando la exposición de la canopia, impactando positivamente sobre la composición de la uva y el vino (David et al., 2001; Maigre y Aerny, 2001a y b; Wheeler et al., 2005; Carsoulle, 1995; Morlat y Jacquet, 2003; Tesic et al., 2007; Monteiro y Lopes, 2007; Hickey, 2016; Reynolds et al., 2005). En general ha enconrado que ocurre un incremento de la concentración de azúcares y antocianos tanto en mosto como en vino (Agulhon, 1998; Bourde et al., 1999; Morlat y Jacquet, 2003; Wheeler et al., 2005; Nazrala, 2008). Además se ha observado una reducción significativa de la infección con *Botrytis* (Carsoulle, 1995; Morlat y Jacquet, 2003; Tesic et al., 2007) en los tratamientos con cultivos de cobertura permanente respecto a aquellos donde la cubierta vegetal es eliminada.

Sin embargo, en muchos casos esta mejora de la composición y/o sanidad de la uva se encuentra asociada a una pérdida significativa del rendimiento potencial (Agulhon, 1998; David et al., 2001; Morlat y Jacquet, 2003; Wheeler et al., 2005). El establecimiento temprano (floración) de la competencia entre la vid y el cultivo de cobertura no solo afectara el crecimiento vegetativo, sino también dependiendo de su intensidad, podría afectar el cuajado y la inducción de yemas del año siguiente (Spayd y Morris, 1978). Una restricción hídrica establecida durante el periodo cuajado - envero afectaría también el rendimiento potencial a través de la reducción del tamaño de baya, mientras que en el periodo envero – cosecha el impacto esperable sobre el crecimiento vegetativo y la productividad del viñedo sería menor (Ojeda et al., 2002; Matthews et al., 1987). Es decir que reducir el vigor de la vid sin afectar el rendimiento potencial representa un desafío productivo.

El análisis requiere resaltar algunas características claves del desarrollo vegetativo y formación del rendimiento de la vid. El crecimiento vegetativo se encuentra limitado por acumulación de reservas de la planta (Winkler, 1974) pero condicionado fundamentalmente por las condiciones ambientales durante el período más intenso de crecimiento vegetativo (generalmente coincidente con la floración) (Huglin y Schneider, 1998). Por otro lado, el rendimiento de la vid se encuentra establecido además por las condiciones de crecimiento de al menos dos temporadas consecutivas. Los principales componentes del rendimiento de la uva (número de racimos por brote y número de bayas por racimo) están determinados principalmente por la luz, temperatura y suministro de asimilados a los brotes durante la floración anterior y afectados por el estatus hídrico y disponibilidad de nitrógeno (Guilpart et al., 2014; Butrose, 1974; Keller, 2005; Vasconcelos et al., 2009). Por lo tanto, la ocurrencia de cualquier restricción ambiental durante este período, no solo determinará la reducción del desarrollo vegetativo en la estación actual, sino también la reducción del rendimiento de fruta de la siguiente temporada (Hardie y Considine, 1976; Guilpart et al., 2017; Coniberti et al., 2018).

En general, la respuesta encontrada frente a la utilización de coberturas vegetales en viñedos excesivamente vigorosos bajo clima húmedo, ha sido comparable con el normalmente asociado al riego deficitario en climas áridos (reducción del vigor, menor tamaño de bayas, mejor composición de la fruta y menor incidencia de podredumbres) (Guerra y Steenwerth, 2012). El crecimiento vegetativo es extremadamente sensible al déficit hídrico, puesto que para permitir la ampliación de las células en crecimiento, las raíces deberán absorber más agua de la que se pierde por transpiración (Boyer, 1985). Por lo tanto, las tasas de crecimiento cambian rápidamente con fluctuaciones en el potencial de xilema (ΨXilema) y una reducción del crecimiento de brotes y hojas es el primer signo visible de déficit de agua de la vid (Williams et al., 1994).

En la mayoría de los trabajos esta reducción del crecimiento potencial estuvo acompañada de una mejora de la calidad potencial de la uva, entendiendo a los cultivos de cobertura en general como beneficiosos (Guerra y Steenwerth, 2012). El cierre estomático producido a partir de un déficit hídrico significativo, no solo reduce el crecimiento, sino también limita la fotosíntesis, dado que la difusión del $CO_2$, es más sensible que la del vapor de $H_2O$ frente a la apertura estomática (Boyer et al., 1997). Sin embargo, un déficit hídrico leve reducirá el crecimiento vegetativo sin afectar substancialmente la fotosíntesis, lo que supone una mayor disponibilidad de carbohidratos para la uva (Keller, 2005).

Sin embargo, existen contradicciones sobre si exiten diferencias significativas en la composición de la uva, incluso en situaciones donde la cobertura vegetal redujo satisfactoriamente el excesivo vigor de los viñedos (Hickey et al., 2016, Karl et al., 2016; Giese et al., 2015; Afonso et al.; 2003). La relación hoja/fruta o rendimiento/peso de poda (Índice de Ravaz) son indicadores reconocidos del equilibrio de la vid, pudiendo afectar significativamente la composición del fruto

(Bravdo et al., 1985). Es así que una reducción del crecimiento vegetativo e interceptación de luz, con relativo menor impacto en la producción de fruta (proveniente de yemas inducida la temporada anterior), podría generar durante las primeras temporadas de establecimiento de la cobertura vegetal, una significativamente menor relación hoja/fruta afectando la maduración de las bayas (Lakso y Sacks, 2009). Bajo condiciones de excesiva restricción hídrica, el metabolismo fotosintético también se afectaría progresivamente (Escalona et al., 1999), lo que se reflejaría no solo en pérdidas de productividad, sino también de calidad de uva (Escalona et al., 1999; Lawlor y Cornic, 2002). El umbral en el que los efectos negativos de los cultivos de cobertura comienzan a sobrepasar los beneficios depende del genotipo de la vid, del cultivo de cobertura, de las características del suelo y clima, además de los objetivos productivos (Hatch et al., 2011; Steenwerth et al., 2013; Wolpert et al., 1993).

Aunque los principales factores de competencia variaran con las condiciones ambientales (Giese et al., 2015), la mayoría de la literatura coincide en que además de la restricción hídrica (Lopes et al., 2011; Morlat y Jacquet, 2003; Lopes et al., 2004; Pellegrino et al., 2004; Hatch et al., 2011; Celette et al., 2005; Monteiro y Lopes, 2007; Sweet y Schreiner, 2010; Tesic et al., 2007), la disminución de la disponibilidad de nitrógeno durante la primavera es otro de los factores determinantes de la restricción del crecimiento vegetativo observado (Rodriguez-Lovelle et al., 2000; Afonso et al., 2003; Agulhon, 1996; Le Goff et al., 2000; Maigre y Aerny, 2001b; Celette et al., 2005; Celette et al., 2009; Saayman y Van Huyssteen, 1983).

Los efectos estimulantes de la disponibilidad de nitrógeno (particularmente nitrógeno) sobre el crecimiento vegetativo de la vid, ha sido ampliamente estudiado en la literatura (Ver Keller, 2005). El crecimiento vegetativo de la vid presenta frente a incrementos en su disponibilidad, respuesta "tipo-saturación" (Spayd et al., 1993). La vid absorbe nitrógeno principalmente bajo la forma de nitrato ($NO_3^-$) (Conradie y Saayman, 1989), estando su tasa de absorción, regulada por el nivel de circulación de aminoácidos y péptidos en el floema quienes actúan como intermediarios entre lo demandado por los tejidos en crecimiento y la absorción de $NO_3^-$ (Cooper y Clarkson, 1989). Tanto el metabolismo del nitrógeno como su eficiencia de absorción, presentan un fuerte componente genético, estando el comportamiento vegetativo de la vid frente a determinados niveles de péptidos, aminoácidos y $NO_3^-$ en pecíolos de hojas, esta muy ligado altamente al cultivar (Christensen, 1984). Sin embargo en cualquier caso suprimir la disponibilidad de N, en teoría, permitiría controlar el crecimiento vegetativo (Christensen et al., 1994). Según Tregoat et al. (2000) citado por Filgueira (2005) en climas o temporadas secas, el comportamiento vegetativo de la viña está determinado fundamentalmente por el régimen hídrico, mientras que la disponibilidad de nitrógeno es un importante factor de control de vigor y calidad de la uva en regiones o temporadas lluviosas. Esta disminución del vigor es explicada por una reducción del flujo de citoquininas producidas en la raíz, en respuesta a la deficiencia de nitrógeno, afectando principalmente la división celular (Kakimoto, 2003; Boyer, 1985; Gastal y Lemaire, 2002).

Al igual que frente a un déficit hídrico controlado, una deficiencia leve de nitrógeno suprimirá el crecimiento sin afectar significativamente la fotosíntesis, lo que supone una mayor disponibilidad de carbohidratos (Gastal y Lemaire, 2002), los que podrán ser translocados a la uva, favoreciendo la maduración, acumulación de sólidos solubles y producción de metabolitos secundarios en uvas (Lemaire y Millard, 1999; Keller et al., 1998; Wade et al., 2004) lo que supone una mejora en la composición de la uva. Por el contrario, deficiencias importantes de N podrían limitar significativamente la fotosíntesis debido a la reducción de proteínas enzimáticas como la Rubisco (Chen y Cheng, 2003). Deficiencias aún más severas, promoverían además la redistribución de nutrientes hacia hojas en desarrollo y la abscisión de hojas viejas (Lawlor y Cornic, 2002). En consecuencia, el contenido de sólidos solubles y metabolitos secundarios en bayas se verían negativamente afectados (Keller et al., 1998).

Adicionalmente a la restricción de la disponibilidad hídrica y de nutrientes asociada al consumo de la pastura, la competencia de cobertura vegetal podría también afectar el desarrollo de las raíces de la vid en suelos poco profundos (Celette et al., 2009; Smart et al., 2006) y/o restringiendo el crecimiento radicular, en horizontes superficiales, fundamentalmente de raíces finas (<1 mm de diámetro) (Centinari et al., 2016; Morlat y Jacquet, 1993), lo que afectaría la captación potencial de agua y nutrientes (Van Huyssteen, 1988; Champagnol, 1984; Byers et al., 2005; Giese y Wolf, 2009; Celette et al., 2008).

Los cultivos de cobertura pueden modificar también la distribución espacial y temporal del agua en el perfil del suelo (Celette et al., 2008) además de las reservas de nitrato y amonio a través de la tasa de mineralización del N del suelo (Steenwerth y Belina, 2008b). Esta desaparición de parte de las raíces superficiales sumado al establecimiento de formas más estables de la materia orgánica en viñedos empastados, podrían determinar deficiencias de nitrógeno durante períodos de restricción hídrica (Morlat y Jacquet, 2003). Esto es particularmente relevante cuando se trata de viñedos en secano y/o bajo suelos superficiales, dado que una excesiva competencia durante períodos críticos podría resultar en pérdidas importantes de productividad (Bugg y Van Horn, 1998; Celette et al., 2005; Wolpert et al.; 1993). El impacto de las cubiertas vegetales sobre el crecimiento vegetativo, rendimiento y composición de la uva ha sido en general más pronunciado en climas secos que en regiones húmedas (≈300 mm vs >500 mm de precipitación anual), por lo que fue sugerido que prácticas como el riego y fertilización permitirían compensar el efecto del establecimiento de una cobertura vegetal (Colugnati et al., 2004). Sin embargo, el desarrollo de la vid es afectado por muchos factores interrelacionados, existiendo riesgos e incoherencias en la presunción de que una determinada restricción del contenido de agua del suelo, resultará en una reducción del crecimiento vegetativo de la vid (Zufferey y Smart, 2012). Estudios realizados en la Estación Experimental de Changins (Suiza) por Maigre y Aerny (2001a) muestran que incluso en condiciones de suelos profundos y buena disponibilidad hídrica, la competencia ejercida por la gramínea perenne (*Festuca arundinacea*) redujo significativamente el peso de poda y la absorción de N del viñedo durante los cuatro años de estudio. Esta tendencia se mantuvo incluso cuando el viñedo fue fertilizado con 100 kg/Ha N cada año.

A su vez, no todos los estudios encuentran la respuesta esperada de reducción de vigor frente a la instalación de cobertura vegetal permanente en entrefila, obteniéndose en algunos casos nulo o modesto impacto sobre en el crecimiento y desarrollo de la vid (Baumgartner et al., 2008; Steenwerth et al., 2013; Sweet y Schreiner, 2010; Ingels et al., 2005). En suelos fértiles, sin limitación de agua, la competencia establecida por la cobertura podría ser mínima, resultando en un limitado o nulo efecto sobre el desarrollo radicular y vegetativo de la vid (Pugnaire y Luque, 2001; Firth et al., 2003; Wheeler et al., 2005).

En estudios más recientes, se propone que se extienda la proporción del suelo cubierto al área debajo de las plantas, con el objetivo de incrementar la competencia entre las raíces de la cobertura y de la vid (Hatch et al., 2011; Jordan et al., 2016; Giese et al., 2015; Centinari et al., 2016; Hickey, 2016; Karl et al., 2016). De esta forma el potencial de reducir la disponibilidad de agua y nutrientes del perfil del suelo aumentaría substancialmente (Sánchez et al., 2007; St. Laurent et al., 2008; Atucha et al., 2011; Centinari et al., 2016), lo que alteraría la generación de raíces, su distribución vertical (Yao et al. 2009, Atucha et al 2013) y longevidad (Comas et al., 2010). Al igual que lo mencionado para los casos donde el establecimiento de la cubierta vegetal es restringida a la entrefila, la especie utilizada como cultivo de cobertura es determinante en la respuesta observada de la vid (Centinari et al., 2016; Karl at al., 2016; Jordan et al., 2016; Giese et al., 2015). Dado que la restricción del vigor de viñedos excesivamente vigorosos es en general el principal objetivo perseguido cuando se plantea completa cubertura vegetal del suelo, son muy pocos los estudios que han evaluado el uso de cultivos anuales plantados directamente bajo la fila (Centinari et al., 2016; Karl et al., 2016; Jordan et al. 2016). Algunos estudios provienen del noreste de Estados Unidos donde debido a la necesidad de cubrir el tronco de la vid para evitar en daño por frío invernal, no es posible la instalación de coberturas permanentes (Jordan et al., 2016). Los resultados encontrados en Cabernet Franc (Centinari et al., 2016; Karl at al., 2016) y Riesling (Jordan et al., 2016) bajo esas condiciones edafoclimáticas (suelos fértiles, precipitación estacional > 500mm), han sido inconsistentes, generando cierta incertidumbre sobre la capacidad de las coberturas anuales de restringir el excesivo vigor de la vid (Centinari et al., 2016; Jordan et al., 2016). En general no se ha detectado una reducción significativa del desarrollo vegetativo en viñedos excesivamente vigorosos (Centinari et al., 2016; Jordan et al., 2016). Por el contrario Karl et al. (2016), encontraron una significativa reducción del crecimiento vegetativo y rendimiento de fruta en similares condiciones de estudio. Cabe destacar que en este último trabajo el viñedo experimental podría considerarse en equilibrio bajo la forma tradicional de producción con herbicidas (peso de poda por metro de hilera < 0,6 Kg), por lo que escaparía a la situación productiva que típicamente justificaría esta práctica.

Por otra parte, dado que en ninguno de estos trabajos se detectaron efectos negativos sobre la composición de la uva, y considerando el impacto sobre la salud del suelo y el riesgo de contaminación ambiental que conlleva la utilización de herbicidas (Landry et al., 2006; Martinez-Casanovas y Sánchez-Bosch, 2000), esta práctica es actualmente recomendada como una alternativa sostenible a la utilización de herbicidas en esta región (Centinari et al., 2016; Jordan et al., 2016).

Contrariamente a lo reportado para cultivos anuales, las gramíneas perennes en general redujeron efectivamente el excesivo crecimiento vegetativo cuando se mantuvo el 100% del suelo cubierto, con un mínimo impacto sobre el rendimiento de viñedos situados en ambientes de alta disponibilidad hídrica (capacidad de retención de agua de los suelos + pluviometría estacional > 700 mm) (Giese et al., 2015; Hickey et al., 2016). En un estudio realizado durante seis vendimias consecutivas sobre un viñedo experimental de Carolina del Norte, USA (Giese et al., 2015), se requirió el completo establecimiento de la cobertura vegetal y una estación de crecimiento con precipitaciones estacionales por debajo de la media (300 mm), para que se produzca una reducción significativa del excesivo crecimiento vegetativo detectado en el tratamientos tradicional con herbicidas. Existe cierto consenso en que la competencia por agua es también la causa principal de la disminución del vigor observado frente a la instalación de coberturas vegetales completa (Hatch et al., 2011; Celette et al., 2009; Hickey et al 2016; Giese et al., 2015; Centinari et al., 2016; Jordan et al., 2016).

Sin embargo es importante destacar que en este estudio, los pesos de poda se mantuvieron dentro de valores aceptables (<0,6 kg/m) en siguientes temporadas, incluso cuando la precipitación fue similar o superiores a la media para la región (> 600 mm) y no fueron registrados periodos de significativa restricción hídrica (nunca inferior a -0,6 MPa). Resultados comparables fueron obtenidos por Hickey et al. (2016) en Virginia, USA, en estos estudios la mayor diferencia de peso de poda observada entre dos temporadas consecutivas fue ~ 20% independientemente del tratamiento de gestión del suelo (Hickey et al., 2016; Giese et al., 2015). Dado que, el desarrollo vegetativo de la vid está estrechamente condicionada por su estatus hídrico, pero su potencial de crecimiento permanecerá limitado por su condición previa (Winkler et al., 1974), la completa cobertura del suelo del viñedo con gramíneas perennes (*Festuca arundinacea* Schreb.), podría representan una estrategia efectiva para el control del excesivo desarrollo vegetativo incluso en condiciones edafoclimáticas promotoras del desarrollo vegetativo y del régimen de precipitación variable como el Uruguay. También se ha sugerido que dado que la vid y los cultivos de cobertura coexisten en los viñedos, el manejo del riego y la fertilización deberían incorporarse al sistema de forma de satisfacer las necesidades de ambos (Colugnati et al., 2004). Adicionalmente al riego y la fertilización, otras prácticas de manejo como el corte o la eliminación (parcial o total) del cultivo de cobertura son consideraciones que deben ser tomadas a escala de tiempo estacional, especialmente bajo regímenes de precipitación variable (Garcia et al., 2018). Garantizar la sustentabilidad del sistema de producción vitícola en este caso, solo es posible si se aplica un manejo adaptativo (Jackson et al., 2010).

# 3. UNDER-TRELLIS COVER CROP AND PLANTING DENSITY TO ACHIEVE VINE BALANCE IN A HUMID CLIMATE[1]

## 3.1. ABSTRACT

The goal of our study was to improve Tannat (*Vitis vinifera* L.*)* grape and wine composition, by achieving vine balance in a humid climate. We tested under-trellis cover crops (UTCC) compared to a standard floor management of alleyway cover crops and under-vine herbicide. This strategy was tested in combination with variable planting density over three growing seasons in Southern Uruguay. Two factors were evaluated in a split plot design with five replicates. Treatments were, (1) UTCC (full cover of the vineyard soil with tall fescue (*Festuca arundinacea Shreb*) versus conventional alleyway tall fescue with 1.0 m wide weed-free strips under the trellis, and (2) two spacings between vines in the row (0.8 vs 1.5 m). To avoid excessive vine water stress, supplemental irrigation was used during water deficit periods. Shoot growth rate, mid-day stem water potential, berry size and berry composition were monitored over the season as well as final yield, cluster and pruning weights. Results showed that UTCC reduced vegetative growth as expressed by pruning weight/m while closer PD resulted in greater vegetative growth parameters. UTCC reduced vine vegetative growth to recommended values of pruning weight per m of row under both plating densities. It also reduced berry size, cluster weight and bunch rot incidence as well as increased total soluble solids and anthocyanin concentration in grapes compared to the standard herbicide treatment. The use of UTCC with supplemental irrigation, showed promise for achieving vine balance in high vine capacity conditions.

**Key words:** Tannat, vegetative growth, bunch rot, water potential, viticultural practice.

[1] Este capítulo fue publicado en la revista Sientia Horticulturae: A. Coniberti, E. Disegna, V. Ferrari, M. García Petillo, A. N. Lakso, 2018. Under-trellis cover crop and planting density to achieve vine balance in a humid climate. Scientia Horticulturae, v.: 227 p.:65 – 74.

## 3.2. INTRODUCTION

Research has generally shown that grapevine productivity, fruit quality and management efficiency are optimal with moderate vegetative vigor (Smart and Robinson 1991). Excessive vine growth includes dense, shaded canopies that not only negatively impact fruit and wine quality potential (Dry et al. 2005, Smart and Robinson 1991), but also foster bunch rot incidence (Guilpart et al. 2017). Conversely, very small vines that are limited by inadequate water or nutrients have reduced vine capacity for ripening the crop due to reduced light interception and reduced leaf function (Lakso and Sacks 2009, Kliewer and Dokoozlian 2005). Thus, the goal in many vineyards is to have a good vegetative/reproductive balance giving good vine capacity for yield potential, and having appropriate crop for desired ripening and fruit quality. Several metrics of an appropriate range of vegetative growth have been developed (Dry et al. 2005, Kliewer and Dokoozlian 2005). Mean pruning weights of vine length greater than 0.5 kg per meter of row or canopy length or dormant cane weights above 50 grams tend to indicate excessive vegetative vigor and imbalance.

Although the appropriate balance may vary with variety and desired yield or wine style, regulating vegetative growth is a key vineyard practice. Increasing growth tends to be easier to achieve as adding adequate water, nutrients or soil amendments are practical. Reducing excessive growth, however, is often difficult as removing or limiting resources is more difficult especially in humid climates and deep fertile soils. The use of under-trellis cover crops (UTCC) has been studied in cool humid climates that often experience abundant water availability and fertile soils. Apparently due to competition for soil moisture and mineral nutrients, UTCC has been reported to consistently reduce vine vigor (Hickey et al. 2016, Karl et al. 2016, Giese et al. 2015, Giese et al. 2014, Lopes et al. 2008, Tesic et al. 2007), and improve fruit sunlight exposure (Giese et al. 2015, Giese et al. 2014, Hatch et al. 2011). Additionally, increases of total soluble solids and/or berry skin phenols and anthocyanins have been reported (Hickey et al. 2016, Tesic et al. 2007).

Planting density (PD) is another technique used to control excessive vegetative growth of perennial fruit crops including grapevines. In general, it has been reported that under dry land, restricted and low potential soil conditions, increases in plant density resulted in less vegetative growth and less dense canopy (Archer and Strauss 1990, Champagnol 1984). With equal shoots per meter of trellis, however generally the opposite has been reported under high capacity conditions. Jackson and Lombard (1993), reported that it is not possible to control excessive vine vigor under high capacity condition by planting density, and recommended the use of divided canopies trellis systems under these conditions.

We propose a change in the current local commercial production practices in the humid climate of Uruguay that, (1) accepts unpredictable periods of water deficit or excess in non-irrigated vineyards, and (2) reduces competition from cover crops. The goal of our study was to evaluate integrated systems of cover cropping with supplemental irrigation to regulate canopy growth to optimize vine balance resulting in improved Tannat grape and wine composition. Our approach was to use under-trellis cover crops (UTCC) to limit vine water availability, reduce vine growth rate and limit final canopy size and density. To avoid excessive water stress due to the cover crop competition, supplemental irrigation was applied during moderate water deficit periods to regulate the stress and thus vine growth and function. Treatments were tested under two different inter-vine planting densities.

## 3.3. MATERIALS AND METHODS

### 3.3.1. Experimental site

The experiment was conducted over three consecutive growing seasons from 2011 to 2014 in Southern Uruguay (34°44' S, 56°13' W). Uruguayan climate can be classified as temperate – humid without a prolonged dry season, Cfa by the Köppen-Geiger system (http,//en.climate-data.org/location/3741/). Historical mean total annual rainfall in Southern Uruguay (1972-2015) is 1100 mm/year, with 650 mm occurring the growing season (Table 1). Further weather data details can be accessed at http,//www.inia.org.uy/gras/agroclima/cara_agro/index.html. The soil has been classified as a Tipic Argiudolls (USDA soil classification system), with a variable depth of 0.90 to 1.1 m and silty clay texture. A restrictive clay layer (Bt) is located at 40 to 50 cm, so most of root system is developed above. The total soil available water (field capacity-permanent wilting point) to 1.0 m depth was 117 mm.

**Table 1**. Irrigation by treatment and evapotranspiration, precipitation and growing degree-days (>10ºC), from Las Brujas weather station located at 200 m from the experimental site.

| | Phonological stage | Degree-days (>10ºC) | Eto Penman (mm) | Precipitation (mm) | Irrigation (mm) | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | | H0.8 | H1.5 | UTCC0.8 | UTCC1.5 |
| Historical | budbreak - bloom | 338 | 208 | 227 | | | | |
| | bloom - veraison | 718 | 359 | 291 | | | | |
| | veraison - harvest | 664 | 257 | 266 | | | | |
| | post-harvest | 421 | 136 | 252 | | | | |
| Season 2011/12 | budbreak - bloom | 328 | 224 | 132 | 0 | 0 | 0 | 0 |
| | bloom - veraison | 722 | 391 | 177 | 16.7 | 22.7 | 103.5 | 110.9 |
| | veraison - harvest | 665 | 271 | 212 | 23.7 | 37.9 | 70.7 | 80.5 |
| | post-harvest | 435 | 157 | 147 | 0 | 0 | 0 | 0 |
| Season 2012/13 | budbreak - bloom | 403 | 211 | 359 | 0 | 0 | 0 | 0 |
| | bloom – veraison | 753 | 381 | 288 | 0 | 0 | 42.2 | 59.0 |
| | veraison - harvest | 641 | 291 | 199 | 0 | 0 | 9.7 | 9.7 |
| | post-harvest | 352 | 139 | 173 | 0 | 0 | 0 | 0 |
| Season 2013/14 | budbreak - bloom | 332 | 214 | 204 | 0 | 0 | 51.2 | 51.2 |
| | bloom - veraison | 804 | 398 | 284 | 7.3 | 12 | 60.2 | 69.2 |
| | veraison - harvest | 692 | 228 | 641 | 0 | 0 | 0 | 0 |
| | post-harvest | 395 | 146 | 194 | 0 | 0 | 0 | 0 |

### 3.3.2. Experimental vineyard and general vine management

Vines were trained to a vertical shoot positioning system (VSP) in north-south oriented rows (2.8 m row spacing). Cordon-trained vines were pruned to seven two-bud spurs per meter during dormancy. The height of the cordon was 1.0 m, and the top of the canopy was approximately 2.1 m above the ground. At approximately 30 cm shoot length, all shoots not located on spurs were removed. During the growing season, shoots were positioned by hand vertically above the spurs and topped 30 cm above the top wire. Catch wires were used to keep shoots in position. To avoid overcropping during ripening, crop level was adjusted by cluster thinning in each experimental plot at veraison (Stage 35 - Eichhorn and Lorentz 1977). Based on prior research (Coniberti et al. 2011), an optimal crop level was estimated to be about 1 cluster/shoot. With a full canopy this provides a ratio of at least 1.8 m2 leaf area/kg fruit weight needed for maximize sugar and anthocyanin accumulation.

To estimate the potential yield in every plot, thinned clusters were counted and weighed. Standard disease control fungicide programs were applied for downy mildew, powdery mildew, and *Botrytis* bunch rot. Irrigation water was applied with drip emitters (4 L/minute) located directly under the vines and distributed 0.3 m apart. The irrigation system was designed to allow independently-irrigated single experimental plots.

3.3.3. Treatments

The experiment was conducted on Tannat grapevines grafted on to SO4 rootstock. The vineyard was 7-years-old when an under-trellis cover crop (UTCC) was established in March 2011 (seeding rate, 60 kg/Ha of tall fescue, *Festuca arundinacea*). Two factors were evaluated in a split plot design with five replicates. Main plots compared under-trellis cover crop (UTCC) with conventional under-trellis herbicide floor management (H); and subplots compare the effects of two planting density (PD) (0.8 and 1.5 m between plants), giving a total of four treatments: 0.8H, 1.5H, 0.8UTCC and 1.5UTCC. Since in most Uruguayan vineyards between vines spacing ranged from 0.8 to 1.1 m apart, the combination H treatment and 0.8 PD is considered the Control or standard treatment (0.8H-control). The UTCC treatment consisted of the full cover of the vineyard soil with tall fescue. The conventional management scheme used the same inter-row groundcover except with a 1.0 m wide weed-free strip under the trellis. The under-trellis, weed-free strip was maintained with a combination of herbicides. The five replicate subplots were each comprised of eight adjacent vines but only the central six were evaluated. Buffer rows separated ground cover (GC) treatments, following the same vine spacing as evaluated plots.

To avoid the effect of the treatment due to nitrogen (N) competition, in every UTCC plot ammonium nitrate (NH4NO3) was applied twice at a rate of 20 kg/ha N when shoots reached approximately 30 cm and after fruit set (stage 29 - Eichhorn and Lorentz, 1977). No statistically significant differences among treatments were detected in leaf N%, P%, K%, Ca% and Mg% at bloom or veraison (data not shown). Average leaf N% content ranged from 2.1 to 2.5%. No visual nutrient deficiency symptoms were detected.

To monitor water stress in the treatments, midday stem water potential ($\Psi$stem) was periodically measured (~ bi-weekly) between 1400 and 1600 h using a leaf pressure chamber (Soil Moisture Equipment Corp., Santa Barbara, CA) on two leaves per treatment replication (Chone et al. 2001). Plastic bags covered with aluminum foil were placed on well-exposed mid-shoot leaves one hour before the onset of Ψstem measurements. The time from cutting leaf petioles to insertion and chamber pressurization was less than 30 sec. The chamber was pressurized with nitrogen gas at a rate of 2 to 4 sec/0.1 MPa.

Normally no irrigation was needed pre-bloom. However, pre-bloom irrigation was needed only during the last season in UTCC treatments, for a 10 days period (from 20 to 10 days before bloom) at an average rate of 5 mm/day to ensure adequate early canopy development. This was done in plots with average pruning weight per meter of trellis < 0.4 Kg/m as without pre-bloom irrigation the vine growth and capacity would have been excessively reduced.

In order to avoid severe post-bloom water stress, deficit irrigation (70% ETc) was applied for all treatment replicates once the plants reached -0.9 MPa mid-day stem water potential (Ψstem). To avoid the Ψstem becoming more negative than -1.1 MPa after a prolonged period of deficit irrigation, occasionally the amount of water the vines had consumed the previous week was applied at 100% ETc.

ETo was obtained from the weather station of INIA Las Brujas Experiment Station located < 200 m from the experimental site (Allen et al., 1998). Mid-day canopy shaded area (light interception) was estimated at solar noon using a Solar panel (Paso panel, http,//cesanluisobispo.ucanr.edu/Viticulture/Paso_Panel/) and each plot's crop coefficient (Kc) was calculated using the formula proposed by Williams and Ayars (2005); Kc = (0.017 * Shaded percentage of field) – 0.008. ETc was calculated by multiplying ETo by Kc and used to estimate weekly vine water consumption.

3.3.4. Vegetative measurements

Shoot growth rates were obtained by repeated measures of shoot length. Two shoots from two representative vines per plot were tagged and measured on a ~ weekly basis starting shortly after bud-break. Total leaf area (TLA) of tagged shoots was estimated based on the relationship between leaf blade length and leaf area, according to the equation developed specifically for Tannat by Disegna et al. (2003), LA/leaf (cm2) = 44.4x2 + 1541.3x – 4381.1 R2= 0.963. Leaf blade length of those tagged shoots was measured at veraison and prior to harvest. Measured canes were later weighed at pruning time. The relationship between cane pruning weight and leaf area of the tagged shoots was determined via regression analysis. Quadratic equation was used to fit the data, with R2= 0.91. Based on this relationship, total leaf area per vine was estimated by weighing individually every cane from each vine. Canopy surface area (SA) at each side of two plants per plot was also estimated according to Schneider (1989). The width and height of the canopy were collected at four different spots along the vine using a measurement tape. To estimate vine canopy density, Leaf index (LI) of those plants was calculated using the equation, LI = SA/TLA as proposed by Schneider (1989).

At veraison, photosynthetically active radiation (PAR) available in the fruit zone, was estimated with an average of three readings taken in the canopy fruit zone, with the ceptometer (AccuPAR L80; Decagon Devices, Pullman, WA) inserted parallel to and 15 cm above the cordon. Since the clusters are not flat, the light availability was estimated ate three angles one with the sensors face angled 45° to the east, a second vertically upright, and the third angled 45° to the west. Incident radiation was measured by orienting the ceptometer vertically upright, at the fruit zone height outside the canopy in the alleyway (ambient). PAR measurements were made ±2 hours of solar noon on cloudless days. In the winter, the prunings of every experimental plant were weighed and averaged by plot. The Ravaz index of crop load (RI, fruit yield/pruning weight) and average shoot pruning weight were then calculated. Every cane from tagged plants was individually weighed and average cane pruning weight of those selected plants was calculated as an expression of shoot development.

3.3.5. Berry development and fruit composition

From fruit set to harvest, berry samples were randomly collected from each treatment replicate. Eighty berries from fruit set to veraison and one hundred berries from veraison to harvest were collected on a bi-weekly basis. In pre-veraison samples only berry weight was analyzed. In post veraison samples, berry weight, total soluble solids (TSS), titratable acidity (TA), pH, tartaric and malic acids and total anthocyanin were analyzed. TSS was determined using a hand refractometer (Atago, model N1, Tokyo, Japan). Must pH was measured with a pH meter (Horiba, model F13, Kyoto, Japan) and titratable acidity (TA) was determined by titration (NaOH 0.1 N) and expressed as tartaric acid equivalents (w/w). The concentration of malic and tartaric acids was measured by HPLC. Total anthocyanin analyses were performed according to Glories (1984). From veraison to harvest, the percentage of bunches infected by *Botrytis* bunch rot (incidence) as well as the percentage of each bunch that was infected (severity) was determined by visual inspection using a six-point scale (0, 5, 25, 50, 75 and 100%). *Botrytis* severity (S) was calculated as follows, $S = \Sigma Si/n$; where Si = % severity for the ith bunch and n = the total number of affected bunches.

3.3.6. Harvest and yield

All treatments were harvested at the same date. In addition to chemical evaluations, bunch rot incidence and severity were monitored to help define the requirement for early harvest to avoid excess loss due to bunch rot. Total fruit yield and clusters per vine were determined for each experimental plot. Mean cluster weight was calculated. From the harvested fruits, about 10 kg of grapes per plot were crushed and juice TSS, pH, TA, malic and tartaric acids were analyzed. In addition, samples of 200 berries per replication were taken, to prepare homogenized fruit extracts. From these extracts, total anthocyanin was analyzed as previously described.

3.3.7. Wine making and analysis

Approximately 10 kg of fruit per treatment replicate were retained for winemaking. After storage for 16hr at 5 ºC, grapes were crushed and destemmed. Must samples were taken, and grapes from adjacent field blocks (B) were combined (B1 + B2 + B3/2 and B3/2 + B4 + B5) giving two wine replicates for each treatment.

Sulfur dioxide was added at a rate of 60 mg/kg and the must was inoculated with *Saccharomyces cerevisiae* (ALG111, DSM, Delft, The Netherlands) at 25 g/hL. Fermentation temperature was maintained between 26 and 30 ºC. Alcoholic fermentation was completed six to eight days after the beginning of fermentation, but maceration was allowed for 10 days. After maceration was completed, wines were pressed, and placed into sterile 10-L glass containers where malolactic fermentation was completed. After completing malolactic fermentation, wines were cold-stabilized at 0 ºC for two weeks, $SO_2$ was added and the wines were stored in sterile 5-L glass containers at 11 ºC. For each wine, a 125 mL sample was taken for analysis (alcohol concentration, TA, pH, tartaric and malic levels) by standard methods (OIV 2009).

Total anthocyanins were calculated from the absorbance at 520 nm as described by Glories (1984).

3.3.8. Statistics

A split plot design was used to analyze the significance of treatments main effects and their interactions using MSTAT-C computer software (Michigan State University). The fixed effects of the model were under-trellis ground cover (UTCC vs. H), plant density (0.8 vs. 1.5 m between plants) and their interactions; the random effects were block interactions with main effects. A Tukey HSD test (5% significance level) was used to compare treatment means.

## 3.4. RESULTS

3.4.1. Weather conditions

Accumulated growing degree days, base 10C (GDD) and precipitation occurred during the three growing seasons are presented in Table 1. The three growing seasons were characterized by variable rainfall amounts and patterns. Precipitation accumulated during 2011/12 was below average in every phonological period, and irrigation was needed also in H treatment to avoid severe vine water stress. During 2012/13 precipitations occurred early in season was almost double of the historical value, while from veraison to harvest was below average. Precipitation occurred during 2013/14 was close to average until veraison end extremely rainy from veraison to harvest (Table 1). On the other hand total GDD accumulated during the three growing seasons was similar to the historic average for the region (≅2150 GDD).

3.4.2. Vegetative growth

Both the planting density (PD) and the under-trellis cover crop (UTCC) significantly affected vegetative growth (cane pruning weight, TLA/m, pruning weight/m, shaded area/m) and its associated parameters of canopy density (PAR% in fruit zone, LI) (Table 2).

During the first season, final PW/m reflected the different shoot growth rate among treatments during the 30 days period after bloom. No substantial differences in shoot growth rate were detected previous to or after this period among treatments (Figure 1). Final average shoot length of individual plots was well correlated with the mid-day Ψstem of that period ($R^2$= 0.69) (average of three measuring dates) (Figure 2). Shoot growth rates post-bloom were correlated to mid-day Ψstem between -0.35 and -0.60 MPa, while no significant growth was detected in any period when the Ψstem was lower than -0.9 MPa. During the last two seasons, vine water status during the 30 days period after bloom was always above -0.5 Mpa Ψstem in all treatments, so the correlations observed were not as strong.

Vine pruning weights in the second and third years maintained the differences observed in the first year even though the water stress was less severe, especially in the third year (Table 2).

**Table 2** Canopy characteristics and *Botrytis* bunch rot incidence and severity of Tannat grapevines as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

| | | **Herbicide** | | **[a] UTCC** | | **Significance** | | |
|---|---|---|---|---|---|---|---|---|
| | | **0.8 m** | **1.5 m** | **0.8 m** | **1.5 m** | **GC** | **PD** | **GC*PD** |
| Harvest 2012 | Cluster weight (g) | 393 | 381 | 359 | 357 | ns | ns | ns |
| | Berry weight (g) | 2.12 a [c] | 2.17 a | 1.88 b | 1.72 b | <0.01 | ns | ns |
| | Vine yield (kg/m) | 3.60 | 3.53 | 3.31 | 2.78 | ns | ns | ns |
| | [b] Potential vine yield (kg/m) | 5.54 a | 4.82 ab | 4.66 b | 3.47 c | <0.01 | <0.01 | ns |
| | Pruning weight (kg/m) | 0.88 a | 0.68 b | 0.50 c | 0.40 d | <0.01 | <0.01 | ns |
| | Cane pruning weight (g) | 82.9 a | 58.4 b | 47.6 bc | 32.0 c | 0.07 | 0.05 | ns |
| | Total leaf area ($m^2$/m) | 10.2 a | 8.02 b | 7.14 c | 5.34 d | <0.01 | <0.01 | ns |
| | Leaf Index | 0.35 c | 0.46 b | 0.49 b | 0.60 a | <0.01 | <0.01 | 0.02 |
| | PAR % | 3.93 c | 7.35 b | 12.07 a | 18.33 a | <0.01 | 0.03 | ns |
| | Shaded area $m^2$/m | 0.97 a | 0.85 a | 0.70 b | 0.63 b | <0.01 | ns | ns |
| | Ravaz index | 4.2 c | 5.2 b | 6.7 ab | 8.5 a | 0.02 | 0.05 | ns |
| | *Botrytis* incidence (%) | 13.2 a | 11.8 a | 2.9 b | 2.7 b | <0.01 | ns | ns |
| | *Botrytis* severity (%) | 12.0 a | 13.2 a | 3.6 b | 0.8 b | <0.01 | ns | ns |
| Harvest 2013 | Cluster weight (g) | 430 a | 370 ab | 300 b | 260 b | <0.01 | ns | ns |
| | Berry weight (g) | 1.77 | 1.79 | 1.6 | 1.68 | ns | ns | ns |
| | Vine yield (kg/m) | 3.94 a | 3.52 a | 2.89 b | 2.32 b | <0.01 | <0.01 | ns |
| | [b] Potential vine yield (kg/m) | 5.55 a | 5.98 a | 4.37 b | 3.70 c | <0.01 | 0.03 | 0.03 |
| | Pruning weight (kg/m) | 0.92 a | 0.71 b | 0.50 c | 0.34 d | <0.01 | <0.01 | ns |
| | Cane pruning weight (g) | 65.5 a | 50.0 b | 39.3 c | 35.1 c | <0.01 | ns | ns |
| | Total leaf area ($m^2$/m) | 10.2 a | 7.56 b | 6.06 c | 3.99 d | <0.01 | <0.01 | ns |
| | Leaf Index | 0.37 c | 0.47 b | 0.55 b | 0.72 a | <0.01 | <0.01 | ns |
| | PAR % | 3.83 d | 7.03 c | 13.9 b | 17.11 a | <0.01 | 0.01 | ns |
| | Shaded area $m^2$/m | 0.91 a | 0.76 ab | 0.65 bc | 0.57 c | <0.01 | ns | ns |
| | Ravaz index | 4.58 c | 4.99 bc | 6.24 ab | 6.80 a | 0.04 | ns | ns |
| | *Botrytis* incidence (%) | 45.2 a | 18.0 b | 3.8 c | 0.8 d | <0.01 | <0.01 | ns |
| | *Botrytis* severity (%) | 32.6 a | 31.8 a | 15.6 b | 10.0 b | <0.01 | ns | ns |
| Harvest 2014 | Cluster weight (g) | 274 a | 283 a | 243 b | 248 b | <0.01 | ns | ns |
| | Berry weight (g) | 1.97 | 2.00 | 2.07 | 2.06 | ns | ns | ns |
| | Vine yield (kg/m) | 2.69 a | 2.44 ab | 1.93 b | 1.74 b | <0.01 | ns | ns |
| | [b] Potential vine yield (kg/m) | 3.45 a | 3.03 a | 2.39 b | 2.23 b | <0.01 | ns | 0.02 |
| | Pruning weight (kg/m) | 0.81 a | 0.76 a | 0.56 b | 0.33 c | <0.01 | <0.01 | <0.01 |
| | Cane pruning weight (g) | 74.7 a | 73.9 a | 59.6 b | 44.5 c | <0.01 | ns | ns |
| | Total leaf area ($m^2$/m) | 8.04 a | 7.64 a | 5.68 b | 3.38 c | <0.01 | <0.01 | ns |
| | Leaf Index | 0.49 c | 0.51 c | 0.61 b | 0.88 a | <0.01 | <0.01 | 0.02 |
| | PAR % | 4.9 c | 6.6 c | 10.1 b | 21.2 a | <0.01 | 0.01 | ns |
| | Shaded area $m^2$/m | 0.88 a | 0.83 a | 0.81 a | 0.69 b | <0.01 | 0.04 | ns |
| | Ravaz index | 3.28 b | 3.27 b | 3.56 b | 5.37 a | <0.01 | <0.01 | ns |
| | *Botrytis* incidence (%) | 55.9 a | 45.6 b | 8.1 c | 8.0 c | <0.01 | <0.01 | ns |
| | *Botrytis* severity (%) | 13.9 a | 5.9 b | 0.1 c | 0.2 c | <0.01 | <0.01 | ns |

[a] UTCC: Under trellis cover crop; PD: Planting density; GC: Under trellis ground cover. [b] Potential yield = Vine yield at harvest + thinned clusters weight at veraison.[c] Significance of treatments and interactions (p >F; ns: not significant). Values with different letters in single rows are significantly different at $p < 0.05$

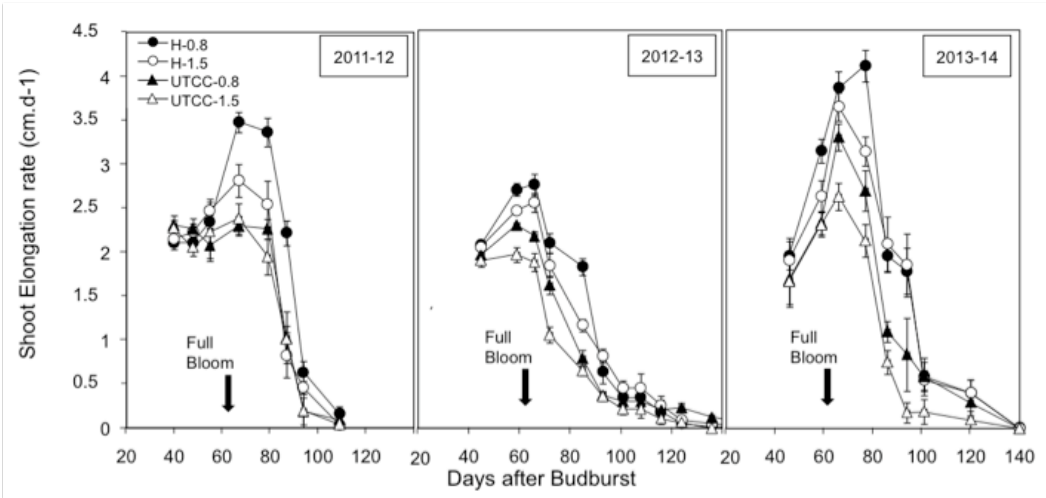


Figure 1. Shoot elongation rate of Tannat grapevines as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

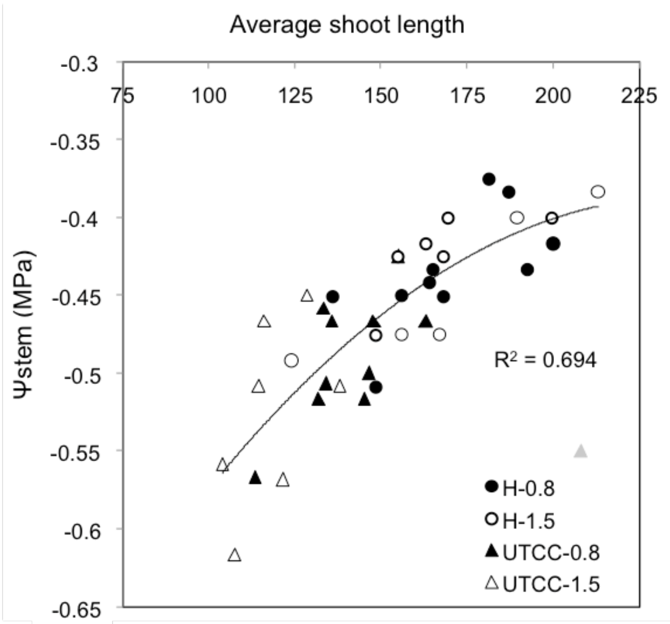


Figure 2. Relationship between Ψstem (average of four measurements during the 30 days after bloom period) and average shoot length of Tannat grapevines as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop) during spring 2011 in southern Uruguay.

Associated with vine water status the UTCC factor exhibited a more pronounced effect over vegetative growth as compared to PD (Figures 1 and 2). Over the three seasons UTCC treated vines produced lower PW/m than the H treatments (Table 2). The 0.8 m spacing produced higher PW/m compared to the 1.5 m spacing in both herbicide and UTCC treatments. The combination PD1.5-UTCC exhibited every season the most pronounced reduction of vegetative growth (PW/m in PD1.5- UTCC ~ 40% of that observed in PD-0.8-H treatment), even though it was the most irrigated treatment (Figure 3, Tables 1 and 2). A significant interaction between UTCC and PD factors was detected only in the last season when 1.5m spacing treatment had a stronger reduction in PW/m in UTCC than in H treatment. Vine PW/m over three seasons were consistent and did not show cumulative effects (Table 2) even in the most restrictive treatment (1.5-UTCC). Measurements of canopy ground shade (vine capacity) showed that above about 0.6 Kg of PW/m there was minimum increase in vine light interception and thus vine capacity, while LI observed in those vines was lower than 0.65 (Figure 4 and Table 2).

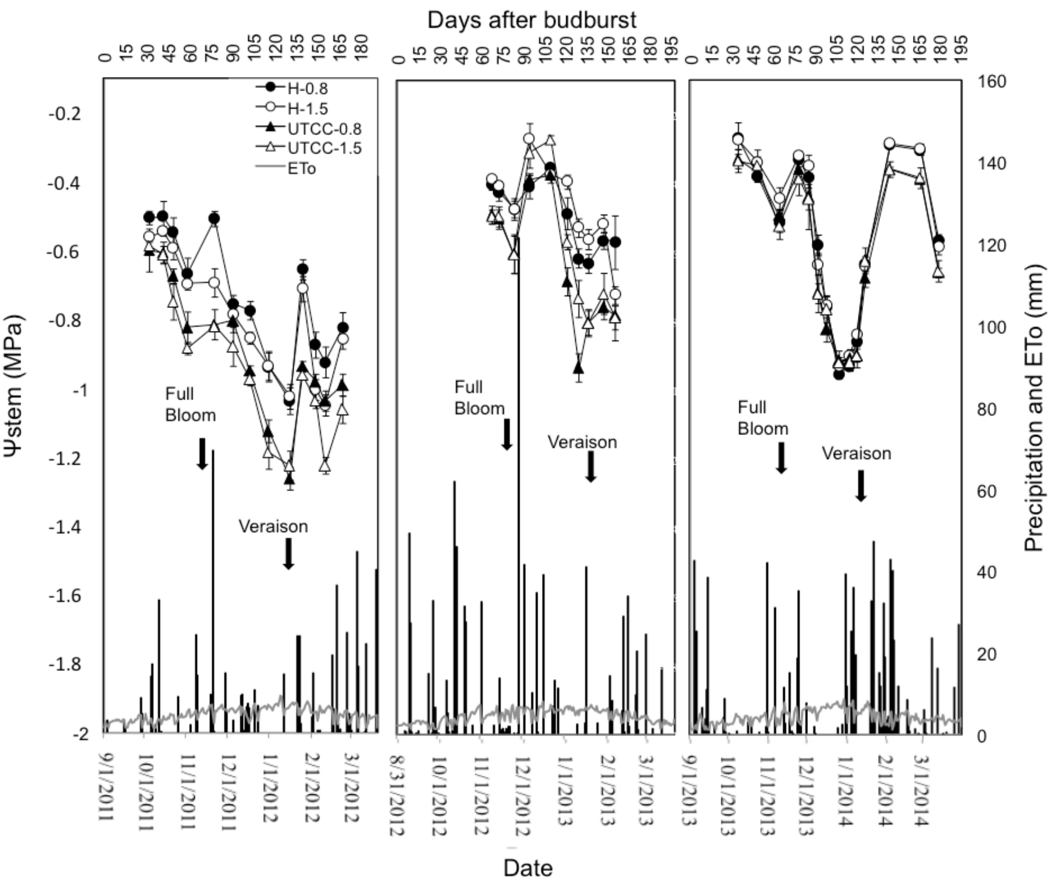

Figure 3. Midday Ψstem of Tannat grapevines as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay. Daily precipitation and ETo from budburst to harvest.

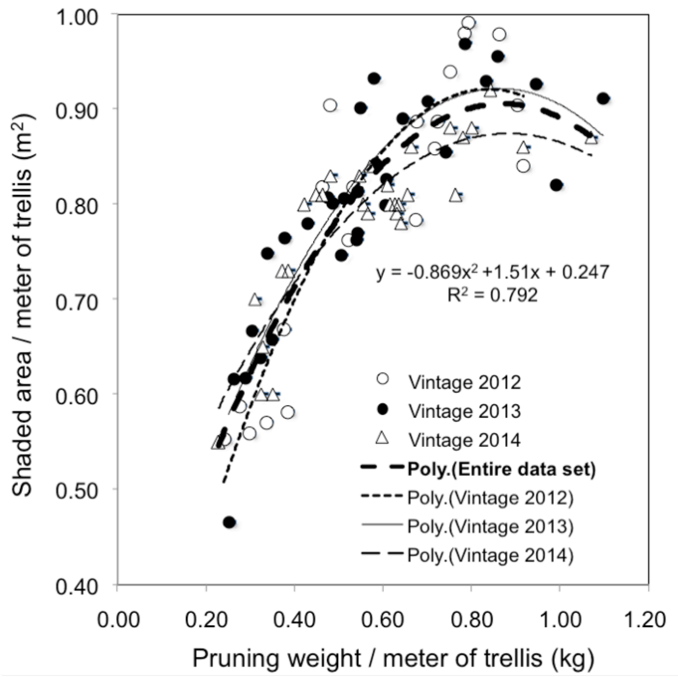


Figure 4. Relationship between pruning weight and shaded area per meter of canopy length of Tannat grapevines trained under VSP system, during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

3.4.3. Fruit yield

Final berry size in 2012 harvest, the first year of treatment, was positively correlated with vegetative growth as expressed by PW/m (R2=0.69 - data not shown). Independent of PD, berry weight was significantly reduced by UTCC (Table 2). Differences observed in berry weight where not detected in cluster weight or yield/m (1 cluster/shoot) (PD significance = 0.053) at harvest 2012. In contrast at harvest in 2013 and 2014 the berry weight was not affected by treatments, while UTCC significantly reduced cluster weight, and yield/m.

The average potential yield (harvested + thinned clusters) of the seasons was 17.3, 16.4, 13.6 and 11.2 Ton/ha for 0.8H-control, 1.5H, 0.8UTCC and 1.5UTCC treatments respectively. In general UTCC had greater effect on yield components as compared to PD, while a significant interaction PD x Ground Cover for potential yield was detected the last two seasons. The combination 1.5UTCC exhibited the most pronounced reduction of potential yield (~ 35% less fruit than the highest yield observed in the 0.8H-control treatment). The 0.8UTCC potential yield was ~ 20% lower fruit compared with 0.8H-control treatment. Treatments potential yield/m (harvested + thinned fruits at veraison) was related to final canopy size (Shaded area/m, TLA/m or PW/m) (Table 2).

3.4.4. Berry must and wine composition

With the exception of total anthocyanins in UTCC treatments at the 2013 harvest, PD did not significantly affect fruit composition (Table 3). The combination Block-PD factors produced a significant variation in vegetative expression among plots (PW/m from 0.53 to 1.07). However, when yields of individual plots were adjusted to >1.8 m2 of leaf area/kg of fruit (by cluster thinning), no significant correlations between vigor of individual H plots (PW/m or individual shoots pruning weight) and fruit composition were detected (data not shown). As compared to the H treatment, UTCC treatment significantly increased every season the fruit TSS (Table 3). Total anthocyanin concentration at 2012 harvest was also significantly increased by UTCC, however in 2013 harvest anthocyanin concentration was significantly higher only in the PD0.8-UTCC treatment. Associated with similar water availability during the entire season, treatments had not a significant effect over berry size or anthocyanin concentration at harvest 2014. No delays or advances in maturation were detected between treatments. The evolution of soluble solids and anthocyanin accumulation in grapes during maturation in 2014 is presented as an example (Figure 5).

Wine anthocyanins were higher for the UTCC treatments as compared to the H treatment every season. This was the only wine parameter evaluated, which was consistently increased by UTCC treatment, while no effect of PD was detected. UTCC treatment reduced must TA, while tartaric/malic ratio and pH was not significantly affected (Table 3). Even though fertilization was applied in UTCC plots (40 Kg/Ha N each season) to avoid nitrogen deficiency effects, must YAN concentration was significantly reduced by UTCC treatment. With the exception of anthocyanin concentration in 2013 harvest, planting density did not significantly affect berry or must composition (Table 3).

**Table 3.** Grape must and wine composition as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

| | | | Herbicide | | [a] UTCC | | Significance | |
|---|---|---|---|---|---|---|---|---|
| | | | 0.8 m | 1.5 m | 0.8 m | 1.5 m | GC | PD |
| Harvest 2012 | Grapes and must composition | Soluble solids (Brix) | 24.2 b [b] | 23.9 b | 25.4 a | 25.2 a | <0.01 | ns |
| | | Titratabe acidity (g/L) | 6.16 a | 6.18 a | 5.67 b | 5.61 c | 0.02 | ns |
| | | pH | 3.46 | 3.43 | 3.41 | 3.41 | ns | ns |
| | | Total anthocyanins (mg/L) | 1389 b | 1369 b | 1504 a | 1511 a | 0.01 | ns |
| | | Tartaric / Malic | 2.27 | 2.35 | 2.55 | 2.46 | ns | ns |
| | | YAN (mg/L) | 91.2 a | 82.4 ab | 70.7 b | 72.6 b | 0.025 | ns |
| | Wine composition | Titratable acidity (g/L) | 6.14 | 6.15 | 6.14 | 5.69 | ns | ns |
| | | pH | 3.72 | 3.67 | 3.72 | 3.66 | ns | ns |
| | | Ethanol (% v/v) | 14.1 | 13.9 | 14.6 | 14.7 | ns | ns |
| | | Anthocyanins (mg/L) | 1077 | 1065 | 1296 | 1570 | <0.01 | ns |
| Harvest 2013 | Grapes and must composition | Soluble solids (Brix) | 25.8 b | 26.0 b | 27.4 a | 27.4 a | <0.01 | ns |
| | | Titratable acidity (g/L) | 7.61 a | 7.71 a | 6.90 b | 6.65 b | <0.01 | ns |
| | | pH | 3.43 | 3.41 | 3.44 | 3.42 | ns | ns |
| | | Total anthocyanins (mg/L) | 1563 b | 1495 b | 1615 a | 1527 b | ns | <0.01 |
| | | Tartaric / Malic | 1.87 | 1.80 | 1.95 | 1.91 | ns | ns |
| | | YAN (mg/L) | 101.0 a | 108.2 a | 78.7 b | 70.1 b | <0.001 | ns |
| | Wine composition | Titratable acidity (g/L) | 6.72 | 7.78 | 7.08 | 7.24 | ns | ns |
| | | pH | 4.00 | 3.9 | 4.0 | 3.91 | ns | ns |
| | | Ethanol (% v/v) | 15.4 | 15.4 | 15.5 | 15.6 | ns | ns |
| | | Anthocyanins (mg/L) | 1385 | 1430 | 1540 | 1474 | <0.01 | ns |
| Harvest 2014 | Grapes and must composition | Soluble solids (Brix) | 22.1 b | 21.8 b | 22.7 a | 22.5 a | <0.01 | ns |
| | | Titratable acidity (g/L) | 8.03 a | 8.00 a | 7.53 ab | 6.98 b | <0.01 | ns |
| | | pH | 3.23 | 3.25 | 3.22 | 3.22 | ns | ns |
| | | Total anthocyanins (mg/L) | 1284 | 1259 | 1282 | 1208 | ns | ns |
| | | Tartaric / Malic | 3.03 | 3.26 | 3.09 | 3.33 | ns | ns |
| | | YAN (mg/L) | 106.4 a | 103.9 a | 84.5 b | 81.3 b | <0.01 | ns |
| | Wine composition | Titratable acidity (g/L) | 7.33 | 7.48 | 6.50 | 6.42 | <0.01 | ns |
| | | pH | 3.69 | 3.63 | 3.79 | 3.78 | ns | ns |
| | | Ethanol (% v/v) | 12.2 | 12.3 | 11.7 | 12.5 | ns | ns |
| | | Anthocyanins (mg/L) | 807 | 841 | 860 | 951 | <0.01 | ns |

[a] UTCC: Under trellis cover crop; PD: Planting density; GC: Under trellis ground cover. [b] Significance of treatments and interactions (p >F; ns: not significant). Values with different letters in single rows are significantly different at $p < 0.05$. No significant interactions (GC*PD) were detected.

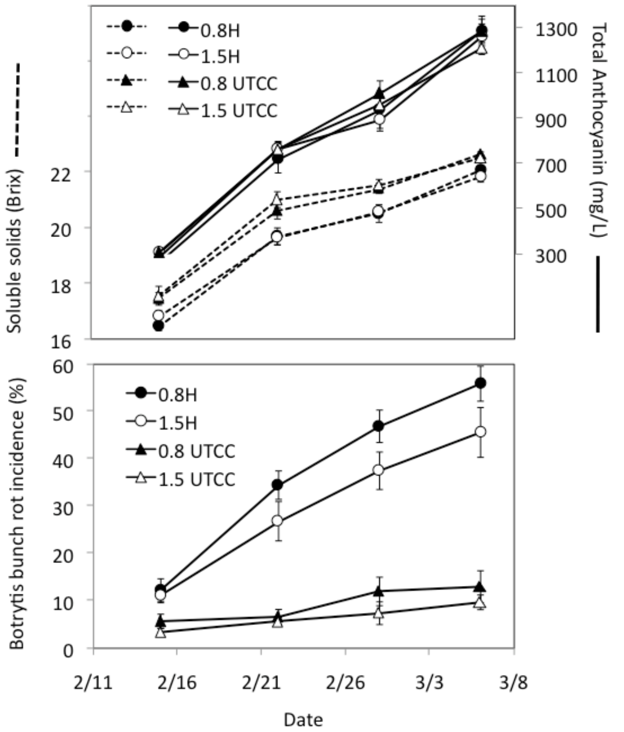


Figure 5. Evolution of total anthocyanin, soluble solids, and *Botrytis* bunch root incidence of Tannat grapevines growing as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop) during 2013/14 growing season in southern Uruguay.

### 3.4.5. *Botrytis* bunch rot

During the study bunch rot development was positively related to vine vegetative growth (Figure 6). High *Botrytis* bunch rot incidence was detected when pruning weight per meter of canopy length increased over about 0.5 kg/m. Below this threshold bunch rot incidence was always under 10% (Figure 6). Therefore, bunch rot incidence in the UTCC treatment was in all 3 years 80 to 85 % lower than in the H treatment (Table 3). During the last two seasons, PD also affected *Botrytis* bunch rot incidence. *Botrytis* incidence was significantly lower in 1.5 versus 0.8 vine spacing. Bunch rot severity was every season significantly lower in UTCC treatments as compared to H treatments while higher PD resulted in greater bunch rot severity in 2013 and 2014 (Table 3). Differences of *Botrytis* bunch root incidence and severity were already significant in every season at least 15 days previous to harvest. Data for 2014 vintage is presented as an example (Figure 5), but disease development was comparable every season.

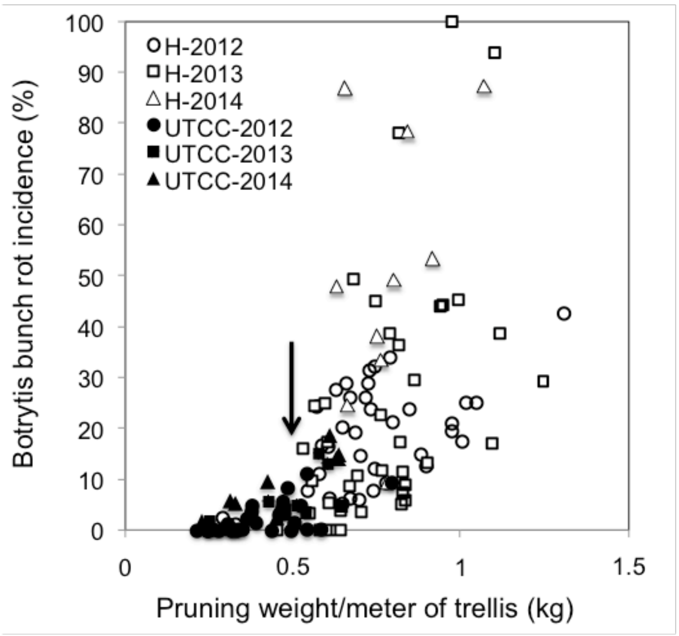


Figure 6. Relationship between pruning weight per meter of trellis and *Botrytis* bunch root incidence of Tannat grapevines as affected by planting density (0.8 and 1.5 m) and groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

## 3.5. DISCUSSION

### 3.5.1. Planting Density

Since there were very few interactions of Planting Density and UTCC, the main effects will be discussed separately. Of the two treatments designed to provide competition to reduce vegetative growth, the planting density had many fewer effects than UTCC. The primary effect of closer vine spacing to 0.8m was to increase shoot vigor, in terms of post-bloom growth rate, final cane weight and leaf area, rather than reduce it.

This suggests that in these conditions of climate and soil, the closer spacing led to a root/shoot imbalance. The shoot numbers per vine and vine light interception were reduced by almost 50%, yet the reduction of 50% in planting density did not appear to proportionately reduce the ability of the root system to support growth. Hatch et al. (2012) used small root restriction bags to reduce soil volume to about 1% compared to controls, yet found only about a 30-40% of control pruning weight. Thus, the greater growth rate of the reduced shoot numbers may be expected, in many ways analogous to a severe pruning effect on root/shoot balance.

Potential average yield was also significantly reduced as planting density was reduced. This suggests that the vines balanced their crop to the available resources (canopy size) (Winkler et al. 1974). Contrarily to what has been reported (Dry et al. 2005, Smart and Robinson 1991) the significant differences observed in vegetative growth and its associated parameters of canopy density between PD treatments, did not consistently affect fruit or wine composition.

3.5.2. UTCC Effects

As previously reported UTCC effectively reduced excessive vegetative growth of grapevines (Giese et al. 2014, Hatch et al. 2012). This provided the ability to reduce excessive growth to the desired benchmark range of 0.30–0.60 kg PW/m (Smart and Robinson, 1991) which translates to 25-45 grams cane weights. PW/m values in H treatments were always above this optimal range, which would indicate an excessively shaded canopy with the standard H practice while not increasing vine potential yield, but increasing *Botrytis* incidence. Vegetative growth in the 0.8UTCC treatment would be considered near optimal range (PW/m ~ 0.5 Kg, ~35-40 gram canes) since it results in a minor reduction of vine capacity (shaded area under the vine), and reduced canopy density (LI) yet a major reduction in *Botrytis* incidence. The Leaf Index observed in the 0.8 UTCC vines (LI >0.65) is associated with the fruiting zone being well exposed to light (Coniberti et al. 2012). The 0.33-0.34 kg PW/m values observed in the 1.5UTCC treatment were near the minimum recommended value, suggesting an excessive depression of vegetative growth and thus vine capacity (shaded area) in this study. Further declines were prevented by the stress-based supplemental irrigation. The strong vigor control achieved for the combined effects of UTCC and 1.5m spacing could be useful for reducing vigor to optimal levels in sites with even higher vine capacity potential.

Although other factors, such as cover crops sequestering mineral nutrients (especially N), have been reported affecting vegetative development (Hatch et al. 2011, Giese et al. 2014), we did not find any clear effects on tissue nutrient status as mentioned earlier. However, the first season vegetative growth was well correlated with vine Ψstem ($R^2$= 0.69). Similarly, Hatch et al. (2011) reported that the use of perennial UTCC decreased shoot growth through bloom. Tesic et al. 2007, also reported that associated with the early reduction of soil water availability, UTCC decreased shoot growth from bud-break through bloom compared with inter-row cover or bare soil strip in semi-arid climates. Centinari et al. (2016), testing the effect of root pruning and the use of annual UTCC on grapevine vegetative growth, reported that over the course of the entire experiment a decrease in grapevine size was linearly related to the decrease in Ψstem ($R^2$= 0.71) but poorly correlated with petiole N concentration at bloom or veraison.

During the last two seasons, vine post bloom period vine Ψstem did not fall below −0.5 MPa in any treatment during bloom-30 days after bloom, however vegetative growth parameters (cane pruning weight, PW/m, LI) were comparable with first season in all treatments. Similar results were previously reported by Giese et al. (2014), when in a 6-year study, consistent effects on vine vegetative growth were achieved after a below average rainfall season (300 mm), even when in following seasons vine Ψstem did not fall below −0.6 MPa on any measured date. The greatest average difference observed in PW/m between consecutive seasons vine size in this study was ∼10%. Similar results were also reported in previous studies (Hickey et al. 2016, Giese et al. 2015) when from season to season PW/m differences were always less than 30%. Therefore, vine capacity may be initially related to water stress, but apparently remains limited not just by current environmental conditions but also by its previous history (Winkler et al. 1974). The mechanism of this effect is not clear.

UTCC may provide one strategy to control vine size, even when there is no restriction of water availability as seen in 2012/13 and 2013/14 seasons. Morlat and Jacquet (2003) found that the greater the number of Festuca roots in the root zone the fewer the grape roots. Competition from cover crop roots can decrease vine root growth in shallow soils (Centinari et al. 2016, Morlat and Jacquet 2003) and alter grapevine-rooting patterns (Celette et al., 2008), which may impact the vine's uptake of water and nutrients as soil water and nutrient availability generally varies with depth.

This suggests that soil depth, water holding capacity and nutrient levels may affect the outcome of any cover crop treatment. The soil structure and depth may be important in this competition. When the soil is deep and fertile grape roots may be able to re-distribute deeper over time to avoid the direct competition from the cover crop (Pool and Lakso, unpublished data). In shallow restricted soils or those with poor water or nutrient supplies at depth, the vines may be limited to the zone of competition. The spatial distribution of root growth as affected by treatments was not determined in this study.

No blade or petiole N content differences were detected during our study. However, must YAN was every season significantly reduced by UTCC by about 20 mg/L. The higher YAN content observed in H treatments (average values for the region) suggests that vines from UTCC have a lower nitrogen uptake (Table 2).

According with many other studies (Centinari et al. 2016, Hickey et al. 2016, Karl et al. 2016, Hatch et al. 2011), potential yield was significantly reduced by UTCC. In the present study berry size for the first season was well correlated with PW/m ($R^2$=0.69), however differences in berry weight, did not translate into significant cluster weight or yield/m differences. For harvest 2013 and 2014 berry weight was not affected by treatments, but UTCC significantly reduced cluster weight via reductions in berry number. Higher precipitation rates occurred during 2012-13 and 2013-14 seasons (above the average) may explain no significant differences in berry size. Centinari et al. (2016) have shown a correlation between decreased vine vegetative growths and yield in UTCC, associated with vine water status. However, even with no water deficit in this study, yield was significantly reduced with UTCC in 2012-13 and 2013-14 seasons. The analysis requires highlighting some key features of yield formation. Grapevine yield is affected by current season climatic conditions, water and nitrogen status, but yield formation extends over two consecutive years and the main components of grape yield (bunch number per vine and berry number per bunch) are determined primarily by light, temperature, water status and assimilate supply to the buds in the previous bloom (Guilpart et al. 2017; Keller 2005, Buttrose 1974). So the occurrence of any environmental constraint during this period may be expected to have an important impact on next season yield.

No or limited impacts on fruit composition were reported in previous studies of UTCC (Hickey et al. 2016, Karl et al. 2016, Giese et al. 2015). In our study UTCC treatment consistently increased grape TSS and reduced TA while pH and tartaric to malic ratio were not affected. It should be noted that due to the cluster thinning at veraison, the yields in these trials were low to moderate crop levels generally varying from about 6 to 14 tonnes/ha and Ravaz Indices <7. Thus, excessively heavy crops were avoided. Associated with a lower berry size, during the first seasons UTCC also increased grape total anthocyanins by about 10% compared to the herbicide treatments. Probably related to similar water status among treatments during the entire 2013/14 season, no effect over berry size or grape total anthocyanin concentration was detected at harvest 2014. On the other hand, wine anthocyanins concentration was always enhanced by UTCC.

Bunch rot development was highly related to vine vegetative growth. The results suggest that susceptibility of clusters/grapes to bunch rot may increase when PW/m exceeds about 0.5 kg/m or cane weights of 40-50 grams. Although *Botrytis cinerea* damage can be reduced as a result of an increased light exposure (PAR %) of the fruit zone (Reynolds et al. 1996), the strong effect observed in this study could not be explained by fruit exposure alone. The significantly lower berry weight (reducing cluster compactness) and cluster weight (Hed et al. 2009) and also nitrogen grape content (YAN) (Van Zyl and Van Huyssteen 1980), may play an important role on the disease occurrence.

When evaluating vine vegetative balance in a humid climate, it is relevant to consider that in many seasons *Botrytis* bunch rot may be the main factor defining harvest time. In many cases fruit is harvested without achieving full maturation to avoid excessive bunch rot. In dry areas, vines are considered being in vegetative balance when growth fills the trellis with shoots of 1.2-1.4 m to ensure enough light interception to fully support the crop development while avoiding unnecessary additional growth that causes excessive shade (Kliewer and Dokoozlian 2005) or additional canopy management. PW/m of canopy length is widely used as an indicator of vine vegetative balance. A relatively wide range of pruning weights of 0.3 to 0.6 Kg/m has been associated with balanced vines (Smart and Robinson 1991), though optimal values would be specific for a given variety, region and trellis system (Dry et. al. 2005).

In our study, bunch rot progressively increased when PW/m exceeded 0.5 kg/m. UTCC led to a reduction of potential yield compared to excessively vigorous H vines (PW/m > 0.6 kg), with a consistent improvement of fruit or wine composition (TSS increases from 0.5 to 1.5 Brix, fruit anthocyanin from 0 to 8%, and wine anthocyanin concentration from 7 to 20% when fruit is picked at the same time.

However, in a commercial vineyard, due to the high bunch rot incidence (20% bunch rot incidence is commonly used as maximum threshold), fruit from H treatments in our study (vigorous vines) would have been harvested in every season prior to full maturation. To evaluate the bunch rot effect on harvest date decisions, the evolution of TSS and anthocyanin accumulation in grapes and bunch rot incidence and severity during maturation in 2014 was compared. Data for 2014 vintage is presented as an example, but the results were similar for all three seasons. Note that in a commercial context H treatment would be harvested at least two weeks before a target harvest date of 19 Brix and 760 mg/kg anthocyanin concentration. UTCC treatments could be harvested even later (since the incidence and severity of *Botrytis* bunch rot never reached 10%), allowing the achievement of 22.7 Brix and 1282 mg/kg anthocyanins at harvest. Such large differences between treatments in fruit composition at harvest are not only statistically significant, but have been found to be relevant in a commercial context by grower testing of UTCC in Tannat vineyards in this region.

## 3.6. CONCLUSIONS

The effects of planting density were relatively minimal except that closer spacing generally invigorated shoot growth leading to larger cane weights and pruning weights/m. This was likely due to the heavy soil in this site and the humid climate that was less limiting than the canopy. The use of UTCC to limit growth in combination with supplemental deficit irrigation to support growth was an effective tool to regulate vine vegetative growth and canopy size in a humid environment. We found that water availability from bloom to 30 days after bloom period was highly correlated to vegetative growth. The impact of UTCC with supplemental irrigation limit excessive water stress as a vine growth regulation tool increased while planting density is reduced. The fact that in our experimental conditions, most of root system were developed above a restrictive clay layer (Bt) located at 40 to 50 cm, the UTCC effects observed in this study are not necessarily transferrable to areas with similar rainfall conditions but deep soils where root systems can grow to avoid competition with the cover crop. Vegetative growth by itself did not correlate strongly to Tannat fruit composition. PW/m below 0.60 Kg reduced vine potential yield. On the other hand, the susceptibility of Tannat to bunch rot, increased strongly when PW/m increases over 0.5 kg/m. So, under Uruguayan conditions an optimal growth for a Tannat vineyard was found to be approximately 0.50 to 0.60 kg PW/m with cane weights of 40-45 grams to balance yield potential, fruit ripening and bunch rot susceptibility. TSS and anthocyanin accumulation in grapes and wines was modestly increased by UTCC treatment due to reduced berry size; however, the opportunity to delay harvest to accomplish full maturation in UTCC plots, due to lower bunch rot incidence, was a desirable outcome in this climate. Future research will be oriented to better understand the long-term effects of UTCC, and to optimize other management practices like nitrogen fertilization and irrigation strategies.

## 3.7. ACKNOWLEDGMENTS

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## 4. UNDER–TRELLIS COVER CROP AND DEFICIT IRRIGATION TO REGULATE WATER AVAILABILITY AND ENHANCE TANNAT WINE SENSORY ATTRIBUTES IN A HUMID CLIMATE[2]

### 4.1. ABSTRACT

The goal of our study was to improve Tannat grapes and wine composition by achieving vine balance in high capacity conditions. Current Uruguayan grape-growing paradigm accepts unpredictable periods of water deficit, or excess in non-irrigated vineyards only applying herbicides to eliminate weed competition (H). We used an under-trellis cover crop (UTCC) to limit vine water availability, reduce vine growth rate and final canopy size. However, to avoid excessive vine water stress due to the UTCC competition, irrigation was used as needed during water deficit periods. The experiment was conducted over three consecutive growing seasons from 2011 to 2013, in two experimental vineyards located in Southern Uruguay (34º S 56º W). Treatments were: UTCC (full cover of the vineyard soil with tall fescue) versus conventional alleyway tall fescue with 1.0 m wide weed-free strip under the trellis. Deficit drip irrigation was provided at mid-day stem water potential (SWP) thresholds of -0.9 MPa early and -1.1 MPa later in season. Shoot growth rate, SWP, berry size and berry composition (Brix, organic acids, total anthocyanins) were monitored over the season as well as final yield, cluster, pruning weights and wine sensory attributes. UTCC regulated vine vegetative growth and final canopy size, reduced bunch-rot incidence as well as increased fruit Brix and anthocyanin concentration in grapes and wines. Wines from UTCC treatments increased fruity aroma, overall aroma intensity levels and had distinctive sensory characteristics that exceeded H wines during overall palatability test (liking test).

**Keywords:** Tannat, vigor, vine balance, wine sensory attributes.

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## 4.2. INTRODUCTION

In areas as Uruguay where the temperature, water availability and soil fertility, induce high growth rates, it is common to observe dense and unbalanced canopies giving unfavorable microclimate for fruit maturation and disease management (Smart and Robinson, 1991). The need to maintain a good vegetative development capable to maximize vine capacity and yield potential, in a traditional non-irrigated vineyard, has led to that the commonly adopted strategy for most Uruguayan growers to use medium to high vigor rootstocks combined with a 1.0 m wide weed-free strip under the trellis.

Although the appropriate balance may vary with variety and desired yield or wine style, regulating vegetative growth is a key vineyard practice. Increasing growth tends to be easier to achieve as adding adequate water, nutrients or soil amendments are practical. Reducing excessive growth, however, is often difficult as removing or limiting resources is more difficult especially in humid climates and deep fertile soils. The current local commercial production practices in the humid climate of Uruguay (1) accepts unpredictable periods of water deficit or excess in non-irrigated vineyards, and (2) reduces competition from cover crops.

The use of under-trellis cover crops (UTCC) has been studied in cool humid climates that often experience abundant water availability and fertile soils. Apparently due to competition for soil moisture and mineral nutrients, UTCC has been reported to reduce vine growth (Hickey et al. 2016, Karl et al. 2016, Giese et al. 2015, Giese et al. 2014, Lopes et al. 2008, Tesic et al. 2007), improving fruit sunlight exposure (Giese et al. 2015, Giese et al. 2014, Hatch et al. 2011). Additionally, increases of total soluble solids and/or berry skin phenols and anthocyanins have been reported (Hickey et al. 2016, Tesic et al. 2007), though it was not clear if those were direct or indirect effects of the reduction in canopy density.

The goal of our study was to evaluate an integrated management of cover cropping with supplemental irrigation to regulate canopy growth in order to optimize vine balance and improve Tannat grape and wine composition and wine sensory characteristics. Tannat is noted as a very tannic and often harsh wine. Our approach was to use under-trellis cover crops (UTCC) to limit vine water availability via competition, thus reducing vine growth rate and limiting final canopy size and density.

To avoid excessive water stress due to the cover crop competition during dry periods, supplemental irrigation was applied to regulate the stress and thus vine growth, function and fruit development. Wines were made to evaluate wine composition and characteristics.

## 4.3. MATERIALS AND METHODS

### 4.3.1. Experimental sites

The experiment was conducted over three consecutive growing seasons from 2011 to 2014 in two experimental vineyards located in Southern Uruguay (34°44' S, 56°13' W). Uruguayan climate can be classified as temperate – humid without a prolonged dry season, Cfa by the Köppen-Geiger system (http//en.climate-data.org/location/3741/). Historical mean total annual rainfall in Southern Uruguay (1972-2015) is 1100 mm/year, with 650 mm occurring the growing season (Table 1). Further weather data details can be accessed at http//www.inia.org.uy/gras/agroclima/cara_agro/index.html. Both sites soils were similar and characteristic of the region. Soils were classified as a silty clay loam Tipic Argiudoll by the USDA soil classification system), with a variable depth of 0.90 to 1.1 m and 0.90 to 1.0 m for Sites 1 and 2 respectively. The total soil available water (field capacity-permanent wilting point) to 1.0 m depth was from 100 to 110 mm for both soils.

**Table 1.** Irrigation by treatment and site and evapotranspiration, precipitation and growing degree-days (>10ºC), from Las Brujas weather station located at 400 m from experimental sites.

| | Phenological stage | Degree-days (>10ºC) | Eto Penman (mm) | Precipitation (mm) | Irrigation (mm) Site 1 H | Site 1 UTCC | Site 2 H | Site 2 UTCC |
|---|---|---|---|---|---|---|---|---|
| Historical | budbreak - bloom | 338 | 208 | 227 | | | | |
| | bloom - veraison | 718 | 359 | 291 | | | | |
| | veraison - harvest | 664 | 257 | 266 | | | | |
| | post-harvest | 421 | 136 | 252 | | | | |
| Season 2011/12 | budbreak - bloom | 328 | 224 | 132 | 0 | 0 | 0 | 0 |
| | bloom - veraison | 722 | 391 | 177 | 16.7 | 103.5 | 14.7 | 98.5 |
| | veraison - harvest | 665 | 271 | 212 | 23.7 | 70.7 | 20.3 | 76.8 |
| | post-harvest | 435 | 157 | 147 | 0 | 0 | 0 | 0 |
| Season 2012/13 | budbreak – bloom | 403 | 211 | 359 | 0 | 0 | 0 | 0 |
| | bloom – veraison | 753 | 381 | 288 | 0 | 42.2 | 0 | 27.1 |
| | veraison - harvest | 641 | 291 | 199 | 0 | 9.7 | 0 | 8.2 |
| | post-harvest | 352 | 139 | 173 | 0 | 0 | 0 | 0 |
| Season 2013/14 | budbreak - bloom | 332 | 214 | 204 | 0 | 51.2 | 0 | 20.1 |
| | bloom - veraison | 804 | 398 | 284 | 7.3 | 60.2 | 0 | 50.2 |
| | veraison - harvest | 692 | 228 | 641 | 0 | 0 | 0 | 0 |
| | post-harvest | 395 | 146 | 194 | 0 | 0 | 0 | 0 |

In both locations Tannat grapevines, grafted on to SO4 rootstock, were trained on vertical shoot positioned training system (VSP) in north-south oriented rows (0.8 m x 2.8 m; vine x row spacing at Site 1 and 1.0 m x 2.6 m; vine x row spacing at Site 2). The Site 1 and 2 vineyards were 7 and 10 years old respectively when cover crop was installed in March 2011(seeding rate, 60 kg/Ha of tall fescue, Festuca arundinacea).

4.3.2. General vine management

Cordon-trained vines were pruned to seven two-bud spurs per meter during dormancy. The height of the cordon was 1.0 m, and the top of the canopy was approximately 2.1 m above the ground. At approximately 30 cm shoot length, all shoots not located on spurs were removed. During the growing season, shoots were positioned by hand vertically above the spurs and topped 30 cm above the top wire. Catch wires were used to keep shoots in position.

To avoid overcropping during ripening, crop level was adjusted by cluster thinning in each experimental plot at veraison (Stage 35 - Eichhorn and Lorentz, 1977). Based on prior research (Coniberti et al. 2011), an optimal crop level was estimated to be about 1 cluster/shoot. With a full canopy this provides a ratio of at least 1.8 m2 leaf area/kg fruit weight needed for maximize sugar and anthocyanin accumulation. To estimate the potential yield in every plot, thinned clusters were counted and weighed. Since no berry weight significant differences were detected from veraison to harvest, it was assumed to develop as the remaining clusters, and the assumed final weights were thus added to the harvested yield. The potential yield provides insight to the crop load during periods before thinning.

Standard disease control fungicide programs were applied for downy mildew, powdery mildew, and *Botrytis* bunch rot. Irrigation water was applied with drip emitters (4 L/minute) located directly under the vines and distributed 0.3 m apart. The irrigation system was designed to allow independently-irrigated single experimental plots.

4.3.3. Treatments

Two treatments (UTCC versus conventional floor management) were evaluated in a complete random block design with five and four replicates for Site 1 and Site 2 respectively.

At both experimental sites replications comprised eight adjacent grapevines, the outer vines serving as guard vines. Buffer rows separated ground cover treatments. The UTCC treatment consisted in the full cover of the vineyard soil with tall fescue (Festuca arundinacea). The UTCC treatment was compared with a conventional herbicide management scheme with the same inter-row groundcover but combined with a 1.0 m wide weed-free strip under the trellis (H). The under-trellis, weed-free strip was maintained with a combination of herbicides.

To avoid the effect of the treatment due to nitrogen (N) competition, in every UTCC plot ammonium nitrate ($NH_4NO_3$) was applied twice at a rate of 20 kg N /ha when shoots reached approximately 30 cm and after fruit set (stage 29 - Eichhorn and Lorentz, 1977). No statistically significant differences among treatments were detected in leaf N%, P%, K%, Ca% and Mg% at bloom or veraison (data not shown). Average leaf N% content ranged from 2.1 to 2.5% at both Sites. No visual nutrient deficiency symptoms were detected.

To monitor water stress in the treatments, midday stem water potential ($\Psi$stem) was periodically measured (~ bi-weekly) between 1400 and 1600 hr using a leaf pressure chamber (Soil Moisture Equipment Corp., Santa Barbara, CA) on two leaves per treatment replication (Chone et al. 2001). Plastic bags covered with aluminum foil were placed on well-exposed mid-shoot leaves one hour before the onset of Ψstem measurements. The time from cutting leaf petioles to insertion and chamber pressurization was less than 30 sec. The chamber was pressurized with nitrogen gas at a rate of 2 to 4 sec/0.1 MPa.

Normally, no irrigation was needed pre-bloom. However, pre-bloom irrigation was needed only during the last season in UTCC treatments, for a 10 days period (from 20 to 10 days before bloom) at an average rate of 5 mm/day to ensure adequate early canopy development. This was done in plots with weak growth as the average pruning weight per meter of trellis < 0.4 Kg/m since without pre-bloom irrigation the vine growth and capacity would have been excessively reduced. In order to avoid severe post-bloom water stress, deficit irrigation (70% ETc) was applied for all treatment replicates once the plants reached -0.9 MPa mid-day stem water potential (Ψstem). To avoid the Ψstem becoming more negative than -1.1 MPa after a prolonged period of deficit irrigation, occasionally the amount of water the vines had consumed the previous week was applied at 100% ETc.

ETo was obtained from the weather station of INIA Las Brujas Experiment Station located < 200 m from the experimental sites (Allen et al., 1998). Mid-day canopy shaded area (light interception) was estimated at solar noon using a Solar panel (Paso panel, http,//cesanluisobispo.ucanr.edu/Viticulture/Paso_Panel/) and each plot's crop coefficient (Kc) was calculated using the formula proposed by Williams and Ayars (2005); Kc = (0.017 * Shaded percentage of field) – 0.008. ETc was calculated by multiplying ETo by Kc and used to estimate weekly vine water consumption.

4.3.4. Vegetative measurements

Shoot growth rates were obtained by repeated measures of shoot length. Two shoots from two representative vines per plot were tagged and measured on a ~ weekly basis starting shortly after bud-break. At veraison, photosynthetically active radiation (PAR) available in the fruit zone was estimated with an average of three readings taken in the canopy fruit zone, with a ceptometer (AccuPAR L80; Decagon Devices, Pullman, WA) inserted parallel to and 15 cm above the cordon. Since the clusters were not flat, the light availability was estimated at three angles: one with the sensors face angled

45° to the east, a second vertically upright, and the third angled 45° to the west. Incident radiation was measured by orienting the ceptometer vertically upright, at the fruit zone height outside the canopy in the alleyway (ambient). PAR measurements were made ±2 hours of solar noon on cloudless days. In the winter, the pruning of every experimental plant were weighed and averaged by plot. The Ravaz index of crop load (RI, fruit yield/pruning weight) and average shoot pruning weight were then calculated. Every cane from tagged plants was individually weighed and average cane pruning weight of those selected plants was calculated as an expression of shoot development.

4.3.5. Berry development, fruit composition and disease development

From veraison to harvest; berry samples of one hundred berries were randomly collected from each treatment replicate on a bi-weekly basis. Berry weight, total soluble solids (TSS), titratable acidity (TA), pH and total anthocyanins were analyzed. TSS was determined using a hand refractometer (Atago, model N1, Tokyo, Japan). Must pH was measured with a pH meter (Horiba, model F13, Kyoto, Japan) and titratable acidity (TA) was determined by titration (NaOH 0.1 N) and expressed as tartaric acid equivalents (w/w). Total anthocyanins analyses were performed according to Glories (1984). From veraison to harvest, the percentage of bunches infected by *Botrytis* bunch rot (incidence) as well as the percentage of each bunch that was infected (severity) was determined by visual inspection using a six-point scale (0, 5, 25, 50, 75 and 100%). *Botrytis* severity (S) was calculated as follows, $S = \Sigma Si/n$; where $Si$ = % severity for each bunch and n = the total number of affected bunches. All this information was used to define harvest date.

4.3.6. Harvest and yield

In both experimental sites, treatments were harvested at the same date. In addition to chemical evaluations, bunch rot incidence and severity were monitored to help define the requirement for early harvest to avoid excess loss due to bunch rot. Total fruit yield and clusters per vine were determined for each experimental plot. Mean cluster weight was calculated. From the harvested fruits, about 10 kg of grapes per plot were crushed and juice TSS, pH and TA were analyzed. In addition, samples of 200 berries per replication were taken, to prepare homogenized fruit extracts. From these extracts, total anthocyanins was analyzed as previously described.

4.3.7. Wine making and analysis

Approximately 10 kg of fruit per treatment replicate were retained for winemaking. After storage for 16hr at 5 ºC, grapes were crushed and destemmed. Must samples were taken, and grapes from adjacent field blocks (B) were combined (B1 + B2 + B3/2 and B3/2 + B4 + B5 in Site 1; and B1 + B2 and B3 + B4 in Site 2) giving two wine replicates for each treatment. Sulfur dioxide was added at a rate of 60 mg/kg and the must was inoculated with Saccharomyces cerevisiae (ALG111, DSM, Delft, The Netherlands) at 25 g/hL. Fermentation temperature was maintained between 26 and 30 ºC. Alcoholic fermentation was completed six to eight days after the beginning of fermentation, but maceration was allowed for 10 days.

After maceration was completed, wines were pressed, and placed into sterile 10-L glass containers where malolactic fermentation was completed. After completing malolactic fermentation, wines were cold-stabilized at 0 ºC for two weeks, SO2 was added and the wines were stored in sterile 5-L glass containers at 11 ºC. For each wine, a 125 mL sample was taken for analysis (alcohol concentration, TA, pH, tartaric and malic levels) by standard methods (OIV, 2009). Total anthocyanins were calculated from the absorbance at 520 nm as described by Glories (1984).

4.3.8. Wine testing

Prior to descriptive analysis wines were compared. At the time, terms were gathered as possible descriptors for flavor profiles. After discussion, seven sensory attributes (two aroma attributes, overall aroma intensity, and four attributes by mouth) were selected to be used by panelists. Finally, a hedonic (like–dislike) assessment was included in the study.

Considering that professionals are trained to differentiate their own preferences from typicality of the wine style under consideration, high liking ratings would correspond with wines perceived to be high in varietal definition. Each component was evaluated on a five-point scale (1 = low, 5 = high). A group of eight enologists was assembled as a descriptive testing panel. Experts were drawn from different local wineries and were selected on the basis of having extensive experience with the product of interest, namely Tannat wines. Prior to participation and in keeping with ethical requirements, each person was provided information about the impending study. The study was conducted in two one-hour sessions, separated by a 20 min break. To compare the effect of treatments, four rounds of two wines from each experimental treatment and Site were tasted per session. Samples in random order in two-digit coded (ISO 3591, 1977) standard tasting glasses (50 mL) and assessed at room temperature.

Statistics. A complete random Block design was used to analyze the significance of the treatments main effects. For canopy characteristics, fruit composition, wine composition and disease, variance analyses were performed with the model: Year (three years), Site (two sites) and Treatment (H and UTCC) with interaction Site*Year, Treatment*Year and Site*Treatment. For each attribute of wine sensory analysis, descriptive analysis data were analyzed by analysis of variance with the model: Wine, Panelist with interaction Wine*Panelist. All the statistics analyses were performed using MSTAT-C computer software (Michigan State University).

## 4.4. RESULTS

### 4.4.1. Weather conditions

Accumulated growing degree days, base 10C (GDD) and precipitation during the three growing seasons is summarized in Table 1. The three growing seasons were characterized by variable rainfall amounts and patterns. Precipitation accumulated during 2011/12 was below average in every phonological period, and irrigation was needed also in H treatment to avoid severe vine water stress. During 2012/13 precipitations occurred early in season was almost double of the historical value; while from veraison to harvest was below average.

Precipitation occurred during 2013/14 was close to average until veraison end extremely rainy from veraison to harvest (Table 1). On the other hand total GDD accumulated during the three growing seasons was similar to the historic average for the region (≅2150 GDD).

4.4.2. Vegetative growth

Under-trellis cover crop (UTCC) significantly reduced vegetative growth (cane pruning weight, pruning weight/m, and shaded area/m) and its associated parameters of canopy density (PAR% in fruit zone) (Table 2). Final PW/m reflected the different shoot growth rate among treatments during the 30 days period after bloom. No substantial differences in shoot growth rate were detected previous to or after this period among treatments (Fig. 1).

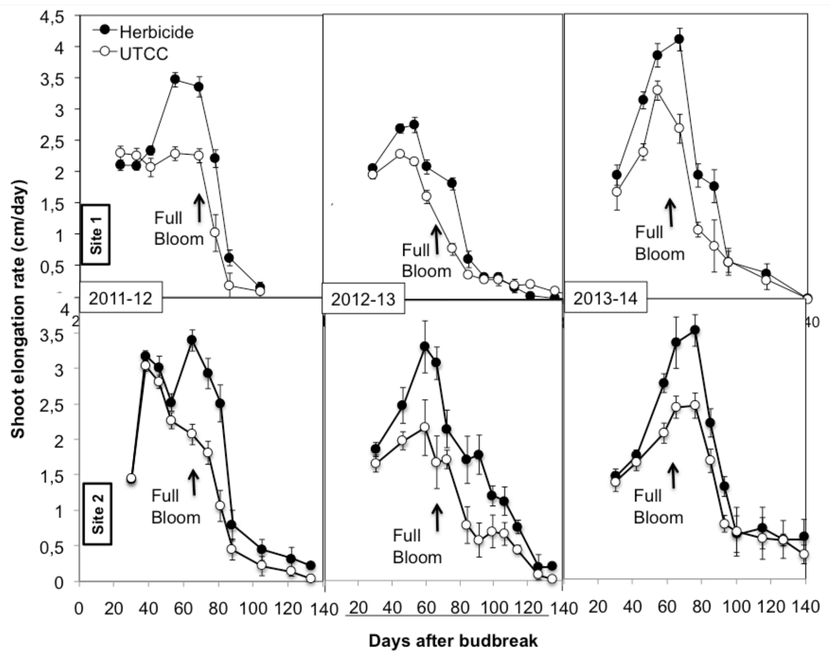


**Figure 1.** Shoot elongation rate of Tannat grapevines as affected by groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in two sites in southern Uruguay.

**Table 2.** Canopy characteristics of Tannat grapevines as affected by groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

| | | Site 1 | | Site 2 | |
|---|---|---|---|---|---|
| | | H | UTCC | H | UTCC |
| Season 2011/12 | Cluster weight (g) | 393 | 359 | 287 a | 263 b |
| | Berry weight (g) | 2.12 a | 1.88 b | 1.90 a | 1.66 b |
| | Vine yield (kg/m) | 3.60 | 3.31 | 3.21 | 3.20 |
| | Potential yield (kg/m) | 5.54 a | 4.66 b | 4.88 a | 4.39 b |
| | Pruning weight (kg/m) | 0.88 a | 0.50 b | 0.79 a | 0.49 b |
| | Pruning weight/cane (g) | 82.9 a | 47.6 b | 54.4 a | 31.7 b |
| | PAR % | 3.93 b | 12.07 a | 2.64 b | 4.69 a |
| | Shaded area m2/m | 0.97 a | 0.70 b | 0.92 a | 0.69 b |
| | Ravaz index | 4.2 b | 6.7 a | 4.6 b | 6.5 a |
| Season 2012/13 | Cluster weight (g) | 430 a | 300 b | 352 a | 305 b |
| | Berry weight (g) | 1.77 | 1.6 | 1.72 a | 1.54 b |
| | Vine yield (kg/m) | 3.94 a | 2.89 b | 4.08 a | 2.79 b |
| | Potential yield (kg/m) | 5.55 a | 4.37 b | 4.18 a | 3.42 b |
| | Pruning weight (kg/m) | 0.92 a | 0.50 b | 0.71 a | 0.36 b |
| | Pruning weight/cane (g) | 65.5 a | 39.3 b | 51.0 a | 31.0 b |
| | PAR % | 3.83 b | 13.9 a | 1.86 b | 8.45 a |
| | Shaded area m2/m | 0.91 a | 0.65 b | 1.03 a | 0.84 b |
| | Ravaz index | 4.58 b | 6.24 a | 5.7 b | 7.8 a |
| Season 2013/14 | Cluster weight (g) | 274 a | 243 b | 268 a | 241 b |
| | Berry weight (g) | 1.97 | 2.07 | 1.77 | 1.66 |
| | Vine yield (kg/m) | 2.69 a | 1.93 b | 2.61 | 2.71 |
| | Potential yield (kg/m) | 3.45 a | 2.39 b | 3.30 a | 2.92 b |
| | Pruning weight (kg/m) | 0.81 a | 0.56 b | 0.65 a | 0.41 b |
| | Pruning weight/cane (g) | 74.7 | 59.6 | 71.2 a | 43.9 b |
| | PAR % | 4.9 b | 10.1 a | 3.1 b | 11.3 a |
| | Shaded area m2/m | 0.88 a | 0.81 a | 0.89 a | 0.67 b |
| | Ravaz index | 3.28 | 3.56 | 4.1 b | 6.6 a |

Values from each experimental site, with different letters in single rows are significantly different at $p \leq 0.05$ by Tukey's test.

Grapevine water status varid with treatment and year during the three growing seasons (Fig. 2). At both sites shoot growth rates post-bloom (bloom +30 days) were correlated to mid-day Ψstem between -0.35 and -0.60 MPa (average of three measurements; R2= 0.69 and 0.88 for Site 1 and Site 2 respectively) in the first year. No significant growth was detected in any period when the Ψstem was lower than -0.9 MPa. Note how the first year was the only season when moderate water stress was detected at the beginning of the season at both sites (Fig. 2). During the last two seasons, vine water status during the 30 days period after bloom was always above -0.5 MPa Ψstem in every experimental plot at both sites, so the correlations observed were not as strong (Fig. 2).

Vine pruning weights in the second and third years maintained the differences observed in the first year even though the water stress was less severe, especially in the third year (Table 2). Vine PW/m over three seasons were consistent and did not show cumulative effects (Table 2) even in the most restrictive treatment, however in order to prevent excessive vegetative growth restriction due to water availability (<-0.6MPa Ψstem) in UTCC treatment, irrigation was applied the last season (Table 1 and Fig. 1). Note how the significant reductions in vine pruning weight observed in UTCC compared to H treatment at both sites (from 31 to 49%) produce a relatively minor reduction in vine light interception (from 8 to 28% of canopy ground shade) (Table 2).

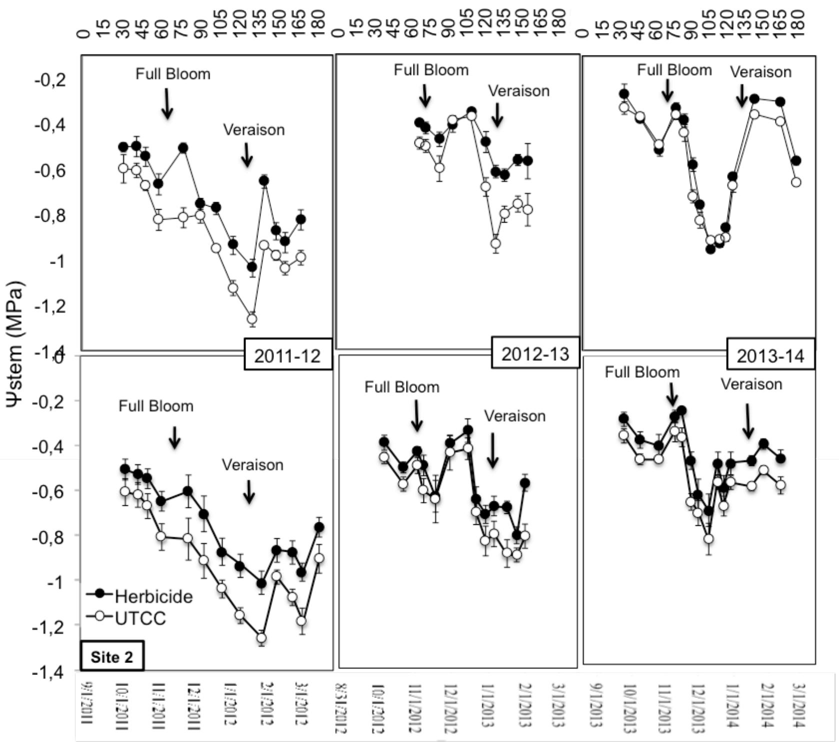


**Figure 2.** Midday Ψstem of Tannat grapevines as affected by groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in two experimental sites in southern Uruguay.

<u>4.4.3. Fruit yield</u>

Final berry weight was consistently reduced by UTCC in both experimental sites only at 2012 harvest (Table 2). Those differences were not detected in Site 1 during 2013, and in any case at harvest 2014. Note how significant Treatment*Year interactions were detected for berry size (Table 3). Differences observed in berry weight where translated to cluster weight at harvest 2012 only in Site 2. In contrast to berry weight, cluster weight was consistently reduced by UTCC the last two seasons. Associated with a significant reduction of cluster weight and/or berry weight, potential yield (harvested + thinned clusters) was consistently reduced by UTCC in this study. Seasonal potential yield reductions were from 16 to 31% in Site 1 and 11 to 19% in Site 2, while the accumulated potential yield losses in UTCC compared to H treatment reached 21% and 13% for Site 1 and Site 2 respectively.

**Table 3.** Analysis of variance for intensity ratings of sensory attributes of wines made from different treatments of groundcover management (H: herbicide and UTCC: under trellis cover crop) in two experimental sites in southern Uruguay.

| | ***F* values** | | | | | |
|---|---|---|---|---|---|---|
| | **Year** | Site | Treatment | Site*Year | Treatment*Year | Site*Treatment |
| **Canopy Characteristics** | | | | | | |
| Cluster weight (g) | 88.55*** | 83.27*** | 9.20** | 20.13*** | 2.37 | 1.93 |
| Berry weight (g) | 27.76*** | 62.42*** | 34.02*** | 9.78*** | 10.18*** | 3.34 |
| Vine yield (kg/m) | 11.59*** | 0.04 | 10.48** | 1.85 | 3.48* | 0.75 |
| Potential yield (kg/m) | 26.45*** | 5.30* | 13.46*** | 3.81* | 0.19 | 1.38 |
| Pruning weight (kg/m) | 0.36 | 8.24** | 77.76*** | 1.82 | 1.09 | 0.13 |
| Pruning weight/cane (g) | 16.90*** | 42.44*** | 126.38*** | 3.31* | 1.32 | 0.32 |
| PAR % | 0.65 | 1.56 | 57.35*** | 1.89 | 0.37 | 0.54 |
| Shaded area m2/m | 2.05 | 0.21 | 68.38*** | 7.31** | 1.01 | 1.08 |
| Ravaz index | 11.16*** | 4.12* | 26.07*** | 1.79 | 1.11 | 1.51 |
| **Fruit composition and disease** | | | | | | |
| Brix | 146.40*** | 152.34*** | 45.90*** | 20.17*** | 2.63 | 0.80 |
| Titratable acidity (g/L) | 38.88*** | 142.46*** | 25.39*** | 42.78*** | 0.01 | 0.04 |
| pH | 36.41*** | 15.82*** | 1.00 | 20.65*** | 0.96 | 0.28 |
| Anthocyanins (mg/L) | 18.90*** | 13.54*** | 6.10* | 0.28 | 3.13 | 0.17 |
| YAN (mg/L) | 4.16* | 6.40* | 35.32*** | sd | 0.22 | 0.21 |
| Bunch rot incidence (%) | 3.12 | 0.96 | 73.71*** | 5.72** | 2.07 | 1.47 |
| Bunch rot severity (%) | 5.68** | 4.72* | 61.91*** | 15.13*** | 2.60 | 0.52 |
| **Wine composition** | | | | | | |
| Ethanol (% v/v) | 48.59*** | 75.01*** | 5.44* | 16.76*** | 1.94 | 2.96 |
| Titratable acidity (g/L) | 8.81** | 6.85* | 8.2E-4 | 0.74 | 3.75 | 0.31 |
| pH | 50.45*** | 0.12 | 0.12 | 1.53 | 2.06 | 0.12 |
| Anthocyanins (mg/L) | 27.95*** | 2.49 | 6.2* | 5.45* | 0.33 | 0.22 |

*, ** and *** Indicate statistical significance at the $p < 00.5$, $<0.01$ and $<0.001$ level of confidence respectively.

4.4.4. Berry composition and bunch rot

With the only exception of Site 2 at 2014 harvest (Table 4), UTCC treatment significantly increased the fruit TSS in both Sites. Total fruit anthocyanins concentration was consistently increased by UTCC, at 2012 harvest, however differences were detected only in one of the two experimental sites the last two seasons. Differences detected in anthocyanin concentration among treatments were not directly associated to berry size. Note how during 2013 harvest, berry size was not significantly affected by treatments in Site 1 but significant differences in anthocyanin concentration were detected, while the opposite occurred in Site 2.

At the same time, even when associated with similar water availability during the entire season, berry weight was not significantly affected by treatments at harvest 2014, UTCC increased anthocyanin concentration in Site 2. As compared with H treatment, UTCC reduced must TA the first two seasons with no effect on must pH (Table 4). Must YAN concentration was consistently reduced by UTCC treatment, even though fertilization was applied in UTCC plots (40 Kg/Ha N each season) to avoid vine nitrogen deficiency and no visual deficiency symptoms were observed.

Bunch rot incidence was strongly reduced by UTCC. Bunch rot incidence in UTCC was from 8 to 21% of it observed in H treatment in Site 2 and 18% to 21% in Site 2 (Table 4). Similarly, with the only exception of 2013 harvest – Site 2, bunch rot severity was consistently reduced by UTCC.

**Table 4.** Grape must and wine composition and *Botrytis* bunch rot incidence and severity as affected by groundcover management (H: herbicide and UTCC: under trellis cover crop), during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay.

| | | Site 1 | | Site 2 | |
|---|---|---|---|---|---|
| | | **H** | **UTCC** | **H** | **UTCC** |
| **Harvest 2012** | **Berry composition** | | | | |
| | Brix | 24.2 b | 25.4 a | 20.7 b | 22.0 a |
| | Titratable acidity (g/L) | 6.16 a | 5.67 b | 3.94 a | 3.37 b |
| | pH | 3.46 | 3.41 | 3.39 | 3.36 |
| | Anthocyanins (mg/L) | 1389 b | 1504 a | 1156 b | 1446 a |
| | YAN (mg/L) | 91.2 a | 70.7 b | | |
| | Bunch rot incidence (%) | 13.2 a | 2.9 b | 34.3 a | 6.0 b |
| | Bunch rot severity (%) | 12.0 a | 3.6 b | 36.4 a | 7.0 b |
| | **Wine composition** | | | | |
| | Ethanol (% v/v) | 14.1 | 14.6 | 11.7 b | 12.6 a |
| | Titratable acidity (g/L) | 6.14 | 6.14 | 4.00 a | 3.45 b |
| | pH | 3.72 | 3.72 | 3.65 | 3.68 |
| | Anthocyanins (mg/L) | 1077 b | 1296 a | 1199 b | 1317 a |
| **Harvest 2013** | **Berry composition** | | | | |
| | Brix | 25.8 b | 27.4 a | 23.9 b | 25.0 a |
| | Titratable acidity (g/L) | 7.61 a | 6.90 b | 5.25 a | 4.89 b |
| | pH | 3.43 | 3.44 | 3.52 | 3.49 |
| | Anthocyanins (mg/L) | 1563 b | 1615 a | 1437 | 1401 |
| | YAN (mg/L) | 101.0 a | 78.7 b | 113.7 a | 92.8 b |
| | Bunch rot incidence (%) | 45.2 a | 3.8 b | 30.5 a | 3.4 b |
| | Bunch rot severity (%) | 32.6 b | 15.6 b | 16.5 | 14.5 |
| | **Wine composition** | | | | |
| | Ethanol (% v/v) | 15.4 | 15.5 | 12.2 b | 13.5 a |
| | Titratable acidity (g/L) | 6.72 | 7.08 | 4.03 | 4.41 |
| | pH | 4.00 | 4.00 | 4.05 | 3.98 |
| | Anthocyanins (mg/L) | 1385 b | 1540 a | 1308 | 1385 |
| **Harvest 2014** | **Berry composition** | | | | |
| | Brix | 22.1 b | 22.7 a | 21.3 | 21.8 |
| | Titratable acidity (g/L) | 8.03 | 7.53 | 6.05 | 5.76 |
| | pH | 3.23 | 3.22 | 3.38 | 3.42 |
| | Anthocyanins (mg/L) | 1284 | 1282 | 1559 b | 1810 a |
| | YAN (mg/L) | 106.4 a | 84.5 b | | |
| | Bunch rot incidence (%) | 55.9 a | 8.1 b | 32.7 a | 6.1 b |
| | Bunch rot severity (%) | 13.9 a | 1.0 b | 26.7 a | 6.9 b |
| | **Wine composition** | | | | |
| | Ethanol (% v/v) | 12.2 | 11.7 | 11.6 b | 12.0 a |
| | Titratable acidity (g/L) | 7.33 | 6.50 | 4.40 | 4.33 |
| | pH | 3.69 | 3.79 | 3.80 | 3.78 |
| | Anthocyanins (mg/L) | 807 b | 860 a | 1057 | 1151 |

Values from each experimental site, with different letters in single rows are significantly different at $p \leq 0.05$ by Tukey's test.

### 4.4.5 Wine composition

Treatment effects on wine ethanol and anthocyanin concentration were not always consistent with that expected from must TSS and grapes total anthocyanins concentration. UTCC consistently increased wine ethanol concentration in Site 2, even when no significant differences in fruit TSS were detected the last season. Fruit TSS was increased every season by UTCC in Site 1; nevertheless wine ethanol concentration was not significantly affected by treatments. In the same way, wine anthocyanin concentration was always increased by UTCC at Site 1, even when no effect was detected on fruit the last season. Similarly, in Site 2 wine anthocyanin concentration was not significant affected by treatments the last two seasons, however fruit anthocyanin concentration was higher in UTCC treatment. Wine and berry anthocyanin concentration were consistently increased by UTCC in both Sites only the first season, probably associated to a consistently reduction in berry size and vine water availability compared to H treatments. With the only exception of wine TA at 2012 harvest - Site 2, no significant effect was detected in wine TA or pH in this study.

#### 4.4.6 Wine sensory attributes

Highly significant differences were found among wines in fruity aroma while no differences were detected for green characters (Table 5). Note that even the excessively vigorous vines from H treatment produced wines with relatively low green characters scores during sensory analysis (Table 6). Descriptive analysis revealed that wines from UTCC treatments increased fruity aroma, overall aroma intensity levels and also had distinctive sensory characteristics that were preferred compared to H wines (Table 6). The sensory panel was able to detect differences for most wines gustatory descriptors (except persistence) (Table 5), however no significant differences were detected among treatments (except wine body; Site 2-2012) (Table 5 and 6). Means of overall palatability ratings showed that with the only exception of Site 2 at harvest 2013 where no significant differences were detected, the experts preferred UTCC treatment wines. The experts also identified an enhanced wine color in UTCC treatment (Table 6).

| Table 5. Analysis of variance for intensity ratings of sensory attributes of wines made from different treatments of groundcover management (H: herbicide and UTCC: under trellis cover crop) in two experimental sites in southern Uruguay. | | | |
|---|---|---|---|
| | *F* values | | |
| | Wine | Panelist | Wine*Panelist |
| **Olfactory descriptors** | | | |
| Green (herbacius) | 2.89* | 13.77*** | 1.04 |
| Fruity | 4.95*** | 4.84*** | 1.40 |
| Overall aroma intensity | 3.11** | 11.04*** | 1.34 |
| | | | |
| **Gustatory descriptors** | | | |
| Acidity | 2.26* | 7.83*** | 0.99 |
| Astringent (Tanins) | 3.02** | 13.22*** | 0.59 |
| Body | 3.55** | 5.72*** | 0.81 |
| Persistence | 1.20 | 9.29*** | 1.11 |
| | | | |
| **Color** | 2.71* | 1.66 | 0.38 |
| | | | |
| **Preference test** | 4.19*** | 2.80* | 1.51 |
| *, ** and *** Indicate statistical significance at the p < 00.5, <0.01 and <0.001 level of confidence respectively. | | | |

| **Table 6.** Effect of groundcover management treatments (H: herbicide and UTCC: under trellis cover crop) on average intensity rating for wine aroma and gustatory attributes. | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | ***Harvest 2012*** | | | | ***Harvest 2013*** | | | |
| | **Site 1** | | **Site 2** | | **Site 1** | | **Site 2** | |
| | H | UTCC | H | UTCC | H | UTCC | H | UTCC |
| **Olfactory descriptors** | | | | | | | | |
| Green (herbacius) | 2.36 | 2.14 | 2.36 | 2.00 | 2.36 | 2.07 | 2.71 | 2.00 |
| Fruity | 3.07* | 3.71 | 3.00** | 3.79 | 3.29* | 3.93 | 2.79** | 3.79 |
| Overall aroma intensity | 3.50* | 4.07 | 3.50* | 4.07 | 3.36* | 4.07 | 3.57 | 4.00 |
| **Gustatory descriptors** | | | | | | | | |
| Acidity | 3.21 | 3.00 | 3.50 | 3.14 | 3.21 | 2.93 | 3.79 | 3.57 |
| Astringent (Tanins) | 3.36 | 2.86 | 3.29 | 3.00 | 3.43 | 2.86 | 2.64 | 2.43 |
| Body | 3.14 | 3.64 | 3.36* | 3.79 | 3.50 | 3.79 | 2.93 | 3.21 |
| Persistence | 3.36 | 3.71 | 3.50* | 3.86 | 3.50 | 3.64 | 3.36 | 3.50 |
| | | | | | | | | |
| **Color** | 4.43** | 5.00 | 4.44* | 4.86 | 4.57 | 4.86 | 3.86 | 4.36 |
| | | | | | | | | |
| **Preference test** | 3.14** | 3.93 | 3.21*** | 4.21 | 3.36* | 4.07 | 3.57 | 3.79 |
| *, ** and *** Indicate statistical significance at the p < 00.5, <0.01 and <0.001 level of confidence respectively. | | | | | | | | |

## 4.5. DISCUSSION

### 4.5.1 Vegetative growth and yield

As previously reported UTCC effectively reduced excessive vegetative growth of grapevines (Hatch et al. 2011, Giese et al. 2014). This provided the ability to reduce excessive growth to the desired benchmark range of 0.30–0.60 kg PW/m (Smart and Robinson, 1991) which translates to 30-60 grams individual cane weights at 10 shoots/m. PW/m values in H treatments were always above this optimal range, which would indicate an excessively shaded canopy with the standard H practice (Table 2 and 3). Vegetative growth in the UTCC treatment in Site 1, would be considered near optimal range for Tannat conducted to VSP under experimental conditions (PW/m ~ 0.5 Kg) since it results in a minor reduction of vine capacity (shaded area under the vine), and reduced canopy density (LI) yet a major reduction in *Botrytis* incidence (Coniberti et al. 2017). On the other hand, the PW/m reduction observed in UTCC plots at Site 2 the last two seasons were near the minimum recommended value, suggesting an excessive depression of vegetative growth and thus vine capacity in this study. Further declines were prevented by the stress-based supplemental irrigation.

Although other factors, such as cover crops sequestering mineral nutrients (especially N), have been reported affecting vegetative development (Hatch et al. 2011, Giese et al. 2014), we did not find any clear effects on tissue nutrient status as mentioned earlier. The first season vegetative growth was well correlated with vine Ψstem ($R2$= 0.69 and 0.63 for site 1 and Site 2 respectively) (data non shown). During the last two seasons, during the critical period for vegetative development (bloom-30 days after bloom), vine Ψstem did not fall below −0.6 MPa in any treatment or Site, however vegetative growth and associated parameters of canopy density (cane pruning weight, PW/m, PAR%) were comparable with first season in both sites. Similar results were previously reported by Giese et al. (2014), when in a 6-year study, consistent effects on vine vegetative growth were achieved after a below average rainfall season (300 mm), even when in following seasons vine Ψstem did not fall below −0.6 MPa on any measured date. Our results suggest that vine capacity may be initially related to water stress, but apparently remains limited not just by current environmental conditions but also by its previous history (Winkler et al. 1974).

UTCC may provide one strategy to control vine size, even when there is no restriction of water availability as seen in 2012/13 and 2013/14 seasons. Morlat and Jacquet (2003) found that the greater the number of Festuca roots in the root zone the fewer the grape roots.

Competition from cover crop roots can decrease vine root growth in shallow soils (Morlat and Jacquet 2003; Centinari et al. 2016) and alter grapevine-rooting patterns (Celette et al., 2008), which may impact the vine’s uptake of water and nutrients as soil water and nutrient availability generally varies with depth. The results, however, may be different if the soil is deep with good water and fertility at depth, allowing vine roots to grow away from the cover crop competition. No blade or petiole N content differences were detected during our study. However, must YAN was every season consistently reduced by UTCC. The higher YAN content observed in H treatments (average values for the region) suggests that vines from UTCC have a lower nitrogen uptake (Table 2).

According with many other studies (Coniberti et al. 2017, Centinari et al. 2016, Hickey et al. 2016, Karl et al. 2016, Hatch et al. 2011), potential yield was significantly reduced by UTCC. In the present study berry size was the first season consistently reduced by UTCC however differences in berry weight, did not translate into significant cluster weight or yield/m differences. The opposite happened at harvest 2014 when berry weight was not affected by treatments, but UTCC significantly reduced cluster weight via reductions in berry number. The analysis requires highlighting some key features of yield formation. Grapevine yield is affected by current season climatic conditions, water and nitrogen status, but yield formation extends over two consecutive years and the main components of grape yield (bunch number per vine and berry number per bunch) are determined primarily by light, temperature, water status and assimilate supply to the buds in the previous bloom (Guilpart et al. 2017; Keller, 2005; Buttrose, 1974). So the occurrence of any environmental constraint during this period may be expected to have an important impact on next season yield. Higher precipitation rates occurred during 2012-13 and 2013-14 seasons (over the average) may explain its no consistent effect of under trellis ground management in berry size and fruit composition.

4.5.2 Grapes and wine composition and wine sensory attributes

No or limited impacts on fruit composition were reported in previous studies of UTCC (Hickey et al. 2016, Karl et al. 2016, Giese et al. 2015). In our study UTCC treatment generally increased grape TSS and reduced TA while pH was not affected (Tables 3 and 4). It should be noted that due to the cluster thinning at veraison, the yields in these trials were low to moderate crop levels (Ravaz Indices <8). UTCC increased also grape and/or wine total anthocyanins concentration at both sites and in every season compared to the herbicide treatments (Tables 3 and 4). Those differences were later appreciated for experts during wine testing.

Nitrogen is required for yeast growth and completion of alcoholic fermentation in grape juice; concentrations of 130-160 mg/L of yeast fermentable nitrogen (primary amino acids and $NH_4+$) are normally required for complete fermentation (Bell and Henschke, 2005, Spayd et al., 1995). A significant relationship between must amino acids and wine aromatic composition has also been described (Hernandez-Orte at al. 2002; Trigo-Córdoba at al. 2015b). In this context, the low must YAN concentration consistently achieved in UTCC (from 70 to 100 mg/L) may be considered as a negative outcome of the treatment (Table 4).

Therefore, new research is beginning to focus on adapting vineyard practice in order to increase the concentration of amino acids in grapes at harvest (Bindon et al. 2008, Teles Oliveira et al. 2012, Trigo-Córdoba et al., 2015a).

A limitation to under-trellis cover crops studies has been the relative lack of a sensory evaluation of the resulting wines. The impact of floor management practices on wine quality in studies conducted in several French regions including both white and red varieties, have been discussed by Guerra and Steenwerth (2012). These trials compared wines from bare plots against those produced from plots with permanent inter-row cover crop. Common to most trials, wine quality decreased as fermentation length increased due to lower juice N, which resulted from permanent cover cropping that reduced the quality of the vines (Guerra and Steenwerth, 2012). Similary David et al. (2001), reported a loss of aromas intensity in response to cover crop treatments (Festuca arundinacea, Festuca rubra, or a F. rubra/Lolium perenne mix) attributed to the longer fermentations brought about by the reduced juice N levels, however, it produced a better mouth balance and a lower acidity than those from bare-soil. On the other hand, Agulhon (1998) reported that in the cases where the red macerations were prolonged for two additional days to compensate for the cover crop, the results were reversed, and the wines from the cover crop plots were liked the best by the panelists.

In our study, no negative effects associated to a reduced must YAN availability of UTCC treatment were detected. Alcoholic fermentation was completed six to eight days after the beginning of independently of treatments. The maceration was allowed for 10 days so the extended maceration may have an impact in our results; however, overall aroma intensity was one of the more significantly discriminated sensory descriptors (Table 5) and in most cases UTCC overall aroma intensity significantly exceeded H treatment wines (Table 6).

The association of excessive vine vigor and canopy density with undesirable green character of red wines has been widely reported (Ristic et al. 2007, Guerra and Steenwerth 2012). However, in our trial, UTCC consistently reduced excessive vegetative growth of H treatment, but no effect of treatments on green aromas of Tannat was detected during wine testing. It is important to consider that even the excessively vigorous vines from H treatment produced wines that obtain relatively low green characters scores during sensory analysis. Higher preference scores were defined for the experts more in terms of overall aroma intensity and fruity aroma abundance than of green aroma, showing the relative importance of those descriptors on Tannat wine acceptance (Table 6).

4.5.3 Bunch rot incidence and severity

Bunch rot development was strongly reduced by UTCC (Table 3 and 4). It has been widely reported that cover crops decreased *Botrytis* incidence by opening up the vine canopy by decreasing the leaf layer number and percentages of internal clusters and leaves, and increasing the percentage of gaps (Morlat and Jacquet, 2003; Tesic et al., 2007) and cluster compactness (Hed et al., 2009). *Botrytis cinerea* damage can be reduced as a result of an increased light exposure (PAR %) of the fruit zone (Reynolds et al. 1996), berry weight (reducing cluster compactness), cluster weight (Hed et al. 2009) and nitrogen grape content (YAN) (Van Zyl and Van Huyssteen 1980, Mundy and Beresford, 2007). When evaluating vine vegetative balance in a humid climate, it is relevant to consider that in many seasons *Botrytis* bunch rot may be the main factor defining harvest time. In our study, UTCC led to a reduction of potential yield compared to excessively vigorous H vines (from 11 to 31%), with a relatively consistent improvement of fruit or wine composition (TSS increases from 0.5 to 1.5 Brix, fruit anthocyanin from 0 to 25%, wine anthocyanin concentration from 6 to 20%) and wine sensory attributes when fruit is picked at the same time. However, it is important to remark that in a commercial vineyard, due to the high bunch rot incidence (20% bunch rot incidence is commonly used as maximum threshold), fruit from H treatments in our study (vigorous vines) would have been harvested in every season prior to full maturation. Consequently in commercial context larger improvements of wine composition and sensory attributes would be expected in UTCC treatment.

## 4.6. CONCLUSION

UTCC in combination with supplemental irrigation was an effective tool to regulate vine vegetative growth and finally canopy size in a humid environment. The grapevine-UTCC water competition during the period of maximum growth rate (bloom ± 20 days period) appeared to be the main factor affecting vine growth in the first year. The continued reduction in vine pruning weight in subsequent years suggest that vine capacity may be initially related to water stress, but remains limited not just by current environmental conditions but also by its previous condition. Consequently UTCC may provide a strategy to consistently control excessive vegetative growth of grapevine even in areas where in some seasons no restriction of water availability during critical periods is expected. UTCC significantly reduce accumulated potential yield (from 13 to 20%) and must YAN, but significantly increased TSS in grapes and anthocyanin accumulation in grapes and wine as well as enhanced wine sensory attributes. Fruity aroma, and overall aroma intensity, was the more significant affected sensory descriptors. Higher liking scores of UTCC treatment were defined for the experts more in terms of fruity aroma abundance than green aroma absence. Bunch rot incidence and severity were strongly reduced by UTCC. Even the enhancement achieved by UTCC treatment in grapes and wine composition may be relevant, the opportunity to delay harvest to accomplish full maturation, due to lower bunch rot incidence in UTCC treatments, is considered the most significant outcome for humid climate viticulture. Even UTCC has a detrimental effect on yield, it may offer an economic and environmentally sustainable alternative to consistently produce high quality wines. Future research will be oriented to better understand the long-term effects of UTCC, and to optimize other management practices like nitrogen fertilization and irrigation strategies to avoid potential yield losses.

## 4.7. ACKNOWLEDGMENTS

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# 5. COMPLETE VINEYARD FLOOR COVER CROP TO REDUCE GRAPEVINE SUSCEPTIBILITY TO BUNCH ROT[3]

## 5.1. ABSTRACT

Excessive vine growth not only negatively impacts fruit composition but also fosters bunch rot incidence. The goal of our study was to improve *Vitis vinifera* (Tannat) grape and wine composition by achieving adequate vine vegetative growth in a humid climate. We tested under-trellis cover crops (UTCC) consisting of full cover of the vineyard soil with red fescue (Festuca rubra) versus conventional alleyway red fescue with 1.0m wide weed-free strips under the trellis (H). As excessive competition with grapevines remains the main reason for UTCC rejection, this strategy was tested in combination with two irrigation schedules—irrigation to avoid water restriction at bloom (Ir) vs. no early irrigation—and two nitrogen inputs (0 vs. 100 Kg N ha-1) over three growing seasons in southern Uruguay. Treatments were arranged in a split-split-plot randomized block design with cover crop schemes as main plots, water availability as subplots and nitrogen inputs as sub-subplots. Shoot growth rate, mid-day stem water potential (Ψstem), berry size and berry composition were monitored over the season, as well as final yield, cluster and pruning weights. UTCC significantly reduced vine vegetative growth, while no significant differences were detected between H and UTCC when irrigation took place early in the season. Even nitrogen input showed positive effects on grapevine vegetative growth in some cases, water availability at bloom was the key driver of vegetative growth. UTCC treatments increased grape soluble solids (TSS) in the last two out of three seasons and consistently increased anthocyanin concentration in grapes. Independent of vegetative growth, strong differences in bunch rot incidence were detected between H and UTCC treatments. Seasonal variations in water status and/or free amino nitrogen content of grapes may have a relevant impact on disease susceptibility at harvest.

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**Keywords:** vegetative growth, water potential, nitrogen, bunch rot, grape composition, under-trellis cover crop

## 5.2. INTRODUCTION

*Botrytis* bunch rot occurrence is one of the most important limitations for the wine industry in humid environments. Under these conditions, decrease of fruit bunch rot susceptibility is the key to consistently achieve the desired wine quality. *Botrytis* bunch rot in grapes is present in all vineyards to some extent, but only results in a damaging bunch rot epidemic when physiological triggers cause berry tissues to become susceptible (Mundy and Beresford, 2007). Berry sugar concentration, senescing tissues, cluster architecture, physical damage and microclimatic conditions are known to be key *Botrytis* bunch rot infection triggers (Hill et al., 1981; Valdes-Gomez et al., 2008). A positive correlation between grapevine growth and susceptibility to fungal pathogens has been found (Coniberti et al., 2018; Guilpart et al., 2017; Valdes-Gomez et al., 2008). It has been reported that cover crops decreased *Botrytis cinerea* incidence by opening up the vine canopy through decreasing the leaf layer number and the percentages of internal clusters and leaves (Coniberti et al., 2018; Guilpart et al., 2017; Morlat and Jacquet, 2003; Tesic et al., 2007; Valdez-Gomez et al., 2008), and reducing cluster weight and compactness (Hed et al., 2009; Valdes-Gomez et al., 2008). A positive correlation was also found between canopy aeration (often enhanced under service crop treatments) and berry skin strength, a source of grape resistance to disease infection (Jacometti et al., 2010). The number and thickness of epidermal and hypodermal cell layers and the rarity of pores have been positively correlated with resistance to *B. cinerea* (Miklota-Gabler et al., 2003). Cover crops may also improve soil biological activity, leading to a faster decomposition of vine residues, which are habitats for *B. cinerea* primary inoculum (Garcia et al., 2018).

In the literature, cover cropping has been extensively assessed in a variety of soil and climate conditions across the world (Garcia et al., 2018). It has been proved that cover cropping may provide solutions to a large number of issues in viticulture (Garcia et al., 2018; Guerra and Steenwerth, 2012). Several strategies of cover cropping are possible: temporary or permanent, total or partial, sown or spontaneous. The main considerations for the choice of cover crops in vineyards may include their degree of competition for water and nutrients, their growth cycle (perennial vs. annual), their adaptation and persistence in the wine environment, and their propensity to house organisms that can positively or negatively affect the vine (Giese et al., 2015; Guerra and Steenwerth, 2012). Structure of cropping systems (e.g. spatial arrangement, rotations) may also lead to different sets of potential services achieved by agroecosystems (Garcia et al., 2018). The number of targeted purposes of using cover crops will determine the level of complexity of the cropping system and its management. The standard practice in Uruguay is to maintain vegetation in the alleys between rows and spray herbicide or cultivate under the trellis to avoid excessive competition for water and nutrients in traditional non-irrigated vineyards (Coniberti et al., 2018). Different cover crops have been tested in the alleys between rows with relative success to regulate excessive vegetative growth of the vine (Guerra and Steenwerth, 2012). However, in areas such as Uruguay, with fertile soils and a high precipitation rate, excessive vine vegetative growth often results anyway (Coniberti et al., 2018; Vanden Heuvel et al., 2017).

Under these conditions, grapevine producers invest considerable effort and expense in vine canopy management measures such as shoot thinning and leaf removal, but these practices do not affect other factors which are also extremely relevant for disease development, such as cluster compactness (Hed et al., 2009). Proactive measures to avoid excessive vine growth, such as more extensive use of vineyard floor cover crops, has received increasing attention over the past few years (Giese et al., 2014; Hatch et al., 2011; Karl et al., 2016). The use of under-trellis cover crops (UTCC) has been studied in vineyards from regions such as the Finger Lakes and North Carolina, USA, and New Zealand that often experience excessive water availability and/or fertile soils (Vanden Heuvel at al., 2017). UTCC cover crops have been reported as a potential tool to reduce excessive vegetative growth of the grapevine (Coniberti et al., 2018; Giese et al., 2014; Karl et al., 2016). In this case, the grapevine should experience moderate water stress after the flowering stage to limit excessive grapevine vegetative development (Coniberti et al., 2018; Pellegrino et al., 2006). However, the introduction of UTCC in vineyards may also induce excessive competition with the grapevine for water and nutrients during dry seasons (Coniberti et al., 2018), resulting in a significant reduction of fruit yield with no major implications for fruit composition (Giese et al., 2015; Hickey et al., 2016; Karl et al., 2016). With adequate mineral nutrition, vegetative growth is determined in the current year by growing conditions during the most intense vegetative growth period of the grapevine (generally during the period from flowering to fruit set) (Huglin and Schneider, 1998). At the same time, grapevine yield formation extends over two consecutive years and the main components of grape yield (bunch number per vine and berry number per bunch) are determined primarily by light, temperature and assimilated supply to the buds in the previous post-bloom period, but are also affected by water and nitrogen status (Buttrose, 1974; Guilpart et al., 2014; Keller, 2005; Vasconcelos et al., 2009). Therefore, over-intense water stress may jeopardize grape yield at year n but also at year n+1 (Guilpart et al., 2014).

Managing the balance between grape yield and berry quality is of particular importance in wine production. Both excessive water availability and severe water stress can alter berry development and the resulting wine quality (Garcia et al., 2018). For that reason, in variable precipitation regimen conditions such as in Uruguay, flexibility and adaptive management may be recognized as relevant to reach an adequate balance between cover crop ecosystem services and disservices. Previous results showed that rigid strategies that do not allow for annual adjustments increased the risk of failure (Coniberti et al., 2018). Technical operations on a seasonal time scale depending on the climate and state of the system during the crop(s) cycle(s)—e.g. irrigation or fertilization—may also contribute to driving ecosystem services and disservices (Garcia et al., 2018).

The main objective of this study was to investigate the potential of using under-vine cover crops as an alternative to herbicides in humid climate vineyards. Our approach was to use UTCC to limit vine water availability and reduce excessive vine growth and bunch rot incidence. However, to avoid excessive vine vegetative growth and yield restriction due to water stress and nutrient deficiency, UTCC treatments were tested in combination with supplemental irrigation treatments and nitrogen inputs. As excessive competition with grapevines for soil resources (water and nitrogen) remains the main reason for cover crop rejection (Garcia et al., 2018), the combination of treatments applied in this study may provide a framework to better define tactical management options to achieve a compromise between UTCC ecosystem services and farmers' economic return. The relationship between grapevine vegetative growth and bunch rot susceptibility is at the centre of our analysis. The field experiment, encompassing three growing seasons (2012–2015), was located in southern Uruguay.

## 5.3. MATERIALS AND METHODS

### 5.3.1. Experimental site

The experiment was conducted over three consecutive growing seasons from 2012/13 to 2014/15 in southern Uruguay (34°44' S, 56°13' W). The Uruguayan climate can be classified as temperate— humid without a prolonged dry season according to the Köppen-Geiger classification (http://en.climate-data.org/location/3741/). Historical mean total annual rainfall in southern Uruguay (1972–2015) is 1100 mm/year, with 650 mm occurring during the growing season (Table 1). Further weather data details can be accessed at http://www.inia.org.uy/gras/agroclima/cara_agro/index.html. The soil has been classified as Typic Argiudoll according to the USDA soil classification system (Soil Survey Staff, 1999), with a variable depth of 0.90 to 1.1 m and silty clay texture (clay 423 g kg-1, silt 444 g kg-1, sand 133 g kg-1), organic matter 24.6 g kg-1, pH (H2O) 6.25, potassium (K) 0.94 cmolc kg-1, and cation exchange capacity (CEC) 28.7 cmolc kg-1. A restrictive clay layer (Bt) is located at 40 to 50 cm, so most of the root system is developed above. The total soil available water (field capacity-permanent wilting point) to 1.0 m depth was 122.4 mm. Total soil available water was estimated from samples with undisturbed structure, collected in volumetric rings (cores of 68.7 cm3) from six trenches (on each block) at depths of 20, 40, 60, and 80 cm.

**Table 1.** Irrigation by treatment and Eto, precipitation and growing degree-days, from Las Brujas weather station located at 5 km from the experimental site.

| | Phonological stage | Degree-days (>10°C) | Eto Penman (mm) | Precipitation (mm) | Irrigation (mm) | | |
|---|---|---|---|---|---|---|---|
| | | | | | Herbicide | UTCC | UTCC + early Irr. |
| **Historical** | budbreak - bloom | 338 | 208 | 227 | | | |
| | bloom - veraison | 718 | 359 | 291 | | | |
| | veraison - harvest | 664 | 257 | 266 | | | |
| | post-harvest | 421 | 136 | 252 | | | |
| **2012/13** | budbreak - bloom | 403 | 211 | 359 | 0 | 0 | 0 |
| | bloom - veraison | 753 | 381 | 288 | 0 | 0 | 31 |
| | veraison - harvest | 641 | 291 | 199 | 0 | 0 | 0 |
| | post-harvest | 352 | 139 | 173 | 0 | 0 | 0 |
| **2013/14** | budbreak - bloom | 332 | 214 | 204 | 0 | 0 | 54 |
| | bloom - veraison | 804 | 398 | 284 | 0 | 34 | 34 |
| | veraison - harvest | 692 | 228 | 641 | 0 | 0 | 0 |
| | post-harvest | 395 | 146 | 194 | 0 | 0 | 0 |
| **2014/15** | budbreak - bloom | 411 | 232 | 334 | 0 | 0 | 0 |
| | bloom - veraison | 732 | 357 | 222 | 0 | 0 | 22 |
| | veraison - harvest | 659 | 275 | 145 | 0 | 0 | 0 |
| | post-harvest | 467 | 169 | 39 | 0 | 0 | 0 |

[a] UTCC: Under trellis cover crop; Ir: Irrigation; Historical: Average from 1971.

## 5.3.2. Experimental vineyard and management

The experiment was conducted on *Vitis vinifera* (Tannat) grapevines grafted on to 3309 rootstocks. Vines were trained to a vertical shoot positioning system (VSP) in north-south oriented rows (1.2 x 2.8 m, vine x row spacing). Cordon-trained vines were pruned to six two-bud spurs per metre of row during dormancy. The height of the cordon was 1.0 m, and the top of the canopy was approximately 2.1 m above the ground. During the growing season, shoots were positioned by hand vertically above the spurs and topped 30 cm above the top wire. Catch wires were used to keep shoots in position. A standard disease control programme was applied and included combinations of Phthalimides, Iprodione, Cyprodinil + Fludioxonil and specific downy mildew fungicides, resulting in an average of ten treatments in each growing season. Specifically for *Botrytis* bunch rot, three spray applications were applied, two applications from stages 21–25 and an additional application at stage 33 (Eichhorn and Lorenz, 1977). Irrigation water was applied with drip emitters (4 L minute-1) located directly under the vines and distributed 0.3 m apart. The irrigation system was designed to allow independently irrigated single experimental plots. As (1) sugar concentration is known to be a key *Botrytis* bunch rot infection trigger (Hill et al.,

1981) and (2) treatments were expected to affect grapevines' yield to pruning weight ratio, to avoid an overcropping affecting on maturation, cluster thinning was done in five vines from every experimental plot. Crop level was adjusted by cluster thinning at veraison (one cluster per shoot at stage 35; Eichhorn and Lorentz, 1977). Based on prior research (Coniberti et al., 2011), an optimal crop level was estimated to be about one cluster per shoot. With a full canopy, this requires a yield to pruning weight ratio of 5 to 7 (Ravaz index) to maximize sugar and anthocyanin accumulation.

### 5.3.3. Treatments

The vineyard was ten years old at the beginning of the experiment when the UTCC was established in March 2012 (seeding rate: 20 kg ha-1 of red fescue, Festuca rubra). The UTCC treatment consisted of full coverage of the vineyard soil with red fescue (Festuca rubra). In H treatment, a conventional management scheme was used with the same inter-row groundcover except with a 1.0 m wide weed-free strip under the trellis (Coniberti et al., 2018). The under-trellis weed-free strip was maintained with a combination of herbicides.

#### 5.3.3.1. Experimental design

Treatments were arranged in a split-split-plot, randomized block design with six replicates. Main plots, comprising two adjacent rows, compared UTCC with conventional under-trellis herbicide floor management (H); subplots (individual rows) compared the effects of two water regimens at full bloom—no early irrigation (UTCC) vs. irrigation at bloom (UTCC+Ir). In UTCC treatment, supplementary irrigation (70% ETc) was applied for all treatment replicates once the plants reached -0.9 MPa mid-day stem water potential (Ψstem) in order to avoid severe water stress. In UTCC+Ir, early irrigation was also applied from bud-break to ten days after full bloom in order to avoid significant differences in water availability compared to H treatments. Additionally, sub-subplots compared nitrogen input (NI) (0 vs. 100 UN ha-1). The six replicate sub-subplots each consisted of twelve adjacent vines, but only the central ten were evaluated. Buffer rows separated ground cover treatments (H from UTCC) receiving the same nitrogen input treatment as the evaluated plots alongside. Ten adjacent grapevines separated nitrogen input treatments on each row.

5.3.3.2. Water and nitrogen applications

ETo was obtained from the weather station of INIA Las Brujas experiment station located < 5 km from the experimental site. The mid-day canopy shaded area (light interception) was estimated at solar noon using a solar panel (Paso panel, http://cesanluisobispo.ucanr.edu/Viticulture/Paso_Panel/) and each plot's crop coefficient (Kc) was calculated using the formula proposed by Williams and Ayars (2005): Kc = (0.017 * Shaded percentage of field) – 0.008. ETc was calculated by multiplying ETo by Kc and used to estimate weekly vine water consumption. Supplementary irrigation was applied as needed regardless of treatment or plot. Since values below -0.7 MPa Ψstem were not detected during the experiment, no irrigation was applied in H treatments (Table 1).

Nitrogen inputs were applied twice at a rate of 25 kg N ha-1 when shoots reached approximately 40 cm and after fruit set (stage 29; Eichhorn and Lorentz, 1977), with one application immediately after harvest as a rate of 50 kg N ha-1.

5.3.4. Vegetative growth measurements

Shoot growth rates were obtained by repeated measures of shoot length. Two shoots from two representative vines per plot were tagged and measured on a roughly weekly basis starting shortly after bud-break. At veraison, photosynthetic active radiation (PAR) received in the fruit zone was estimated by an average of three readings taken in the canopy fruit zone (±2 hours from solar noon) with the ceptometer (AccuPAR L80; Decagon Devices, Pullman, WA). Pruning weight (PW) was determined in the winter and averaged by plot. Ravaz index (RI = fruit yield/pruning weight) as an estimate of vine capacity per kg of crop and pruning weight per metre (PW/m) as an estimate of canopy density were later calculated.

5.3.5. Plant nutrient status

Plant tissue samples were collected every season to evaluate the effects of the treatments on plant nutrient status. Twenty leaves opposite to the basal inflorescence at bloom (90 to 100% of cap fall) or opposite a fruit cluster at veraison (90 to 100% of coloured berries) were collected from every experimental plot. Leaf blades and petioles were separated, washed with tap water and rinsed with distilled water. Tissue samples were oven-dried (70°C) and analysed for essential mineral elements (N, P, K, Ca and Mg) at the INIA Analytical Services Laboratory (INIA, Uruguay).

5.3.6. Water status

Midday stem water potential (Ψstem) was periodically measured from approximately 40 days after bud-break until harvest (~ bi-weekly) between 1400 and 1600 hr using a leaf pressure chamber (Soil Moisture Equipment Corp., Santa Barbara, CA) on two leaves per treatment replication (Chone et al., 2001). Plastic bags covered with aluminium foil were placed on well-exposed mid-shoot leaves one hour before the onset of Ψstem measurements. The time from cutting leaf petioles to insertion and chamber pressurization was less than 30 seconds. The chamber was pressurized with nitrogen gas at a rate of 2 to 4 sec/0.1 MPa (Allen et al., 1998).

5.3.7. Berry development and fruit composition

From fruit set to harvest, berry samples were randomly collected from each treatment replicate. Eighty berries from fruit set to veraison and one hundred berries from veraison to harvest were collected on a bi-weekly basis. In pre-veraison samples, only berry weight was analysed. In post-veraison samples, berry weight, total soluble solids (TSS), titratable acidity (TA), pH and total anthocyanin were analysed. TSS was determined using a hand refractometer (Atago, model N1, Tokyo, Japan). Must pH was measured with a pH meter (Horiba, model F13, Kyoto, Japan) and titratable acidity (TA) was determined by titration (NaOH 0.1 N) and expressed as tartaric acid equivalents (w/w). Total anthocyanin analyses were performed according to Glories (1984).

5.3.8. Harvest evaluation

In addition to chemical evaluations, once fruit reached 20° Brix to harvest, bunch rot occurrence was monitored to help define the requirement for early harvest to avoid excess loss due to bunch rot. All treatments were harvested on the same date. The percentage of bunches infected by *Botrytis* bunch rot (incidence) as well as the percentage of each bunch that was infected (severity) was determined by visual inspection using a seven-point scale (0, 1, 5, 25, 50, 75 and 100%). *Botrytis* severity (S) was calculated as follows: $S = \Sigma Si/n$; where Si = % severity for the ith bunch and n = the total number of affected bunches. Total fruit yield and clusters per vine were determined for each experimental plot. Mean cluster weight was calculated. From the harvested fruits, about 10 kg of grapes per plot were crushed and juice TSS, pH, TA and free amino nitrogen (YAN) were analysed. In addition, samples of 200 berries per replication were taken to prepare homogenized fruit extracts. From these extracts, total anthocyanin was analysed as previously described.

5.3.9. Weather conditions

Precipitation occurring during the three growing seasons was variable in amounts and patterns (Table 1). Compared to the historical value, precipitation that occurred from bud-burst to harvest 2012/13 was close to average (≅800 mm), but significantly higher from bud-burst to bloom (≅160%) and lower from veraison to harvest (≅75%). In season 2013–14, accumulated precipitation from bud-burst to veraison was near average but markedly higher from veraison to harvest (≅240%). In season 2014–15, precipitation was above average until bloom (≅150%) but it was the driest season from veraison to harvest (≅50% of historical value). Accumulated growing degree-days (base 10°C GDD) during the three growing seasons was similar to the historic average for the region (≅2200 GDD) (Table 1).

5.3.10. Statistics

The effects of under-trellis ground cover (GC), water availability (Ir), nitrogen inputs (NI) and their interactions were assessed using linear mixed effects models following a split-split-plot design with a repeated measure. Were included: Year, GC, Ir, NI and their interactions as fixed effects. As random effects, were included Block, GC (main plot), Ir (Subplot) and individual GC*Ir*NI plots (included to account for the repeated measures). The appropriateness of the random and correlation structures was analyzed by comparing nested models with and without the structures with the likelihood ratio test using the restricted maximum likelihood estimation procedure. Since significant year*treatments interactions were detected, treatments effects were also analyzed by season, including the same fixed effects used for repeated measure analysis, and Block, GC and Ir as random effects. A Tukey HSD test (5% significance level) was used to compare treatment means. Statistical analyses were performed using InfoStat computer software (Di Rienzo, et al. 2017).

## 5.4. RESULTS

### 5.4.1. Vine water status

During the study, precipitation at the beginning of the season was average or above average (> 350 mm). However, in order to avoid excessive vegetative growth restriction due to water availability (< -0.6MPa Ψstem) in UTCC+Ir treatment, irrigation was needed every season (Table 1 and Figure 1). This shows the potential scope of UTCC as a tool to regulate water availability under the experimental conditions. On the other hand, in the 2012/13 and 2014/15 seasons, precipitation from veraison to harvest was below average (50% and 75% of the historical value), but even in these conditions, H treatment never reached values more negative than -0.7Mpa MDSWP (Figure 1), and consequently irrigation was not needed. Irrigation aimed to avoid severe vine water stress (more negative than -0.9Mpa MDSWP; Coniberti et al., 2018) was applied only one time during pre-veraison 2013/14 in both UTCCs (Table 1 and Figure 1).

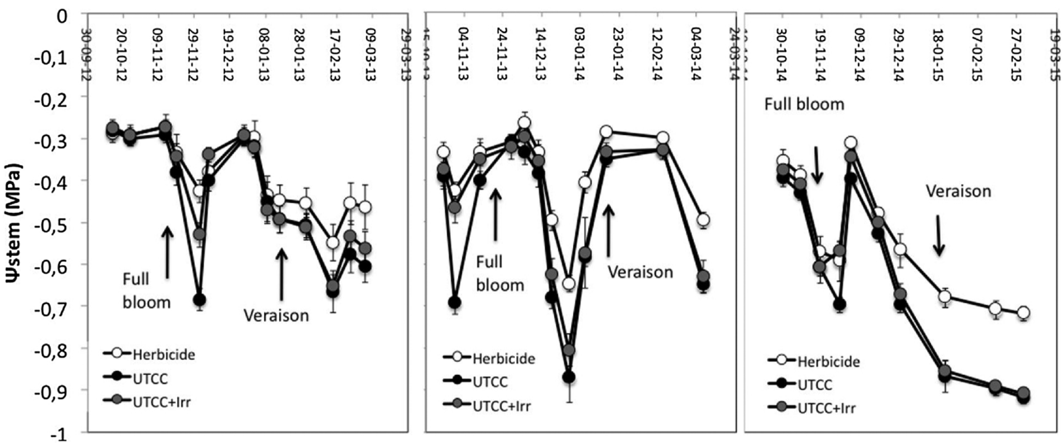


**Fig 1.** Midday stem water potential of Tannat grapevines as affected by groundcover management and irrigation treatments during 2012/13, 2013/14 and 2014/15 seasons in southern Uruguay.

### 5.4.2. Vegetative growth

After three seasons, under-trellis ground cover (GC), irrigation (I) and nitrogen inputs (NI) all significantly affected vegetative growth (PW/m) (Table 2). Since no significant differences in shoot elongation rate were detected among nitrogen input treatments during this study (with the exception of H treatment in spring 2013), for clarity only the average values of groundcover/irrigation treatments during the 2012/13, 2013/14 and 2014/15 seasons are presented in Figure 2. Significant differences in shoot growth rate among treatments were detected only from bloom to the next three-week period (Figure 2). After three years, the final PW/m in the UTCC treatment was significantly lower than in H; however, those differences were not detected before the second season (Table 3). Those PW/m differences were never detected between H and UTCC+Ir treatment. UTCC exhibited more pronounced effects on vegetative growth than irrigation or N inputs (Table 2). Additionally, in H or UTCC+Ir treatments, no differences in vegetative growth were detected between full crop vines versus those whose yield was adjusted by cluster thinning at veraison; however, in UTCC treatment, vegetative growth was reduced by yield.

**Table 2.** Analysis of variance for canopy characteristics, fruit composition and Bunch rot incidence and severity at harvest of Tannat grapevines as affected by groundcover management (GC), irrigation (Ir), and nitrogen inputs (NI).

| | ***F* values** | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Year (Y) | GC | Ir | NI | Y*GC | Y*Ir | Y*NI | GC*NI | Ir*NI | Y*GC*NI | Y*Ir*NI |
| **Cluster thinned vines** | | | | | | | | | | | |
| Berry wt. (g) | 11.40*** | 10.19* | 2.57 | 0.77 | 0.31 | 0.40 | 0.06 | 0.05 | 0.76 | 0.07 | 0.22 |
| Cluster w. (kg) | 43.53*** | 1.89 | 4.60 | 0.69 | 0.56 | 0.99 | 0.59 | 0.31 | 0.51 | 0.25 | 0.11 |
| Yield/m (kg) | 12.09** | 0.76 | 0.40 | 1.68 | 0.28 | 0.63 | 0.08 | 0.48 | 0.01 | 0.11 | 0.01 |
| Pw/m (kg) | 42.97*** | 12.38* | 11.56* | 3.64 | 1.82 | 5.87** | 2.13 | 0.03 | 0.01 | 0.20 | 0.60 |
| PAR (%) | 5.20* | 1.85 | 0.63 | 1.44 | 0.27 | 0.20 | 0.09 | 0.13 | 0.11 | 0.31 | 0.11 |
| Ravaz Index | 37.99*** | 1.63 | 3.89 | 0.27 | 0.21 | 2.98 | 0.48 | 0.18 | 0.09 | 0.11 | 0.53 |
| Brix | 10.99*** | 9.31* | 0.14 | 7.10* | 7.95** | 0.10 | 2.22 | 1.67 | 0.95 | 2.68 | 0.28 |
| pH | 234.82*** | 0.34 | 0.50 | 0.47 | 10.83*** | 0.58 | 0.19 | 7.27* | 0.01 | 6.42** | 0.16 |
| Total Ant (mg/g) | 27.06*** | 17.91** | 0.01 | 2.33 | 1.01 | 1.88 | 1.78 | 4.89* | 0.13 | 2.11 | 2.61 |
| YAN (mg/L) | 119.51*** | 23.15** | 0.30 | 0.91 | 6.22** | 4.69* | 12.95*** | 0.72 | 0.12 | 0.08 | 0.70 |
| **Full crop vines** | | | | | | | | | | | |
| Yield/m (kg) | 178.15*** | 14.88 | 35.26* | 4.78 | 11.37*** | 2.19 | 1.09 | 2.46 | 0.02 | 0.43 | 0.21 |
| Pw/m (kg) | 25.05*** | 4.41 | 13.41 | 1.80 | 2.33 | 7.88** | 0.06 | 0.64 | 0.19 | 0.69 | 1.16 |
| Ravaz Index | 40.18*** | 1.07 | 2.00 | 0.27 | 0.46 | 3.213 | 0.23 | 0.01 | 0.50 | 0.49 | 1.85 |
| Brix | 45.31*** | 1.66 | 2.70 | 0.27 | 5.12* | 1.53 | 0.79 | 0.30 | 0.75 | 2.07 | 0.31 |
| Total Ant (mg/g) | 8.58** | 1.82 | 0.05 | 1.8E-05 | 1.41 | 0.08 | 0.08 | 0.13 | 2.0E-03 | 0.03 | 0.06 |
| **Bunch rot** | | | | | | | | | | | |
| Incidence (%) | 33.08*** | 67.55*** | 0.38 | 6.54* | 26.05*** | 0.49 | 5.32** | 5.34* | 0.22 | 2.25 | 0.08 |
| Severity (%) | 43.16*** | 44.38** | 0.23 | 1.41 | 7.65** | 0.14 | 3.31 | 0.10 | 0.01 | 0.08 | 1.6E-03 |

*, ** and *** Indicate statistical significance at the p < 00.5, <0.01 and <0.001 level of confidence respectively.

**Table 3.** Canopy characteristics and fruit composition of Tannat grapevines as affected by groundcover management, early irrigation, and nitrogen inputs.

| Cover crop / irrigation schemes | | | Herbicide | | UTCC | | UTCC + Irrigation | |
|---|---|---|---|---|---|---|---|---|
| Nitrogen inputs (kg/ha) | | | 0 | 100 | 0 | 100 | 0 | 100 |
| Season 2012/13 | Cluster thinning | Berry wt. (g) | 1.76 ab | 1.78 a | 1.67 b | 1.66 b | 1.66 b | 1.72 ab |
| | | Cluster w. (kg) | 0.45 | 0.43 | 0.44 | 0.44 | 0.44 | 0.45 |
| | | Yield/m (kg) | 4.12 | 4.37 | 4.21 | 4.27 | 4.21 | 4.23 |
| | | PW/m (kg) | 0.67 | 0.69 | 0.62 | 0.65 | 0.64 | 0.67 |
| | | PAR (%) | 12.9 | 12.2 | 13.9 | 12.9 | 13.6 | 12.6 |
| | | Ravaz Index | 6.1 | 6.6 | 6.3 | 6.6 | 6.7 | 6.4 |
| | | Brix | 24.3 | 24.4 | 24.4 | 24.4 | 24.6 | 24.4 |
| | | pH | 3.25 ab | 3.27 a | 3.20 b | 3.22 ab | 3.23 ab | 3.24 ab |
| | | Total Ant (mg/g) | 1544 b | 1454 b | 1709 a | 1712 a | 1742 a | 1792 a |
| | | YAN (mg/L) | 154 a | 148 a | 130 ab | 115 b | 129 ab | 114 b |
| | Full crop | Yield/m (kg) | 6.46 | 6.34 | 6.28 | 6.60 | 6.98 | 7.01 |
| | | PW/m (kg) | 0.63 | 0.68 | 0.64 | 0.66 | 0.62 | 0.66 |
| | | Ravaz Index | 10.4 | 9.8 | 9.9 | 10.1 | 11.3 | 10.6 |
| | | Brix | 20.1 | 20.3 | 19.8 | 20.0 | 20.8 | 20.3 |
| | | Total Ant (mg/g) | 1240 | 1217 | 1223 | 1177 | 1188 | 1186 |
| Season 2013/14 | Cluster thinning | Berry wt. (g) | 1.84 a | 1.87 a | 1.78 b | 1.75 b | 1.84 a | 1.85 a |
| | | Cluster w. (kg) | 0.40 | 0.42 | 0.36 | 0.38 | 0.40 | 0.41 |
| | | Yield/m (kg) | 3.55 | 3.78 | 3.19 | 3.44 | 3.63 | 3.80 |
| | | PW/m (kg) | 0.65 ab | 0.73 a | 0.50 c | 0.57 bc | 0.64 ab | 0.69 ab |
| | | PAR (%) | 9.3 ab | 4.4 b | 12.1 a | 9.8 ab | 7.8 ab | 8.8 ab |
| | | Ravaz Index | 5.7 | 5.2 | 6.6 | 6.0 | 5.7 | 5.6 |
| | | Brix | 23.5 ab | 21.9 b | 24.2 a | 24.3 a | 24.5 a | 23.9 a |
| | | pH | 3.49 ab | 3.37 b | 3.48 ab | 3.53 a | 3.46 ab | 3.54 a |
| | | Total Ant (mg/g) | 1442 a | 1159 b | 1658 a | 1542 a | 1474 a | 1540 a |
| | | YAN (mg/L) | 128 a | 142 a | 82 b | 90 b | 95 b | 116 ab |
| | Full crop | Yield/m (kg) | 4.45 a | 4.18 ab | 3.10 b | 3.46 ab | 4.04 ab | 4.35 a |
| | | PW/m (kg) | 0.62 a | 0.59 a | 0.35 b | 0.49 ab | 0.62 a | 0.64 a |
| | | Ravaz Index | 7.9 ab | 7.0 b | 9.3 a | 7.0 b | 6.6 b | 7.0 b |
| | | Brix | 21.5 c | 21.8 bc | 22.6 ab | 22.2 bc | 23.4 a | 23.5 a |
| | | Total Ant (mg/g) | 918 | 904 | 1060 | 1141 | 1107 | 1128 |
| Season 2014/15 | Cluster thinning | Berry wt. (g) | 1.75 ab | 1.77 a | 1.66 b | 1.69 b | 1.69 b | 1.72 ab |
| | | Cluster w. (kg) | 0.35 a | 0.36 a | 0.30 b | 0.33 ab | 0.35 a | 0.36 a |
| | | Yield/m | 4.32 | 4.80 | 4.24 | 4.34 | 4.23 | 4.35 |
| | | PW/m (kg) | 0.51 ab | 0.57 a | 0.40 c | 0.46 bc | 0.52 ab | 0.53 ab |
| | | PAR (%) | 12.4 ab | 10.9 b | 16.2 a | 13.8 ab | 14.7 ab | 12.6 ab |
| | | Ravaz Index | 8.7 ab | 8.5 b | 10.7 a | 9.3 ab | 9.0 ab | 8.2 b |
| | | Brix | 23.6 bc | 23.2 c | 24.4 a | 24.1 ab | 24.6 a | 24.1 ab |
| | | pH | 3.27 a | 3.25 ab | 3.22 b | 3.23 ab | 3.23 ab | 3.23 ab |
| | | Total Ant (mg/g) | 1407 b | 1422 b | 1537 ab | 1613 a | 1638 a | 1537 ab |
| | | YAN (mg/L) | 107 a | 103 a | 85 b | 81 b | 87 b | 83 b |
| | | Yield/m (kg) | 6.41 ab | 6.79 a | 4.49 c | 5.08 bc | 5.82 ab | 6.35 ab |
| | | PW/m (kg) | 0.53 a | 0.56 a | 0.35 b | 0.39 b | 0.50 a | 0.55 a |
| | | Ravaz Index | 11.5 b | 12.2 b | 13.1 a | 13.0 a | 11.6 b | 11.7 b |
| | | Brix | 22.7 | 21.8 | 22.3 | 22.2 | 21.8 | 22.3 |
| | | Total Ant (mg/g) | 913 | 869 | 983 | 980 | 918 | 949 |
| Accumulated Yield (kg) | | | 17.3 a | 17.3 a | 13.9 c | 15.1 b | 16.8 a | 17.7 a |

[a] UTCC: Under trellis cover crop; Values with different letters in single rows are significantly different at $p < 0.05$.

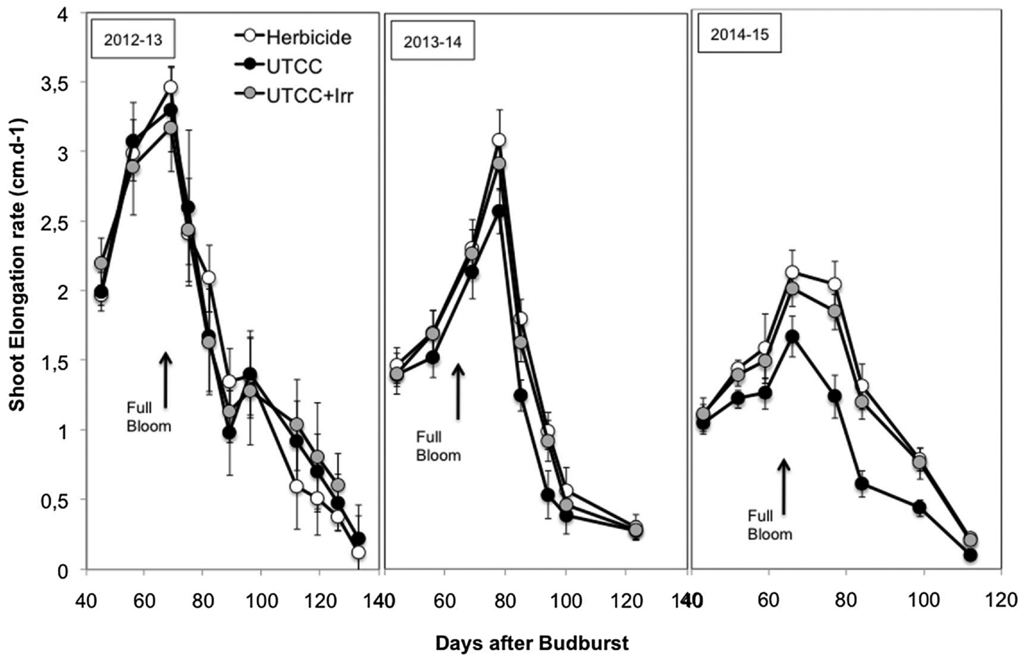


**Fig 2.** Shoot elongation rate of Tannat grapevines as affected groundcover management (H: herbicide and UTCC: under trellis cover crop) and irrigation treatments during 2012/13, 2013/14 and 2014/15 seasons in southern Uruguay.

5.4.3. Fruit yield

Under-trellis ground cover significantly affected berry weight (Table 2). Independent of N inputs, final berry weight was generally significantly reduced by UTCC and UTCC+Ir treatments, the only exception being UTCC+Ir during the 2014 harvest, when no significant differences were detected from H treatment (Table 3). Differences observed in berry weight (5–10%) were not translated into significant differences in cluster weight and fruit yield per metre (full crop vines) until the 2015 harvest, when they were reduced by UTCC compared with H and UTCC+Ir treatments. Yield of H and UTCC+Ir was comparable every season (Table 3). After three seasons, UTCC with no nitrogen or water input reduced the accumulated yield in ≅ 20% compared with the potential yield (estimates as the average of H treatments) and ≅15% when 100 units of N were applied. Nitrogen input did not consistently affect yield components (berry weight, cluster weight or yield/m) in our study (Table 2); however, a significant 8% increase with added N was detected when total UTCC accumulated yield was compared (Table 3).

5.4.4. Berry and must composition

With the exception of total anthocyanins in H treatments in the 2014 harvest, nitrogen inputs did not significantly affect fruit composition (Table 3). Even though nitrogen fertilization applied in this experiment would be considered high for the region (100 Kg N ha-1 each season), YAN concentration in grapes was also not consistently affected by nitrogen inputs (Table 2). A strong interaction Year-NI was detected (Table 2). On the other hand, ground cover (GC) treatments consistently affected every analysed fruit composition parameter except pH (Table 2). Compared to the H treatment, UTCC significantly increased fruit TSS in the last two seasons (Table 3) and total anthocyanin concentration every season (Table 3). Must YAN concentration at harvest was not affected by treatments in the first season while, independent of irrigation or nitrogen inputs, it was significantly reduced by UTCC treatments in the 2014 and 2015 harvests.

5.4.5. Vine nutrients status

No significant differences among treatments were detected in leaf blade P%, K%, Ca% and Mg% at bloom or veraison (data not shown). No visual nutrient deficiency symptoms were detected either. Leaf blade nitrogen concentration at bloom or veraison for all treatments could be considered as high in the first two seasons (> 2.9 and 3.8% in bloom and > 2.2 and 1.7% in the 2012/13 and 2013/14 seasons respectively) and relatively low or marginal in the last (< 1.9% and 0.6% at bloom and veraison respectively) (Table 4). Differences in leaf blade nitrogen concentration were not detected until the last season (2014/15), when nitrogen concentration was higher in plots with nitrogen inputs independent of under-trellis ground cover treatment (H vs. UTCC treatments). Even tissue analysis indicates an increased nitrogen uptake in fertilized plots (> leaf area with equal or higher leaf blade N concentration); values were in any case below optimum at bloom (from 1.76 to 1.86%) and clearly low at veraison (<0.58%). Nitrogen input shows a relatively low impact on leaf blade nitrogen concentration compared with differences observed from season to season.

| **Table 4.** Nitrogen leaf blade (%) as affected by groundcover, irrigation, and nitrogen inputs. | | | | | | | |
|---|---|---|---|---|---|---|---|
| **Treatment** | | **Herbicide** | | **UTCC** | | **UTCC + Irr** | |
| **Nitrogen input** | | **0** | **100** | **0** | **100** | **0** | **100** |
| Nitrogen leaf blade (%) | | | | | | | |
| **2012/13** | **Bloom** | 3.01 | 3.16 | 2.91 | 3.01 | 2.98 | 2.95 |
| | **Veraison** | 2.32 | 2.42 | 2.23 | 2.37 | 2.25 | 2.41 |
| **2013/14** | **Bloom** | 3.88 | 4.02 | 3.88 | 4.02 | 3.81 | 3.92 |
| | **Veraison** | 2.14 | 2.30 | 1.96 | 2.14 | 1.77 | 1.98 |
| **2014/15** | **Bloom** | 1.60 b | 1.86 a | 1.54 b | 1.76 ab | 1.60 b | 1.81 a |
| | **Veraison** | 0.47 b | 0.58 a | 0.39 b | 0.58 a | 0.43 b | 0.58 a |

[a] UTCC: Under trellis cover crop; Ir: Irrigation; 0 and 100: Nitrogen imput (kg/Ha). Values with different letters in single rows are significantly different at $p < 0.05$.

5.4.6. Bunch rot incidence and severity

Bunch rot incidence was significantly affected by under trellis ground cover (GC) and nitrogen inputs (NI) in our study (Table 2). Associated with a low disease occurrence in the 2013 harvest (bunch rot incidence < 3% and severity < 1%), no significant differences were detected among treatments. However, during the last two seasons, bunch rot incidence in the most affected treatment (H-100UN) reached 40% approximately (Table 5). H treatment bunch rot incidence and severity were in both seasons significantly higher than in UTCC treatments. In the 2014 harvest, nitrogen inputs increased bunch rot incidence only in H treatment and had no effect on bunch rot severity. In the 2015 harvest, nitrogen inputs increased bunch rot incidence and severity independently of under-trellis ground cover (Table 5). The effect was significant even in cases where vegetative growth was not affected by nitrogen inputs (H and UTCC treatments) (Table 3). In 2014, bunch rot incidence differences detected among ground cover treatments (H and UTCC) were already significant 20 days before harvest (Figure 3). Bunch rot incidence in H treatments progressively increased when PW/m increased over about 0.5 kg/m; below this threshold, bunch rot incidence was always under 10% (Figure 4). On the other hand, in UTCC treatments, vegetative growth (PW/m) showed a relatively low impact on bunch rot incidence (Figure 4).

**Table 5.** Bunch rot incidence and severity at harvest as affected by groundcover management, irrigation schemes, and nitrogen inputs (Kg/ha).

| **Treatment** | | **Herbicide** | | **UTCC** | | **UTCC+Irr** | |
|---|---|---|---|---|---|---|---|
| **Nitrogen input** | | **0** | **100** | **0** | **100** | **0** | **100** |
| **Harvest 2013** | **Incidence (%)** | 0.35 | 1.75 | 0.92 | 2.26 | 0.33 | 1.32 |
| | **Severity (%)** | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| **Harvest 2014** | **Incidence (%)** | 23.1 b | 38.0 a | 4.4 c | 4.5 c | 6.8 c | 8.7 c |
| | **Severity (%)** | 6.7 a | 7.5 a | 1.8 b | 1.8 b | 1.9 b | 2.3 b |
| **Harvest 2015** | **Incidence (%)** | 20.9 b | 42.7 a | 3.7 d | 7.5 cd | 3.2 d | 11.0 c |
| | **Severity (%)** | 30.0 a | 32.5 a | 3.5 c | 12.2 bc | 7.5 c | 16.9 b |

[a] UTCC: Under trellis cover crop; Irr: Irrigation;Values with different letters in single rows are significantly different at $p < 0.05$. *Botrytis* bunch rot severity was determined by visual inspection using a seven-point scale (0, 1, 5, 25, 50, 75 and 100%). *Botrytis* severity (S) was calculated as follows: $S = \Sigma Si/n$; where Si = % severity for the ith bunch and n = the total number of affected bunches.

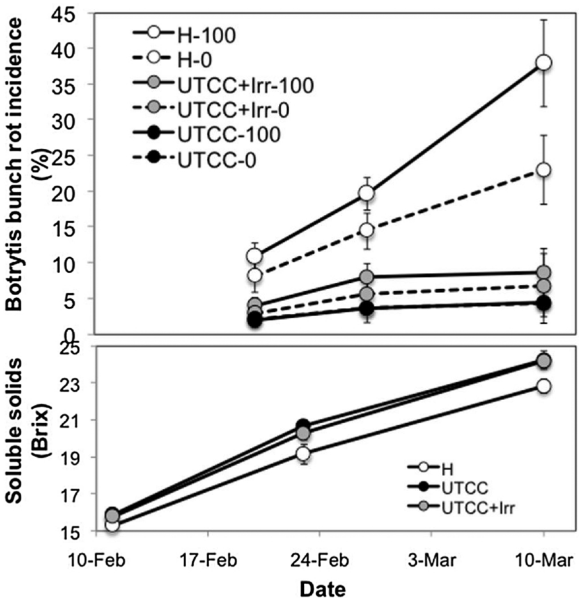


**Fig 3.** Evolution of soluble solids, and botrytis bunch root incidence of Tannat grapevines as affected by groundcover management and irrigation treatments during 2013/14 growing season in southern Uruguay. a UTCC: Under trellis cover crop; H: Herbicide; Irr; Suplementary irrigation; 0 and 100: Nitrogen imput (kg/ha).

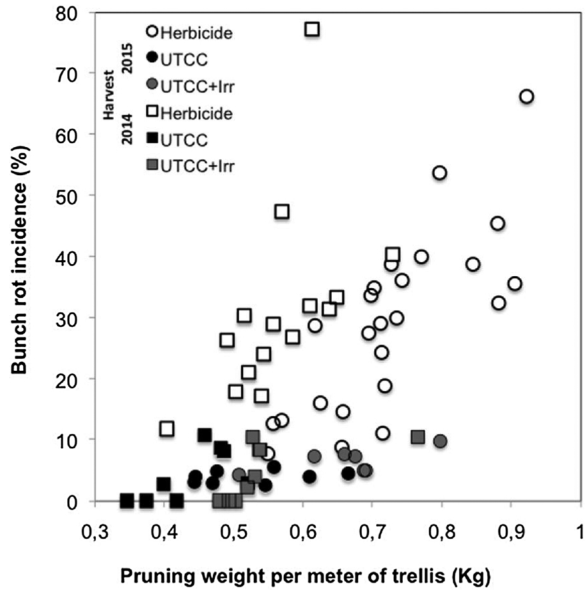


**Fig 4.** Relationship between pruning weight per meter of trellis and Botrytis bunch root incidence of Tannat grapevines as affected by groundcover management and irrigation treatments, during 2013/14 and 2014/15 seasons in southern Uruguay.
[a] UTCC: Under trellis cover crop; Irr; Suplementary irrigation.

## 5.5. DISCUSSION

### <u>5.5.1. Vegetative growth and yield</u>

Our results are consistent with literature as they show a positive relationship between grape yield and grapevine vegetative development (Coniberti et al., 2018; Guilpart et al., 2017). As previously reported, UTCC effectively reduced excessive vegetative growth of grapevines (Coniberti et al., 2018; Giese et al., 2014; Karl et al., 2016). However, the (0.40 PW/m) value observed in the UTCC-0 treatment after three seasons suggests an excessive depression of vegetative growth (as light intersection and fruit yield were significantly reduced) (Coniberti at al., 2018; Smart and Robinson, 1991) (Table 3). The most rapid growth period for grapevines was coincident with bloom (Figure 2), so the occurrence of environmental constraint during this period (water or nitrogen availability in this experiment) may explain the significant yield declines observed in UTCC-0 treatment after the first season (Table 3). A similar response was previously observed in Tannat grapevines under similar experimental conditions (Coniberti et al., 2018). It appears that water availability at bloom is a key driver of yield components and vegetative growth (Guilpart et al., 2017). Note that (1) relatively small water status differences observed between UTCC treatments (with and without early irrigation) had a great impact on accumulated yield (> 15%) and pruning mass (> 20%) (Table 3); and (2) no differences were detected between H and UTCC+Ir when water restriction (around bloom) was avoided through irrigation (Table 3).

Nitrogen input showed minor effects on grapevine vegetative growth as compared with ground cover treatment (PW/m differences were detected only for H treatment in the 2013/14 season). Previous studies reported that permanent cover crops reduced leaf N at bloom (Agulhon, 1996; Giese et al., 2014; Le Goff et al., 2000; Tesic et al., 2007), and even with deep soil and abundant water the competition exerted by the tall fescue was excessive, as determined by the associated pale-green colour of the canopy. Significant soil nitrogen uptake (e.g. up to 40 kg N ha-1) was observed for well-established Festuca arundinacea Shreb cover, reducing the available N for the grapevine (Celette et al., 2009). Cover crops may also compete indirectly for nitrogen by reducing soil nitrogen mineralization, as they might change soil temperature slightly and strongly reduce soil water content (Celette and Gary, 2013). However, in this study, due to the high soil N pools, vines' N storage and/or the use of a less competitive cover crop (red fescue), no effects on nitrogen concentration on leaf samples or visual green colour were detected in the first two seasons. Note that the tissue sample values were independent of treatment (> 2.9 and > 1.7% for bloom and veraison respectively for the first wo seasons) but strongly affected by the year (Table 2). The last season when the blade N status in all treatments indicated deficiency (leaf bade N concentration < 1.9% and 0.6% at bloom and veraison respectively) (Weir and Cresswell, 1993) nitrogen inputs consistently but modestly increased tissue N concentrations (Table 3). Nonetheless, the vines' vegetative growth was generally not significantly affected by N inputs even under restrictive conditions (the only exception being the UTCC-0 treatment).

Similarly, potential yield (full crop vines) was not significantly affected by nitrogen inputs, but the three seasons' accumulated yield of UTCC-100 was modestly but significantly higher than in UTCC with no nitrogen fertilization. In our experimental site, vines may have significant N and carbohydrates reserves even in our more restrictive treatments (UTCC-0 PW/m> 0.50 and N leaf blade content > 3.88 and 1.96 at bloom and veraison respectively). Thus cumulative effects of vineyard floor management practices on grape growth and yield may be expected over the long term

(Morlat and Jacquet, 2003; Smith et al., 2008; Winkler et al., 1974). The strength of this effect on grapevine growth may vary among years or growing conditions, but it is expected to be greater in years and/or sites where water availability is more limited (Celette et al., 2009).

5.5.2. Grape and must composition

No or limited impact on fruit composition was reported in previous studies of UTCC in humid climates (Giese et al., 2015; Karl et al., 2016). In our study, UTCC treatments increased grape TSS in two out of three seasons and consistently increased anthocyanin concentration in berry skins (Table 3). With the exception of H treatments in the 2014 harvest, the rainiest season (1129 mm from bud-break to harvest: 44% above historical value), nitrogen inputs did not significantly affect fruit composition (Table 3). The excessively shaded canopy (PW/m = 0.73 kg) in H+100N treatment may explain this result (Smart and Robinson, 1991). As expected, cluster thinning shows a positive effect on TSS and anthocyanin accumulation in grapes at the same time that yield (thinned vs. full crop vines) does not affect the treatments' relative responses. Must YAN was low overall (80–150 mg L-1) and generally reduced by both UTCC treatments compared to H. Interestingly, N additions had no consistent effect on YAN. Nitrogen is required for yeast growth and completion of alcoholic fermentation in grape juice; concentrations of 130–160 mg L-1 of yeast fermentable nitrogen (primary amino acids and NH4+) are normally required for complete fermentation (Bell and Henschke, 2005; Spayd et al., 1995). A significant relationship between must amino acids and wine aromatic composition has also been described (Hernandez-Orte at al., 2002; Trigo-Córdoba at al., 2015). In this context, the low must YAN concentration consistently achieved in UTCC (from 80 to 100 mg L-1) may be considered a negative outcome of the treatment (Table 3). Clearly, interpreting N effects is complex as many of the expected relationships and tissue measurements gave unexpected results, regardless of other treatments. This is an area that greatly needs better understanding.

5.5.3. Bunch rot incidence and severity

As expected, bunch rot incidence was progressively reduced with PW/m independent of treatments (Figure 4) (Coniberti et al., 2018; Guilpart et al., 2017; Valdes-Gomez et al., 2008). This important factor involved in *Botrytis* development interacts with climate and microclimate, so the effect of grapevine vegetative growth on grey mould expression results from direct effects (increased size and number of leaves and berries) and indirect ones (via microclimate), which are difficult to dissociate in such field experiments (Valdes-Gomez et al., 2008). However, the novelty of our results is the strong difference detected in bunch rot incidence between H and UTCC+Ir treatments, even when vegetative development (shoot elongation rate, PW/m, PAR%), berry size and fruit maturation (TSS and pH) were in most cases comparable. It has been reported that sites with typically low yeast available nitrogen (YAN) tend to have a lower incidence of *Botrytis* bunch rot; nevertheless, this may be related to an indirect response to low vine nitrogen levels and vigour rather than a decreased susceptibility of berries because of low YAN levels (Mundy and Beresford, 2007). In our study, bunch rot incidence was significantly higher in treatments with nitrogen inputs even when vegetative growth or canopy density (PW/m, PAR%) were not affected. Additionally, UTCC significantly reduced juice N levels at harvest. Our results therefore suggest that grapevine nitrogen status could be one of the direct factors affecting bunch rot susceptibility. Nevertheless, the strong difference observed in disease occurrence between UTCC and H treatment could be explained by more than just this factor, since no significant YAN differences were detected between H and UTCC-100 treatments in the 2014 harvest, but strong differences in bunch rot occurrence were detected. Guilpart et al. (2017) showed that water stress at flowering was strongly correlated with susceptibility to diseases (powdery mildew and grey mould), suggesting that cover crop management could be a relevant lever to drive soil water status and reduce disease incidence. Valdes-Gomez et al. (2008) investigated the relationship between grey mould expression, climate and microclimate, canopy development, and associated morphological and biochemical features of grape clusters and berries. In order to create different levels of canopy development, the authors test two types of cropping system: (i) inter row perennial cover crop (a mixture of Festuca arundinacea Shreb and Lolium perenne L) (CC) and (ii) chemical weed control with glyphosate all over the soil surface (H) and an additional treatment, where in order to generate higher vegetative growth, vines from H treatment were irrigated once a week from bud-break to harvest and fertilized with approximately 100 NU. They report that in vigorous vines, which were both irrigated and fertilized, grey mould developed even during dry summer conditions. In the same study, no disease was developed in low vigour vines, even when, to modify microclimate favouring the disease, water was sprayed at the fruit zone (for 24 h ten days before harvest). Emerging evidence indicates that drought stress tolerance in grapevines involves the activation of polyamine oxidation, suggesting an improved immune response and reduced susceptibility to *Botrytis cinerea* (Hatmi et al., 2014). Seasonal variations in water status observed in our study could therefore also have a relevant impact on disease susceptibility at harvest via such a molecular basis. Note that, during the study, water status in H treatment was never below -0.6 MPa Ψstem, while UTCC treatments reached values every season below -0.9 MPa Ψstem (Figure 1). Confirming a direct link between seasonal grapevine water stress and disease susceptibility at harvest would be extremely relevant to define tactical management options in humid environments.

5.5.4. Adaptive strategy

As was previously reported, grapevines should experience moderate water stress after the flowering stage to limit excessive grapevine vegetative development (Coniberti et al., 2018; Pellegrino et al., 2006). However, under variable weather conditions, the introduction of UTCC in vineyards may also induce excessive competition with the grapevine in dry periods (Coniberti et al., 2018). This competition may be particularly risky in non-irrigated vineyards. For instance, in our study, UTCC accumulated yield was reduced by more than 20% in a relatively short-term period, even when precipitation from bud-break to veraison was average or above average and irrigation was provided to avoid severe water stress periods (below -0.9 MPa Ψstem). Our results demonstrate how such excessive competition between the cover crop and grapevines is less critical to consider in irrigated and/or fertilized conditions (UTCC+Ir). It was possible through ground cover and irrigation management to reduce grey mould incidence even when vegetative growth and yield were not significantly affected (UTCC+Ir). Previous studies (Guilpart et al., 2017; Hatmi et al., 2014; Valdes-Gomez et al., 2008) also suggest that, besides vine vegetative growth, seasonal water status may have a relevant impact on grape disease susceptibility at harvest. Therefore inducing water stress at particular stages during the season (e.g. after post-bloom period to yield formation) may be a suitable strategy leading to a reduced use of pesticides in humid environments without impairing farmers' economic return. In this context, flexibility and adaptive management such as complementary irrigation and mineral nutrition may be recognized as relevant to reach an adequate balance between cover crops' ecosystem services and disservices.

## 5.6. CONCLUSION

Although the interaction between a cover crop and the vine is complex and dynamic, we demonstrated how, under the experimental conditions, it is possible to control vegetative growth of grapevines through a combination of cover cropping, nitrogen inputs and supplemental irrigation if needed. The grapevine-UTCC water competition during the period of maximum growth rate (bloom ± 20 days period) was the main factor affecting vine growth and yield. Nitrogen inputs slightly increased the accumulated potential yield of UTCC, but water availability around bloom was a key regulator of vegetative growth and yield components as H and UTCC+Ir accumulated yields were comparable. TSS and anthocyanin accumulation in grapes was modestly increased by UTCC treatment due to reduced berry size; however, the opportunity to delay harvest to accomplish full maturation in UTCC plots, due to lower bunch rot incidence, was the most practically significant outcome in this climate. Although a reduction in bunch rot incidence can be achieved in some circumstances associated with reduced growth and better cluster microclimate, we found that reduced susceptibility can be achieved without major growth reduction. A direct physiological process may be involved and seasonal variations in water status may have a relevant impact on disease susceptibility at harvest. These findings may be relevant for vineyards located in humid areas in which bunch rot incidence may be a main limitation for full ripening of the crop. Site-specific tactical decisions may depend not only on environmental conditions but also on the objective of grape yield and quality, where short- and long-term temporal scales have to be also considered in order to satisfy the farmer's objectives. However, even though under some circumstances UTCC may have detrimental effects, in combination with complementary irrigation and/or mineral nutrition it may offer an economic and environmentally sustainable alternative to consistently produce high quality wines in a humid climate. Further investigation should be conducted to better understand the direct factors affecting bunch rot susceptibility.

## 5.7. ACKNOWLEDGMENTS

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# 6. UNDER TRELLIS COVER CROP VERSUS DIVIDED CANOPY TRELLIS SYSTEM AS POTENTIAL TOOLS TO CONTROL EXCESSIVE VINE GROWTH IN TEMPERATE HUMID CLIMATE[4]

## 6.1. ABSTRACT

The main objective of this work was to improve Tannat grapes and wine composition, by achieving appropriate vine balance in high capacity growing conditions. We tested Vertical Shoot Positioned (VSP) versus Lyra trellis systems with conventional floor management consisting of alleyway tall fescue with 1.0 m wide weed-free strips under the trellis (VSP-H and Lyra-H), and VSP with under-trellis cover crop (VSP-UTCC). UTCC consists in the full cover of the vineyard soil with tall fescue (Festuca arundinacea). Deficit drip irrigation was provided at mid-day stem potential (SWP) thresholds of -0.9 MPa. Treatments were arranged in a split-split-plot randomized block design with trellis system (Lyra vs VSP) as main plots, flour management schemes (H vs UTCC) as subplots and crop load (thinned cluster vs. full crop vines) as sub-subplots. The experiment was conducted over three growing seasons in Southern Uruguay. Shoot growth rate, SWP, berry size and berry composition were monitored over the seasons, as well as final vine yield, cluster weights, and pruning weights. VSP-H pruning weight/m values were always above 0.65 kg/m associated with excessively shaded canopies. UTCC limited vine water availability, reduced vine growth rate and final canopy size to optimal values, while reducing berry size and increasing soluble solids and anthocyanin concentration in grapes. The Lyra-H system produced, for most evaluated parameters of canopy growth and microclimate, comparable values with VSP-UTCC, however fruit composition was not consistently improved compared to VSP-H treatment. Our results suggest that an improved fruit composition can be achieved in some circumstances associated to an improved microclimate of leaves and clusters, but the physiological process involved in vegetative growth regulation is even more relevant for fruit composition. Even though the specific mechanism by which UTCC induced a higher tolerance to *Botrytis* bunch

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rot was not identifying, a clear effect on fruit firmness and more relevant pathogen or host plant behavior was detected.

**Key words:** Tannat, *Botrytis* bunch rot, grape composition, Fescue, vine vigor.

## 6.2. INTRODUCTION

In areas as Uruguay where the temperature, water availability and soil fertility, induce high growth rates, it is common to observe dense and unbalanced canopies generating unfavorable microclimates for fruit maturation and disease management (Smart and Robinson 1991). The effects of the amount of exposed leaf area on yield and fruit composition are best appreciated by recognizing the role played by photosynthesis and the factors influencing its efficiency (Reynolds and Vanden Heuvel, 2009). The proportion of interior leaves to exterior leaves may also affect carbon balance of the vine (Vanden Heuvel et al. 2002). There is no universally accepted recommendation for leaf layer number of a canopy as it is affected by geography and cultivar, but it is highly accepted that elimination of internal canopy shading of excessively large vines leads to improved fruit composition at a given crop size (Dry et al. 2005). Values of 0.3 to 0.6 kg per m of canopy length are generally considered to be within the optimal range of vegetative growth (Kliewer and Dokoozlian 2005). The main grower's challenge is to avoid excessive vine growth while reaching the highest sunlight interception as possible in order to maximize each vineyard's potential yield. However, in regions with unpredictable and high precipitation rate, it isn't simple to consistently maintain optimal vine growth and in most cases corrective practices (shoot thinning, leaf removal, and hedging) represent a large part of grower's job and production costs. Canopy division involves a modification to the configuration of the trellis so that generally two canopies are created from the initial single canopy or curtain. Compared to VSP, Lyra may have similar leaf area and pruning weights but considerable less vigor (Shoot growing rate, cane pruning weight and canopy density), higher light intersection yields, enhanced node per vine, node fruitfulness resulting from a reduction in canopy shade, and improved fruit composition (Carbonneau et al. 1978). However, even though under Uruguayan conditions Lyra system effectively reduced vegetative growth of excessively vigorous vines, fruit composition was in general not affected by canopy division (Lyra vs VSP) (Disegna et al. 2005a). Due to the high influence of professor Carbonneau in Uruguayan viticulture, Uruguay is probably the country were Lyra system has been more adopted by the industry, reaching in 2008 close to 30% of the area with vineyards (INAVI, 2015). However, associated with the increased mechanization, most new vineyards (<5 years old) are trained on VSP (90%) and only 8% in Lyra system (INAVI, 2015).

Many techniques were tested in national research trials with the objective of controlling excessive vegetative growth of VSP vineyards. The use of less vigorous rootstocks as Riparia Gloire (Disegna et al. 2001) and root pruning, have shown a limited vigor reduction in most seasons and a poor performance under water deficit periods (Disegna et al. 2001). The effect on vine vigor and grape composition from the use of highly competitive cover crops (Festuca arundinacea) in the alleyways has been limited and inconsistent (Filgueira 2005).

More recently, the effect of under-trellis cover crops (UTCC) has been studied mostly in cool climate regions that experience excessive water availability and fertile soils. As a result of the competition for soil moisture and nutrients, UTCC has been reported to reduce vine vigor and yield (Giese et al. 2015), improve fruit exposure and canopy microclimates indicators (Hatch et al. 2011), however fruit composition has not consistently improved (Giese et al. 2015, Karl et al. 2016, Lopez et al. 2008). In most studies, effects of nutrients (mostly vine nitrogen status) and soil moisture on vine growth cannot be clearly distinguished (Giese et al. 2014, Giese et al. 2015, Lopez et al. 2008, Karl et al. 2016). However, water availability seems to be the main factor affecting grapevines responses to under vine ground cover (Centinari et al. 2015, Coniberti et al.2018).

In this study, our first goal was to evaluate UTCC as a potential tool to regulate vine vegetative growth in a humid climate condition. Our approach was to use UTCC to limit vine water availability, reduce vine growth rate and final canopy size. However, to avoid excessive water stress due to the cover crop competition, supplemental irrigation was applied when water deficit reached a minimum level (-0.9 MPa) independently of the treatment. The different treatments were applied on VSP vineyards and compared with the Lyra trellis system, which is the most effective tool used to control excessive vine vigor in our region. The effects of UTCC on vine size, yield components and fruit composition were determined. The second goal was to test the general hypothesis that, fruit composition is affected by vine vegetative expression but the mechanism involved in the vine growth control (water versus shoot competition) could be even more relevant.

## 6.3. MATERIALS AND METHODS

### 6.3.1. Experimental site

The experiment was conducted during 2011-14 growing seasons in Southern Uruguay (latitude 34º South). Uruguayan climate can be classified as temperate – humid without a prolonged dry season according to the Köppen-Geiger classification. Historical mean total annual rainfall in Southern Uruguay is 1100 mm/yr, but 650 mm of those fall during the growing season (Table 1). Further weather data details can be accessed at http://www.inia.org.uy/gras/agroclima/cara_agro/index.html. Soil was classified as a Tipic Argiudolls according to the USDA soil classification system (Soil Survey Staff, 1999), with a variable depth of 0.90 to 1.0 m and silty clay texture. The total soil available water (field capacity-permanent wilting point) to 1.0 m depth was 110 mm.

### 6.3.2. Experimental vineyard and general vine management

The experiment was conducted on Tannat grapevines grafted on to SO4 rootstock. The vineyard was ten years old when cover crop was installed in March 2011. Vines were in rows north-south oriented and 2,6 m apart. In both trellis systems (VSP and Lyra) plants were pruned to seven two-bud spurs per meter of cordon during dormancy. At approximately 30 cm shoot length, all infertile shoots as well as shoots not located on spurs were removed. During the growing season, shoots were positioned by hand vertically above the spurs and topped 30 cm above the top wire. Standard disease control program was applied for downy mildew, powdery mildew, and Botrytis bunch rot. Supplementary irrigation water was applied with drip emitters (4 L/m) located directly under the vine row and separated 0.3 m from each other. The irrigation system allowed to independently irrigate single experimental plots.

**Table 1.** Irrigation by treatment and evapotranspiration, precipitation and growing degree-days (>10ºC), from Las Brujas weather station located at 200 m from the experimental site.

| | Phenological stage | Degree-days (>10ºC) | Eto Penman (mm) | Precipitation (mm) | Irrigation (mm) VSP-H | L-H | VSP-UTCC |
|---|---|---|---|---|---|---|---|
| Historical | budbreak - bloom | 338 | 208 | 227 | | | |
| | bloom - veraison | 718 | 359 | 291 | | | |
| | veraison - harvest | 664 | 257 | 266 | | | |
| | post-harvest | 421 | 136 | 252 | | | |
| Season 2011/12 | budbreak - bloom | 328 | 224 | 132 | 0 | 0 | 0 |
| | bloom - veraison | 722 | 391 | 177 | 14.7 | 18.3 | 98.5 |
| | veraison - harvest | 665 | 271 | 212 | 20.3 | 33.5 | 76.8 |
| | post-harvest | 435 | 157 | 147 | 0 | 0 | 0 |
| Season 2012/13 | budbreak - bloom | 403 | 211 | 359 | 0 | 0 | 0 |
| | bloom – veraison | 753 | 381 | 288 | 0 | 0 | 27.1 |
| | veraison - harvest | 641 | 291 | 199 | 0 | 0 | 8.2 |
| | post-harvest | 352 | 139 | 173 | 0 | 0 | 0 |
| Season 2013/14 | budbreak - bloom | 332 | 214 | 204 | 0 | 0 | 0 |
| | bloom - veraison | 804 | 398 | 284 | 0 | 0 | 50.2 |
| | veraison - harvest | 692 | 228 | 641 | 0 | 0 | 0 |
| | post-harvest | 395 | 146 | 194 | 0 | 0 | 0 |

[a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system.

### 6.3.3. Treatments

Treatments were arranged in a split-split-plot, randomized block design with four replicates. Main plots, comprising two adjacent rows compared Trellis systems (Lyra vs VSP), subplot compared UTCC with conventional under-trellis herbicide floor management (H), and sub-subplots compared the effect of crop load (cluster thinning vs full crop vines). Treatments consisted of (1) VSP trained vines with conventional floor management (VSP-H), (2) Lyra trained vines with conventional floor management (Lyra-H) and (3) VSP trained vines with under-trellis cover crop (VSP-UTCC). Each treatment plot consisted of 18 grapevines, the outer 2 serving as guard vines. To avoid overcropping affecting maturation, crop level was adjusted by cluster thinning at veraison (1 cluster per shoot at stage 35 - Eichhorn and Lorentz 1977) in half of the vines. It allowed to evaluate in adjusted yield subplot the potential fruit composition enhancement expected in more vegetative balanced canopies (Lyra-H and VSP-UTCC) and the potential yield in full crop sub-subplots.

In the VSP system, vines were spaced 0.9 x 2.6 m (vine x row spacing). Unilateral cordon-trained vines were pruned to seven two-bud spurs per meter. The height of the cordon was 1.0 m, and the last wire was located 1.9 m above the ground. In the Lyra system, vines were spaced 1.0 x 3.4 m (vine x row spacing) in north-south oriented rows. Bilateral cordon-trained vines were pruned to seven two-bud spurs per meter of trellis. The two parallel cordons were located 0.9 m apart at a height of 1.0 m. The last wire was located 1.8 m above the ground. The UTCC treatment consisted in the full cover of the vineyard soil with tall fescue (Festuca arundinacea). To avoid the effect of the treatment due to nitrogen (N) competition, in every UTCC plot ammonium nitrate ($NH_4NO_3$) was applied twice at a rate of 20 kg/ha N when shoots reached approximately 30 cm and after fruit set (stage 29 - Eichhorn and Lorentz, 1977). No statistically significant differences among treatments were detected in leaf N%, P%, K%, Ca% and Mg% at bloom or veraison (data not shown). Average leaf N% content ranged from 2.1 to 2.5%. No nutrient deficiency symptoms were detected. The conventional management scheme (H) consisted in the same inter-row groundcover, but combined with a 1.0 m wide weed-free strip under the trellis. The under-trellis, weed-free strip was maintained with a combination of herbicides.

### 6.3.4. Vegetative measurements

Shoot growth rates were obtained by repeated measures of shoot length. Two shoots from two representative vines per plot were tagged and measured on a ~ weekly basis starting shortly after bud-break. At veraison, photosynthetic active radiation (PAR) available in the fruit zone, was estimated with an average of three readings taken in the canopy fruit zone, with the ceptometer (AccuPAR L80; Decagon Devices, Pullman, WA) inserted parallel to and 15 cm above the cordon: one with the sensor face angled 45° to the east, a second vertically upright, and the third angled 45° to the west. Incident radiation was measured by orienting the ceptometer vertically upright, at the bunch zone height outside the canopy (ambient). PAR measurements were made ±2 hours of solar noon on cloudless days. The pruning of every experimental plant was weighed and averaged by plot. Ravaz index (RI, fruit yield/pruning weight) and average shoot pruning weight was later calculated. Every shoot from tagged plants was individually weighed and average cane pruning weight of those selected plants was calculated.

6.3.5. Water status and supplementary irrigation.

Midday stem water potential (Ψstem) measurements were periodically performed (~ bi-weekly) between 1400 and 1600 hr using a leaf pressure chamber (Soil Moisture Equipment Corp., Santa Barbara, CA) in two leaves from two vines per treatment replication (Chone et al. 2001). Plastic bags covered with aluminum foil were placed on well-exposed mid-shoot leaves one hour before the onset of Ψstem measurements. The time from cutting leaf petioles to insertion and chamber pressurization was less than 30 sec. The chamber was pressurized with nitrogen gas at a rate of 2 to 4 sec/0.1 MPa. Independently of treatments, supplementary irrigation water was applied once the plants reached -0.9 MPa mid-day stem water potential (Ψstem) in order to avoid severe water stress. With the goal of avoiding a Ψstem more negative than -1.1 MPa, when this occurred the amount of water the vines had consumed the previous week was applied (100% ETc). ETo was obtained from the weather station of INIA Las Brujas Experiment Station located < 200 m from the experimental site. Mid-day canopy shaded area (light interception) was estimated using a Paso Panel and each plot's crop coefficient (Kc) was calculated using the formula proposed by Williams and Ayars (2005); Kc = (0.017 * Shaded percentage of field) – 0.008. ETc was calculated by multiplying ETo by Kc and used to estimate weekly vine water consumption.

6.3.6. Soil moisture

To better control irrigation, in every field plot, Frequency Domain Reflectance sensors (10HS, Decagon Devices, Inc., Pullman, WA) were installed under the trellis. Two sensors were installed between selected plants at ~25 and ~50 cm depth for continuous data logging (60 min intervals) during the growing season using Em50 Digital/Analog Data Logger (Decagon Devices, Inc., Pullman, WA) (data collected are not presented in this study).

6.3.7. Berry development and fruit composition

From fruit set to harvest, randomly collected berry samples were taken from each treatment replicate. Eighty berries from fruit set to veraison and one hundred berries from veraison to harvest were collected on a bi-weekly basis. In pre-veraison samples only berry weight was analyzed. In post veraison samples, berry weight, total soluble solids (TSS), titratable acidity (TA), pH and total anthocyanin were analyzed. SS was determined using a hand refractometer (Atago, model N1, Tokio, Japan). Must pH was measured with a pH meter (Horiba, model F13, Kyoto, Japan) and titratable acidity (TA) was determined by titration (NaOH 0.1 N) and expressed as tartaric acid equivalents (w/w). Total anthocyanin analyses were performed according to Glories (1984). From veraison to harvest, the percentage of bunches infected by Botrytis bunch rot (incidence) as well as the percentage of each bunch that was infected (severity) was determined by visual inspection using a six-point scale (0, 5, 25, 50, 75 and 100%).

6.3.8. Fruit firmness.

Due to the clear effect of UTCC on berry firmness observed during the first two seasons, at harvest 2014 fruit firmness evaluation was included. Randomly collected berry samples were taken at harvest time from each treatment replicate. Firmness as deformation (mm) was evaluated by compressing the berry equator (pedicel positioned horizontally) using a 6.3 mm diameter flat stainless steel probe (applied force 1N and test speed 0.5 mm/s) (TaXT2i Texture Analyzer; Texture Technologies). The test was conducted on 40 healthy berries (whole berry), positioned on the base of the texture analyzer at ambient temperature (20 ± 2 ºC). To guarantee the desired degree of confidence in the results, ripening stages were confirmed by visual analysis of external fruit color and TSS measurements. The relationship between fruit firmness and TSS of individual berries was analyzed (not significant). In the same way, the relationship between fruit firmness and berry size (berry weight) was analyzed and discarded as a possible cause of differences detected.

6.3.9. Harvest evaluation

All treatments were harvested at the same date. Additionally to chemical evaluation, bunch rot incidence and severity was considered to define harvest date. Total fruit yield and clusters per vine were determined for each experimental plot. Mean cluster weight was calculated. The incidence and severity of Botrytis bunch rot were visually estimated. From the harvested fruits, about 10 kg of grapes per plot were crushed and juice SS, pH, TA were analyzed. In addition, samples of 200 berries per replication were taken, to prepare homogenized fruit extracts. From these extracts, total anthocyanin was analyzed as previously described.

6.3.10. Wine making and analysis

Approximately 10 kg of fruit per treatment replicate were retained for winemaking. After storage for 16 hr at 5 ºC, grapes were crushed and destemmed. Must samples were taken, and grapes from adjacent field blocks (B) were combined (B1 + B2 and B3 + B4) giving a total of two wine replicates for each treatment. Sulfur dioxide was added at a rate of 60 mg/kg and the must was inoculated with Saccharomyces cerevisiae (ALG111, DSM, Delft, The Netherlands) at 25 g/hL. Fermentation temperature was maintained between 26 and 30 ºC. Alcoholic fermentation was completed six to eight days after the beginning of fermentation, but maceration was allowed for 10 days. After maceration was completed, wines were pressed, and placed into sterile 10-L glass containers where malolactic fermentation was completed. After completing malolactic fermentation, wines were cold stabilized at 0 ºC for two weeks, SO2 was added and the wines were stored in sterile 5-L glass containers at 11 ºC. For each wine, a 125 mL sample was taken for analysis (alcohol concentration, TA, pH levels) by standard methods (OIV 2009). Total anthocyanins were calculated from the absorbance at 520 nm as described by Glories (1984).

6.3.11. Weather conditions

Precipitation occurring during the three growing seasons was variable in amounts and patterns (Table 1). Compared to the historical value, precipitation accumulated during 2011/12 was below average in every phenological stage, making it necessary to irrigate all treatments. Precipitation that occurred from bud-burst to harvest 2012/13 was close to average (≅800 mm), but significantly higher from bud-burst to bloom (≅160%) and lower from veraison to harvest (≅75%). In season 2013–14, accumulated precipitation from bud-burst to veraison was near average but markedly higher from veraison to harvest (≅240%). Accumulated growing degree-days (base 10°C GDD) during the three growing seasons was similar to the historic average for the region (≅2200 GDD) (Table 1).

6.3.12. Data analysis

The effects of trellis system (TS), ground cover (GC), Crop load (CL) and their interactions were assessed using linear mixed effects models following a split-split-plot design with a repeated measure. Year, TS, GC, CL and their interactions were considered as fixed effects. As random effects: Block, TS (main plot) and GC (Subplot), were included. The appropriateness of the random and correlation structures was analyzed by comparing nested models with and without the structures with the likelihood ratio test using the restricted maximum likelihood estimation procedure. Since significant year*treatments interactions were detected, treatments effects were also analyzed by season to allow seasonal patterns to be observed. A Tukey HSD test (5% significance level) was used to compare treatment means. Statistical analyses were performed using InfoStat computer software (Di Rienzo, et al. 2017).

## 6.4. RESULTS

### 6.4.1. Vine water status

During the study, precipitation at the beginning of the season was below average only during spring 2011 when irrigation was needed in every treatment. Precipitation at seasons 2012/13 and 2013/14 was average for the region previous to veraison and below average from varaison to harvest 2012/13 (75% of the historical value), however vines under the conventional floor management treatment (H) never reached the -0.9 MPa Ψ stem threshold and consequently irrigation was not applied. On the other hand, it was necessary to irrigate VSP-UTCC treatment every season, showing the potential scope of UTCC as a tool to regulate water availability under the experimental conditions. No significant differences were detected among trellis systems water status in our study, however during the only longer period of water deficit occurred in our study (from budbreak to veraison 2011/12), Ψstem was significantly lower in Lyra-H compared to VSP-H treatment (Figure 1). Those differences were significant even when due to its higher Kc, irrigation amount provided in the Lyra-H was higher compared to VSP-H treatment (Table 1).

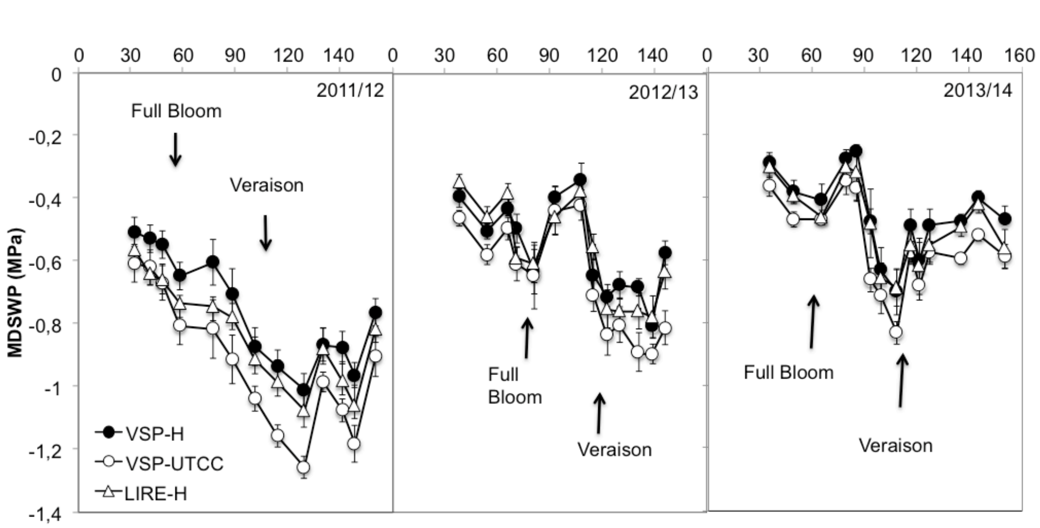


Fig 1. Midday ψstem of Tannat grapevines as affected by trellis system and groundcover managements during 2011/12, 2012/13 and 2013/14 seasons in southern Uruguay. [a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system.

## 6.4.2 Vegetative growth

After three seasons, under-trellis ground cover (GC) and trellis systems significantly affected vegetative growth (PW/m, Cane pruning weight, shoot growth) while no effect on crop was detected (Table 2). During the first season, final canopy size (PW/m) in both trellis systems reflected the differential shoot growth rate among treatments during the 30 days period after bloom (Figure 2). No significant differences were detected previous to or after this period among treatments (Figure 3). Vine water status was the main environmental factor affecting vegetative growth. Shoot growth rate was well correlated to Ψstem between -0.3 and -0.8 Mpa, (R2= 0.65 and 0.88 for Lyra and VSP respectively - average of three measuring dates), while no significant growth was detected in any period when the vine Ψstem was lower than -0.9 Mpa. Shoot growth rate was also significantly affected by trellis system (Figure 2). At equal water status (Ψstem) VSP vines showed a higher shoot growth rate than vines in Lyra (Figure 2). The last two seasons, vine water status during this critical period was not remarkably different among treatments (and always above -0.5 Mpa SWP) (Figure 1), so no strong correlations were observed. Finally canopy size was more associated to previous seasons vine vegetative expression ($R^2$ > 0.70). Consequently VSP-H was every season the most vigorous treatment (Shoot growth rate, cane pruning weight or PW/m). Over the three seasons VSP-H vines produced consistently lower PW/plant but higher PW/m compared to vines trained in Lyra-H. In hectare basis both trellis system produced comparable PW while light intersection (shaded area) was significantly higher in Lyra (from 35% to 42% higher compared to VSP-H). UTCC consistently reduced both pruning weight and shaded area compared to values observed in VSP-H treatment (control). PW/m in VSP-UTCC and Lyra-H treatments were comparable until the thirst season when Lyra-H PW/m was significantly higher than VSP-UTCC treatment. Both techniques were effective reducing vine vigor (shoot growth rate, cane pruning weight) and increasing its associated parameters of canopy density (PW/m and PAR%), however vine capacity estimated through the shaded area under the vine, was significantly higher in Lyra-H. Lyra system produced similar vigor and canopy density as VSP_UTCC but with drastically higher light intersection either per plant or hectare (around 135% and 80% respectively).

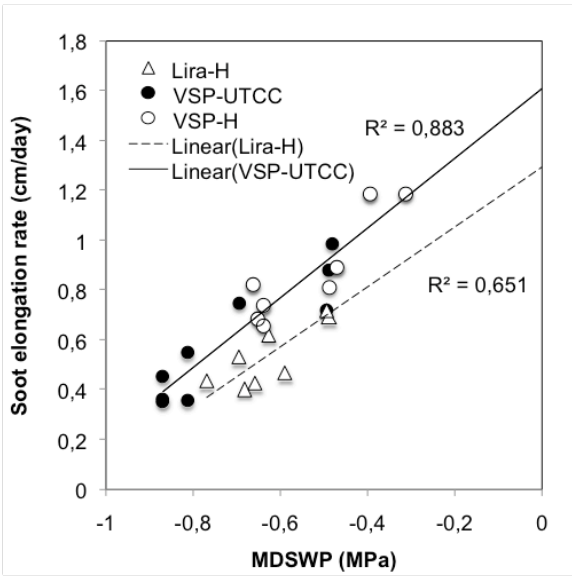


Fig. 2. Shoot elongation rate of Tannat grapevines under different trellis and groundcover management treatments. [a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system.

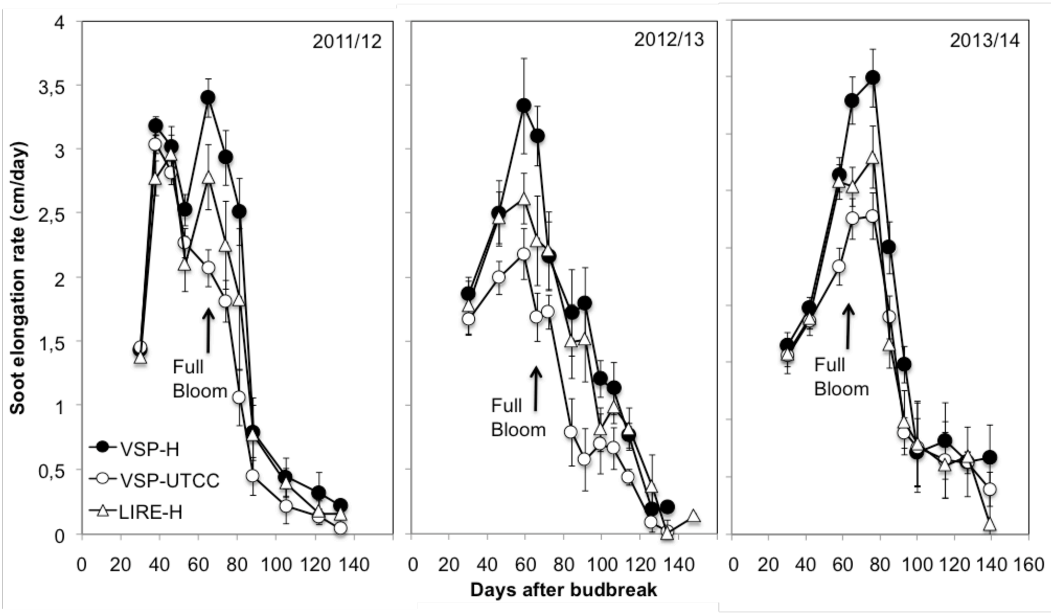


Fig 3. Shoot Elongation rate of Tannat grapevines under different trellis systems and groundcover management treatments. [a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system.

**Table 2.** Analysis of variance for canopy characteristics, fruit yields, grape must and wine composition and bunch rot incidence and severity of Tannat grapevines as affected by trellis system (VSP vs. Lyra), groundcover management (herbicide and under-trellis cover crop) and crop load (cluster thinning vs. full crop vines) from an experimental site in southern Uruguay.

| | Year (Y) | Trellis (TS) | Ground cover (GC) | Crop load (CL) | Y*TS | Y*GC | Y*CL | TS*CL | GC*CL |
|---|---|---|---|---|---|---|---|---|---|
| **Canopy characteristics** | | | | | | | | | |
| Pruning weight (kg/m) | 3.44* | 10.38** | 87.31*** | 0.10 | 2.34 | 0.70 | 1.52 | 0.10 | 2.1E-03 |
| Shoots/m | 20.95*** | 0.90 | 1.07 | 0.17 | 4.62* | 0.03 | 1.21 | 0.04 | 0.16 |
| Pruning weight/cane (g) | 18.11*** | 10.76** | 114.94*** | 1.2E-0.3 | 0.56 | 0.64 | 0.60 | 0.05 | 0.01 |
| PAR % | 19.74*** | 0.31 | 80.16*** | 1.06 | 1.08 | 7.67** | 0.35 | 0.15 | 2.23 |
| Shaded area m2/m | 8.75*** | 0.31 | 60.20*** | 1.43 | 0.21 | 0.12 | 0.18 | 0.44 | 0.02 |
| Pruning weight/ha (kg) | 3.41* | 29.80*** | 63.98*** | 0.06 | 3.08* | 0.51 | 1.61 | 0.09 | 1.6E-03 |
| Shaded area/ha (m2) | 7.24** | 234.8*** | 37.45*** | 1.54 | 0.23 | 0.08 | 0.25 | 0.89 | 0.02 |
| **Fruit yields** | | | | | | | | | |
| Cluster weight (g) | 59.03*** | 8.64** | 16.16*** | 2.09 | 5.35** | 0.42 | 2.86 | 1.70 | 0.22 |
| Berry weight (g) | 15.60*** | 41.84*** | 45.96*** | 1.77 | 2.42 | 6.69 | 0.91 | 0.85 | 0.01 |
| Cluster/shoots | 9.81*** | 18.49*** | 0.96 | 28.63*** | 4.00* | 1.10 | 3.57* | 0.05 | 0.45 |
| Cluster/m | 33.54*** | 36.04*** | 1.9E-05 | 57.79** | 2.1E-03 | 1.13 | 4.26* | 0.44 | 0.31 |
| Vine yield (kg/m) | 37.94*** | 21.62*** | 6.61* | 44.25*** | 0.12 | 2.01 | 7.93*** | 1.10 | 0.24 |
| Yield (Ton/Ha) | 49.77*** | 36.46*** | 6.16* | 53.29*** | 3.38* | 1.97 | 7.94*** | 0.05 | 0.22 |
| Ravaz index | 11.84*** | 1.77 | 42.72*** | 16.82*** | 2.82 | 0.21 | 4.12* | 0.03 | 1.33 |
| **Grape and must composition** | | | | | | | | | |
| Brix | 190.60*** | 2.01 | 9.24** | 25.01*** | 0.81 | 0.22 | 0.98 | 0.55 | 7.55** |
| TA (g/L) | 59.86*** | 1.01 | 9.65** | 0.44 | 0.15 | 0.70 | 0.94 | 0.80 | 0.29 |
| pH | 31.68*** | 0.39 | 0.52 | 6.03** | 0.84 | 0.11 | 0.99 | 1.6E-03 | 0.12 |
| Anthocyanins (mg/L) | 138.60*** | 5.40* | 12.17** | 52.33*** | 0.03 | 2.66 | 10.75*** | 1.66 | 2.91 |
| YAN (mg/L) | | | | | | | | | |
| **Wine composition** | | | | | | | | | |
| TA (g/L) | 4.77* | 1.88 | 0.26 | 1.12 | 3.00 | 3.56 | 2.16 | 0.31 | 0.01 |
| pH | 54.58*** | 0.82 | 0.04 | 2.28 | 0.30 | 0.09 | 2.67 | 3.1E-03 | 0.73 |
| Ethanol (% v/v) | 48.92*** | 14.62*** | 35.44*** | 42.94*** | 10.69*** | 1.98 | 5.49* | 21.09*** | 16.67*** |
| Anthocyanins (mg/L) | 35.17*** | 24.33*** | 2.79 | 22.87*** | 3.58* | 0.08 | 4.94* | 3.08 | 0.73 |
| **Botrytis bunch rot** | | | | | | | | | |
| Incidence (%) | 4.41* | 25.84*** | 158.40*** | 1.95 | 1.25 | 0.08 | 0.45 | 0.21 | 3.13 |
| Severity (%) | 1.01 | 15.99*** | 24.69*** | 0.44 | 2.57 | 6.06** | 1.23 | 0.02 | 1.06 |

*, ** and *** indicate statistical significance at $p < 0.05$,<0.01 and<0.001 levels of confidence, respectively.

### 6.4.3. Fruit yield

Trellis systems and under-trellis ground cover significantly affected potential fruit yield in our study (full crop vines) (Table 2). All treatments were pruned to seven two-bud spurs per meter, however, even no significant differences in shoots per meter, clusters per shoot, and cluster weight were detected between both H treatments (VSP-H and Lira-H), vines trained on Lyra system ended the first and last seasons with a lower Yield/m (Table 3). UTCC also consistently reduced fruit yield compared to the conventional floor management (H). Differences observed between VSP-UTCC and VSP-H treatments were mostly explained by a reduction in cluster size (since shoots per meter and number of clusters per shoot were not affected by ground cover management). Berry weight in UTCC treatment was significantly lower than H treatment (Table 3). Differences in berry weight were already detected at the first sampling date (data not shown) and well correlated to PW/m in both trellis systems ($R^2$ >0.69). On the other hand, associated with its higher vine capacity (shaded area per hectare) plants trained on Lyra produced higher yield/ha than those on VSP. The three season's average yield (non-thinned vines) was 16.4, 15.0 and 19.3 Ton/ha for VSP-H, VSP-UTCC and Lyra-H treatments respectively. VSP-UTCC reduced in average 15% while Lyra-H increased 18% the yield achieved in VSP-H treatment.

**Table 3** Canopy characteristics as affected by under trellis ground cover and trellis system treatments.

| | VSP-H | VSP-UTCC | L-H |
|---|---|---|---|
| | Season 2011/2012 | | |
| Cluster weight (g) | 295 a | 268 b | 296 a |
| Berry weight (g) | 1.93 a | 1.64 b | 1.98 a |
| Pruning weight (kg/m) | 0.80 a | 0.50 b | 0.52 b |
| Shoots/m | 14.4 | 13.7 | 12.4 |
| Pruning weight/cane (g) | 55.4 a | 33.3 c | 42.2 b |
| PAR % | 2.64 c | 4.70 a | 3.16 b |
| Shaded area m2/m | 0.92 a | 0.69 c | 0.81 b |
| Pruning weight/ha (kg) | 3066 a | 1924 b | 3058 a |
| Shaded area/ha (m2) | 3523 b | 2659 c | 4749 a |
| | Season 2012/2013 | | |
| Cluster weight (g) | 341 ab | 300 b | 369 a |
| Berry weight (g) | 1.74 a | 1.58 b | 1.81 a |
| Pruning weight (kg/m) | 0.74 a | 0.40 b | 0.43 b |
| Shoots/m | 12.9 | 12.3 | 11.6 |
| Pruning weight/cane (g) | 57.5 a | 32.9 b | 37.7 b |
| PAR % | 1.89 c | 8.70 a | 5.93 b |
| Shaded area m2/m | 1.01 a | 0.80 b | 0.90 ab |
| Pruning weight/ha (Kg) | 2847 a | 1521 b | 2556 a |
| Shaded area/ha (m2) | 3880 b | 3042 c | 5447 a |
| | Season 2013/2014 | | |
| Cluster weight (g) | 267 a | 242 b | 250 ab |
| Berry weight (g) | 1.76 a | 1.70 b | 1.81 a |
| Pruning weight (kg/m) | 0.68 a | 0.43 b | 0.54 ab |
| Shoots/m | 9.6 | 9.6 | 11.1 |
| Pruning weight/cane (g) | 69.0 a | 44.4 c | 50.1 b |
| PAR % | 4.63 b | 9.05 a | 6.00 b |
| Shaded area m2/m | 0.88 a | 0.69 b | 0.82 a |
| Pruning weight/ha (kg) | 2613 b | 1642 c | 3226 a |
| Shaded area/ha (m2) | 3397 b | 2654 c | 4830 a |

[a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system. Values with different letters in single rows are significantly different at $p <$

6.4.4. Berry, must and wine composition

Vegetative growth by itself did not have a significant effect over fruit composition. The combination Block-Training system produced a significant variation in vegetative expression among plots (PW/m, cane pruning weight). However, even when yields of individual plots were adjusted to >1.8 m2 of leaf area/kg of fruit to avoid over-cropping effect, no significant correlation between vigor of individual plots (PW/m or individual shoots pruning weight) and any analyzed fruit composition parameter was detected. Trellis system had no consistent effect over any evaluated parameter of fruit or wine composition (Table 2). Fruit composition in VSP-H and Lyra-H treatments was comparable every season. On the other hand, UTCC significantly affected all fruit composition parameters evaluated except pH (Table 2). Compared to the conventional management, UTCC generally reduced most TA and YAN, and consistently increased Brix (thinned vines) (Table 4). Associated with a lower berry size, total anthocyanins accumulation was also significantly increased by UTCC treatment in two of the three harvests (2012 and 2014), Differences observed in anthocyanins accumulation in grapes were detected in wine just in 2012 harvest (Table 4). Crop load significantly affected fruit composition (Table 2). Yield negatively affected most sugar and grape anthocyanins accumulation. The effect of UTCC on fruit composition was not as consistent in full crop vines (Table 3).

**Table 4** Fruit yields, grape and wine composition as affected by under trellis ground cover and trellis system treatments.

| | Thinned cluster vines | | | Full crop vines | | |
|---|---|---|---|---|---|---|
| | VSP-H | VSP-UTCC | L-H | VSP-H | VSP-UTCC | L-H |
| **Harvesy 2012** | | | | | | |
| **Fruit yields** | | | | | | |
| Cluster/shoots | 0.81 b | 0.93 ab | 0.90 ab | 1.27 a | 1.34 a | 1.13 ab |
| Cluster/m | 11.8 c | 12.1 bc | 11.9 c | 18.0 a | 18.6 a | 13.7 b |
| Vine yield (kg/m) | 3.37 d | 3.25 d | 3.17 d | 5.48 a | 5.12 b | 4.05 c |
| Yield (Ton/Ha) | 13.0 d | 12.2 d | 19.1 c | 21.1 b | 19.7 c | 23.9 a |
| Ravaz index | 4.3 c | 7.3 b | 6.3 bc | 6.8 b | 10.7 a | 8.1 ab |
| **Grape composition** | | | | | | |
| Brix | 20.8 b | 22.0 a | 21.2 ab | 20.6 b | 20.4 b | 20.8 b |
| TA (g/L) | 4.39 a | 3.75 b | 4.63 a | 4.74 a | 3.78 b | 4.27 ab |
| pH | 3.39 a | 3.36 ab | 3.38 ab | 3.35 ab | 3.33 b | 3.38 ab |
| Anthocyanins (mg/L) | 1156 bc | 1446 a | 1261 b | 953 cd | 994 cd | 891 d |
| YAN | | | | | | |
| **Wine composition** | | | | | | |
| TA (g/L) | 4.00 | 3.45 | 4.16 | 4.35 | 3.93 | 4.30 |
| pH | 3.65 | 3.68 | 3.72 | 3.70 | 3.68 | 3.69 |
| Ethanol (% v/v) | 11.7 b | 12.6 a | 11.5 b | 11.1 b | 11.1 b | 11.1 b |
| Anthocyanins (mg/L) | 1119 b | 1317 a | 930 bc | 923 bc | 941 bc | 804 c |
| **Harevest 2013** | | | | | | |
| **Fruit yield** | | | | | | |
| Clusters/shoot | 0.92 | 0.81 | 0.76 | 0.92 | 0.91 | 0.78 |
| Clusters/m | 11.6 ab | 9.2 cd | 7.4 d | 11.9 a | 12.0 a | 9.6 bc |
| Vine yield (kg/m) | 4.08 a | 2.79 b | 2.75 b | 3.96 a | 3.32 ab | 3.23 ab |
| Yield (Ton/Ha) | 15.7 ab | 10.7 c | 16.2 ab | 15.2 ab | 13.4 bc | 19.0 a |
| Ravaz index | 6.0 ab | 8.1 a | 7.0 ab | 5.3 b | 8.4 a | 7.5 ab |
| **Grape composition** | | | | | | |
| Brix | 23.9 b | 25.0 a | 24.2 ab | 23.5 b | 23.8 ab | 23.1 b |
| TA (g/L) | 5.25 ab | 4.89 ab | 5.59 a | 5.09 ab | 4.66 b | 5.14 ab |
| pH | 3.52 | 3.49 | 3.49 | 3.48 | 3.49 | 3.45 |
| Anthocyanins (mg/L) | 1437 a | 1401 a | 1407 a | 1240 ab | 1291 ab | 1136 b |
| YAN | | | | | | |
| **Wine composition** | | | | | | |
| TA (g/L) | 4.03 c | 4.41 a | 3.96 c | 3.97 c | 4.28 b | 3.94 c |
| pH | 4.05 | 3.98 | 4.01 | 3.85 | 3.95 | 3.97 |
| Ethanol (% v/v) | 12.2 b | 13.5 a | 12.1 bc | 12.0 bc | 12.4 b | 11.7 c |
| Anthocyanins (mg/L) | 1308 | 1385 | 1219 | 1258 | 1283 | 1126 |
| **Harvest 2014** | | | | | | |
| **Fruit yield** | | | | | | |
| Cluster/shoots | 1.02 ab | 1.07 ab | 0.59 b | 1.40 a | 1.53 a | 0.95 ab |
| Cluster/m | 9.4 b | 10.6 b | 6.8 c | 12.8 a | 12.9 a | 10.4 b |
| Vine yield (kg/m) | 2.58 b | 2.56 b | 1.75 c | 3.33 a | 3.12 ab | 2.53 b |
| Yield (Ton/Ha) | 9.9 c | 9.9 c | 10.3 bc | 12.8 ab | 12.0 bc | 14.9 a |
| Ravaz index | 3.9 b | 5.9 ab | 3.2 b | 5.0 ab | 8.7 a | 5.0 ab |
| **Grape composition** | | | | | | |
| Brix | 21.2 b | 21.9 a | 21.1 b | 20.9 b | 21.1 b | 20.8 b |
| TA (g/L) | 6.14 | 5.42 | 6.22 | 5.79 | 5.86 | 6.49 |
| pH | 3.37 | 3.41 | 3.38 | 3.35 | 3.31 | 3.30 |
| Anthocyanins (mg/L) | 1559 b | 1812 a | 1649 ab | 1620 ab | 1704 ab | 1578 b |
| YAN | | | | | | |
| **Wine composition** | | | | | | |
| TA (g/L) | 4.40 | 4.33 | 4.83 | | | |
| pH | 3.80 | 3.78 | 3.77 | | | |
| Ethanol (% v/v) | 11.6 | 12.0 | 12.2 | | | |
| Anthocyanins (mg/L) | 1056 | 1151 | 1107 | | | |

[a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system. Values with different letters in single rows are significantly different at $p < 0.05$.

6.4.5. Botrytis bunch rot

In VSP-H and Lyra-H treatments Botrytis bunch rot incidence progressively increased when pruning weight per meter of canopy length increased. With equal PW/m, botrytis incidence was higher in Lyra compared to VSP (Figure 4): However due to the average PW/m was lower in Lyra no significant effect of trellis system on bunch rot incidence was detected (Tables 2 and 5). Bunch rot severity was also not consistently affected by trellis system (a significantly lower bunch rot incidence in Lyra-H vs VSP-H treatment was detected just at 2012 harvest) (Table 5). Contrarily, ground cover management consistently reduced Botrytis bunch rot occurrence (Tables 2 and 5). Bunch rot incidence was remarkably lower in VSP-UTCC compared to VSP-H (between 15% and 23% of those affected in VSP-H treatment) (Table 5). Contrarily with what was observed under the conventional H treatment, no correlation was detected between pruning weight per meter of canopy length and pruning weight in VSP-UTCC plots (Figure 4). Note how in Lyra-H and VSP-H treatments Botrytis incidence progressively increased with pruning weight, however VSP-UTCC incidence was never above 15% in any individual plot and year independently of vine vegetative growth (PW/m) (Figure 4). These results were consistent every season. Differences of Botrytis bunch rot incidence described at harvest were already significant in every season at least 15 days previous to harvest. Data for 2014 vintage is presented as an example, but disease development was comparable every season (Figure 5). Botrytis bunch rot severity was also consistently lower in VSP-UTCC treatment (Table 5). Crop load had no effect on Botrytis incidence or severity (Table 2).

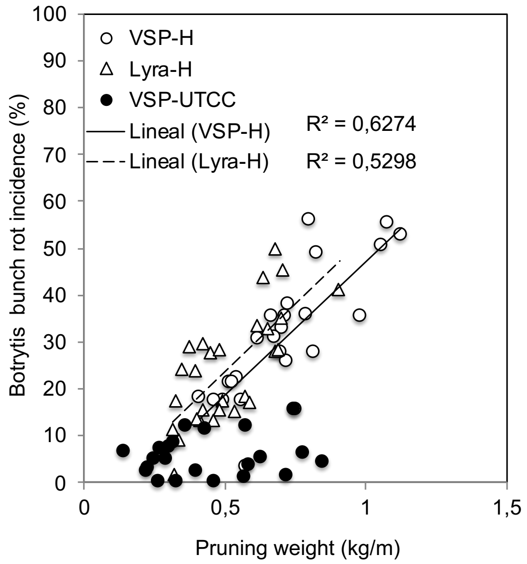


Fig 4. Relationship between pruning weight per meter of trellis and Botrytis bunch rot incidence of Tannat grapevines subjected to different groundcover management and trellis system treatments. UTCC: Under trellis cover crop; Lyra: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system

**Table 5** Botrytis bunch rot incidence and severity as affected by under trellis ground cover and planting density treatments.

| | | | |
|---|---|---|---|
| | VSP-H | VSP-UTCC | L-H |
| | **Harvest 2012** | | |
| Bunch rot incidence (%) | 32.3 a | 7.4 b | 30.1 a |
| Bunch rot severity (%) | 31.0 a | 7.0 c | 21.0 b |
| | **Harvest 2013** | | |
| Bunch rot incidence (%) | 28.1 a | 4.22 b | 20.9 a |
| Bunch rot severity (%) | 19.1 | 18.5 | 33.3 |
| | **Harvest 2014** | | |
| Bunch rot incidence (%) | 29.6 a | 6.4 b | 28.2 a |
| Bunch rot severity (%) | 27.0 a | 9.3 b | 31.1 a |

[a] UTCC: Under trellis cover crop; L: Lyra trellis system; H: Herbicide; VSP: Vertical shoot position trellis system. Values with different letters in single rows are significantly different at $p < 0.05$.

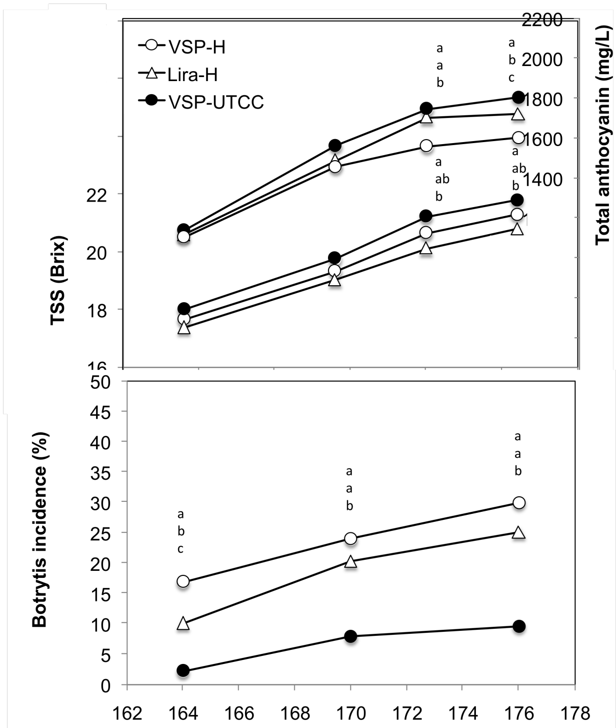


Figure 5. Evolution of total anthocyanin, soluble solids, and botrytis bunch rot incidence and severity of Tannat grapevines growing under different trellis systems and groundcover management treatments (season 2013-14).

6.4.6. Berry firmness

Texture analysis was not performed until last season but a clear effect of UTCC on berries was observed during post-veraison at every berry sampling date. A significant increase of berry firmness as deformation was confirmed at 2014 harvest. During the single compression test, berry firmness defined as mm of deformation under a force of 1 N was significantly affected by the under trellis ground cover treatment (VSP-H vs VSP-UTCC) while no differences were detected between trellis systems (Lyra-H vs VSP-H). Average deformation values were 3.67b, 3.75b and 4.03a for VSP-H, Lyra-H and VSP-UTCC respectively. Berry firmness measured in the 160 berries per treatment was not significantly correlated with its TSS or berry weight. No significant correlation between berry firmness and individual plots vegetative growth was detected.

## 6.5. DISCUSSION

VSP-H treatments PW/m values were always above the benchmark range of 0.30–0.60 kg/m (Smart and Robinson, 1991), which would indicate an excessively shaded canopy (Table 3). As previously reported, UTCC effectively reduces excessive vegetative growth of grapevines under the conventional ground cover management (H) (Coniberti et al., 2018; Giese et al., 2014; Karl et al., 2016). There is no universally accepted recommendation for leaf layer number of a canopy as it is affected by geography and cultivar, but the resulting reduction of canopy density (PW/m<0.50) was previously associated with well light exposed canopies and maximum accumulation of fruit TSS and anthocyanins in Tannat (Coniberti et al. 2012) (Table 3). However, the value observed in the VSP-UTCC treatment the last two seasons (0.40 PW/m) suggests an excessive depression of vegetative growth (as light intersection and fruit yield were significantly reduced) (Coniberti at al., 2018; Smart and Robinson, 1991) (Table 3). Although other factors, as cover crop's sequestration of mineral nutrients, especially N, have been reported affecting vegetative development (Hatch et al. 2011, Giese et al. 2014), the occurrence of water deficit during spring 2011 may explain the significant vegetative growth and yield declines observed in VSP-UTCC treatment (Table 3). First season shoot elongation rate of vines from individual plots was well correlated with post bloom period vine Ψstem ($R^2$= 0.88 and 0.65 for VSP and Lyra respectively) (Figure 1). Similar results were previously reported by Tesic et al. (2007), when associated to the early reduction of soil water content, UTCC decreased shoot growth from bud-break through bloom compared with inter-row cover or bare soil strip in semi-arid climates.

During the last two seasons, vine Ψstem did not fall below −0.6 MPa until veraison in any treatment, however vegetative growth parameters (cane pruning weight, PW/m) were comparable with first season in all treatments. Comparable findings were previously reported by Giese et al. (2014), when in a long-term study (6 seasons), consistent effects on vine vegetative growth were achieved after a below average rainfall season (300 mm), even when in following seasons vine Ψstem did not fall below −0.6 MPa on any measured date. Vegetative growth was more associated to previous seasons vine size (pruning weight). PW/m of vines with UTCC are likely to be increased in humid seasons (year), however vine capacity in a given season is not just limited by its environmental conditions but also by its previous history (Winkler et al. 1974). Our results suggest that UTCC should be considered as a strategy to consistently control vine size, even when no restriction of water availability during some seasons above average rainfall may be expected (as 2012/13 and 2013/14 seasons). Even though water availability should be the main factor affecting vegetative growth (since no blade or petiole N content differences were detected during our study), must YAN was every season significantly reduced by UTCC. The higher YAN content observed in H treatments (average values from the region) suggests that vines from UTCC should have a lower nitrogen uptake, which could be partially contributing to the vegetative growth differences observed.

Although vine PW/m over three seasons was consistent and did not show cumulative effects of UTCC (Table 3), this could be expected in the long term (Hatch et al. 2011). Competition from cover crop roots can decrease vine roots in shallow soils (Centinari et al. 2016) and alter grapevine-rooting patterns (Celette et al., 2008), which may impact the vine's uptake of water and nutrients. In our study, no effect of UTCC on nutrients status was detected on leaf samples. However, considering that nutrient uptake is also function of water availability (Tesic et al. 2007) and during pre-bloom period, the last two seasons Ψstem did not fall below −0.5 MPa in any treatment we are not able in this short-term study to predict vine root distribution and potential change in nutrients uptake in the long term. On the other hand, in a commercial setting, in case of an excessive reduction of vegetative growth or yield, water deficit could be avoided during post-bloom period through irrigation, when grapevine is most susceptible to decreased reproductive yield from water stress (Hardie and Considine 1976); and/or to return to the traditional floor management (H treatment) or cultivation. For sure this is an advantage of under vine management, versus other more permanent strategies (rootstock, divided canopies trellis systems or planting density).

A volume of literature has acknowledged that vine growth, development, yield, and fruit composition are extremely affected by the amount of leaf area that can be consistently exposed to the sun, and it should be a major consideration in the choice of a training system (Reynolds and Vanden Heuvel, 2009). As expected (Carboneau, 1978, Kliewer and Dokoozlian, 2005), the Lyra-H effectively reduced excessive vegetative growth observed on VSP-H. After three seasons vegetative expression of plants under Lyra-H were located in the optimal range (PW/m from 0.41 to 0.58 kg/m). In theory the overall effects of reducing excessively large vines through horizontal canopy division are typically higher yields (increased buds per vine and bud fruitfulness resulting from a reduction in canopy shade - Shaulis et al. 1966), and increased harvest juice TSS at a given crop size (Shaulis et al. 1966, Smart et al. 1985a, 1985b). In our study, associated with the increased number of buds and increased light intersection (shaded area), full crop vines trained in Lyra consistently produced higher yield per vine and hectare than those trained in VSP, however bud fertility (number of cluster/bud) was always lower in Lyra.

The arrangement and volume of the canopy impacts on light interception by both leaves and clusters, and on the microenvironment in which the fruit grows and matures. It is highly accepted that by increasing the exposed leaf area fruit composition improves (Reynolds and Vanden Heuvel, 2009). In fact, most pruning and training practices are based on these concepts. However in our study, the improvement achieved for most indicators of canopy microclimate in Lyra-H treatment was not consistently translated into an improvement on fruit composition even when crop loads were adjusted and comparable among treatments (Ravaz index varied from 4 to 7). Many previous reports indicate that, with the appropriate choice of training system, yield can be increased (generally through an increase in exposed leaf area) with simultaneous improvements in fruit composition (Carbonneau et al. 1978, Morris and Cawthon 1980, Kliewer and Dokoozlian 2005, Shaulis et al. 1966). Although the literature indicates an accepted relationship exists among training, vine microclimate, and fruit composition (Reynolds and Vanden Heuvel, 2009), there are many studies of training systems where yield components were increased but fruit composition was not affected (Shaulis and May 1971, May et al. 1973, Peterlunger et al. 2002). As demonstrated in the review on trellis systems by Reynolds and Vanden Heuvel (2009), both higher yield and improved fruit composition can be achieved with some training systems in some circumstances; however, the relationships existing among training, vine microclimate, and fruit composition are not yet demonstrated conclusively. Indicators such as cane pruning weight, PW/m, cluster zone PAR% and Ravaz index were widely used to define the effect of different treatments on vine balance (Dry et al. 2005), however those do not directly address which physiological processes were involved. In our study, through two different management practices (trellis system and ground cover), VSP-UTCC and Lyra-H produced comparable and optimum indicators of canopy microclimate, however compared to the excessively vigorous VSP-H treatment fruit composition was generally not affected by Lyra-H and consistently improved by VSP-UTCC. It has been demonstrated that associated to an enhancement on cluster light exposure, management practices such as leaf removal consistently increase Tannat grapes anthocyanins concentration in our region (Disegna et al. 2005b). However, in our study, fruit composition seems to be more intimately related to the physiological processes involved and probably water status, than to the improved leaves and clusters microclimate.

A positive correlation between grapevine growth and susceptibility to bunch rot has often been observed (Coniberti et al., 2018; Guilpart et al., 2017, Valdes-Gomez et al., 2008). The effect of grapevine vegetative growth on grey mould expression results from direct effects (increased size and number of shoots, leaves, and berries) and indirect ones (via microclimate), which are difficult to dissociate in field experiments (Valdes-Gomez et al., 2008). Concerning indirect effects in our study, differences detected in canopy density among treatments (PW/m and PAR%) have been clearly associated with changes in the microclimatic conditions (Evaporation rate). The main effects are associated with an increased wind penetration (Savage and Sall, 1984), solar radiation, and evaporative potential into the canopy (Coniberti et al, 2013) that occurs as the number of leaf layers within the canopy is reduced (English et al., 1990). The benefit of improved fungicide coverage on clusters of less vigorous vines should be also taken into consideration (Chellemi and Marois, 1992). In our experiment, bunch rot susceptibility of clusters/grapes to bunch rot of herbicide treatments progressively increased with PW/m ($R^2$ = 0.63 and 0.53 for VSP-H and Lyra-H respectively). However the novelty of our results is that bunch rot incidence of VSP-UTCC was consistently lower than the other treatments, even when vegetative development (shoot elongation rate, PW/m, PAR%) and fruit maturation (TSS, titratable acidity) compared to Lyra-H. The fact that no correlation was detected among individual plots Pw/m and bunch rot incidence under VSP-UTCC treatment (Figure 4), suggests that other factors other than vine vigor are playing a significant roll on the three-way interactions of bunch rot epidemic (host, environment and pathogen). Valdes-Gomez et al. (2008) investigated the relationship between grey mould expression, climate and microclimate, canopy development, and associated morphological and biochemical features of grape clusters and berries. Bunch mass has been shown to make the largest contribution to cluster compactness among various cluster measurements and can be considered as the key morphological feature afecting *B. cinerea* infection in grape clusters (Valdes-Gomez et al., 2008). The reduction in size and number of host organs such as grape berries result in a lower probability of contact with spores and mycelial colonization from one berry to the next (Elmer and Michailides, 2004). Bunch mass is affected by seasonal changes in vine growth (particularly shoot vigour) (Valdes-Gomez et al., 2008), however in our experimental conditions, with vines pruned to produce around 12 to 14 shoots per meter, cane-pruning weight in Lyra-H was significantly reduced with no major effect on bunch mass compared to VSP-H treatment. This may contribute to explain the comparable botrytis infection in both H treatments (VSP and Lyra). Associated with a reduction on berry weight, bunch mass was significantly reduced by UTCC treatment. Bunch rot incidence and severity were also significantly lower in VSP-UTCC treatment, even when its canopy density was comparable with Lyra-H treatment. This may underline the important role of cluster architecture in the lower *B. cinerea* infection. The strong difference observed in disease occurrence between UTCC and H treatment in our study could not be explained by just this factor. It has been reported that sites with typically low yeast available nitrogen (YAN) tend to have a lower incidence of Botrytis bunch rot; nevertheless, this may be related to an indirect response to low vine nitrogen levels and vigour rather than a decreased susceptibility of berries because of low YAN levels (Mundy and Beresford, 2007). In our study, UTCC significantly reduced juice N levels at harvest compared to H treatments with comparable canopy density, so grapevine nitrogen status could not be discarded as other direct factor affecting bunch rot susceptibility. Physical damage is known to be other key *Botrytis* bunch rot infection trigger (Hill et al. 1981). A positive correlation was also found between canopy aeration (often enhanced under service crop treatments) and berry skin strength, a source of grape resistance to disease infection (Jacometti et al., 2010). Texture analysis performed in our experiment also shows a clear effect of UTCC on berry firmness, (VSP-H vs VSP-UTCC) while no differences were detected between trellis systems

(Lyra-H vs VSP-H). Emerging evidence indicates that drought stress tolerance in grapevines involves the activation of polyamine oxidation, suggesting an improved immune response and reduced susceptibility to *Botrytis cinerea* (Hatmi et al., 2014). Seasonal variations in water status observed in our study could therefore also have a relevant impact on disease susceptibility at harvest via such a molecular basis, since UTCC significantly reduced water availability compared with both H treatments. On the other hand, during the first season (2011/2012), all treatments reached values below -0.9 MPa Ψstem which may suppose some level of water stress (Coniberti et al. 2018) (Figure 1) and bunch rot infection at harvest 2012 was comparable with the other seasons when no periods of stress occurred in H treatments. On the other hand, considering that in our study bunch rot incidence in H treatments was the main parameter defining harvest date, it is not possible to discard a direct link between seasonal grapevine water stress and disease susceptibility at harvest. Cover crops may also improve soil biological activity, leading to a faster decomposition of vine residues, which are habitats for *B. cinerea* primary inoculum (Garcia et al., 2018). Botrytis bunch rot is a complex disease, and many of the three-way interactions (host, environment and pathogen) are poorly understood. Our results don't allow identifying the specific mechanism by which UTCC induced a higher tolerance to botrytis bunch rot, however a clear effect on pathogen or host plant behavior was detected, and its seems to be more related to direct factors than indirect ones associated with canopy microclimate.

In our study, compared to VSP-H and Lyra-H a significant yield reduction with a significant but not always commercially relevant improvement of fruit or wine composition was observed in VSP-UTCC treatment (TSS increases from 0.5 to 1.3 Brix, fruit anthocyanin from 0 to 25%, and wine anthocyanin concentration from 0 to 41% (when fruit is picked at the same time - Table 3). However, in order to define the vineyard management in a humid climate, it is relevant to consider that in most seasons botrytis bunch rot is the main factor defining harvest time, causing in many cases to harvest fruit without achieving full maturation. Taking that into account, in most commercial wineries, 20% of bunch rot incidence is commonly used as maximum threshold to accept fruit from growers or define harvest. Fruit from H treatments in our study would be harvested in every season prior to full maturation. The evolution of TSS and anthocyanin accumulation in grapes and bunch rot incidence and severity during maturation in 2014 was compared (Figure 6). Data for 2014 vintage is presented as an example, but the analysis applies for the three seasons. Note that in a commercial context H treatment would be harvested at least two weeks before the experimental harvest date with a substantially lower TSS and anthocyanins concentration in grapes. Such large differences between treatments in fruit composition at harvest are not only statistically significant, but also certainly relevant in a commercial context.

## 6.6. CONCLUSIONS

Through the appropriate choice of training system, yield can be increased (through an increase in exposed leaf area) however reducing excessively large vines through horizontal canopy division may not consistently improve fruit composition even with adjusted crop loads (Ravaz index varied from 4 to 7). UTCC in combination with supplemental deficit irrigation is also an effective tool to regulate vine vegetative growth and canopy size in humid environments. The vegetative growth restriction achieved with UTCC is primarily due to vine water status during the post-bloom period. Subsequently in even distributed rainfall conditions, severe vine water deficit should be expected later in season. As a result: in any site where due to (1) climate conditions, (2) soil fertility and water holding capacity and (3) selected cultural practices (rootstock or trellis system), UTCC is considered as a potential tool to control excessive vegetative growth, deficit irrigation should be applied. Independently of the training system, average fruit yield/ha is well correlated with light intersection (shaded area/ha) and a reduction of potential yield should be expected when UTCC is used to regulate excessively high vegetative growth at least in varieties like Tannat where bud fertility is not remarkably affected by light exposure. TSS and anthocyanin accumulation in grapes and wines was significantly increased by UTCC treatment. The literature indicates an accepted relationship exists among training, vine microclimate, and fruit composition; in fact, most pruning and training practices are based on these concepts. However our results suggest that an improved fruit composition can be achieved in some circumstances associated to an improved leaf and cluster microclimate, but the physiological process involved in vegetative growth regulation could be even more relevant for fruit composition. Even though, canopy microclimatic factors, berry weight (reducing cluster compactness), cluster weight, and nitrogen grape content were reported affecting bunch rot occurrence, the strong reduction of bunch rot incidence observed associated to the ground cover management in this study could not be explained by just vine vigor and associated parameters. Our results didn't allow identifying the specific mechanism by which UTCC induced a higher tolerance to botrytis bunch rot, however a clear effect on pathogen or host plant behavior was detected. Considering that achieving a consistent wine quality is the first limitation of viticulture industry in humid environments, the opportunity to delay harvest to accomplish full maturation due to lower bunch rot incidence observed in UTCC plots, should be considered extremely relevant under these climatic conditions.

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# 7. CONCLUSIONES GLOBALES

Para las condiciones edafoclimaticas del sur de Uruguay, el uso de pastura permanente bajo la fila es una herramienta eficaz para restringir el excesivo crecimiento vegetativo de la vid observado con el manejo tradicional con herbicidas. El potencial de control del vigor (tasa de elongación de brotes, peso de poda de sarmientos) de la pastura permanente es superior al de otras prácticas de manejo estudiadas (sistema de conducción o la densidad de plantación). La restricción de la disponibilidad hídrica en el periodo floración ± 20 días se identificó como el factor determinante del crecimiento vegetativo. En las condiciones de estudio no se detectó una respuesta consistente a la aplicación de nitrógeno en plantas con buena disponibilidad hídrica durante el periodo crítico (-0,6 MPa MDSWP), sin embargo asociado a la interacción agua-nitrógeno, la respuesta al nitrógeno fue significativa en parcelas donde ésta se produjo.

La reducción del crecimiento vegetativo no implica pérdidas significativas de rendimiento potencial de la variedad Tannat hasta los 0,6 kg/m de peso de poda. Por el contrario, frente a mayores restricciones seria de esperar pérdidas significativas. Dado que los principales componentes del rendimiento de la vid (número de racimos por planta y número de bayas por racimo) están determinados durante la floración anterior, estas pérdidas de potencial productivo serán de significancia a partir del segundo año de establecida la competencia.

La interacción entre el cultivo de cobertura y la vid es compleja y dinámica y en consecuencia controlar el crecimiento de la vid mediante el establecimiento de competencia sin afectar el rendimiento potencial del viñedo, se trata de un desafío importante. Nuestros resultados muestran que en las condiciones de estudio es posible mediante el aporte complementario de agua y nitrógeno, regular el crecimiento vegetativo de la vid bajo cultivo de cobertura (manteniéndolo próximo a los 0,6 kg/m), sin afectar significativamente el potencial de captación de luz y rendimiento de fruta alcanzado en plantas bajo el manejo tradicional. El aporte de agua de riego requerido para evitar restricciones hídricas durante el periodo crítico (floración ± 20 días; > -0,6 MPa MDSWP) fue el equivalente al consumo de agua del viñedo de entre 4 a 10 días (20 a 50 mm; temporadas de precipitación próximas a la media; suelos de 90 y 110 mm de agua disponible).

Es importante destacar que el factor determinante de la restricción del crecimiento vegetativo en tratamientos con pastura bajo las condiciones de estudio es la disponibilidad hídrica en un periodo donde la demanda ambientan no es máxima (noviembre). Por lo tanto, en cualquier situación productiva donde el régimen de precipitación, fertilidad, agua disponible del suelo y otras prácticas culturales (porta-injerto y sistema de conducción) hagan viable la utilización de coberturas vegetales como herramienta para regular el vigor del viñedo, la utilización de riego será necesaria para evitar pérdidas de rendimiento y/o calidad asociados al estrés severo mas avanzada la temporada (< -1.0 MPa MDSWP).

La relevancia que tiene el microclima de la canopia sobre la calidad de la uva ha sido ampliamente reportada, siendo muchas de las prácticas de manejo de los viñedos orientada a establecer un correcto "equilibrio". En nuestro estudio las diferentes combinaciones de tratamientos, bloques y sitios experimentales se tradujeron en plantas con un amplio rango de condiciones de vigor (pero de poda/m, pero de sarmiento), sin embargo no fueron detectadas correlaciones significativas entre la expresión vegetativa de las plantas (peso de sarmiento, peso de poda) o microclima de la canopia (Índice foliar, PAR%) y los parámetros de composición de la uva. La mejora de la calidad de uvas y vinos observadas en los tratamientos con pastura, estuvo asociada fundamentalmente a la restricción hídrica durante el periodo envero-cosecha y no a un mejor equilibrio vegetativo, por lo que en temporadas con precipitaciones por encima de la media durante este período, no se obtuvieron mejoras significativas respecto al manejo convencional. Nuestros resultados indicarían que si bien la composición de la fruta podría estar afectada por el vigor de brotes y microclima de la canopia, el mecanismo por el cual se alcanza este equilibro vegetativo es mucho más determinante en la composición de la uva. Ello explicarían el porque no se han identificado diferencias significativas en la composición de uva provenientes de plantas excesivamente vigorosas en sistemas de conducción de canopia simple (Espaldera) vs. canopia dividida (Lira) en nuestras condiciones productivas.

La competencia establecida por la cubierta vegetal en la fila permite incrementar el potencial de control de agua del viñedo durante la maduración, incrementando también así su potencial de calidad comparado al manejo convencional con herbicida. La acumulación de azúcar y concentración de antocianinas en bayas fue

significativamente superior en el tratamiento con completa cobertura vegetal + riego deficitario que en tratamientos con herbicida. Los vinos elaborados presentaron una mayor intensidad aromática y aromas frutados que el tratamiento convencional con herbicida, siendo en general preferido por los expertos en el test de preferencia.
Por otro lado, la susceptibilidad de plantas de Tannat a las podredumbres de racimo se vio fuertemente asociada al desarrollo vegetativo de las plantas. El riesgo de ocurrencia de la enfermedad se incrementó progresivamente a medida que el peso de poda aumentó por sobre los 0,5 kg/m. Esta respuesta fue muy evidente sobre en el tratamiento convencional (con herbicida). Sin embargo, lo sorprendente de nuestros resultados es la reducida incidencia de Botrytis observada en los tratamientos con pasturas, independientemente de la condición de vigor. El efecto de la densidad de la canopia de vid sobre la expresión de podredumbres de racimo, es el resultado de efectos directos e indirectos (a través del microclima), los cuales son difíciles de disociar en experimentos de campo. Nuestros resultados no permiten identificar el mecanismo por el cual la pastura induce una mayor tolerancia a las podredumbres de racimo, sin embargo se identifica una clara modificación de la interacción húesped-patógeno. Ello permite al menos relativizar la relevancia atribuida a los efectos indirectos (modificación del microclima), frente a los directos en la bibliografía.
Factores como una mayor firmeza (probablemente desencadenado en respuesta al déficit hídrico) y/o la reducción del nitrógeno en bayas (YAN), se plantean como posibles responsables de la respuesta observada. Futuros estudios deberían estar dirigidos a comprender mejor tales relaciones, a fin de desarrollar estrategias de gestión y MIP que conduzcan a un uso mas reducido de plaguicidas. Como ya fue mencionado, el manejo bajo pastura + riego deficitario produjo una mejora significativa de la calidad de la uva respecto al manejo con herbicida (tratamientos cosechados la misma fecha), sin embargo la posibilidad de dilatar la cosecha en plantas con una significativamente menor incidencia de Botrytis, se considera de extrema relevancia para la viticultura de clima húmedo. Por otra parte, la fertilización nitrogenada incrementó la incidencia de Botrytis en todos los tratamientos ensayados, incluso en casos donde no se detectó respuesta en términos de crecimiento vegetativo (peso de poda) o concentración de nitrógeno en hojas o bayas.

Finalmente, la necesidad de utilización de riego en viticultura para vino en Uruguay es un tema controversial, no obstante nuestros resultados indican que sería una herramienta fundamental para el ajuste del manejo del suelo de nuestros viñedos. La producción tradicional en secano requiere un manejo del suelo orientado a preservar el agua, asegurando la disponibilidad hídrica incluso en temporadas secas. Es relevante considerar que además de la retracción del crecimiento vegetativo y potencial productivo asociado al déficit hídrico (1) la disponibilidad de nitrógeno se encuentra estrechamente ligada a la oferta hídrica y (2) diferencias no fácilmente detectables de los niveles de nitrógeno en planta pueden limitar tanto el rendimiento potencial del viñedo como su susceptibilidad a podredumbres de racimo. En un régimen de precipitaciones variable como el de Uruguay, esto trae importantes implicancias prácticas, dado que la oportunidad de ajustar factores determinantes del rendimiento y calidad (agua y nitrógeno) es extremadamente reducida. Asegurar entonces el potencial productivo de la vid en secano, implica la sobre-fertilización nitrogenada, y por lo tanto la obtención de calidad en forma sostenida estará limitada a las temporadas con pluviometría inferior a la media y supeditada a un fuerte manejo en verde.

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