Using diluted winery effluent for irrigation of *Vitis vinifera* L. *cv.* Cabernet Sauvignon and the impact thereof on soil properties with special reference to selected grapevine responses

by

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DECLARATION

By submitting this dissertation electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the authorship owner thereof (unless to the extent explicitly otherwise stated) and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

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SUMMARY

Wine production is an important industry in the Western and the Northern Cape regions of South Africa. Wineries produce large volumes of poor quality wastewater, particularly during harvest. International requirements, as well as national legislation, are putting pressure on wine producers regarding the responsible management of winery wastewater, which may have a large-scale detrimental impact on the environment. Currently, the Department of Water and Sanitation is drafting new legislation aimed at wineries to allow beneficial crop irrigation as a General Authorisation. In this regard, a multidisciplinary research project to investigate the impact of diluted winery wastewater on soils, crop growth and product quality was initiated and funded by the Water Research Commission of South Africa. The project was also co-funded by Winetech and the Agricultural Research Council. The possible re-use of winery wastewater for vineyard irrigation was investigated in a Cabernet Sauvignon vineyard in a sandy soil near Rawsonville in the Breede River Valley. Wastewater obtained from a co-operative winery was diluted to levels of 100, 250, 500, 1000, 1500, 2000, 2500 and 3000 mg/L chemical oxygen demand (COD), using water obtained from the Holsloot River. The dilution was carried out individually for each concentration in 15 m^3 tanks at the vineyard. Control grapevines were irrigated with river water. In addition to the field trial, a pot trial was also included to determine the effect of diluted winery wastewater on near-saturation hydraulic conductivity (K) of four different soils.

In general, soil potassium and sodium increased with an increase in COD level of the diluted winery wastewater, *i.e.* a decrease in dilution of the wastewater. Although irrigation using diluted winery wastewater had almost no other effects, element accumulation particularly with respect to potassium and sodium, might be more prominent in soils with higher clay contents or in regions with low winter rainfall. After three years, near-saturation hydraulic conductivity of shale-derived soil, alluvial and aeolian sands decreased with a decrease in the level of wastewater dilution. This indicated that severe degradation in hydraulic properties can occur if diluted winery wastewater is used for irrigation, and might even be aggravated if undiluted winery wastewater is used.

Irrigation of grapevines using diluted winery wastewater did not affect grapevine water status, vegetative growth, production or evapotranspiration, irrespective of the level of dilution. Results showed that irrigation of grapevines using diluted winery wastewater did not have detrimental effects on juice characteristics with regard to ripeness parameters and ion content. Wine sensorial characteristics were not affected by irrigation using diluted winery wastewater. The grapevines did not respond to level of COD *per se*. This indicated that sufficient aeration occurred between irrigations which allowed organic carbon breakdown. Although salinity and sodicity levels in the diluted winery wastewater were below the

thresholds where growth and yield reductions are expected for grapevines, it should be monitored frequently. The low salinity and sodicity levels in the diluted winery wastewater could be a further explanation why the grapevines did not respond negatively to the wastewater irrigation.

Based on the above-mentioned results, the following criteria should be considered for possible amendments to the General Authorisations for wineries when using diluted wastewater for vineyard irrigation: (i) COD must be diluted to 3000 mg/L or less, preferably to less than 2000 mg/L to avoid unpleasant odours in the vineyard during irrigations, (ii) electrical conductivity (EC_{iw}) must be less than 0.75 dS/m, (iii) sodium adsorption ratio (SAR_{iw}) must be less than 5, (iv) the soil must have a low cation exchange capacity, (v) unrestricted internal drainage in the root zone, (vi) irrigation water must not percolate beyond the root depth, (vii) only micro-sprinklers should be used, (viii) irrigation must be applied in such a way that bunches are not wetted, (ix) at least 50% plant available water depletion should be allowed between irrigations to allow sufficient aeration for oxidation of organic material applied *via* the irrigation water and (x) irrigation frequency and volumes must be such that wine quality is not reduced.

OPSOMMING

Die produksie van wyn is ŉ belangrike industrie in die Wes- en Noordkaap streke van Suid-Afrika. Kelders produseer groot volumes swak gehalte afvalwater, veral tydens oes. Aangesien kelderafvalwater ŉ grootskaalse nadelige impak op die omgewing kan hê, word daar hoë internasionale vereistes, sowel as druk deur nasionale wetgewing op wynprodusente geplaas om kelderafvalwater verantwoordelike te bestuur. Die Departement van Water en Sanitasie is tans besig om nuwe wetgewing te formuleer wat daarop gemik is om kelders deur middel van ŉ Algemene Magtiging toe te laat om voordelige gewasse te besproei met afvalwater. ŉ Multidissiplinêre navorsingsprojek om die impak van verdunde kelderafvalwater op gronde, gewas groei en produk gehalte te ondersoek is deur die Water Navorsing Kommissie van Suid-Afrika geïnisieer en befonds. Die projek is ook gedeeltelik deur Winetech en die Landbou Navorsingsraad befonds. Die moontlike hergebruik van kelderafvalwater vir wingerdbesproeiing is in ŉ Cabernet Sauvignon wingerd in ŉ sandgrond naby Rawsonville in die Breede Rivier Vallei ondersoek. Kelderafvalwater van ŉ koöperatiewe wynkelder was met onbehandelde rivierwater van die Holsloot Rivier verdun tot konsentrasies van 100, 250, 500, 1000, 1500, 2000, 2500 en 3000 mg/L chemiese suurstof aanvraag (chemical oxygen demand in Engels of COD in kort). Die verdunnings is afsonderlik vir elk van die konsentrasies in 15 m^3 tenke by die wingerd gedoen. Kontrole wingerdstokke is met die rivierwater besproei. Benewens die veldproef is ŉ potproef ook ingesluit om die effek van verdunde kelderafvalwater op naby versadigde hidrouliese geleiding (K) van vier verskillende tekstuur gronde te bepaal.

Oor die algemeen het die kalium- en natriuminhoud van die grond toegeneem met ŉ toename in COD van die verdunde afvalwater, *m.a.w.* ŉ afname in die verdunning van die afvalwater. Alhoewel besproeiing met verdunde kelderafvalwater byna geen ander effekte gehad het nie, mag element aansameling, veral met betrekking tot kalium en natrium, dalk meer prominent wees in gronde met hoër klei inhoude of in gebiede waar die winter reënval laag is. Na drie jaar het K van die skaliegrond, asook alluviale en eoliese sande afgeneem met ŉ afname in die vlak van afvalwaterverdunning. Dit het daarop gedui dat K drasties kan afneem indien verdunde kelderafvalwater vir besproeiing gebruik sou word, en dat hidroliese eienskappe drasties kan benadeel indien onverdunde kelderafvalwater gebruik word.

Besproeiing van wingerdstokke met verdunde kelderafvalwater het nie die wingerd se waterstatus, vegetatiewe groei, produksie of evapotranspirasie beïnvloed nie, ongeag die vlak van verdunning. Besproeiing met verdunde kelderafvalwater het nie suikertoename, totale titreerbare suur en elemente in sap benadeel nie. Derhalwe is die element samestelling en sensoriese gehalte van wyn ook nie nadelig beïnvloed nie. Die wingerdstokke het nie gereageer op COD vlak *per se* nie. Dit het aangedui dat daar

voldoende deurlugting tussen besproeiing toedienings was wat die afbreek van organiese koolstof toegelaat het. Alhoewel die brak- en natriumvlakke in die verdunde kelderafvalwater laer as die drumpelwaardes vir die groei en produksie afnames van wingerdstokke was, moet dit dikwels gemonitor word. Die lae brak- en natriumvlakke in die verdunde kelderafvalwater kan ʼn verdere rede wees vir die gebrek aan wingerdstokke se reaksie op die besproeiing met afvalwater.

Op grond van die voorafgaande resultate, behoort die volgende kriteria in aanmerking geneem te word by moontlike aanpassings vir die Algemene Magtigings vir kelders wanneer verdunde afvalwater vir wingerdbesproeiing gebruik word: (i) COD moet tot 3000 mg/L of minder verdun word, verkieslik minder as 2000 mg/L om onaangename ruike in die wingerd tydens besproeiing te vermy, (ii) elektriese geleiding (EC_{iw}) moet minder as 0.75 dS/m wees, (iii) natrium adsorpsie verhouding (NAV_{iw}) moet minder as 5 wees, (iv) die grond moet ʼn lae katioonuitruilkapasiteit hê, (v) interne dreinering in die wortelsone moet vrylik kan plaasvind, (vi) besproeiingswater moet nie verby die worteldiepte loog nie, (vii) slegs mikrospuite moet gebruik word, (viii) besproeiing moet op so ʼn manier toegedien word dat die trosse nie benat word nie, (ix) ‘n minimum van 50% plant beskikbare water onttrekking moet gehandhaaf word sodat voldoende deurlugting verkry word sodat oksidasie van organiese materiaal wat deur die besproeiingswater toegedien, kan plaasvind en (x) besproeiingfrekwensies en -volumes moet sodanig wees dat wyngehalte nie benadeel word nie.

This dissertation is dedicated to Philip Myburgh and Vink Lategan.

BIOGRAPHICAL SKETCH

Carolyn Louise Howell was born on 8 October 1974 in Port Nolloth. She started her school career at De Villiers Graaff Primary in Villiersdorp, and matriculated from Rhenish Girls' High School, Stellenbosch in 1992. In 1993, she enrolled at Stellenbosch University and obtained a B.Sc. Agric. degree in 1996, majoring in Agricultural Economics and Horticulture. In 1998, she started working at ARC Infruitec-Nietvoorbij (Agricultural Research Council) as a Research Technician. In 2004 she obtained her M.Sc. Agric. degree (Soil Science) from the Stellenbosch University. In 2006 and 2007, she worked as a Senior Research Officer (Viticulture) for South Australian Research and Development Institute in Adelaide, Australia. She obtained her Hons. B.Sc. Agric. (Viticulture) from Stellenbosch University in 2008. She has been a Researcher in the Soil and Water Science division at ARC Infruitec-Nietvoorbij since April 2010.

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1.1. BACKGROUND

Wine production is an important industry in the Western Cape and the Lower Orange River region in the Northern Cape. Wineries produce large volumes of low quality wastewater, particularly during harvest. In South Africa, historically most of the winery wastewater has been disposed of to land through irrigation of pastures (J. Rossouw, personal communication, 2014). With regard to the National Water Act (NWA) (1956), if a person was unable to comply with the conditions of Section 21(1), the Minister could exempt such a person from compliance with either one, or both the requirements of Section 21(1)(a) and 21(1)(b). South African wineries found compliance with the requirements of Sections 21(1)(a) and 21(1)(b) of the NWA (1956) technically and financially challenging. Therefore, most wineries applied for exemption from compliance with these requirements as they are based in the rural areas, where land is available for irrigation and the resulting grazing could be used (J. Rossouw, personal communication, 2014). The use of untreated winery wastewater was also considered suitable for irrigation of kikuyu grass for grazing purposes, as it consists mainly of biodegradable organic pollutants. The soil, and more particularly, bacteria present in the soil were found to be able to break down organic components in the wastewater under aerobic conditions. However, there are certain secondary environmental pollution risks associated with such irrigation, namely the potential occurrence of unpleasant odours and seepage due to over-irrigation which can result in saturated, anaerobic soils. Subsequently, many wineries were exempt from the requirements of Section 21(1), and there were certain conditions which limited the volume of wastewater that could be irrigated (J. Rossouw, personal communication, 2014). However, many of the exemptions did not specify a minimum area that had to be irrigated, nor a requirement for water quality analysis. In addition to this, quite often there was no expiry date for the exemption. Consequently, over-irrigation and soil saturation occurred which led to an accumulation of organic material, as well as inorganic salts such as phosphorus (P), sodium (Na^+) and potassium (K^+) in the soil (Mulidzi *et al*., 2009b). This resulted in soil degradation and possible seepage of wastewater into underground resources at certain irrigation areas.

In 1997, the Department of Water Affairs and Forestry published a Water Policy White Paper which repealed the NWA of 1956, and a new NWA (Act 36) was promulgated in 1998. This Act, in Section 22, did not make provision for exemption from compliance with the Act. The Act did define the irrigation of land with waste or water containing waste as a specific type of water use, in terms of section 37(1)(a). The controlled activity could be deemed a permissible water use activity. In 2004, the Department of Water Affairs and Forestry

(DWAF), recognising that the lack of conditions requiring at least the monitoring of the quality and quantity of wastewater being irrigated did not allow for the responsible management of the irrigation practice, incorporated these conditions into all new authorisations under the NWA (J. Rossouw, personal communication, 2014). These General Authorisations require not only the monitoring of wastewater quantity and quality, but also specify limits for wastewater being irrigated based on the volume irrigated per day. There is no restriction in the General Authorisations on the area being irrigated using wastewater.

In 1999, Winetech initiated research projects into improving the understanding of the composition and handling of winery wastewater (J. Rossouw, personal communication, 2014). In 2005, this research culminated in guidelines for the management of winery wastewater that not only focussed on assisting wineries in developing wastewater management plans, but also advised wineries as to how they could improve the quality of the winery wastewater by employing cleaner production principles. Winetech also started to engage more pro-actively with DWAF and through their co-operation secured their input into the guideline development process to ensure that the industry was fully aware of the legal requirements of the NWA. They also actively engaged in securing compliance with these conditions. In 2004, there was a series of meetings in June and August between various stakeholders to discuss a proposed General Authorisation for the wine industry. The DWAF indicated the need of the wine industry to move away from disposal irrigation of untreated wastewater.

In a meeting in August 2004, a treatment and disposal methodology was agreed upon on which the proposed General Authorisations could be based. Furthermore, treated winery wastewater, in combination with other water should be used for beneficial irrigation of agricultural crops such as vineyards. If winery wastewater could be used in a sustainable way, it would have the following benefits:

(i) Reducing the energy presently required for wastewater treatment, *e.g.* using pumps to aerate the water in ponds.

(ii) The presence of plant nutrients in the wastewater, *e.g.* nitrogen (N), P, and K^+, could also reduce cost of fertilization.

(iii) Where irrigation water is limited, the re-use of wastewater will have a positive impact on grape yields if additional irrigation could be applied.

(iv) If possible, the water saving and higher yields will contribute to the sustainability and economic viability of wine production.

Considering the foregoing, winery wastewater should be treated to specific quality standards, whereafter it could be stored in irrigation dams, and used for irrigation of crops.

Until now, the impact of this practice has, however, not been studied comprehensively. Adding winery wastewater to the current irrigation water (dilution) may well become a necessity in the future with current water shortages becoming more and more an alarming reality. Thus, knowledge on the impact of irrigating using winery wastewater on the chemical composition and physical structure of the soil, grapevine performance and wine quality is indispensable. It is envisaged that irrigating vineyards using irrigation water with added winery wastewater could alter the wine characteristics and overall quality, compared to vineyards irrigated using river water. In this regard, a solicited research project (Project K5/1881) to investigate the impact of wastewater irrigation by wineries on soils, crop growth and product quality was initiated and funded by the Water Research Commission (WRC) of South Africa. The project was co-funded by Winetech, THRIP and the Agricultural Research Council (ARC). During a workshop held on 15 May 2008 in Stellenbosch, the terms of reference for the project, including the range of chemical oxygen demand (COD) levels to which the winery wastewater had to be diluted for the different treatments, were finalized. It should be noted that COD was the preferred indicator of winery wastewater quality of the Steering Committee for the project.

1.2. PROJECT OBJECTIVES

The specific objectives of the project were:

- To standardise/optimise the dilution procedure to achieve large volumes of winery wastewater with different COD levels.
- To determine the effect of winery wastewater diluted to different COD levels with river water on soil characteristics with reference to amongst others: Physical, biological and chemical properties, whilst considering electrical conductivity (EC_e), pH and sodium adsorption ratio (SAR), as well as N, P and K^+ concentrations.
- To determine the effect of winery wastewater diluted to different COD levels with river water on the response of vineyards, cover crops and weeds to irrigation with winery wastewater with reference to amongst others: Vegetative growth, reproductive growth, grape composition, juice characteristics and wine quality.

With regard to this dissertation, the impact of diluted winery wastewater irrigation on soil, selected grapevine responses and wine parameters will be presented.

1.3. KNOWLEDGE REVIEW

1.3.1. Introduction

In South Africa, grapes are an important crop in regions such as the Western Cape and the Lower Orange River in the Northern Cape. The wine industry makes a significant contribution to the economy in these regions. In 2012 there were 3440 primary wine grape growers (South African Wine Industry Statistics, 2013). Furthermore, the wine industry provides a large number of employment opportunities, particularly in the rural areas. In 2012, the vineyards planted for wine production in South Africa amounted to 100 093 hectares, of which *c.* 92% is considered as producing, *i.e.* four years and older (South African Wine Industry Statistics, 2013). The number of wineries which crush grapes almost doubled from 1991 to 2002 (Table 1.1). Since 2005, the number of wineries appeared to be more or less stable. During this period, the industry produced around one billion litres of grape related products annually (Table 1.2).

Table 1.1. Growth trends in the South African wine industry (South African Wine Industry Statistics, 2013).

Role player	**Number**							
	1991	**2002**	**2005**	**2008**	**2009**	**2010**	**2011**	**2012**
Wine cellars which crush grapes	212	427	581	504	527	593	505	509
Co-operatives	70	66	65	58	57	54	52	50
Wine producing wholesalers	6	11	21	23	23	26	25	23

Table 1.2. Wine production trends in the South African wine industry (South African Wine Industry Statistics, 2013).

Product	**Production (million litres)**							
	2005	**2006**	**2007**	**2008**	**2009**	**2010**	**2011**	**2012**
Wine	628.5	709.7	730.4	780.2	739.0	779.8	831.2	870.9
Rebate	82.9	82.1	101.5	88.0	75.8	39.6	34.2	62.3
Juice	64.6	73.2	65.2	66.9	55.0	52.1	40.2	40.1
Distilling wine	129.2	147.9	146.4	157.9	152.2	113.3	107.2	121.8
Total	905.2	1012.9	1043.5	1093.0	1022.0	984.8	1012.8	1095.1

Using raw water is an integral part of the wine production processes. However, these processes generate wastewater of low quality that cannot be disposed of in natural systems. Winery wastewater can cause salinization and eutrophication of water resources, *i.e.* natural streams, rivers, dams, groundwater and wetlands (Van Schoor, 2005 and references therein; Laurenson *et al.*, 2012). Furthermore, wastewaters can cause soil sodicity, salinity, contamination with a wide range of chemicals, waterlogging and anaerobiosis, as well as loss of soil structure and increased susceptibility to erosion. Where solid wastes are present, offensive odours may be generated and seepage may result in the contamination of soil and water resources that can inhibit vegetative performance (Van Schoor, 2005 and references therein).

1.3.2. Volume of water involved in the winemaking industry

1.3.2.1. Water used for winemaking

Information on the actual amounts of water used by wineries is limited and appears to be inconsistent. A survey carried out in South Africa, which included wineries that crush up to 22 000 tonnes of grapes annually, showed that the volume of raw water increased significantly with the amount of grapes crushed (Sheridan *et al*., 2005). Although the variability between wineries was high, the slope of the relationship indicated that approximately 2 m^3 of water was required to crush one tonne of grapes. The Lutzville Vineyards' winery uses a measured average 100 000 m^3 of water to produce between 30 million and 40 million litres of wine annually (Kriel, 2008). Since this particular winery crushes approximately 47 500 tonnes per year (G. Theron, personal communication), about 2.1 m^3 of raw water is required to process one tonne of grapes. Although the amount of grapes crushed is substantially higher, the amount of water used by Lutzville Vineyards' winery agrees with the results of the survey carried out by Sheridan *et al*. (2005). According to Mosse *et al.* (2011), wineries in Australia generally require 3 m^3 to 5 m^3 water to crush a tonne of grapes. The average annual grape production in South Africa was 1.33 million tonnes from 2010 until 2012 (SAWIS, 2013). If it is assumed that winemaking in South Africa requires approximately 2 m^3 of water to process one tonne of grapes, it can be roughly estimated that the wine industry is currently using 2.66 million litres of raw water annually.

1.3.2.2. Volume of wastewater generated during winemaking

Reports on the actual volumes of wastewater that are generated by wineries are also extremely limited. It is estimated that medium to large wineries generate more than 15 000 m^3 of wastewater annually, whereas small wineries generate less than 15 000 m^3 annually (Van Schoor, 2005 and references therein). Australian wineries generate about 5 m^3 of wastewater per tonne of grapes crushed (Chapman *et al.*, 1995). Crushing approximately 50 000 tonnes of grapes annually generate about 175 000 m^3 of wastewater at the Berri estates' winery in the Riverland region of South Australia (Anonymous, 2010). Hence, their wastewater generation amount to *c.* 3.5 m^3 per tonne of grapes. Usually most of the raw water entering wineries ends up as wastewater. In contrast, it is estimated that 50%, *i.e.* 50 000 m^3, of the raw water used by the Lutzville Vineyards' winery ends up as wastewater (Kriel, 2008). The other half of the water is presumably lost to evaporation under the warm windy atmospheric conditions. This means that this particular winery generates about 1.1 m^3 of wastewater per tonne of grapes crushed. In comparison, substantially lower volumes, *i.e.* 0.359 m^3 and 0.357 m^3 wastewater per tonne of grapes crushed was generated for off-skin white wine making, and rosé and thermo-vinification of red wines, respectively, in French cellars (Bories & Sire, 2010). An even lower value of 0.262 m^3 of wastewater generated per tonne of grapes crushed, was reported for on-skin vinification of red wines (Bories & Sire, 2010).

1.3.3. Origin of winery wastewater and associated pollutants

1.3.3.1. Sources of pollutants

Wineries vary in size, operational procedures and management practices. They undertake similar, yet highly site-specific processes. The variations result in the production of different qualities and quantities of wastewater (Van Schoor, 2005). According to Bories and Sire (2010), wine making methods can have an impact on the quality of the wastewater generated. In off-skin wine making, wastewaters are produced which contain mainly sugars. In comparison, in cellars where classical red wine making methods are followed, wastewaters are generated which have high ethanol levels. In South Africa, the typical wine production process can be divided into various stages (Table 1.3). Medium to large wineries with year-round operations generate approximately 50% of their wastewater during the vintage period, whereas small wineries may generate up to 80% of their wastewater during harvest (Van Schoor, 2005 and references therein). The major form of wastewater from wineries is water that is used for cleaning processes (Van Schoor, 2005). The primary winemaking processes related to winery wastewater generation and their associated contribution to wastewater quantity and quality, as well as possible effects on legal wastewater quality parameters are summarized in Table 1.4. The primary water quality parameters are COD, EC, SAR and pH.

Table 1.3. Typical stages of winery activities and their role in wastewater generation (after Van Schoor, 2005 and references therein).

Stage	Activities	Duration (weeks)
1. Pre-harvest	Bottling takes place and tanks are washed out with sodium or potassium hydroxide. Other equipment is also washed to prepare for the harvest period.	1 to 4
2. Early harvest	Wastewater generation increases drastically during this period and reaches 40% of the maximum weekly rate measured at peak. White wine production dominates harvest activities.	2 to 3
3. Peak harvest	Wastewater generation and harvest activities reach their peak.	3 to 14
4. Late harvest	Wastewater generation decreases to 40% of the maximum (peak) weekly flow and red wine production dominates harvest activities. Distillation of ethanol may take place.	2 to 6
5. Post-harvest	Pre-fermentation activities come to an end and maximum usage of hydroxide occurs.	6 to 12
6. None harvest	Wastewater volume is at its minimum (less than 30% of the peak weekly flow). Wastewater quality depends on daily activities.	10 to 20

Table 1.4. Major processes related to winery wastewater generation and their associated contribution to wastewater quantity and quality as well as possible effects on legal wastewater quality parameters (after Van Schoor, 2005).

Winery operation	Contribution to total wastewater quantity	Contribution to wastewater quality	Effect on legal wastewater quality parameters
Cleaning water			
Alkali washing (removal of K-bitartrate) and neutralization	Up to 33%	Increase in Na^+, K^+, COD and pH Decrease in pH	Increase in EC, SAR, COD Variation in pH
Rinse water (tanks, floors, transfer lines, bottles, barrels, *etc.*)	Up to 43%	Increase in Na^+, P, Cl^-, COD	Increase in EC, SAR, COD Variation in pH
Process water			
Filtration with filter aid	Up to 15%	Various contaminants	Increase COD and EC
Acidification and stabilization of wine	Up to 3%	H_2SO_4 or NaCl	Increase COD and EC Decrease in pH
Cooling tower waste	Up to 6%	Various salts	Increase COD and EC
Other sources			
Laboratory practices	Up to 5-10%	Various salts, variation in pH, *etc.*	Increase COD and EC

1.3.3.2. Quality of wastewater generated in wineries

In contrast to the volumes of wastewater produced, there are many reports on the quality thereof, particularly in terms of COD or biological oxygen demand (BOD) (Chapman *et al*., 1995; Ryder, 1995; Deans, 2003; Jeison *et al.*, 2003; Sheridan *et al*., 2005; Baker & Hinze, 2007; Kriel, 2008; Matthews, 2008; Arienzo *et al*., 2009; Mulidzi *et al*., 2009a). The BOD is estimated as 66% of the COD (Van Schoor, 2005). Winery wastewaters also contain high levels of K^+ and Na^+ (Laurenson *et al.*, 2012). Although various parameters may be used to evaluate winery wastewater, COD, pH, SAR, EC, chloride (Cl^-), K^+ and Na^+ are considered to be important. A survey was carried out in 2000 to evaluate the winery wastewater generated by the South African industry in terms of these variables (Mulidzi *et al*., 2009a). Results of this survey showed that there is considerable variation in wastewater quality parameters between wineries, but there is also a strong seasonal variation at most wineries. A similar seasonal trend was reported for winery wastewater in Australia (Arienzo *et al*., 2009). These trends were confirmed where effluents of two wineries were monitored frequently (Sheridan *et al.*, 2011). Considering the legal requirements for irrigation water quality in South Africa (Table 1.5), results of the survey confirmed that the majority of South African wineries are not able to irrigate crops beneficially as part of the General Authorisation with wastewater unless the water is first subjected to an effective form of pre-treatment, or unless there is relaxation of the General Authorisations.

Table 1.5. General Authorisations for legislated limits for chemical oxygen demand (COD), faecal coliforms, pH, electrical conductivity (EC) and sodium adsorption ratio (SAR) for irrigation using wastewater in South Africa (Department of Water Affairs, 2013).

Parameter	**Maximum irrigation volume allowed (m^3/day)**		
	< 50	**< 500**	**< 2000**
COD (mg/L)	5 000	400	75
Faecal coliforms (per 100 ml)	1 000 000	100 000	1 000
pH	6-9	6-9	5.5-9.5
EC (mS/m)	200	200	70-150
SAR	<5	<5	Other criteria apply

Different winemaking processes also affect the composition of winery wastewater. In the case of off-skin winemaking, sugars are the main component of the organic load in the effluent water, whereas classical winemaking methods generate wastewaters containing high levels of ethers and ethanol (Bories & Sire, 2010). However, it is also possible that spikes of extremely low quality can be caused by process interruptions. Power failure, fire, flood, storms, over- or under-loading of wastewater treatment systems, temporary unavailability of wastewater holding dam capacity and the absence of trained operators may cause process interruptions (Campos *et al*., 2000; Van Schoor, 2005; Baker & Hinze, 2007).

1.3.4. Management of winery wastewater

1.3.4.1. Wastewater treatment

Wastewater is usually collected in one or more sumps at the wineries. The first step in the treatment of winery wastewater is usually to remove the solids such as grape pips, skins and stems. This is obtained by passing the water through a screen filter. The latter is a simple, but effective step and helps to prevent other treatment machinery from getting clogged with solids (Mosse *et al.*, 2011). The wastewater is normally acidic and the pH can be less than 3. Therefore lime is added to the water in order to increase the pH to the legal or crop requirement (Van Schoor, 2005). The water is then pumped to sedimentation or maturation ponds to allow settling of the remaining solids. Depending on the quality of the wastewater at this stage, the water can be used to irrigate selected crops, such as Kikuyu grass, in specific soils. A further step could be to circulate and aerate the wastewater in dams using an aeration pump system. If these steps are managed correctly, the treatment of the wastewater can be fairly successful, particularly in reducing the COD levels (Tables 1.6 & 1.7 & Fig. 1.1).

Table 1.6. Mean winery wastewater quality during the crushing season and in aerated storage ponds in California's North Coast region (after Ryder 1995).

Parameter	Crushing season	Reclaimed water
COD[1] (mg/L)	3780	15
pH	4.1	7.7
Nitrogen (mg/L)	20	5
Phosphorus (mg/L)	10	2
Dissolved solids (mg/L)	800	500

[1] Adjusted from biological oxygen demand (BOD) where BOD = 66% of COD.

Table 1.7. Variation of chemical oxygen demand (COD) and total suspended solids (TSS) in raw and treated winery effluent (after Baker & Hinze, 2007).

Sampling date	COD[1] (mg/L)		TSS (mg/L)	
	Wastewater	Final effluent	Wastewater	Final effluent
18 November 2005	9091	16	1700	92
19 December 2005	2727	28	265	66
13 February 2006	3788	8	280	16
23 March 2006	6621	788	940	1080
28 April 2006	644	72	319	683
08 June 2006	5788	64	245	460
18 January 2007	4848	14	400	53
28 March 2007	6712	379	1040	617

[1] Adjusted from biological oxygen demand (BOD) where BOD = 66% of COD.

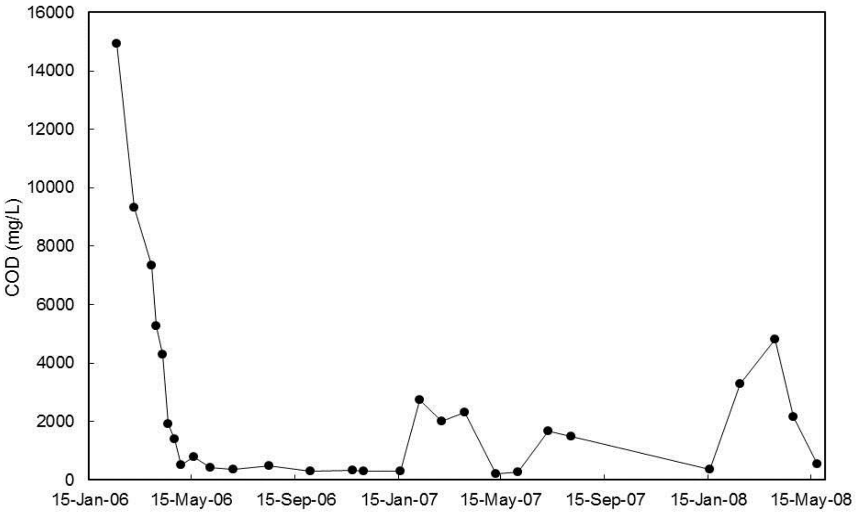

Figure 1.1. Seasonal variation in level of chemical oxygen demand (COD) in treated winery wastewater following aeration of the water which commenced in January 2006 (data supplied by the courtesy of the Botha winery).

Up-flow anaerobic sludge blanket (UASB) technology can also be used to treat winery wastewater (Matthews, 2008). This technology relies on anaerobic digestion, a biological process in which organic matter is converted to methane and carbon dioxide in the absence of air. The process involves a synergistic relationship between four different groups of bacteria, namely hydrolytic, fermentative-acidogenic, acetogenic and methanogenic. The bacteria cluster into granules which settle out to form a dense bed of sludge that is retained in the system. This is a distinct advantage over aerobic systems which produce masses of surplus sludge that must be disposed of. However, disadvantages are that nutrient removal is not feasible in anaerobic systems and trained staff are needed to operate UASB systems. Anaerobic digestion is often limited by the presence of refractory and toxic compounds in the wastewater, but ozone helps counter this effect. Pre-ozonation enhances the biodegradability of organic matter by converting these compounds into simpler molecules. Post-ozonation may be used as a “polishing” step. In addition, installation costs are relatively high (Mosse *et al.*, 2011).

Worldwide, most UASB plants have operational volumes of 100 000 litres to 10 million litres (Matthews, 2008). Only a few operate on less than 50 000 litres. A winery near Franschhoek operates a relatively small, fully automatic UASB system which can treat 25 000 litres per day. This particular wastewater treatment plant reduces the COD to *c.* 250 mg/L throughout the year. It was also shown that UASB technology can be used for the successful treatment of wastewater generated in the production of Chilean pisco, an aged drink distilled from grapes (Jeison *et al*., 2003). Expanded granular sludge bed (EGSB) technology was also tested in this study, but it was more difficult to operate and required higher capital investment, as well as operational costs compared to the UASB technology.

1.3.4.2. Disposal or utilization of winery wastewater

1.3.4.2.1. Return to natural resources

In terms of the General Authorisations published in Government Notice No. 665 (Department of Water Affairs, 2013) in terms of section 39 of the National Water Act (1998), untreated wastewater from wine cellars would rarely, if ever, qualify for discharge into natural water resources (Van Schoor, 2005). Given the quality of the treated water, most wastewaters would still not be suitable for discharge into natural water resources. Consequently, this practice is not really considered as a disposal option.

1.3.4.2.2. Disposal ponds

Some wineries pump the treated wastewater into ponds or storage dams. If the water is not re-used for irrigation, it evaporates or seeps into deeper soil layers when the ponds or dams are unlined (Mulidzi *et al.*, 2009b). Multi-stage facultative aerobic ponds have been used successfully for some 30 years for treatment and storage of winery wastewater in California (Ryder, 1995). These ponds are lined to prevent seepage of water into underground water streams and are aerated sufficiently to prevent objectionable odour generation.

1.3.4.2.3. Irrigation with winery wastewater

In South Africa more than 93% of wine cellars dispose of their effluent by means of land application (Van Schoor, 1995). The majority of cellars currently dispose effluent by irrigation as the primary disposal option. Land application systems are ideally suited for the treatment of organic carbon contained in winery effluents because the water in the soil system transports the organic contaminants to the aerobic microbial populations. However, it is important that waterlogging should be avoided. Consequently, it is essential to allow sufficient time between irrigations for the soil to become aerobic (Chapman *et al.*, 1995).

1.3.4.2.3.1. Crops irrigated with winery wastewater

In most cases, the wastewater is used for the irrigation of small permanent pasture grazing paddocks close to the wineries. The pastures mainly consist of Kikuyu grass, but Fescue grass can also be irrigated with winery wastewater (Arienzo *et al.*, 2009). There are also cases in Australia where treelots, *e.g. Eucalyptus camaldulensis*, are irrigated with winery wastewater (Chapman *et al.*, 1995; Deans, 2003; Anonymous, 2010). Research results have also shown that lemon nursery trees could successfully be irrigated with wastewater generated by a pisco distillery after the water had been treated using UASB technology (Jeison *et al.*, 2003). The pisco distillery wastewater was also disposed of in a Eucalyptus tree lot on a commercial scale.

Winery wastewater stored in lined and aerated ponds is used for vineyard irrigation during the rain-free spring and summer in California (Ryder, 1995). At a winery in the Clare Valley in Australia, treated wastewater of which the COD contents are presented in Table 1.7, is recycled into the raw irrigation water to be used for irrigation of grapevines (Baker & Hinze, 2007). In this particular case, the treated wastewater constituted only 10% of the annual irrigated volume. The actual wastewater applied was less than 10 mm. Although some vineyards have been irrigated using winery wastewater for long periods, the effect thereof on the soils and grapevines have not been reported. In one study, where grapevines were irrigated with simulated winery wastewater, it was reported that it had little, to no impact on grapevines after one season (Mosse *et al.*, 2013).

A range of leaf analyses has been carried out from representative areas in the Eucalyptus plantation where the Berri Estate's winery dispose their wastewater (Anonymous, 2010). The relatively low nutrient levels in the winery wastewater reflected in declining, but still acceptable, levels of N, P and K^+ in the leaves. However, it was concluded that some nutrient addition might be necessary during the lifespan of the trees. During the first weeks after planting, leaves of Eucalyptus saplings that were irrigated with wastewater treated in a UASB reactor showed symptoms of Na^+ toxicity (Jeison *et al*., 2003). The lemon trees used in the experiments showed similar symptoms. The problem was caused by the sodium hydroxide (NaOH) required for pH control in the UASB reactor. Approximately 2 g/L NaOH had been applied during the first weeks after reactor start up. However, after a few weeks the biogas production provided a significant level of alkalinity by CO_2 dissolution. Consequently, NaOH application was reduced to less than 20% of its original level. The Eucalyptus saplings recovered without any permanent damage. Unfortunately, the Na^+ concentrations in the treated wastewater were not reported. There are also no other reports on the effects of irrigation with winery wastewater on the performance of most of the different species mentioned above.

Recently, research has shown that potted fodder beet grown during summer in sandy soil collected from the field trial at Rawsonville, absorbed 38% of the Na^+ applied *via* Na^+-enriched irrigation water (Myburgh & Howell, 2014). The concentration of Na^+ applied was equal to that of the Na^+ concentration in the irrigation water of the 3000 mg/L COD treatment in the field trial. Furthermore, the fodder beet reduced exchangeable soil K^+ (K^+_{ex}) by 50%, thereby indicating that it could also absorb K^+ applied *via* winery wastewater.

1.3.4.2.3.2. Irrigation systems used to dispose winery wastewater

High volume sprinklers are commonly used for applying irrigation to grazing paddocks. Full surface flood irrigation was used to dispose winery wasterwater onto Fescue grass (Arienzo *et al.*, 2009) and a Eucalyptus plantation (Anonymous, 2010). It must be noted that in the latter case, diatomaceaous earth solids entered the pipeline used to transport the winery wastewater to the plantation during the grape harvesting period. This required annual flushing and/or pigging to avoid blockages. Unfortunately, most other reports on the disposal of winery wastewater by means of irrigation did not mention the systems used to irrigate the different species. Since vineyards in Australia are almost invariably drip irrigated, it can be assumed that the winery in the Clare Valley in Australia disposed of the treated wastewater by means of a drip irrigation system (Baker & Hinze, 2007). Aboveground as well as subsurface drip irrigation was used in a field experiment in Israel to irrigate grapevines with water from sewerage waste stabilization ponds (Campos *et al*., 2000). The rationale for using drip irrigation, and particularly subsurface drip, was to minimise the risk of disease contamination by preventing direct contact between the wastewater and the edible part of the crop.

1.3.4.2.3.3. Effects of winery wastewater on soil conditions

Soil chemical status: Land application of wastewaters can increase the levels of soluble and exchangeable forms of potassium (K^+ & K^+_{ex}) more rapidly than with conventional inorganic fertilizers (Arienzo *et al.*, 2009). Furthermore, most of the K^+ in wastewater is immediately available. The effects of excessively high K^+ application on soils have not been extensively researched and are still unclear (Mosse *et al.*, 2011; Laurenson *et al.*, 2012). In addition, the fate of K^+ in soils and on grapevines irrigated with winery wastewater has received limited attention (Laurenson *et al.*, 2012). Irrigation using K^+-rich wastewaters could be beneficial to overall soil fertility, although long-term application could affect soil chemical and physical properties (Laurenson *et al.*, 2011; Mosse *et al.*, 2011). A further advantage of using winery wastewater as a source of K^+ over the use of conventional fertiliser is that it could be an efficient recycling practice in areas where the soil has low K^+. In addition to Na^+ and K^+ ions, winery wastewater can also contain calcium (Ca^{2+}) and magnesium (Mg^{2+}) ions (Mosse *et al.*, 2011). Neither of these ions are harmful to soil structure and can ameliorate the impacts of Na^+ application indirectly *via* their role in reducing the SAR. Wastewater with high levels of organic material can clog soil pores, which could decrease soil hydraulic conductivity. Due to the high K^+ levels in winery wastewater, adsorption of Na^+, and exchangeable sodium percentage (ESP), will be less than in wastewaters of comparable Na^+ concentration with no K^+ (Laurenson *et al.*, 2011). In a study investigating the irrigation of an established vineyard using artificial winery wastewater, grapevines were either irrigated with water from Lake Berryessa or artificial wastewaters containing high K^+, high K^+ plus wine, low K^+, and Na^+ (Mosse *et al.*, 2013). All treatments caused an increase in soil K^+ and Na^+. The accumulation of K^+ was restricted to the 0-20 cm soil layer, with the exception of the treatment where wine was added to the irrigation water. The addition of wine enhanced K^+ transport into the subsoil. Elevated Na^+ levels were found in the 0-20 cm and 20-40 cm soil layers. Therefore, the presence of wine, *i.e.* organic material, facilitated the transport of the K^+ within the profile. In a study investigating the long-term use of winery wastewater, *i.e.* 30 years, an accumulation of K^+, Na^+, Ca^{2+}, Mg^{2+} and boron (B^{3+}) was reported (Mosse *et al.*, 2012). Furthermore, soil that had been irrigated with winery wastewater for 30 years showed a decrease in soil pH with depth. The increased concentrations of the cations were attributed to higher levels encountered in the wastewater.

A survey was carried out in South Africa to assess the soil chemical status where winery wastewater had been disposed over prolonged periods (Mulidzi *et al*., 2009b). Control soil samples were collected close to the area of land where the wastewater was disposed. Unfortunately, there was no history about the volumes of water or the quality of the wastewater that had been applied to the disposal sites. However, by comparing the disposal site to the control samples it was shown that the winery wastewater almost invariably induced negative effects, irrespective of soil type (Mulidzi *et al*., 2009b). Furthermore, it was concluded that (i) in general, effluent disposal is poorly planned and managed, and disposal sites rarely seem to have been selected, because their soil properties are inappropriate for effluent disposal. In particular, deep sandy soils are unsuitable for disposal by ponding, mainly because of their high infiltration rates, high permeability and low water storage capacity and (ii) many disposal sites are too limited in area to permit the large volumes of effluent to be absorbed without surface runoff. This problem invariably persists despite the presence of Kikuyu swards and sandy subsoil (Mulidzi *et al*., 2009b).

Soil physical status: Unfortunately, no references on the effect(s) of irrigation using winery wastewater on in-field soil physical properties could be found in the literature. Soil chemical properties can be altered by wastewater irrigation (Vogeler, 2009; Lado & Ben-Hur, 2010), and this could influence soil structure and hydraulic properties (Sort & Alcaniz, 1999; Tarchitzky *et al.*, 1999). Dissolved and suspended solids, both organic and inorganic, may induce soil clogging through physical, chemical, and biological processes (Viviani & Iovino, 2004). Degradation of soil hydraulic properties due to physical clogging of the surface layer of soil is one of the expected risks involved in wastewater reuse for irrigation (Viviani & Iovino, 2004). Their investigations, conducted in soil columns, showed that the reductions of saturated hydraulic conductivity were only restricted to the 0-2 cm depth layer, and the lower part of the column was not affected by wastewater application.

Irrigation using olive mill wastewaters increased soil hydrophobicity and reduced drainable porosity, because of increasing organic matter content (Mahmoud *et al.*, 2010). Furthermore, soil hydraulic conductivity was reduced compared to a control site. After 15 years of application of such wastewater, the highest infiltration rate was observed because of the presence of large and deep shrinkage cracks. According to Barbera *et al.* (2013), irrigation using olive mill wastewaters can have a temporary positive effect on soil. However, in clay soils, salt accumulation could lead to disintegration of the soil structure. Subsequently the hydraulic conductivity would decrease. Regarding the use of wastewater generated by oil production, research showed that the use of such water created a sodicity problem, which had negative effects on soil physical properties such as infiltration rate, saturated hydraulic conductivity and pore size distribution (Al-Haddabi *et al.*, 2004).

After four years of irrigation using secondary-treated municipal wastewater, saturated and near saturated hydraulic conductivity of a soil decreased from 567 mm/h and 40 mm/h to 56 mm/h and 3 mm/h, respectively (Sparling *et al.*, 2006). In a study on a sewage farm to investigate the effects of long-term irrigation using sewage effluent on soil physical properties, bulk density was significantly lower compared to soil which was irrigated with well-water. Furthermore, the longer the irrigation with sewage water took place, the lower the bulk density became (Mathan, 1994). Subsequently, hydraulic conductivity increased. In a study to evaluate the long-term effect of wastewater application on soil physical properties, it was also found that this practice increased organic matter content and reduced bulk density. In addition to this, long-term wastewater irrigation resulted in a higher aggregate stability and saturated hydraulic conductivity (Vogelier, 2009). In contrast, leaching a loamy and a clay soil with treated sewage effluent reduced saturated hydraulic conductivity (Lado & Ben-Hur, 2009). This was attributed to the plugging of the pores with suspended solids. However, the saturated hydraulic conductivity of a sandy soil was not affected because of its large average pore size. In a non-calcareous, sandy soil, the higher sodicity enhanced seal formation, reduced infiltration, and increased runoff. However, there were no effects of the effluent on a calcareous soil under similar conditions. According to Tarchouna *et al*. (2010), irrigation using wastewater from a sludge treatment plant reduced both saturated and unsaturated hydraulic conductivity of a very sandy soil, but it was still high enough to allow water percolation.

The negative effects of high Na^+ levels in irrigation water on the hydraulic properties of soils are well known. According to Levy and Van der Watt (1990), increasing the amount of K^+ resulted in a decrease in hydraulic conductivity and infiltration rate of soils. There is a broad spectrum of possible effects of K^+ on infiltration, ranging from being similar to Na^+, to being similar to Ca^{2+} (Arienzo *et al.*, 2009). Furthermore, it was concluded that, relative to exchangeable Ca^{2+} and Na^+, K^+ had an intermediate effect on the soil hydraulic properties. Since winery wastewater can contain high Na^+ and/or K^+ concentrations, the effect of SAR and potassium adsorption ratios (PAR) on the soil hydraulic conductivity at a wastewater disposal site was investigated in a laboratory study (Arienzo *et al*., 2009). The results showed that the soil hydraulic conductivity was considerably reduced when the SAR or the PAR exceeded 20. These negative effects even occurred when the electrolyte concentrations in the soil were relatively high, *i.e.* > 40 meq/L. It was also shown that the negative effect of Na was more pronounced compared to K^+ at the same electrolyte concentration.

Laurenson *et al*. (2012) used a combination of solutions with known SAR and PAR to investigate the binding of Na^+ and K^+ for a Barossa Chromosol. Their conclusions were that ESP corresponding to a given SAR was increasingly lowered at higher K^+ concentrations. Subsequently, if SAR in wastewater remains similar during vintage, reductions in soil ESP may occur because of increasing K^+ and exchangeable potassium percentage (EPP). Changes in soil structure will therefore be less pronounced compared to wastewaters with comparable monovalent concentrations of only Na^+. Therefore, in the case of winery wastewater, replacing Na^+-based cleaners with K^+-based cleaners can contribute towards decreasing clay dispersion risks.

In contrast, substitution of K^+-based cleaning agents with Na^+-based ones has been proposed due to the high K^+ content in winery wastewater (Arienzo *et al*., 2009). Using Na^+-based cleaning agents might reduce the K^+, but in the long run increased Na^+ levels in soil will certainly cause more structural damage compared to K^+. In addition, Na^+ could reach toxic levels in soils. On the other hand, K^+ accumulation in the soil could be reduced through uptake and removal by crops grown on winery wastewater disposal sites. Furthermore, it should be kept in mind that the cost of potassium hydroxide is substantially higher than NaOH (Mosse *et al.*, 2011).

1.3.4.2.3.4. Present guidelines if winery wastewater is re-cycled for irrigation

Wastewater quality standards were proposed for irrigation of vineyards using treated winery wastewater stored in aerated ponds in California (Ryder, 1995). The maximum COD, faecal coliforms, pH, EC_e and SAR standards (Table 1.8) are more or less comparable to the legislated limits for irrigation with wastewater in South Africa, *i.e.* if less than 2 000 m^3 is irrigated per day (Table 1.5).

Table 1.8. Proposed reclaimed effluent water quality standards for vineyard re-use (after Ryder, 1995).

Parameter	Optimum value	Maximum values
pH	6.5 - 8.4	6.0 – 9.0
EC_e (dS/m)	< 0.75	< 1.50
TDS (mg/L)	< 500	< 1000
Alkalinity (mg/L $CaCO_3$)	< 150	< 250
Hardness (mg/L $CaCO_3$)	< 250	< 400
Ca (mg/L)	< 60	< 100
Mg (mg/L)	< 25	< 50
Na (mg/L)	< 65	< 100
K (mg/L)	< 5	< 10
Fe (mg/L)	< 5	< 5
Mn (mg/L)	< 0.2	< 0.5
Cu (mg/L)	< 0.01	< 0.05
Zn (mg/L)	< 2	< 5
Bicarbonate (mg/L)	< 200	< 300
Carbonate (mg/L)	< 5	< 10
Chloride (mg/L)	< 70	< 120
Sulfate (mg/L)	< 150	< 250
N (mg/L)	< 5	< 10
P (mg/L)	< 5	< 10
B (mg/L)	< 0.5	< 1
SAR	< 6	< 9
COD(1) (mg/L)	< 60	< 100
Coliforms (MPN(2)/100ml)	< 23	< 230

(1) Adjusted from biological oxygen demand (BOD) where BOD = 66% of COD.

(2) Most probable number.

1.3.5 CONCLUSIONS

Wineries generate large volumes of poor quality wastewater, particularly during harvest. The use of winery wastewater for vineyard irrigation could have many potential benefits for the wine industry. Since water is becoming increasingly scarce, the use of winery wastewater as an alternative source of irrigation water for vineyards could reduce the pressure on water resources. However, there is no available information to guide legislators regarding what specific quality of the winery wastewater could be permitted to be applied for vineyard irrigation under a specific set of conditions to minimize the effects on soil and grapevine responses. In this regard, a research project to investigate the possible use of diluted winery wastewater for vineyard irrigation was initiated and funded by the WRC of South Africa, and co-funded by Winetech, THRIP and the ARC. With regard to the hypothesis of the study, it was expected that irrigating vineyards using diluted winery wastewater rather than river water could alter soil chemical properties after wastewater application due to the high levels of K^+ and Na^+ in winery wastewater. This could affect juice characteristics and, subsequently, wine character and overall quality.

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CHAPTER 2: EXPERIMENTAL LAYOUT, INFRASTRUCTURE FOR IRRIGATION OF GRAPEVINES WITH DILUTED WINERY WASTEWATER IN A FIELD EXPERIMENT AND PRELIMINARY MEASUREMENTS

2.1. INTRODUCTION

Wineries produce large volumes of wastewater, particularly during the harvest period from February until March. Generally, the chemical oxygen demand (COD) in the wastewater (Arienzo *et al.*, 2009; Mulidzi *et al.*, 2009a; Conradie *et al.*, 2014) is higher than the allowable limits for irrigation of agricultural crops as stipulated by the General Authorisations for legislated limits for irrigation using wastewater in South Africa (Department of Water Affairs, 2013). Surveys have shown that soil chemical conditions deteriorated where grazing paddocks were irrigated with winery wastewater over a period of time (Mulidzi *et al*., 2009b). Furthermore, the sodium (Na^+) in the water could accumulate in the soil, which could have negative effects on the soil physical properties in the long run (Arienzo *et al.*, 2009; Laurenson *et al.*, 2012). This could be more pronounced in dry regions where winter rainfall is inadequate to leach accumulated salts from the soil. An alternative to the grazing paddocks would be to re-use diluted winery wastewater for irrigation of agricultural crops. Since many wineries are close to, or even surrounded by vineyards, it would be a logical alternative to re-use the diluted wastewater to irrigate grapevines. To assess the effects of diluted wastewater on grapevine growth, yield and wine quality, a research project was initiated and funded by the Water Research Commission (WRC). The project was co-funded by Winetech and the Agricultural Research Council (ARC). The Infruitec-Nietvoorbij institute of the ARC at Stellenbosch was contracted to carry out the field experiment.

The grapevines had to be subjected to irrigation with winery wastewater containing a range of COD levels to determine a possible threshold concentration for sustainable use. Control grapevines were irrigated with river water (raw water). Since one of the major objectives was to assess the effect of the wastewater irrigation on wine quality characteristics, the experimental plots had to be large enough to produce at least 40 kg of grapes, *i.e.* the minimum required for small scale winemaking. Furthermore, the water had to be applied with micro-sprinklers rather than drip to (i) distribute the water over the total surface and (ii) reduce the risk of emitter clogging. Therefore, relatively large volumes of water were required to apply a single irrigation for the three replication plots of each treatment. Consequently, the wastewater had to be diluted in 15 m^3 tanks. Since previous wastewater studies were carried out in laboratories (Laurenson, 2010; Laurenson *et al*., 2012), there were no guidelines for diluting winery wastewater on such a large scale. In a field study using simulated winery wastewater to irrigate grapevines by means of drip irrigation, the concentrated solutions were made in 189 L tanks, and injected into the irrigation system

(Mosse *et al.*, 2013). In this particular trial, grapevines received 227 mm of irrigation per season. In other winery wastewater trials, a survey type approach is often used. In general, soil is sampled from sites which have received winery wastewater and compared to control sites which did not receive winery wastewater (Kumar *et al.*, 2006; Kumar *et. al.*, 2014). Although Kumar *et al.* (2014) investigated the effect of winery wastewater irrigation in a paired field trial, no details regarding the quality of the irrigation water were presented.

The experimental layout, infrastructure for diluting winery wastewater and its accuracy, as well as preliminary measurements are described in this chapter. Since there are no guidelines to dilute winery wastewater to a certain water quality, the objective of the study was to evaluate the efficiency of the dilution procedure where grapevines were subjected to irrigation with winery wastewater containing different levels of COD. Various materials, *e.g.* organic material, limestone as well as iron and manganese oxides, can be deposited on the inside of irrigation lines (Myburgh, 1989 and references therein). These deposits may cause clogging of drippers and micro-sprinklers, and in severe cases reduce water flow in irrigation lines. Given the poor quality of winery wastewater, particularly in terms of the high COD, it could be expected that foreign material may cause problems if the water is re-cycled for irrigation of crops. Therefore, a further objective of this study was to determine the formation of foreign material deposits in irrigation lines where diluted winery wastewater is re-cycled for irrigation.

2.2. MATERIALS AND METHODS

2.2.1. Irrigation treatments

According to the terms of reference for the project, the winery wastewater had to be diluted to obtain a range COD levels for the different treatments (Table 2.1). It should be noted that COD was the preferred indicator of wastewater quality of the Steering Committee of the project, therefore treatments were applied in terms of COD levels.

Table 2.1. Range of target chemical oxygen demand (COD) levels to which the winery wastewater was diluted for the different treatments.

Control	Target COD level in diluted winery wastewater (mg/L)							
T1	T2	T3	T4	T5	T6	T7	T8	T9
River water(1)	100	250	500	1000	1500	2000	2500	3000

(1) Abstracted from the Holsloot River.

2.2.2. Vineyard characteristics

The field trial was carried out in an eight-year-old commercial Cabernet Sauvignon/99 Richter vineyard at the Goudini winery near Rawsonville in the Breede River grape growing region of the Western Cape at 33° 41′ S latitude (Fig. 2.1). The region has a Mediterranean climate, and based on the growing degree days (GDD) from September until March (Winkler, 1962), the specific locality is in a class V climatic region (Le Roux, 1974). The vineyard is located on an alluvial flood plain of the Du Toitskloof Mountains. The sandy soil belongs to the Longlands form (Soil Classification Working Group, 1991). The soil was deep delved to 1.0 m before planting. Grapevines were planted 2.4 m × 1.2 m and trained onto a four-strand lengthened Perold trellis (Booysen *et al*., 1992). Vertical shoot positioning was carried out to prevent the development of a sprawling canopy. The vineyard was previously irrigated by means of drippers. In addition to the standard viticultural practices, measures were taken to prevent erinose mite infestation in the vineyard. This consisted of a lime sulphur spray prior to bud break, as well as three additional foliar sprays of MicroThiol™.

After an evaluation of the soil chemical status at bud break, *i.e.* mid September, potassium (K^+) fertilisation was applied at a rate of 30 kg K^+/ha in all three seasons. In the 2010/11 season, potassium chloride (KCl) was applied to all treatments in the middle of November. However, in the 2011/12 season, 30 kg K^+/ha was only applied to T1 to T6 in the middle of December as the soil K contents of T7, T8 and T9 were deemed sufficient for the grapevines. In the 2012/13 season, K fertilisation was applied to all plots in December. A standard winter cover crop of *Avena sativa* L. cv. Pallinup (oats) was cultivated and produced 5.4±0.3, 4.7±1.0, 6.7±1.2 and 7.5±1.1 t/ha dry matter for the 2009/10, 2010/11, 2011/12 and 2012/13 seasons, respectively (Fourie & Theron, 2014). In addition to the normal winter cover crop, an interception crop of *Pennisetum glaucum* (pearl millet) was cultivated in the work rows in summer to intercept salts applied *via* the diluted winery wastewater. It produced 10.4±0.8, 6.0±1.0 and 6.4±0.9 t/ha dry matter for the 2010/11, 2011/12 and 2012/13 seasons, respectively.

2.2.3. Experimental layout

All treatments were replicated three times in a randomised block design. The experimental plots were marked in July 2009. Experimental plots comprised two rows of six grapevines each, with two buffer grapevines at each end and a buffer row on each side. Each experimental plot covered 104 m^2.

2.2.4. Preliminary measurements

Grapevines in the experimental plots were pruned to two bud spurs and the baseline cane mass per plot determined on 3 August 2009. A surface plot was created to obtain an indication of the growth vigour variation in the field trial part of the vineyard. To determine the baseline soil chemical status, soil samples were collected over 300 mm increments to a depth of 1.8 m in selected plots during August 2009. The sample sites represented the variation in vegetative growth.

2.2.5. Installation and commissioning of infrastructure

2.2.5.1. Irrigation system in the experimental vineyard

The micro-sprinkler irrigation system, which allowed irrigation of the individual treatments, was installed in the vineyard during August 2009. To prevent possible damage to grapevine shoots during installation of the irrigation system, this task was completed before bud break in September. The micro-sprinklers (White base/white swivel, Gyro Sprinklers, Brackenfell) had a 30 L/h flow rate at 100 KPa.

2.2.5.2. Wastewater mix and distribution plant

The 400 m long, 110 mm diameter PVC pipeline required to convey the water from the wastewater pit at the winery to the experimental vineyard was also installed in August 2009 (Fig. 2.1). The pipeline installation was completed on 7 August. At the same time, four rows of an adjacent vineyard were pulled out to create space for the wastewater mix and distribution plant. Since the COD levels in winery wastewater can vary considerably as the winery activities change over the course of a day, wastewater was first collected in a 20 m^3 stock dam to obtain water with a stable concentration. The wastewater was pumped from the stock dam using a 30 m^3/h pump (Fig. 2.2) to the eight plastic tanks, one for each dilution treatment, at the mix and distribution facility near the vineyard (Fig. 2.3). The water mix and distribution facility was completed on 12 February 2010.

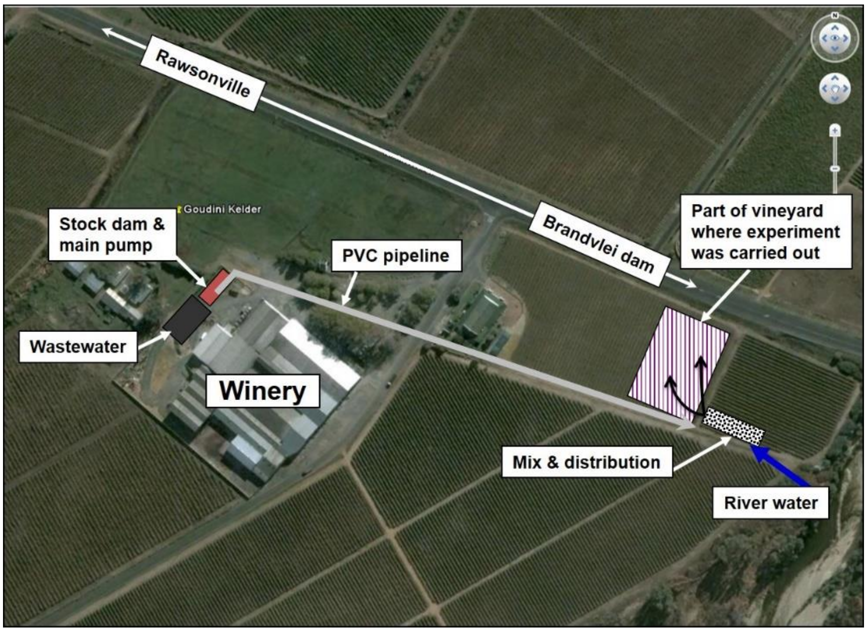


Figure 2.1. Layout of the infrastructure and experimental vineyard where irrigation using diluted winery wastewater was carried out.

Figure 2.2. Main pump and stock dam in which wastewater from the winery was collected.

Figure 2.3. Tanks in which winery wastewater was diluted with river water before it was pumped to the different treatment plots.

2.2.5.3. Procedure for the dilution of the winery wastewater

Wastewater coming from the winery is first screened to remove coarse particles (Fig. 2.4). During this process, lime is added to increase the pH of the water. The wastewater then flows through a concrete sedimentation pond to allow settling of substances, *e.g.* tartaric acid (Fig. 2.5). The treated water is collected in a pit from where it is pumped onto a grass paddock. Wastewater for the experiment was abstracted from this pit. The dilution and mixing procedure was carried out according to the following steps: First, the 20 m^3 stock dam was filled with wastewater from the collection pit at the winery (Fig. 2.1). Following this, the COD in the stock dam water, as well as in the water from the Holsloot River, *c.* 80 m from the tanks was measured. The COD in the samples was measured using a portable spectrophotometer (Aqualitic COD-reactor®, Dortmund) with the appropriate test kits (COD, CSB, 0-15000 mg/L). This procedure requires a two hour oxidation time. The COD levels were used to calculate the volumes of winery wastewater and river water required to obtain the different target COD levels. The volume (m^3) of wastewater required from the stock dam (V_S) to obtain a certain target COD concentration (COD_T) was calculated as follows:

$$V_S = (COD_T - COD_R) \times V_T \div (COD_S - COD_R) \qquad \text{(Eq. 2.1)}$$

where COD_R and COD_S are the COD concentrations (mg/L) in the river water and the stock dam, respectively, and V_T is the tank volume (m^3).

Figure 2.4. Screening equipment used to remove coarse particles from the winery wastewater.

Figure 2.5. Sedimentation pond used to settle substances such as tartaric acid from the winery wastewater.

Since the COD_R was undetectably low, a value of 1 mg/L was used in the calculations. The required volume of winery wastewater for a specific treatment was first pumped into the designated tank. Following this, the tank was filled by pumping water from the river. Since the inlets were near the bottom of the tanks, the river water was forced to flow through the wastewater already inside the tanks. Furthermore, the inlet on the inside of the tank was set at an angle of *c.* 45 degrees to create a swirling effect while water was flowing into the tank (Fig. 2.6). The total volume pumped into the tanks was only 13.5 m^3 to reduce the risk of wastewater spills. Once the tanks were filled, the water was pumped onto the treatment plots using 2.5 m^3/h pumps (Fig. 2.6). Approximately one hour after the irrigation commenced, the COD in the diluted water was measured at the tank outflows. The foregoing steps took almost three days to complete. To avoid the settling of substances, the diluted water was never allowed to stand overnight in the tanks. Irrigations were applied almost immediately after the tanks were filled.

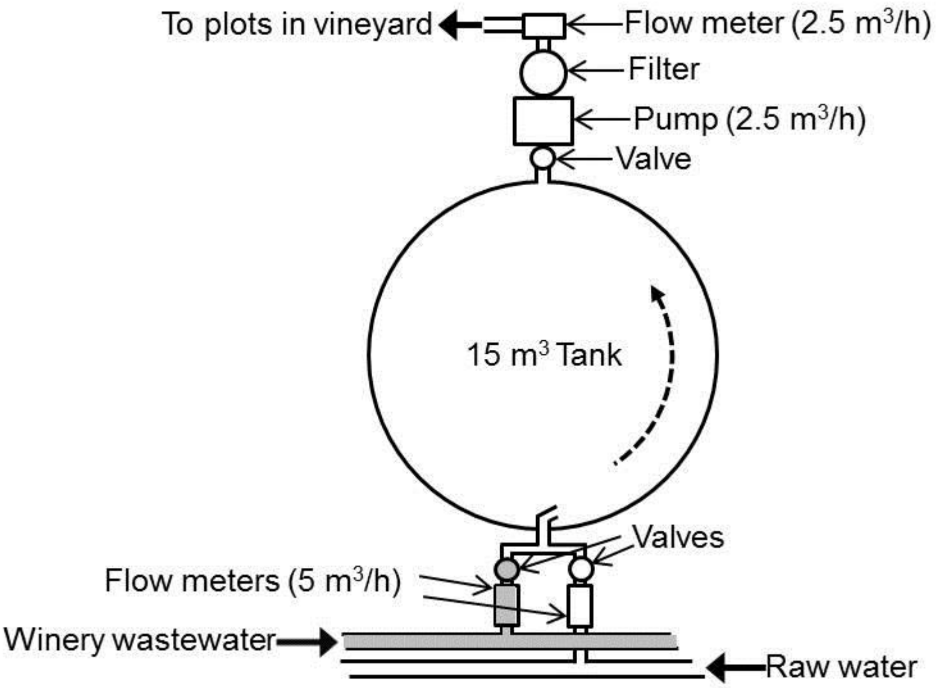


Figure 2.6. Diagram to illustrate the components used to dilute and mix the winery wastewater in the tanks.

2.2.6. Monitoring deposits in irrigation lines

2.2.6.1. Non-destructive method

During the 2010/11, 2011/12 and 2012/13 seasons, foreign material accumulation on the inside surface of the irrigation lines was determined by means of non-destructive deposit detectors containing removable glass slides. The idea was to measure the deposits on the glass slides. The detectors were installed in the tubes which connected the micro-sprinklers to the lateral irrigation lines (Fig. 2.7). The deposit detectors were installed at the last micro-sprinkler in a lateral, *i.e.* where the highest level of deposits is to be expected. Before installation, the glass slide in each detector was weighed to the nearest 0.001 mg using an electronic balance. The detectors were installed in November of each season, *i.e.* before irrigation with river water started. The detectors were only installed in the three replications of T1, T3, T5, T7 and T9. When the irrigation season ended in May, the glass slides were carefully removed from the detectors and placed in bottles for transportation to the laboratory. Following drying at 60°C for 24 hours in an oven, the slides were weighed again. The deposits were calculated as the difference between the initial mass and the mass recorded after drying the glass slide, and were expressed as mass of dry material per unit surface (mg/m^2).

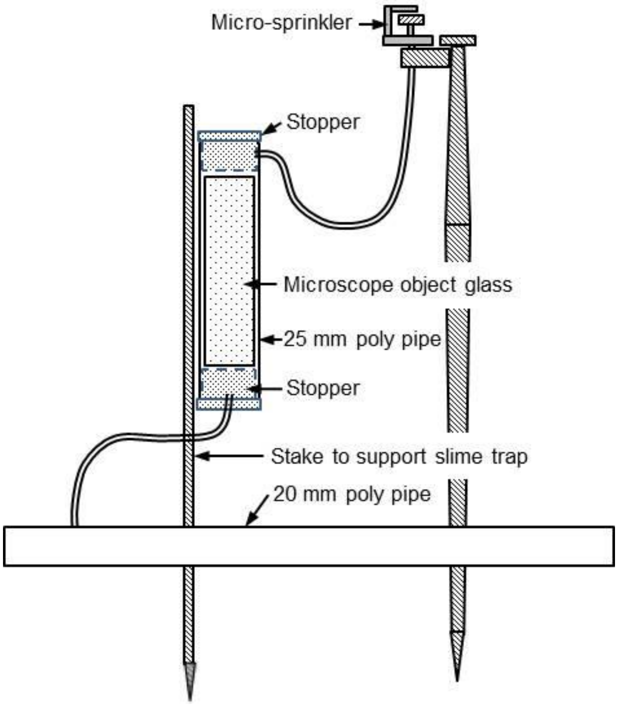


Figure 2.7. Schematic diagram illustrating the components of the deposit detectors.

2.2.6.2. Destructive method

Following the last wastewater irrigations in April 2013, 50 cm sections of poly pipe were removed from the irrigation laterals, and replaced with new pipe. Samples were collected from each experimental plot. The pipe samples were carefully cleaned on the outside, dried at 60°C for 24 hours and weighed. The pipe sections were then cleaned on the inside using a bottle brush. After rinsing under running water, the pipe samples were again dried at 60°C for 24 hours and weighed. The deposits accumulated over the four years on the inside surface of the pipes were calculated as the difference between the initial mass and the mass recorded after drying. The deposits were expressed as mass of dry material per unit surface (mg/m^2).

2.3. RESULTS AND DISCUSSION

2.3.1. Experimental layout

Before the wastewater treatments were applied, mean cane mass per experimental plot varied between 2.25 kg/grapevine and 0.25 kg/grapevine (Fig. 2.8). Results indicated that the pruning mass variation between the replications was related to the Bray II-P, extractable K^+ and organic carbon contents in the soil (Tables 2.2 & 2.3). The role of organic carbon is not a direct nutritional effect, but serves as an indication of the N availability in the soil. The surface plot was used to determine the layout for the three replications required for reliable statistical analyses of the data as indicated in Figure 2.9. In addition to the river water control and eight diluted winery wastewater treatments, there was also a treatment representative of all the grower's vineyard management practices (T10). The ten treatments were randomly distributed within each replication. The treatment representative of the grower's vineyard management practices was only to be used as a legal control and will not be discussed further in the dissertation.

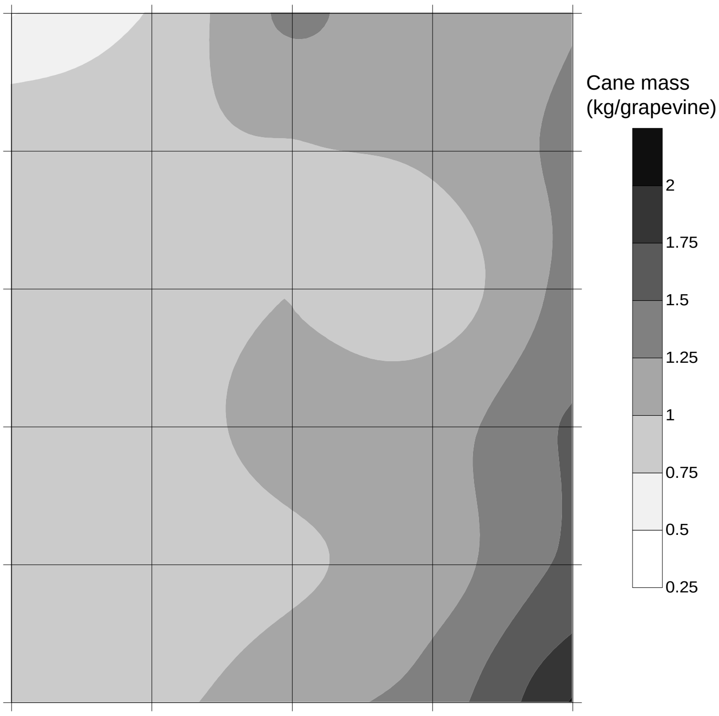


Figure 2.8. Surface plot indicating the variation in grapevine growth vigour in the part of the vineyard before the winery wastewater experiment commenced.

Table 2.2. Cane mass at pruning in August 2009, particle size analyses, estimated soil water retention (SWR) and extractable cation content in the sandy soil where the wastewater treatments were applied.

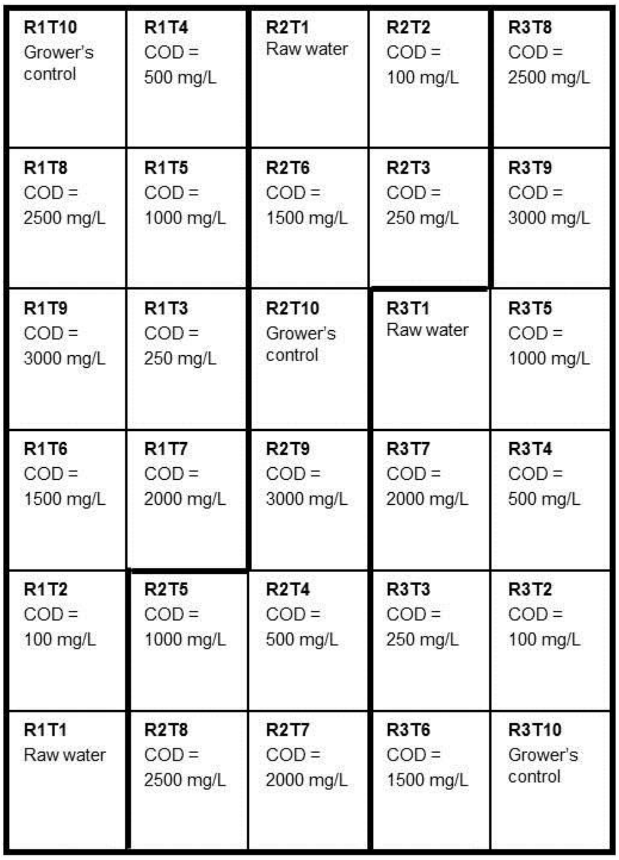
Replication	Cane mass (kg/grapevine)	Clay (%)	Silt (%)	Sand (%)			SWR (mm/m)	Bray II-P (mg/kg)	Extractable cations (cmol$^{(+)}$/kg)			
				Fine	Medium	Coarse			Na^{+}	K^{+}	Ca^{2+}	Mg^{2+}
1	0.66	1.7(1)	0.7	49.6	38.1	9.8	100	128	0.06	0.08	1.56	0.73
2	0.98	1.7	0.9	53.6	33.4	10.5	109	137	0.07	0.09	1.76	0.75
3	1.54	1.8	0.8	49.3	36.6	11.6	100	143	0.06	0.13	1.58	0.77

(1) Data are the means for the 0-1.8 m soil depth.

Table 2.3. Baseline organic carbon, micro nutrients and selected heavy metal contents in the sandy soil where the wastewater treatments were applied.

Replication	Organic carbon (%)	Micro nutrients (mg/kg)					Heavy metals (mg/kg)		
		Cu^{2+}	Zn^{2+}	Mn^{2+}	B^{3+}	Fe^{2+}	Cd^{2+}	Pb^{2+}	Hg^{2+}
1	0.54(1)	2.66	1.38	2.32	0.06	14.75	0.06	0.12	0
2	0.61	1.99	1.57	2.87	0.11	13.84	0.02	0.02	0
3	0.95	7.62	4.72	2.32	0.06	20.05	0.04	0.12	0

(1) Data are the means for the 0-1.8 m soil depth.



R1T10 Grower's control	**R1T4** COD = 500 mg/L	**R2T1** Raw water	**R2T2** COD = 100 mg/L	**R3T8** COD = 2500 mg/L
R1T8 COD = 2500 mg/L	**R1T5** COD = 1000 mg/L	**R2T6** COD = 1500 mg/L	**R2T3** COD = 250 mg/L	**R3T9** COD = 3000 mg/L
R1T9 COD = 3000 mg/L	**R1T3** COD = 250 mg/L	**R2T10** Grower's control	**R3T1** Raw water	**R3T5** COD = 1000 mg/L
R1T6 COD = 1500 mg/L	**R1T7** COD = 2000 mg/L	**R2T9** COD = 3000 mg/L	**R3T7** COD = 2000 mg/L	**R3T4** COD = 500 mg/L
R1T2 COD = 100 mg/L	**R2T5** COD = 1000 mg/L	**R2T4** COD = 500 mg/L	**R3T3** COD = 250 mg/L	**R3T2** COD = 100 mg/L
R1T1 Raw water	**R2T8** COD = 2500 mg/L	**R2T7** COD = 2000 mg/L	**R3T6** COD = 1500 mg/L	**R3T10** Grower's control

Figure 2.9. Experimental layout and treatments to determine the effects of diluted winery wastewater on soil properties, as well as grapevine yield and wine quality. (R = Replication and T = treatment; COD = target chemical oxygen demand level).

2.3.2. Efficacy of wastewater dilution

The seasonal COD in the diluted winery wastewater was generally close to the treatment target values presented in Table 2.1. However, in some cases the COD in the diluted water differed from the target treatment values. Therefore, the standard deviation from the mean was relatively large, particularly during the 2009/10 season (Fig 2.10). Possible reasons for the deviation from the target COD levels were as follows: The main pipeline was initially filled with river water after the system had been completed. The COD concentration in the water in the pipeline was not accounted for, and therefore, probably had a diluting effect when the water was mixed. From the second irrigation onwards, the main pipeline was filled with winery wastewater before the water was pumped into the tanks. The water meters used to monitor the volumes of water flowing into the tanks also presented problems, particularly when the flow rates increased when only one or two tanks were being filled at a time. This problem was overcome by marking the required water levels on the outside of the translucent tanks.

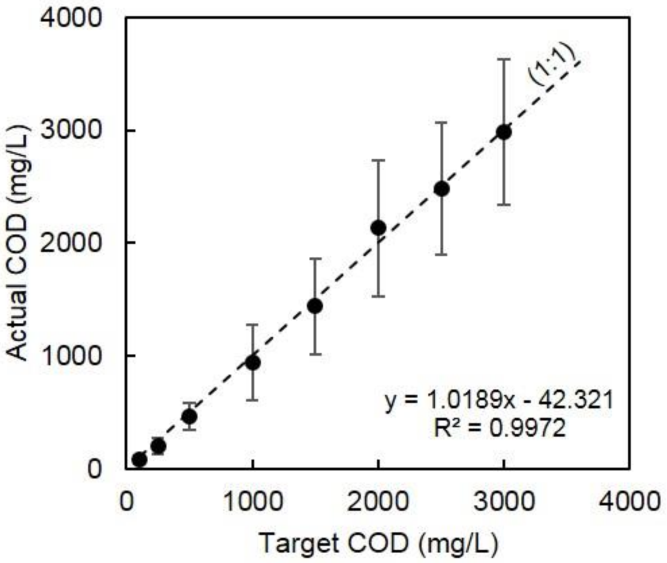


Figure 2.10. Relationship between mean actual chemical oxygen demand (COD) in diluted winery wastewater applied to grapevines and the target COD levels of treatments T2 to T9 during the 2009/10 season. Vertical bars indicate standard deviation (n = 3).

An operator error also occurred when the plunger of the mechanical pipette was pressed too deep when the water samples were transferred into the test kit vials. This caused an overestimation of the COD level in the stock dam, which in turn resulted in too low COD levels in the diluted water in the tanks, particularly when low COD in the water required sample volumes of 2 ml compared to the 0.2 ml required for the higher COD levels, *i.e.* > 1500 mg/L. Since the dilution and irrigation procedures took almost three days to complete, the project team investigated ways to save time. According to a technical advisor of a company that supplies the test kits, one hour oxidation time would be adequate for most COD levels in water samples. If the oxidation time could be reduced, it would significantly reduce the time required to carry out the whole procedure as described above, particularly the two hour waiting period after the stock dam had been filled. Consequently, it was decided to reduce the oxidation time for the samples from the stock dam to one hour for the second and third treatment applications in 2009. Since the target COD levels after mixing were higher than the target levels in the second and third irrigations, the possibility that the one hour oxidation time could have underestimated the COD concentrations, particularly in the stock dam, was investigated. The COD in three water samples containing different COD levels were measured after different oxidations times. These results showed that the COD reading became constant in less than one hour when the COD concentrations were below *c.* 2000 mg/L (Fig. 2.11). However, in the case of the high concentrations, *e.g.* in the stock dam, the COD readings only reached a plateau after *c.* 90 minutes. Based on these findings it was decided to standardize the oxidation time for all COD analyses to two hours.

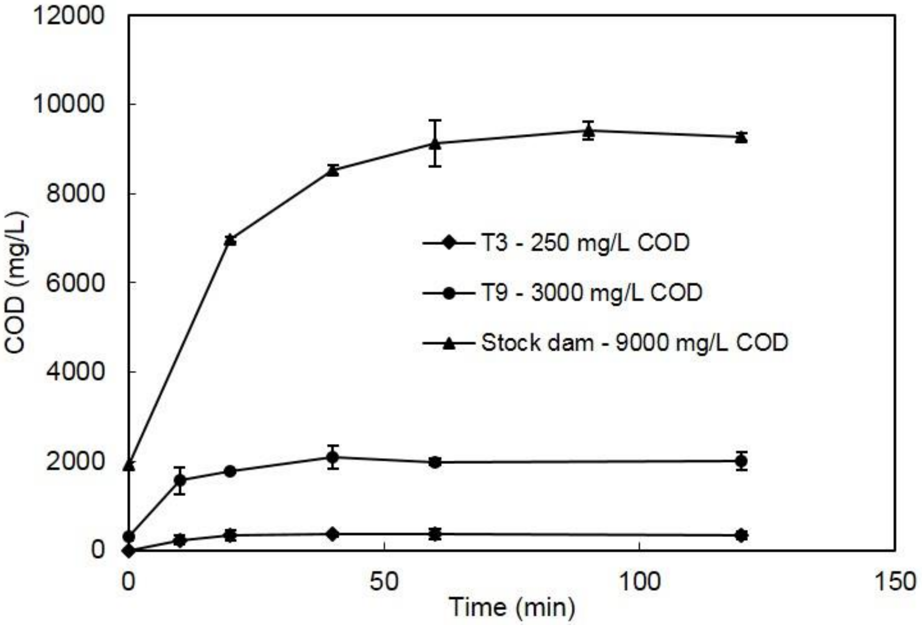


Figure 2.11. The effect of oxidation time on spectrophotometrically measured chemical oxygen demand (COD) in winery wastewater that was diluted to different COD levels. Vertical bars indicate standard deviation (n = 3).

As the project progressed, the above mentioned problems and possible causes for error were addressed and eliminated where possible. Due to this, the accuracy of the treatment application improved substantially in the subsequent seasons compared to 2009/10 (Figs. 2.12 to 2.14). Furthermore, after each irrigation, the target COD for the following irrigation was adjusted as follows:

$$C_A = (C_T \times n) - \Sigma C_P \qquad \text{(Eq. 2.2)}$$

where C_A is the adjusted COD concentration, C_T is the target COD for a specific winery wastewater dilution treatment, n is the number of the irrigation to be applied in a particular season and C_P is the sum of the actual COD concentrations for the previous irrigations applied in the season. This continuous adjustment contributed to the fact that the mean actual COD was close to the target COD required for the different treatments for each of the four seasons. This means that the actual total COD in the water applied was similar to the ideal situation, *i.e.* if the actual COD in the diluted winery wastewater had been exactly the same as the target for each irrigation.

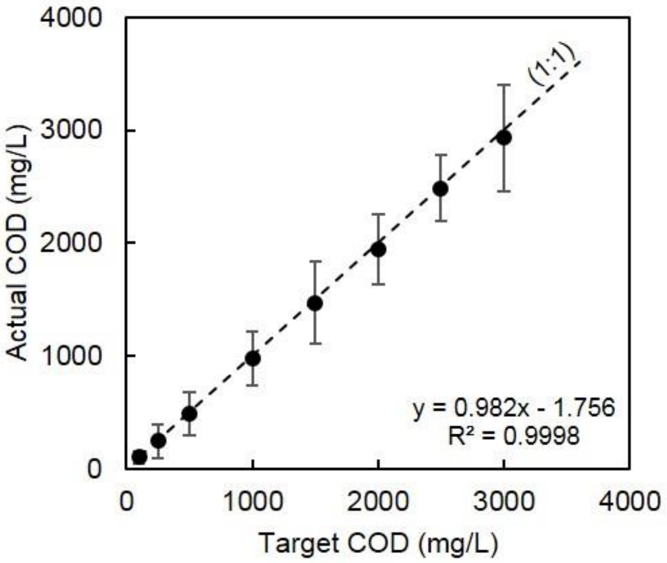


Figure 2.12. Relationship between mean actual chemical oxygen demand (COD) in diluted winery wastewater applied to grapevines and the target COD levels of treatments T2 to T9 during the 2010/11 season. Vertical bars indicate standard deviation (n = 6).

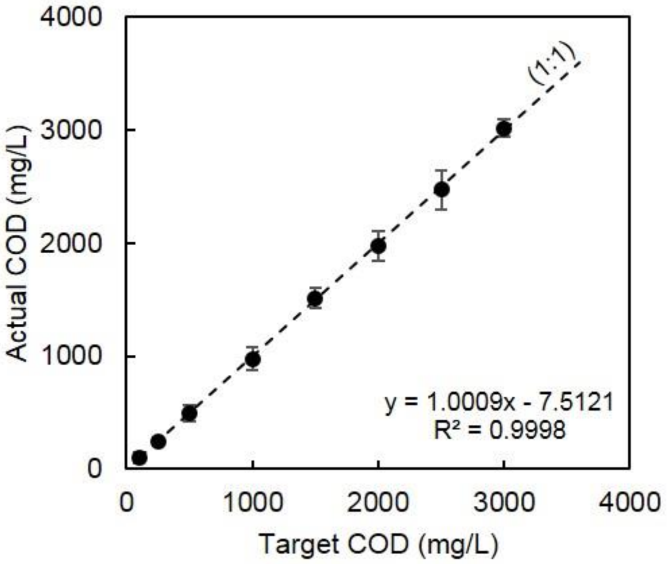


Figure 2.13. Relationship between mean actual chemical oxygen demand (COD) in diluted winery wastewater applied to grapevines and the target COD levels of treatments T2 to T9 during the 2011/12 season. Vertical bars indicate standard deviation (n = 6).

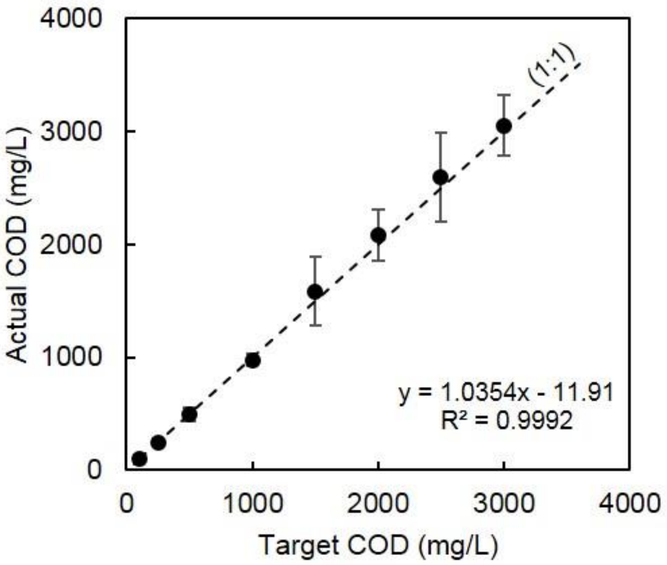


Figure 2.14. Relationship between mean actual chemical oxygen demand (COD) in diluted winery wastewater applied to grapevines and the target COD levels of treatments T2 to T9 during the 2012/13 season. Vertical bars indicate standard deviation (n = 6).

Before the field work commenced, one of the major concerns was the efficiency of the mixing process in the tanks. On 12 April 2010, the variation in COD was measured as the irrigation progressed. The duration of the irrigations varied between 4.5 hours and 5.5 hours. Hence, water samples were collected one hour, 2.5 hours and 5 hours after the irrigations started. Water was sampled in triplicate only at the T4 and T9 tanks. Analyses of the water showed that the COD levels remained reasonably constant as the irrigation progressed, irrespective of the COD concentration (Fig. 2.15). Furthermore, it indicated that the concentrations of the diluted wastewater within the tanks were fairly homogeneous, and that effective mixing occurred while the tanks were being filled.

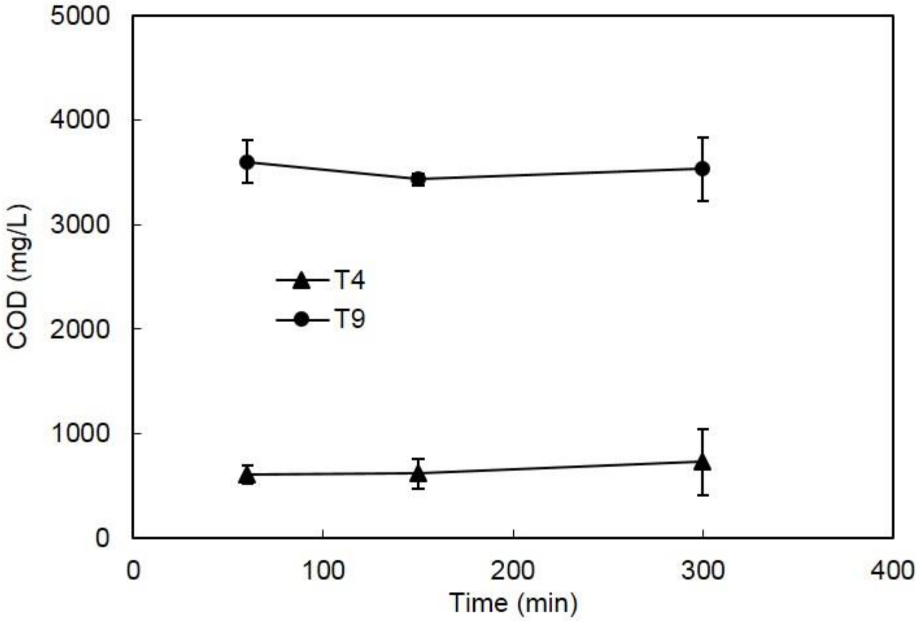


Figure 2.15. Temporal variation in chemical oxygen demand (COD) in diluted winery wastewater pumped from the mixing tanks for the irrigation of grapevines of two respective treatments, *i.e.* T4 and T9. Vertical bars indicate standard deviation (n = 3).

2.3.3. Deposits in irrigation lines

2.3.3.1. Non-destructive method

In the first season, material deposited increased with increasing concentration of the winery wastewater as indicated by increasing COD level and decreasing dilution (Fig. 2.16). However, this increase was non-linear, and the highest deposits occurred where the target COD concentration was 2000 mg/L (T7). The high standard deviation from the means was the result of variation between treatment replications. In the second and third seasons, this variability between treatment replications increased to the extent that the amount of deposits could not be related to level of COD (data not shown). A possible explanation for the inconsistency between treatment replications could be that the pipe containing the glass slides did not drain completely following irrigations. Between irrigations, algae growth could have caused additional deposits on the slides. The foregoing indicated that the methodology was not accurate enough to detect differences in the small amount of material on the glass slides.

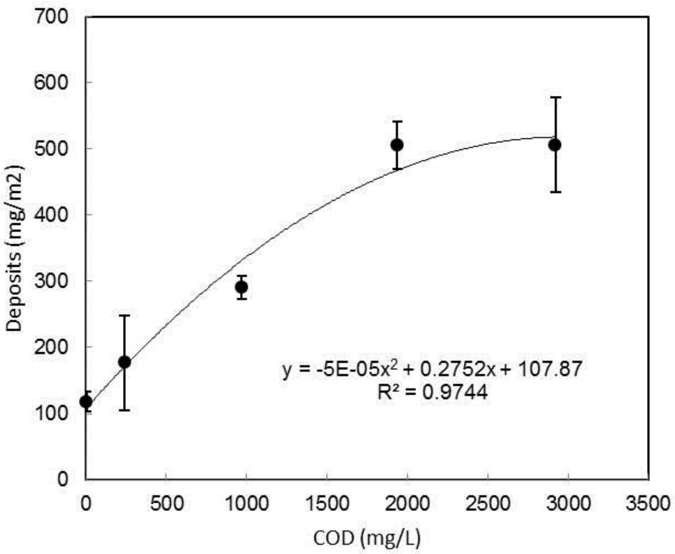


Figure 2.16. Effect of chemical oxygen demand (COD) level in diluted winery wastewater on foreign material deposits accumulated in the 2010/11 season on the inside of the irrigation lines. Vertical bars indicate standard deviation (n = 3).

2.3.3.2. Destructive method

After four seasons, material deposits in the pipe samples increased with a decrease in the level of dilution of the wastewater (Fig. 2.17). Similar to results obtained with the glass slides, the deposit increase was also non-linear. The non-linear increase of deposits to level of COD indicated that possibly some constituent(s) in the diluted wastewater suppressed deposits in the irrigation lines over the three years.

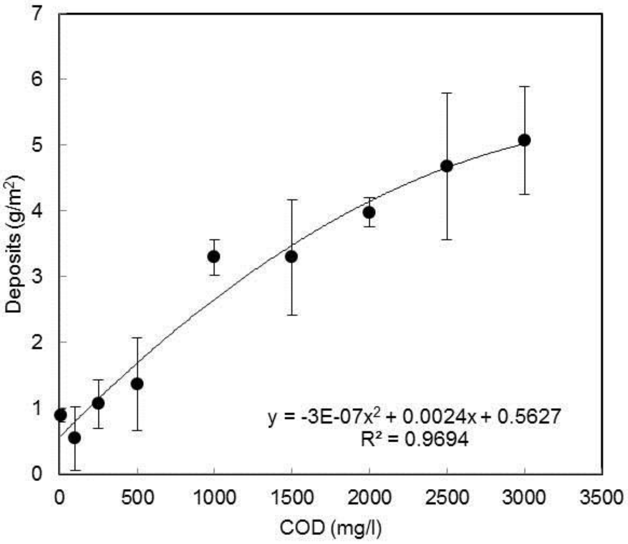


Figure 2.17. Effect of chemical oxygen demand (COD) level in diluted winery wastewater on foreign material deposits accumulated over four seasons, *i.e.* 2009/10 to 2012/13 on the inside of the irrigation lines. Vertical bars indicate standard deviation (n = 3).

Iron and manganese oxides are known to form deposits which can cause clogging of irrigation systems. Perusal of the data revealed that the deposits increased with the mean Fe^{2+} concentration in the diluted waters (Refer to Appendix 2.18). Furthermore, visual observation revealed that the deposits on the insides of the irrigation lines had reddish, brown colour. However, the deposit increase with increasing Fe^{2+} was also non-linear (Fig. 2.18), which suggested that the contribution of Fe^{2+} to the deposits was suppressed in the less diluted waters. Since the water pH decreased as the level of wastewater dilution decreased (Refer to Appendix 2.1), the foregoing suggested that the lower pH levels might have suppressed Fe-oxide formation.

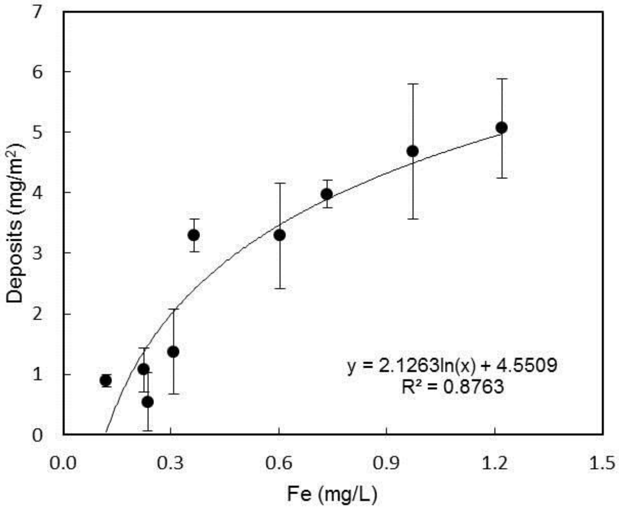


Figure 2.18. Effect of iron (Fe^{2+}) concentration in diluted winery wastewater on foreign material deposits accumulated over four seasons, *i.e.* 2009/10 to 2012/13 on the inside of the irrigation lines. Vertical bars indicate standard deviation (n = 3).

2.4. CONCLUSIONS

As previous studies used artificial “winery wastewater”, mostly on a laboratory scale, this study was the first where wastewater from a commercial winery was diluted with raw river water to obtain a range of COD levels for vineyard irrigation at the field level. The relatively simple mix and distribution facility allowed dilution of large volumes of winery wastewater to a range of COD levels required for irrigating grapevines in a field trial. After initial practical problems and sources of error were eliminated, accuracy in terms of treatment application of the target COD concentrations was acceptable. Measuring COD concentrations while the irrigation water was being pumped from the tanks confirmed that the concentrations of diluted winery wastewater within the tanks were homogeneous, and that effective mixing had taken place while the tanks were being filled. By adjusting the COD level for the next irrigation according to the total COD applied *via* the preceding irrigations in a particular season, close agreement was obtained between the mean actual COD and the target values. When doing COD measurements, results will be more reliable if the oxidation time is standardized at two hours, irrespective of the level of COD in the water.

Measuring cane mass at pruning on a per plot basis before the field work commenced indicated that the vegetative growth varied naturally across the experimental vineyard. However, this allowed the treatment replications to be allocated accordingly.

The formation of deposits on the inside of the irrigation lines appeared to be a function of the organic matter and Fe^{2+} as affected by pH in the diluted winery wastewater. In practical terms, the deposited material formed a relatively thin layer, which implies that it could not have caused significant clogging of the irrigation lines or micro-sprinklers after three years. Therefore, negative effects on grapevine growth and/or yield due to increased clogging of micro-sprinklers would not be expected under the prevailing conditions. Since drippers have narrow flow paths and/or small orifices, they are more susceptible to clogging than micro-sprinklers. Therefore, drip irrigation systems should be avoided where winery wastewater is to be re-cycled for irrigation of crops. The destructive method provided more reliable results than monitoring annual deposits with the non-destructive method. More reliable and accurate monitoring of deposit formation in irrigation lines by means of non-destructive methods requires further investigation.

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CHAPTER 3: WATER QUALITY, IRRIGATION VOLUMES AND AMOUNT OF ELEMENTS APPLIED *VIA* WASTEWATER IRRIGATION

3.1. INTRODUCTION

Wineries produce large volumes of low quality wastewater, particularly during harvest. Reports on the actual volumes of wastewater generated by wineries are extremely limited. However, it is estimated that medium to large wineries generate more than 15 000 m^3 of wastewater annually, whereas small wineries generate less than 15 000 m^3 annually (Van Schoor, 2005 and references therein). Australian wineries generate about 5 m^3 of wastewater per tonne of grapes crushed (Chapman *et al.*, 1995). Crushing approximately 50 000 tonnes of grapes annually generates about 175 000 m^3 of wastewater at the Berri estates' winery in the Riverland region of South Australia (Anonymous, 2010). Hence, their wastewater generation amounts to *c.* 3.5 m^3 per tonne of grapes. It can be estimated that the Lutzville Vineyards' winery generates about 1.1 m^3 of wastewater per tonne of grapes crushed. However, this relatively low value is misleading, since 50% of the wastewater is presumably lost to evaporation (Kriel, 2008).

Winery wastewater contains high levels of potassium (K^+) and sodium (Na^+) (Laurenson *et al.*, 2012). Although various parameters may be used to evaluate winery wastewater, chemical oxygen demand (COD), pH, sodium adsorption ratio (SAR), electrical conductivity of the irrigation water (EC_{iw}), chloride (Cl^-), K^+ and Na^+ are considered to be important. A survey carried out to evaluate winery wastewater generated by the South African wine industry revealed that the water quality parameters vary substantially between wineries (Mulidzi *et al*., 2009). The variation in water quality parameters also occur in wastewater produced by wineries all over the world (Conradie *et al.*, 2014 and references therein). Furthermore, a strong seasonal variation in winery wastewater quality was observed in the South African industry (Mulidzi *et al*., 2009). A similar seasonal trend was reported for winery wastewater in Australia (Arienzo *et al*., 2009a). These trends were confirmed where effluents of two wineries were monitored frequently (Sheridan *et al.*, 2011). Considering the legal requirements for irrigation water quality in South Africa, results of the survey confirmed that the majority of South African wineries are not able to irrigate crops beneficially as part of the General Authorisations for irrigation with winery wastewater unless the water is first subjected to an effective form of pre-treatment, or unless there is relaxation of the General Authorisations (Department of Water Affairs & Forestry, 1996; Department of Water Affairs, 2013).

International requirements, as well as national legislation, are putting pressure on wine producers regarding the responsible management of their wastewater, which may have large-scale detrimental impact on the environment. In the Western Cape, most vineyards need irrigation. Therefore, the ideal situation would be to implement sustainable use of winery wastewater for wine grape irrigation by adding winery wastewater to existing irrigation water resources. This practice has already been performed by various wineries for decades. Until now, the impact of this practice has, however, not been studied comprehensively. Currently, the Department of Water and Sanitation is drafting a new Authorisation for wineries. Depending on the permitted water quality limits and volumes stipulated by these authorisations, adding winery wastewater to current irrigation water may well become a more viable practice in the future. Re-using winery wastewater in this way will be beneficial, particularly in situations where water shortages occur.

The objectives of the study were (i) to assess the quality of wastewater produced by a commercial winery, (ii) to determine whether this quality could be improved by dilution with raw river water for irrigation and (iii) to quantify the amount of plant nutrients applied *via* irrigation with the diluted water.

3.2. MATERIALS AND METHODS

3.2.1. Irrigation volumes applied

Details of the experimental layout and viticultural aspects are presented in Chapter 2. Irrigation with winery wastewater diluted to 100 mg/L, 250 mg/L, 500 mg/L, 1000 mg/L, 1500 mg/L, 2000 mg/L, 2500 mg/L and 3000 mg/L COD with river water was compared to a control irrigated with river water (Appendix 2.1). Refer to Chapter 2 for more details regarding the irrigation infrastructure, as well as the dilution procedures. The wastewater irrigation treatments were applied from mid-February when high volumes of wastewater usually become available when the harvest period begins in the Breede River valley. After each wastewater irrigation application, grapevines were also irrigated with river water to flush the irrigation pipes. Grapevines were generally irrigated at *c.* 50% plant available water (PAW) depletion to prevent excessive vegetative growth and yield reduction. Irrigation had to be applied every two weeks to maintain this PAW depletion level. Irrigation was terminated either in mid-April or the beginning of May each year, when the wastewater volumes decreased and/or the first winter rains began. Irrigation was applied by means of micro-sprinklers in order to apply larger volumes of water than what the case would have been with drip irrigation. Water meters were used to monitor the irrigation volumes applied to each treatment. Grapevines of all treatments, including those of the control, received the same volume of water per irrigation.

3.2.2. Water quality

The annual wastewater quality dynamics of the winery where the wastewater for the field experiment was sourced, was determined. For this purpose, a 2 L sample of undiluted treated winery wastewater was abstracted from the collection pit at the winery on a monthly basis from January 2010 until mid-December 2013. The COD in the undiluted winery wastewater was determined as described in Chapter 2. The samples were also analysed by a commercial laboratory (Bemlab, Strand) for pH, EC, ammonium nitrogen (NH_4^+-N), nitrate nitrogen (NO_3^--N), phosphorus (P), calcium (Ca^{2+}), magnesium (Mg^{2+}), K^+, Na^+, Cl^-, bicarbonate (HCO_3^-), sulphate (SO_4^{2-}), boron (B^{3+}), iron (Fe^{2+}), manganese (Mn^{2+}), copper (Cu^{2+}), zinc (Zn^{2+}), fluoride (F^-) as well as heavy metals according to methods described by Clesceri *et al.* (1998). The potassium adsorption ratio (PAR) was calculated as follows:

$$PAR = K^+ \div [(Ca^{2+} + Mg^{2+}) \div 2]^{0.5} \qquad \text{(Eq. 3.1)}$$

where K^+ is the potassium concentration (mg/L) divided by the molecular mass, *i.e.* 39 g/mol, Ca^{2+} is the calcium concentration (mg/L) divided by the equivalent molecular mass, *i.e.* 20 g/mol and Mg^{2+} is the magnesium concentration (mg/L) divided by the equivalent molecular mass, *i.e.* 12.15 g/mol. Similarly, the sodium adsorption ratio (SAR) was calculated as follows:

$$SAR = Na^+ \div [(Ca^{2+} + Mg^{2+}) \div 2]^{0.5} \qquad \text{(Eq. 3.2)}$$

where Na^+ is the sodium concentration (mg/L) divided by the molecular mass, *i.e.* 23 g/mol. The NH_4^+-N and NO_3^--N concentrations were summed to obtain the total nitrogen (Total-N). No N determinations were carried out in the 2009/10 season.

Approximately one hour after a wastewater irrigation started, a 500 mL water sample was collected from each of the eight dilution tanks to verify actual COD levels obtained for each of the eight dilution treatments. A 500 mL sample of the river water was collected at the same time. Samples of the undiluted winery wastewater from the stock dam and river, as well as the samples from the eight dilution tanks were analysed by a commercial laboratory as mentioned above. Assessment of the microbial status of the winery wastewater, as well the diluted waters, was beyond the scope of the study.

3.2.3. Amount of elements applied

For the diluted wastewater treatments, *i.e.* T2 to T9, the amount of wastewater applied was converted from mm to L per ha as follows:

$$V = I \times 10^4 \qquad \text{(Eq. 3.3)}$$

where I is the amount of irrigation applied (mm) and 10^4 is the factor used to convert depth of water (mm) to volume (L) per hectare (1 mm = 10 m^3 per ha = 10^4 L per ha).

For each treatment, the element concentrations in the diluted wastewater were used to calculate the amounts of elements added to the soil per irrigation per hectare as follows:

$$m = V \times C_e \qquad \text{(Eq. 3.4)}$$

where m is amount of element (mg/ha), V is the volume of water applied per hectare (L/ha) and C_e is the element concentration (mg/L) in the irrigation water.

In addition, the contribution of the elements deposited by the river water was taken into account. For T1, *i.e.* the river water control treatment, the same procedure was followed. The amount of element in milligram per hectare was converted to kilogram per hectare (M) as follows:

$$M = m \div 10^6 \qquad \text{(Eq. 3.5)}$$

The amount of elements applied per irrigation were summed to obtain the seasonal applications.

3.3. RESULTS AND DISCUSSION

3.3.1. Irrigation volumes applied

3.3.1.1. Amounts per irrigation: The amounts of diluted winery wastewater are presented in Appendix 1.1. A full tank of diluted winery wastewater applied *c.* 41 mm irrigation on the three replication plots of each treatment. Grapevines of all treatments received the same mean volume of wastewater per irrigation when a full tank of wastewater was applied to the plots. Due to low COD levels in the winery wastewater in the middle of April in the 2010/11 season, T3 to T9 had to be applied in two irrigations on consecutive days to apply the required COD. The average COD applied on these two days was calculated. Due to low COD levels in the winery wastewater, as well as limited available water, only one third of a tank of diluted water could be applied on 2 May in the 2011/12 season. This data was therefore not considered for the calculation of the average mean volume of wastewater applied per irrigation.

Amounts of river water applied per irrigation are presented in Appendix 1.2. Grapevines of the diluted wastewater treatments received on average 25 mm, 14 mm and 8 mm of river water per irrigation in 2010/11, 2011/12 and 2012/13, respectively. The amount of river water applied following wastewater irrigations was considerably less in 2012/13 than in previous years, since it was decided to follow a continuous deficit irrigation strategy to minimise drainage losses, and to curtail possible excessive vegetative growth. The total amounts per irrigation, *i.e.* wastewater plus river water, are presented in Appendix 1.3.

3.3.1.2. Seasonal amounts: Total seasonal amounts of wastewater applied to the diluted wastewater treatments are presented in Appendix 1.4. On average, seasonal wastewater irrigation amounted to 247 mm, 177 mm and 246 mm in 2010/11, 2011/12 and in 2012/13, respectively. The total seasonal amounts of river water applied to grapevines of all treatments are presented in Appendix 1.5. On average, seasonal river water amounted to 152 mm in 2010/11, 70 mm in 2011/12 and 48 mm in 2012/13. The total seasonal amounts of irrigation water, *i.e.* wastewater plus river water, applied to grapevines of all treatments are presented in Appendix 1.6. On average, the total amounts of irrigation water applied to grapevines amounted to 398 mm in 2010/11, 247 mm in 2011/12 and 294 mm in 2012/13.

3.3.2. Water quality

3.3.2.1 Undiluted winery wastewater

3.3.2.1.1 Annual dynamics of the undiluted winery wastewater quality

pH: The annual mean monthly pH in the undiluted wastewater ranged from 4.2 to 6.8 (Fig. 3.1). The narrow pH range was most likely due to the addition of lime to the wastewater by the particular winery. The pH variation was between values of 3 to 12 previously reported for winery wastewater (Mosse *et al.*, 2011 and references therein). Likewise, the undiluted wastewater pH was within the range of 3.5 to 7.9 according to a more recent study (Conradie *et al.*, 2014 and references therein). The pH levels were also below the recommended pH for irrigation water, which ranges from 6.5 to 8.4 (Department of Water Affairs & Forestry, 1996; Howell & Myburgh, 2013). According to the General Authorisations of 2013, up to 500 m^3 of wastewater may be irrigated on any given day provided that the pH is between 6 and 9 (Department of Water Affairs, 2013). In general, pH in the undiluted winery wastewater was below these norms (Fig. 3.1). Since the pH in the undiluted winery wastewater was lower than the limits prescribed by the General Authorisations, the undiluted water would not be suitable for irrigation without treatment.

The pH in the undiluted winery wastewater tended to be lower during harvest, *i.e.* from February to May, than in the rest of the year (Fig. 3.1). In annual dynamics monitored at two wineries in Stellenbosch, winery wastewater pH was also lower during harvest (Sheridan *et al.*, 2011). Similar results were reported by Kumar *et al.* (2006). The lower pH was probably due to the organic acids in grapes (Mosse *et al.*, 2011) which could have spilled into the washwater during the winemaking process. Since the pH in grape juice and wine ranges from 3 to 4 (Sheridan *et al.*, 2011), juice and wine spills could also have reduced the wastewater pH. In annual dynamics monitored at two wineries in Stellenbosch, it was also observed that the winery wastewater pH was lower during harvest which were ascribed to low pH in grape juice and wine, as well as wine handling operations (Sheridan *et al.*, 2011). Furthermore, the ethanol in the wine is degraded to acetic acid, which further could reduce the pH. In a study observing the composition of winery wastewater from ten different wineries across South Africa, pH in winery wastewater during harvest was frequently below 4 (Mulidzi *et al.*, 2009).

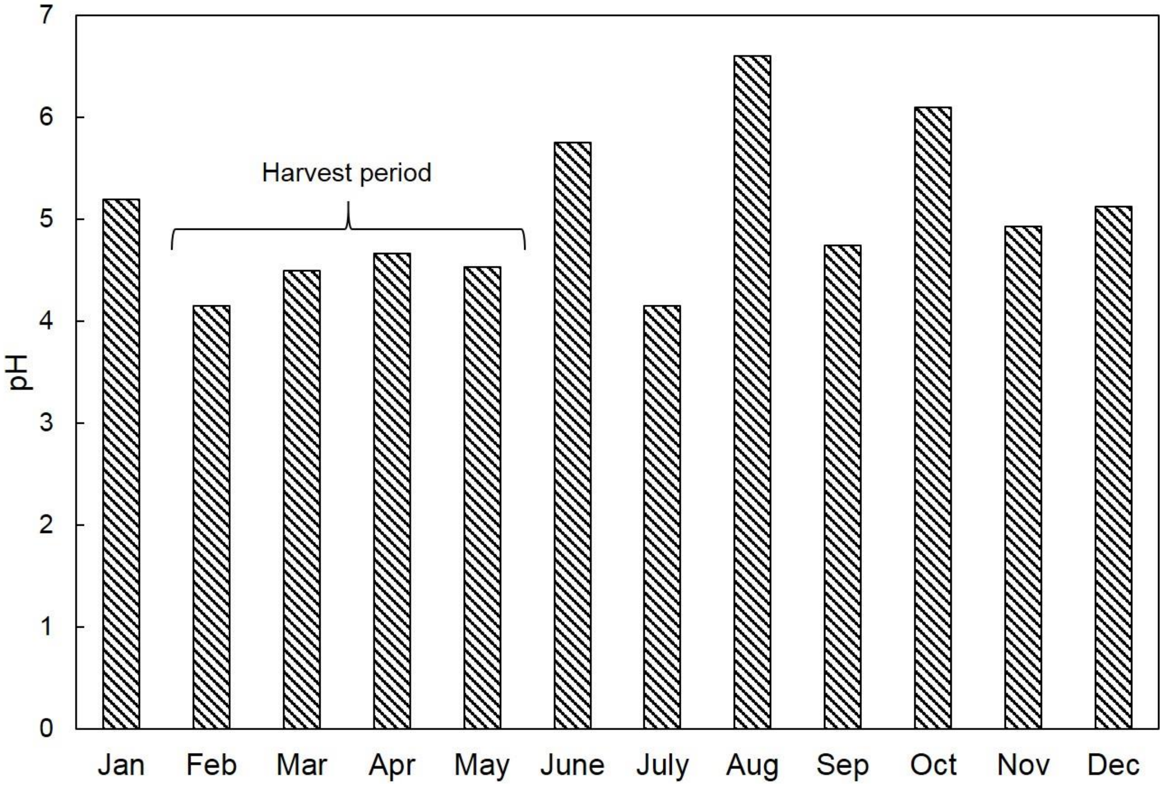

Figure 3.1. Mean monthly pH measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

EC: The annual mean monthly EC in the undiluted wastewater from the collection pit ranged from 0.68 dS/m to 2.15 dS/m (Fig. 3.2). The EC variation was similar to values of 0.8 dS/m to 3.1 dS/m (Mosse *et al.*, 2011 and references therein). However, the variation was wider than the 1.3 dS/m to 1.6 dS/m reported for winery wastewater in South Africa (Laurenson *et al.*, 2012 and references therein). Although the lower limit was comparable to 0.44 dS/m for undiluted wastewater reported by Mulidzi *et al.* (2009), the upper limit of 25.7 dS/m observed in that study was appreciably higher compared to 2.15 dS/m in the current study. With the exception of August, EC exceeded the critical value of 0.75 dS/m which is the salinity threshold for water used for grapevine irrigation (Van Zyl, 1981; Myburgh, 2012). With regard to the General Authorisations of 2013 (Department of Water Affairs, 2013), up to 500 m^3 of wastewater may be irrigated on any given day provided that the EC_{iw} is less than 2 dS/m. Since the EC in the undiluted winery wastewater was lower than the limit prescribed by the General Authorisations, the EC of the undiluted water would render it suitable for irrigation without treatment to reduce the salinity level. Although the EC during the harvest period was lower than the norm prescribed where up to 500 m^3 is irrigated per day, it was higher than the prescribed norms of 0.70 dS/m and 1.50 dS/m where 2000 m^3 of wastewater is irrigated on any given day (Department of Water Affairs, 2013).

The EC in the undiluted winery wastewater tended to increase from the start of harvest in February and reached a maximum in May, followed by a decline to a minimum in August (Fig. 3.2). This increase in EC probably originated from cleaning agents used in the winery, as well as K^+ in grape lees and spillages from the grape fermentation process.

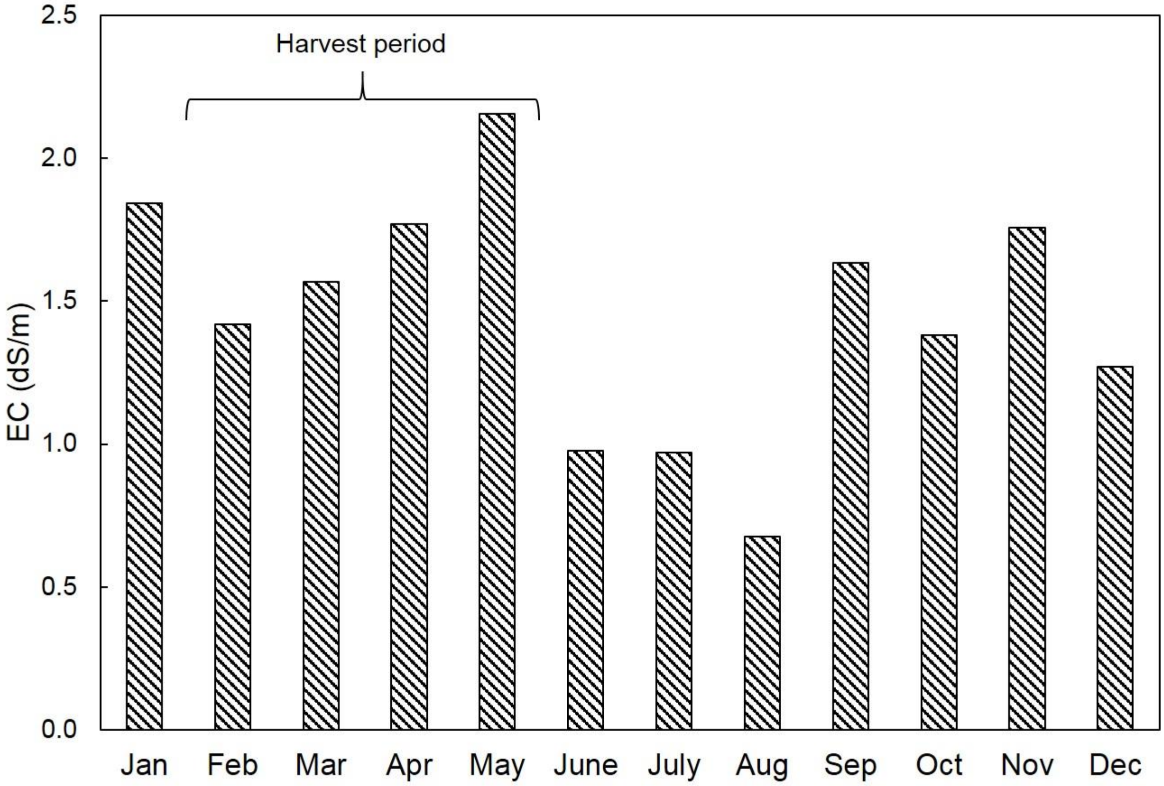

Figure 3.2. Mean monthly electrical conductivity (EC) measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

COD: The annual mean monthly COD levels in the undiluted wastewater ranged from 1815 mg/L to 13286 mg/L (Fig. 3.3), which fall between the values of 320 mg/L to 12000 mg/L previously reported for winery wastewater in South Africa (Mosse *et al.*, 2011 and references therein). Likewise, COD levels were within the range of 340 mg/L to 49105 mg/L according to a more recent study (Conradie *et al.*, 2014 and references therein). Up to 50 m^3, 500 m^3 and 2000 m^3 of wastewater may be irrigated on any given day provided that the COD is lower than 5000 mg/L, 400 mg/L and 40 mg/L, respectively (Department of Water Affairs, 2013). In general, COD levels in the undiluted winery wastewater were higher than these norms. Since the COD levels in the undiluted winery wastewater was higher than the limits prescribed by the General Authorisations, particularly in the harvest period, the undiluted water would not be suitable for irrigation without treatment to reduce the COD.

The mean monthly COD level in the undiluted winery wastewater increased from January and was the highest during peak harvest in March, exceeding 10000 mg/L (Fig. 3.3). Reported COD values in a survey of ten different wineries across South Africa ranged from

3370 mg/L for a winery in Paarl to 47024 mg/L for a winery in the Olifants River region (Mulidzi *et al.*, 2009). Sheridan *et al.* (2011) reported much lower COD values for a cellar in Stellenbosch, which peaked at *c.* 3800 mg/L. Lower COD values tended to occur in the pre- and post-harvest periods. Similar findings were reported by Sheridan *et al.* (2011). Following the maximum COD levels at the peak of harvest, levels decreased until June. This decrease reflected the end of the peak harvesting period. In July, the COD level in the winery wastewater was high probably due to stabilisation of the wine, which can increase COD levels (Conradie *et al.*, 2014). The COD levels in the undiluted winery wastewater were low in August, September and October. Thereafter, the COD levels increased in the undiluted wastewater, and this increase can be attributed to preparations in the cellar for the forthcoming harvest period.

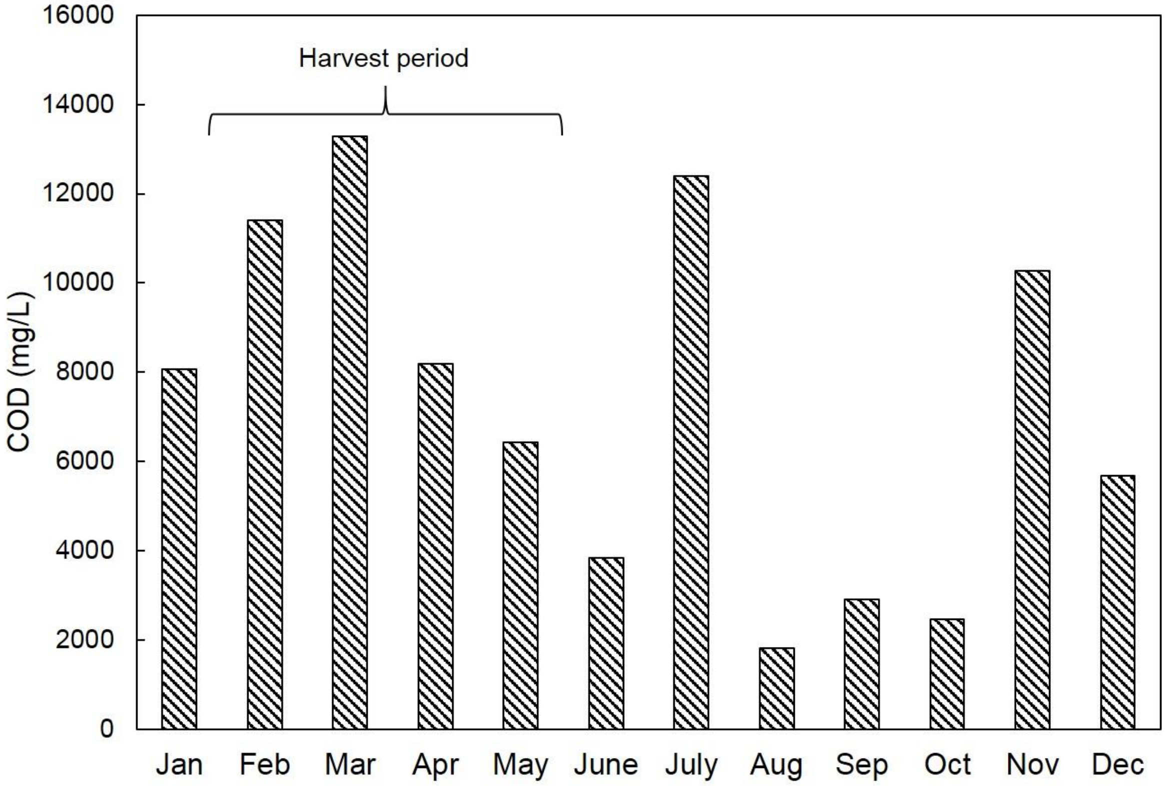

Figure 3.3. Mean monthly chemical oxygen demand (COD) measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

Potassium: The annual mean monthly K^+ in the undiluted wastewater ranged from 44 mg/L to 506 mg/L (Fig. 3.4). Mulidzi *et al*. (2009) tentatively classed 200 mg/L as high for K^+. Using this norm, the K^+ levels in the undiluted wastewater from the collection pit was high from January to May. The K^+ levels were higher than the range of 29 mg/L to 353 mg/L previously reported for winery wastewater (Mosse *et al.*, 2011 and references therein). Likewise, the undiluted wastewater K^+ was higher than the range of 20 mg/L to 220 mg/L reported for a study carried out at two wineries near Stellenbosch (Sheridan *et al.*, 2011).

The K^+ levels in the undiluted winery wastewater increased substantially from the beginning of harvest in early February to May (Fig. 3.4.). The higher K^+ probably originated from cleaning agents used in the winery, as well as grape lees and spillage from the grape fermentation process (Arienzo *et al.*, 2009a; Laurenson *et al.*, 2012). The increase in K^+ during the harvest period at this particular winery was similar to findings reported by Sheridan *et al.* (2011). In a survey on the composition of winery wastewater, reported values for a winery in the Orange River region ranged from 49 mg/L in January to 296 mg/L in March, and values were high for most of the sampling period (Mulidzi *et al.*, 2009). Excessively high values of 4119 mg/l also occurred during March at the winery in the Olifants River region.

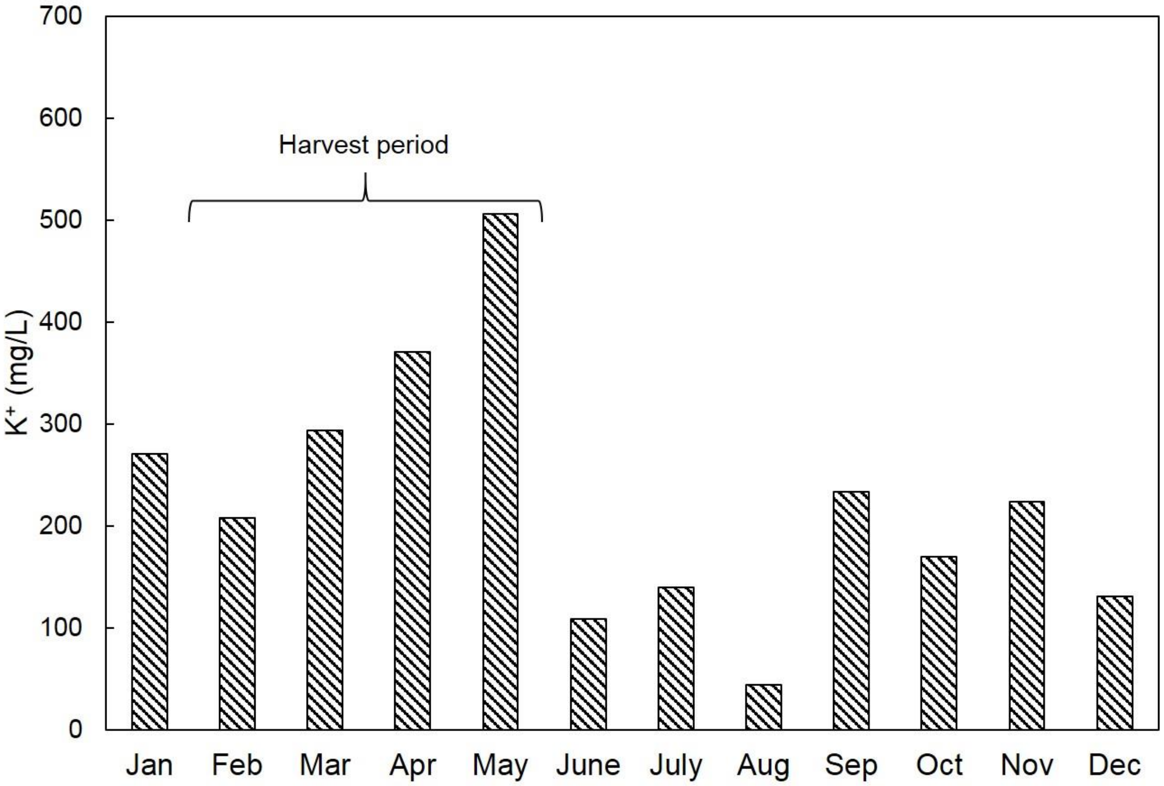

Figure 3.4. Mean monthly potassium (K^+) concentration measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

Potassium adsorption ratio: The annual mean monthly PAR in the undiluted wastewater ranged from 1.7 to 10.8 (Fig. 3.5). The PAR levels were higher than the values of 2.1 to 3.2 that were reported for winery wastewater, particularly during the harvest period (Laurenson *et al.*, 2012 and references therein). However, for a winery in Australia it was previously reported that PAR values ranged from 3.7 to 43.0 (Arienzo *et al.*, 2009b). As the K^+ levels increased during harvest, probably due to K^+ in cleaning agents used in the winery, grape lees and spillage from the grape fermentation process (Arienzo *et al.*, 2009a; Laurenson *et al.*, 2012), the PAR levels in the undiluted winery wastewater increased substantially from the beginning of harvest in early February to May (Fig. 3.5).

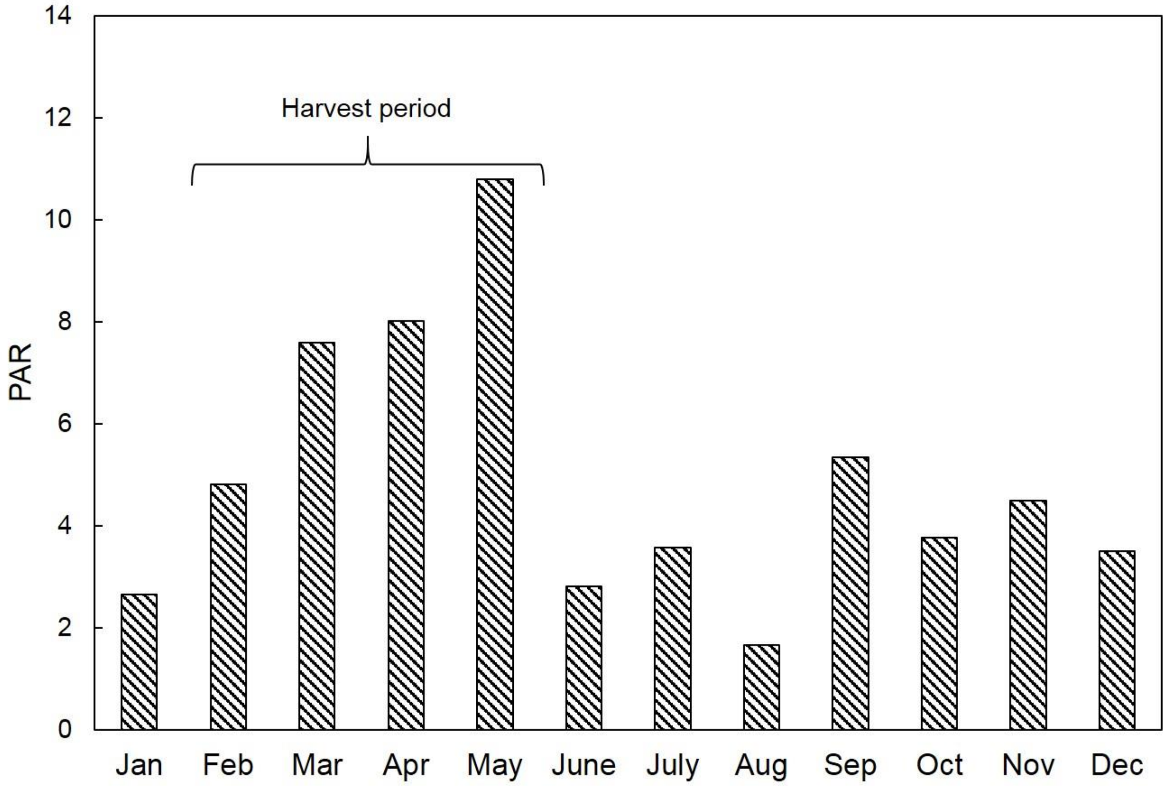

Figure 3.5. Mean monthly potassium adsorption ratio (PAR) measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

Sodium: The annual mean monthly Na^+ levels in the undiluted wastewater varied from 76 mg/L to 224 mg/L (Fig. 3.6), and fell in the range of 7 mg/L and 470 mg/L previously reported for Na^+ in winery wastewater (Mosse *et al.*, 2011 and references therein). Since grapevines are considered moderately sensitive to foliar injury from Na^+, a concentration of 115 mg/L is recommended as the upper threshold when overhead irrigation is applied (Department of Water Affairs & Forestry, 1996; Howell & Myburgh, 2013). It is important to note that Na^+ levels in the undiluted winery wastewater generally exceeded this threshold from September to November. As in the case of the K^+, Na^+ levels increased from February to May (Fig. 3.6), and were highest in October and November. This is probably related to cleaning actions within the winery before the harvest period commences.

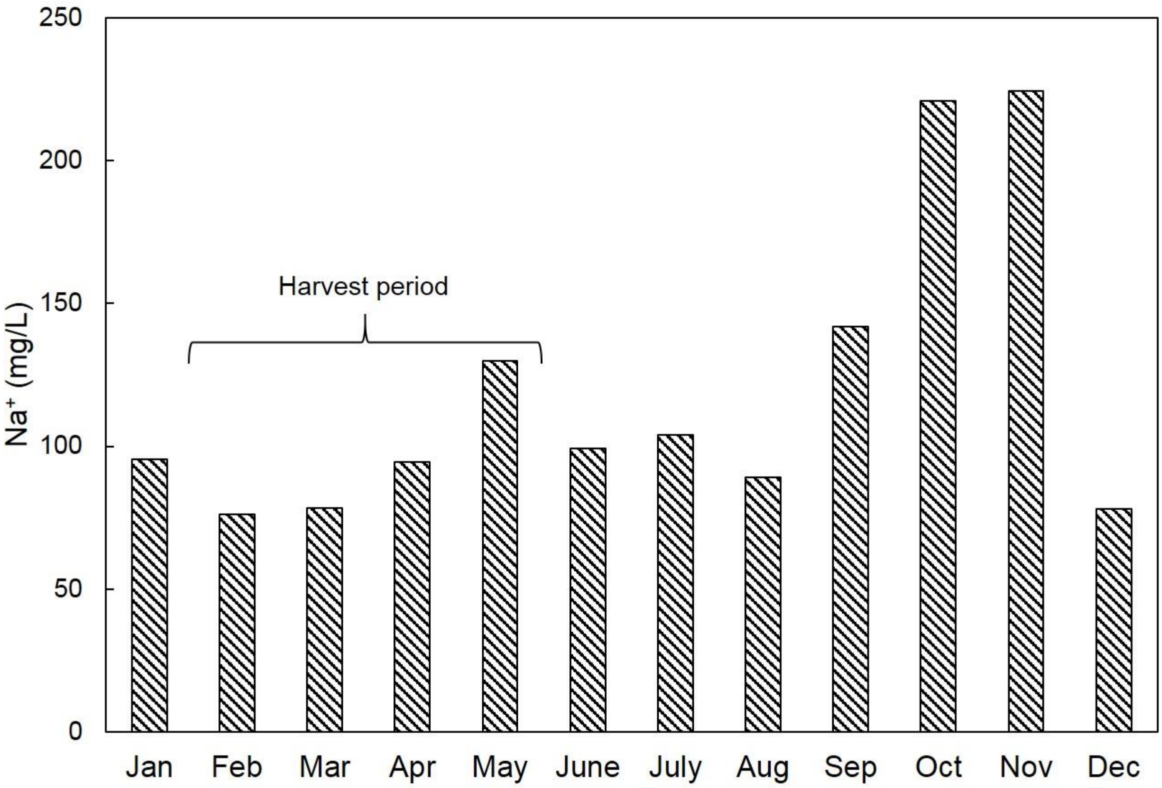

Figure 3.6. Mean monthly sodium (Na^+) concentration measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

Sodium adsorption ratio: The annual mean monthly SAR in the undiluted wastewater from the collection pit ranged from 2.4 to 9.0 (Fig. 3.7), which fall within the the SAR variation of 0.3 to 33.1 reported for winery wastewater (Mulidzi *et al.*, 2009). However, the SAR in the undiluted wastewater from the collection pit fell outside the range of 3.5 to 7.9 reported in a more recent study (Conradie *et al.*, 2014 and references therein). The SAR was generally within acceptable limits for the irrigation of grapevines, *i.e*. < *c.* 10 (Richards, 1954; Myburgh, 2012). With regard to the General Authorisations of 2013, up to 500 m^3 of wastewater may be irrigated on any given day provided that the SAR is less than 5 (Department of Water Affairs, 2013). In general, SAR in the undiluted winery wastewater was below these norms. Since the SAR in the undiluted winery wastewater was lower than the limits prescribed by the General Authorisations, the undiluted water would be suitable for irrigation without treatment to reduce the sodicity hazard. The SAR increased gradually from January to September, with high values in October and November which was in agreement with the annual Na^+ dynamics.

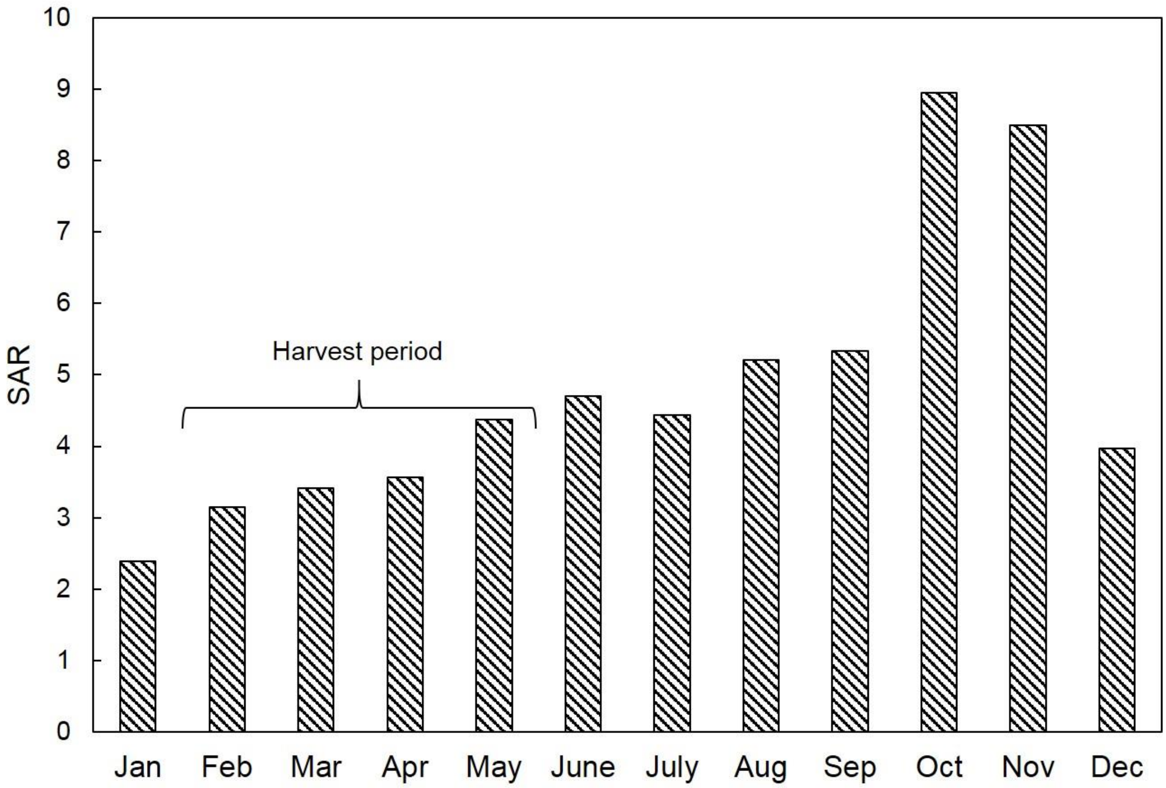

Figure 3.7. Mean monthly sodium adsorption ratio (SAR) measured in undiluted winery wastewater at the Goudini winery (data are means for 2010, 2011, 2012 and 2013).

3.3.2.1.2 Correlations between the different water quality parameters for the undiluted winery wastewater obtained from the collection pit at the winery

During the planning phase of the project, it was decided by industry representatives that the dilution treatments had to be applied in terms of COD concentration. However, there was doubt as to whether the COD level *per se* would provide a realistic indication of the overall water quality, since other variables, *e.g.* pH, EC, K^+, Na^+ and SAR also play an important role. Results of the study clearly showed that the pH (Fig. 3.8A) and EC in the undiluted winery wastewater could not be related to COD level (Fig. 3.8B). There was also no relationship between K^+ concentration and COD level in the undiluted winery wastewater (Fig. 3.8C). Furthermore, at a specific COD level, the K^+ concentration in the undiluted winery wastewater differed substantially. Consequently, there was also no relationship between PAR and COD (Fig. 3.8D). As in the case of the K^+, there was no relationship between Na^+ in the winery wastewater and level of COD (Fig. 3.8E). Similar to K^+, Na^+ concentration varied substantially at a specific COD level. The SAR was not related to COD level (Fig. 3.8F). Although it was decided that the dilution treatments had to be applied in terms of COD, COD level does not give an accurate indication of the other water quality variables. Taking all of the above-mentioned into consideration, it is clear that the COD level in the winery wastewater can not be used to predict K^+ and Na^+ concentrations in winery wastewater diluted to different levels of COD.

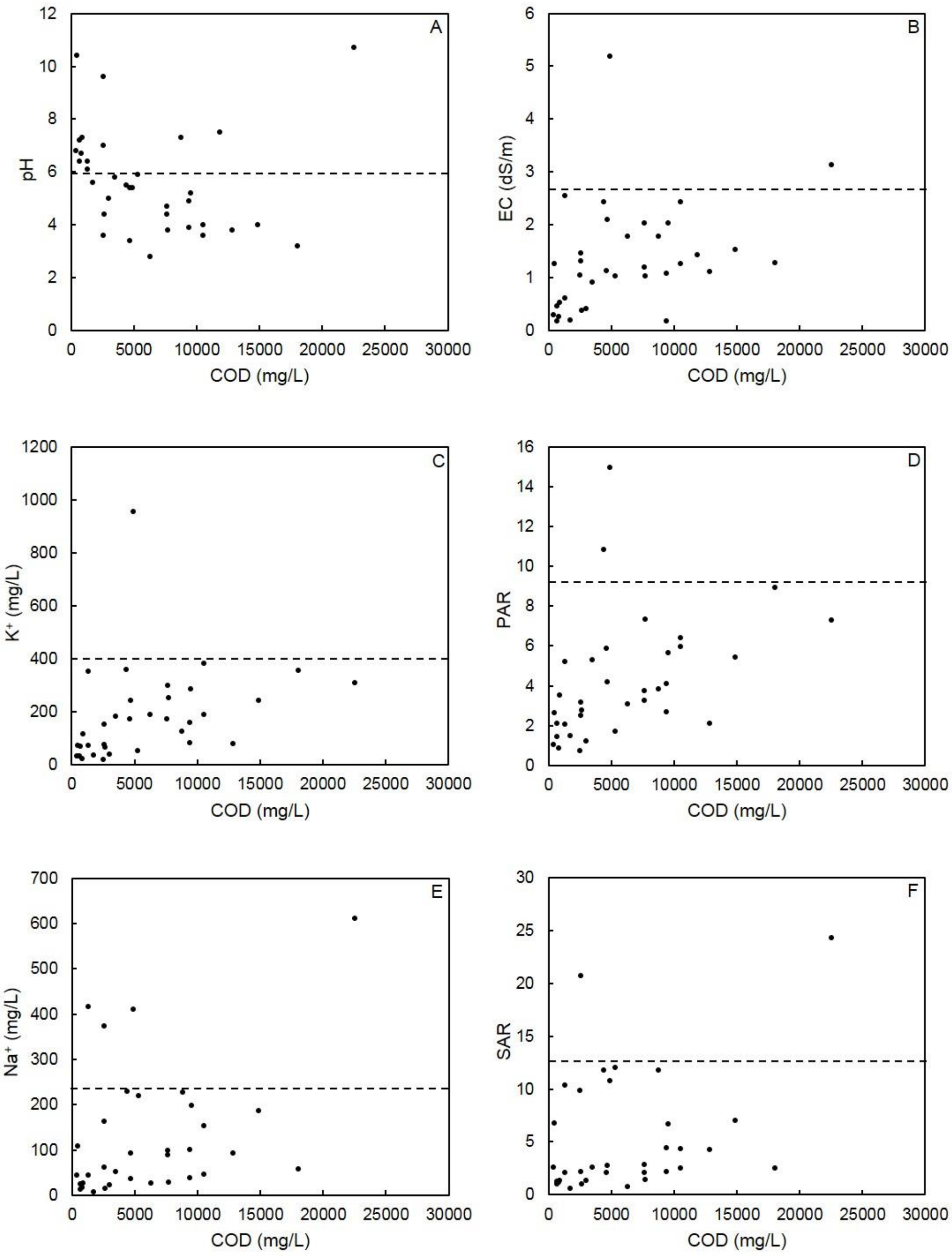

Fig. 3.8. Relationship between (A) pH, (B) electrical conductivity (EC), (C) potassium (K^+), (D) potassium adsorption ratio (PAR), (E) sodium (Na^+) and (F) sodium adsorption ratio (SAR) and chemical oxygen demand (COD) of undiluted winery wastewater abstracted from the collection pit at the Goudini winery over a four year period.

There was no relationship between Na$^+$ and K$^+$ in the undiluted winery wastewater (Fig. 3.9A). Although there was a strong correlation between EC and K$^+$ in the winery wastewater (Fig. 3.9B), the correlation between EC and Na$^+$ was not as good (Fig. 3.9C). The best correlation was obtained between EC and K$^+$ plus Na$^+$ concentration in the undiluted winery wastewater (Fig. 3.9D). These results indicated that the EC in the winery wastewater was strongly determined by the K$^+$ concentration. This was to be expected, since K$^+$ is usually the most abundant cation in winery wastewater. Furthermore, it was clear that the level of COD provided no indication of the salinity or sodicity hazard. Where irrigation is scheduled in such a way that that the organic matter is allowed to break down between irrigations, EC would be a more reliable indicator of the suitability for irrigation of vineyards and other crops than COD. The measurement of EC is also much quicker and less expensive than COD measurements, thereby making EC measurements more suitable for water quality assessment by wineries.

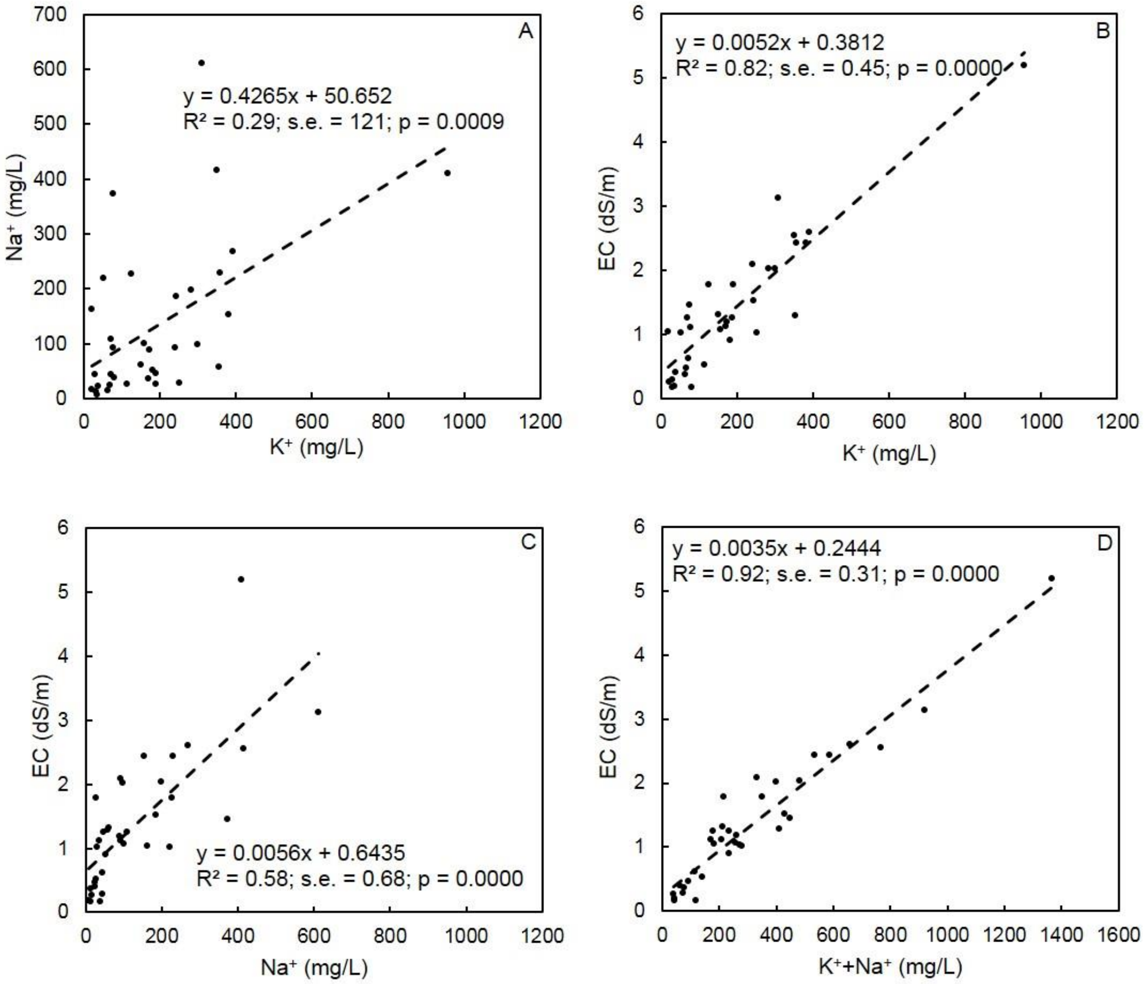

Fig. 3.9. Relationship between (A) sodium (Na$^+$), (B) electrical conductivity (EC) and potassium (K$^+$) and (C) EC and Na$^+$ and (D) EC and K$^+$ plus Na$^+$ of undiluted winery wastewater abstracted from the collection pit at the Goudini winery over a four year period.

3.3.2.2 Diluted winery wastewater

pH: During the four seasons, water pH tended to decrease with a decrease in the dilution of the wastewater, *i.e.* an increase in the COD level of the water (Appendix 2.1). The pH of the river water (T1) was generally the highest, whereas pH in winery wastewater diluted to COD levels of 1500 mg/L and higher were comparable to the undiluted winery wastewater, *i.e.* T5 to T9 (Appendix 2.1). In general, water pH levels tended to be below the recommended pH for irrigation water (Department of Water Affairs & Forestry, 1996; Howell & Myburgh, 2013), well as the as the General Authorisations of 2013 (Department of Water Affairs, 2013) as discussed in Section 3.3.2.1.1. Due to the low pH, problems with corrosion of metals and concrete used in irrigation systems and infrastructure can be expected when the irrigation water has a pH below 6.5 (Department of Water Affairs & Forestry, 1996). The diluted wastewater could also cause problems such as aluminium, manganese and heavy metals being mobilized to concentrations high enough to be toxic to grapevines if used for irrigation (Howell & Myburgh, 2013). The foregoing indicated that dilution of winery wastewater did not have a sufficient positive effect on pH in terms of irrigation water quality, irrespective of the level of dilution.

EC_{iw}: As expected, EC_{iw} increased with a decrease in the level of dilution of the wastewater (Appendix 2.2). This indicated that there was an increase in salt levels with a decrease in dilution of the wastewater. Furthermore, the EC_{iw} in the least diluted water, *i.e.* wastewater diluted to a COD level of 3000 mg/L, was still substantially lower compared to the undiluted winery wastewater. This trend was consistent for all four seasons. On 12 April 2010, the EC_{iw} in the wastewater diluted to COD levels of 2000 mg/L and higher, *i.e.* T7, T8 and T9 (data not shown), exceeded the critical value of 0.75 dS/m which is the salinity threshold for water used for grapevine irrigation (Van Zyl, 1981; Myburgh, 2012). This salinity threshold level was also exceeded in wastewater diluted to a COD level of 3000 mg/L (T9) on 3 May 2010 (data not shown). The EC_{iw} in the T7, T8 and T9 waters, *i.e.* wastewater diluted to COD levels of 2000 mg/L and higher, exceeded the critical value of 0.75 dS/m on 14 April 2011 and 2 May 2012 (data not shown). In 2012/13, the EC_{iw} did not exceed the critical value of 0.75 dS/m. In general, EC_{iw} in the diluted winery wastewaters was well below 2 dS/m, *i.e.* the norm according to the General Authorisations of 2013 (Department of Water Affairs, 2013) as discussed in Section 3.3.2.1.1. Given that the EC in the undiluted water was close to 2 dS/m, dilution of winery wastewater had a positive effect on the salinity hazard in terms of irrigation water quality.

COD: With the exception of the first season, the mean COD levels in the diluted winery wastewater were generally close to the target values (Appendix 2.3). Reasons for the COD deviations from the target values were discussed in Chapter 2. In general, COD in the diluted winery wastewaters was well below the norms according to the General Authorisations of 2013 (Department of Water Affairs, 2013) as discussed in Section 3.3.2.1.1. Given that the COD in the undiluted winery water was generally not lower than 7624 mg/L, dilution of winery wastewater had a positive effect on the COD in terms of irrigation water quality.

Nitrogen: The fact that NO_3^--N and NH_4^+-N were not determined in 2009/10 was identified as a shortcoming, and this was included in the water analyses in subsequent seasons. In general, there were no consistent patterns with regard to NH_4^+-N, NO_3^--N and total N concentrations which could be related to the level of dilution. However, it was evident that the N levels in the river water were higher compared to that of some of the diluted wastewater treatments (Appendix 2.4 to 2.6). At this stage there is no explanation for the latter trend.

Phosphorus: Levels of P in the diluted wastewater treatments increased with a decrease in level of dilution (Appendix 2.7). Although there are no guidelines for P levels in irrigation water, there is a long term critical value of 0.05 mg/L recommended by ANZECC (2000). This norm has been established to minimise the risk of algal blooms developing in storage facilities and to reduce the likelihood of bio-fouling (biological fouling) in irrigation equipment. Levels in diluted wastewaters, as well as undiluted winery wastewater, generally exceeded this norm (Appendix 2.7), thereby indicating that P in winery wastewater may induce algal blooms.

Calcium: The levels of Ca^{2+} increased with a decrease in the dilution of the winery wastewater (Appendix 2.8). There are no guidelines for Ca^{2+} levels in irrigation water (Department of Water Affairs & Forestry, 1996). However, it is important to determine the Ca^{2+} levels to calculate the SAR. As Ca^{2+} is beneficial, rather than harmful to soil structure, it may mitigate the impacts of Na application indirectly *via* its role in reducing the SAR. However, if the Ca^{2+} to Mg^{2+} ratio in the water is less than one, the potential negative effects of Na^+ may be exacerbated (Ayers & Westcott, 1985).

Magnesium: Magnesium concentrations increased with a decrease in dilution of the winery wastewater, but there were no substantial differences in Mg^{2+} levels with regard to the dilution treatments (Appendix 2.9). There are no guidelines for Mg^{2+} levels in irrigation water (Department of Water Affairs & Forestry, 1996). Similar to Ca^{2+}, Mg^{2+} can also play an indirect, positive role in reducing the SAR. On the negative side, crops irrigated with water containing high levels of Mg^{2+} may produce low yields due to Mg^{2+}-induced Ca^{2+}

deficiencies, but there is still insufficient data to make the Ca^{2+} to Mg^{2+} ratio an evaluation factor (Ayers & Westcott, 1985). However, should this ratio be less than one or the Ca^{2+} to total cation ratio less than 0.15, the potential negative effects of Na^+ may be exacerbated (Ayers & Westcott, 1985). If this is the case, a further evaluation of the water is required. Under the conditions of this study, the Ca^{2+} to Mg^{2+} ratio was consistently higher than one (data not shown). Therefore, it is expected that the high Ca^{2+} to Mg^{2+} ratios would have suppressed possible negative effects of Na^+.

Potassium: The K^+ concentrations in the diluted wastewater increased substantially with a decrease in dilution (Appendix 2.10). Mulidzi *et al*. (2009) tentatively considered 200 mg/L K^+ in winery wastewater as being high. In terms of this norm, the K^+ levels in the diluted wastewater were generally not high. Given that the K^+ concentrations in the diluted wastewater were still substantially lower than in the undiluted wastewater, dilution had a positive effect on the K^+ concentration. It should be kept in mind that the K^+ in the wastewaters could make an important contribution to the K^+ nutrient requirements of the grapevine. On the negative side, it must be noted that increasing the amount of K^+ may result in a decrease in hydraulic conductivity and infiltration rate of soils (Levy & Van der Watt, 1990). There is a broad spectrum of possible effects of K^+ on infiltration, ranging from being similar to Na^+, to being similar to Ca^{2+}. Furthermore, it was concluded that, relative to exchangeable Ca^{2+} and Na^+, exchangeable K^+ had an intermediate effect on the soil hydraulic properties (Arienzo *et al*., 2009b).

Potassium adsorption ratio: According to Laurenson *et al*. (2012), the PAR has been less widely adopted than the SAR as K^+ is low in most wastewaters. However, the PAR has proved important for wastewaters of high K^+ concentration, *i.e.* piggery, meat processing and winery wastewaters (Smiles & Smith, 2004). The PAR in the diluted wastewater increased with a decrease in level of dilution (Appendix 2.11). All the values, with only one exception, were lower than values of 2.1 to 3.2 reported for winery wastewater by Laurenson *et al.* (2012). These values were generally lower than PAR values ranging from 3.7 to 43 reported for a winery in Australia (Arienzo *et al.*, 2009b). It was previously shown that soil hydraulic conductivity was considerably reduced when the PAR exceeded 20 in a laboratory study (Arienzo *et al.*, 2009b). These negative effects even occurred when the electrolyte concentrations in the soil was relatively high, *i.e.* > 40 meq/L. However, it was also shown that the negative effect of Na^+ was more pronounced compared to K^+ at the same electrolyte concentration.

Sodium: As the field work progressed, it became evident that the Na^+ concentrations in the river water used for dilution was consistently high. Sampling water at three places along the course of the Holsloot River during the 2012/13 season (Table 3.1) revealed that the Na^+ levels increased substantially during summer along the floodplain up to the point where the water was abstracted near the Goudini winery. However, the Na^+ concentrations in spring (September) were more comparable along the course of the river, probably due to high flow that had occurred in winter. Since no definite point of contamination could be identified, the Na^+ probably became more concentrated under low flow conditions.

Table 3.1. Temporal and spatial variation in sodium (Na^+) concentration in mg/L in water of the Holsloot River, a tributary of the Breede River.

Sampling date	**Locality**		
	In mountain	**Beginning of floodplain**	**Near Goudini winery**
4 December 2012	2.3	3.0	16.6
28 March 2013	1.8	2.2	11.6
30 September 2013	2.3	2.4	3.0

As expected, Na^+ levels in the diluted wastewaters increased substantially with a decrease in dilution of the winery wastewater (Appendix 2.12). Grapevines are considered moderately sensitive to foliar injury from Na^+. Therefore, a concentration of 115 mg/L Na^+ in the water is recommended as the upper threshold for overhead irrigation (Department of Water Affairs & Forestry, 1996). Since the experimental grapevines were irrigated by means of micro-sprinklers, the leaves were not wetted during irrigation. However, the Na^+ levels in the diluted wastewater treatments were so low that even if leaves were wetted, no substantial damage would be expected. The above-mentioned Na^+ threshold was exceeded only on one occasion during the 2010/11 season when the Na^+ concentration amounted to 124 mg/L in winery wastewater diluted to a COD level of 3000 mg/L (data not shown).

Sodium adsorption ratio: As expected, the SAR increased with a decrease in level of dilution of the wastewater (Appendix 2.13). Since the Ca^{2+} and Mg^{2+} also increased, it suggested that the increase in these cations could not counterbalance the effect of increased Na^+ on the SAR. The SAR norm stipulated by the General Authorisations of 2013 (Department of Water Affairs, 2013) was only exceeded on 14 April 2011 where the SAR of irrigation waters diluted to COD levels of 1500 mg and higher was more than 5 (data not shown). Furthermore, the SAR in the diluted winery wastewaters was still within acceptable limits for grapevine irrigation, *i.e*. less than *c.* 10 (Richards, 1954; Myburgh, 2012). It must be noted that the SAR in the undiluted winery wastewater also did not exceeded the acceptable threshold for grapevine irrigation.

Chloride: Similar to Na^+, it became evident that the Cl^- concentrations in the river water used for dilution was consistently high. Sampling water along the course of the Holsloot River during the 2012/13 season revealed that the Cl^- levels increased substantially during summer along the floodplain up to the point where water was abstracted near the winery (Table 3.2). However, the Cl^- concentrations in spring were more comparable along the course of the river, probably due to high flow in winter. Since no definite point of contamination could be identified, the Cl^- probably became more concentrated under low flow conditions.

Table 3.2. Temporal and spatial variation in chloride (Cl^-) concentration in mg/L in water of the Holsloot River, a tributary of the Breede River.

Sampling date	**Locality**		
	In mountain	**Beginning of floodplain**	**Near Goudini winery**
4 December 2012	8.9	8.8	19.6
28 March 2013	10.0	9.9	31.0
30 September 2013	6.8	7.7	9.8

Although the Cl^- concentrations in the diluted waters were lower compared to the undiluted winery wastewater, the range of dilutions did not cause any consistent trends with respect to the level of dilution (Appendix 2.14). Levels in both the diluted and undiluted winery wastewater were substantially lower than 150 mg/L and 700 mg/L which are the threshold for overhead irrigation and grapevine root uptake, respectively (Van Zyl, 1981). This suggested that Cl^- levels in the diluted wastewater would not have caused damage to the grapevines, even if the leaves were wetted.

Bicarbonate: Although the HCO_3^- concentration increased with a decrease in the dilution of the wastewater, the concentrations varied considerably between seasons (Appendix 2.15). The HCO_3^- concentration also showed large variation within a season, *i.e.* high standard deviation values. Similarly, the HCO_3^- concentration also showed large variation within a season. The reason for the variability is uncertain. However, the concentration in the least diluted water (T9) was still substantially lower compared to the undiluted winery wastewater. When irrigations were applied on 9 February 2011, 30 March 2011, 6 March 2012 and 19 March 2012, no trends with regard to the level of dilution were evident for HCO_3^- concentrations in the water (data not shown). Throughout the 2012/13 season no trends were evident for HCO_3^- levels in the water with the exception of 30 April 2013 (data not shown). At this stage, there is no explanation why the HCO_3^- level did not increase with a decrease in the level of dilution on these days.

Irrigation water containing high levels of HCO_3^- can negatively affect plants, soils and irrigation systems. Irrigation waters that contain high levels of HCO_3^- and CO_3^{2-} can increase HCO_3^- in the soil solution. Consequently, Ca^{2+} and Mg^{2+} can precipitate as insoluble carbonates when the soil dries out (Van Zyl, 1981; McCarthy *et al.*, 1988). According to norms proposed for HCO_3^- concentrations in overhead irrigation water, values lower than 91.5 mg/L (1.5 me/L) indicate no restriction when used, whereas levels between 91.5 mg/L and 518.6 mg/L (8.5 me/L) indicate a slight to moderate degree of restriction when used (Ayers & Westcott, 1985). In the first season, dilutions above 500 mg/L COD contained excessive levels of HCO_3^-, whereas in the second and third seasons, dilutions above 1000 mg/L COD contained excessive HCO_3^- (Appendix 2.15). In contrast, HCO_3^- levels fell into the no restriction category during the 2012/13 season. There are no recently recommended guidelines (Department of Water Affairs & Forestry, 1996; ANZECC, 2000).

Sulphate: Although the SO_4^{2-} in the diluted irrigation waters generally increased with a decrease in the level of dilution of the wastewater, the levels in the least diluted water (T9) was substantially lower compared to the winery wastewater (Appendix 2.16). Similar to HCO_3^-, the SO_4^{2-} concentrations varied considerably between seasons, and within a season. The reason for the variability is also uncertain. In general, SO_4^{2-} levels in the diluted winery wastewater were below the proposed level of 150 mg/L and lower for reclaimed effluent water quality standards for vineyard re-use (Ryder, 1995). In the 2009/10 and 2011/12 seasons, SO_4^{2-} levels in the undiluted wastewater were above this optimum level, but still lower than the maximum threshold of 250 mg/L.

Boron: Boron levels in the diluted wastewater increased with a decrease in the level of dilution (Appendix 2.17). Concentrations in the least diluted water were still substantially lower compared to the undiluted winery wastewater. Although B^{3+} is essential for the growth of plants, it reaches toxic levels at low concentrations. Grapevines have been classed as sensitive (Ayers & Westcott, 1985; Department of Water Affairs & Forestry, 1996; ANZECC, 2000) to highly sensitive (Van Zyl, 1981) with regard to B toxicity. In general, B^{3+} levels of 0.5 mg/L are considered ideal for vineyard irrigation (McCarthy *et al.*, 1988), whereas levels under 0.75 mg/L have been recommended by Ayers and Westcott (1985). On 13 April 2011, the limit of 0.5 mg/L was only exceeded where winery wastewater was diluted to 3000 mg/L COD (T9), whereas on 14 April 2011, winery wastewater diluted to 1500 mg/L and more (T6 to T9) exceeded this limit (data not shown). It should be noted that due to low COD levels in the winery wastewater in April 2011, the range of COD levels had to be made up and applied on two consecutive days. On 2 May 2012, diluted winery wastewater of 1000 mg/L and higher (T5 to T9) also exceeded the limit of 0.5 mg/L The foregoing showed that diluted winery wastewater has a sporadic risk of inducing B^{3+} toxicity if used for vineyard irrigation.

Iron: The Fe^{2+} levels increased with a decrease in the dilution of the wastewater, but the level in the least diluted wastewater was still substantially lower compared to the undiluted winery wastewater (Appendix 2.18). Recommended maximum levels of Fe^{2+} in irrigation water for continuous irrigation on all soils is 5 mg/L (Van Zyl, 1981). With the exception of the 2011/12 season, Fe^{2+} levels in the undiluted wastewaters never exceeded this value. However, the Fe concentration becomes important in the case of drip irrigation where major clogging problems can be expected when Fe^{2+} levels are higher than 1.5 mg/L (Department of Water Affairs & Forestry, 1996). Given the low Fe^{2+} concentrations in the diluted wastewater, it would not cause clogging of micro-sprinkler systems under the prevailing conditions.

Heavy metals: Although the heavy metal concentrations in the diluted wastewater tended to be lower compared to the undiluted winery wastewater (Appendix 2.19 to 2.21), they did not show any consistent trends with respect to the different levels of dilution. Levels were also generally low, therefore they were not considered in this dissertation.

3.3.3. Amount of elements applied

Nitrogen: In terms of NH_4^+-N, total amounts added *via* the irrigation water were generally higher where wastewater was diluted to 1500 mg/L and higher compared to the river water control (Appendix 3.1) With regard to NO_3-N, total amounts added *via* the irrigation water were generally higher where wastewater was diluted to 2500 mg/L and higher compared to control (Appendix 3.2). For total-N, amounts added *via* the irrigation water were higher where wastewater was diluted to 2000 mg/L and higher compared to control (Appendix 3.3). Trends across the range of COD levels were inconsistent. The total-N applied *via* the winery wastewater diluted to 2000 mg/L COD and higher was similar to the estimated *c.* 5 kg N per ha applied *via* winery wastewater based on an irrigation depth of 100 mm (Laurenson *et al.*, 2012). Full bearing grapevines annually require 50 kg N where 10 to 15 tonne of fruit is produced per ha (Conradie, 1994). Based on this recommendation, the amount of N applied *via* the diluted wastewater appeared to be completely inadequate to supply the grapevine’s annual N requirement under the prevailing conditions (Fig. 3.10A). Therefore, winery wastewater cannot be considered as a sufficient source of N for grapevines.

Phosphorus: The amount of P applied *via* the irrigation water increased with a decrease in wastewater dilution (Appendix 3.4). The P applied *via* the winery wastewater diluted to 2000 mg/L COD and higher was similar to the estimated *c.* 5.3 kg P per ha applied *via* winery wastewater based on an irrigation depth of 100 mm (Laurenson *et al.*, 2012). Full bearing grapevines annually require 0.7 kg P per tonne of fruit produced (Conradie, 1994). Based on this recommendation, the amount of P applied *via* the winery wastewater diluted to 2500 mg/L COD and higher would supply adequate P during most seasons if the grape yield amounts to 10 t/ha under the prevailing conditions (Fig. 3.10B).

Potassium: The amount of K^+ applied per hectare increased substantially with a decrease in dilution of the winery wastewater (Appendix 3.5). In 2010/11 and 2012/13, similar amounts were applied in the pre- and post-harvest periods. Since only one irrigation was applied in the post-harvest period of 2011/12, amounts of K^+ added *via* the irrigation water were substantially less than during the pre-harvest period. In general, K^+ applied *via* winery wastewater diluted to 2500 mg/L and higher was more than the estimated 129 kg K^+ per hectare applied *via* winery wastewater based on an irrigation depth of 100 mm (Laurenson *et al.*, 2012).

Full bearing grapevines annually require 3 kg K^+ per tonne of fruit produced (Conradie, 1994). Based on this recommendation, the amount of K^+ applied *via* winery wastewater diluted to 250 mg/L COD and higher would supply more than adequate K^+ if the grape yield amounts to 10 t/ha under the prevailing conditions (Fig. 3.10C). The K^+ supplied *via* diluted wastewater will only be beneficial for a month after harvest as the grapevine's nutrient requirements are generally low after this (Conradie, 1981). The K applied will also only be beneficial in the following season if it is not leached from the root zone during winter.

On average, between 47 kg/ha and 164 kg/ha K^+ was applied in excess to the soil where the winery wastewater was diluted to COD levels ranging from 1000 mg/L (T5) to 3000 mg/L (T9). The effect of high concentrations of K^+ applied to soils as well as the fate of K^+ in soils and grapevines irrigated with winery wastewater has received limited attention (Mosse *et al.,* 2011; Laurenson *et al.*, 2012). However, excessive K^+ applied *via* the diluted winery wastewater could have several implications. Excessive K^+ in grape berries can be detrimental to wine quality, as it decreases free tartaric acid (Mpelasoka *et al.*, 2003). Subsequently pH in grape juice, must and wine increases (Saayman, 1981; Mpelasoka *et al.*, 2003). Excessive K^+ in fruit also causes the formation of insoluble potassium bitartrate (Laurenson *et al.*, 2012). The increase in pH causes unstable musts and wines, as well as a reduction in the degree of ionisation of anthocyanins (Mpelasoka *et al.*, 2003). The increase in berry pH in hot climates produces grape juice with a high pH which has a flat taste and possible brown hue (Kodur, 2011; Laurenson *et al.*, 2012). In addition to these grapevine responses, excessive K^+ can reduce juice N (Saayman, 1981), thereby increasing the risk of stuck fermentation during the winemaking process. Excessive K^+ can also reduce Ca^{2+} and Mg^{2+} in the grapevine indicating antagonisms between K^+ and these elements (Morris & Cawthon, 1982; Wolf *et al.,* 1983; Myburgh & Howell, 2014). Given that the amounts of K^+ applied *via* the diluted winery wastewater were much higher than the requirements of the grapevine, the cultivation of an interception crop during summer might be useful to absorb excessive K^+.

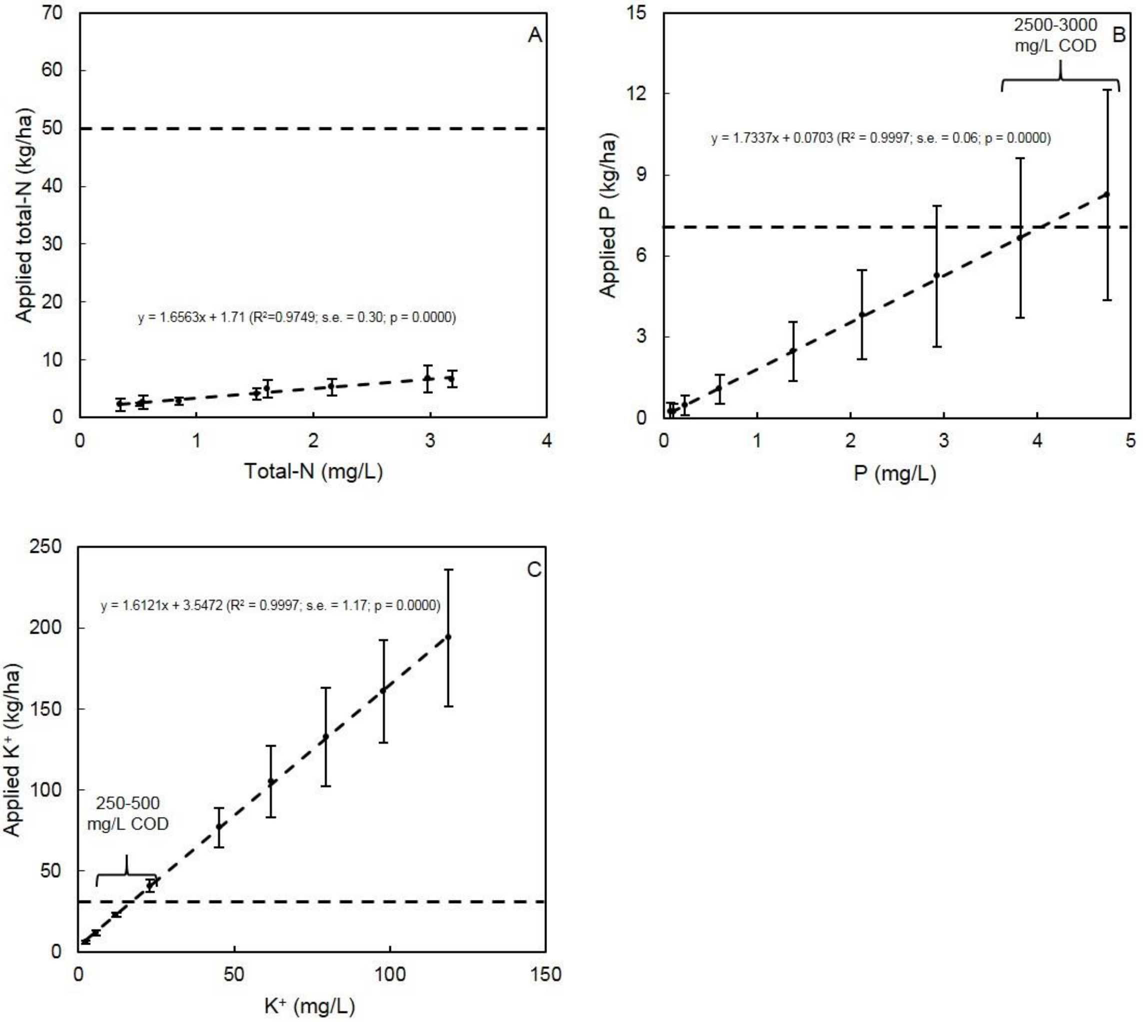

Figure 3.10. Relationship between amounts of (A) total nitrogen (total-N), (B) phosphorus (P) and (C) potassium (K^+) applied *via* irrigation using diluted winery wastewater and the respective total-N, P and K^+ concentration in the water. Dashed horizontal lines indicate the annual grapevine requirement for a grape yield of 10 t/ha according to Conradie (1981, 1994). With the exception of total-N, data are means for four seasons. Vertical bars indicate standard deviation.

Calcium and magnesium: The amounts of Ca^{2+} and Mg^{2+} applied increased with a decrease in dilution of the winery wastewater (Appendix 3.6 & 3.7), but differences between the highest level of dilution, *i.e.* T2, and the lowest level of dilution, *i.e*. T9, were not as substantial as in the case of K^+ and Na^+. Full bearing grapevines require *c.* 2 kg Ca^{2+} annually per tonne of fruit produced (Conradie, 1981). Based on this recommendation, the amount of Ca^{2+} applied *via* winery wastewater diluted to 500 mg/L COD and higher would supply more than adequate Ca^{2+} if the grape yield amounts to 10 t/ha under the prevailing conditions (Fig. 3.11A). As the grapevine's nutrient requirements are generally low during the harvest and post-harvest periods (Conradie, 1981), the Ca^{2+} supplied *via* the wastewater will only be beneficial if it is not leached from the root zone during winter. Furthermore, bunches require no Ca^{2+} from véraison and harvest. With regard to Mg^{2+}, 0.7 kg is required per tonne of grapes produced. Under the prevailing conditions, all of the treatments supplied sufficient Mg^{2+} to supply the grapevine's requirements (Fig. 3.11B).

Sodium: Amounts of Na^{+} applied per hectare also increased substantially with a decrease in level of dilution of the winery wastewater (Appendix 3.8). In 2010/11 and 2012/13 similar amounts were applied in the pre- and post-harvest periods. As there was only one irrigation in the 2011/12 post-harvest period, amounts of Na^{+} added *via* the irrigation water were substantially less than during the pre-harvest period. The total amounts of Na^{+} added *via* the irrigation water ranged from 32 kg/ha for the river water control (T1) to 85 kg/ha for the least diluted water, *i.e.* 3000 mg/L COD (Fig. 3.11C). Although there are no threshold values for grapevines with regard to amount of Na^{+} applied per hectare, it is well known that excessive Na^{+} can reduce vegetative growth, yield and suppress Ca^{2+} uptake (Myburgh & Howell, 2014 and references therein).

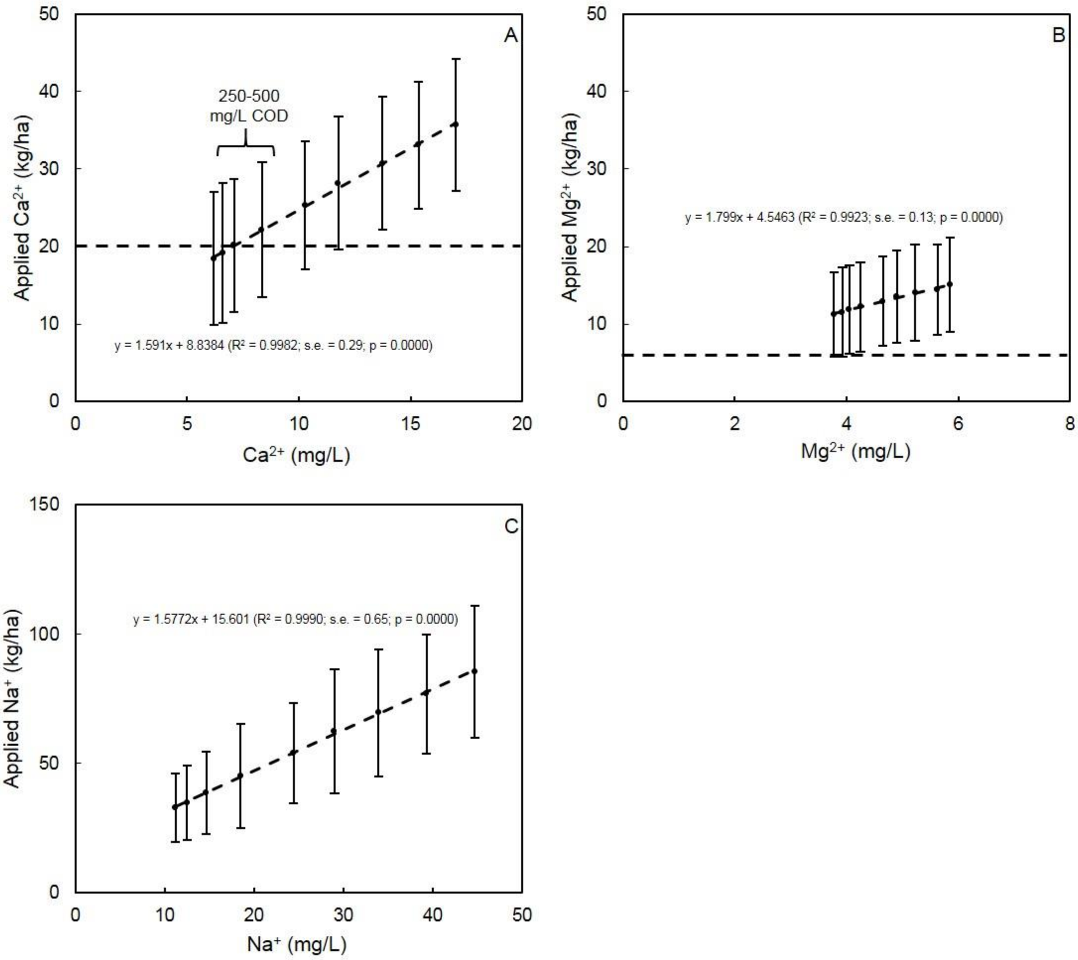

Figure 3.11. Relationship between amounts of (A) calcium (Ca^{2+}), (B) magnesium (Mg^{2+}) and (C) sodium (Na^{+}) applied *via* irrigation using diluted winery wastewater and the respective Ca^{2+}, Mg^{2+} and Na^{+} concentration in the water. Dashed horizontal lines indicate the annual grapevine requirement for a grape yield of 10 t/ha according to Conradie (1981, 1994). Data are means for four seasons. Vertical bars indicate standard deviation.

Chloride: Although amounts of Cl^- added *via* the irrigation water were higher for T9 compared to the river water control (T1), increases across the COD levels were inconsistent (Appendix 3.9). It should be noted that the river water also contained relatively high levels of Cl^-. The total amounts of Cl^- added *via* the irrigation water ranged from 76 kg/ha for the river water control to 87 kg/ha for the least diluted water, *i.e.* 3000 mg/L COD (Fig. 3.12A). Although there are no threshold values for grapevines with regard to amount of Cl^- applied per hectare, excessive Cl^- can reduce vegetative growth and yield (Myburgh & Howell, 2014 and references therein).

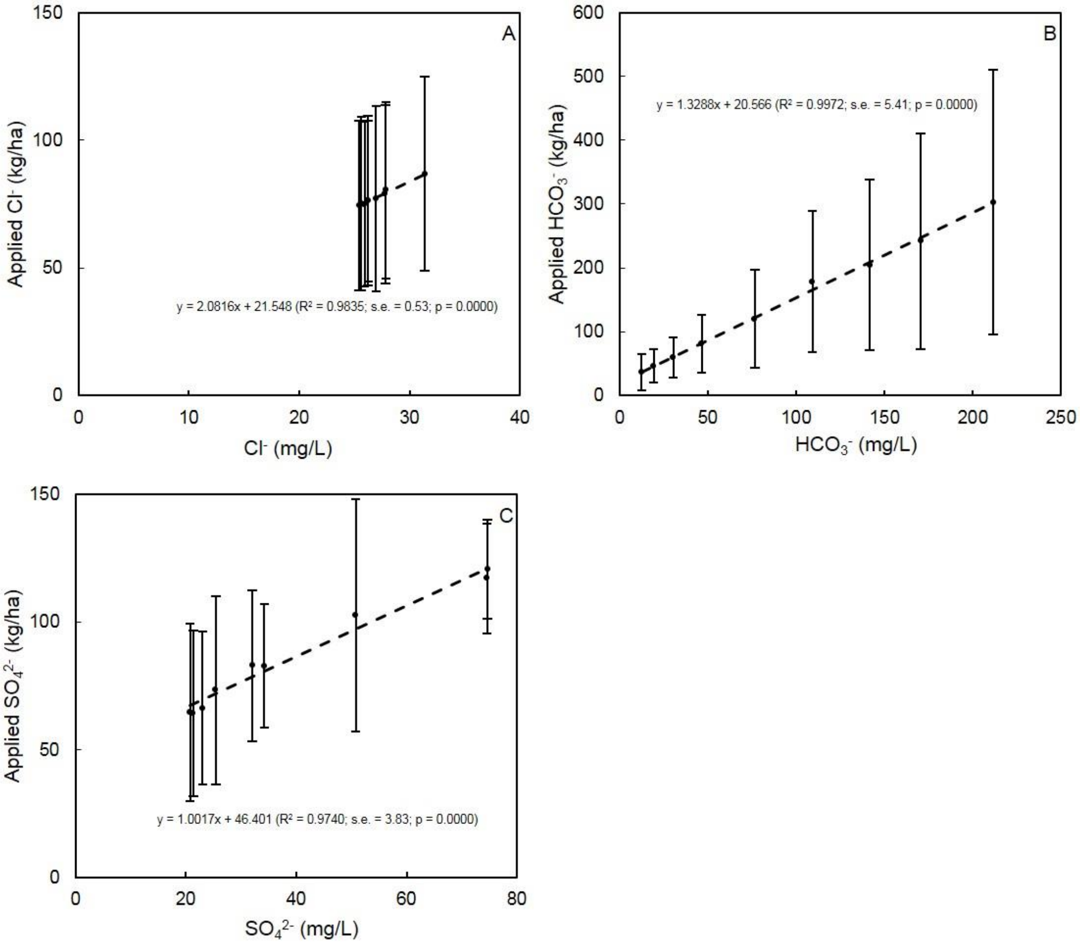

Figure 3.12. Relationship between amounts of (A) chloride (Cl^-), (B) bicarbonate (HCO_3^-) and (C) sulphate (SO_4^{2-}) applied *via* irrigation using diluted winery wastewater and the respective Cl^-, HCO_3^- and SO_4^{2-} concentration in the water. Data are means for four seasons. Vertical bars indicate standard deviation.

Bicarbonate: In 2010/11 and 2011/12, there was a substantial increase in HCO_3^- applied per hectare with a decrease in dilution of the wastewater (Appendix 3.10). In the 2012/13 season, amounts of HCO_3^- applied per ha in the pre-harvest period did not show any trends. However, in the post-harvest period, there was a substantial increase in HCO_3^- applied per ha with a decrease in dilution of the wastewater. The total amounts of HCO_3^- added *via* the irrigation water ranged from 36 kg/ha for the river water control to 303 kg/ha for the least diluted water, *i.e.* 3000 mg/L COD (Fig. 3.12B).

Sulphate: In general, the amount of SO_4^{2-} applied *via* the irrigation water increased with increasing level of COD (Appendix 3.11). The total amounts of SO_4^{2-} added *via* the irrigation water ranged from 66 kg/ha for the river water control to 117 kg/ha for the least diluted water, *i.e.* 3000 mg/L COD (Fig. 3.12C).

Boron: Despite low levels of B^{3+} being applied *via* the irrigation water, amounts increased with a decrease in level of dilution of the winery wastewater (Appendix 3.12). According to Conradie (1994), the requirements of the grapevine for B^{3+} is relatively low. Under the prevailing conditions, less than one kg of B^{3+} per ha was applied *via* the diluted winery wastewater (Fig. 3.13A).

Iron: The amounts of Fe^{2+} applied per hectare increased with a decrease in level of dilution of the winery wastewater (Appendix 3.13). According to Conradie (1994), the requirements of the grapevine for Fe^{2+} is relatively low. Under the prevailing conditions, *c.* two kg of Fe^{2+} per ha was applied *via* the diluted winery wastewater (Fig. 3.13B).

Heavy metals: The concentrations of Cd^{2+}, Cr^{2+} and As^{3-} in the undiluted winery wastewater were extremely low, therefore amounts applied *via* the diluted winery wastewater were low (data not shown). Furthermore, the heavy metals did not show any trends with regard to level of dilution.

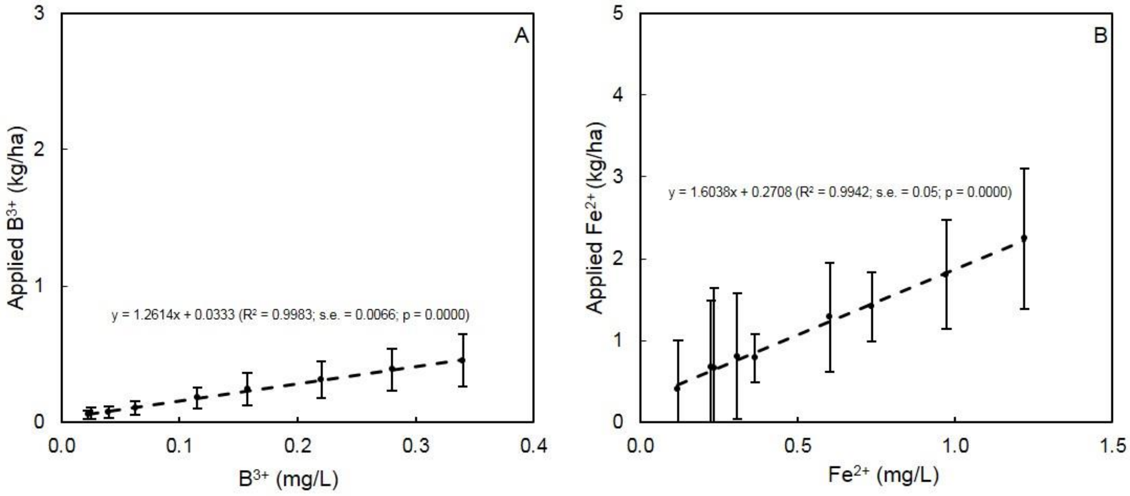

Figure 3.13. Relationship between amounts of (A) boron (B^{3+}) and (B) iron (Fe^{2+}) applied *via* irrigation using diluted winery wastewater and the respective B^{3+} and Fe^{2+} concentration in the water. Data are means for four seasons. Vertical bars indicate standard deviation.

3.4. CONCLUSIONS

In previous studies, artificial “winery wastewater” was used. Furthermore, most of these studies were carried out in laboratories. This study was the first where wastewater from a commercial winery was diluted with raw river water to a range of COD levels for vineyard irrigation at the field scale. Although the COD concentration in winery wastewater was the preferred indicator of water quality of the Steering Committee for the project, it did not provide a reliable indication of wastewater pH and EC. Furthermore, the COD level could not be used to estimate K^+ and Na^+ concentrations in the winery wastewater as these levels differed substantially at a specific COD level. Level of COD also provided no indication of the salinity or sodicity hazard of the wastewater. The EC in the undiluted winery wastewater was strongly determined by the K^+ concentration. This was to be expected, since K^+ is usually the most abundant cation in winery wastewater. Therefore, in future research projects investigating the re-use of winery wastewater for irrigation, EC would be a more reliable indicator of the quality of the winery wastewater than COD concentration, particularly with regard to cation concentrations such as K^+ and Na^+. In addition, EC would be easier for winery staff to measure. The ratio of these monovalent cations to bi-valant Ca^{2+} and Mg^{2+} is also an important consideration in the suitability of water for irrigation purposes. In addition to conforming to pH, EC and sodicity criteria, water application needs to be scheduled in such way that the applied organic matter is allowed to oxidise between irrigations. The foregoing aspects are critical for the sustainable irrigation of vineyards or other crops with diluted winery wastewater.

Since the pH in the diluted wastewater was lower than 6, it could induce nutrient toxicity, if used for irrigation of vineyards or other crops. The results indicated that dilution of winery wastewater did not have any positive effect on pH with respect to irrigation water quality. The diluted winery wastewater did not pose any salinity hazard, since EC_{iw} was well below 2 dS/m. As the EC in the undiluted water was close to 2 dS/m, dilution of winery wastewater reduced the salinity hazard with respect to irrigation water quality. Given the fact that the COD in the undiluted winery water was generally not lower than 7624 mg/L, dilution of winery wastewater had a positive effect on the COD in terms of irrigation water quality. The K^+ concentrations in the diluted wastewater were substantially lower than in the undiluted wastewater. The K^+ levels in the diluted wastewater were generally not high for winery wastewater, *i.e.* less than 200 mg/L. It must be noted that the K^+ in the wastewater could make a contribution to the K^+ requirements of the grapevine, *i.e.* if it is not lost *via* leaching during winter. For the given range of wastewater dilutions, the SAR never exceeded 10, which indicated that the diluted wastewater posed little sodicity hazard. Sodium and Cl^- never exceeded 115 and 150 mg/L, *i.e.* the respective upper toxicity thresholds of these elements for grapevines. Considering the classical water quality criteria, *i.e.* pH, EC and SAR, dilution of winery wastewater up to a COD level of 3000 mg/L produced irrigation water of which the quality would permit sustainable vineyard irrigation under the prevailing conditions, *i.e.* Mediterranean climate with high winter rainfall and sandy soil.

As expected, levels of P, K^+, Na^+, Ca^{2+}, Mg^{2+}, HCO_3^-, SO_4^{2-} and B^{3+} in the diluted wastewater increased with a decrease in the level of dilution. In contrast, levels of N and Cl^- were inconsistent with regard to the level of dilution. The B^{3+} concentrations in undiluted winery wastewater indicated a potential risk of inducing B^{3+} toxicity if used for vineyard irrigation. Since B^{3+} levels in the least diluted wastewater exceeded the recommended norm sporadically, there is still a slight B^{3+} toxicity risk which should not be ignored. Results indicated that SO_4^{2-} levels in the diluted winery wastewater were below the proposed level of 150 mg/L for effluent water quality standards for vineyard re-use.

Since one of the incentives for diluting winery wastewater is that it could serve as a possible nutrient source, it is important to note that the N load in the diluted winery wastewater was completely inadequate to supply the grapevine's annual requirement. On the positive side, P loads in the winery wastewater diluted to 2500 mg/L COD and higher could supply more than adequate P if the grape yield amounts to 10 t/ha. Likewise, dilution of winery wastewater to 250 mg/L COD and higher could supply more than adequate K^+ if grape yield amounts to 10 t/ha. However, the excessive K^+ applied *via* the diluted wastewater could increase juice pH that could cause unstable musts and wines, as well as a reduction in the degree of ionisation of anthocyanins in the wine. Furthermore, excessive K^+ application could induce nutrient imbalances in the grapevine tissues, particularly antagonisms with respect to N, Ca^{2+} and Mg^{2+}. Given that the amounts of K^+ applied *via* the diluted winery wastewater were considerably higher than the grapevine's requirements, the cultivation and removal of a suitable interception crop during summer might be useful to absorb excessive K^+.

In practice it would be essential to know the mass of nutrients applied *via* the diluted winery wastewater irrigation in order to adjust the normal nutrition program of grapevines or other crops accordingly. However, in addition to the annual, and even daily variation, the composition of winery wastewater can vary considerably between wineries. Therefore, chemical analyses of the water and measuring the volumes of water applied per irrigation would be necessary to calculate the exact amount of nutrients applied where diluted winery wastewater is used for irrigation. Considering the foregoing, monitoring the water quality and irrigation volumes will have to be included in the overall wastewater management programs employed by wineries. It is recommended that wineries should assess the wastewater quality and volumes at least fortnightly if the water is to be used for irrigation of crops.

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CHAPTER 4: EFFECT OF IRRIGATION USING DILUTED WINERY WASTEWATER ON SOIL CHEMICAL STATUS OF A SANDY ALLUVIAL SOIL WITH PARTICULAR REFERENCE TO POTASSIUM AND SODIUM

4.1. INTRODUCTION

During the grape harvest period, wineries produce large volumes of low quality wastewater which can contain high levels of potassium (K^+) and sodium (Na^+). The chemical status of this water is generally worse than the legislated limits for irrigation with wastewater (Department of Water Affairs, 2013). Information on actual volumes of wastewater generated by wineries is extremely limited. However, medium to large wineries generate more than 15 000 m^3 of wastewater annually, whereas small wineries generate less than 15 000 m^3 annually (Van Schoor, 2005 and references therein). For Lutzville Vineyards' winery in the Olifant's River region of South Africa, *c.* 1.1 m^3 of wastewater is produced per tonne of grapes crushed (Kriel, 2008). However, this relatively low value is misleading, since 50% of the wastewater is presumably lost to evaporation. In Australia, it is estimated that approximately 3 to 5 m^3 of winery wastewater, with high organic load and variable salinity and nutrient levels, is produced per tonne of grapes crushed (Mosse *et al*., 2011). On the other hand, limited irrigation water supplies could be further restricted in future allocations of irrigation water (Van Zyl & Weber, 1981; Petrie *et al.*, 2004). If winery wastewater could be re-used to irrigate vineyards, with no detrimental impacts on soil chemical status, it could be a viable alternative to using water abstracted from natural resources.

Currently, the Department of Water and Sanitation is drafting new General Authorisations for wineries. Depending on the permitted water quality limits and volumes stipulated by the new authorisations, diluting winery wastewater with current irrigation water may well become a more viable practice in the future. Re-using winery wastewater in this way will be beneficial, particularly where there are water shortages. In such situations, re-using winery wastewater will have a positive impact on grape yields if additional irrigation could be applied. Water saving and higher yields will also contribute to sustainability and economic viability of wine production. Presently, there is also increasing pressure on producers to use water in a more environmentally friendly way. If winery wastewater could be re-used in a sustainable way, it could also have other benefits, such as a reduction in the energy required for wastewater treatment and the availability of nutrients. Plant nutrients in the wastewater, such as K^+, calcium (Ca^{2+}) and magnesium (Mg^{2+}) could reduce fertiliser requirements, thereby reducing fertilization costs. In addition, land application of wastewaters can increase soluble and exchangeable forms of K^+ more rapidly than with conventional inorganic fertilizers and most of the K^+ is available immediately (Arienzo *et al.*, 2009). Although it appears that the nitrogen (N) load in diluted winery wastewater would be inadequate to supply the grapevine's N requirement, phosphorus (P) and K^+ applied *via* diluted winery wastewater should be adequate for a grape yield of *c.* 10 t/ha (Refer to Chapter 3).

Using treated municipal wastewater for crop irrigation increased Na^+ to a depth of 1.8 m, as well as K^+ and Ca^{2+} to a depth of 0.6 m (Hulugalle *et al.*, 2006). In the case of Na^+, the additional amount added *via* the municipal wastewater was 736 kg/ha and 1834 kg/ha during 2001/02 and 2003/04, respectively. For K^+, 49 kg/ha and 71 kg/ha were added in same two seasons. Where 50 mm of wastewater was used weekly for irrigation over 53 years, soil pH and organic matter increased considerably (Walker & Lin, 2008). Over the short term, *i.e.* after one year, land application of treated municipal wastewater had no effects on Na^+, Ca^+ and Mg^{2+} in sandy clay loam containing *c.* 32% clay (Duan *et al.,* 2010). When treated municipal wastewater was used for grapefruit tree irrigation on sandy and clay soils, soil pH and EC increased (Lado *et al.*, 2011), and there was an increase in organic matter content, P and K^+ in the sandy soil. When treated sewage wastewater was applied to a sandy loam and a sandy clay loam soil, there was a substantial increase in the exchangeable sodium percentage (ESP) and clay dispersion after 16 months (Blum *et al*., 2012). Furthermore, soil pH increased in sub-soil layers. Although Na^+ increased substantially, levels returned to low concentrations after rainfall. Using treated municipal wastewater for six years to irrigate table grapes increased soil Na^+ compared to irrigation with fresh water (Netzer *et al.,* 2014). In an in-field survey in a drip irrigated vineyard in South Australia, municipal wastewater increased soil Na^+ and Mg^{2+} in the top 20 cm soil layer (Laurenson, 2010). With regard to the long-term effects of irrigation with untreated sewage water on agricultural fields, Ca^{2+}, Mg^{2+}, chloride (Cl^-) and bicarbonate (HCO_3^-) increased in the 0-10 cm soil layer (Rana *et al.*, 2010).

When olive mill wastewater was used for irrigation, annual applications of 4161 kg/ha *via* the wastewater increased soil K^+ after five years (Moraetis *et al*., 2011). It must be noted that the olive mill wastewater contained substantially more K^+ than winery wastewater diluted to *c.* 3000 mg/L chemical oxygen demand (COD) used in the current study (Refer to Chapter 3). The acidic nature of olive mill wastewaters will also result in the long-term loss of carbonate from the top soil (Barbera *et al.,* 2013) and reduce soil pH (Di Bene *et al.,* 2013). The use of olive mill wastewaters increases soil EC_e because of its high salt concentration. Several authors have reported an increase in both soil K^+ and P levels (Barbera *et al.,* 2013; Di Bene *et al.,* 2013). Piggery wastewater, which also contains high K^+ levels, increased soil K^+ compared to where no wastewater was applied on six piggery farms (Smiles & Smith, 2004). The duration of application of the wastewater ranged from six to 30 years.

Although there is extensive literature available regarding the effect of irrigation with wastewaters of various origins on soil chemical properties, there is no information regarding the re-use of winery wastewater diluted to pre-determined levels of COD, for any crop. In pastures irrigated with undiluted winery wastewater for over 100 years, total organic carbon, N, K^+, Na^+, Mg^{2+} and Ca^{2+} levels increased relative to the controls (Kumar *et al.*, 2006). Although soil K^+, Na^+, Mg^{2+} and Ca^{2+} of pastures irrigated with undiluted winery wastewater for 15 to 20 years increased, these increases were not as substantial as where pastures which had been irrigated for 100 years (Kumar *et al.*, 2006). However, there were no differences with regard to Ca^{2+} and Mg^{2+} in the soil. Where winery wastewater was applied to a silty clay loam soil for 30 years, soil K^+ and Na^+ were substantially higher compared to soil where no winery wastewater was applied (Mosse *et al*., 2012). Although irrigation with winery wastewater for three years increased soil K^+ in the surface layer, sub-surface soil K^+ remained unchanged (Quale *et al.*, 2010). Irrigation using winery wastewater containing high levels of organic carbon increased total soilorganic carbon content (Kumar *et al.*, 2009). In addition, soil K^+, as well as salinity and sodicity levels were higher in wastewater treated plots compared to control plots, particularly woodlot and pasture sites at certain wineries. Irrigation using undiluted winery wastewater increased soil K^+ to a depth of 90 cm (Mulidzi *et al.*, 2009). According to Kumar *et al.* (2014), both soil K^+ and sodium adsorption ratio (SAR) increased throughout the soil profile where winery wastewater was used for irrigation. The latter practice also resulted in higher Na^+ and K^+ in vineyard soils than a control vineyard which was irrigated with river water (Kumar *et al.*, 2006). In a field study, where grapevines were irrigated with simulated winery wastewater, soil Na^+ levels in the 0-20 cm and 20-40 cm layers increased (Mosse *et al*., 2013). The addition of wine to the simulated winery wastewater enhanced K^+ movement to the sub-soil. In a laboratory study, irrigation with winery wastewater increased soil Na^+ and K^+ in a loamy sand, a loam and a clayey soil (Kumar *et al.*, 2006). It should be noted that these soils were collected from areas where winery wastewater is currently being used for irrigation of woodlots, pastures or vineyards. Winery wastewater used for irrigation also increased soil $pH_{(1:5)}$, K^+ and Ca^{2+} of a deep sand, clay loam and a hard setting sandy loam in a laboratory study (Laurenson, 2010).

Irrigation with wastewaters containing high levels of K^+, such as winery wastewater, could be beneficial to overall soil fertility, although long-term application could have negative effects on soil chemical and physical properties (Smiles & Smith, 2004; Kumar & Christen, 2009; Laurenson *et al*., 2011; Mosse *et al.*, 2011). The effects of high K^+ concentrations on soil properties have not been extensively researched and are still unclear (Kumar *et al.*, 2009; Mosse *et al.*, 2011; Laurenson *et al*., 2012). However, accumulation of monovalent ions in the soil can deteriorate soil structure and hydraulic conductivity, thereby negating soil productivity (Smiles & Smith, 2004; Kumar *et al.*, 2006; Laurenson *et al.*, 2011). In addition to K^+ and Na^+, winery wastewater can contain Ca^{2+} and Mg^{2+} (Mosse *et al*., 2011). Neither of the latter mentioned ions are harmful to soil structure and can ameliorate the impacts of Na^+

via their role in reducing the SAR. However, a matter of potential concern is Na^+ and Mg^{2+} accumulation in surface soils and subsequent loss of Ca^{2+} (Laurenson, 2010). A literature search revealed that the effect of irrigation with winery wastewater on soil P is not well-documented. With respect to P, Mulidzi *et al.* (2009) reported that land application of undiluted winery wastewater increased soil P, but that the P in the different soil horizons fluctuated throughout the season.

The objectives of the study were to (i) determine the effect of irrigation with winery wastewater diluted to eight different levels of COD on the soil chemical status to determine a possible threshold concentration for sustainable use and (ii) develop empirical mathematical models to estimate soil Bray-II K and Na^+ at bud break and after wastewater application in a sandy, alluvial vineyard soil.

4.2. MATERIALS AND METHODS

4.2.1. Collection of samples

For details of the experimental vineyard and layout refer to Chapter 2. The water quality as well as amounts of irrigation and elements applied are presented in Chapter 3. Soil samples were collected using an auger in August 2009 before the trial commenced to determine the baseline chemical status before treatments were applied. Samples were taken over 30 cm increments to a depth of 1.8 m. After the first season of wastewater application, soil samples were collected over the same depth increments in the work rows of all experimental plots in May 2010. Soil from each of the three replications of each treatment was pooled for analysis. Soil samples were also collected at bud break in October 2010, September 2011 and 2012 in the work rows of all plots. Soil from the three replications of each treatment was also pooled for analysis. In April 2011, soil samples were collected from each of the three replications of the river water control (T1) as well as where winery wastewater was diluted to 250 (T3), 1000 (T5), 2000 (T7) and 3000 mg/L COD (T9), respectively. The samples were collected *c.* one week after the end of the wastewater application. In order to determine possible differences in the soil chemical properties within the vineyard due the water distribution pattern of the irrigation system, samples were taken in the work row and in the grapevine row. In contrast to soil samples taken in early May 2010, no rain occurred in the period preceding the collection of these samples. The same procedure was followed for samples collected after the wastewater application had stopped in May 2012 and 2013. At the end of the trial in September 2013, soil samples were collected in the work rows of all the experimental plots over 30 cm increments to a depth of 3.0 m using an extended soil auger (Fig. 4.1).

4.2.2 Analysis

All samples were analysed by a commercial laboratory (Bemlab, Strand). Soil $pH_{(KCl)}$ was measured in 1M KCl. Electrical conductivity of the saturated extract (EC_e) was determined in a US Bureau of Standards cup. To determine exchangeable acidity of the soil, aluminium (Al^{3+}) and hydrogen (H^+) was extracted with 1N KCl and titrated to the end-point with NaOH (0.01M). The acidity was expressed as an equivalent of H^+ in $cmol^{(+)}/dm^3$ soil (The Non-Affiliated Soil Analyses Work Committee, 1990). The Bray No. 2 method, *i.e.* extraction with 0.03 M NH_4F (ammonium-fluoride) in 0.01 M HCl (hydrochloric acid) was used to determine P and K^+. The P and K^+ concentration in the extract was determined by inductively coupled plasma optical emission spectrometry (ICP-OES) using a spectrometer (PerkinElmer Optima 7300 DV, Waltham, Massachusetts). The Ca^{2+}, Mg^{2+}, K^+ and Na^+ were only extracted with 1 M ammonium acetate at pH 7 and their concentrations in the extract were determined by inductively coupled plasma optical emission spectrometry (ICP-OES) using a spectrometer (PerkinElmer Optima 7300 DV, Waltham, Massachusetts). Since the amounts of soluble cations were not determined, the amount of exchangeable cations, which is the extractable minus the soluble amounts (Richards, 1954), could not be calculated. Therefore, the cation exchange capacity (CEC) could not be calculated. Most South African laboratories only determine extractable cations due to the tedious process of determining the exchangeable cations and CEC (Conradie, 1994). Therefore, most laboratories calculate the sum of the extractable cations to obtain an estimated CEC, which is also referred to as the S-value. Given the above-mentioned, the exchangeable potassium percentage (EPP) and exchangeable sodium percentage (ESP) of the soil could not be calculated. However, the extractable potassium percentage (ExPP) was calculated as follows:

$$ExPP = (K^+ \div S) \times 100 \qquad \text{(Eq. 4.1)}$$

where K^+ is the extractable potassium ($cmol^{(+)}/kg$) and S is the S-value ($cmol^{(+)}/kg$), *i.e.* the sum of the Ca^{2+}, Mg^{2+}, K^+ and Na^+.

The extractable sodium percentage (ExSP) was calculated as follows:

$$ExSP = (Na^+ \div S) \times 100 \qquad \text{(Eq. 4.2)}$$

where Na^+ is the extractable sodium ($cmol^{(+)}/kg$) and S is the S-value ($cmol^{(+)}/kg$), *i.e.* the sum of the Ca^{2+}, Mg^{2+}, K^+ and Na^+.

The designation ExPP is used so as not to confuse extractable potassium percentage, which includes both adsorbed K^+ and K^+ in solution, with EPP. Likewise, the designation ExSP is used so as not to confuse extractrable sodium percentage, which includes both adsorbed Na^+ and Na^+ in solution, with ESP. Total organic C contents were determined using the method described by Walkley and Black (1934).

4.2.3. Statistical analysis

Linear and multiple linear regressions were calculated using STATSGRAPHICS® version XV (StatPoint Technologies, Warrenton, Virginia, USA) to develop models to estimate various soil chemical parameters after wastewater application and at bud break. Data for the 2010/11, 2011/12 and 2012/13 seasons was used for the development of the models after wastewater application, whereas data for the 2009/10, 2010/11, 2011/12 and 2012/13 seasons was used to develop models at bud break. Input parameters after wastewater application included the amounts of elements applied *via* the diluted winery wastewater and the dry matter production (DMP) of the pearl millet interception crop. With regard to the rainfall input parameters, the average winter (June-August) rainfall was 300±95 mm, whereas the average rainfall from February to the end of August was 447±78 mm.

Figure 4.1. The extended soil auger, which was used to collect soil samples to 3 m depth in September 2013.

4.3. RESULTS AND DISCUSSION

4.3.1. pH$_{(KCl)}$, acidity and EC$_e$

After wastewater application, there were no clear trends in soil pH$_{(KCl)}$ that could be related to the different levels of dilution of winery wastewater compared to the river water control (data not shown). Similarly, there was no change in soil pH where winery wastewater was used for irrigation of two soils typical of the South Eastern Australia Riverine plains (Quale *et al.*, 2010). In contrast, soil pH$_{(H2O)}$ of a silty clay loam soil that received solid and liquid winery waste for 30 years tended to increase compared to soil where no waste was applied (Mosse *et al.*, 2012). In two case studies where pastures and a vineyard were irrigated with winery wastewater, soil pH also increased (Kumar *et al.*, 2014). However, comparing the results with a historical data set of soil chemical properties, it seemed that irrigation with winery wastewater actually caused a decrease in soil pH. In a laboratory study where mains water, municipal water or winery wastewater was used for irrigation of three different soils, an increase in soil $pH_{(1:5)}$ occurred (Laurenson, 2010). However, it should be kept in mind that the winery wastewater pH in that particular study was 8.5. There has also been conflicting reports of either an increase or decrease in soil pH (Laurenson *et al.*, 2012 and references in). It was suggested that these soil pH changes can be related to the characteristics of the wastewater. If wastewaters contain high concentrations of bicarbonate, application to soils will increase pH whereas acidic wastewaters could reduce soil pH.

Where winery wastewater was diluted to 3000 mg/L COD, soil $pH_{(KCl)}$ increased at bud break after winter rainfall (Fig. 4.2). Since irrigation using winery wastewater is likely to increase soil K^+ and Na^+, soil pH will consequently increase *via* alkaline hydrolyses. This reaction is primarily caused by the hydrolysis of exchangeable cations in soils, *e.g.* K^+_{ex} and Na^+_{ex}, or salts, *e.g.* $CaCO_3$, $MgCO_3$ and Na_2CO_3 (Abrol *et al.*, 1988). Hydrogen ions (H^+) are inactivated by exchange adsorption in the place of exchangeable K^+ and Na^+. These displaced cations do not inactivate the hydroxide anions (OH^-), which in turn cause soil pH to increase (Abrol *et al.*, 1988). The extent to which exchangeable cations hydrolyse depends on their ability to compete with H^+ for exchange sites. Exchangeable Ca^{2+} and Mg^{2+} are more tightly adsorbed to the exchange complex than K^+ and Na^+ (Abrol *et al.*, 1988). Therefore, K^+ and Na^+ are more readily hydrolyzed and produce a higher pH than do exchangeable Ca^{2+} or Mg^{2+}. Hydrolysis of exchangeable Ca^{2+} and Mg^{2+}, in fact, is so limited that it results in a soil having only a mildly alkaline reaction. In the present study, excessive soil K^+ after wastewater application in conjunction with the relatively high winter rainfall in this region induced alkaline hydrolysis, thereby increasing soil $pH_{(KCl)}$ at bud break.

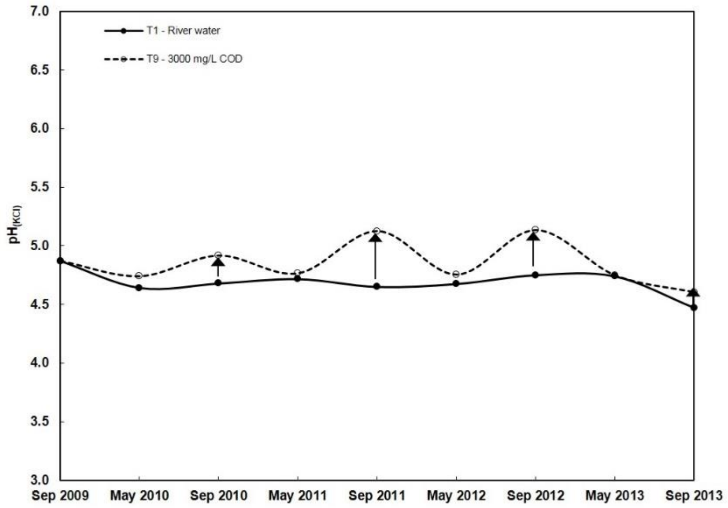

Figure 4.2. Seasonal variation in soil $pH_{(KCl)}$ (0-180 cm depth) for two water qualities in the work rows of a vineyard in a sandy soil near Rawsonville from the beginning to the end of the trial.

The baseline values for $pH_{(KCl)}$ were 5.3 and 4.7 for the 0-90 cm and 90-180 cm soil layer, respectively. Soil $pH_{(KCl)}$ levels remained similar to baseline values until the end of the trial in September 2013 (Tables 4.1 & 4.2) when the soil $pH_{(KCl)}$ in both the 0-90 cm and 90-180 cm layers tended to be lower than the baseline values. In particular, the $pH_{(KCl)}$ in the 90-180 cm layer tended to be below the norm of 5.0 to 7.5 recommended by Saayman (1981) for optimal grapevine growth. However, under the prevailing conditions, visual observations indicated that there were no adverse effects of low sub-soil pH on grapevine performance.

Soil H^+ did not respond to the different levels of dilution of the winery wastewater (data not shown). The baseline values for H^+ were 0.60 $cmol^{(+)}$/kg and 0.85 $cmol^{(+)}$/kg, for the 0-90 cm and 90- 180 cm soil layer, respectively. At the end of the trial in September 2013, H^+ in the 0-90 cm layer was similar to the baseline value whereas the H^+ in the 90-180 cm layer was slightly higher than the baseline value (Tables 4.1 & 4.2).

There were no clear trends in soil EC_e that could be related to the different levels of dilution compared to the river water control (data not shown). However, EC_e was considerably higher after wastewater application compared to bud break. This suggested an accumulation of salts during the grapevine growing season which was mainly due to irrigation with diluted winery wastewater which contains salts (Laurenson *et al.*, 2012). Although there were no consistent trends with regard to EC_e in April 2011, EC_e was more than double that in May 2010. This difference can be attributed to the heavy rainfall in May 2010 before the soil was sampled. In a laboratory study, soil $EC_{(1:5)}$ was not affected by irrigation with either mains water, municipal- or winery wastewater regardless of soil type (Laurenson, 2010). Similarly, also in a laboratory study, soil EC of a loam and loamy sandy soil did not respond to winery wastewater irrigation (Kumar *et al.*, 2006). However, soil EC was higher where woodlots were irrigated with winery wastewater compared to a control (Kumar *et al.*, 2009). In September 2013, *i.e.* the end of the trial, EC_e in the 0-90 cm soil layer was similar to the baseline levels whereas the EC_e in the 90-180 cm layer was slightly higher than the baseline levels (Table 4.1). Therefore, under the prevailing conditions, irrigation using diluted winery wastewater did not cause a long-term accumulation of salts in the soil. However, this does not rule out the possibility that winter rainfall could have leached salts beyond the measured depth. These results confirm the necessity for sufficient rainfall to reduce soil EC_e where winery wastewater, which is known to contain high Na^+ and K^+ levels, is used for irrigation. Furthermore, results emphasize the importance of only irrigating where the roots occur, *i.e.* within the root zone. In heavier textured soils or in regions with lower winter rainfall, less effective leaching is more likely to result in more salt accumulation, and consequently, higher EC_e. During simulated rainfall cycles in a laboratory study, the drainage water EC was substantially higher than that of the input rainwater (Laurenson, 2010), which indicated that there was a net loss of salts during rainfall. Their results emphasized the importance of regular rainfall cycles in reducing high soil EC_e, especially where municipal and winery wastewaters, which contain high levels of salts, are used for irrigation.

4.3.2. Phosphorus (Bray II)

On average, the soil contained 114 mg/kg, 135 mg/kg and 153 mg/kg Bray II-P in the 0-90 cm layer in the vine rows after wastewater application in the 2010/11, 2011/12 and 2012/13 seasons, respectively (Table 4.2). These values were substantially higher than the norm of 20 mg/kg P for sandy soils (*i.e.* ≤ 6% clay) based on Bray II extraction for soils with a $pH_{(KCl)}$ of 5.5 as proposed by Conradie (1994). On average, the soil contained 22 mg/kg, 26 mg/kg and 46 mg/kg Bray II-P in the 90-180 cm soil layer after wastewater application in the 2010/11, 2011/12 and 2012/13 seasons, respectively (Table 4.2). These values were also higher than the norm for sandy soils. Since the grapevines would have absorbed only a small fraction of the available P, the steady incline over time in the profile probably reflected the P applied *via* the wastewater irrigation as well as the 40.5 kg P applied for the cover crops on 30 March 2010 and 30 November 2011.

Perusal of the data revealed soil Bray II-P in the 0-30 cm layer of the work rows increased linearly as the P applied *via* the diluted winery wastewater increased, particularly in the 2011/12 and 2012/13 seasons (Fig. 4.3). The P in the 0-30 cm layer of grapevine rows showed a similar trend (data not shown). However, this trend did not occur in the deeper soil layers. This suggested that the P attenuation only occurred in the top 30 cm of this sandy, alluvial soil which only contained *c.* 3.3% clay. There were no further relationships between soil Bray II-P in the sub-soil layers of both the work and vine rows and P applied *via* the diluted winery wastewater under the prevailing conditions (data not shown).

Baseline values for soil Bray II-P were 153 mg/kg and 29 mg/kg, for the 0-90 cm and 90-180 cm soil layer, respectively. Although soil Bray II-P in the 0-90 cm layer was substantially lower than the baseline values, P levels in the 90-180 cm layer were similar in September 2013 (Tables 4.1). Since the amount of P applied *via* diluted winery wastewater appears to be generally low and would only sustain a grape yield of *c.* 10 t/ha, application of P fertilizers will probably still be necessary to ensure an adequate supply for grapevines.

Table 4.1. Mean values for selected soil chemical parameters as measured in the work rows for the duration of the trial. Data are means for all treatments on a specific date.

Soil parameter	Aug 09	May 2010	Oct. 2010	April 2011	Sep. 2011	May 2012	Sep. 2012	May 2013	Sep. 2013
					0-90 cm				
$pH_{(KCl)}$	5.3	5.1±0.5	6.4±0.9	5.1±0.6	5.1±0.4	5.1±0.5	5.2±0.5	5.1±0.4	4.8±0.5
EC_e (dS/m)	0.058	0.068±0.014	0.059±0.010	0.150±0.050	0.042±0.011	0.200±0.009	0.080±0.020	0.110±0.080	0.050±0.010
H^+ (cmol$^{(+)}$/kg)	0.60	0.69±0.22	0.73±0.24	0.48±0.23	0.67±0.19	0.76±0.34	0.61±0.24	0.43±0.20	0.63±0.32
P (mg/kg)	153	116±74	193±116	110±82	133±72	137±91	116±87	124±91	99±86
Ca^{2+} (cmol$^{(+)}$/kg)	1.72	1.58±0.76	1.52±0.70	1.78±1.08	1.66±0.68	1.73±0.98	1.78±0.90	1.66±0.85	1.55±0.95
Mg^{2+} (cmol$^{(+)}$/kg)	0.80	0.66±0.30	0.74±0.27	0.67±0.35	0.72±0.27	0.73±0.33	0.73±0.33	0.74±0.28	0.73±0.35
C (%)	0.74	0.66±0.17	0.74±0.13	0.66±0.17	0.72±0.22	0.66±0.17	0.63±0.20	0.67±0.27	0.67±0.32
					90-180 cm				
$pH_{(KCl)}$	4.7	4.5±0.1	5.9±0.9	4.4±0.1	4.4±0.3	4.3±0.1	4.4±0.1	4.4±0.2	4.1±0.2
EC_e (dS/m)	0.057	0.067±0.015	0.072±0.022	0.170±0.090	0.064±0.026	0.150±0.050	0.130±0.050	0.110±0.080	0.070±0.050
H^+ (cmol$^{(+)}$/kg)	0.85	0.93±0.36	0.98±0.31	0.75±0.34	1.01±0.29	1.17±0.38	0.97±0.33	0.78±0.39	1.08±0.51
P (mg/kg)	29	27±6	46±32	25±8	47±36	29±13	22±4	35±41	23±10
Ca^{2+} (cmol$^{(+)}$/kg)	0.30	0.37±0.11	0.33±0.31	0.34±0.16	0.52±0.41	0.21±0.17	0.31±0.09	0.44±0.45	0.23±0.13
Mg^{2+} (cmol$^{(+)}$/kg)	0.17	0.15±0.04	0.20±0.13	0.14±0.06	0.24±0.15	0.15±0.10	0.15±0.04	0.24±0.22	0.15±0.11
C (%)	0.44	0.35±0.18	0.47±0.16	0.44±0.20	0.53±0.16	0.56±0.24	0.42±0.19	0.50±0.27	0.41±0.24

Table 4.2. Mean values for selected soil chemical parameters as measured on the grapevine rows for the duration of the trial. Data are means for all treatments on a specific date.

Soil parameter	Aug 09	April 2011	May 2012	May 2013
0-90 cm				
pH$_{(KCl)}$	5.3	5.2±0.5	5.3±0.6	5.1±0.4
EC$_e$ (dS/m)	0.058	0.100±0.040	0.110±0.030	0.060±0.030
H^+ (cmol$^{(+)}$/kg)	0.60	0.59±0.31	0.74±0.37	0.44±0.26
P (mg/kg)	153	114±61	135±77	153±89
Ca^{2+} (cmol$^{(+)}$/kg)	1.72	2.00±1.04	2.01±0.99	1.88±0.92
Mg^{2+} (cmol$^{(+)}$/kg)	0.80	0.84±0.41	0.76±0.34	0.80±0.30
C (%)	0.74	0.71±0.26	0.85±0.26	0.74±0.26
90-180 cm				
pH$_{(KCl)}$	4.7	4.6±0.4	4.6±03	4.6±0.3
EC$_e$ (dS/m)	0.057	0.070±0.020	0.100±0.040	0.060±0.030
H^+ (cmol$^{(+)}$/kg)	0.85	0.77±0.31	0.97±0.29	0.57±0.27
P (mg/kg)	29	22±8	26±9	46±44
Ca^{2+} (cmol$^{(+)}$/kg)	0.30	0.36±0.31	0.42±0.19	0.50±0.50
Mg^{2+} (cmol$^{(+)}$/kg)	0.17	0.17±0.18	0.15±0.09	0.25±0.22
C (%)	0.44	0.38±0.21	0.49±0.16	0.36±0.22

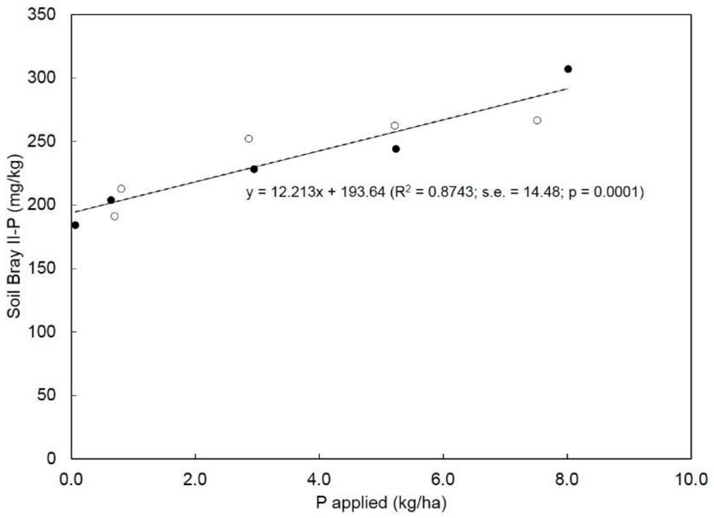

Figure 4.3. Effect of phosphorus (P) applied *via* diluted winery wastewater on soil Bray II-P contents in the 0-30 cm layer in the work rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over two seasons.

4.3.3. Potassium (Bray II)

4.3.3.1 Actual soil Bray II-K

Soil Bray II-K increased linearly with a decrease in wastewater dilution (Figs. 4.4 & 4.5). This was expected since the additional K^+ applied *via* the diluted winery wastewater ranged, on average, from 6.6 kg/ha/year for the river water control (T1) to 177.3 kg/ha/year for the lowest level of dilution (T9). Furthermore, the additional K^+ applied *via* the diluted winery wastewater was applied in the post-veraison period of the grapevine. Most of the K^+ uptake by the grapevine takes place prior to veraison, with almost no uptake from five weeks after harvest (Conradie, 1981). In particular, there was a good correlation between soil Bray II-K in the 0-30 cm layer of the work rows and amounts of K^+ applied *via* the diluted wastewater (Fig. 4.4A). In the 2011/12 and 2012/13 seasons, soil Bray II-K in the 30-60 cm soil layer responded to the amount of K^+ applied *via* the diluted winery wastewater (Fig. 4.4B). With the exception of 2011/12, there were no clear trends with regard to soil Bray II-K in the 60-90cm as well as 90-120 cm soil layer and the amount of additional K^+ applied *via* the diluted winery wastewater (Figs. 4.4C & D).

Similar results were observed in the grapevine rows (Fig. 4.5). It should be noted that the magnitude of the soil Bray II-K in the work and grapevine rows were similar except that the soil Bray II-K in the 60-90 cm laer in the vine row responded better to wastewater dilution levels than on the work rows. Similar results with regard to an accumulation of soil K^+ in response to irrigation with winery wastewater has been reported previously. Where winery wastewater was used for irrigation for over 30 years, an accumulation of K^+ was reported (Mosse *et al*., 2012). Likewise, soil surface K^+ increased where winery wastewater was used for irrigation of two soils typical of the South Estern Australia Riverine plains.for three years (Quale *et al.,* 2010). However, there were no changes in sub-soil K^+ due to slow mobility of K^+ in the soils, which contained *c.* 50% to 60% clay. Soil K^+ levels were also higher in vineyards which were irrigated with winery wastewater compared to control vineyard soils (Kumar *et al.*, 2006). Furthermore, land application of wastewaters can increase the levels of soluble and exchangeable forms of K^+ more rapidly than conventional, inorganic fertilizers (Arienzo *et al.*, 2009). In the only field study of its kind, where simulated winery wastewater was used for vineyard irrigation, the addition of wine to the wastewater enhanced K^+ movement to the sub-soil. Although the fate of K^+ in soils and grapevines irrigated with winery wastewater has received limited attention (Laurenson *et al.,* 2012), it is almost certain that high soil K^+ could lead to an increase in K^+ uptake by grapevines. This could have negative consequences on grapevine responses, such as musts with high pH, malate concentrations and poor colour (Jackson & Lombard, 1993; Mpelasoka *et al.*, 2003; Kodur, 2011). However, the effect of soil K^+ on K^+ concentrations in must is often negligible unless excessive amounts are applied (Jackson & Lombard, 1993).

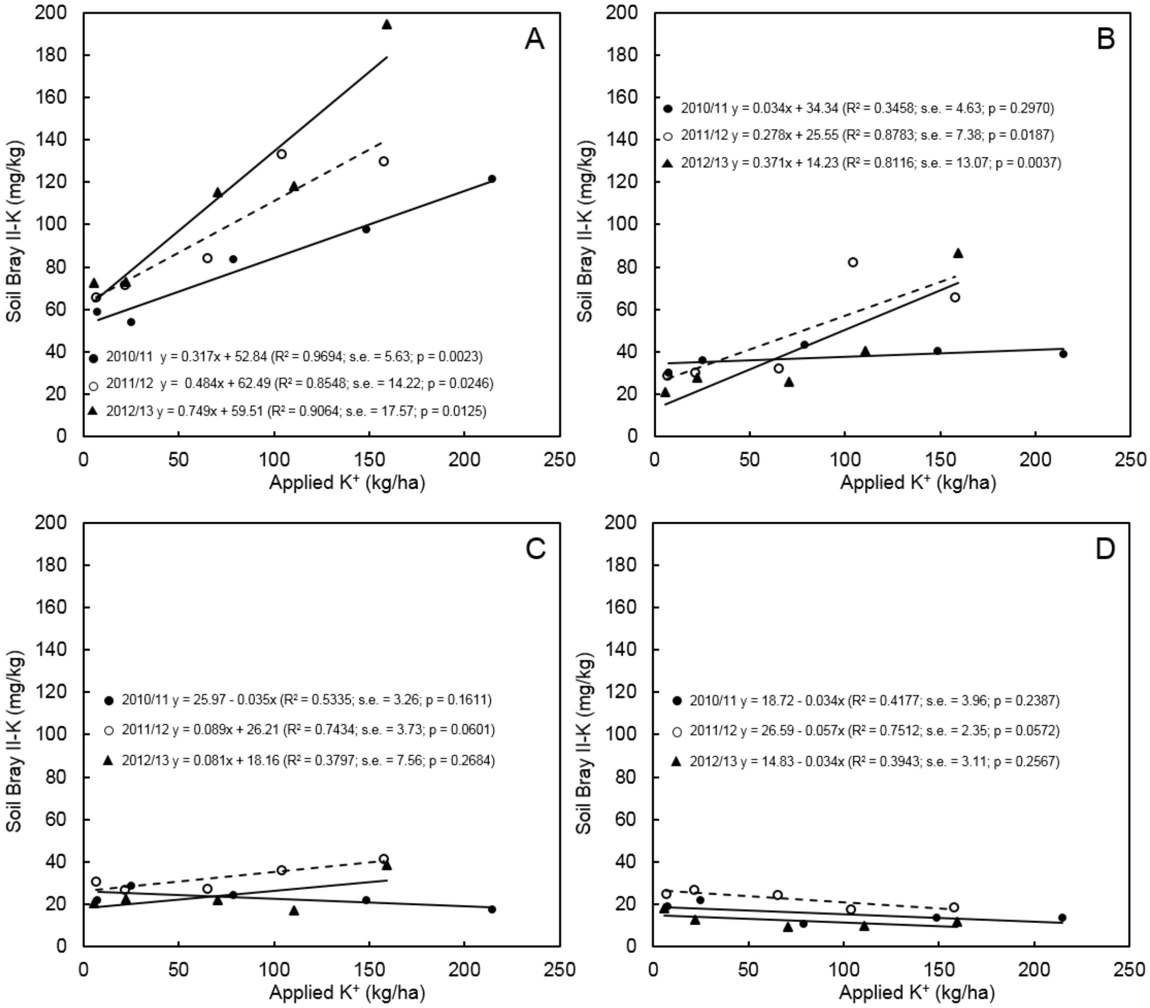

Figure 4.4. Effect of potassium (K^+) applied *via* diluted winery wastewater on soil Bray II-K contents in the (A) 0-30 cm, (B) 30-60 cm, (C) 60-90 cm and (D) 90-120 cm layers in the work rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over three seasons.

In the 0-30 cm soil layer, when the winter rainfall was higher than the average of 300 mm, *i.e.* in the 2011/2012 and 2012/2013 seasons, soil Bray II-K at bud break was substantially lower than after wastewater application, particularly where winery wastewater was diluted to 2000 mg/L COD and higher (data not shown). However, in 2010 and 2011, when there was much less winter rain, soil Bray II-K levels at bud break were similar to levels after wastewater application. Furthermore, soil Bray II-K where winery wastewater was diluted to 2000 mg/L and higher were such that these treatments did not require any K fertilization in the 2010/11 season. With regard to deeper soil layers, during the wetter winters, there was less soil Bray II-K in the 30-60 cm and 60-90 cm soil layers at bud break compared to after wastewater application. In contrast, for the drier winters there were no differences in soil

Bray II-K at bud break compared to after wastewater application. However, it should be noted that in the 2010/11 season, there was, in fact, an accumulation of soil Bray II-K at bud break in the 30-60 cm and 60-90 cm soil layer. Although there is no explanation for this trend, it could be possible that the roots of the pearl millet interception crop absorbed K^+ during wastewater application. Due to favourable, dry winter conditions the roots of the interception crop mineralized, releasing K^+. However, rainfall was too low to leach away the K^+. This indicated insufficient leaching under the prevailing conditions. It should be noted that quantification of interception crop root mineralization was beyond the scope of the study.

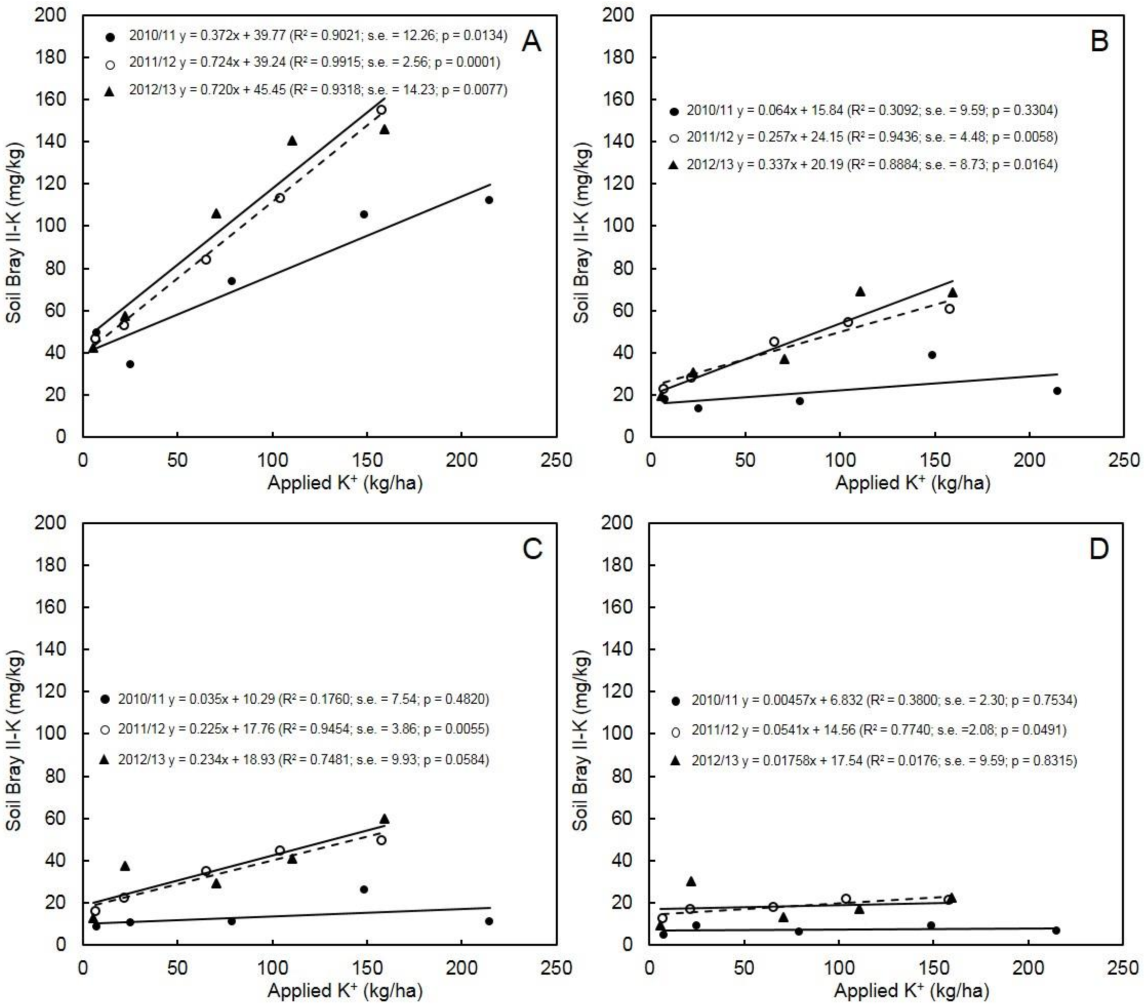

Figure 4.5. Effect of potassium (K^+) applied *via* diluted winery wastewater on soil Bray II-K contents in the (A) 0-30 cm, (B) 30-60 cm, (C) 60-90 cm and (D) 90-120 cm layers in the grapevine rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over three seasons.

4.3.3.2. Modelling soil Bray II-K within the profile

Based on the consistent differences observed between treatments, as well as seasons, models were developed to estimate soil Bray II-K from simple input parameters which could be easily measured. The models were developed for soil Bray II-K levels after wastewater application, as well as at bud break after the winter rainfall. With regard to the prediction models for soil Bray II-K after wastewater application, the amounts of K^+ applied *via* the diluted winery wastewater as well as the dry matter production (DMP) of the pearl millet interception crop were strong input parameters (Table 4.3).

Based on the foregoing, it would be possible to estimate soil Bray II-K in the 0-30 cm layer after wastewater application by means of the following simple, multiple linear regression model:

$$\text{Soil Bray II-K} = 97.5 + 0.534 \times K_{appl} - 6.276 \times DMP_{Pm} \quad \text{(Eq. 4.3)}$$

where K_{appl} is the amount of K^+ applied *via* the diluted winery wastewater (kg/ha) and DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

For the 30-60 cm layer, the simple multiple linear regression model is:

$$\text{Soil Bray II-K} = 51.0 + 0.198 \times K_{appl} - 3.711 \times DMP_{Pm} \quad \text{(Eq. 4.4)}$$

where K_{appl} is the amount of K^+ applied *via* the diluted winery wastewater (kg/ha) and DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

For the 60-90 cm layer, the simple multiple linear regression model is:

$$\text{Soil Bray II-K} = 43.4 + 0.0924 \times K_{appl} - 3.158 \times DMP_{Pm} \quad \text{(Eq. 4.5)}$$

where K_{appl} is the amount of K^+ applied *via* the diluted winery wastewater (kg/ha) and DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

For the 90-120 cm layer, the simple linear regression model is:

$$\text{Soil Bray II-K} = 1 \div (0.0455 + 0.000384739 \times DMP_{Pm}^2) \quad \text{(Eq. 4.6)}$$

where DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

It should be noted that in the case of the estimation of soil Bray II-K in the 90-120 cm layer, K^+ applied *via* the diluted winery wastewater was not significant, and was therefore excluded as an input parameter from the model given in Eq. 4.6.

In general, there was a good relationship between the estimated and actual soil soil Bray II-K contents of three selected treatments, namely river water (Fig. 4.6A), winery wastewater diluted to 1000 mg/L (Fig. 4.6B) and 3000 mg/L COD (Fig. 4.6C), respectively.

Table 4.3. The two independent variables, namely amount of potassium (K^+) applied *via* diluted winery wastewater and the dry matter production (DMP) of a pearl millet interception crop, used in the estimation of soil Bray II-K after wastewater application in a sandy soil.

Dependent variable	Independent variables							
	Applied K^+ (kg/ha)		Pearl millet DMP (t/ha)		Model			
	Coefficient	p	Coefficient	p	N	R^2	s.e	p
Soil K 0-30 cm	0.534	0.0000	-6.276	0.0031	15	0.86	15.30	0.0000
Soil K 30-60 cm	0.198	0.0006	-3.711	0.0077	15	0.68	10.40	0.0011
Soil K 60-90 cm	0.0924	0.0080	-3.157	0.0017	15	0.64	7.10	0.0023
Soil K 90-120 cm			0.000385	0.0039	15	0.49	0.01	0.0039

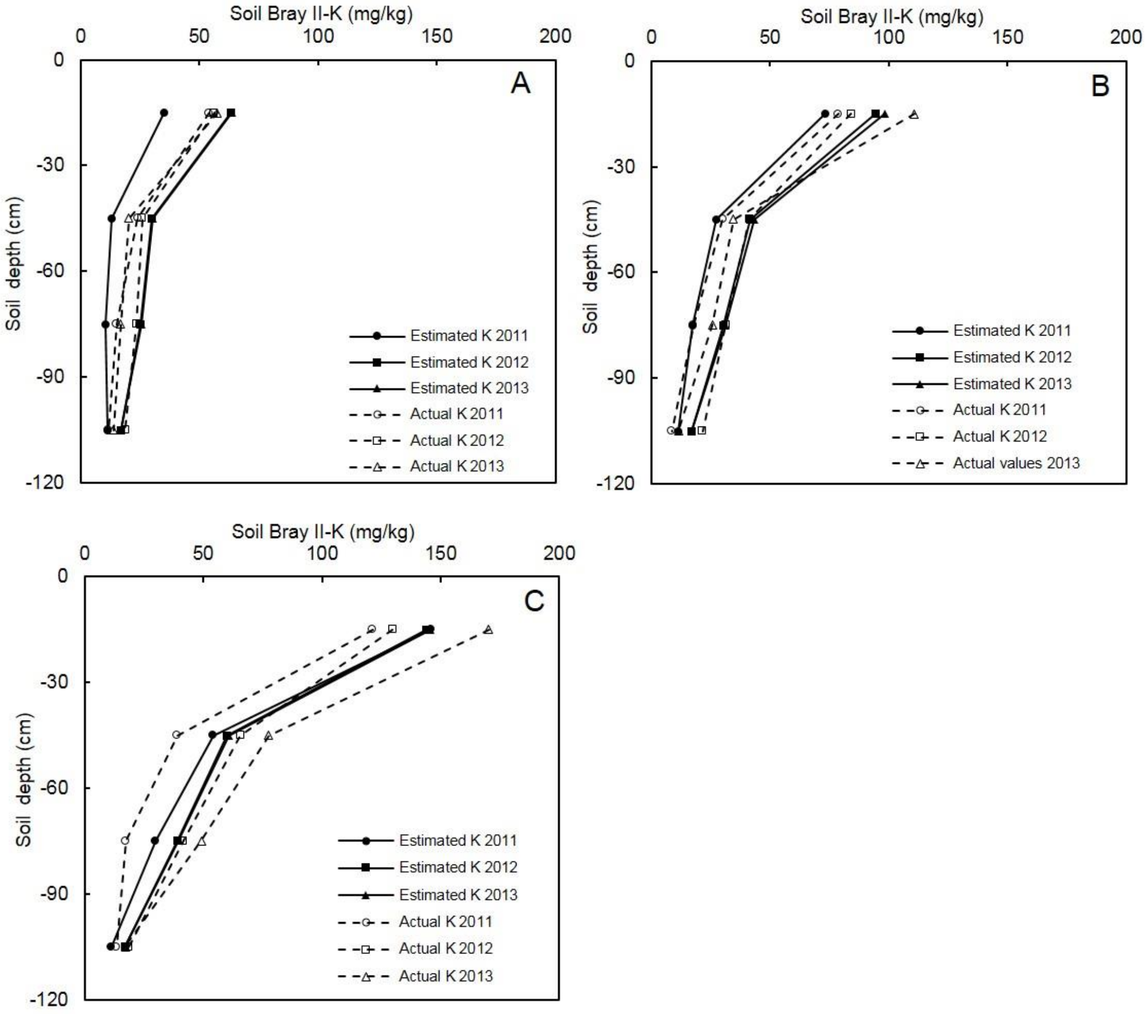

Figure 4.6. Estimated and actual soil Bray II-K contents after wastewater application for (A) river water (T1), (B) 1000 mg/L (T5) and (C) 3000 mg/L chemical oxygen demand (COD) (T9) in a vineyard in a sandy soil near Rawsonville over three seasons.

Using the prediction models given in Eqs. 4.3 to 4.6, soil Bray II-K was calculated for an alluvial, sandy soil after wastewater application for low (Fig. 4.7A), intermediate (Fig. 4.7B) and high (Fig. 4.7C) levels of K^+ applied *via* the diluted winery wastewater. In combination with level of K^+ applied *via* the diluted wastewater, there was either no interception crop, or a pearl millet interception crop of either 5 t/ha and 10 t/ha DMP, respectively. Under the prevailing conditions, the additional K^+ applied *via* the diluted winery wastewater would be sufficient to supply the grapevine's K^+ requirements, if no interception crop were to be cultivated based on the proposed norm of Conradie (1994) for this particular soil. In addition to this, soil Bray II-K levels where winery wastewater is diluted to 3000 mg/L COD, and where there is no interception crop would nearly exceed 200 mg/kg. This is substantially higher than the range of 120 mg/kg to 150 mg/kg recommended by Conradie (1994) for alluvial soils in the Lower Orange River region. This could have negative impacts on grapevine responses as well as soil structure and physical properties. In soils with heavier texture than in the current study, the rate of K^+ accumulation is likely to be even greater. Therefore, in terms of the refinement of the General Authorisations for wineries, the use of diluted winery wastewater for irrigation of vineyards should only be recommended for sandy soils. In addition to this, taking regular water samples of diluted wastewater which is used for irrigation, and monitoring volumes of water applied would be an important management tool.

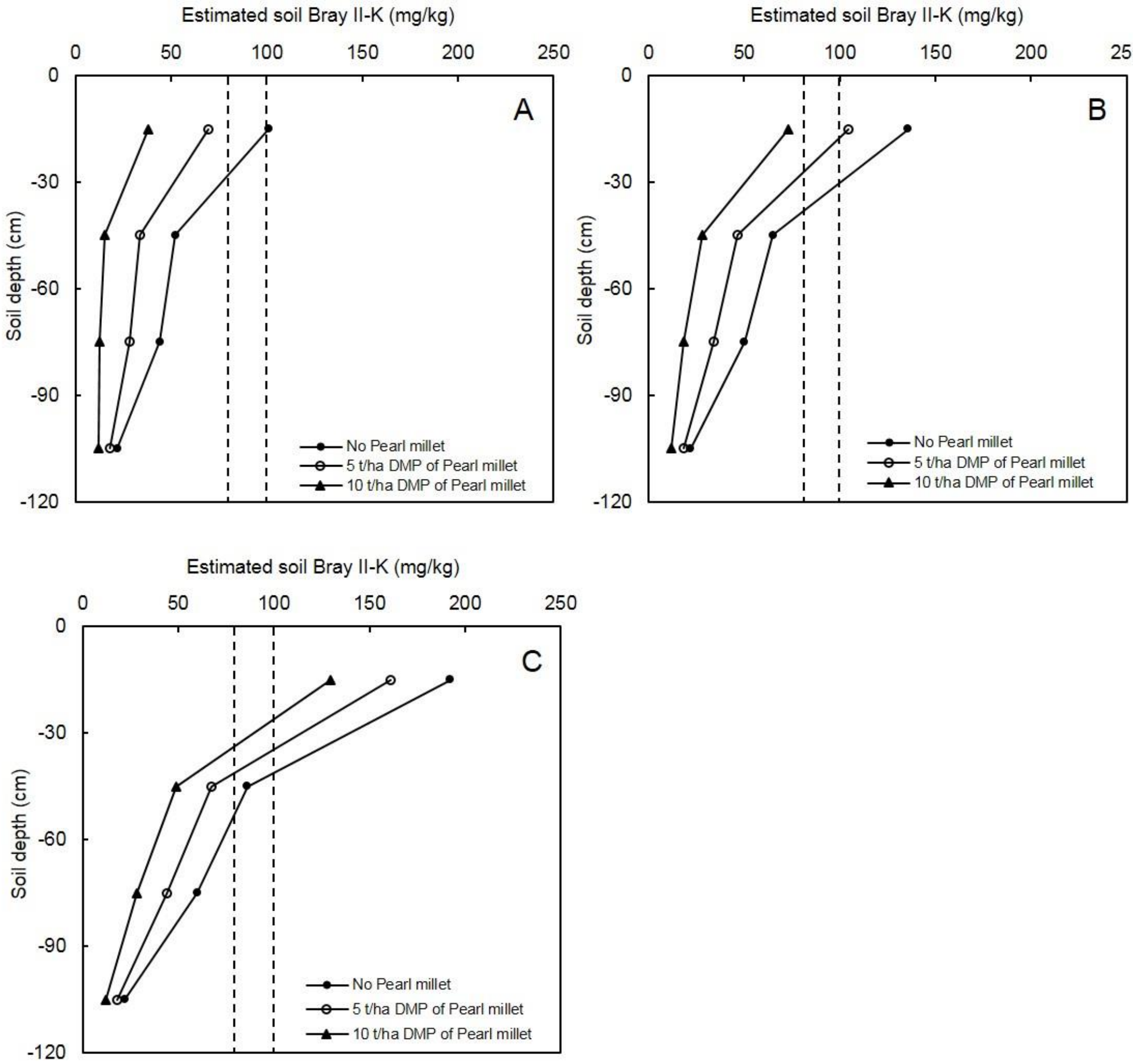

Figure 4.7. The estimated effect of potassium (K^+) applied *via* diluted winery wastewater on Bray II-K in the soil profile after wastewater application where (A) is 6.6 kg, (B) is 71.6 kg and (C) is 177.3 kg K^+ per ha applied *via* diluted winery wastewater in the work rows of a vineyard in a sandy soil where there is no interception crop, or a pearl millet interception crop of 5 t/ha or 10 t/ha dry matter production (DMP), respectively. Dashed vertical lines indicate the optimal norm for Bray II-K in alluvial soils of the Breede River Valley (Conradie, 1994).

Where an interception cover crop of pearl millet of 5 t/ha DMP is cultivated on a sandy soil irrigated using diluted winery wastewater, even an amount of 50 kg/ha additional K^+ applied *via* the diluted wastewater would increase soil Bray II-K (Fig. 4.8) to above the recommended level for an alluvial sandy soil in the Breede River Valley (Conradie, 1994). Under the prevailing conditions, the ideal amount of K^+ applied would range between 50 kg/ha to 100 kg/ha. Results confirm that the K^+ in diluted winery wastewater could be an important source of K^+ for the grapevine. Furthermore, fertilizer costs could be reduced. In soils with heavier texture than in the current study, the K^+ accumulation, particularly in the 0-60 cm layer, is likely to be even more pronounced.

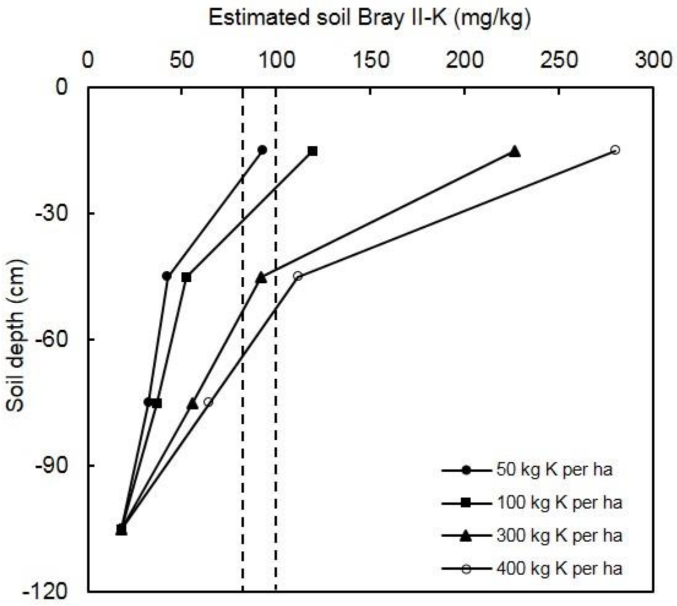

Figure 4.8. The estimated effect of potassium (K^+) applied *via* diluted winery wastewater on Bray II-K contents in the soil profile where 50 kg, 100 kg, 300 kg and 400 kg K^+ per ha is applied to a vineyard in a sandy soil where a pearl millet interception crop of 5 t/ha dry matter production (DMP) is produced. Dashed vertical lines indicate the optimal norm for Bray II-K in alluvial soils of the Breede River Valley (Conradie, 1994).

The following models can be used to predict soil Bray II-K at bud break after the winter rainfall (Table 4.4). It would be possible to estimate soil Bray II-K in the 0-30 cm layer by means of the following simple, multiple linear regression model:

$$\text{Soil Bray II-K} = 91.2 + 0.349 \times \text{Soil Bray II-K}_{\text{AWA0-30}} - 0.174 \times \text{Rainfall}_{\text{JA}} \qquad \text{(Eq. 4.7)}$$

where Soil Bray II-$K_{\text{AWA0-30}}$ is soil Bray II-K in the 0-30 cm layer after wastewater application and $\text{Rainfall}_{\text{JA}}$ is the winter rainfall from June to August (mm).

For the 30-60 cm layer, the simple linear regression model is:

$$\text{Soil Bray II-K} = \exp(1.320 + 1059.66 \div \text{Rainfall}_{\text{FA}}) \qquad \text{(Eq. 4.8)}$$

where $\text{Rainfall}_{\text{FA}}$ is the rainfall from February to August (mm).

For the 60-90 cm layer, the simple linear regression model is:

$$\text{Soil Bray II-K} = 1 \div (0.124 - 39.896 \div \text{Rainfall}_{\text{FA}}) \qquad \text{(Eq. 4.9)}$$

where $\text{Rainfall}_{\text{FA}}$ is the rainfall from February to August (mm).

For the 90-120 cm layer, the simple linear regression model is:

$$\text{Soil Bray II-K} = \exp(1.249 + 868.716 \div \text{Rainfall}_{\text{FA}}) \qquad \text{(Eq. 4.10)}$$

where $\text{Rainfall}_{\text{FA}}$ is the rainfall from February to August (mm).

Table 4.4. The three independent variables, namely soil Bray II-K in the 0-30 cm layer after wastewater application, rainfall from June to August (mm) and from February to August (mm), used in the estimation of soil Bray II-K at bud break after the winter rainfall in a sandy soil.

Dependent variable	Independent variables									
	Soil Bray II-K (mg/kg)		Rainfall (June - August) (mm)		Rainfall (February - August) (mm)		Model			
	Coefficient	p	Coefficient	p	Coefficient	p	N	R^2	s.e	p
Soil K 0-30 cm	0.349	0.0012	-0.174	0.0002			20	0.64	13.16	0.0002
Soil K 30-60 cm					1059.66	0.0001	20	0.60	0.28	0.0001
Soil K 60-90 cm					-39.896	0.0000	20	0.75	0.01	0.0000
Soil K 90-120 cm					868.716	0.0167	20	0.28	0.05	0.0167

Using Eqs. 4.7 to 4.10 to estimate soil Bray II-K at bud break where there was either lower or higher, or equivalent to average rainfall, should 11.46 kg K^+ per ha be applied *via* diluted wastewater, soil Bray II-K would range from 49 mg/kg to 82 mg/kg (Fig. 4.9A). This is lower than the recommended norm which ranges from 80 mg/kg to 100 mg/kg for this particular soil (Conradie, 1994). In contrast, should 177 kg/ha and 300 kg/ha K^+ be applied *via* diluted wastewater, soil Bray II-K would range from 80 mg/kg to 113 mg/kg (Fig. 4.9B) and 103 mg/kg to 135 mg/kg (Fig. 4.9C) in the 0-30 cm soil layer, respectively, at bud break. With regard to the magnitude of the rainfall, as expected, higher rainfall leads to more extensive leaching and lower soil Bray II-K, whereas rainfall lower than the average rainfall would lead to less leaching of K^+, and, consequently, higher soil Bray II-K. Therefore, in the case of the highest level of dilution of winery wastewater or, alternatively, where low levels of K^+ are applied *via* the wastewater, should the amount of rain be similar or one standard deviation wetter than average, soil K^+ would be leached from the profile to such an extent that it would be necessary to apply K fertiliser to supply the vineyard with sufficient K^+ (Fig. 4.9A). However, in drier winters the amount of K^+ applied *via* the wastewater could be sufficient to maintain the soil at the norm recommended by Conradie (1994). For higher levels of K^+ applied *via* diluted winery wastewater, soil Bray II-K, irrespective of the magnitude of rainfall, would be sufficient to supply the grapevine's requirement (Fig. 4.9C). Therefore, in this particular case, should *c.* 177 kg/ha of K be applied *via* diluted wastewater, it would not be necessary to apply K^+ fertiliser in the following season. For substantially higher levels of K^+ applied, *i.e.* 300 kg/ha, soil Bray II-K, irrespective of the magnitude of rainfall, the risks of excessive K^+ accumulation are much higher than in the other two cases, even on a sandy, alluvial soil,would be sufficient to supply the grapevine's requirement. These results clearly illustrate the effect of magnitude of winter rainfall on soil Bray-II K at bud break. Furthermore, with regard to the refinement of the General Authorisations for wineries, results show that the re-use of diluted winery wastewater can be recommended in regions where the average rainfall is similar to that of the Breede River Valley region in order to prevent excessive accumulation of K^+ in the soil. It should be noted that the soil should have a similar texture to the alluvial soil in this study. However, this does not exclude the possibility that drier winters in this region could cause excessive K^+ levels to develop should more than 177 kg K^+ per ha be applied *via* diluted winery wastewater. In order to calculate the amount of K^+ applied via the wastewater, it is important that samples of diluted wastewater used for irrigation are taken regularly to monitor element concentrations. Also, the amount of water applied *via* irrigation should be measured by means of water meters. Using this information, the amount of element applied *via* the diluted wastewater can be accurately assessed.

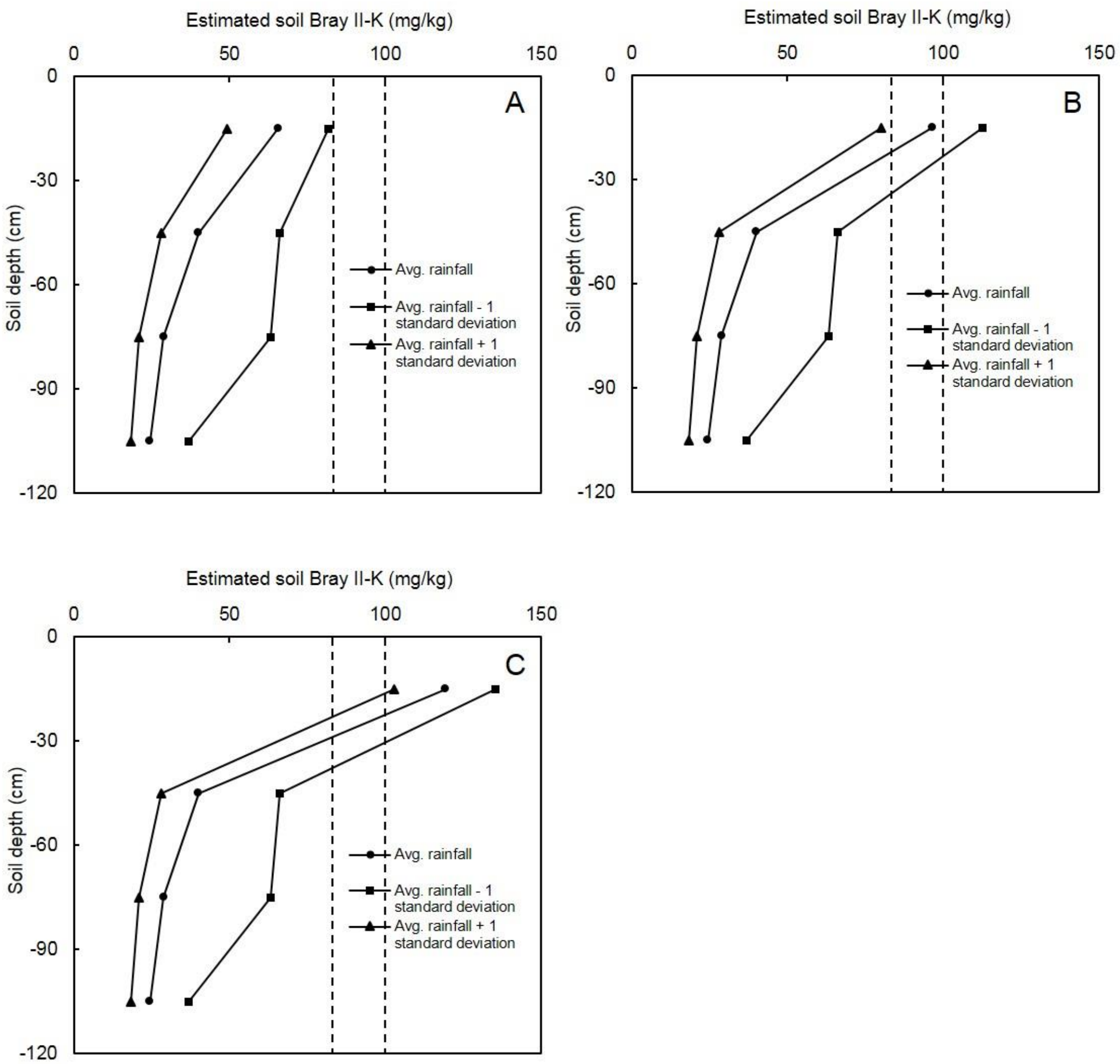

Figure 4.9. The estimated effect of rainfall on soil Bray II-K in the soil profile at bud break after winter rain where (A) 11.46 kg, (B) 177.34 kg and (C) 300 kg potassium (K^+) per ha is applied *via* diluted winery wastewater to a vineyard in a sandy soil where a pearl millet interception crop of 5 t/ha dry matter production (DMP) is produced. Dashed vertical lines indicate the optimal norm for Bray II-K in alluvial soils of the Breede River Valley (Conradie, 1994).

4.3.3.2 Soil Bray II-K at the end of the trial

With the exception of the 0-30 cm soil layer, it was evident that the heavy winter rainfall had negated treatment differences with respect to soil Bray II-K at bud break in September 2013 (Fig. 4.10). This implied that the heavy winter rainfall probably leached K^+ from the soil profile, and this was substantiated by mineral analysis of soil samples collected deeper than 180 cm with a modified soil auger. At the end of the trial in September 2013, soil Bray II-K levels in the 0-30 cm soil layer where winery wastewater was diluted up to 2000 mg/L COD, were lower than the baseline level. In contrast, where winery wastewater was diluted to 3000 mg/L, baseline levels were maintained in the 0-30 cm soil layer. Therefore, under the prevailing conditions, using winery wastewater diluted to 3000 mg/L COD was beneficial with regard to soil K^+ status.

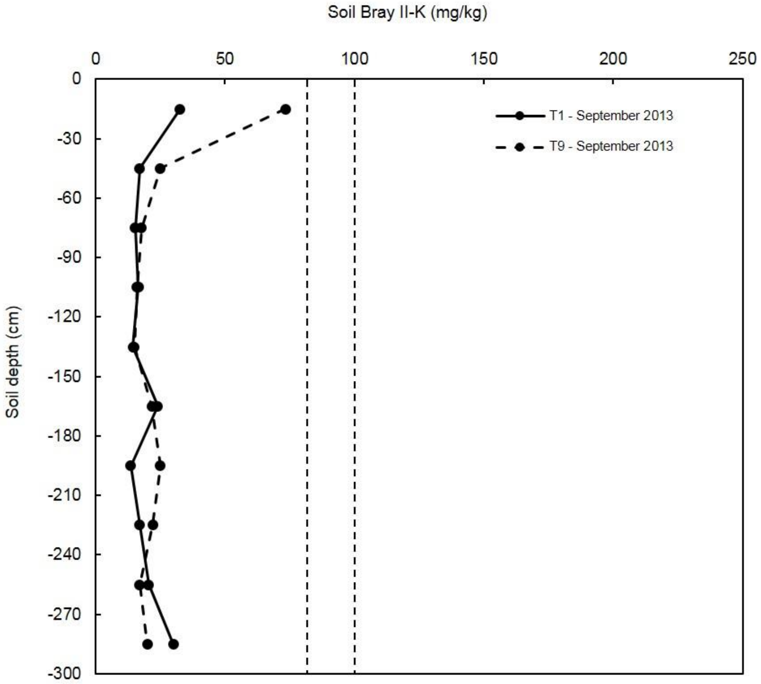

Figure 4.10. Soil Bray II-K contents over 3 m depth measured near Rawsonville in September 2013. Dashed vertical lines indicate the optimal norm for Bray II-K in alluvial soils of the Breede River Valley (Conradie, 1994).

4.3.4. Extractable cations

4.3.4.1. Calcium and magnesium

Soil Ca^{2+} and Mg^{2+} did not show any consistent trends with respect to the different levels of wastewater dilution (data not shown). The lack of response could be expected since there were no substantial differences with regard to the amounts of additional Ca^{2+} and Mg^{2+} applied to the vineyard *via* the diluted winery wastewater. Similar results for soil Ca^{2+} were also reported where winery wastewater was used for irrigation for three years on two soils typical of the South Estern Australia Riverine plains (Quale *et al.,* 2010). However, in that particular study soil Mg^{2+} tended to decrease. The baseline values for Ca^{2+} were 1.72 $cmol^{(+)}$/kg and 0.30 $cmol^{(+)}$/kg, for the 0-90 cm and 90-180 cm soil layer, respectively. The baseline values for Mg^{2+} were 0.80 $cmol^{(+)}$/kg and 0.17 $cmol^{(+)}$/kg, for the 0-90 cm and 90-180 cm soil layer, respectively. At bud break in September 2013, soil Ca^{2+} and Mg^{2+} were similar to these baseline values (Tables 4.1 & 4.2). This confirmed that irrigation of vineyards with diluted winery wastewater is unlikely to be beneficial with regard to increased Ca^{2+} and Mg^{2+} supply for grapevines. In addition to this, if applied in such small amounts, these elements will not be able to counter the negative effects should high levels of Na^+ be applied *via* diluted winery wastewater.

4.3.4.2. Potassium

Extractable K^+ exhibited similar trends to soil Bray II-K (data not shown) and will therefore not be discussed further. Since exchangeable K^+ was not determined in the laboratory, the ExPP rather than the EPP was calculated. For the Western Cape fruit industry, the recommended ratio of exchangeable K^+ to other cations is 3% to 4% (Conradie, 1994). The ExPP in the 0-30 cm soil layer could consistently be related to the amount of K^+ applied *via* the diluted winery wastewater (Fig. 4.11), and values were near the upper threshold of the recommended norm (Conradie, 1994). This implies that if even more K^+ is applied to the soil *via* diluted winery wastewater, excessive K^+ could accumulate, causing even higher ExPP. Depending on the timing of the wastewater applications, there is the risk that the excessive K^+ could be applied when the grapevine is actively absorbing K^+ in the post veraison period (Conradie, 1981). In the case of red wine production, this could cause high wine pH, and wine instability (Mpelasoka *et al.*, 2003; Kodur, 2011).

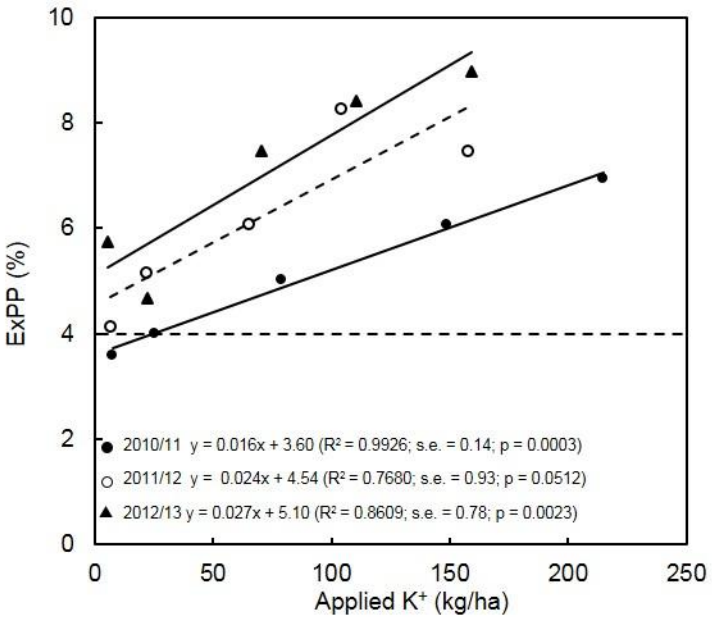

Figure 4.11. Effect of potassium (K^+) applied *via* diluted winery wastewater on soil extractable potassium percentage (ExPP) in the 0-30 cm layer in the work rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over three seasons. Dashed horizontal line indicates the critical extractable potassium percentage (ExPP) threshold for grapevines (Conradie, 1994).

Approximately 75% of the variation in ExPP in the 0-30 cm soil layer after wastewater application could be explained by means of the following multiple linear regression model:

$$\text{Soil ExPP} = 6.99535 + 0.0218053 \times K_{appl} - 0.347127 \times DMP_{Pm} \qquad \text{(Eq. 4.11)}$$

where K_{appl} is the amount of K^+ applied *via* the diluted winery wastewater (kg/ha) and DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

For the 30-60 cm layer, the simple linear regression model could explain 48% of the variation in soil ExPP:

Soil ExPP = $1 \div (0.225693 + 1.03855 \div K_{appl})$ ……. (Eq. 4.12)

where K_{appl} is the amount of K^+ applied *via* the diluted winery wastewater (kg/ha).

With regard to ExPP at bud break, 57% of the variation in ExPP in the 0-30 cm soil layer could be explained by means of the following multiple linear regression model:

Soil ExPP = $4.49851 + 0.559417 \times$ soil $ExPP_{AWA0\text{-}30}$ - $0.00952629 \times Rainfall_{JA}$ (Eq. 4.13)

where soil $ExPP_{AWA0\text{-}30}$ is the soil extractable potassium percentage in the 0-30 cm layer after wastewater application and $Rainfall_{JA}$ is the winter rainfall from June to August (mm).

For the 30-60 cm layer, the simple linear regression model could explain 59% of the variation in soil ExPP:

Soil ExPP = $9.79224 - 0.0175314 \times Rainfall_{JA}$ (Eq. 4.14)

where $Rainfall_{JA}$ is the winter rainfall from June to August (mm).

For the 60-90 cm layer, the simple linear regression model could explain 49% of the variation in soil ExPP:

Soil ExPP = $13.0863 - 0.0207164 \times Rainfall_{JA}$ (Eq. 4.15)

where $Rainfall_{JA}$ is the winter rainfall from June to August (mm).

With regard to the refinement of the General Authorisations for wineries, the potassium adsorption ratio (PAR) of the wastewater has not been adopted as a quality parameter. Considering that winery wastewater contains high levels of K^+ and that the soil K^+ responded consistently to the K^+ applied *via* the diluted winery wastewater, the use of the PAR of the wastewater should be considered as a further indicator of the wastewater quality. However, further research is needed to refine PAR norms for wastewater quality.

4.3.4.3. Sodium

4.3.4.3.1 Actual sodium

Work row soil Na^+ in the 0-30 cm as well as the 60-90 cm soil layers increased linearly with a decrease in wastewater dilution, *i.e.* an increase in the COD level of the irrigation water (Figs. 4.12A & C). This was expected since the additional Na^+ applied *via* the diluted winery wastewater ranged, on average, from 37.6 kg/ha/year for the river water control to 92.6 kg/ha/year for the lowest level of dilution. The relatively high Na^+ in the Holsloot River has already been discussed in Chapter 3. At this stage, there is no explanation why soil Na^+ levels in the 30-60 cm soil layer did not respond consistently to the different levels of dilution compared to the 0-30 cm as well as 60-90 cm soil layers. Several studies have also reported an increase in soil Na^+ as a response to irrigation with wastewater (Mosse *et al*., 2012). In a field study where grapevines were irrigated with simulated winery wastewater, soil Na^+ levels increased in the 0-20 cm, as well as the 20-40 cm soil layer (Mosse *et al*., 2013).

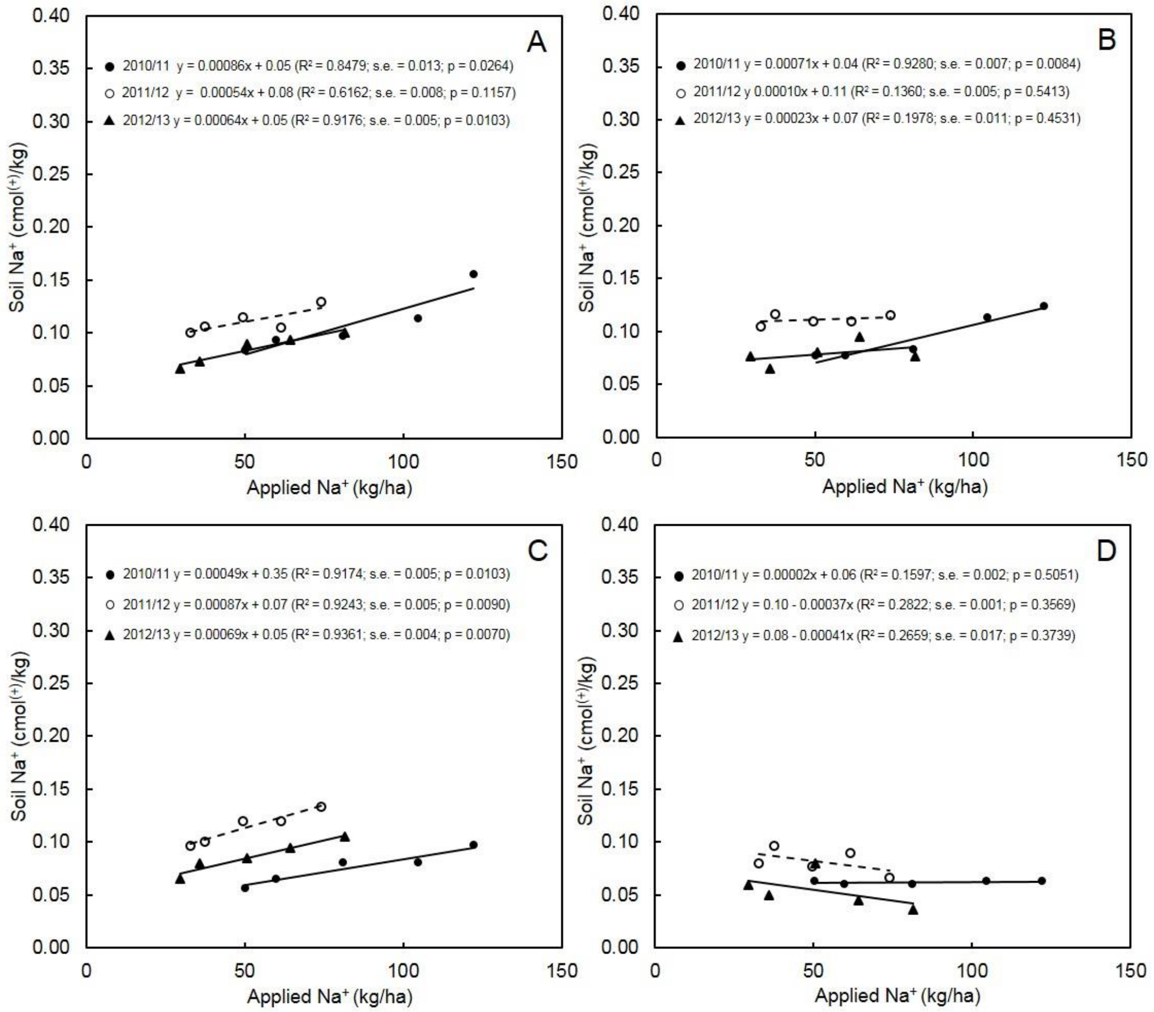

Figure 4.12. Effect of sodium (Na^+) applied *via* diluted winery wastewater on soil Na^+ contents in the (A) 0-30 cm, (B) 30-60 cm, (C) 60-90 cm and (D) 90-120 cm layers in the work rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over three seasons.

At bud break, soil Na^+ in the 0-30 cm soil layer was consistently lower than after wastewater application (data not shown). Likewise, when winters were wetter, the soil Na^+ in the 30-60 cm and 60-90 cm soil layers were lower at bud break compared to after wastewater application. However, when winters were drier, *i.e.* 2010 and 2011, trends were not consistent in deeper soil layers. Therefore, when winter rainfall was higher, there was sufficient leaching to remove the Na^+ from the root zone. However, the redistribution and accumulation of Na^+ in the root zone during the drier winters is a cause for concern.

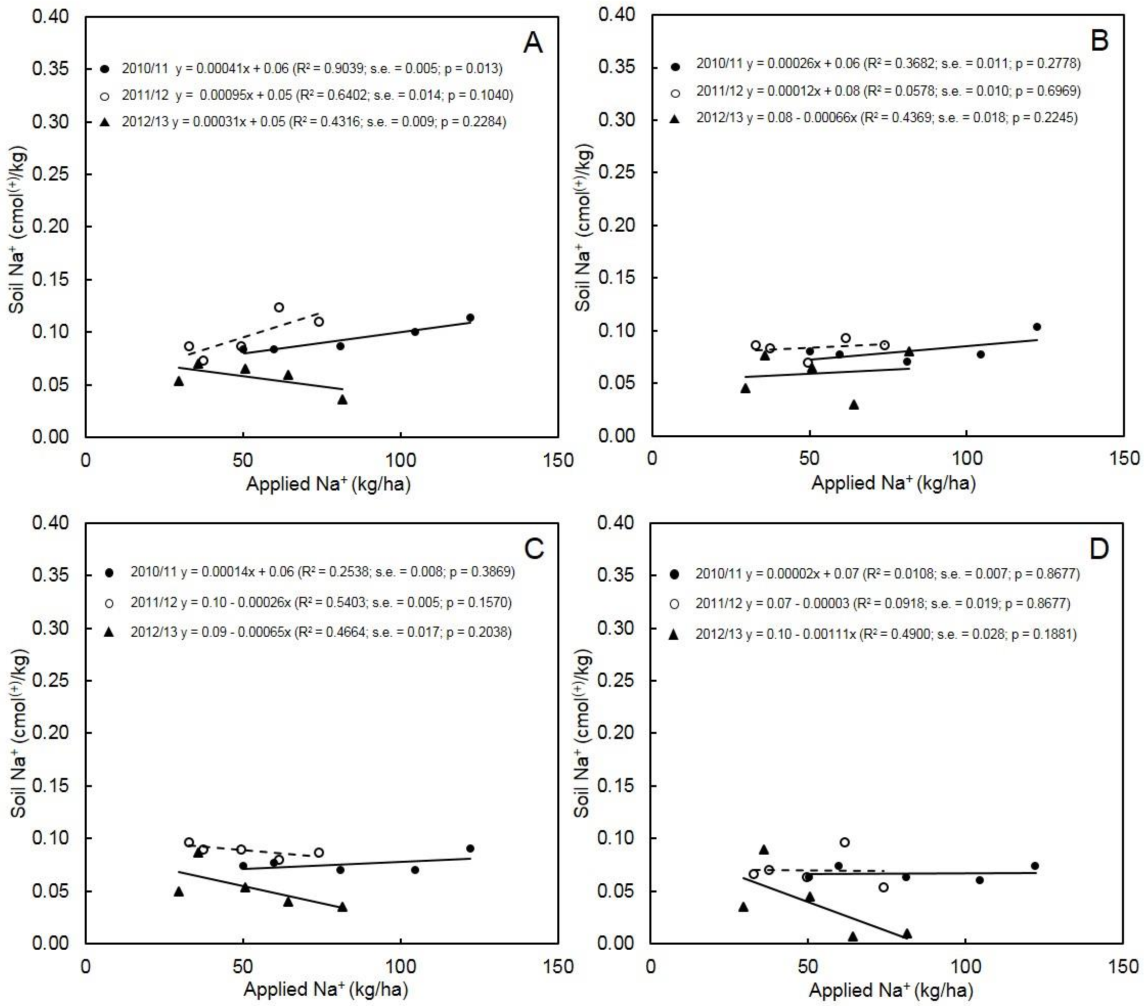

Figure 4.13. Effect of sodium (Na^+) applied *via* diluted winery wastewater on soil Na^+ contents in the (A) 0-30 cm, (B) 30-60 cm, (C) 60-90 cm and (D) 90-120 cm layers in the grapevine rows of a vineyard in a sandy soil near Rawsonville measured after wastewater application over three seasons.

4.3.4.3.2 Modelling Na^+ within the profile

Based on the consistent differences observed between treatments, as well as seasons, it was decided to develop models to estimate soil Na^+ from simple input parameters which could be easily measured. The models were developed to predict soil Na^+ levels after wastewater application (Table 4.5), as well as at bud break after the winter rainfall (Table 4.6). With regard to the prediction models for soil Na^+ after wastewater application, the amounts of Na^+ applied *via* the diluted winery wastewater was an important input parameters. However, in contrast to the estimation of soil Bray II-K where DMP of the pearl millet interception crop emerged as a strong input parameter, DMP of pearl millet was generally an insignificant parameter when estimating soil Na^+.

Based on the foregoing it would be possible to estimate the soil Na^+ in the 0-30 cm layer after wastewater application by means of the following simple, linear regression model:

Soil $Na^+ = \mathrm{sqrt}(0.00648338 + 9.33858E\text{-}7 \times Na_{appl}^2)$ (Eq. 4.16) where Na_{appl} is the amount of Na^+ applied *via* the diluted winery wastewater (kg/ha).

For the 30-60 cm layer, the simple linear regression model is:

$$\text{Soil } Na^+ = \text{sqrt}(0.00740477 + 4.26821E\text{-}7 \times Na_{appl}^2) \qquad \text{(Eq. 4.17)}$$

where Na_{appl} is the amount of Na^+ applied *via* the diluted winery wastewater (kg/ha).

For the 60-90 cm layer, the simple multiple linear regression model is:

$$\text{Soil } Na^+ = 0.114418 + 0.000616649 \times Na_{appl} - 0.00813162 \; DMP_{Pm} \qquad \text{(Eq. 4.18)}$$

where Na_{appl} is the amount of Na^+ applied *via* the diluted winery wastewater (kg/ha) and DMP_{Pm} is the dry matter production of the pearl millet interception crop (t/ha).

For the 90-120 cm layer, the simple linear regression model is:

$$\text{Soil } Na^+ = \text{sqrt}(0.00796653 - 0.000432853 \times \text{sqrt}(Na_{appl}) \qquad \text{(Eq. 4.19)}$$

where Na_{appl} is the amount of Na^+ applied *via* the diluted winery wastewater (kg/ha).

There was a good relationship between estimated and actual soil Na^+ of three selected treatments, namely river water, winery wastewater diluted to 1000 mg/L and 3000 mg/L COD, respectively (data not shown). Under the prevailing conditions, Na^+ application *via* diluted winery wastewater in excess of 400 kg/ha would increase the soil Na^+ to levels higher than the threshold of 0.4 $cmol^{(+)}$/kg recommended for this particular soil (W.J. Conradie, personal communication) (Fig. 4.14).

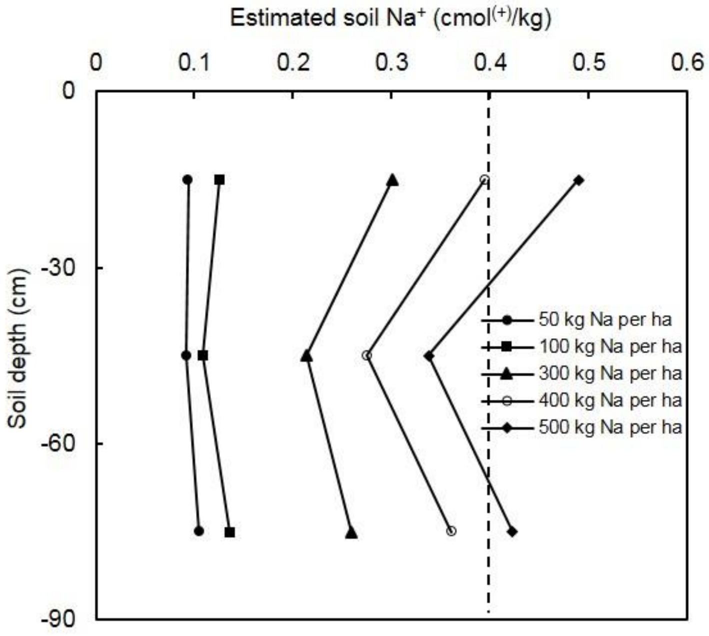

Figure 4.14. The estimated effect of sodium (Na^+) applied *via* diluted winery wastewater on the soil Na^+ contents in the soil profile after wastewater application where 50 kg, 100 kg, 300 kg, 400 kg and 500 kg Na^+ per ha is applied to a vineyard in a sandy soil where a pearl millet interception crop of 5 t/ha dry matter production (DMP) is produced. Dashed vertical line indicates the critical Na^+ threshold for this particular soil (Conradie, personal communication).

Table 4.5. The two independent variables, namely amount of sodium (Na$^+$) applied *via* diluted winery wastewater and the dry matter production (DMP) of a pearl millet interception crop, used in the estimation of soil Na$^+$ after wastewater application in a sandy soil.

Dependent variable	Independent variables							
	Applied Na$^+$ (kg/ha)		Pearl millet DMP (t/ha)		Model			
	Coefficient	p	Coefficient	p	N	R^2	s.e	p
Soil Na$^+$ 0-30 cm	9.34E-7	0.0009			15	0.58	0.003	0.0009
Soil Na$^+$ 30-60 cm	4.27E-7	0.0828			15	0.21	0.003	0.0828
Soil Na$^+$ 60-90 cm	0.0006	0.0125	-0.008	0.0035	15	0.53	0.016	0.0102
Soil Na$^+$ 90-120 cm	0.0004	0.2436			15	0.10	0.002	0.2436

Table 4.6. The three independent variables, namely soil sodium (Na$^+$) in the 30-60 cm and 60-90 cm soil layer after wastewater application and rainfall from February to August (mm) used in the estimation of soil Na$^+$ at bud break after the winter rainfall in a sandy soil.

Dependent variable	Independent variables									
	Soil Na$^+$ (30-60 cm) (cmol$^{(+)}$/kg)		Soil Na$^+$ (60-90 cm) (cmol$^{(+)}$/kg)		Rainfall (February - August) (mm)		Model			
	Coefficient	p	Coefficient	p	Coefficient	p	N	R^2	s.e	p
Soil Na$^+$ 0-30 cm					0.00004	0.0001	20	0.59	2.38	0.0001
Soil Na$^+$ 30-60 cm	0.628	0.0002			-0.00016	0.0005	20	0.73	0.01	0.0000
Soil Na$^+$ 60-90 cm			0.495	0.0011	-0.00023	0.0001	20	0.68	0.01	0.0001
Soil Na$^+$ 90-120 cm					-0.00449	0.0000	20	0.65	0.23	0.0000

Should the additional Na^+ applied *via* the diluted winery wastewater increase to 500 kg/ha, there would also be a substantial increase in soil Na^+ in the sub-soil after wastewater application. It is expected that Na^+ accumulation would occur to a greater extent in soils with heavier textures than the sandy, alluvial soil containing *c.* 3.3% clay which was used in the current study.

In order to prevent the accumulation of soil Na^+, a halophytic interception crop such as fodder beet (*Beta vulgaris* L. Brigadier) could be used to absorb excess Na^+ as it has been reported that it can absorb up to 178 kg Na^+ per ha (Myburgh & Howell, 2014).

It would be possible to estimate the soil Na^+ in the 0-30 cm layer at bud break by means of the following simple, linear regression model:

$$\text{Soil Na}^+ = 1/(4.81792 + 0.0000423129 \times \text{Rainfall}_{FA}^2) \qquad \text{(Eq. 4.20)}$$

where Rainfall_{FA} is the rainfall from February to August (mm).

For the 30-60 cm layer, the simple multiple linear regression model is:

$$\text{Soil Na}^+ = 0.100904+0.627604 \times \text{Soil Na}_{AWA30\text{-}60} - 0.000164219 \times \text{Rainfall}_{FA} \qquad \text{(Eq. 4.21)}$$

where $\text{Soil Na}_{AWA30\text{-}60}$ is soil Na^+ in the 30-60 cm layer after wastewater application and Rainfall_{FA} is the rainfall from February to August (mm).

For the 60-90 cm layer, the simple multiple linear regression model is:

$$\text{Soil Na}^+ = 0.138816 +0.495405 \times \text{Soil Na}_{AWA60\text{-}90} - 0.000232851 \times \text{Rainfall}_{FA} \qquad \text{(Eq. 4.22)}$$

where $\text{Soil Na}_{AWA60\text{-}90}$ is soil Na^+ in the 60-90 cm layer after wastewater application and Rainfall_{FA} is the rainfall from February to August (mm).

For the 90-120 cm layer, the simple linear regression model is:

$$\text{Soil Na}^+ = \exp(-0.776641 - 0.00448983 \times \text{Rainfall}_{FA} \qquad \text{(Eq. 4.23)}$$

where Rainfall_{FA} is the rainfall from February to August (mm).

Using the models given in Eqs. 4.20 to 4.23 to estimate soil Na^+ at bud break, with the exception of two data points, there was a good relationship between estimated and actual soil Na^+ contents of three selected treatments, namely river water, winery wastewater diluted to 1000 mg/L and 3000 mg/L COD (data not shown). Regardless of the amount of Na^+ applied *via* diluted winery wastewater and magnitude of rainfall, levels of soil Na^+ were still below 0.4 cmol(+)/kg (Fig. 4.15). However, should substantially more Na^+ be applied *via* the wastewater, there is a more pronounced accumulation and redistribution in the deeper soil layers (Fig. 4.15C). Therefore, in the case of heavier textured soil it seems likely that the accumulation and redistribution of Na^+ would occur to an even greater extent. Results confirm that using diluted winery wastewater for vineyard irrigation of a deep, sandy soil with excessive drainage could lead to pollution of natural water sources.

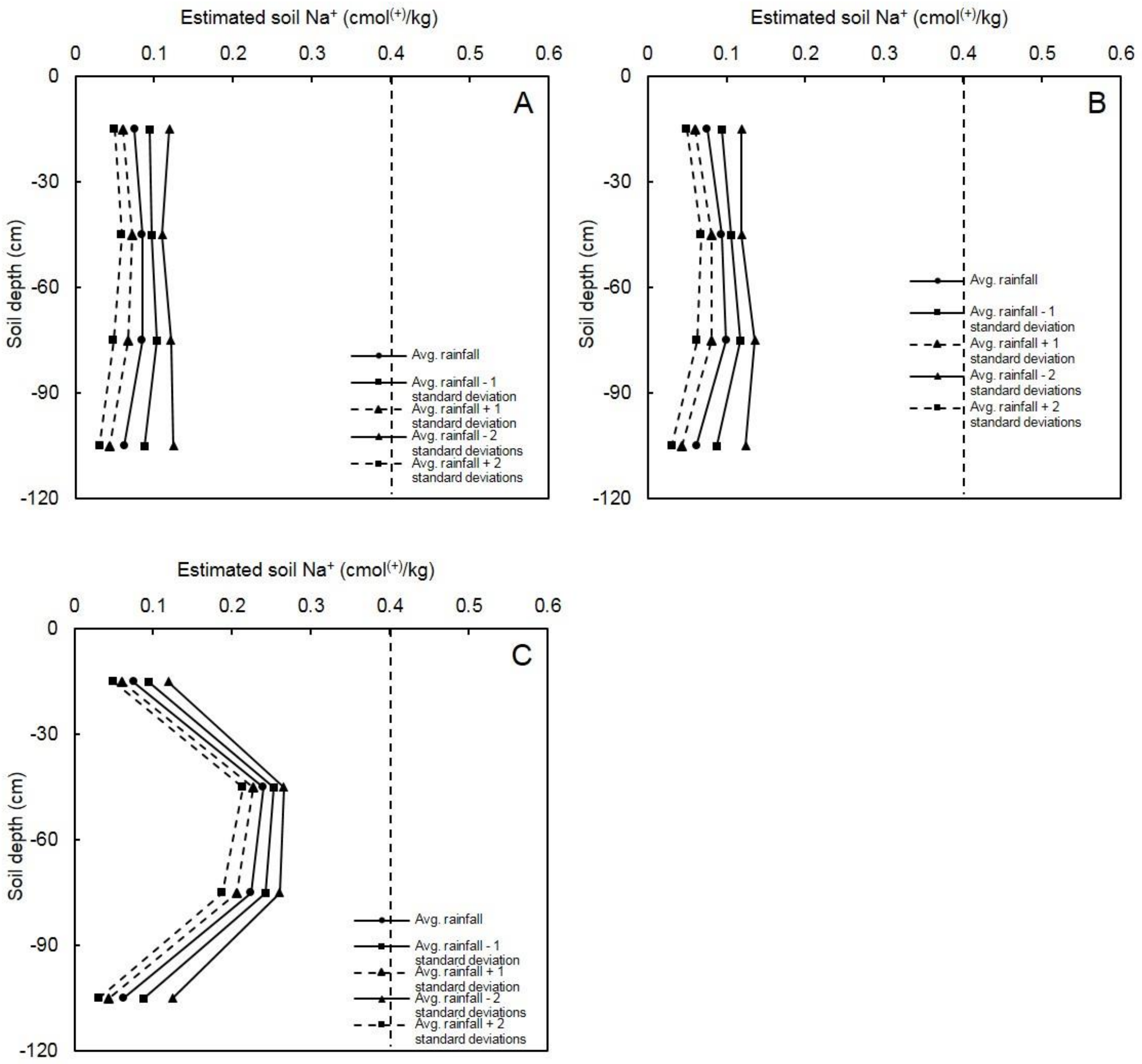

Figure 4.15. The estimated effect of rainfall on the distribution of sodium (Na^+) contents in the soil profile at bud break after the winter rain where (A) 44 kg, (B) 93 kg and (C) 500 kg per ha Na^+ is applied *via* diluted winery wastewater to a vineyard in a sandy soil where a pearl millet interception crop of 5 t/ha dry matter production (DMP) was produced. Dashed vertical line indicates the critical Na^+ threshold for this particular soil (Conradie, personal communication).

In general, the ExSP did not exceed the critical threshold of 15% for sustainable agricultural use (Laker, 2004; Seilsepour *et al.*, 2009). The models developed to estimate ExSP after wastewater application in the respective soil layers were insignificant, with low R^2.

It would be possible to estimate the soil ExSP in the 0-30 cm layer at bud break means of the following simple, linear regression model, where R^2 was 42%:

$\text{Soil ExSP} = 1/(0.216304 + 0.00000138595423129 \times \text{Rainfall}_{FA}^2)$ (Eq. 4.24)

where Rainfall_{FA} is the rainfall from February to August (mm).

For the 30-60 cm layer, the simple multiple linear regression model could explain 36%of the variation in ExSP:

$\text{Soil ExSP} = 3.33178 + 0.366519 \times \text{Soil ExSP}_{AWA30\text{-}60} - 0.00529303 \times \text{Rainfall}_{JA}$ (Eq. 4.25) where Soil $ExSP_{AWA30-60}$ is soil extractable sodium percentage in the 30-60 cm layer after wastewater application and $Rainfall_{JA}$ is the rainfall from June to August (mm).

4.3.4.3.2. Sodium at the end of the trial

After the heavy rainfall of winter 2013, Na^+ levels of T1, T3 and T5 were lower than baseline levels (data not shown). Soil Na^+ at the end of the trial for the lowest level of dilution, *i.e.* winery wastewater diluted to 3000 mg/L COD was not substantially more than that of the river water control (Fig. 4.16). Therefore, under field conditions, using diluted winery wastewater for vineyard irrigation did not have any long term negative consequences on soil Na^+. However, for a heavier textured soil or where rainfall is substantially less than that of the Breede River valley, the accumulation of Na^+ in the soil could be more prominent.

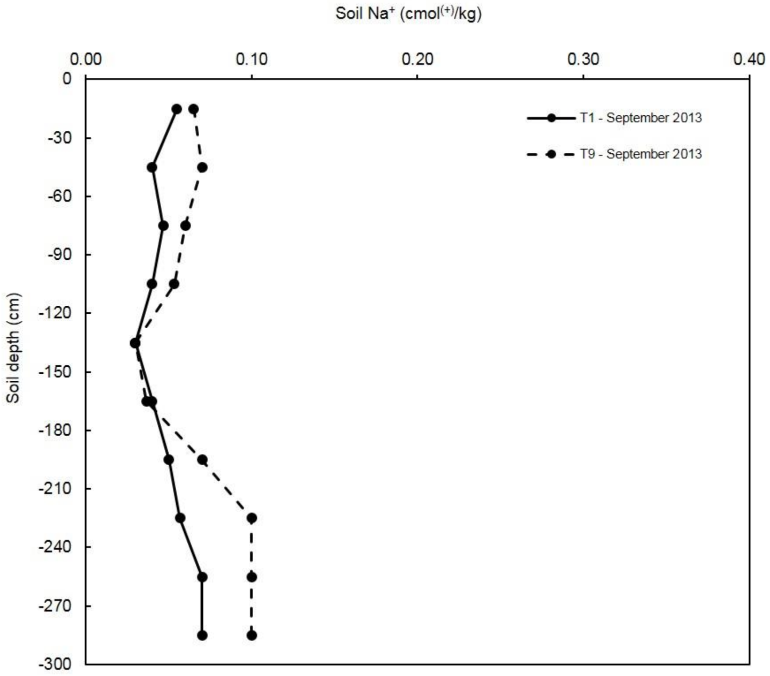

Figure 4.16. Soil sodium (Na^+) contents over 3 m depth measured near Rawsonville in September 2013.

4.3.5. Organic carbon

There were no consistent trends with regard to soil organic C that could be related to the level of dilution of the winery wastewater (data not shown). This indicated that the organic C content in the diluted wastewaters was still too low to have had a positive effect on soil fertility. It is also possible that organic material in the diluted wastewaters, which could have led to an accumulation of organic soil C, decomposed when the soil was aerated between irrigations. In contrast, Kumar *et al.* (2009) reported that higher organic carbon content of winery wastewater resulted in an increased total organic carbon content in soils irrigated using such wastewater. The baseline values for C were 0.74% and 0.44% for the 0-90 cm and 90-180 cm soil layer, respectively. At the end of the trial in September 2013, soil C was

0.67% and 0.41% for the 0-90 cm and 90-180 cm soil layer, respectively (Table 4.1). These levels were therefore similar to baseline values.

4.4. CONCLUSIONS

Where diluted winery wastewater was used for irrigation of a vineyard in a sandy, alluvial soil there was a consistent increase in soil Bray II-K after wastewater application. The increase in soil Bray II-K was linearly related to the additional amounts of K^+ applied *via* the diluted winery wastewater. Soil K^+ increases could have a negative impact on wine colour stability should it be taken up by the grapevine in sufficient quantities, particularly if soil K^+ accumulates to such an extent that it is luxuriously adsorbed by grapevines. Soil Ca^{2+} and Mg^{2+} did not respond to levels of dilution of the winery wastewater. This was probably due their low levels in the diluted winery wastewater. Soil Na^+ also increased linearly as the level of wastewater dilution decreased, particularly in the top-soil. In heavier textured soils or in regions with lower winter rainfall, soil K^+ and Na^+ could accumulate to levels where it could impact negatively on soil physical conditions or grapevine growth and yield. In addition, natural water resources could be polluted with these elements during winter. Changes in cation ratios due to the accumulation of K^+ and Na^+ with no consequent increase in Ca^{2+} and Mg^{2+} could be detrimental in terms of soil physical properties. It should be noted that the results represent a specific in-field situation, *i.e.* in the presence of rainfall and crops. Under the prevailing conditions, results indicated that the General Authorisations for wineries could be refined to permit irrigation of vineyards in sandy soils using winery wastewater diluted up to 3000 mg/L COD.

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CHAPTER 5: EFFECT OF IRRIGATION USING DILUTED WINERY WASTEWATER ON SELECTED GRAPEVINE AND WINE RESPONSES

5.1. INTRODUCTION

Although wineries produce large volumes of low quality wastewater which can contain high levels of organic matter, potassium (K^+) and sodium (Na^+), information on the actual volumes produced is extremely limited. Recent studies have shown that *c.* 3 to 5 m^3 of winery wastewater is produced per tonne of grapes crushed (Mosse *et al*., 2011). The chemical status of winery wastewater is generally worse than the legislated limits for irrigation with wastewater (Department of Water Affairs, 2013). On the other hand, limited irrigation water supplies could be restricted further in future irrigation water allocations (Van Zyl & Weber, 1981; Petrie *et al.*, 2004). Where wineries are surrounded by vineyards, irrigation using diluted winery wastewater could be used instead of water from natural resources. If winery wastewater could be re-used to irrigate vineyards, with no detrimental impacts on either grapevines or subsequent wine quality and chemical composition, it could be a possible viable alternative to using either river or recycled municipal water.

Currently, the Department of Water and Sanitation is drafting new General Authorisations for wineries. Depending on the permitted water quality limits and volumes stipulated by the new authorisations, diluting winery wastewater with current irrigation water may well become a more viable practice in the future. Re-using winery wastewater in this way will be beneficial, particularly where there are water shortages. In such situations, re-using winery wastewater will have a positive impact on grape yields if additional irrigation could be applied. Water saving and higher yields will also contribute to sustainability and economic viability of wine production. In addition to these benefits of re-using diluted winery wastewater for irrigation, the nutrients in the wastewater could reduce the necessity to apply fertilisers, and, consequently, reduce the cost of fertilization (Neilsen *et al.*, 1989a; Kumar *et al.*, 2014). In particular, K^+ in winery wastewater could make a meaningful contribution to the annual K^+ requirement of the grapevine. Where winery wastewater was diluted on a field-scale for vineyard irrigation, additional K^+ applied to the vineyard *via* the diluted winery wastewater ranged, on average, from 6.6 kg/ha/year for the river water control to 177.3 kg/ha/year where winery wastewater was diluted to 3000 mg/L chemical oxygen demand (COD) (Refer to Chapter 3). The land application of wastewaters can increase soluble and exchangeable forms of K^+ more rapidly than with conventional, inorganic fertilizers, and most of the K^+ is available immediately (Arienzo *et al.*, 2009). Although it appears that the nitrogen (N) load in diluted winery wastewater would be inadequate to supply the grapevine’s requirement, phosphorus (P) and K^+ applied *via* diluted winery wastewater should be adequate for a grape yield of *c.* 10 t/ha (Refer to Chapter 3). Presently, there is also increasing pressure on producers to use water in a more environmentally friendly way.

In the first study of its kind, winery wastewater diluted up to 3000 mg/L chemical oxygen demand (COD) did not pose any salinity hazard, since the irrigation water electrical conductivity (EC_{iw}) was well below 2 dS/m (Refer to Chapter 3). Considering the other classical water quality criteria, *i.e.* pH and sodium adsorption ratio (SAR), dilution of winery wastewater up to 3000 mg/L COD produced irrigation water of which the quality would permit sustainable vineyard irrigation under the prevailing conditions, *i.e.* Mediterranean climate with high winter rainfall and sandy soil. Although there is extensive literature available regarding the effect of irrigation with wastewaters of various origins on plant responses, there is much less available information for fruit trees and grapevines. Where sewage water was used to irrigate grapevines by means of drip irrigation of *c.* 22 mm water per week from September until March there was an increase in yield compared to good quality reservoir water for one season (McCarthy, 1981). The use of sewage water rather than good quality reservoir water did not affect cane mass. However, when sewage water was used for irrigation, harvest petiole magnesium (Mg^+), sodium (Na^+) and chloride (Cl^-) increased (McCarthy, 1981). Although wine P, K^+ and Mg^{2+} were higher where grapevines were irrigated in response to irrigation with sewage water, concentrations were not excessively high (McCarthy & Downton, 1981). In contrast, wine Na^+ and Cl^- were substantially higher. There were no differences with regard to wine quality. Irrigation with municipal wastewater increased N, P and K^+ in apple tree leaves, and increased trunk diameter (Neilsen *et al.*, 1989b). In a similar trial where Okanagan Riesling grapes were irrigated with municipal wastewater, petiole P, K^+ and calcium (Ca^{2+}) increased (Neilsen *et al.*, 1989a). Furthermore, wastewater irrigation increased yield. With regard to sweet cherries, municipal wastewater increased leaf N, P, K^+, boron (B^{3+}) and manganese (Mn^{2+}) whereas Ca^{2+} and Mg^{2+} were reduced (Neilsen *et al.*, 1991). Where table grape vineyards were irrigated with treated wastewater, yield was not affected after six years (Netzer *et al.*, 2014) but petiole Na^+ increased substantially. The use of recycled municipal wastewater for irrigation reduced leaf N of Soultanina grapevines, whereas leaf P and K^+ increased (Paranychianakis *et al.*, 2006). Yield was also reduced substantially, and this was probably due to a reduction in average leaf area (Paranychianakis *et al.*, 2004).

Although there is extensive literature available regarding the irrigation of grapevines with saline water (Walker *et al*., 1997; Stevens *et al*., 1999; Ben-Asher *et al*., 2006; Stevens *et al.*, 2011), there is no information on using winery wastewater diluted to a pre-determined COD level on grapevine growth, yield and juice responses. Where "simulated" winery wastewater was used for vineyard irrigation, there were no substantial differences in ripeness parameters, yield and vegetative growth after one year (Mosse *et al.*, 2013). Although high K^+ concentrations in artificial wastewater promoted the accumulation of harvest petiole K^+, petiole Ca^{2+} was reduced substantially. When artificial wastewater contained organic matter together with high K^+ levels, petiole Ca^{2+} was not reduced to the same extent. The use of Na^+ based artificial wastewater increased petiole Na^+ levels substantially. In a glass house study, where winery wastewater was applied either undiluted, or diluted in different ratios to potted Shiraz grapevines, irrespective of level of dilution, petiole K^+ contents were below recommended levels (Kumar *et al.*, 2014). In addition to the different levels of winery wastewater dilution, there were also treatments where solutions of differing K^+ and Na^+ nutrient loads were used to irrigate grapevines. Increasing K^+ concentrations increased petiole K^+ (Kumar *et al.*, 2014). The authors concluded that their results indicated that solutions should not be used to study winery wastewater effects. On a field-scale, in two paired field trials where grapevines were irrigated with either main water or winery wastewater, there was no difference in sensorial evaluation of the wines (Kumar *et al.*, 2014). Furthermore, where grapevines were irrigated with winery wastewater, wine Na^+ levels were still below 100 mg/L, whereas wine K^+ ranged from 1220 mg/L to 1400 mg/L, and were within industry norms for red wines in Australia (Kumar *et al.*, 2014).

It has been reported previously that winemakers are reluctant to use winery wastewater for vineyard irrigation due to high Na^+ and K^+ (Kumar *et al.*, 2014). Potassium is the predominant cation involved in the pH balance of grape juice or wine, and there is a good correlation between pH and K^+ concentration in juice and wine (Kodur, 2011 and references therein). During winemaking, high wine K^+ increases the precipitation of tartaric acid, consequently reducing free tartaric acid (Kodur, 2011). Therefore, a high concentration of K^+ in wine makes pH adjustment difficult and expensive (Kumar *et al.*, 2014). High juice K^+ can lead to a reduced tartaric/malic acid ratio which is undesirable for the production of high quality wines (Mpelasoka *et al.*, 2003). Elevated berry K^+ will influence the effect of other cations present and is thought to have impacts on fermentation and microbial activity as well as other wine properties such as taste, bitterness and sourness (Boulton, 1980; Kumar *et al.*, 2014). According to Jackson and Lombard (1993), high juice K^+ are not only associated with high pH but also poor colour of red wines. Although high concentrations of K^+ in the soil are correlated with levels in the plant, the effect of soil K^+ on juice levels is small unless excessive K^+ is applied. Although application of wastewater with high K^+ levels will increase soil fertility, long-term application may cause an accumulation of soil K^+ (Kumar *et al.*, 2014) and decrease the soil hydraulic conductivity (Arienzo *et al.*, 2009).

The objective of this study was to determine the effect of using diluted winery wastewater for vineyard irrigation rather than river water on grapevine water status, growth, yield, evapotranspiration (ET_c) as well as juice and wine quality characteristics in order to make recommendations for the refinement of the General Authorisations for wineries.

5.2. MATERIALS AND METHODS

Grapevines were irrigated with winery wastewater diluted to different COD levels (Table 5.1). Details of the viticultural aspects, experimental layout, water quality, amounts of elements applied *via* the diluted winery wastewater and soil responses are presented in Chapters 2, 3 and 4.

5.2.1. Soil water content

The objective was to apply irrigation only within the grapevine root zone, *i.e.* < 90 cm, in order to prevent leaching to the deeper layers. The soil water content in the experimental vineyard was measured using the neutron scattering technique. Access tubes were installed in the grapevine row in all plots. Soil water content was measured over 30 cm increments to a depth of 1.8 m. A field calibration was carried out to convert neutron counts to volumetric soil water content. Soil water content was measured weekly from October, as well as before and after irrigations. After irrigations stopped in either April or May, soil water content was measured every two weeks throughout winter.

5.2.2. Grapevine water status

Grapevine water status was quantified by measuring grapevine water potential in mature, unscathed leaves on primary shoots by means of the pressure chamber technique (Scholander *et al.*, 1965), according to the protocol described by Myburgh (2010). Predawn (Ψ_{PD}) and midday (Ψ_L) leaf water potentials, as well as midday stem (Ψ_S) water potential were measured in one leaf per plot. For Ψ_S measurements, leaves were covered in aluminium bags (Choné *et al.*, 2001; Myburgh, 2010) for at least one hour before the measurements were carried out. Since the diluted wastewater irrigations only commenced after harvest in 2010, grapevine water status was not determined in the 2009/10 season. During the 2010/11 season, Ψ_{PD}, Ψ_L, and Ψ_S were measured during berry development (December) and berry ripening (March). During the 2011/12 and 2012/13 growing seasons, Ψ_L and Ψ_S were only measured on selected days during berry ripening.

5.2.3. Vegetative growth

5.2.3.1. Cane mass

To quantify growth vigour, cane mass at pruning (July) was weighed per experimental plot using a hanging balance. Shoot mass per plot (kg) was converted to tonnes per hectare.

5.2.3.2. Leaf and shoot chemical status

In order to allow maximum exposure to the wastewater *via* the irrigation, leaf samples were collected prior to harvest in the 2010/11 to 2012/13 seasons. Thirty mature, unscathed leaves opposite a bunch on the second spur were sampled per plot in accordance with the protocol of Conradie (1994). Petioles were immediately separated from the leaf blade. Due to high costs of chemical analyses, only leaf blade and shoot samples of Replication 2 were analysed. Shoot samples consisting of four primary canes per plot were collected at pruning in July. All of the samples were dried in a fan oven at 60˚C for 24 hours. The dried leaf blade and shoot contents were determined by a commercial laboratory (BEMLAB, Strand). Leaf and shoot nitrogen (N) was measured by means of a nitrogen analyser using the methods decribed by Horneck and Miller (1998). Samples were prepared for analysis of P, K^+, Ca^{2+}, Mg^{2+}, Na^+, Mn^{2+}, iron (Fe^{2+}), copper (Cu^{2+}), zinc (Zn^{2+}) and B^{3+} and analysed by means of an ICP-OES spectrometer (PerkinElmer Optima 7300 DV, Waltham, Massachusetts, U.S.A.) using methods described by Isaac and Johnson (1998).

5.2.4. Yield and its components

To determine berry mass at harvest, ten randomly selected bunches were picked from each experimental plot for all the treatments. Twenty berries were sampled from each of these bunches in order to obtain a sample of 200 berries. Berry mass was determined in the laboratory by weighing the samples using an electronic balance. At harvest, all bunches of the experimental grapevines on each plot were picked and counted. Grapes were weighed using a top loader mechanical balance to obtain the total mass per experimental plot. The number of bunches per grapevine was calculated by dividing the total number of bunches per plot by the number of experimental grapevines per plot. Grape mass per grapevine (kg/grapevine) was calculated and converted to yield (t/ha).

5.2.5. Evapotranspiration

The ET_c was determined by calculating a soil water balance on a weekly basis as described by Myburgh and Howell (2007). Monitoring soil water content to 1.8 m showed that almost no deep percolation occurred during the irrigation season. Consequently, drainage losses were not accounted for in the soil water balance equation. Daily ET_c was used to calculate mean monthly values.

5.2.6. Juice characteristics

Grape samples were collected at harvest from all experimental plots, and analysed for total soluble solids (TSS), total titratable acidity (TTA) and pH according to standard procedures of the winery at the Infruitec-Nietvoorbji Institute of the Agricultural Research Council (ARC) near Stellenbosch. Juice was obtained by gently crushing berries sampled at harvest and the resultant juice squeezed through cheese cloth. To determine total N, juice was digested with selenic acid and concentrated sulphuric acid. Total N was then determined by means of a nitrogen analyser using methods described by Clesceri *et al*. (1998). To determine P, K^+, Ca^{2+}, Mg^{2+} and Na^+, juice samples were digested by adding concentrated nitric acid, allowing it to stand overnight and then adding perchloric acid. Following the nitric acid/perchloric acid digestion, the above-mentioned elements were determined using an inductively coupled plasma emission spectrometer (Liberty 200 ICP AES, Varian, Australia).

5.2.7. Wine quality

Grapes were harvested when they reached the target sugar content of 24°B. In 2010/11 and 2011/12, four wastewater irrigations were applied prior to harvest whereas in 2012/13, three wastewater irrigations were applied prior to harvest. Wines were made from the grapes (*c.* 40 kg) of each experimental plot according to the standard procedure for making red wine used by the experimental winery at ARC Infruitec-Nietvoorbij as described by Myburgh (2011b). After six months, the wines were evaluated sensorially by a panel of at least 12 industry experts. In order to determine whether the wines were safe for tasting, *i.e.* free of harmful bacteria, the wine samples were first analysed for the presence of total bacteria, coliforms and *E. coli* by a commercial laboratory (BEMLAB, Strand) during all three seasons. Wines were evaluated on an unmarked line scale of 100 mm for wine colour, overall intensity, vegetative character, berry character, spicy character, acidity, body, astringency and overall quality. The panel was also asked to give an indication of the occurrence of off-flavours (off-odours and off-tastes) and any other atypical red wine characteristics. Following tasting, the alcohol, extract, residual sugar, glucose, fructose, volatile acidity, tartaric acid, malic acid, total acidity and pH of the wines were analysed by a commercial laboratory (Koelenhof winery, Stellenbosch) as described by Schoeman (2012). The ion composition of the wine was analysed using the same procedure as described above for the juice.

5.2.8. Statistical analyses

The data were subjected to an analysis of variance. Least significant difference (LSD) values were calculated to facilitate comparison between treatment means. Means that differed at $p \leq 0.05$ were considered to be significantly different. STATGRAPHICS® version XV (StatPoint Technologies, Warrenton, Virginia, USA).was used for the analyses of variance.

5.3. RESULTS AND DISCUSSION

5.3.1. Soil water content

During the four seasons, irrigation using diluted winery wastewater had no effect on the soil water status compared to river water. Therefore, only trends in the mean soil water content for each season will be presented and discussed.

2009/10 season: When the field work commenced, the vineyard was drip irrigated once a week for 12 hours from the end of November until February when the micro-sprinkler system was installed. For the duration of the field trial, all irrigations were applied by means of micro-sprinklers. Since the grower applied the drip irrigation according to a continuous deficit strategy, the soil was relatively dry at that stage (Fig. 5.1). Consequently, the objective of the first micro-sprinkler irrigation was to wet the total soil volume thoroughly using river water. Since the infrastructure was only completed at the end of January 2010, irrigation using diluted winery wastewater only commenced after the grapes had been picked.

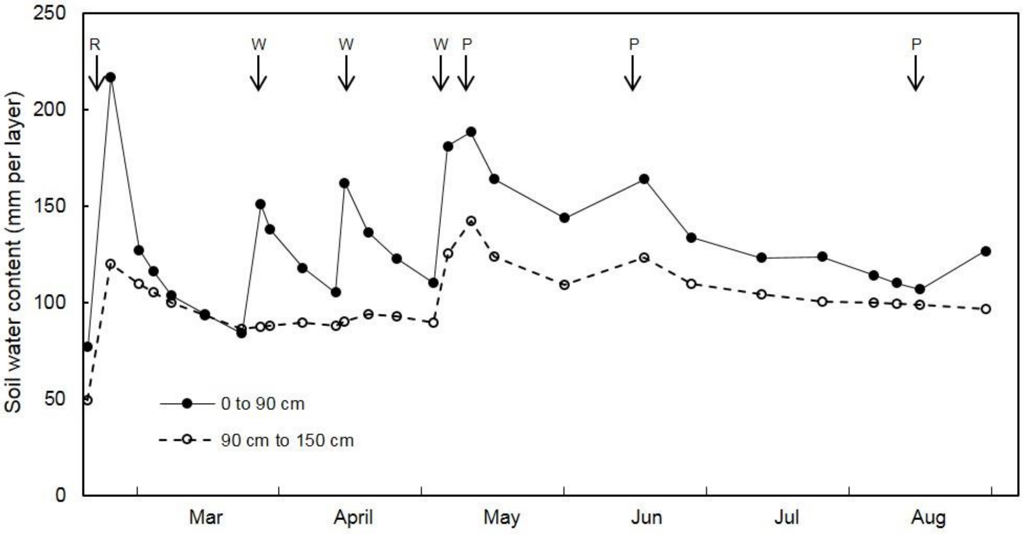

Figure 5.1. Variation in soil water content during the 2009/10 season where diluted winery wastewater was used to irrigate Cabernet Sauvignon grapevines in a sandy soil near Rawsonville. (P = precipitation, R = river water irrigation and W = wastewater irrigation).

Three irrigations were applied during the post-harvest period. Due to late ripening of the 2010 harvest in general, the winery was still crushing grapes when the first two irrigations were applied. The relatively high soil water content indicated that most layers were still saturated when the soil water content was measured shortly after the irrigation was stopped (Fig. 5.1). However, the soil water content in the 150-180 cm layer only showed an increase six days later (data not shown). This indicated that percolation from the saturated shallower soil layers into the deep layer must have occurred in the first few days following the irrigation. Smaller irrigations, *i.e.* approximately 55 mm each, were applied when the wastewater treatments commenced. These irrigations only wetted the soil to a depth of *c.* 90 cm and the soil water content measurements showed that no percolation into the deeper layers occurred (Fig. 5.1). As a result, the soil water content in the deepest layers remained fairly constant in the period following the first irrigation. The day after the third irrigation was applied in May, there was 85 mm rainfall. The combined effect of the irrigation and the rainfall saturated the upper soil layers to such an extent that deep percolation substantially increased the soil water content in the deepest layers.

2010/11 season: Due to the relatively low winter rainfall in 2010, the soil was relatively dry at bud break in September (Fig. 5.2). Despite the relatively dry soil conditions, grapevine vegetative growth did not show any visual signs of water constraints, and the first irrigation was only applied in December 2010. The first of the six wastewater irrigations was applied on 9 February 2011. The wastewater irrigations were applied at *c.* 14 day intervals. Although the objective was to apply irrigations to the root zone only, rainfall in May (94 mm), June (150 mm) and July (56 mm) seemed to have caused percolation into the deeper layers (Fig. 5.2).

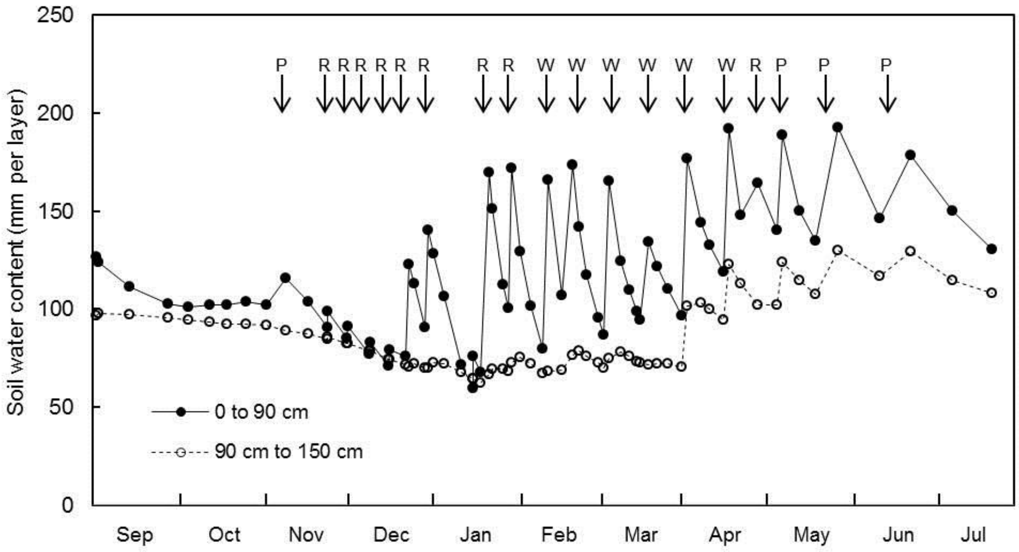

Figure 5.2. Variation in soil water content during the 2010/11 season where diluted winery wastewater was used to irrigate Cabernet Sauvignon grapevines in a sandy soil near Rawsonville. (P = precipitation, R = river water irrigation and W = wastewater irrigation).

2011/12 season: Due to the winter rains during 2011, the soil was relatively wet at bud break in September (Fig. 5.3). The first river water irrigation was only applied in middle December 2011. The second river water irrigation was required early January 2012, followed by three weekly river water irrigations of 16 mm each for the pearl millet summer interception crop. The grapevines were irrigated twice during February 2012 with river water. Since inadequate volumes of suitable winery wastewater were produced in February, the first of the five wastewater irrigations could only be applied on 6 March 2012. The wastewater irrigations were applied at *c.* 14 day intervals.

When established in November 2010, the pearl millet interception crop increased the ET_c from November until February compared to the same period in the 2009/10 season. Since the pearl millet was only established in January 2012, ET_c for January and February was lower compared to the previous season. These results confirmed that a summer interception crop established earlier in the season, *e.g.* in November, will increase the ET_c of vineyards substantially compared to clean cultivated or mulched soil surfaces.

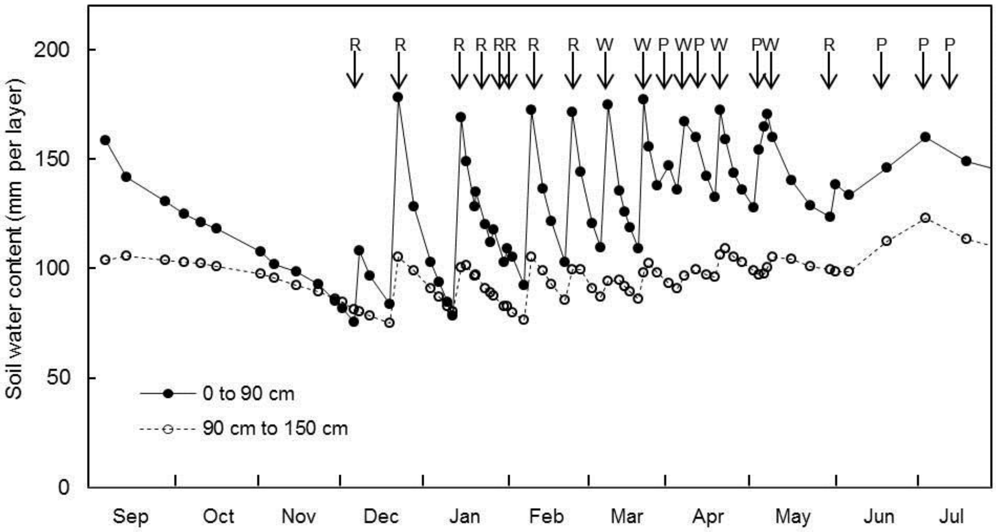

Figure 5.3. Variation in soil water content during the 2011/12 season where diluted winery wastewater was used to irrigate Cabernet Sauvignon grapevines in a sandy soil near Rawsonville. (P = precipitation, R = river water irrigation and W = wastewater irrigation).

2012/13 season: Due to the winter rains during 2012, the soil was relatively wet at bud break in September (Fig. 5.4) and the first river water irrigation was only applied towards the end of December 2012. In early January 2013, river water irrigation was applied to facilitate the soil cultivation for the planting of the pearl millet summer crop and was followed by three weekly river water irrigations of 16 mm each for this crop. The second river water irrigation for the vineyard was applied in early February 2013.

The first of the six wastewater irrigations was applied on 14 February, and thereafter these irrigations were applied at *c.* 14 day intervals until the end of April. However, on 25 March the COD level of the wastewater in the stock dam was too low (< 5000 mg/L) to have sufficient wastewater to make up all of the 15 m^3 tanks. On 26 March, the COD levels were higher than 30 000 mg/L. It was therefore decided to postpone the irrigation for a week to obtain more suitable water. On 2 April, the COD level of the stock dam was *c*. 9750 mg/L and wastewater irrigations were applied every two weeks until the end of April. Irrigation was applied only to the upper soil layers, *i.e.* 0-60 cm depth, to prevent leaching of elements into the deeper layers. In addition, such a continuous deficit irrigation strategy would reduce excessive growth and enhance ripening. However, a rainfall event of 67 mm after the wastewater irrigation in mid-April probably leached elements into the deeper layers. It was evident that the continuous deficit irrigation strategy also reduced ET_c in January 2013 compared to January 2012. Furthermore, the pearl millet interception crop did not increase ET_c substantially during the ripening period. In May 2013, river water irrigation was applied to the oats cover crop.

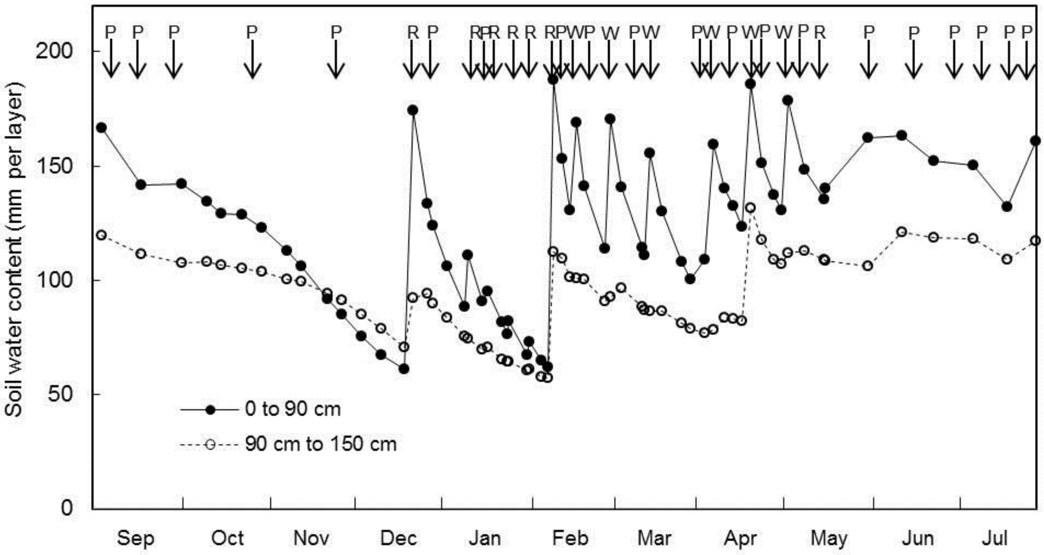

Figure 5.4. Seasonal variation in soil water content during the 2012/13 season where diluted winery wastewater was used to irrigate Cabernet Sauvignon grapevines in a sandy soil near Rawsonville. (P = precipitation, R = river water irrigation and W = wastewater irrigation).

5.3.2. Grapevine water status

Measurements in the 2010/11 season showed that Ψ_{PD} was around -0.2 MPa (Table 5.1) which is the lower threshold for no water constraints (Deloire *et al.*, 2004). This confirmed that the water status in the grapevines was able to fully recover during the night under the prevailing conditions. During daytime, the grapevines only experienced low water constraints, *i.e.* Ψ_L ranged between -1.0 MPa and -1.4 MPa, *i.e.* the Ψ_L thresholds according to Greenspan (2005). The low daytime water constraints were substantiated by Ψ_S that ranged between -0.4 MPa and -1.0 MPa, *i.e.* the thresholds proposed by Van Leeuwen *et al.* (2009). The foregoing indicated that grapevine only experienced low water constraints. Furthermore, irrigation using diluted winery wastewater, regardless of level of dilution, clearly had no effect on the grapevine water status compared to where grapevines were irrigated using river water. This was to be expected, since winery wastewater diluted up to 3000 mg/L COD had EC_{iw} well below 2 dS/m (Refer to Chapter 3). Furthermore, the pH and SAR of

3000 mg/L COD diluted winery wastewater produced irrigation water of which the quality would permit sustainable vineyard irrigation under the prevailing conditions.

Table 5.1. Predawn (Ψ_{PD}), as well as midday leaf (Ψ_L) and stem (Ψ_S) water potential in Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11 season.

Treatment no. & target COD (mg/L)	Ψ_{PD} (MPa)		Ψ_L (MPa)		Ψ_S (MPa)	
	08 Dec	02 Mar	08 Dec	15 Mar	08 Dec	15 Mar
T1 - River water	-0.20 a(1)	-0.24 a	-1.23 a	-1.33 a	-0.81 a	-0.68 a
T2 - 100	-0.18 a	-0.20 a	-1.13 a	-1.15 a	-0.70 a	-0.54 a
T3 - 250	-0.19 a	-0.23 a	-1.24 a	-1.15 a	-0.79 a	-0.65 a
T4 - 500	-0.23 a	-0.25 a	-1.11 a	-1.17 a	-0.74 a	-0.63 a
T5 - 1000	-0.18 a	-0.23 a	-1.22 a	-1.14 a	-0.76 a	-0.68 a
T6 - 1500	-0.18 a	-0.23 a	-1.25 a	-1.18 a	-0.78 a	-0.60 a
T7 - 2000	-0.20 a	-0.23 a	-1.26 a	-1.18 a	-0.73 a	-0.68 a
T8 - 2500	-0.21 a	-0.25 a	-1.08 a	-1.18 a	-0.68 a	-0.64 a
T9 - 3000	-0.21 a	-0.25 a	-1.18 a	-1.23 a	-0.75 a	-0.60 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

In the 2011/12 season, midday Ψ_L and Ψ_S values again ranged between the low constraints thresholds as mentioned above (Tables 5.2 & 5.3). When deficit irrigation was applied in the 2012/13 season, midday Ψ_L and Ψ_S also remained between the thresholds for low water constraints (data not shown). These results confirmed that the grapevines experienced low water constraints throughout the study period, and that the different diluted winery wastewater treatments clearly had no effect on the grapevine water status compared to the river water control. Since irrigation of grapevines using diluted winery wastewater, irrespective of dilution level did not induce any grapevine water constraints, it can be assumed that the functioning of other physiological processes would not have been negatively affected by water deficits. Given the low levels of water constraints, poor wine quality would be expected (Lategan, 2011).

Table 5.2. Midday leaf water potential (Ψ_L) in Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2011/12 season.

Treatment no. & target COD (mg/L)	Ψ_L (MPa)				
	08 March	19 March	21 March	02 April	18 April
T1 - River water	-1.03 a(1)	-1.03 a	-1.10 a	-0.92 a	-1.20 a
T2 - 100	-0.90 a	-0.91 a	-1.14 a	-0.99 a	-1.13 a
T3 - 250	-0.88 a	-0.83 a	-1.21 a	-1.03 a	-1.11 a
T4 - 500	-0.91 a	-1.02 a	-1.18 a	-0.94 a	-1.05 a
T5 - 1000	-1.01 a	-1.03 a	-1.20 a	-0.94 a	-1.25 a
T6 - 1500	-0.90 a	-1.16 a	-1.15 a	-0.88 a	-1.25 a
T7 - 2000	-0.85 a	-0.83 a	-1.14 a	-1.03 a	-1.10 a
T8 - 2500	-0.87 a	-1.24 a	-1.15 a	-0.98 a	-1.17 a
T9 - 3000	-0.89 a	-1.02 a	-1.23 a	-1.03 a	-1.10 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.3. Midday stem (Ψ_S) water potential in Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2011/12 season.

Treatment no. & target COD (mg/L)	Ψ_S (MPa)				
	08 March	19 March	21 March	02 April	18 April
T1 - River water	-0.61 a(1)	-0.58 a	-0.56 a	-0.51 a	-0.64 a
T2 - 100	-0.53 a	-0.43 a	-0.51 a	-0.45 a	-0.51 a
T3 - 250	-0.54 a	-0.55 a	-0.53 a	-0.49 a	-0.60 a
T4 - 500	-0.48 a	-0.53 a	-0.57 a	-0.59 a	-0.55 a
T5 - 1000	-0.55 a	-0.56 a	-0.55 a	-0.58 a	-0.67 a
T6 - 1500	-0.49 a	-0.53 a	-0.53 a	-0.47 a	-0.66 a
T7 - 2000	-0.52 a	-0.55 a	-0.50 a	-0.50 a	-0.57 a
T8 - 2500	-0.52 a	-0.56 a	-0.55 a	-0.55 a	-0.68 a
T9 - 3000	-0.58 a	-0.63 a	-0.53 a	-0.58 a	-0.63 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

5.3.3. Vegetative growth

5.3.3.1. Leaf and shoot chemical status

Since leaf blade and shoot samples of only Replication 2 were analysed, only the standard deviation from the mean is presented in Tables 5.4 to 5.6. According to norms for grapevine nutrient levels in leaves (Conradie, 1994), *i.e.* 1.6% to 2.7% for N, 0.14% to 0.55% for P, 0.65% to 1.3% for K^+, 1.2% to 2.2% for Ca^{2+}, and 0.16% to 0.55% for Mg^{2+}, none of the macro elements were at deficient levels during any of the seasons, except for low K^+ in 2012/13. The latter was probably due a competition effect of the pearl millet interception crop in summer. Otherwise, the pearl millet incerception crop and oats combination in winter did not seem to have any negative effects on grapevine nutrient status under the prevailing conditions. In addition to this, the nutrient levels were also not excessively high. This indicated that the additional amounts of elements applied *via* the diluted winery wastewater, in particular K^+ and Na^+, were not taken up by the grapevine to such an extent that negative effects would be expected. There were no trends in N that could be related to the different levels of wastewater dilution. This was probably due to the fact that the N load in the diluted winery wastewater was completely inadequate to supply the grapevine's annual requirement (Refer to Chapter 3). Leaf blade P could also not be related to the different levels of wastewater dilution, as P loads in the winery wastewater diluted to 2500 mg/L COD and lower could not supply adequate P for a grape yield of 10 t/ha. (Refer to Chapter 3).

Although soil Bray II-K increased substantially in the 0-30 cm as well as 30-60 cm soil depth layer, and the increase was strongly related to the additional amounts of K^+ *via* the diluted winery wastewater (Refer to Chapter 4), there were no substantial differences in the leaf blade K^+ measured prior to harvest. Similarly, even though soil K^+ increased substantially where 2 t/ha K_2SO_4 was applied (Dundon & Smart, 1984), *i.e.* 800 to 880 kg/ha K^+, there were no consistent responses in petiole contents at flowering (Dundon *et al.*, 1984). Since most of the K^+ uptake by the grapevine takes place prior to véraison, with almost no uptake from five weeks after harvest (Conradie, 1981), it could be that the additional K^+ applied *via* the diluted winery wastewater was applied too late in the growing season to have had an impact on leaf K^+ uptake. It has been shown that leaf K^+ becomes less from véraison to harvest, whereafter it increases (Conradie, 1981). High K^+ concentrations in "simulated" wastewater promoted the accumulation of harvest petiole K^+ (Mosse *et al.*, 2013). However, in that particular study, grapevines were irrigated with the artificial wastewater in the pre-véraison period as well. Where Shiraz grapevines were irrigated with winery wastewater at different dilutions, petiole K^+ was not affected (Kumar *et al.*, 2014), whereas the use of undiluted winery wastewater for vineyard irrigation increased petiole K^+ (Kumar *et al.*, 2014). Excessive levels of K^+, *i.e.* 450 kg/ha, applied to Concord grapevines increased petiole K^+ substantially (Morris & Cawthon, 1982). Where no K^+ and either 225 kg K^+, 450 kg K^+ or 900 kg K^+ per ha was applied to Concord grapevines, petiole K^+ already responded in the first year of the study (Morris *et al.*, 1980). Even though substantially less K^+ fertilizer was applied, increasing K^+ fertilizer from 0 kg to 90 kg increased both leaf blade and petiole K^+ (Conradie & Saayman, 1989). For Seyval Blanc grapevines growing in four nutrient solutions with different K^+ concentrations, there was an increase in petiole K^+ (Wolf *et al.*, 1983).

In the 2011/12 and 2012/13 seasons, leaf Ca^{2+} tended to decrease with a decrease in wastewater dilution (Tables 5.5 & 5.6) and could be related to the increase in the K^+ applied *via* the diluted winery wastewater up to harvest. Therefore, it seems that there was a K^+-induced suppression of Ca^{2+} absorption. Since leaf blade Ca^{2+} of grapevines irrigated using winery wastewater diluted to 3000 mg/L COD (T9) was still substantially higher than the minimum norm for Ca^{2+} (Conradie, 1994), the reduction in Ca^{2+} did not reduce leaf Ca^{2+} to insufficient levels. A similar response was observed where high K^{+} concentrations in artificial wastewater reduced harvest petiole Ca^{2+} substantially (Mosse *et al.*, 2013). However, when the artificial wastewater contained organic matter together with high K^{+} levels, petiole Ca^{2+} was not reduced to the same extent. It has been reported previously that there was a reduction in petiole Ca^{2+} where excessive levels of K^{+}, *i.e.* 450 kg/ha, were applied to Concord grapevines (Morris & Cawthon, 1982) after five years. For Seyval Blanc grapevines growing in four nutrient solutions, an increase in the solution K^{+} from 0 mg/L to 235 mg/L increased petiole Ca^{2+} (Wolf *et al.*, 1983). However, a further increase in the K^{+} concentration to 700 mg/L reduced petiole Ca^{2+}. It seemed that leaf blade Ca^{2+} tended to be more sensitive than petiole Ca^{2+}, with a reduction in Ca^{2+} as K^{+} application increased (Conradie & Saayman, 1989). In addition to the K^{+}/Ca^{2+} antagonism, it could also be that the leaf blade Ca^{2+} levels in the present study also decreased due to a Na^{+}/Ca^{2+} antagonism (Prior *et al*., 1992; Garcia & Charbaji, 1993; Fisarakis *et al*., 2005).

Leaf blade Mg^{2+} tended to decrease with an increase in the additional K^{+} added *via* the diluted winery wastewater up to harvest (Tables 5.4, 5.5 & 5.6). This indicated a possible K^{+}-induced suppression of Mg^{2+} absorption (Saayman, 1981). Similar results were reported by Morris *et al.* (1980) where grapevines were fertilized with excessive amounts of K^{+}. Large applications of K^{+} have been known to reduce Mg^{2+} to deficiency levels (Morris & Cawthon, 1982 and references therein), and it is possible that a K^{+}-induced Mg^{2+} deficiency could develop from continued use of high levels of K (Morris *et al.*, 1980). Where Seyval Blanc grapevines were growing in four nutrient solutions, petiole Mg^{2+} decreased in response to increasing K^{+} (Wolf *et al.*, 1983). Likewise, when 45 kg K^{+} was applied per ha compared to no K^{+}, leaf blade and petiole Mg^{2+} decreased (Conradie & Saayman, 1989). However, increasing K^{+} from 45 kg/ha to 90 kg/ha did not induce further Mg^{2+} reductions. Although substantial amounts of Na^{+} were applied *via* the diluted winery wastewater, leaf blade Na^{+} contents were well below 0.25%, *i.e.* the maximum for grapevines (Conradie, 1994), thereby reflecting the low sodicity risk of the diluted winery wastewater under the prevailing conditions. In contrast, for Shiraz grapevines, Na^{+} based artificial wastewater as a source of irrigation water increased petiole Na^{+} levels substantially (Mosse *et al.*, 2013). With the exception of Ca^{2+}, irrigation using diluted winery wastewater did not affect shoot chemical composition compared to the river water control. Although leaf blade Mg^{2+} responded negatively to an increase in K^{+} there was no such response with regard to shoot Mg^{2+}.

Table 5.4. Nutrient status of Cabernet Sauvignon leaf blades sampled prior to harvest in March and shoots sampled in the winter of 2011.

Treatment no. & target COD (mg/L)	N (%)	P (%)	K^+ (%)	Ca^{2+} (%)	Mg^{2+} (%)	Na^+ (mg/kg)	Mn^{2+} (mg/kg)	Fe^{2+} (mg/kg)	Cu^{2+} (mg/kg)	Zn^{2+} (mg/kg)	B^{3+} (mg/kg)
Leaf blades											
T1 - River water	2.04±0.08(1)	0.18±0.02	0.78±0.08	2.17±0.3	0.54±0.03	187±19	160±26	376±60	281±58	84±13	36±4
T2 - 100	1.82±0.08	0.16±0.02	0.77±0.08	1.82±0.3	0.51±0.03	202±19	95±26	271±60	142±58	49±13	46±4
T3 - 250	1.82±0.08	0.17±0.02	0.75±0.08	1.68±0.3	0.48±0.03	193±19	89±26	206±60	138±58	49±13	45±4
T4 - 500	1.85±0.08	0.16±0.02	0.67±0.08	2.09±0.3	0.55±0.03	225±19	84±26	223±60	151±58	47±13	39±4
T5 - 1000	1.90±0.08	0.15±0.02	0.70±0.08	1.90±0.3	0.51±0.03	184±19	101±26	205±60	105±58	53±13	34±4
T6 - 1500	1.95±0.08	0.16±0.02	0.75±0.08	1.89±0.3	0.49±0.03	172±19	92±26	220±60	116±58	47±13	41±4
T7 - 2000	1.84±0.08	0.14±0.02	0.69±0.08	1.42±0.3	0.46±0.03	172±19	75±26	189±60	97±58	43±13	47±4
T8 - 2500	1.82±0.08	0.13±0.02	0.55±0.08	1.43±0.3	0.49±0.03	163±19	75±26	177±60	91±58	43±13	40±4
T9 - 3000	1.95±0.08	0.17±0.02	0.82±0.08	1.78±0.3	0.51±0.03	178±19	96±26	223±60	114±58	52±13	42±4
Shoots											
T1 - River water	2.11±0.05	0.13±0.01	0.52±0.05	0.42±0.05	0.15±0.02	242±23	13±0.9	55±15	4±0.5	26±10	13±2
T2 - 100	1.96±0.05	0.14±0.01	0.49±0.05	0.41±0.05	0.15±0.02	189±23	12±0.9	56±15	4±0.5	30±10	11±2
T3 - 250	2.07±0.05	0.15±0.01	0.51±0.05	0.41±0.05	0.16±0.02	190±23	11±0.9	47±15	3±0.5	42±10	11±2
T4 - 500	2.00±0.05	0.14±0.01	0.51±0.05	0.41±0.05	0.16±0.02	197±23	13±0.9	45±15	4±0.5	48±10	11±2
T5 - 1000	1.98±0.05	0.12±0.01	0.45±0.05	0.37±0.05	0.13±0.02	204±23	14±0.9	37±15	3±0.5	47±10	10±2
T6 - 1500	2.01±0.05	0.12±0.01	0.47±0.05	0.34±0.05	0.11±0.02	170±23	12±0.9	22±15	3±0.5	21±10	9±2
T7 - 2000	1.99±0.05	0.13±0.01	0.40±0.05	0.31±0.05	0.13±0.02	171±23	12±0.9	16±15	3±0.5	37±10	8±2
T8 - 2500	1.99±0.05	0.12±0.01	0.37±0.05	0.31±0.05	0.13±0.02	165±23	12±0.9	19±15	3±0.5	29±10	8±2
T9 - 3000	2.02±0.05	0.11±0.01	0.42±0.05	0.34±0.05	0.12±0.02	187±23	12±0.9	28±15	3±0.5	29±10	9±2

(1) Standard deviation.

Table 5.5. Nutrient status of Cabernet Sauvignon leaf blades sampled prior to harvest in March and shoots sampled in the winter of 2012.

Treatment no. & target COD (mg/L)	N (%)	P (%)	K^{+} (%)	Ca^{2+} (%)	Mg^{2+} (%)	Na^{+} (mg/kg)	Mn^{2+} (mg/kg)	Fe^{2+} (mg/kg)	Cu^{2+} (mg/kg)	Zn^{2+} (mg/kg)	B^{3+} (mg/kg)
Leaf blades											
T1 - River water	1.73±0.05(1)	0.15±0.03	0.76±0.08	2.39±0.19	0.68±0.04	262±23	116±17	332±51	28±3.4	30±3.2	45±2
T2 - 100	1.80±0.05	0.22±0.03	0.72±0.08	2.28±0.19	0.64±0.04	227±23	106±17	360±51	27±3.4	32±3.2	47±2
T3 - 250	1.70±0.05	0.13±0.03	0.59±0.08	2.02±0.19	0.63±0.04	191±23	91±17	204±51	23±3.4	32±3.2	43±2
T4 - 500	1.74±0.05	0.14±0.03	0.56±0.08	2.14±0.19	0.66±0.04	238±23	83±17	223±51	25±3.4	32±3.2	46±2
T5 - 1000	1.68±0.05	0.13±0.03	0.58±0.08	2.24±0.19	0.62±0.04	206±23	137±17	238±51	33±3.4	33±3.2	44±2
T6 - 1500	1.77±0.05	0.15±0.03	0.62±0.08	2.15±0.19	0.60±0.04	202±23	88±17	276±51	32±3.4	28±3.2	42±2
T7 - 2000	1.77±0.05	0.12±0.03	0.50±0.08	1.93±0.19	0.63±0.04	192±23	93±17	241±51	25±3.4	23±3.2	44±2
T8 - 2500	1.75±0.05	0.13±0.03	0.62±0.08	1.83±0.19	0.61±0.04	209±23	105±17	251±51	30±3.4	33±3.2	42±2
T9 - 3000	1.84±0.05	0.17±0.03	0.71±0.08	1.90±0.19	0.54±0.04	212±23	94±17	254±51	26±3.4	31±3.2	39±2
Shoots											
T1 - River water	0.83±0.05	0.13±0.01	0.61±0.05	0.35±0.19	0.12±0.02	115±35	16±3	54±11	5±0.5	18±8	12±2
T2 - 100	0.84±0.05	0.13±0.01	0.59±0.05	0.36±0.19	0.14±0.02	162±35	9±3	37±11	5±0.5	28±8	11±2
T3 - 250	0.77±0.05	0.12±0.01	0.50±0.05	0.38±0.19	0.14±0.02	156±35	6±3	26±11	5±0.5	23±8	10±2
T4 - 500	0.81±0.05	0.12±0.01	0.47±0.05	0.36±0.19	0.13±0.02	150±35	13±3	24±11	5±0.5	24±8	10±2
T5 - 1000	0.73±0.05	0.11±0.01	0.49±0.05	0.39±0.19	0.12±0.02	130±35	15±3	23±11	5±0.5	20±8	9±2
T6 - 1500	0.84±0.05	0.13±0.01	0.53±0.05	0.35±0.19	0.14±0.02	158±35	13±3	27±11	6±0.5	30±8	10±2
T7 - 2000	0.78±0.05	0.12±0.01	0.51±0.05	0.38±0.19	0.16±0.02	177±35	12±3	37±11	6±0.5	29±8	17±2
T8 - 2500	0.87±0.05	0.12±0.01	0.54±0.05	0.37±0.19	0.17±0.02	220±35	11±3	22±11	6±0.5	43±8	13±2
T9 - 3000	0.88±0.05	0.13±0.01	0.57±0.05	0.44±0.19	0.16±0.02	213±35	14±3	22±11	6±0.5	38±8	13±2

(1) Standard deviation.

Table 5.6. Nutrient status of Cabernet Sauvignon leaf blades sampled prior to harvest in March and shoots sampled in the winter of 2013.

Treatment no. & target COD (mg/L)	N (%)	P (%)	K^+ (%)	Ca^{2+} (%)	Mg^{2+} (%)	Na^+ (mg/kg)	Mn^{2+} (mg/kg)	Fe^{2+} (mg/kg)	Cu^{2+} (mg/kg)	Zn^{2+} (mg/kg)	B^{3+} (mg/kg)
Leaf blades											
T1 - River water	1.91±0.09(1)	0.22±0.03	0.45±0.04	2.88±0.25	1.01±0.10	144±21	51±17	258±50	39±6.1	25±1.9	44±4.4
T2 - 100	2.18±0.09	0.25±0.03	0.49±0.04	2.87±0.25	0.88±0.10	139±21	46±17	424±50	43±6.1	22±1.9	49±4.4
T3 - 250	2.20±0.09	0.23±0.03	0.43±0.04	2.48±0.25	0.80±0.10	158±21	45±17	282±50	40±6.1	26±1.9	44±4.4
T4 - 500	2.15±0.09	0.19±0.03	0.46±0.04	2.47±0.25	0.75±0.10	191±21	45±17	365±50	45±6.1	27±1.9	45±4.4
T5 - 1000	2.03±0.09	0.16±0.03	0.51±0.04	2.32±0.25	0.68±0.10	175±21	62±17	321±50	53±6.1	25±1.9	41±4.4
T6 - 1500	2.03±0.09	0.19±0.03	0.49±0.04	2.48±0.25	0.79±0.10	135±21	46±17	301±50	37±6.1	23±1.9	55±4.4
T7 - 2000	2.07±0.09	0.18±0.03	0.53±0.04	2.30±0.25	0.77±0.10	146±21	48±17	344±50	45±6.1	25±1.9	52±4.4
T8 - 2500	2.08±0.09	0.17±0.03	0.49±0.04	2.19±0.25	0.79±0.10	187±21	54±17	361±50	55±6.1	28±1.9	49±4.4
T9 - 3000	2.11±0.09	0.19±0.03	0.56±0.04	2.30±0.25	0.71±0.10	156±21	47±17	342±50	48±6.1	27±1.9	49±4.4
Shoots											
T1 - River water	1.09±0.09	0.12±0.01	0.34±0.04	0.38±0.06	0.19±0.03	288±41	16±4	84±57	6±0.7	27±10	11±1.2
T2 - 100	0.90±0.09	0.08±0.01	0.19±0.04	0.27±0.06	0.12±0.03	169±41	6±4	49±57	4±0.7	13±10	7±1.2
T3 - 250	0.96±0.09	0.10±0.01	0.25±0.04	0.37±0.06	0.16±0.03	180±41	16±4	149±57	5±0.7	25±10	10±1.2
T4 - 500	1.05±0.09	0.11±0.01	0.28±0.04	0.40±0.06	0.18±0.03	226±41	18±4	180±57	5±0.7	44±10	11±1.2
T5 - 1000	1.05±0.09	0.12±0.01	0.24±0.04	0.37±0.06	0.21±0.03	238±41	17±4	115±57	5±0.7	38±10	10±1.2
T6 - 1500	0.88±0.09	0.09±0.01	0.25±0.04	0.30±0.06	0.14±0.03	224±41	9±4	52±57	4±0.7	16±10	10±1.2
T7 - 2000	0.83±0.09	0.09±0.01	0.28±0.04	0.27±0.06	0.15±0.03	178±41	10±4	48±57	6±0.7	25±10	10±1.2
T8 - 2500	0.96±0.09	0.10±0.01	0.25±0.04	0.31±0.06	0.20±0.03	228±41	11±4	29±57	5±0.7	34±10	9±1.2
T9 - 3000	0.92±0.09	0.09±0.01	0.25±0.04	0.24±0.06	0.13±0.03	167±41	12±4	12±57	5±0.7	27±10	9±1.2

(1) Standard deviation.

5.3.3.2. Cane mass

Irrigation using diluted winery wastewater had no effect on vegetative growth of the grapevines compared to river water (Table 5.7). The mean cane mass over the three years was 2.51±0.15 t/ha. The lack of differences was to be expected, since irrigation using diluted winery wastewater did not affect grapevine water status, or the chemical status of the leaves and shoots as discussed above. In addition to this, the N load in the diluted winery wastewater was totally inadequate to supply the grapevine's N requirement (Refer to Chapter 3). Results confirmed that, under the prevailing conditions, winery wastewater diluted up to 3000 mg/L COD did not pose any salinity hazard to grapevine growth. Where artificial winery wastewater was used for vineyard irrigation, there were no differences in cane length and diameter at harvest (Mosse *et al.*, 2013). Similarly, the use of sewage water rather than good quality reservoir water for vineyard irrigation did not effect cane mass (McCarthy, 1981). Cane mass was slightly higher in the 2011/12 season compared to the 2010/11 season, but comparable to the 2009/10 season. In the 2012/13 season, cane mass was slightly higher compared to the 2010/11 season, but comparable to the values reported for the 2009/10 and 2011/12 seasons. Cane mass was comparable to values reported for Cabernet Sauvignon in the Breede River Valley (Roux, 2005) and Lower Olifants River Valley (Bruwer, 2010) but was substantially higher than that of non-irrigated grapevines in the Swartland region (Mehmel, 2010). The foregoing suggested that the interception crop did not seem to have a pronounced negative effect on grapevine vegetative growth.

Table 5.7. Effect of irrigation using diluted winery wastewater on cane mass at pruning in July of Cabernet Sauvignon/99R measured during four seasons near Rawsonville.

Treatment no. & target COD (mg/L)	Cane mass (t/ha)			
	2009/10	2010/11	2011/12	2012/13
T1 - River water	2.9 a⁽¹⁾	2.3 a	2.7 a	2.8 a
T2 - 100	3.1 a	2.5 a	2.8 a	2.8 a
T3 - 250	2.8 a	2.4 a	2.7 a	2.7 a
T4 - 500	3.0 a	2.4 a	2.8 a	2.7 a
T5 - 1000	2.9 a	2.2 a	2.6 a	2.7 a
T6 - 1500	2.6 a	1.9 a	2.5 a	2.6 a
T7 - 2000	2.9 a	2.0 a	2.4 a	2.3 a
T8 - 2500	2.8 a	2.3 a	2.5 a	2.7 a
T9 - 3000	3.0 a	2.2 a	2.8 a	2.8 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

5.3.4. Yield and its components

Bunches per grapevine: Low grapevine fertility occurred throughout the Breede River region in the 2010/11 season. This was probably caused by unfavourable atmospheric conditions during bunch initiation in the preceding year. Irrigation using diluted winery wastewater did not affect mean grapevine fertility, *i.e.* number of bunches per grapevine, except in the 2010/11 season (Table 5.8). In this particular season, the bunch numbers of T7 grapevines were lower compared to some of the other treatments. This result was co-incidental, and probably occurred since many grapevines had to be pruned severely in July 2010 to obtain a more correct grapevine structure. The mean number of bunches over the three years where wastewater was applied from véraison, was 28±1.

Table 5.8. Effect of irrigation using diluted winery wastewater on number of bunches per grapevine of Cabernet Sauvignon/99R measured during four seasons near Rawsonville.

Treatment no. & target COD (mg/L)	Bunches per grapevine			
	2009/10	2010/11	2011/12	2012/13
T1 - River water	28 a(1)	23 ab	33 a	29 a
T2 - 100	26 a	24 a	31 a	28 a
T3 - 250	27 a	23 ab	32 a	30 a
T4 - 500	28 a	22 ab	32 a	29 a
T5 - 1000	30 a	24 a	33 a	31 a
T6 - 1500	29 a	23 ab	30 a	27 a
T7 - 2000	28 a	19 b	32 a	28 a
T8 - 2500	29 a	24 a	31 a	30 a
T9 - 3000	26 a	23 ab	31 a	28 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Berry size: In 2010/11 and 2011/12 seasons, berry development of selected treatments showed the typical double sigmoid curve expected for grapes and diluted wastewater irrigation had no effect on berry size development, regardless of level of dilution (Schoeman, 2012). This was probably due to a lack of differences in grapevine water status (Tables 5.1, 5.2 & 5.3). Irrigation using diluted winery wastewater had no effect on berry mass at harvest compared to the river water control (Table 5.9). Although Mosse *et al.* (2013) observed some differences in berry weight at harvest where different artificial winery wastewaters were used for vineyard irrigation, these differences were very small and no conclusions could be made. Similarly, the use of undiluted winery wastewater for vineyard irrigation at Oxford Landing had no detrimental effect on berry size (Kumar *et al.*, 2014). In contrast, in a similar study at Angaston by the same researchers, the use of undiluted winery wastewater for vineyard irrigation consistently reduced berry weight substantially. It could be that the quality of the winery wastewater differed between the two sites. It should also be noted that the amounts of irrigation water applied to the vineyard were substantially greater where winery wastewater was used. Mean berry mass at harvest ranged between 1.2 and 1.5 g/berry, which was comparable to values for drip irrigated Cabernet Sauvignon in the Breede River valley (Roux, 2005). Where Cabernet is subjected to severe water constraints, *i.e.* Ψ_L below 1.6 MPa, berry mass is expected to be *ca.* 1 g/berry (Bruwer, 2010; Mehmel, 2010). The foregoing confirmed that the grapevines experienced low levels of water constraints. Since there were no differences in grapevine water status, there is no explanation for the smaller berries of T9 grapevines compared to T2 grapevines as determined at harvest in March 2011 (Table 5.9).

Table 5.9. Effect of irrigation using diluted winery wastewater on berry mass of Cabernet Sauvignon/99R measured at harvest during four seasons near Rawsonville.

Treatment no. & target COD (mg/L)	Berry mass (g)			
	2009/10	2010/11	2011/12	2012/13
T1 - River water	1.19 a(1)	1.31 ab	1.36 a	1.38 a
T2 - 100	1.28 a	1.38 a	1.38 a	1.48 a
T3 - 250	1.22 a	1.34 ab	1.31 a	1.38 a
T4 - 500	1.25 a	1.31 ab	1.34 a	1.34 a
T5 - 1000	1.19 a	1.28 ab	1.33 a	1.32 a
T6 - 1500	1.25 a	1.36 ab	1.36 a	1.41 a
T7 - 2000	1.23 a	1.32 ab	1.36 a	1.30 a
T8 - 2500	1.28 a	1.36 ab	1.41 a	1.42 a
T9 - 3000	1.24 a	1.27 b	1.35 a	1.27 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Bunch mass: The lower berry mass reported in Table 5.9 reflected in smaller bunches (Table 5.10) and lower yield (Table 5.11) compared to some of the other treatments. In the case of the T7 grapevines, the low bunch number (Table 5.8) seemed to have caused the lower yield compared to most of the other treatments. Since yield showed no trend with respect to level of dilution in this particular season, the lower yield was definitely not the result of irrigation with diluted winery wastewater. The mean bunch mass did not differ over the three years and was, on average, 155±6.12 g.

Table 5.10. Effect of irrigation using diluted winery wastewater on bunch mass of Cabernet Sauvignon/99R measured during four seasons near Rawsonville.

Treatment no. & target COD (mg/L)	Bunch mass (g)			
	2009/10	2010/11	2011/12	2012/13
T1 - River water	148 a(1)	126 ab	153 a	174 a
T2 - 100	156 a	135 ab	156 a	167 a
T3 - 250	142 a	139 a	155 a	164 a
T4 - 500	153 a	139 a	163 a	167 a
T5 - 1000	149 a	126 ab	152 a	173 a
T6 - 1500	137 a	136 ab	165 a	169 a
T7 - 2000	146 a	123 ab	150 a	154 a
T8 - 2500	148 a	134 ab	173 a	169 a
T9 - 3000	149 a	119 b	142 a	164 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Yield: Irrigation using diluted winery wastewater did not affect grapevine yield compared to the river water control, except in the 2010/11 season (Table 5.11). However, the observed differences in the 2010/11 season were not related to level of wastewater dilution. It was probably a result of the severe pruning applied in July 2010 to obtain more correct grapevine structures. As in the case of cane mass, results confirmed that winery wastewater diluted up to 3000 mg/L COD did not pose any salinity hazard to grape yield. Furthermore, considering the other classical water quality criteria such as pH and SAR, dilution of winery wastewater up to 3000 mg/L COD produced irrigation water of which the quality would permit sustainable vineyard irrigation under the prevailing conditions, *i.e.* Mediterranean climate with high winter rainfall and sandy soil. Although Mosse *et al.* (2013) observed some differences in yield with regard to different types of artificial winery wastewaters, the magnitude of these differences were very small. It should, however, be noted that only one year of application of the artificial wastewaters took place. Mean yield was lower in the 2010/11 season compared to 2009/10 (Table 5.11). Lower grapevine fertility in the region, as well as the severe pruning probably caused the generally lower yields in the 2010/11 season. During the other seasons, yield was comparable to *c.* 15 t/ha reported for irrigated Cabernet Sauvignon (Roux, 2005), but substantially higher than non-irrigated ones (Mehmel, 2010). This serves as confirmation that the cover crop combination of pearl millet in summer and oats in winter did not seem to have any negative effects on grapevine yield under the prevailing conditions. The mean yield did not differ over the three years where diluted winery wastewater was applied and was, on average, 14.9±0.9 t/ha.

Table 5.11. Effect of irrigation using diluted winery wastewater on yield of Cabernet Sauvignon/99R measured during four seasons near Rawsonville.

Treatment no. & target COD (mg/L)	Yield (t/ha)			
	2009/10	2010/11	2011/12	2012/13
T1 - River water	13.9 a(1)	9.8 abc	17.2 a	17.6 a
T2 - 100	14.1 a	11.2 a	17.0 a	16.1 a
T3 - 250	13.4 a	11.0 ab	17.4 a	17.2 a
T4 - 500	14.9 a	10.6 ab	18.0 a	18.1 a
T5 - 1000	15.6 a	10.6 ab	17.5 a	18.3 a
T6 - 1500	13.7 a	10.6 ab	16.9 a	15.7 a
T7 - 2000	13.9 a	8.2 c	16.6 a	15.0 a
T8 - 2500	14.6 a	11.2 a	18.6 a	18.9 a
T9 - 3000	13.6 a	9.4 bc	16.5 a	16.2 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

5.3.5. Evapotranspiration

Since irrigation using diluted winery wastewater did not affect soil water status or grapevine growth and yield compared to river water irrigation, there were no differences in daily vineyard ET_c between treatments (data not shown). Under the prevailing conditions, vineyard ET_c was comparable to that of micro-sprinkler irrigated Pinotage near Robertson in the Breede River valley (Myburgh, 2011a), except in January and February 2011 (Table 5.12). Following sowing in November 2010, vegetative growth of the pearl millet interception crop was extremely vigorous and, at full canopy cover, was almost as tall as the grapevine canopies (Fig 5.5). This indicated that the interception crop increased the vineyard ET_c from November until February compared to the same period in the other seasons (Table 5.12). The ET_c declined considerably in March 2011, *i.e.* after the interception crop had been slashed and removed. In the 2011/12 season, when the pearl millet was sown later, *i.e.* in January 2012, ET_c during January and February was lower compared to the 2010/11 season.

Table 5.12. Mean monthly daily evapotranspiration of Cabernet Sauvignon grapevines in a sandy soil near Rawsonville during four seasons.

Season	Evapotranspiration (mm/day)											
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug
2009/10	(1)	0.7	0.9	2.1	5.2	7.2	6.1	3.0	5.3	3.3	0.7	0.9
2010/11	0.9	0.3	1.7	3.5	8.1	9.4	5.7	5.5	5.1	3.4	1.4	0.9
2011/12	1.4	0.8	1.2	3.8	5.9	6.5	6.8	4.4	2.2	0.9	1.0	0.8
2012/13	1.8	0.9	1.8	4.2	4.2	5.8	5.0	3.2	2.4	1.2	1.2	1.3

(1) Not determined.

Figure 5.5. Stand of the pearl millet interception crop during February 2011.

When the diluted wastewater was applied in the 2012/13 season, when the continuous deficit irrigation strategy was followed, vineyard ET_c was slightly lower in 2013 compared to the other years (Table 5.12). It must be noted that the continuous deficit irrigation did not have any negative effects on grapevine yield under micro-sprinklers compared to the other seasons. This was in contrast to yield reductions where continuous deficit irrigation was applied to drip irrigated Shiraz grapevines in the Breede River valley (Lategan, 2011).

5.3.6. Juice characteristics

In the 2009/10 season, the grapevines were not subjected to diluted winery wastewater irrigation before harvest. In this particular season, mean juice TSS, TTA and pH were 22.4±0.4°B, 4.5±0.3g/L and 3.7±0.1, respectively when the grapes were harvested. In the 2010/11 and 2011/12 seasons, the selected diluted winery wastewater irrigation treatments had no effect on berry size development during ripening compared to the raw water control (Schoeman, 2012). The rate of sugar loading into the berries during ripening was not affected by the wastewater irrigation compared to the river water control (Schoeman, 2012). Similarly, the wastewater irrigation had no effect on acid breakdown. The fact that ripening was comparable to the control indicated that the winery wastewater had no effect on the physiological functioning of the grapevines, irrespective of the level of dilution. Consequently, there were no differences in the TSS and TTA in the juice at harvest in the 2010/11, 2011/12 or 2012/13 seasons (Table 5.13). Due to berry ripening being considerably slower in the 2011/12 season compared to the previous ones, grapes were harvested five weeks later than in 2010/11. In the 2011/12 season, juice pH in T9 grapes was higher than for control grapevines, as well as grapevines that were irrigated with

Table 5.13. Total soluble solids (TSS), total titratable acidity (TTA) and pH in juice of Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	TSS (°B)			TTA (g/L)			pH		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	23.6 a⁽¹⁾	23.2 a	22.9 a	5.33 a	5.38 a	4.93 a	3.49 a	3.68 bc	3.60 a
T2 - 100	23.5 a	22.5 a	22.8 a	5.45 a	5.43 a	5.33 a	3.47 a	3.67 bc	3.63 a
T3 - 250	23.6 a	22.9 a	22.8 a	5.47 a	4.90 a	4.67 a	3.45 a	3.62 c	3.57 a
T4 - 500	24.3 a	23.0 a	22.8 a	5.20 a	4.98 a	4.73 a	3.52 a	3.70 bc	3.58 a
T5 - 1000	24.1 a	24.2 a	23.0 a	5.17 a	4.17 a	4.93 a	3.49 a	3.76 ab	3.64 a
T6 - 1500	23.4 a	22.9 a	23.1 a	5.47 a	4.75 a	4.87 a	3.47 a	3.75 ab	3.66 a
T7 - 2000	23.9 a	22.6 a	22.5 a	5.37 a	6.03 a	5.12 a	3.53 a	3.76 ab	3.61 a
T8 - 2500	23.6 a	22.7 a	23.1 a	5.47 a	4.77 a	5.17 a	3.52 a	3.77 ab	3.65 a
T9 - 3000	24.7 a	24.1 a	24.0 a	4.82 a	4.97 a	4.70 a	3.57 a	3.85 a	3.70 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

winery wastewater containing COD levels lower than 1000 mg/L (Table 5.13). In general, juice pH tended to increase with a decrease in level of dilution with the exception of T1 and T2 in 2012/13. Juice pH was linearly related to the amounts of K^+ applied *via* the irrigation water until harvest in 2010/11 ($R^2 = 0.570$), 2011/12 ($R^2 = 0.838$) and 2012/13 ($R^2 = 0.510$) ($R^2 = 0.690$ if T1 & T2 is not considered). The juice pH could be linearly related to the juice K^+ in 2010/11 ($R^2 = 0.530$), 2011/12 ($R^2 = 0.807$ if T1 & T2 is not considered) and 2012/13 ($R^2 = 0.763$). Likewise, when juice K^+ increased due to K^+ fertilization, juice pH also increased (Morris *et al.*, 1980; Morris & Cawthon 1982). It should be noted that even when 900 kg/ha K was applied to grapevines, juice pH did not increase above 3.57 (Morris *et al.*, 1980). On average, irrigation using diluted winery wastewater did not affect either TSS or TTA (Figs 5.6A & B) over three years. In contrast, average juice pH increased with a decrease in dilution of winery wastewater (Fig. 5.6C).

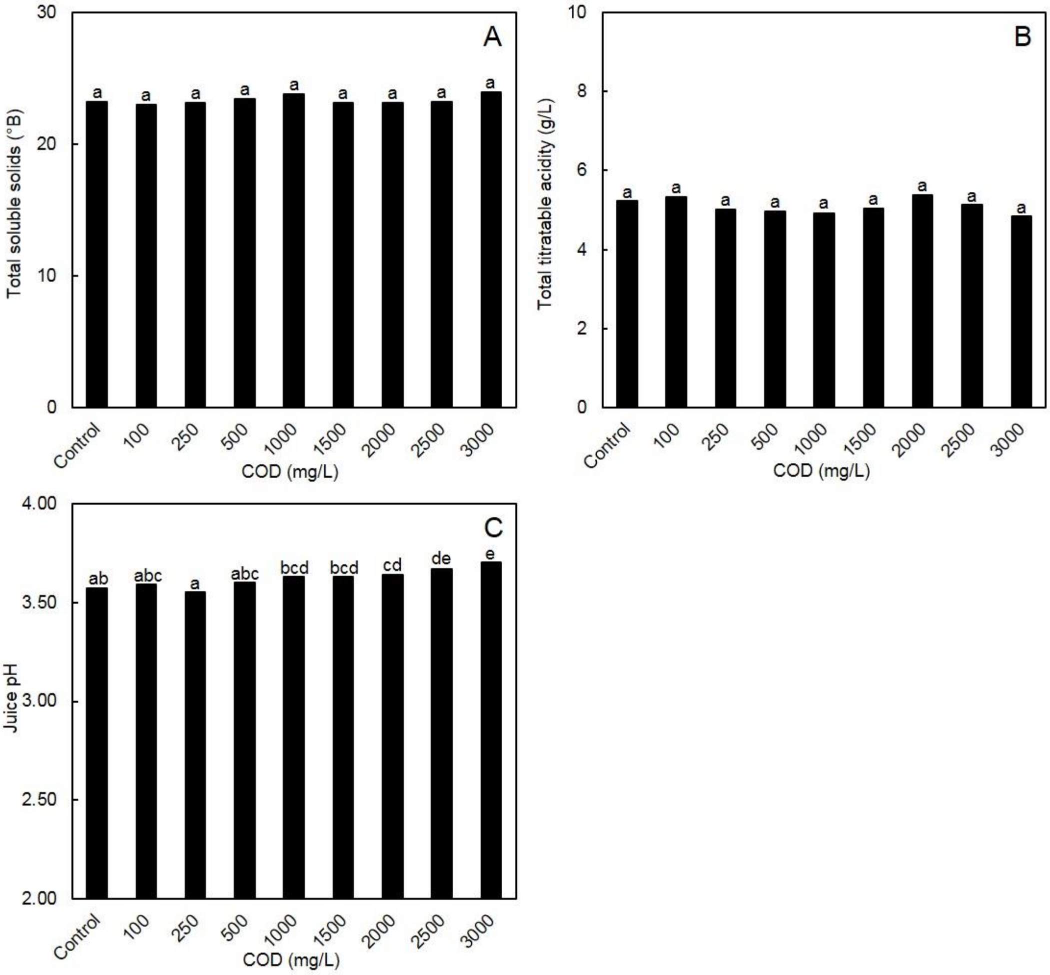

Figure 5.6. Effect of irrigation using diluted winery wastewater on (A) total soluble solids, (B) total titratable acidity and (C) juice pH. Data are means for the 2010/11, 2011/12 and 2012/13 seasons.

In the 2009/10 season, mean juice N, P, K^+, Ca^{2+}, Mg^{2+} and Na^+ were 338±87 mg/L, 96±16 mg/L, 762±149 mg/L, 27±4 mg/L, 48±5 mg/L and 5±1 mg/L, respectively, when the grapes were harvested. In the subsequent seasons, irrigation using diluted winery wastewater generally did not affect juice N, P, K^+, Ca^{2+}, Mg^{2+} and Na^+ compared to the river water control (Tables 5.14 & 5.15). In general, juice N, P, K^+, Ca^{2+}, Mg^{2+} and Na^+ were within the recommended levels (Wooldridge *et al.*, 2010). However, juice element levels tended to be higher than these recommended levels in the 2012/13 season. Although irrigation using diluted winery wastewater did not affect juice element composition, the following exceptions were observed. In the 2010/11 season, there were significant differences with regard to juice N, but there were no trends that could be related to the diluted wastewater irrigation (Table 5.14). In general, juice K^+ tended to be higher when the level of dilution of the winery wastewater was lower, *i.e.* more K^+ was applied *via* the diluted winery wastewater (Table 5.14). Likewise, there was also a tendency to more juice K^+ where undiluted winery wastewater was used for vineyard irrigation (Kumar *et al.*, 2014). It has also been reported that artificial winery wastewaters that contained high K^+ levels as well as wine produced juice with the lowest K^+ compared to wastewaters with high Na^+ and high K^+ (Mosse *et al.*, 2013). This indicated that the prescence of wine in the artificial winery wastewater prevented an increase in juice K^+. In a study investigating the long term use of K^+ fertilizer, juice K^+ increased when 45 kg K^+ was applied per ha compared to no K^+ (Conradie & Saayman, 1989). However, there were no further increases in juice K^+ when the K^+ application increased to 90 kg/ha. Another study indicated that when 450 kg/ha K^+ was applied to grapevines, there was an increase in juice K^+ (Morris & Cawthon, 1982). Juice K^+ of Concord grapevines also increased when K^+ application increased from no application to 225 kg K^+, 450 kg K^+ or 900 kg K^+ per ha (Morris *et al*, 1980).

In the 2010/11 season, juice Na^+ increased with an increase in the COD level of the diluted winery wastewater, whereas juice Ca^{2+} decreased (Table 5.15). There was an increase in extractable soil Na^+ which was related to the different levels of winery wastewater dilution (Refer to Chapter 4). It was previously reported that sodic soil conditions could cause high concentrations of Na^+ in grapevine tissue and concomitantly reduce Ca^{2+} concentrations (McCarthy & Downton, 1981; Stevens *et al*., 2011 and references therein). It should be noted that this trend was only observed in 2010/11. The reason for this is unclear. In the 2010/11 season, *c.* 70.7 kg/ha Na^+ was applied *via* the diluted winery wastewater prior to harvest where winery wastewater was diluted to 3000 mg/L COD compared to 31.0 kg/ha for the river water control (Refer to Appendix 3.8). In addition, a maximum of. 70.7 kg/ha Na^+ was applied to the soil *via* the diluted winery wastewater prior to harvest compared to 62.8 kg/ha and 40.3 kg/ha in 2011/12 and 2012/13, Where artificial wastewater was used for vineyard irrigation, there was an increase in juice Na^+ at harvest where Na^+-based wastewater was used compared to artificial winery wastewaters with high and low K^+, respectively (Mosse *et al.*, 2013). Unfortunately no data pertaining to juice Ca^+ was given.

Lower juice Ca^{2+} could also be due to a K/Ca^{2+} antagonism, as discussed previously for the leaf blades. In 2010/11 *c.* 106.2 kg/ha K^{+} was applied *via* the diluted winery wastewater prior to harvest where winery wastewater was diluted to 3000 mg/L COD which was substantially higher compared to 6.0 kg/ha for the river water control (Refer to Appendix 3.5). When the mean for all three seasons was considered, there tended to be an increase in juice K^{+} (Fig. 5.7A) and a decrease in Ca^{2+} (Fig. 5.7B) with a decrease in dilution level of the winery wastewater. No consistent trends could be observed for juice Na^{+} (Fig. 5.7C). In contrast, juice Na^{+} was higher at harvest where Na^{+}-based wastewater was used compared to artificial winery wastewaters with high and low K^{+}, respectively (Mosse *et al.*, 2013).

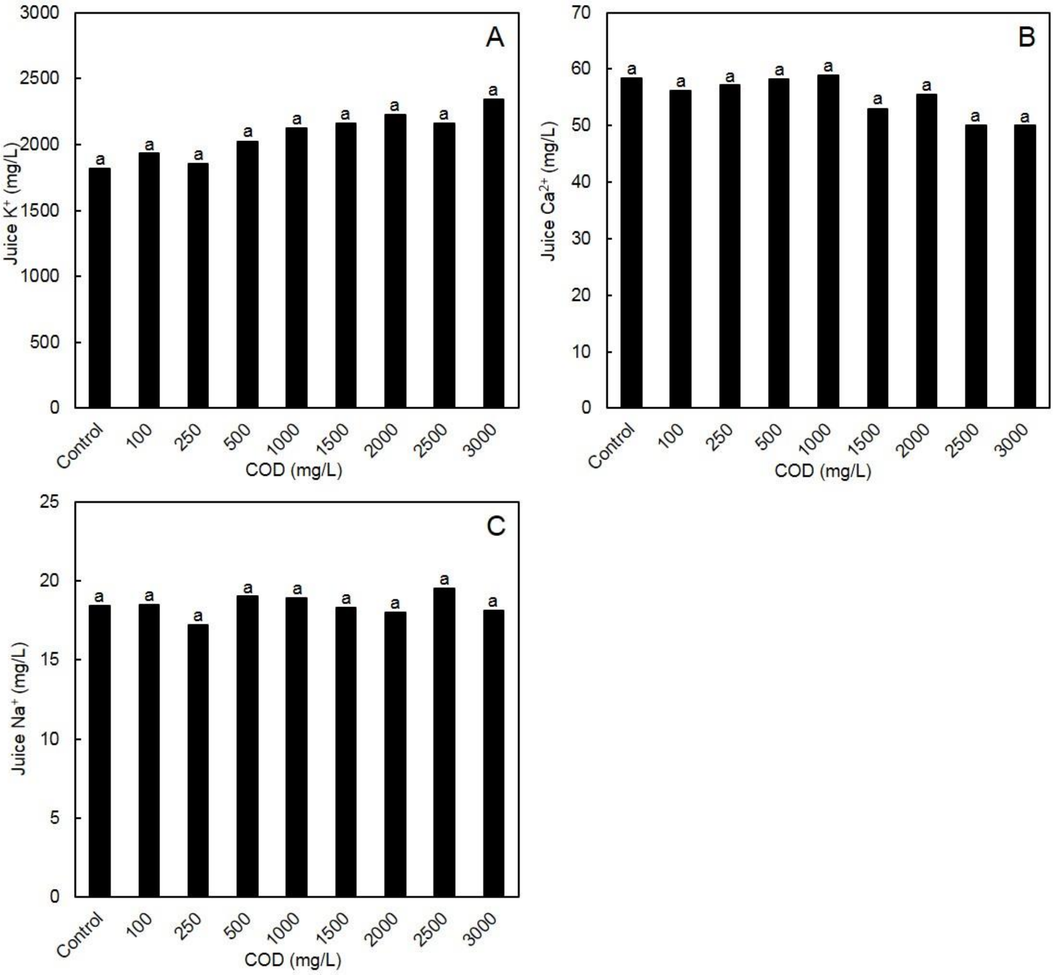

Figure 5.7. Effect of irrigation using diluted winery wastewater on juice (A) potassium (K^{+}), (B) calcium (Ca^{2+}) and (C) sodium (Na^{+}). Data are means for the 2010/11, 2011/12 and 2012/13 seasons.

Although there were differences in juice Cr^{2+} between treatments, they could not be related to the level of dilution (data not shown). Irrigation using diluted winery wastewater did not effect juice Cd^{2+} (data not shown). In addition, no arsenic or other heavy metals (Pb^{2+} & Hg^{2+}) were detected in the grape juice.

Table 5.14. Nitrogen (N), phosphorus (P) and potassium (K^+) in juice of Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	N (mg/L)			P (mg/L)			K^+ (mg/L)		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	202 bc(1)	239 a	1940 a	136 a	182 a	513 a	1626 a	1794 a	2779 a
T2 - 100	148 ab	246 a	1768 a	141 a	202 a	445 a	1780 a	1822 a	3038 a
T3 - 250	147 ab	240 a	2520 a	130 a	187 a	497 a	1675 a	1479 a	2413 a
T4 - 500	127 a	235 a	1688 a	131 a	182 a	480 a	1852 a	1677 a	2532 a
T5 - 1000	123 a	223 a	1573 a	131 a	192 a	471 a	1905 a	1685 a	2789 a
T6 - 1500	136 a	214 a	1582 a	124 a	188 a	487 a	1759 a	1717 a	2996 a
T7 - 2000	135 a	222 a	1630 a	146 a	214 a	435 a	1894 a	1852 a	2931 a
T8 - 2500	137 a	213 a	1830 a	136 a	204 a	503 a	1926 a	1686 a	2863 a
T9 - 3000	221 c	281 a	1913 a	137 a	215 a	382 a	1939 a	1916 a	3179 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.15. Calcium (Ca^{2+}), magnesium (Mg^{2+}) and sodium (Na^{+}) in juice of Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	Ca^{2+} (mg/L)			Mg^{2+} (mg/L)			Na^{+} (mg/L)		
	2010/11	**2011/12**	**2012/13**	**2010/11**	**2011/12**	**2012/13**	**2010/11**	**2011/12**	**2012/13**
T1 - River water	47.2 de(1)	41.1 a	86.6 a	97.0 a	113.4 a	165.1 a	7.6 ab	22.2 a	25.3 a
T2 - 100	49.4 e	46.2 a	86.7 a	102.0 a	124.8 a	170.1 a	7.7 ab	22.6 a	25.1 a
T3 - 250	43.8 cd	44.4 a	83.3 a	94.0 a	114.6 a	159.1 a	7.1 a	21.2 a	23.2 a
T4 - 500	42.1 bc	45.4 a	87.1 a	102.0 a	116.8 a	171.2 a	8.0 ab	23.4 a	25.7 a
T5 - 1000	38.2 ab	46.9 a	91.7 a	97.0 a	116.0 a	177.1 a	8.8 bc	22.6 a	25.4 a
T6 - 1500	34.3 a	44.8 a	80.1 a	92.0 a	113.2 a	169.6 a	7.9 ab	22.9 a	24.1 a
T7 - 2000	35.7 a	48.8 a	82.0 a	100.0 a	121.4 a	172.8 a	8.4 bc	22.5 a	23.0 a
T8 - 2500	37.6 ab	48.1 a	82.6 a	96.0 a	115.4 a	172.0 a	8.5 bc	22.5 a	27.4 a
T9 - 3000	34.5 a	47.1 a	68.3 a	96.0 a	119.8 a	144.9 a	9.6 c	22.3 a	22.4 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

(data not shown). This result was to be expected since these elements were not present in the river or diluted water.

5.3.7. Wine quality

Throughout the study, none of the wines contained any pathogenic bacteria, coliforms and *E. Coli,* and were therefore considered safe for the sensorial evaluation (data not shown). Therefore, results confirmed that wines would not pose a health risk to wine consumers. The application of wastewater irrigation, regardless of level of dilution, consistently had no effect on the sensorial wine characteristics compared to the river water control during 2010/11 (Schoeman, 2012), 2011/12 (Schoeman, 2012) and 2012/13 (Table 5.16). Likewise, although there were slight differences with regard to wine colour and tannin content where winery wastewater was used for vineyard irrigation, there were no differences in the sensorial evaluation of the wines (Kumar *et al.*, 2014). Where Shiraz grapevines were irrigated with sewage water, there were also no differences with regard to wine quality (McCarthy & Downton, 1981). Although mean juice pH increased linearly with increasing amounts of K^+ applied until harvest, it did not reflect in wine colour (Fig. 5.8A). This is probably because juice pH tended to be below 3.8, the norm above which detrimental affects of pH on wine colour, taste and microbial stability may be expected (Kodur 2011, and references therein). Wine vegetative and berry character was not affected by the use of diluted winery wastewater for irrigation (Figs. 5.8B & C). All the wines tended to have a stronger berry-like character than spicy character, consistent with Cabernet Sauvignon wine made from grapes produced in warmer localities such as Klawer in the Lower Olifants River region (Bruwer, 2010). There were no differences in wastewater associated off-flavours and -tastes compared to the river water control (Figs. 5.8D & E), thereby confirming that no contaminants were transferred from the wastewater into the wines. This was expected since visual observations revealed that bunches were not wetted with diluted winery wastewater during the wastewater irrigations. Perusal of the scorecards also revealed that members of the tasting panel were highly inconsistent with respect to their perception of off-tastes. The observed off-odours and off-tastes were all related to frequently occurring off-odours and off-tastes in wines such as volatile acidity and bitterness. However, in a parallel study where bunches were deliberately sprayed with diluted winery wastewater, a winery wastewater-like odour was detected in the wines, and their spicy character reduced (Schoeman, 2012). This highlights the importance of avoiding contact between grapes and winery wastewater. All the wines were of low quality, *i.e.* less than 40% (Fig. 5.8F). This trend was to be expected, since grapevines did not experience any water constraints during the season. Irrigation using diluted winery wastewater did not affect wine quality (Fig. 5.8F). Likewise, although there were slight differences with regard to wine colour and tannin content where winery wastewater was used for vineyard irrigation, there were no differences in the sensorial evaluation of the wines (Kumar *et al.*, 2014). Where Shiraz grapevines were irrigated with sewage water, there were also no differences with regard to wine quality (McCarthy & Downton, 1981).

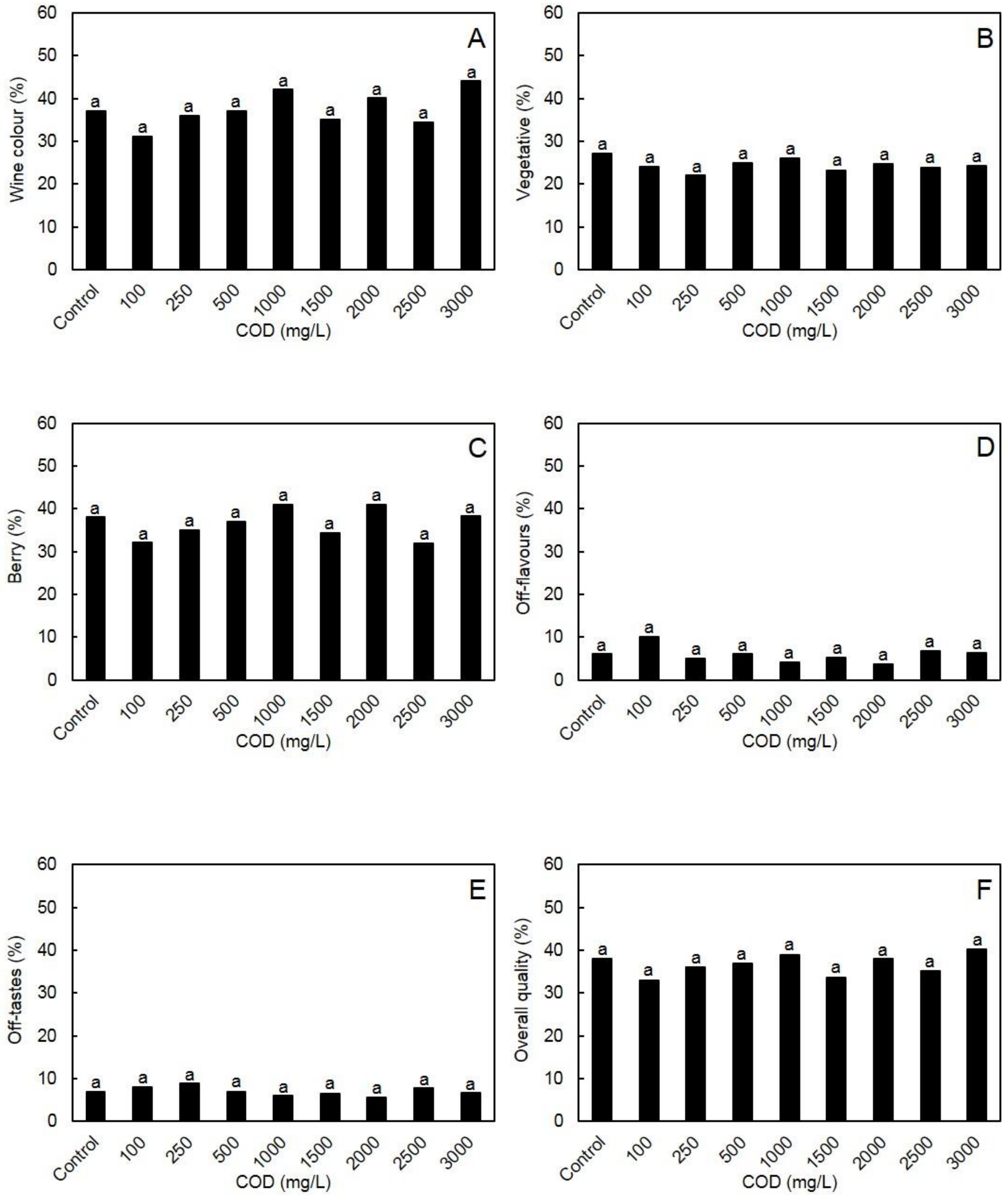

Figure 5.8. Effect of irrigation using diluted winery wastewater on (A) wine colour, (B) vegetative, (C) berry, (D) off-flavours, (E) off-tastes and (F) overall wine quality. Data are means for the 2010/11, 2011/12 and 2012/13 seasons.

Irrigation using diluted winery wastewater did not affect wine chemical properties compared to the river water control during the 2010/11 (Schoeman, 2012) and 2011/12 seasons

(Schoeman, 2012). With the exception of alcohol percentage and tartaric acid, irrigation using diluted winery wastewater did not affect wine chemical properties compared to the river water control in the 2012/13 season (Tables 5.17 & 5.18). At this stage, there is no clear explanation for the higher alcohol percentage where grapevines were irrigated using diluted winery wastewater containing 1000 mg/L and 3000 mg/L COD. Tartaric acid in wines of the 2012/13 season decreased with a decrease in level of dilution (Table 5.17). During wine making, high K^+ increases the precipitation of tartrate in salt form of K^+ bitartaric acid therefore tartrate is reduced (Mpelasoka *et al.*, 2003; Kodur 2011). In conjunction with a reduction in the wine tartaric acid, there was trend towards increased malic acid with an increase in COD level of the diluted winery wastewater (Table 5.17). This is probably due to higher juice K^+, which may decrease the rate of degradation of malic acid through respiration by impeding its transfer from the vacuole to the cytoplasm (Kodur, 2011). It should be noted that berry K^+ levels are often an important consideration for red wine production as the skin is left for some time after crushing for the extraction of anions, during which time more K^+ may be extracted (Mpelasoka *et al.*, 2003). However, in the present study, berry skin K^+ was not measured and it is possible that the berry skin K^+ could have increased in response to the diluted winery wastewater irrigation. Although wine pH increased with a decrease in level of dilution in the 2010/11 season (Schoeman, 2012), this trend was not observed in the 2011/12 and 2012/13 season. Considering the data over three years, irrigation using diluted winery wastewater did not effect malic and tartaric acids in the wine (Figs. 5.9A & B) as well as total acidity (Fig. 5.9C). Although wine pH tended to increase with a decrease in level of dilution (Fig. 5.9D), the pH increase did not have any negative effect on wine colour as determined both chemically and sensorially.

In a study carried out in Robertson, Moolman *et al*. (1998) reported wine Na^+ contents that ranged from 40 mg/L to 190 mg/L. Much higher values were reported for Australian Shiraz wine Na^+ that ranged from 78 mg/L to 533 mg/ (Walker *et al.*, 2003). Wine Na^+ in the current study was much lower than these reported levels in all seasons (Table 5.19). Furthermore, the legal limit for wine Na^+ in South Africa is 100 mg/L (Department of Water Affairs & Forestry, 1996). Wine Na^+ was considerably lower than this norm in all the seasons. Therefore, under the prevailing conditions, wines produced where grapevines were irrigated using diluted winery wastewater still conformed to statutary requirements with regard to Na^+ content. Moolman *et al*. (1998) reported wine Cl^- that ranged from from 50 mg/L to 160 mg/L, whereas much higher values of 98 mg/L to 1788 mg/L were reported for Shiraz in Australia (Walker *et al.*, 2003). The Australian legal limit for wine Cl^- content is 606 mg/L (Leske *et al.*, 1997). Based on this norm, Cl^- contents in the wines were extremely low (Table 5.20). With regard to wine ion composition, no consistent trends were observed (Tables 5.19 & 5.20).

However, in wines from the 2011/12 season, SO_4^{2-} increased with a decrease in level of dilution (Table 5.20). Reasons for this increase are unclear. In 2012/13, wine K^+ increased with an increase in COD level of the irrigation water (Table 5.19). Although wine P, K^+, Mg^{2+}, Na^+ and Cl^- were higher in response to irrigation with sewage water, concentrations were not excessively high (McCarthy & Downton, 1981). In contrast, wine Na^+ and Cl^- were substantially higher where sewage water was used for vineyard irrigation. Although Walker and Blackmore (2012) reported a positive linear relationship for wine K^+ and juice K^+ for two cultivars, the relationship was not 1:1. The R^2 values ranged between 0.80 and 0.86, with the slope of the relationship ranging from 0.40 to 0.89. In the present study, correlations between wine and juice K^+ revealed substantial differences beween the three seasons (data not shown). The slopes of the particular relationships ranged from 0.25 in 2012/13 to 0.65 in the 2010/11 season. Under the prevailing conditions, irrigation using diluted winery wastewater tended to increase wine K^+ (Fig. 5.10A) but had no consistent effect on wine Na^+ (Fig. 5.10B).

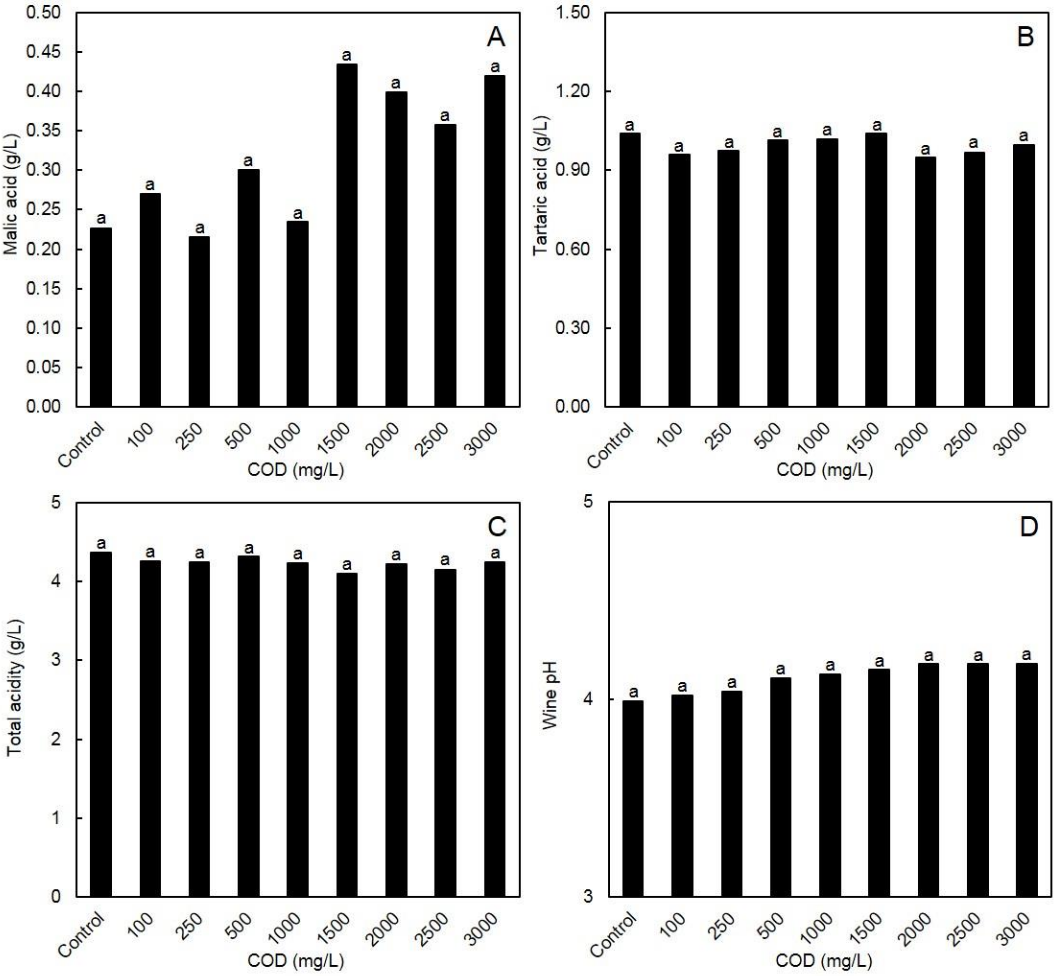

Figure 5.9. Effect of irrigation using diluted winery wastewater on (A) malic acid, (B) tartaric acid, (C) total acidity and (D) pH in wine. Data are means for the 2010/11, 2011/12 and 2012/13 seasons.

Table 5.16. Effect of irrigation using different levels of diluted winery wastewater on sensorial characteristics of Cabernet Sauvignon wine determined in August 2013.

Treatment no. and target COD (mg/L)	Colour (%)	Overall intensity (%)	Vegetative (%)	Berry (%)	Spicy (%)	Off-flavours (%)	Acidity (%)	Body (%)	Astringency (%)	Off-tastes (%)	Overall quality (%)
T1 - River water	32.5 a(1)	40.2 a	24.8 a	34.2 a	16.8 a	4.7 a	38.1 a	30.3 a	28.7 a	4.8 a	31.7 a
T2 - 100	31.5 a	39.9 a	24.5 a	35.4 a	18.5 a	4.7 a	39.5 a	32.3 a	32.2 a	5.4 a	31.5 a
T3 - 250	29.8 a	34.1 a	19.8 a	29.8 a	17.0 a	5.9 a	40.9 a	31.2 a	30.2 a	7.7 a	29.9 a
T4 - 500	36.1 a	37.8 a	22.4 a	34.4 a	16.2 a	3.4 a	42.7 a	32.2 a	30.8 a	3.2 a	35.4 a
T5 - 1000	33.9 a	41.2 a	25.0 a	35.8 a	16.2 a	3.8 a	38.4 a	29.2 a	29.2 a	5.1 a	34.1 a
T6 - 1500	35.1 a	37.6 a	19.4 a	34.2 a	14.7 a	6.8 a	39.8 a	30.0 a	30.1 a	7.4 a	31.6 a
T7 - 2000	31.1 a	43.3 a	22.4 a	40.9 a	17.9 a	2.2 a	38.6 a	31.1 a	28.4 a	4.2 a	34.2 a
T8 - 2500	30.4 a	37.6 a	21.1 a	31.8 a	15.9 a	4.6 a	39.0 a	30.1 a	30.3 a	7.7 a	30.7 a
T9 - 3000	40 4 a	39.6 a	22.9 a	38.3 a	17.5 a	3.5 a	43.0 a	37.3 a	33.1 a	4.2 a	38.7 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.17. Effect of irrigation using different levels of diluted winery wastewater on selected chemical properties of Cabernet Sauvignon wine measured six months after harvest in 2013.

Treatment no. and target COD (mg/L)	Alcohol (%)	Extract	Residual sugar (g/L)	Glucose (g/L)	Fructose (g/L)	Volatile acidity (g/L)	Tartaric acid (g/L)	Malic acid (g/L)	Total acidity (g/L)	pH
T1 - River water	12.7 ab(1)	24.7 a	1.08 a	0.11 a	0.08 a	0.40 a	1.19 e	0.57 a	4.69 a	3.88 a
T2 - 100	12.6 a	24.2 a	1.14 a	0.09 a	0.07 a	0.38 a	1.06 bcde	0.78 a	4.33 a	4.01 a
T3 - 250	12.7 ab	23.8 a	1.14 a	0.11 a	0.06 a	0.39 a	1.06 bcde	0.63 a	4.54 a	3.88 a
T4 - 500	12.8 ab	25.6 a	1.16 a	0.09 a	0.06 a	0.32 a	1.09 cde	0.95 a	5.17 a	3.82 a
T5 - 1000	13.1 bc	26.5 a	1.28 a	0.09 a	0.07 a	0.39 a	1.10 de	0.65 a	4.71 a	3.92 a
T6 - 1500	13.0 ab	26.7 a	1.17 a	0.12 a	0.07 a	0.40 a	1.01 bcd	1.06 a	4.51 a	4.00 a
T7 - 2000	12.7 ab	26.0 a	1.28 a	0.09 a	0.08 a	0.37 a	0.87 a	1.07 a	4.89 a	3.89 a
T8 - 2500	13.0 ab	24.1 a	1.25 a	0.08 a	0.07 a	0.37 a	0.95 ab	1.00 a	4.64 a	3.96 a
T9 - 3000	13.5 c	25.2 a	1.07 a	0.10 a	0.09 a	0.34 a	0.96 abc	1.16 a	5.01 a	3.95 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.18. Effect of irrigation using different levels of diluted winery wastewater on selected chemical properties of Cabernet Sauvignon wine measured six months after harvest in 2013.

Treatment no. and target COD (mg/L)	Free amino nitrogen (mg/L)	Free SO_2 (mg/L)	Total SO_2 (mg/L)	Brown colour (420 nm)	Red colour (520 nm)	Total tannins (510 nm)	Total phenols (280 nm)
T1 - River water	136.3 a(1)	40.7 a	83.0 a	0.92 a	1.10 a	0.52 a	29.9 a
T2 - 100	136.3 a	38.0 a	80.0 a	1.03 a	1.08 a	0.32 a	31.1 a
T3 - 250	128.8 a	35.7 a	78.3 a	0.87 a	1.02 a	0.43 a	29.4 a
T4 - 500	137.2 a	37.7 a	78.7 a	1.15 a	1.45 a	0.48 a	32.7 a
T5 - 1000	136.3 a	38.3 a	80.3 a	1.25 a	1.40 a	0.47 a	33.7 a
T6 - 1500	126.0 a	35.7 a	77.3 a	1.06 a	1.13 a	0.60 a	31.5 a
T7 - 2000	135.3 a	37.3 a	78.0 a	1.08 a	1.25 a	0.22 a	31.9 a
T8 - 2500	135.3 a	40.3 a	77.7 a	0.95 a	1.07 a	0.22 a	30.6 a
T9 - 3000	142.8 a	38.3 a	78.3 a	1.35 a	1.64 a	0.32 a	34.3 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.19. Phosphorus (P), potassium (K^+) and sodium (Na^+) content in wine of Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	P (mg/L)			K^+ (mg/L)			Na^+ (mg/L)		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	412 a(2)	175 a	3034 ab	1740 a	982 a	833 a	32.8 a	19.5 a	14.4 a
T2 - 100	423 a	184 a	3416 a	1971 a	1008 a	971 ab	31.7 a	18.7 a	15.2 a
T3 - 250	419 a	170 a	2498 c	1662 a	845 a	997 ab	28.9 a	17.6 a	13.2 a
T4 - 500	411 a	173 a	3153 ab	1709 a	836 a	1019 ab	30.4 a	17.5 a	14.8 a
T5 - 1000	384 a	183 a	3099 ab	1826 a	949 a	1022 ab	29.9 a	19.7 a	16.0 a
T6 - 1500	374 a	167 a	2906 bc	1879 a	927 a	1162 bc	28.9 a	18.7 a	14.3 a
T7 - 2000	411 a	171 a	3018 ab	2023 a	953 a	1111 bc	28.0 a	17.6 a	14.0 a
T8 - 2500	403 a	168 a	3096 ab	1933 a	987 a	1220 c	28.6 a	18.2 a	15.4 a
T9 - 3000	405 a	183 a	3088 ab	1929 a	1078 a	1224 c	31.8 a	19.1 a	15.6 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

Table 5.20. Chlorine (Cl^-) and sulphate (SO_4^{2-}) content in wine of Cabernet Sauvignon/99R irrigated using diluted winery wastewater during the 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	Cl^- (mg/L)			SO_4^{2-} (mg/L)		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	35.5 a(1)	38.6 a	50.4 a	587 a	108 ab	5311 a
T2 - 100	41.4 a	47.5 a	80.1 a	543 a	98 a	5144 a
T3 - 250	26.6 a	47.5 a	80.1 a	538 a	108 ab	4812 a
T4 - 500	35.5 a	44.5 a	65.3 a	565 a	108 ab	5040 a
T5 - 1000	23.7 a	44.5 a	68.2 a	556 a	126 bc	5022 a
T6 - 1500	35.5 a	38.6 a	86.0 a	549 a	127 bc	5138 a
T7 - 2000	38.5 a	38.6 a	50.4a	550 a	131 c	4920 a
T8 - 2500	32.6 a	44.5 a	86.0 a	566 a	137 cd	5191 a
T9 - 3000	35.5 a	56.4 a	47.5 a	547 a	153 d	5004 a

(1) Values designated by the same letters within a column do not differ significantly ($p \leq 0.05$).

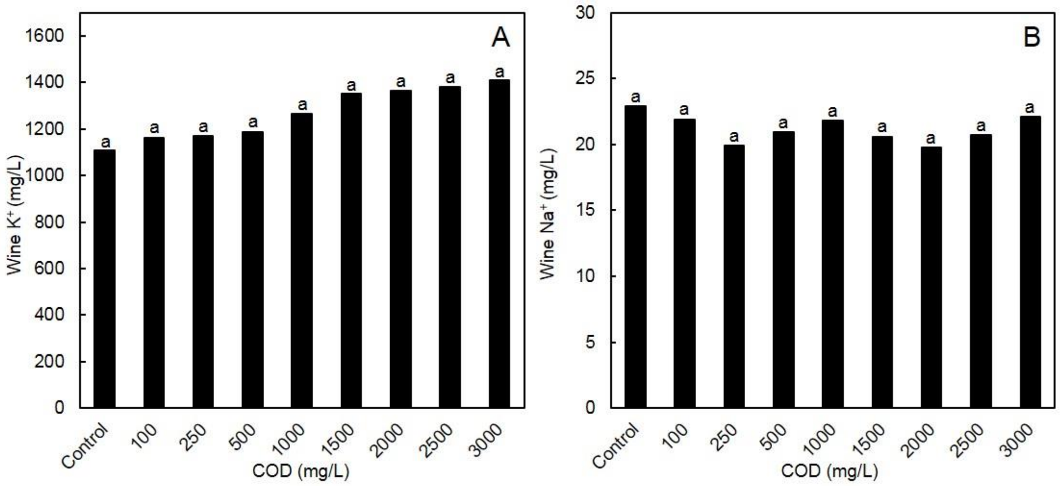

Figure 5.10. Effect of irrigation using diluted winery wastewater on wine (A) potassium (K^+) and (B) sodium (Na^+). Data are means for the 2010/11, 2011/12 and 2012/13 seasons.

5.4. CONCLUSIONS

Irrigation of grapevines using winery wastewater diluted up to a maximum COD level of 3000 mg/L did not affect vegetative growth or any of the yield components compared to the river water control. Consequently, the water use and water status of the grapevines was not affected by the wastewater irrigation under the prevailing conditions. The grapevines did not respond to level of COD *per se*. This indicated that sufficient aeration occurred between irrigations which allowed organic carbon breakdown. Although salinity and sodicity levels in the diluted winery wastewater were below the thresholds where growth and yield reductions are expected for grapevines, it should be monitored frequently. The low salinity and sodicity levels in the diluted winery wastewater could be a further explanation why the grapevines did not respond negatively to the wastewater irrigation. Since vegetative growth and yield of the grapevine were comparable to responses previously reported for vineyards without a summer interception crop, the results suggested that the grapevines were not affected by the pearl millet growing in the work rows during summer. Visual observations revealed that the root system of this cover crop was shallow compared to that of the grapevines. Therefore, the competition for water and nutrients was probably not strong enough to have induced negative effects on grapevine growth and yield. However, results indicated that a summer cover crop may increase the ET_c of vineyards substantially if growing conditions are favourable for the particular crop. The contribution of the slash and removal costs to already high production costs of vineyards is a further aspect that needs consideration.

Results showed that irrigation of grapevines using diluted winery wastewater did not have detrimental effects on juice characteristics with regard to ripeness parameters and ion content, with the exception of juice pH. Wine sensorial characteristics were not affected by irrigation using diluted winery wastewater. Under the conditions of the study, the relatively large irrigation volumes applied during berry ripening resulted in poor wine quality. Since wine quality is an important aspect, particularly if wine needs to be exported, the generally poor quality is of great concern. However, there is ample evidence that less frequent irrigation, which allows higher levels of plant available water (PAW) depletion between irrigations, will enhance wine quality. This implies that the winery wastewater will probably have to be applied over large areas to obtain sufficient PAW depletion between irrigations. Distribution of winery wastewater over large areas will need additional expensive infrastructure. Although the study showed that wine sensorial characteristics were not affected, off-odour due to direct contact with winery wastewater may reduce wine quality. The correct choice of irrigation system, *e.g.* micro-sprinklers or drippers, can eliminate this potential risk. In heavier textured soils, regions with lower winter rainfall, situations where more K^+ is applied *via* diluted winery wastewater or where no interception crop is cultivated during summer, responses with respect to leaf, shoot, juice and wine contents may be more pronounced and consistent. Under the prevailing conditions, it appeared that the General Authorisation for wineries could be revised to permit irrigation using diluted winery wastewater up to 3000 mg/L COD.

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CHAPTER 6: EFFECT OF IRRIGATION USING DILUTED WINERY WASTEWATER ON THE HYDRAULIC PROPERTIES OF FOUR SELECTED SOILS

6.1. INTRODUCTION

In the South African wine industry, most vineyards require irrigation. However, water is becoming an increasingly scarce resource and will become even more so should climate change realise. Wastewater is a potential important source of water for irrigation (Viviani & Iovino, 2004, Arienzo *et al.*, 2009) and in the wine industry, it would be ideal to use winery wastewater for vineyard irrigation by adding winery wastewater to existing irrigation water resources. The availability of such wastewater, in conjunction with the nutrients it could contain, makes it an attractive source of irrigation water (Arienzo *et al.*, 2009). In areas where grapevines do not require much irrigation, but where winery wastewater is being generated, it could potentially be used for irrigation of other crops. The composition of winery wastewater fluctuates with time of season *i.e.* pre-vintage, vintage and post-vintage (Arienzo *et al*., 2009; Bories & Sire, 2010), and potassium (K^+) and sodium (Na^+) concentrations are generally high (Arienzo *et al*., 2009; Samaras *et al*., 2009). Wastewater also contains high concentrations of organic and inorganic suspended and dissolved solids (Tarchitzky *et al*., 1999). Soil chemical properties can be altered by wastewater irrigation (Vogeler, 2009; Lado & Ben-Hur, 2010) and this could influence soil structure (Sparling *et al*., 2006) and hydraulic properties (Mathan, 1994; Sort & Alcaniz, 1999; Tarchitzky *et al*., 1999; Al-Haddabi *et al*., 2004; Coppola *et al.*, 2004; Viviani & Iovino, 2004; Bagarello *et al.*, 2006; Hawke & Summers, 2006; Bhardwaj *et al.*, 2007; Gonçalves *et al.*, 2007; Arienzo *et al*., 2009; Vogeler, 2009). Dissolved and suspended solids, both organic and inorganic, may induce soil clogging through physical, chemical, and biological processes (Viviani & Iovino, 2004).

The effects of wastewater irrigation are closely related to both wastewater and soil properties. An accumulation of monovalent cations, such as K^+ and Na^+ which are generally associated with winery wastewater, can have negative effects on soil structure (Laurenson *et al.*, 2012). Although the effect of irrigation using winery wastewater on soil chemical properties is well documented, its effect on soil physical properties is largely unknown particularly when used for vineyard irrigation (Buelow *et al.*, 2015). This could be due to the fact that changes in soil physical properties are difficult to quantify because they tend to occur only over the long term, and that soil physical properties are greatly variable (Hawke & Summers, 2006). Furthermore, most of the studies were conducted in laboratories using artificial solutions. Results of a laboratory study investigating the effect of sodium adsorption ratio (SAR) and potassium adsorption ratio (PAR) on soil hydraulic conductivity showed that it was considerably reduced when the SAR or the PAR exceeded 20 (Arienzo *et al*., 2009; Arienzo *et al*., 2012). In another study, Laurenson *et al*. (2012) used a combination of solutions with known SAR and PAR to investigate the binding of Na^+ and K^+, and concluded that exchangeable sodium percentage (ESP) corresponding to a given SAR was increasingly lowered at higher K^+ concentrations. Subsequently, if SAR in wastewater remains similar during vintage, reductions in ESP may occur because of increasing K^+ and exchangeable potassium percentage (EPP). Changes in soil structure will therefore be less pronounced compared to wastewaters with comparable monovalent concentrations of only Na^+. Therefore, in the case of winery wastewater, replacing Na^+-based cleaners with K^+-based cleaners can contribute towards decreasing clay dispersion risks. Due to the high K^+ content in winery wastewater, substitution of K^+-based cleaning agents with Na^+-based ones has been proposed (Arienzo *et al*., 2009). Using three soils of contrasting mineralogy packed in soil columns, it was found that soil mineralogy and Na^+ and K^+ concentrations in solutions were key factors influencing the soil hydraulic conductivity (Buelow *et al.*, 2015).

The objective of this study was to determine the effect of irrigation using winery wastewater diluted to five different chemical oxygen demand (COD) levels rather than river water on the near-saturation hydraulic conductivity of four different soils.

6.2. MATERIALS AND METHODS

Since inherent soil properties could influence the soil physical changes when irrigated with winery wastewater, different soils other than the Rawsonville sand, *i.e.* where the field experiment was carried out, were included in the study. The three additional soils were red, aeolic sand from the Lower Olifants River region and two soils from Stellenbosch which were derived from shale and granite parent material. The soil from both Lutzville and Rawsonville were classified as sand, whereas the shale soil was classified as fine sandy clay loam and the granite soil classified as a coarse sandy loam, respectively (Mulidzi *et al.*, 2016). The four different soils were collected and packed into 4.5 dm^3 PVC pots to a bulk density of 1.5 g/cm^3 with the exception of the granitic soil which was packed to a bulk density of 1.55 g/cm^3.

One pot per replication per treatment was used. In total, there were therefore 18 pots per soil. All of the pots were installed at the field experiment at Rawsonville in all three replications of the river water control (T1) and where winery wastewater was diluted to COD levels of 100 mg/L (T2), 500 mg/L (T4), 1000 mg/L (T5), 2000 mg/L (T7) and 3000 mg/L (T9), as described in Chapter 2. These treatments were selected in order to obtain a wide range of COD levels. The pots were installed in free draining pits that were 30 cm wide, 60 cm long and 20 cm deep. The soil at the bottom of the pits was leveled and thereafter coarse material was added to the bottom of each pit. The pots were placed onto the coarse material so that their top ends protruded *c.* 10 mm above the level of the soil surface (Fig. 6.1). A plastic lid was placed on the pot during the installation process to prevent soil and coarse material contaminating the soil surface. The pits were then filled with soil from the vineyard (Fig. 6.2). The level of the soil surface in the pots was the same as the surrounding soil.

The pots remained covered until irrigation using diluted winery wastewater commenced in either early February or March. The lids were only removed during the application of the diluted wastewater irrigations. Before river water was applied to grapevines after the wastewater irrigations to flush the irrigation lines, the lids were also placed back onto the pots. After the completion of each wastewater irrigation, the lids were removed and the soils were allowed to dry out until the next irrigation. The irrigations were usually applied at 14 day intervals. The soils were also covered when rainfall occurred between irrigations. Therefore, the soils were subjected only to the wastewater irrigations, *i.e.* with the exception of the river water control.

Figure 6.1. The PVC pots were covered with lids and placed onto a layer of coarse sand in the pits.

Figure 6.2. Soil from the vineyard was used to fill up the pits.

The four soils were subjected to the same diluted wastewater irrigations that the grapevines were subjected to in 2010/11, 2011/12 and 2012/2013. Further details regarding the amount of irrigation water applied, as well as the water quality and elements applied *via* the irrigation water are presented in Chapter 3. After the end of the application of the wastewater irrigations for each season, the pots were removed from the soil and transported to the irrigation laboratory at the ARC Nietvoorbij campus near Stellenbosch.

Mini disk infiltrometers (Decagon Devices, Pullman, WA) were used to quantify the near-saturation hydraulic conductivity (K_S) of the four different soils (Fig. 6.3). In order to perform measurements on each of the three replications per treatment concurrently, three of these mini disk infiltrometers were used. Mini disk infiltrometer measurements were replicated five times in each pot. Measurements of the Nietvoorbij shale and granitic soils were carried out at a suction head of 2 cm, whereas the measurements of the Rawsonville and Lutzville sands were carried out at a suction head of 4 cm. The K_{ns} values (mm/h) were calculated by means of the following equation:

$$K_S = \{[(V_I - V_E) \div 1000] \div 0.001521\} \text{ X } 60 \div \Delta t_{.} \qquad \text{(Eq.6.1)}$$

where V_I is initial volume reading (mL) at the beginning of the measurement, V_E is volume reading (mL) at the end of the measurement, 0.001521 is the area of the ceramic plate at the bottom of the infiltrometer in m^2 and $\Delta t_{.}$ is the time between measurements (min). It should be noted that data presented is the median value of the five values determined for near-saturation K.

Figure 6.3. Using a mini disk infiltrometer to measure near-saturation hydraulic conductivity in the laboratory at the ARC Nietvoorbij campus near Stellenbosch.

Water collected from the Holsloot River during the winter was used for the hydraulic conductivity studies. Samples of this river water were sent to a commercial laboratory for chemical analysis according to the methods of Clesceri *et al.* (1998). As expected, the water from the Holsloot River was of good quality and levels of elements were generally low (Table 6.1).

Table 6.1. Chemical composition of water from the Holsloot River used for the hydraulic conductivity studies.

Time of sampling	pH	EC (dS/m)	Element (mg/L)								
			Na^+	K^+	Ca^{2+}	Mg^{2+}	Fe^{2+}	Cl^-	HCO_3^-	SO_4^{2-}	P
July 2011	7.6	0.04	5.8	4.0	6.7	1.1	0.10	9.8	10.4	3.0	0.10
July 2012	7.0	0.03	2.3	0.2	0.6	0.7	0.12	9.2	20.4	1.5	0.04
July 2013	5.6	0.02	4.2	1.0	5.2	1. 7	0.02	8.9	1.5	21.5	0.04

6.3. RESULTS AND DISCUSSION

6.3.1. Shale-derived soil

The near-saturation K values were generally within the range expected for fine sandy clay soils (Klute & Dirksen, 1986). During the first two seasons, *i.e.* 2010/11 and 2011/12, irrigation using diluted winery wastewater had no effect on near-saturation K of the fine sandy clay loam shale-derived soil from Stellenbosch (data not shown). However, after the third season, *i.e.* 2012/13, near-saturation K became lower with a decrease in the level of dilution of the winery wastewater, *i.e.* an increase in COD level of the diluted winery wastewater (Fig. 6.4).

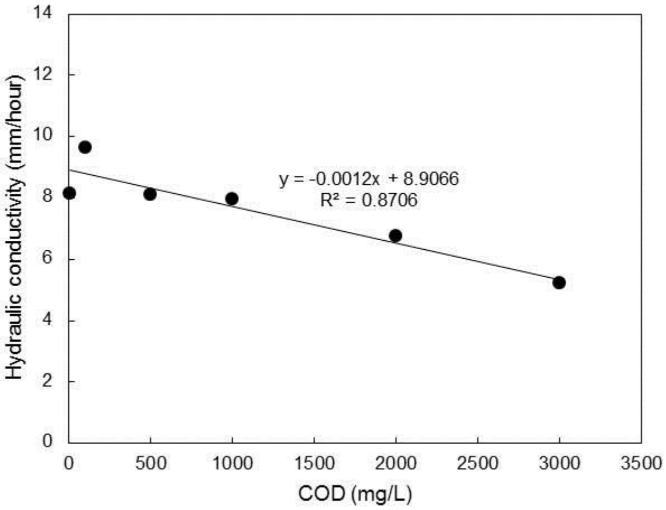

Figure 6.4. Effect of irrigation using diluted winery wastewater on the near-saturation hydraulic conductivity of shale soil from Stellenbosch in 2013, *i.e.* after three years.

Visual observations revealed that the shale-derived soil had a salt-like precipitation on the surface where the winery wastewater was diluted to 3000 mg/L (Fig. 6.5). This precipitation was present on all three replications, and could have been caused by chemical clogging. This includes swelling and dispersion of clay particles induced by Na^+ concentrations higher than that of fresh water. High Na^+ levels in irrigation water combined with low soil-water electrical conductivity can lower soil's permeability, and decrease its infiltration capacity through the swelling and dispersion of clays and slaking of aggregates (Al-Haddabi *et al*., 2004). Swelling of clays can narrow the conducting pores of soils, which could have caused the reduction in near-saturation K with a decrease in the level of winery wastewater dilution, *i.e.* an increase in COD level.

Figure 6.5. Salt-like precipitation on the surface of the fine sandy clay loam soil where winery wastewater diluted to 3000 mg/L was applied.

6.3.2. Granite-derived soil

There were no clear trends in near-saturation K for the granitic coarse sandy loam soil from Stellenbosch after three years with respect to the level of winery wastewater dilution (Fig. 6.6). It could be that the packing procedure *per se* induced a negative effect on the hydraulic properties of the granite soil. The near-saturation K of this particular soil was the lowest of the four soils and was within the range reported for sandy clay soils (Klute & Dirksen, 1986). The near-saturation K values were comparable to 2.32 mm/h reported previously for this particular soil (Louw & Bennie, 1991).

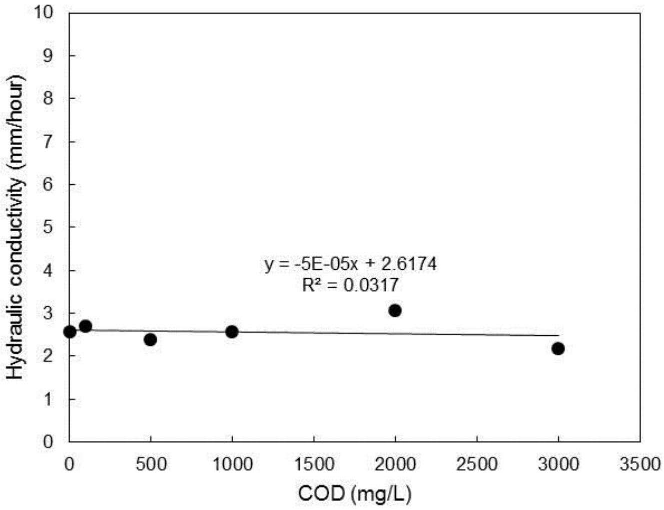

Figure 6.6. Effect of irrigation using diluted winery wastewater on the near-saturation hydraulic conductivity of granitic soil from Stellenbosch in 2013, *i.e.* after three years.

6.3.3. Alluvial sand

The near-saturation K values were generally within the range expected for sandy soils (Hillel, 1980; Klute & Dirksen, 1986). In 2010/11 and 2011/12, no clear trends were observed that could be related to the level of wastewater dilution in near-saturation K for the fine sandy soil from Rawsonville (data not shown). However, similar to the shale-derived soil, near-saturation K of this fine sandy soil showed a clear reduction with a decrease in level of dilution, *i.e.* an increase in COD level, of the winery wastewater after the third year of irrigation (Fig. 6.7). In 2012/13, the near-saturation K of the soil irrigated using winery wastewater diluted to 3000 mg/L COD (T9) was 40% less than that of the river water control (T1). It should be noted that the near-saturation K of the soil irrigated using winery wastewater diluted to 3000 mg/L was still relatively high.

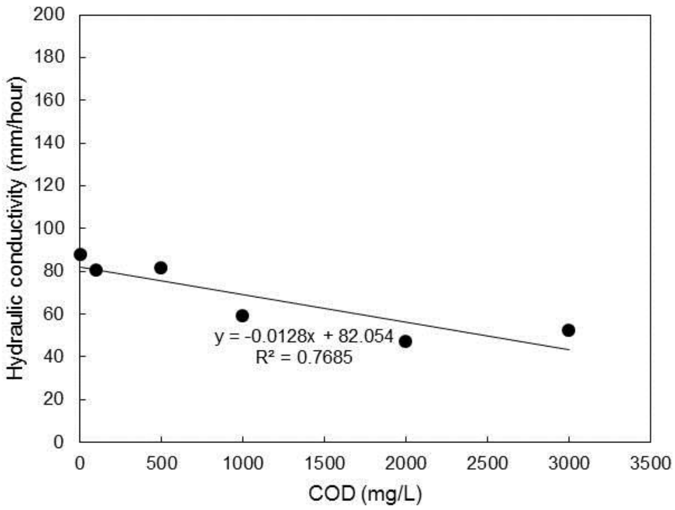

Figure 6.7. Effect of irrigation using diluted winery wastewater on the near-saturation hydraulic conductivity of sandy soil from Rawsonville in 2013, *i.e.* after three years.

6.3.4. Aeolian sand

Generally, near-saturation K was higher than expected for this fine sandy soil from Lutzville, and was more comparable to near-saturation K values reported for coarse sand (Klute & Dirksen, 1986). During the first two seasons, *i.e.* 2010/11 and 2011/12, irrigation using diluted winery wastewater had no effect on near-saturation K of the fine sandy soil (data not shown). However, after the third season, *i.e.* 2012/13, near-saturation K showed a substantial decrease with a decrease in the level of dilution, *i.e.* an increase in COD level of the diluted winery wastewater (Fig. 6.8).

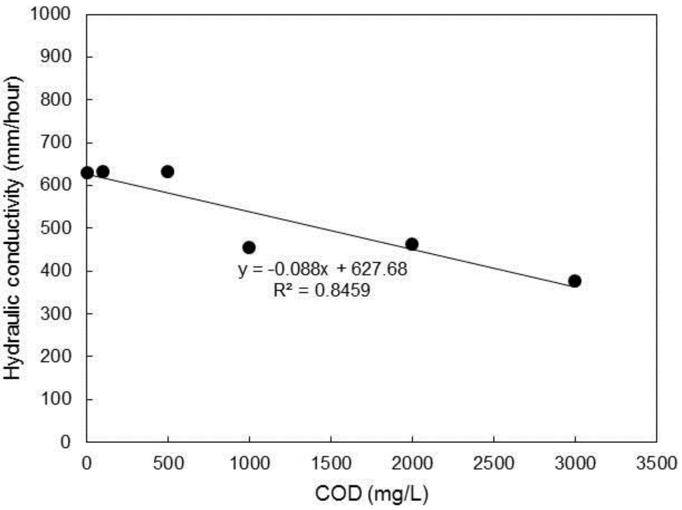

Figure 6.8. Effect of irrigation using diluted winery wastewater on the near-saturation hydraulic conductivity of sandy soil from Lutzville in 2013, *i.e.* after three years.

As in the case of the alluvial Rawsonville soil, near-saturation K was 40% lower where winery wastewater diluted to 3000 mg/L was applied (T9) compared to the river water control (T1).

6.4. CONCLUSIONS

As previous studies used artificial solutions, mostly on a laboratory scale, to investigate hydraulic properties of soil in response to wastewater irrigation, this study was the first where wastewater was diluted to irrigate four different soils in a field vineyard set up. During the first two years, irrigation using diluted winery wastewater did not seem to have had any consistent effect on near-saturation K of the soils, except for the granite-derived soil where there was no effect after three years. At this stage there is no explanation for the behaviour of the granite-derived soil, except that its water permeability is generally low. The packing procedure of the pots could also have induced a negative effect on the hydraulic properties thereof. In the case of the shale-derived soil, as well as the alluvial and aeolian sands, near-saturation K decreased substantially with a decrease in the level of dilution after three years. It should be noted that the soils received no river water irrigation which could have influenced the effect of the wastewater on near-saturation K. In spite of this, the results indicated that severe reductions in near-saturation K will occur in the long run if diluted winery wastewater is used for irrigation on these soils. Furthermore, the reduction in near-saturation K might be more pronounced if undiluted winery wastewater is used for irrigation of crops. Therefore, the absence of leaching by rain and/or irrigation probably explains why the diluted wastewater reduced near-saturation K even on the sandy soils after only three years. The latter result is in contrast with the field trial where irrigation using diluted winery wastewater apparently did not have any effect on hydraulic properties, *e.g.* infiltration rate, of the sandy alluvial soil. Furthermore, there is a possibility that the cover crops probably masked the negative effect of the winery wastewater on soil hydraulic properties in the field trial. However, investigating this aspect was beyond the scope of the study.

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CHAPTER 7: GENERAL CONCLUSIONS AND RESEARCH RECOMMENDATIONS

7.1. GENERAL CONCLUSIONS

Since treatment application in terms of chemical oxygen demand (COD) concentrations was quite accurate, *i.e.* after initial practical problems and sources of error were eliminated, the relatively simple mix and distribution infrastructure can be used where irrigation treatments require a series of water qualities, or nutrient levels in field experiments. However, due to the poor water quality, the tanks will have to be cleaned on the inside from time to time. This will be a costly operation that needs to be budgeted for in future projects of this kind. Results have shown that, although the deposit formation on the inside of the irrigation lines was low, it increased as the level of dilution decreased.

Since one of the incentives for diluting winery wastewater is that it could serve as a possible nutrient source, it is important to note that the nitrogen (N) load in the diluted winery wastewater was completely inadequate to supply the grapevine's annual requirement. On the positive side, phosphorus (P) loads in the winery wastewater diluted to 2500 mg/L COD and higher could supply more than adequate P if the grape yield amounts to 10 t/ha. Likewise, dilution of winery wastewater to 250 mg/L COD and higher could supply more than adequate potassium (K^+) if grape yield amounts to 10 t/ha. However, the excessive K^+ applied *via* the diluted wastewater could increase juice pH that could cause unstable musts and wines, as well as a reduction in the degree of ionisation of anthocyanins in the wine. Furthermore, excessive K^+ application could induce nutrient imbalances in the grapevine tissues, particularly antagonisms with respect to N, Ca^{2+} and Mg^{2+}. Given that the amounts of K^+ applied *via* the diluted winery wastewater were considerably higher than the grapevine's requirements, the cultivation and removal of a suitable interception crop during summer might be useful to absorb excessive K^+.

Under the prevailing conditions, soil K^+ increased with a decrease in the dilution of the wastewater during all four seasons. After four years, only the lowest level of dilution, *i.e.* 3000 mg/L COD, maintained baseline K^+ levels. Soil calcium (Ca^{2+}) and magnesium (Mg^{2+}) did not show any consistent responses to the different levels of wastewater dilution because there were no substantial differences in amounts of these particular elements applied *via* the irrigation water. Generally, soil sodium (Na^+) increased with a decrease in the dilution of the wastewater. There were substantial differences in the amount of Na^+ applied *via* the irrigation water. Although irrigation using winery wastewater had almost no other effects under the prevailing conditions, element accumulation, particularly with respect to K^+ and Na^+, might be more prominent in heavier textured soils or in regions with low winter rainfall. Development of regression models to estimate soil Bray II-K after wastewater application highlighted the necessity of cultivating a pearl millet interception crop, particularly when substantial amounts of K are applied *via* diluted winery wastewater. Estimations using the models also showed that even an amount of 50 kg/ha K^+ applied *via* diluted wastewater would increase soil Bray II-K to above the recommended level for an alluvial sandy soil in the Breede River Valley where an interception cover crop of pearl millet of 5 t/ha DMP is cultivated. Models to estimate soil Bray II-K at bud break confirmed that higher rainfall leads to more extensive leaching and lower soil Bray II-K, whereas rainfall lower than the average rainfall would lead to less leaching of K^+, and, consequently, higher soil Bray II-K. The pot experiment has shown that irrigation with winery wastewater diluted to 3000 mg/L COD has negative effects on the soil hydraulic properties, under no rainfall conditions.

Irrigation of grapevines using diluted winery wastewater did not affect vegetative growth or yield of grapevines compared to the river water control under the prevailing conditions. The grapevines did not respond to level of COD *per se*. This indicated that sufficient aeration occurred between irrigations which allowed organic carbon breakdown. Although salinity and sodicity levels in the diluted winery wastewater were below the thresholds where growth and yield reductions are expected for grapevines, it should be monitored frequently. The low salinity and sodicity levels in the diluted winery wastewater could be a further explanation why the grapevines did not respond negatively to the wastewater irrigation. Since vegetative growth and yield of the grapevine were comparable to responses previously reported for vineyards without a summer interception crop, the results suggested that the grapevines were not affected by the pearl millet growing in the work rows during summer. Visual observations revealed that the root system of this cover crop was shallow compared to that of the grapevines. Therefore, the competition for water and nutrients was probably not strong enough to have induced negative effects on grapevine growth and yield. Results showed that irrigation of grapevines using diluted winery wastewater did not have detrimental effects on juice characteristics with regard to ripeness parameters and ion content. Wine sensorial characteristics were not affected by irrigation using diluted winery wastewater. The relatively large irrigation volumes applied during berry ripening were definitely detrimental to wine quality. Although the study showed that wine colour and common sensory wine descriptors were not affected by the various wastewater irrigation treatments, off-odours due to direct contact with wastewater may reduce wine quality.

Based on the project results, the following criteria should be considered for possible amendments to the General Authorisation for wineries when using diluted wastewater for irrigation of vineyards:

(i) The COD must be diluted to 3000 mg/L or less, preferably to less than 2000 mg/L to avoid unpleasant odours in the vineyard while irrigations are applied.

(ii) The electrical conductivity (EC_{iw}) must be less than 0.75 dS/m.

(iii) The sodium adsorption ratio (SAR_{iw}) must be less than 5.

(iv) The soil must have a low cation exchange capacity.

(v) The internal drainage in the root zone must be unrestricted.

(vi) The irrigation water must not percolate beyond the root depth.

(vii) Only micro-sprinklers should be used, since drippers have narrow flow paths and/or small orifices, and are more susceptible to clogging.

(viii) The irrigation must be applied with micro-sprinklers in such a way that the bunches are not wetted.

(ix) At least 50% plant available water (PAW) depletion should be allowed between irrigations to allow sufficient aeration for oxidation of organic material applied *via* the irrigation water.

(x) The irrigation frequency and volumes (schedule) should enhance, rather than negate, wine quality characteristics.

7.2. RESEARCH RECOMMENDATIONS

The following are more general recommendations and suggestions that need to be considered if diluted winery wastewater is used for vineyard irrigation.

- Since the organic matter in the wastewater seemed to have oxidized between irrigations, it might be more appropriate to evaluate the water quality for vineyard irrigation in terms of pH, EC_{iw} and SAR rather than COD.

- Furthermore, it must be determined if the existing COD norms could be relaxed for winery wastewaters, since the organic material in the latter could break down more readily upon aeration between irrigations compared to organic material in wastewaters produced by other processes, *e.g.* dairies and cheese factories.

- If the industry moves towards K^+ based cleaning agents, further research should be done to determine acceptable potassium adsorption ratio (PAR) norms to avoid excessive K^+ application and accumulation in soils, and subsequently in grapevines.

- Dilution of winery wastewater to a specific COD level for re-use *via* irrigation proved to be a tedious process that requires expertise and specialized equipment. Due to these constraints, and the need for large storage dams, it will be impractical to pre-dilute wastewater on a commercial scale, particularly in the case of larger wineries. In the case of smaller wineries, the wastewater could be diluted by mixing it into existing dams or reservoirs, *i.e.* given that the water quality remains within the statutory quality norms.

- A field study is necessary to investigate the fitness for use of winery wastewater for irrigation of different soil types with varying rainfall quantities and leaching levels on vineyard performance in terms of yield and quality under field conditions. Since climatic conditions range considerably in the Western Cape, it would be possible to investigate the effect of climatic factors such as magnitude of rainfall on the possibility of using winery wastewater for vineyard irrigation. Since it is also well known that soil type can influence nutrient element adsorption and accumulation, it would also be possible to investigate different soil types within the same climatic zone. However, it would be impractical to dilute winery wastewater to a pre-determined level before each irrigation, *i.e.* specifically at the commercial level. The primary reason is that it would be difficult to monitor the winery wastewater quality continuously in order to adjust the volumes of raw and wastewater to obtain a required level of dilution. Therefore, a more practical approach would be to use in-field dilution (augmentation) of winery wastewater with raw water. According to this approach, grapevines would be irrigated as follows: for each irrigation, a certain percentage of the irrigation requirement would be applied as undiluted winery wastewater. Raw water would then be applied for the other part of the irrigation requirement.

- For larger wineries, injecting the wastewater directly into existing irrigation systems on soils and grapevines might be an alternative. However, injecting wastewater directly into irrigation systems will require continuous monitoring of the wastewater quality in order to adjust the injection rate into the irrigation water as the quality of the wastewater changes. Since the quality of winery wastewater changes almost hourly, it will complicate the injection process. Therefore, it might be more viable to inject the wastewater at a fixed dilution ratio, *i.e.* volume per volume. For a specific winery, the ideal dilution ratio will depend on the wastewater quality, as well as the temporal variability thereof. Since spikes of extremely poor water quality could occur within a given dilution ratio, the sustainability of the fixed dilution concept needs to be investigated with respect to soil and grapevine responses in a field trial.

- Given the importance of wine quality, particularly if the wine needs to be exported, the generally poor wine quality obtained under the prevailing conditions is of great concern. Since less frequent irrigation allows higher levels of plant available water depletion between irrigations, the winery wastewater will probably have to be applied over large areas under grapevines to obtain sufficient PAW depletion, *i.e.* at least 80% depletion, between irrigations. The latter approach will require careful planning and design, as well as expensive infrastructure that will inevitably increase the cost of water management.

- Although soil Na^+ and K^+ did not accumulate to excessively high levels in the sandy soil of the field trial, they were reduced to the baseline concentrations following heavy winter rain. This result is of great concern, since it suggests that these elements leached into natural water resources. Leaching due to heavy rainfall also serves as further motivation to investigate the possibility of biological interception of mineral elements. In contrast to pearl millet where the roots remain in the soil, halophytes such as fodder beet have the advantage that almost the entire plant is removed at harvest. Similar to fodder beet, halophytic grasses could be grown to absorb Na^+ from soils. To ensure optimum element uptake from the soil, the halophytes must be able to grow during the period when wineries produce the highest volumes of wastewater. Irrigation of vineyards in heavier textured soils using diluted winery wastewater cannot be recommended, unless Na^+ and K^+ can be successfully intercepted in a biological way with halophytes. If Na^+ could be intercepted effectively, wineries could continue to use Na hydroxide in a judicious way, which will be more efficient and cheaper than K^+ containing cleaning agents. Therefore, the efficacy of selected halophytes, such as fodder beet and grasses, to absorb Na^+ from different textured soils, and the suitability of the halophytes to different soil and climatic conditions in the wine producing regions of South Africa needs to be investigated.

APPENDIX 1 - Diluted winery wastewater applications

Appendix 1.1. Mean amount of diluted winery wastewater (mm) applied per irrigation to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11(1)	2011/12(2)	2012/13
T1 - River water	N/A	N/A	N/A	N/A
T2 - 100	38.8±4.6	41.1±0.3	40.8±0.1	41.0±0.2
T3 - 250	38.9±4.4	41.5±0.2	41.7±0.2	42.0±0.7
T4 - 500	39.1±4.5	41.1±0.3	40.8±0.2	40.9±0.5
T5 - 1000	39.4±4.1	41.3±0.4	41.4±0.1	40.9±1.2
T6 - 1500	39.4±4.0	41.8±0.2	42.0±0.2	41.7±0.2
T7 - 2000	39.2±3.7	41.2±0.7	40.8±0.6	40.8±0.4
T8 - 2500	38.7±4.2	40.7±0.2	40.7±0.2	40.0±1.3
T9 - 3000	38.2±4.5	40.8±0.2	40.7±0.1	40.7±0.4

(1) Due to low COD levels in the winery wastewater at the end of the irrigation season, T3 to T9 had to be diluted and applied in two irrigations on consecutive days in the middle of April. For the calculation of the mean volume, the volumes applied on these consecutive days were added together.

(2) Due to low COD levels in the winery wastewater as well as limited available water, only 1/3 of a tank of diluted water could be applied on 2 May. Therefore, this data was not used for the calculation of the averages.

Appendix 1.2. Mean amount of river water (mm) applied per irrigation to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
T1 - River water	59.5±12.6	65.3±5.2	49.4±20.9	49.1±5.7
T2 - 100	15.4±3.4	25.1±0.6	14.3±13.8	8.1±5.6
T3 - 250	15.5±3.2	25.6±0.5	13.5±13.8	7.5±4.9
T4 - 500	15.8±3.4	25.4±0.7	14.3±13.8	8.1±5.2
T5 - 1000	16.4±3.4	25.3±0.4	13.7±13.9	7.9±5.5
T6 - 1500	16.2±3.1	25.8±0.4	13.3±13.7	7.9±5.2
T7 - 2000	14.5±3.3	25.2±0.4	14.3±13.5	8.1±5.8
T8 - 2500	15.3±2.9	24.9±0.5	14.3±14.0	8.3±5.7
T9 - 3000	15.0±2.9	25.1±0.7	14.7±13.8	8.3±5.8

Appendix 1.3. Mean total amount of irrigation water (mm) applied per irrigation to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
T1 - River water	59.5±12.6	65.3.±5.2	49.4±20.9	49.1±5.7
T2 - 100	54.2±2.1	66.0±0.8	49.4±20.8	49.1±5.7
T3 - 250	54.4±1.9	67.1±0.5	49.4±20.9	49.5±5.5
T4 - 500	54.9±2.2	66.6±0.8	49.4±20.9	49.0±5.8
T5 - 1000	55.8±2.7	66.5±0.3	49.4±20.9	48.9±6.1
T6 - 1500	55.6±2.5	67.6±0.4	49.4±20.8	49.6±5.4
T7 - 2000	53.7±0.5	66.5±0.7	49.4±20.9	48.9±5.9
T8 - 2500	54.0±2.6	65.6±0.5	49.4±20.9	48.3±6.5
T9 - 3000	53.2±2.7	65.9±0.6	49.4±20.9	49.0±5.7

Appendix 1.4. Total amount of diluted winery wastewater (mm) applied per season to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
T1 - River water	N/A(1)	N/A	N/A	N/A
T2 - 100	116.3	205.3	175.8	246.2
T3 - 250	116.7	249.2	179.5	251.7
T4 - 500	117.3	246.6	175.6	245.5
T5 - 1000	118.2	247.5	178.1	245.6
T6 - 1500	118.3	251.0	180.7	250.0
T7 - 2000	117.6	247.3	175.6	244.9
T8 - 2500	116.1	244.1	175.2	239.8
T9 - 3000	114.6	245.0	173.3	244.4

(1) Not applicable.

Appendix 1.5. Total amount of river water (mm) applied per season to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
T1 - River water	178.5	391.6	246.9	294.7
T2 - 100	46.2	190.9	71.3	48.5
T3 - 250	46.5	153.5	67.4	45.1
T4 - 500	47.5	152.1	71.3	48.6
T5 - 1000	49.2	151.7	68.7	47.6
T6 - 1500	48.5	154.7	66.6	47.5
T7 - 2000	43.5	151.4	71.3	48.4
T8 - 2500	46.0	149.4	71.7	50.0
T9 - 3000	45.0	150.4	73.5	49.7

Appendix 1.6. Total amount of irrigation water (mm) applied per season to Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
T1 - River water	178.5	391.6	246.9	294.7
T2 - 100	162.5	395.9	247.0	294.7
T3 - 250	163.2	402.8	246.9	297.0
T4 - 500	164.8	399.6	246.8	294.0
T5 - 1000	167.5	399.2	246.9	293.3
T6 - 1500	166.8	405.7	247.2	297.6
T7 - 2000	161.1	398.8	246.9	293.2
T8 - 2500	162.0	393.7	246.9	289.8
T9 - 3000	159.5	395.3	246.8	294.1

APPENDIX 2 - Water quality

Appendix 2.1. The pH in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	5.4±0.3	4.3±0.8	4.7±0.7	4.4±0.5
T1 - River water	6.6±0.5	5.8±0.5	6.1±0.2	5.5±0.5
T2 - 100	6.6±0.2	5.7±0.8	5.9±0.8	5.6±0.5
T3 - 250	6.4±0.2	5.5±0.9	5.3±0.6	5.1±0.5
T4 - 500	5.8±0.5	4.9±0.8	4.9±0.6	4.7±0.8
T5 - 1000	5.5±0.5	4.4±0.6	4.8±0.6	4.6±0.8
T6 - 1500	5.4±0.7	4.6±0.9	4.7±0.6	4.5±0.8
T7 - 2000	5.3±0.6	4.4±0.9	4.6±0.7	4.5±0.8
T8 - 2500	5.5±0.4	4.4±1.0	4.6±0.8	4.4±0.8
T9 - 3000	5.4±0.4	4.4±0.9	4.6±0.7	4.4±0.8

Appendix 2.2. The electrical conductivity (EC_{iw}) in dS/m in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	2.30±0.93	1.50±0.51	1.63±0.39	1.11±0.28
T1 - River water	0.13±0.02	0.19±0.02	0.13±0.07	0.12±0.06
T2 - 100	0.16±0.02	0.20±0.03	0.16±0.07	0.13±0.06
T3 - 250	0.19±0.03	0.22±0.04	0.19±0.06	0.15±0.06
T4 - 500	0.26±0.04	0.27±0.05	0.25±0.03	0.19±0.06
T5 - 1000	0.42±0.13	0.36±0.08	0.34±0.06	0.24±0.07
T6 - 1500	0.53±0.20	0.43±0.10	0.42±0.11	0.27±0.07
T7 - 2000	0.64±0.26	0.53±0.17	0.50±0.20	0.32±0.09
T8 - 2500	0.74±0.29	0.59±0.21	0.61±0.25	0.37±0.10
T9 - 3000	0.87±0.36	0.69±0.30	0.66±0.24	0.42±0.11

Appendix 2.3. The chemical oxygen demand (COD) in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	7624±854	10098±4163	11990±9178	10560±1567
T1 - River water	0±0	1±3	4±5	0±0
T2 - 100	81±45	100±117	100±59	96±45
T3 - 250	247±70	250±142	247±38	245±30
T4 - 500	545±83	499±181	492±102	499±61
T5 - 1000	1144±353	1004±233	978±225	976±52
T6 - 1500	1560±424	1486±378	1512±91	1587±498
T7 - 2000	2130±601	1994±408	1976±229	2087±273
T8 - 2500	2480±582	2520±610	2474±294	2597±503
T9 - 3000	2975±652	2997±641	3020±342	3053±295

Appendix 2.4. The ammonium nitrogen (NH_4^+-N) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. The NH_4^+-N was not determined in the 2009/10 season.

Treatment no. & target COD (mg/L)	2010/11	2011/12	2012/13
Winery wastewater	2.280±1.777	7.618±8.759	7.845±14.470
T1 - River water	0.369±0.362	0.497±0.656	0.458±0.345
T2 - 100	0.159±0.078	0.135±0.106	0.255±0.213
T3 - 250	0.198±0.123	0.130±0.138	0.172±0.136
T4 - 500	0.172±0.057	0.741±0.932	0.182±0.153
T5 - 1000	0.214±0.056	1.191±1.984	0.420±0.724
T6 - 1500	0.517±0.713	2.168±3.608	0.733±1.216
T7 - 2000	0.298±0.095	3.005±4.854	1.147±1.798
T8 - 2500	0.337±0.172	3.720±6.251	1.453±2.474
T9 - 3000	0.430±0.265	4.579±8.157	0.695±0.635

Appendix 2.5. The nitrate nitrogen (NO_3^--N) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. The NO_3^--N was not determined in the 2009/10 season.

Treatment no. & target COD (mg/L)	2010/11	2011/12	2012/13
Winery wastewater	2.683±4.737	0.092±0.198	8.608±15.473
T1 - River water	0.970±0.362	1.038±1.158	1.493±2.446
T2 - 100	0.492±0.371	0.249±0.227	0.337±0.412
T3 - 250	0.267±0.304	0.166±0.185	0.108±0.185
T4 - 500	0.067±0.151	0.220±0.223	0.162±0.328
T5 - 1000	0.162±0.286	0.224±0.309	0.343±0.747
T6 - 1500	0.282±0.599	0.214±0.304	0.630±1.222
T7 - 2000	0.684±1.286	0.337±0.483	1.008±1.503
T8 - 2500	0.663±1.585	1.090±1.572	1.673±2.558
T9 - 3000	0.850±1.843	1.027±1.927	1.985±3.099

Appendix 2.6. The total nitrogen (Total-N) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. The total-N was not determined in the 2009/10 season.

Treatment no. & target COD (mg/L)	2010/11	2011/12	2012/13
Winery wastewater	4.963±5.611	7.710±8.672	16.453±18.224
T1 - River water	1.339±0.621	1.535±1.010	1.952±2.494
T2 - 100	0.651±0.440	0.383±0.325	0.592±0.583
T3 - 250	0.465±0.410	0.296±0.296	0.280±0.254
T4 - 500	0.239±0.157	0.961±0.877	0.343±0.378
T5 - 1000	0.376±0.258	1.415±1.875	0.763±0.978
T6 - 1500	0.800±1.303	2.381±3.502	1.363±1.551
T7 - 2000	0.982±1.235	3.342±4.694	2.155±2.098
T8 - 2500	1.000±1.493	4.810±5.722	3.127±3.136
T9 - 3000	1.281±1.717	5.606±7.661	2.680±3.219

Appendix 2.7. The phosphorus (P) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	7.43±8.37	22.11±13.03	18.80±8.63	10.82±4.61
T1 - River water	0.00±0.00	0.02±0.04	0.03±0.03	0.24±0.26
T2 - 100	0.00±0.00	0.02±0.04	0.17±0.19	0.23±0.24
T3 - 250	0.00±0.00	0.18±0.22	0.45±0.39	0.28±0.28
T4 - 500	0.20±0.35	0.64±0.44	1.02±0.92	0.54±0.50
T5 - 1000	0.67±1.15	1.64±0.90	2.12±1.74	1.12±0.77
T6 - 1500	1.20±1.82	2.65±1.37	3.11±2.44	1.53±1.04
T7 - 2000	1.67±2.39	3.87±1.81	4.08±3.91	2.08±1.21
T8 - 2500	2.60±2.76	4.85±2.10	5.27±4.91	2.55±1.47
T9 - 3000	3.30±3.40	6.26±3.01	6.40±5.92	3.04±1.58

Appendix 2.8. The calcium (Ca^{2+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	40.4±20.4	24.9±5.0	47.7±17.3	29.3±11.7
T1 - River water	5.4±0.8	7.7±0.6	5.2±3.3	6.5±2.4
T2 - 100	5.8±0.7	7.8±0.5	5.6±3.1	7.2±2.6
T3 - 250	6.3±0.5	7.7±0.9	6.8±2.4	7.6±2.6
T4 - 500	7.5±1.0	8.8±1.2	8.6±0.8	8.5±2.7
T5 - 1000	10.0±2.9	9.7±1.5	11.8±2.3	9.6±2.6
T6 - 1500	12.0±4.7	10.9±1.8	14.2±4.0	10.0±2.3
T7 - 2000	13.9±6.4	11.8±2.0	17.8±8.2	11.5±3.0
T8 - 2500	15.8±6.5	12.4±2.2	20.6±11.2	12.6±3.0
T9 - 3000	17.5±7.5	13.5±2.5	24.1±14.3	13.0±2.8

Appendix 2.9. The magnesium (Mg^{2+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	8.5±1.9	9.8±2.5	11.0±4.9	9.9±5.3
T1 - River water	3.5±0.6	4.9±0.2	3.4±2.2	3.3±1.5
T2 - 100	3.6±0.6	5.0±0.2	3.5±2.1	3.6±1.5
T3 - 250	3.7±0.5	5.0±0.2	3.8±2.1	3.7±1.5
T4 - 500	3.9±0.5	5.2±0.3	3.9±1.9	4.0±1.4
T5 - 1000	4.2±0.4	5.5±0.4	4.4±1.8	4.5±1.5
T6 - 1500	4.5±0.3	5.7±0.5	4.8±1.9	4.6±1.4
T7 - 2000	4.7±0.4	6.1±0.6	5.1±1.5	5.0±1.5
T8 - 2500	5.3±0.1	6.2±0.8	5.5±1.3	5.5±1.8
T9 - 3000	5.3±0.5	6.5±0.9	6.0±1.2	5.6±1.8

Appendix 2.10. The potassium (K^{+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	621.2±264.6	241.6±109.0	313.7±93.0	189.1±58.2
T1 - River water	2.9±1.4	2.0±1.0	2.3±1.6	2.0±1.0
T2 - 100	9.4±5.2	4.3±3.5	5.0±3.5	3.7±2.0
T3 - 250	17.7±10.6	11.3±9.5	11.2±3.8	8.5±3.6
T4 - 500	33.6±12.8	21.3±17.4	20.6±8.6	16.4±7.9
T5 - 1000	74.3±41.9	39.9±33.0	38.3±14.2	28.2±11.6
T6 - 1500	105.6±60.1	55.2±42.0	51.5±20.3	35.0±15.6
T7 - 2000	135.2±76.4	75.3±59.1	62.8±23.0	44.7±16.3
T8 - 2500	162.7±88.9	89.4±70.2	84.4±38.0	55.6±21.2
T9 - 3000	197.8±114.2	112.4±95.3	100.2±46.7	65.1±25.6

Appendix 2.11. Potassium adsorption ratio (PAR) in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	13.51±3.22	6.25±2.94	6.40±1.97	4.74±1.60
T1 - River water	0.14±0.08	0.08±0.04	0.11±0.05	0.10±0.06
T2 - 100	0.46±0.27	0.17±0.14	0.25±0.15	0.18±0.12
T3 - 250	0.84±0.52	0.46±0.38	0.55±0.24	0.39±0.19
T4 - 500	1.47±0.57	0.82±0.65	0.90±0.46	0.72±0.38
T5 - 1000	2.89±1.45	1.47±1.12	1.44±0.55	1.15±0.54
T6 - 1500	3.79±1.77	1.97±1.39	1.77±0.64	1.39±0.70
T7 - 2000	4.59±2.00	2.58±1.87	1.96±0.51	1.67±0.69
T8 - 2500	5.20±2.29	3.01±2.18	2.47±0.74	1.98±0.83
T9 - 3000	6.09±2.76	3.60±2.77	2.73±0.86	2.27±0.95

Appendix 2.12. The sodium (Na^+) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	120.6±49.8	92.8±43.8	90.8±11.4	86.0±48.4
T1 - River water	10.2±0.6	12.8±0.9	11.7±6.0	10.1±3.2
T2 - 100	11.7±0.8	14.0±1.7	13.4±6.1	10.9±3.0
T3 - 250	13.4±1.2	17.7±5.8	15.0±4.6	12.5±3.2
T4 - 500	16.5±0.8	24.6±12.3	18.4±3.0	14.5±2.6
T5 - 1000	24.3±5.5	30.7±20.3	23.8±5.8	18.8±4.3
T6 - 1500	29.7±8.1	37.6±24.2	28.0±10.6	20.6±5.8
T7 - 2000	35.4±10.0	41.5±25.7	34.5±18.4	24.3±7.1
T8 - 2500	42.6±8.7	46.0±31.8	40.6±25.4	27.9±8.6
T9 - 3000	48.9±12.0	52.4±37.5	46.0±31.4	31.5±11.1

Appendix 2.13. Sodium adsorption ratio (SAR) in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	4.49±1.08	4.08±2.16	3.16±0.65	3.40±1.49
T1 - River water	0.84±0.02	0.89±0.07	0.98±0.19	0.81±0.17
T2 - 100	0.94±0.06	0.96±0.11	1.10±0.18	0.83±0.14
T3 - 250	1.05±0.13	1.22±0.39	1.16±0.18	0.93±0.16
T4 - 500	1.21±0.08	1.63±0.81	1.32±0.20	1.05±0.18
T5 - 1000	1.62±0.26	1.92±1.16	1.51±0.40	1.27±0.32
T6 - 1500	1.84±0.31	2.27±1.35	1.63±0.57	1.36±0.41
T7 - 2000	2.09±0.33	2.41±1.37	1.80±0.73	1.51±0.44
T8 - 2500	2.37±0.19	2.63±1.67	1.98±0.89	1.66±0.48
T9 - 3000	2.61±0.27	2.84±1.82	2.08±1.01	1.82±0.58

Appendix 2.14. The chloride (Cl^-) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	32.0±1.0	57.7±21.2	49.7±25.9	63.1±47.0
T1 - River water	22.6±1.8	29.7±3.2	25.3±9.5	27.4±7.8
T2 - 100	22.0±4.1	29.1±3.7	27.0±9.4	25.6±8.5
T3 - 250	20.5±2.5	29.4±2.4	26.3±8.9	26.3±5.9
T4 - 500	19.5±4.2	28.7±2.2	25.9±6.3	27.6±8.6
T5 - 1000	21.7±4.4	29.1±3.3	25.0±8.6	29.0±10.6
T6 - 1500	23.8±1.8	31.1±2.8	25.6±11.6	30.9±9.1
T7 - 2000	23.4±5.2	32.9±4.0	23.1±7.0	28.4±9.6
T8 - 2500	21.1±2.3	31.7±2.2	30.7±9.8	27.7±7.6
T9 - 3000	22.9±3.5	34.6±3.5	31.4±9.7	36.6±13.9

Appendix 2.15. The bicarbonate (HCO_3^-) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	1293.3±505.3	365.0±306.1	382.8±284.4	106.5±260.7
T1 - River water	12.8±4.4	19.4±6.0	13.9±4.3	3.9±4.1
T2 - 100	33.7±9.3	20.7±3.6	16.8±5.2	6.1±4.5
T3 - 250	53.6±27.6	32.1±23.4	28.5±14.2	7.3±9.4
T4 - 500	91.6±48.5	43.9±35.7	43.2±33.1	7.6±15.9
T5 - 1000	164.9±108.1	65.5±71.7	65.9±57.1	10.2±21.4
T6 - 1500	221.0±163.9	110.1±87.1	90.6±78.7	15.5±38.0
T7 - 2000	296.0±194.5	127.3±156.4	124.0±143.0	19.1±46.8
T8 - 2500	373.1±185.3	145.0±173.9	139.0±163.2	25.0±61.1
T9 - 3000	451.7±235.0	190.3±212.4	176.1±185.0	28.0±65.7

Appendix 2.16. The sulphate (SO_4^{2-}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	203.6±122.3	54.4±28.2	179.5±259.7	114.2±119.2
T1 - River water	21.9±6.9	27.8±2.1	21.3±9.1	21.2±11.1
T2 - 100	20.0±4.0	27.8±3.2	17.6±10.0	20.0±8.5
T3 - 250	16.5±4.1	27.8±2.4	18.5±10.6	20.3±7.9
T4 - 500	21.9±6.5	31.5±9.2	19.6±7.6	28.7±20.1
T5 - 1000	29.8±11.8	28.2±1.6	45.9±39.2	24.3±10.0
T6 - 1500	32.8±12.1	25.4±6.9	43.9±36.3	34.7±32.6
T7 - 2000	23.0±9.2	29.4±2.8	107.5±132.1	43.3±49.5
T8 - 2500	89.6±37.8	26.1±8.9	141.3±234.7	41.7±39.6
T9 - 3000	65.5±42.2	38.7±22.7	153.5±290.1	40.7±37.0

Appendix 2.17. The boron (B^{3+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	1.03±1.11	0.90±0.55	0.92±1.01	0.35±0.14
T1 - River water	0.06±0.06	0.02±0.01	0.01±0.01	0.01±0.01
T2 - 100	0.03±0.02	0.03±0.02	0.02±0.03	0.01±0.01
T3 - 250	0.04±0.02	0.05±0.04	0.05±0.08	0.02±0.01
T4 - 500	0.06±0.01	0.08±0.06	0.09±0.15	0.02±0.02
T5 - 1000	0.1±0.05	0.14±0.13	0.18±0.30	0.04±0.03
T6 - 1500	0.15±0.07	0.19±0.17	0.25±0.43	0.04±0.04
T7 - 2000	0.19±0.10	0.25±0.24	0.37±0.67	0.07±0.04
T8 - 2500	0.26±0.09	0.30±0.29	0.47±0.84	0.09±0.04
T9 - 3000	0.33±0.11	0.38±0.39	0.56±1.01	0.09±0.04

Appendix 2.18. The iron (Fe^{2+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12	2012/13
Winery wastewater	4.23±1.90	3.30±1.74	5.63±2.45	2.09±1.58
T1 - River water	0.03±0.06	0.32±0.75	0.10±0.05	0.03±0.04
T2 - 100	0.07±0.06	0.67±1.57	0.14±0.03	0.06±0.06
T3 - 250	0.10±0.10	0.48±1.16	0.21±0.07	0.11±0.07
T4 - 500	0.23±0.15	0.53±1.11	0.30±0.10	0.17±0.07
T5 - 1000	0.43±0.25	0.28±0.16	0.52±0.27	0.23±0.14
T6 - 1500	0.63±0.35	0.65±0.87	0.80±0.51	0.33±0.19
T7 - 2000	0.80±0.46	0.58±0.39	1.08±0.90	0.48±0.29
T8 - 2500	1.00±0.46	0.84±0.79	1.51±1.36	0.54±0.29
T9 - 3000	1.20±0.46	1.12±1.09	1.92±1.58	0.64±0.39

Appendix 2.19. The cadmium (Cd^{2+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11 and 2011/12 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12
Winery wastewater	0.001±0.001	0.001±0.001	0.003±0.003
T1 - River water	0.001±0.001	0.000±0.001	0.002±0.002
T2 - 100	0.001±0.001	0.001±0.001	0.001±0.001
T3 - 250	0.001±0.001	0.000±0.001	0.001±0.001
T4 - 500	0.001±0.001	0.000±0.000	0.001±0.001
T5 - 1000	0.001±0.001	0.001±0.001	0.002±0.001
T6 - 1500	0.001±0.001	0.001±0.002	0.003±0.003
T7 - 2000	0.000±0.001	0.001±0.002	0.001±0.001
T8 - 2500	0.001±0.001	0.001±0.002	0.001±0.001
T9 - 3000	0.001±0.001	0.001±0.001	0.001±0.001

Appendix 2.20. The chromium (Cr^{2+}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11 and 2011/12 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12
Winery wastewater	0.015±0.009	0.038±0.049	0.009±0.009
T1 - River water	0.012±0.007	0.023±0.018	0.004±0.007
T2 - 100	0.010±0.009	0.032±0.025	0.003±0.005
T3 - 250	0.013±0.006	0.024±0.026	0.004±0.004
T4 - 500	0.012±0.005	0.019±0.014	0.003±0.004
T5 - 1000	0.018±0.002	0.023±0.014	0.003±0.003
T6 - 1500	0.013±0.007	0.025±0.020	0.004±0.004
T7 - 2000	0.015±0.009	0.016±0.010	0.005±0.005
T8 - 2500	0.017±0.010	0.021±0.018	0.005±0.004
T9 - 3000	0.015±0.006	0.019±0.013	0.003±0.006

Appendix 2.21. The arsenic (As^{3-}) concentration in mg/L in winery wastewater, river water and diluted winery wastewater used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11 and 2011/12 seasons.

Treatment no. & target COD (mg/L)	2009/10	2010/11	2011/12
Winery wastewater	0.007±0.008	0.001±0.001	0.006±0.009
T1 - River water	0.003±0.002	0.001±0.001	0.003±0.006
T2 - 100	0.002±0.004	0.000±0.001	0.003±0.006
T3 - 250	0.000±0.000	0.000±0.000	0.001±0.002
T4 - 500	0.002±0.004	0.002±0.003	0.006±0.010
T5 - 1000	0.003±0.005	0.000±0.000	0.003±0.005
T6 - 1500	0.003±0.003	0.000±0.001	0.000±0.001
T7 - 2000	0.005±0.004	0.000±0.000	0.003±0.004
T8 - 2500	0.001±0.001	0.001±0.001	0.001±0.002
T9 - 3000	0.007±0.006	0.001±0.001	0.000±0.001

APPENDIX 3 - Amounts of elements applied

Appendix 3.1. The calculated amounts of ammonium nitrogen (NH_4^+-N) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, NH_4^+-N was not determined.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest			Total		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	0.96	0.44	0.48	0.53	0.35	0.90	1.49	0.79	1.38
T2 - 100	0.57	0.38	0.38	0.37	0.14	0.50	0.94	0.53	0.88
T3 - 250	0.70	0.37	0.21	0.34	0.14	0.45	1.04	0.51	0.66
T4 - 500	0.61	0.72	0.21	0.34	0.41	0.48	0.96	1.14	0.69
T5 - 1000	0.69	0.61	0.19	0.37	0.74	1.11	1.06	1.35	1.29
T6 - 1500	0.72	1.05	0.38	1.24	1.22	1.70	1.95	2.27	2.08
T7 - 2000	0.78	1.52	0.65	0.49	1.58	2.43	1.27	3.11	3.08
T8 - 2500	0.86	1.67	0.30	0.48	1.99	3.44	1.35	3.66	3.74
T9 - 3000	0.77	1.68	0.46	0.77	2.21	1.49	1.54	3.89	1.95

Appendix 3.2. The calculated amounts of nitrate nitrogen (NO_3^--N) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, NO_3^--N was not determined.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest			Total		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	2.65	2.52	4.64	1.16	0.01	0.43	3.81	2.53	5.07
T2 - 100	1.58	0.93	1.76	1.09	0.01	0.44	2.67	0.94	2.02
T3 - 250	1.64	0.73	1.25	0.60	0.01	0.25	2.24	0.74	1.50
T4 - 500	1.24	0.88	1.33	0.43	0.01	0.40	1.67	0.89	1.72
T5 - 1000	1.51	0.86	1.37	0.44	0.01	0.83	1.95	0.87	2.20
T6 - 1500	1.24	0.81	1.54	1.10	0.01	1.35	2.34	0.82	2.89
T7 - 2000	2.67	1.14	2.39	0.82	0.01	1.49	3.49	1.15	3.88
T8 - 2500	2.90	2.64	3.62	0.45	0.01	1.86	3.35	2.65	5.48
T9 - 3000	3.20	2.51	4.23	0.68	0.01	2.02	3.89	2.52	6.25

Appendix 3.3. The calculated amounts of total nitrogen (Total-N) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, total-N was not determined.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest			Total		
	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13	2010/11	2011/12	2012/13
T1 - River water	3.61	2.96	5.12	1.69	0.36	1.33	5.30	3.32	6.45
T2 - 100	2.15	1.31	2.14	1.46	0.15	0.94	3.61	1.46	3.08
T3 - 250	2.34	1.10	1.46	0.94	0.15	0.70	3.28	1.25	2.16
T4 - 500	1.85	1.60	1.54	0.77	0.42	0.88	2.62	2.03	2.42
T5 - 1000	2.20	1.47	1.56	0.81	0.75	1.94	3.01	2.21	3.50
T6 - 1500	1.96	1.86	1.92	2.34	1.23	3.05	4.30	3.09	4.97
T7 - 2000	3.45	2.66	3.04	1.31	1.59	3.92	4.76	4.25	6.95
T8 - 2500	3.76	4.31	3.92	0.93	2.00	5.30	4.69	6.31	9.22
T9 - 3000	3.97	4.19	4.69	1.45	2.22	3.51	5.42	6.41	8.20

Appendix 3.4. The calculated amounts of phosphorus (P) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	0.00	0.06	0.28	0.00	0.09	0.01	0.42	0.00	0.09	0.07	0.70
T2 - 100	0.04	0.16	0.29	0.00	0.04	0.06	0.37	0.00	0.08	0.22	0.66
T3 - 250	0.22	0.49	0.29	0.00	0.17	0.15	0.52	0.00	0.39	0.64	0.81
T4 - 500	0.72	1.04	0.47	0.25	0.47	0.33	0.95	0.25	1.19	1.37	1.42
T5 - 1000	2.11	2.30	1.17	0.81	1.07	0.66	1.69	0.81	3.18	2.96	2.86
T6 - 1500	3.67	3.51	1.35	1.49	1.68	0.92	2.59	1.49	5.35	4.43	3.94
T7 - 2000	6.01	3.87	1.96	2.04	2.45	1.37	3.26	2.04	8.46	5.24	5.22
T8 - 2500	7.43	5.08	2.20	3.18	2.95	1.75	4.04	3.18	10.38	6.83	6.24
T9 - 3000	9.40	6.22	2.88	4.04	4.06	1.80	4.64	4.04	13.46	8.02	7.52

Appendix 3.5. The calculated amounts of potassium (K^+) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	5.99	6.37	2.29	4.93	1.47	0.19	3.42	4.93	7.46	6.56	5.71
T2 - 100	12.14	10.32	3.36	12.89	1.38	0.69	6.48	12.89	13.52	11.01	9.84
T3 - 250	16.34	19.81	9.59	23.42	8.80	1.77	12.58	23.42	25.14	21.58	22.17
T4 - 500	24.86	32.15	18.50	43.10	19.00	3.71	22.55	43.10	43.86	35.86	41.05
T5 - 1000	41.05	57.89	29.81	92.84	37.99	7.24	40.84	92.84	79.04	65.13	70.65
T6 - 1500	61.96	77.47	33.84	132.86	50.87	9.85	54.42	132.86	112.83	87.32	88.26
T7 - 2000	78.48	92.37	46.66	166.97	70.31	11.58	63.87	166.97	148.79	103.95	110.53
T8 - 2500	92.61	116.78	57.84	200.08	80.80	17.68	77.53	200.08	173.41	134.46	135.37
T9 - 3000	106.18	140.28	67.56	243.63	108.66	17.39	91.94	243.63	214.84	157.67	159.50

Appendix 3.6. The calculated amounts of calcium (Ca^{2+}) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	19.01	14.67	12.37	9.76	11.01	0.27	6.70	9.76	30.02	14.94	19.07
T2 - 100	20.62	15.05	13.57	9.55	10.12	0.38	7.27	9.55	30.74	15.43	20.84
T3 - 250	20.30	16.90	14.26	10.29	10.49	0.61	7.85	10.29	30.79	17.51	22.11
T4 - 500	21.65	18.98	15.30	11.87	11.25	1.07	8.59	11.87	32.90	20.05	23.89
T5 - 1000	22.68	23.06	16.26	14.91	12.24	1.89	10.27	14.91	34.92	24.95	26.53
T6 - 1500	24.31	26.04	16.90	17.58	14.20	2.53	11.19	17.58	38.51	28.57	28.09
T7 - 2000	25.64	28.75	19.22	19.30	14.30	3.74	12.00	19.30	39.94	32.49	31.22
T8 - 2500	25.77	31.84	20.32	21.74	15.02	4.61	13.03	21.74	40.79	36.45	33.35
T9 - 3000	27.50	35.88	21.03	23.89	15.63	4.81	13.99	23.89	43.13	40.69	35.02

Appendix 3.7. The calculated amounts of magnesium (Mg^{2+}) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	12.25	9.51	5.69	6.44	6.83	0.15	4.13	6.44	19.08	9.66	9.82
T2 - 100	13.28	9.67	6.11	6.05	6.31	0.19	4.50	6.05	19.59	9.86	10.61
T3 - 250	13.32	10.19	6.18	6.22	6.51	0.21	4.84	6.22	19.83	10.40	11.02
T4 - 500	13.46	10.32	6.39	6.55	6.75	0.25	5.14	6.55	20.21	10.57	11.53
T5 - 1000	13.59	11.00	6.67	6.92	7.12	0.34	6.03	6.92	20.71	11.34	12.70
T6 - 1500	14.16	11.79	6.84	7.31	7.48	0.40	6.43	7.31	21.64	12.19	13.27
T7 - 2000	14.51	11.92	7.39	7.28	7.72	0.52	6.70	7.28	22.23	12.44	14.09
T8 - 2500	14.17	12.19	7.62	8.11	7.88	0.63	7.30	8.11	22.05	12.82	14.92
T9 - 3000	14.82	13.02	7.62	8.13	8.09	0.65	7.89	8.13	22.92	13.67	15.51

Appendix 3.8. The calculated amounts of sodium (Na^+) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	31.02	31.90	17.98	18.35	19.34	0.90	11.54	18.35	50.36	32.80	29.52
T2 - 100	35.44	34.25	18.89	19.04	18.15	1.25	12.56	19.04	53.59	35.50	31.45
T3 - 250	38.16	35.67	21.07	21.36	21.67	1.86	14.74	21.36	59.83	37.53	35.81
T4 - 500	48.47	39.74	22.32	25.24	24.58	2.68	17.97	25.24	73.05	42.42	40.29
T5 - 1000	49.72	44.82	26.41	34.99	31.45	4.64	24.30	34.99	81.17	49.46	50.71
T6 - 1500	61.23	48.75	27.65	41.90	36.07	6.15	28.31	41.90	97.30	54.90	55.96
T7 - 2000	64.42	52.74	32.40	47.83	40.23	8.71	31.84	47.83	104.65	61.45	64.24
T8 - 2500	66.99	58.05	35.68	56.85	43.13	11.02	36.01	56.85	110.12	69.07	71.69
T9 - 3000	70.72	62.78	40.27	64.51	51.61	11.19	41.30	64.51	122.33	73.97	81.57

Appendix 3.9. The calculated amounts of chloride (Cl^-) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	73.31	64.38	48.94	40.81	42.93	3.01	31.49	40.81	116.24	67.39	80.43
T2 - 100	78.15	66.88	48.43	37.49	37.98	3.33	27.54	37.49	116.13	70.21	75.97
T3 - 250	78.20	65.08	47.11	35.95	39.99	3.42	31.07	35.95	118.19	68.50	78.18
T4 - 500	76.81	63.97	50.73	34.92	38.36	3.52	29.99	34.92	115.17	67.49	80.72
T5 - 1000	80.36	63.26	53.55	37.95	37.02	3.19	30.31	37.95	117.38	66.45	83.86
T6 - 1500	79.79	65.81	55.96	40.56	42.76	2.79	34.09	40.56	122.55	68.60	90.05
T7 - 2000	80.62	60.37	51.41	38.45	43.51	2.86	31.02	38.45	124.13	63.23	82.43
T8 - 2500	79.67	75.12	49.23	36.27	41.96	3.09	30.29	36.27	121.63	78.21	79.52
T9 - 3000	82.92	75.46	52.29	38.26	44.83	3.35	50.20	38.26	127.75	78.81	102.49

Appendix 3.10. The calculated amounts of bicarbonate (HCO_3^-) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	52.13	33.70	11.57	21.72	23.97	2.38	0.29	21.72	76.10	36.08	11.86
T2 - 100	54.72	38.06	10.37	47.30	25.11	2.89	6.75	47.30	79.83	40.95	17.12
T3 - 250	57.35	46.41	6.44	72.44	35.77	7.76	13.71	72.44	93.12	54.17	20.15
T4 - 500	70.72	57.35	4.62	119.65	42.93	13.35	16.17	119.65	113.65	70.70	20.79
T5 - 1000	85.92	79.30	5.06	208.94	58.34	21.40	22.18	208.94	144.26	100.70	27.24
T6 - 1500	172.40	109.45	2.12	281.46	80.83	27.92	38.95	281.46	253.23	137.37	41.07
T7 - 2000	139.36	115.08	2.22	368.33	101.07	45.81	47.27	368.33	240.43	160.89	49.49
T8 - 2500	150.92	124.84	2.19	461.73	116.92	52.75	59.66	461.73	267.84	177.59	61.85
T9 - 3000	198.62	175.18	4.75	559.12	156.79	52.09	65.41	559.12	355.41	227.27	70.16

Appendix 3.11. The calculated amounts of sulphate (SO_4^{2-}) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	69.53	50.40	45.89	38.28	38.93	4.72	17.58	38.28	108.46	55.12	63.47
T2 - 100	72.81	49.15	41.37	34.27	37.29	2.86	19.22	34.27	110.10	52.01	60.59
T3 - 250	75.49	51.10	41.38	30.19	37.35	2.75	20.24	30.19	112.84	53.85	61.62
T4 - 500	83.70	49.84	60.87	37.01	37.53	3.87	21.17	37.01	121.23	53.71	82.04
T5 - 1000	75.51	96.65	48.02	47.10	35.88	6.15	22.70	47.10	111.39	102.80	70.72
T6 - 1500	66.79	61.78	73.57	51.03	38.29	15.64	24.42	51.03	105.08	77.42	97.99
T7 - 2000	76.27	99.11	93.21	37.35	37.71	43.25	24.06	37.35	113.98	142.36	117.27
T8 - 2500	64.85	76.07	84.38	119.47	38.17	72.05	27.65	119.47	103.02	148.12	112.03
T9 - 3000	99.47	55.07	82.50	89.88	39.03	74.43	28.30	89.88	138.50	129.50	110.80

Appendix 3.12. The calculated amounts of boron (B^{3+}) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	0.05	0.02	0.03	0.12	0.03	0.01	0.01	0.12	0.08	0.03	0.04
T2 - 100	0.07	0.02	0.03	0.06	0.03	0.01	0.01	0.06	0.10	0.03	0.04
T3 - 250	0.09	0.03	0.03	0.07	0.05	0.03	0.01	0.07	0.14	0.06	0.04
T4 - 500	0.11	0.05	0.05	0.10	0.07	0.05	0.01	0.10	0.18	0.10	0.06
T5 - 1000	0.16	0.08	0.07	0.18	0.12	0.09	0.02	0.18	0.28	0.17	0.09
T6 - 1500	0.22	0.11	0.09	0.22	0.18	0.13	0.03	0.22	0.40	0.24	0.12
T7 - 2000	0.27	0.13	0.12	0.26	0.22	0.20	0.05	0.26	0.49	0.33	0.17
T8 - 2500	0.32	0.16	0.14	0.35	0.26	0.25	0.07	0.35	0.58	0.41	0.21
T9 - 3000	0.37	0.19	0.15	0.44	0.33	0.26	0.08	0.44	0.70	0.45	0.23

Appendix 3.13. The calculated amounts of iron (Fe^{2+}) applied *via* river water and diluted winery wastewater (kg/ha) used for irrigation of Cabernet Sauvignon/99R during the 2009/10, 2010/11, 2011/12 and 2012/13 seasons. In 2009/10, irrigations were only applied in the post-harvest period.

Treatment no. & target COD (mg/L)	Pre-harvest			Post-harvest				Total			
	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13	2009/10	2010/11	2011/12	2012/13
T1 - River water	0.04	0.18	0.02	0.06	1.25	0.03	0.05	0.06	1.29	0.21	0.07
T2 - 100	0.08	0.25	0.04	0.10	2.05	0.04	0.11	0.10	2.13	0.29	0.15
T3 - 250	0.10	0.37	0.14	0.14	1.78	0.05	0.15	0.14	1.88	0.42	0.29
T4 - 500	0.21	0.48	0.25	0.31	1.74	0.07	0.18	0.31	1.95	0.55	0.43
T5 - 1000	0.37	0.75	0.40	0.55	0.80	0.13	0.16	0.55	1.17	0.88	0.56
T6 - 1500	0.53	1.04	0.56	0.81	1.71	0.22	0.27	0.81	2.24	1.26	0.83
T7 - 2000	0.73	1.19	0.85	1.00	1.22	0.34	0.32	1.00	1.95	1.53	1.17
T8 - 2500	0.94	1.56	0.93	1.24	1.70	0.50	0.36	1.24	2.64	2.06	1.29
T9 - 3000	1.11	2.10	1.15	1.49	2.18	0.51	0.44	1.49	3.29	2.61	1.59