EVALUATION OF THE STRUCTURAL AND FUNCTIONAL COMPOSITION OF SOUTH AFRICAN TRITICALE CULTIVARS (X *TRITICOSECALE* WITTMACK)

FRANCES DU PISANI

Thesis presented in partial fulfilment of the requirements for the degree of

MASTER OF SCIENCE IN FOOD SCIENCE

Department of Food Science
Faculty of AgriSciences
Stellenbosch University

Study leader: Dr Marena Manley
Co-study leaders: Dr Glen Fox
Ms Nina Muller

March 2009

Declaration

By submitting this thesis electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the owner of the copyright 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.

Date: 25 February 2009

Abstract

Triticale (X *Triticosecale* Whittmack), a cross between durum wheat (*Triticum* sp.) and rye, is a crop with an increasing agronomic and economic potential. Though studies on the functional and compositional quality of triticale have been conducted in other parts of the world, little is known regarding cultivars developed in South Africa in terms of these aspects. South African triticale cultivars from various localities in the Western Cape, obtained for two subsequent harvest seasons, were analysed for moisture, protein and ash contents, as well as falling number (an indication of α-amylase activity), hardness (particle size index), 1000-kernel mass and baking potential (SDS sedimentation). These triticale samples were derived from a breeding program that was not focused on baking quality. The results obtained were found to compare well with those reported on in previous studies.

Significant differences were observed between both cultivars and localities within years, illustrating the effect of genetic as well as environmental factors. Significant differences were also observed between localities when comparing the two harvest seasons, whereas differences between the cultivars for the two seasons were in most cases not significant; illustrating the effect of environment. Interactions between cultivars and localities were found to be significant for all parameters, and trends were observed between protein content and both particle size index (PSI) (negative) as well as SDS sedimentation (positive) results for both years.

Near infrared (NIR) spectroscopy is a rapid method of analysis and is widely used for the quality evaluation of wheat. Limited research has been reported on calibration models for the quality evaluation of triticale, and thus NIR spectroscopy was applied to develop models for the prediction of moisture, protein and ash contents, as well as hardness and baking potential for South African cultivars. Spectra were collected in diffuse reflectance mode and partial least squares (PLS) models developed for both triticale flour and wholegrain using two different instruments (Büchi NIRFlex N-500 and Bruker MPA Fourier transform NIR spectrophometers) and software packages (The Unscrambler and OPUS). Full cross-validations were performed, after which the best prediction models obtained (R^2 > 0.66) were validated using an independent test set (n = 50). The best prediction results were obtained with flour for moisture (Bruker: *SE*P = 0.08%; R^2 = 0.95; *RPD* = 4.65) and protein (Büchi: *SEP* = 0.44%; R^2 = 0.96; *RPD* = 5.23 and Bruker: *SE*P = 0.32%; R^2 = 0.96; *RPD* = 4.88). For whole grain, acceptable results were obtained for protein (Büchi: *SEP* = 0.55%; R^2 = 0.94; *RPD* = 4.18 and Bruker: *SE*P = 0.70%; R^2 = 0.90; *RPD* = 3.23). Though

results for ash content, PSI and SDS sedimentation prediction did not yield models that can be applied as yet, these models form a good basis for further calibration model development and possibly use in early generation screening.

The current limited ranges could be expanded by adding samples from subsequent harvest seasons. By adding more data, a better quality profile for South African triticale can be obtained, which will facilitate better interpretation in terms of the effect of genetic and environmental factors. It would also enable the development of improved NIR prediction models.

Uittreksel

Korog (X *Triticosecale* Whittmack), ‘n kruising tussen durumkoring (*Triticum* sp.) en rog (*Secale* sp.), is ‘n gewas met toenemende agronomiese en ekonomiese potensiaal. Alhoewel studies aangaande die samestelling en funksionele kwaliteit van korog al in ander dele van die wêreld uitgevoer is, is daar min inligting beskikbaar in dié verband oor kultivars wat in Suid-Afrika ontwikkel is. Suid-Afrikaanse korog kultivars, vanaf verskeie lokaliteite in die Wes-Kaap, verkry vir twee opeenvolgende oesseisoene, is in terme van vog-, proteïen- en asinhoud, asook valgetal (‘n aanduiding van α-amilase), hardheid (partikelgrootte indeks), 1000-korrel massa en bakpotensiaal (SDS sedimentatsie) geanaliseer. Hierdie korog kultivars is verkry vanaf ‘n teelprogram wat nie gefokus was op bakkwaliteit nie. Daar is gevind dat die resultate wat verkry is goed vergelyk met dit wat in vorige studies verkry is.

Betekenisvolle verskille is gevind tussen beide kultivars en lokaliteite binne oesjare, wat die effek van genetiese- asook omgewingsfaktore illustreer. Daar is ook betekenisvolle verskille gevind tussen lokaliteite oor die twee oesseisoene, terwyl verskille tussen kultivars oor die twee seisoene meestal nie betekenisvol was nie; wat weereens die effek van omgewing illustreer. Interaksies tussen kultivars en lokaliteite was in alle gevalle betekenisvol. Verder is ’n verwantskap tussen die proteïeninhoud en beide partikelgrootte indeks (PSI) (negatief) en SDS sedimentasie (positief) resultate vir beide jare waargeneem.

Naby infrarooi (NIR) spektroskopie is ‘n vinnige ontledingsmetode wat algemeen gebruik word vir die evaluasie van koring. Beperkte navorsing is al gerapporteer aangaande die ontwikkeling van kalibrasiemodelle vir die kwaliteitsevaluering van korog, en NIR spektroskopie is dus aangewend in hierdie studie om modelle te ontwikkel vir die voorspelling van vog-, proteïen-, en asinhoud, asook die hardheid en bakpotensiaal van Suid-Afrikaanse korog kultivars. Spektra is verkry in diffuse refleksie en parsiële kleinste kwadrate (PLS) modelle is ontwikkel vir beide meel en heelgraan monsters deur gebruik te maak van twee verskillende instrumente (die Büchi NIRFlex N-500 en die Bruker MPA Fourier transformasie NIR spektrofotometers) en sagteware pakette (The Unscrambler en OPUS). Volle kruis-validasie is uitgevoer, waarna die beste voorspellingsmodelle ($R^2 > 0.66$) verkry deur middel van ‘n onafhanklike toetsstel (n = 50) gevalideer is. Die beste resultate is verkry met meel vir voginhoud (Bruker: *SE*P = 0.08%; R^2 = 0.95; *RPD* = 4.65) en proteïeninhoud (Büchi: *SEP* = 0.44%; R^2 = 0.96; *RPD* = 5.23 en Bruker: *SE*P = 0.32%; R^2 = 0.96; *RPD* = 4.88). Met heelgraan is aanvaarbare resultate verkry vir

proteïeninhoud (Büchi: *SEP* = 0.55%; R^2 = 0.94; *RPD* = 4.18 en Bruker: *SEP* = 0.70%; R^2 = 0.90; *RPD* = 3.23). Alhoewel resultate vir die bepaling van asinhoud, PSI en SDS sedimentasie nie modelle gelewer het wat reeds gebruik kan word nie, vorm hierdie modelle ‘n goeie basis vir die ontwikkeling van verdere kalibrasiemodelle wat moontlik gebruik kan word vir rofweg bepaling van vroeë generasies.

Die huidige beperkte reikwydte kan uitgebrei word deur monsters van toekomstige oesseisoene by te voeg. Deur nog data by te voeg, sal ‘n beter kwaliteitsprofiel vir Suid-Afrikaanse korog verkry kan word, wat ‘n beter interpretasie van die effek van genetiese en omgewingsfaktore sal toelaat. Dit sal ook die ontwikkeling van verbeterde NIR modelle moontlik maak.

Acknowledgements

I recognise the following persons and institutions for their contribution to the successful completion of this thesis:

Dr Marena Manley, my study leader, for her amazing guidance, advice and patience, and who made it possible to attend the 14th World Congress of Food Science and Technology in Shanghai, China. Her knowledge of her field is astounding and inspiring, and she is willing to sacrificially do above and beyond what is expected of a study leader to assist her students;

Dr Glen Fox, my co-study leader, for all his excellent advice, and his encouragement. He was always available and willing to help, and taught me almost all I know about NIR spectroscopy;

Ms Nina Muller, my co-study leader, for her advice and support;

Prof Martin Kidd, Centre for Statistical Consultation, Stellenbosch University, for his advice and valuable statistical analyses, and endless patience with me;

Pioneer, Sasko Milling and Baking, Paarl, for the use of their facilities, equipment and staff, especially to Carien Roets and Divan September for their time and help;

The Winter Cereal Trust for a bursary;

The National Research Foundation (NRF) for a bursary and for funding of this project (FA2006031500013);

The PA and Alize Malan Trust for funding of this project;

The Department of Genetics, Stellenbosch University, for kindly supplying all triticale samples used in this study, and specifically Mr Herman Roux, Mr Willem Botes and Mr Henzel Saul for help and advice;

The staff at the Department of Food Science, for their willingness and availability and help and assist, especially Anchen Lombard, Petro du Buisson, Natasja Brown, Daleen du Preez and Eben Brooks;

My fellow NIRDS, Paul, Gerida and Glen, for their friendship, support, and help during crisis management times, without which this thesis would really not have been possible. The lab would be a dull place without you guys;

My close friends for their support, encouragement, patience, prayers and love, especially Aimee, Minke, Mariette and Riaan. I would not have survived the last year without you and I'm very thankful to have you as friends;

My boyfriend Craig for his love, understanding, encouragement, motivation, help, patience and sense of humour during the last year. You made everything seem lighter, and you've come to mean more to me in these lasts months than I can explain. Also sincere thanks to your family;

My family, especially my parents, brother and grandmother for their love, support and encouragement; I am convinced your prayers are responsible for the successful completion of this thesis; and

ABOVE all, I thank my Saviour for walking with me in this time, without Him absolutely nothing would be possible. All glory goes to You.

Contents

	Declaration	ii
	Abstract	iii
	Uittreksel	v
	Acknowledgements	vii
Chapter 1:	Introduction	1
Chapter 2:	Literature review	7
Chapter 3:	Evaluation of the compositional and functional quality of South African triticale (X *Triticosecale* Wittmack) cultivars using conventional methods	30
Chapter 4:	The development of near infrared (NIR) spectroscopy calibration models for the prediction of the moisture, protein and ash content, as well as hardness and baking potential of South African triticale (X *Triticosecale* Whittmack) cultivars	63
Chapter 5:	General discussion and conclusion	89
Appendices:		96
	Appendix 1	97
	Appendix 2	104
	Appendix 3	128
	Appendix 4	139

Language and style used in this thesis are in accordance with the requirements of the *International Journal of Food Science and Technology.* This thesis represents a compilation of manuscripts where each chapter is an individual entity and some repetition between chapters has, therefore, been unavoidable.

CHAPTER 1

Introduction

CHAPTER 1

INTRODUCTION

Triticale (X *Triticosecale* Whittmack), the first cereal crop to be produced by humans by a deliberate action, is a cross between wheat (*Triticum* sp.) and rye (*Secale* sp.) and was attempted for the first time in 1875 (Stallknecht *et al*., 1996; Ammar *et al*., 2004; Oettler, 2005). The aim was to obtain a crop with the beneficial properties of both parent species, including wheat's potential for use in various food products, with rye's hardiness, disease resistance and adaptability to adverse environmental conditions. Originally, this aim proved to be elusive, as triticale had very poor properties relating to its use for baking purposes. This stems from its poor gluten content as well as high α-amylase activity (Stallknecht *et al.,* 1996; Peña, 2004). However, once serious research on this crop began in the 1960's with the establishment of various dedicated research programmes, triticale soon started showing more promise (Kent & Evers, 1994; Ammar *et al*., 2004).

In modern times, it has been reported that triticale is cultivated in more than 30 countries worldwide (Kent & Evers, 1994; Mergoum *et al*., 2004) on around 3.7 million ha in total, yielding more than 12 million tonnes a year (FAO, 2007). While the production of cereals such as rye, oat, sorghum and millet has been decreasing during the last 15 years, the production of triticale increases annually (Salmon *et al*., 2004). This worldwide adoption of triticale can be attributed to its ability to produce a higher yield and biomass than other cereals over a range of soil types as well as under adverse environmental conditions (Mergoum *et al*., 2004). Triticale furthermore shows resistance to many of the pests and diseases affecting wheat (Mergoum *et al*., 2004). Due to this characteristic of triticale, it poses the possibility of expanding agricultural activity into unfavourable areas thereby increasing productivity. In the current unfavourable economic climate, this can be of great value, especially in third world countries facing impending food shortages. These conditions, together with the fact that triticale production is increasing steadily worldwide, seems to indicate that triticale could soon become important in serving as a source of food to the rapidly growing population of the earth (Naeem *et al*., 2002).

Apart from its potential as a source of food to humans, triticale is widely used as animal feed (Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al.*, 2004), and it can be used in the form of grain, forage, silage, hay or straw (Myer & Lozano del Rio, 2004). This is due to its high biomass yield which has been shown to be equal to or higher than that of other cereal grains (Delogu *et al*., 2002). Furthermore it has a good nutritional composition which

compares well with that of wheat, and it is generally a good source of vitamins, minerals and essential amino acids (Lorenz *et al*., 1974). It is high in starch, lipids, dietary fibre and mineral ash, and its protein content is comparable to that of wheat (Kent & Evers, 1994; Stallknecht *et al*., 1996; Dyson, 2006). Furthermore, triticale has a high lysine content, which is significant due to the fact that lysine is usually the limiting amino acid in cereal grains (Kies & Fox, 1970; Villegas *et al*., 1970).

Modern cultivars of triticale have also been found to hold potential as a very competitive raw material for bio-ethanol production (Eudes, 2006). It is more vigorous and adaptable than either of its parent species, as well as oats and barley. Importantly, it also produces a greater biomass when receiving the same input as its parent species, and the high starch content observed in triticale makes it very apt as raw material for bio-ethanol production (Eudes, 2006).

Triticale is thus a crop with great potential, and numerous breeding initiatives around the globe are breeding for improved cultivars. The evaluation of the compositional and functional quality of triticale in order to obtain a profile for cultivars is thus of importance, especially during the breeding of early generations of new lines (Osborne, 2000). A comprehensive study regarding the compositional and functional quality of South African triticale cultivars has not been carried out to date.

During the early stages of the breeding of new cultivars, methods of evaluation are desired that are fast and accurate, and do not require large amounts of sample, as limited sample is usually available for evaluation during this stage of the development of a cultivar. Conventional analysis methods often do not meet these requirements, sometimes resulting in difficulty with the initial evaluation of new cultivars. Near infrared (NIR) spectroscopy, a technology that has been used increasingly in the grain industry since the 1970's (Butler, 1983), is perfectly suited for the analysis of grains both during the breeding of new cultivars and during commercial production (Osborne, 2000).

NIR spectroscopy poses the advantages of being a fast, cheap, non-invasive, non-destructive method of analysis that requires minimal sample preparation and small sample sizes (Butler, 1983; Osborne, 2000; Pasquini, 2003). It is a type of vibrational spectroscopy which operates in the wavelength range from 750 to 2500 nm (Butler, 1983; Pasquini, 2003). The application of NIR spectroscopy is based on the empirical relationship between reference analytical data (conventional analytical methods) and spectral data (NIR methods) to acquire quantitative and/or qualitative information obtained from the interaction between the near infrared electromagnetic waves and the constituents of the sample (Osborne, 1983; Pasquini, 2003).

NIR spectroscopy is currently widely used for the quality evaluation of wheat, and has been used to test for various quality parameters, such as protein (Osborne & Fearn, 1983; Shenk *et al*., 1985; Delwiche, 1998; Manley *et al*., 2002) and moisture contents (Osborne & Fearn, 1983; Law & Tkachuk, 1977; Osborne, 1987; Manley *et al*., 2002), as well as for hardness determination (Osborne & Fearn, 1983; Williams & Sobering, 1986; Norris *et* al., 1989; Osborne, 1991; Manley *et al*., 2002) and ash content (Miralbés, 2004). Limited information is, however, available in literature regarding the use of NIR spectroscopy in the evaluation of triticale quality, and no information has been found on South African cultivars. Studies performed by Igne (2007 a; b) resulted in good prediction models for protein and moisture content, while a study by Viljoen *et al*. (2005) obtained acceptable models for the prediction of moisture, protein and ash content for a sample set containing four South African winter cereals, i.e. oats, barley, wheat and triticale, .

The objectives of this study were therefore:

- to determine the compositional and functional quality of South African triticale cultivars from different localities and two harvest seasons in terms of moisture, protein and ash contents as well as kernel hardness (particle size index), 1000-kernel mass and baking potential (SDS sedimentation); and
- to develop NIR spectroscopy calibrations for the prediction of moisture, protein and ash contents, particle size index (PSI) values (kernel hardness) and sodium dodecyl sulphate (SDS) sedimentation values of these triticale cultivars using two different NIR instruments and software packages.

References

Ammar, K., Mergoum, M., Rajaram, S. (2004). The history and evolution of triticale. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp 1-9. Rome: Food and Agriculture Organisation of the United Nations.

Butler, L.A. (1983). The history and background of NIR. *Cereal Foods World*, **28**(4), 238-240.

Delogu, G., Faccini, N., Faccioli, P., Reggiani, F., Lendini, M., Berardo, N. & Odoardi, M. (2002). Dry matter yield and quality evaluation at two phenological stages of forage triticale grown in the Po valley and Sardinia, Italy. *Field Crops Research*, **74**, 207-215.

Delwiche, S.R. (1998). Protein content of single kernels of wheat by near-infrared reflectance spectroscopy. *Journal of Cereal Science*, **27**, 241-254.

Dyson, C. (2006). Triticale grain for feed - Nutritional information. Alberta Government Agriculture and Food [WWW document]. URL http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/fcd10575. 12 May 2007.

Eudes, F. (2006). Canadian triticale biorefinery initiative. In: *Proceedings of the 6th International Triticale Symposium*. Pp. 85-88. Stellenbosch, South Africa.

FAO (2007). FOASTAT, FAO statistical databases – agriculture [WWW document]. URL http://faostat.fao.org/site/567/default.aspx#ancor. 1 December 2008.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007a). Triticale moisture and protein content prediction by near-infrared spectroscopy (NIRS). *Cereal Chemistry*, **84**(4), 328-330.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007b). Influence of yearly variability of agricultural products on calibration process: a triticale example. *Cereal Chemistry*, **84**(6), 576-581.

Kent, N.L. & Evers, A.D. (1994). *Kent's Technology of Cereals: An Introduction for Students of Food Science and Agriculture*. Pp. 17-18, 50, 96-97, 157. New York, USA: Elsevier Science Ltd.

Kies, C. & Fox, H.M. (1970). Protein nutritive value of wheat and triticale grain for humans, studied at two levels of protein intake. *Cereal Chemistry*, **47**, 671-678.

Law, D.P. & Tkachuk, R. (1977). Determination of moisture content in wheat by near infrared diffuse reflectance spectrophotometry. *Cereal Chemistry*, **54**(4), 874-881.

Lorenz, K., Reuter, F.W. & Sizer, C. (1974). The mineral composition of triticales and triticale milling fractions by X-ray fluorescence and atomic absorption. *Cereal Chemistry*, **51**, 534-541.

Manley, M., Van Zyl, L. & Osborne, B.G. (2002). Using Fourier transform near infrared spectroscopy in determining kernel hardness, protein content and moisture content of whole wheat flour. *Journal of Near Infrared Spectroscopy*, **10**, 71-76.

Mergoum, M., Pfeiffer, W.H., Peña, R.J., Ammar, K. & Rajaram, S., 2004. Triticale crop improvement: the CIMMYT programme. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 11-26. Rome: Food and Agriculture Organisation of the United Nations.

Miralbés, C. (2004). Quality control in the milling industry using near infrared transmittance spectroscopy. *Food Chemistry*, **88**, 621–628.

Myer, R. & Lozano del Rio, A.J. (2004). Triticale as animal feed. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 49-58. Rome: Food and Agriculture Organisation of the United Nations.

Naeem, H.A., Darvey, N.L., Gras, P.W. & MacRitchie, F. (2002). Mixing properties, baking potential, and functionality changes in storage proteins during dough development of triticale-wheat flour blends. *Cereal Chemistry*, **79**(3), 332-339.

Norris, K.H., Hruschka, W.R., Bean, M.M. & Slaughter, D.C. (1989). A definition of wheat hardness using near infrared reflectance spectroscopy. *Cereal Foods World*, **34**(9), 696-705.

Oettler, G. (2005). The fortune of a botanical curiosity – Triticale: past, present and future. *Journal of Agricultural Science*, **143**, 329-346.

Osborne, B.G. (1987). Determination of moisture in white flour, ground wheat and whole wheat by near infrared reflectance using a single calibration. *Journal of the Science of Food and Agriculture*, **38**, 341-436.

Osborne, B.G. (1991). Measurement of the hardness of wheat endosperm by near-infrared spectroscopy. *Postharvest News and Information*, **2**, 331-334.

Osborne, B.G. (2000). Recent developments in NIR analysis of grains and grain products. *Cereal Foods World*, **45**(1), 11-15.

Osborne, B.G. & Fearn, T. (1983). Collaborative evaluation of near infrared reflectance analysis for the determination of protein, moisture and hardness in wheat. *Journal of the Science of Food and Agriculture*, **34**, 1011-1017.

Pasquini, C. (2003). Near infrared spectroscopy: fundamentals, practical aspects and analytical applications. *Journal of the Brazilian Chemical Society*, **14**(2), 198-219.

Peña, R.J. (2004). Food uses of triticale. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 37-48. Rome: Food and Agriculture Organisation of the United Nations.

Salmon, D.F., Mergoum, M. & Gómez-Macpherson, H. (2004). Triticale production and management. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 27-36. Rome: Food and Agriculture Organisation of the United Nations.

Shenk, J.S. & Westerhaus, M.O. (1985). Accuracy of NIRS instruments to analyze forage and grain. *Crop Science*, **25**, 1120-1122.

Stallknecht, G.F., Gilbertson, K.M. & Ranney, J.E. (1996). Alternative wheat cereals as food grains: einkorn, emmer, spelt, kamut, and triticale. In: *Progress in new crops* (edited by J. Janick). Pp. 156-170. Alexandria, Virginia, USA: ASHS Press.

Viljoen, M., Brand, T.S., Brandt, D.A. & Hoffman, L.C. (2005). Prediction of the chemical composition of winter grain and maize with near infrared reflectance spectroscopy. *South African Journal of Plant and Soil*, **22**(2), 89-93.

Villegas, E., McDonald, C.E. & Gilles, K.A. (1970). Variability in the lysine content of wheat, rye, and triticale protein. *Cereal Chemistry*, **47**, 746-757.

Williams, P.C. & Sobering, D.C. (1986). Attempts at standardization of hardness testing of wheat. II. The near-infrared reflectance method. *Cereal Foods World*, **31**(6), 417-420.

CHAPTER 2

Literature review

Contents

1. Introduction 9

2. Triticale (X *Triticosecale* Wittmack) 10

2.1 Origin 10

2.2 Genetics 12

2.3 Cultivar improvements 13

2.4 Nutritional composition 14

2.5 Major producers and yield performance 15

3. Current uses of triticale 16

3.1 Triticale for human consumption 16

3.2 Triticale as animal feed 17

3.3 The use of triticale for the production of biofuels 18

4. Near infrared spectroscopy 18

4.1 Backgound 18

4.2 Principles of NIR spectroscopy 19

4.3 Instrumentation 20

4.4 Calibration development 21

4.4.1 Chemometrics and Multivariate calibration methods 21

4.4.2 Statistical evaluation 22

5. Methods of quality analysis for grains 23

6. Conclusion 24

7. References 24

CHAPTER 2

LITERATURE REVIEW

1. Introduction

Triticale, the first cereal grain to be successfully produced by humans by a deliberate action, was developed in 1875 by crossing durum wheat (*Triticum* sp.) with rye (*Secale* sp.) (Stallknecht *et al*., 1996; Ammar *et al*., 2004; Oettler, 2005). Since then this crop and its development has been avidly studied and followed by scientists all over the world. The aim of the development of such a crop was to merge the positive attributes of both parent species, namely the suitability of wheat for use in the production of numerous food products with rye's ability to adapt to less than ideal soils and climates, as well as its low input requirement. The expectations and excitement regarding triticale in its early years, however, seems to have exceeded its development. Nevertheless, when one considers the thousands of years that have gone into the development of most major crops since their domestication, it can be argued that the results obtained with triticale are rather extraordinary in view of the little time and effort that has gone into its development.

Where research and effort have been continual, modern lines of triticale perform quite comparatively with top wheat cultivars. Moreover, it has been found that triticale often out yields and outperforms even the best wheat cultivars in marginal soils under unfavourable conditions, such as arid and semi-arid areas, as well as acidic soils (Wu *et al*., 1976; Wu *et al*., 1978; Ammar *et al*., 2004; Salmon *et al*., 2004; Tohver *et al*., 2005). This implies that triticale could hold a great deal of advantage from an economic point of view, seeing as it could expand the area available for the cultivation of crops into marginal lands, thereby providing farmers with an additional crop and greater alternatives for production (Mergoum *et al*., 2004).

A negative attribute of triticale, however, is that it does not compare well with wheat when used in baked products, due to poor quality gluten and low levels of it (Stallknecht *et al.,* 1996; Peña, 2004). For this reason most of the triticale produced in the world is currently used for animal feed purposes (Boros, 2006). A great deal of work is, however, being done on the improvement of triticale cultivars for the purpose of human consumption.

Breeding efforts around the world aiming to improve triticale's characteristics have a need to evaluate early generations of each new cultivar. The evaluation of compositional and functional quality, including the determination of the presence of key processing

characteristics, form a large part of the evaluation of early generations of new lines. Often, very small sample sizes are available early in the breeding process, and the desire for a fast and accurate method of determination requiring only small amounts of sample thus exists. It is equally as important that the testing be non-destructive as the small samples of grain may be required for planting in the next generation. This is where technologies such as near infrared (NIR) spectroscopy hold a great deal of promise. NIR spectroscopy evaluation is rapid, accurate, economical, non-destructive, requires minimal or no sample preparation and requires only small amounts of sample (Butler, 1983; Osborne, 2000; Pasquini, 2003).

2. Triticale (X *Triticosecale* Wittmack)

2.1 Origin

Triticale (X *Triticosecale* Wittmack) was first developed in Europe in the latter half of the 19th century, and it is reported that the first cross between durum wheat and rye was successfully attempted in Scotland in 1875 by A. Stephen Wilson (Stallknecht *et al*., 1996; Ammar *et al*., 2004; Oettler, 2005). Wilson managed to obtain plants with attributes that were a combination of those of the two parent species and presented a report on this hybrid plant to the Botanical Society of Edinburgh in 1875 (Ammar *et al*., 2004). These plants were, however, completely sterile due to the fact that they carried dysfunctional pollen grains (Ammar *et al*., 2004). It was only in 1888 that the first stable amphiploid plant was produced from wheat and rye by the German breeder Rimpau. His plants had a uniform appearance and proved to be true breeding through many generations (Ammar *et al*., 2004).

Due to initial confusion regarding the nomenclature and naming of the new hybrid, a large number of names were proposed (Oettler, 2005). A researcher by the name of Wittmack suggested in 1899 that the names of the parent species be put together, and eventually the name *Triticosecale*, or triticale for short, was accepted in accordance with the international code of nomenclature. In 1971 a scientist, Bernard R. Baum, suggested that the full name should be X *Triticosecale* Wittmack, in honour of the researcher who first proposed the name, and it is now the designation used worldwide (Oettler, 2005). The first record of the name triticale was published in literature in Germany in 1935 (Stallknecht *et al*., 1996).

A substantial amount of effort was put into trying to improve triticale's attributes in the decades that followed its initial development, and despite improvements when compared to previous years, triticale was still very much inferior to wheat in terms of yield potential

(Ammar *et al*., 2004). This was mainly due to triticale's partial sterility, shrivelled kernels, tendency to lodge and its susceptibility to sprouting. Due to these results, the potential future of triticale as a cereal crop seemed rather bleak throughout the 1930's and 1940's. A breakthrough came when a method was developed by which the chromosomes of a plant could be doubled using colchicines (Ammar *et al*., 2004), a natural plant alkaloid that has the effect of doubling the number of chromosomes in half of the gametes during meiosis, while leaving the other half of gametes with no chromosomes. Combined with the discovery of applying colchicine for improved plant breeding, came improvements in the methods of embryo culturing on artificial media (Ammar *et al*., 2004). At the same time international attention was also turning towards the development of hexaploid triticales, as more success was achieved with hexaploid than with octoploid triticales (Ammar *et al*., 2004). In-depth scientific research on triticale only began in 1954 at the University of Manitoba in Canada when a privately funded Research Chair was established with the explicit aim to finally develop triticale as a commercial crop (Kent & Evers, 1994; Ammar *et al*., 2004). The aim of plant breeders was to combine the best of both parent plants, i.e. the uniformity and quality of wheat, with the disease resistance, hardiness and yield of rye (Wu *et al*., 1976).

A similar effort to the Canadian one was launched in Hungary at the same time, resulting in the first-ever two cultivars of triticale to be released commercially in 1968 (Ammar *et al*., 2004). They were known as Triticale No. 57 and Triticale No. 64. One year later, these two cultivars were grown on 40 000 hectare (ha) by Hungarian farmers (Ammar *et al*., 2004). The Canadian effort released its first commercial cultivar, known as Rosner, in 1969 (Ammar *et al*., 2004). Other similar triticale breeding programs were initiated in Poland in the 1960's (Varughese *et al*., 1997; Arseniuk & Oleksiak, 2004) and Australia in the 1970's (Cooper *et al*., 2004), which also contributed a great deal to the development of triticale.

The rapid development and spread of triticale since the 1960's can greatly be attributed to the efforts of the International Maize and Wheat Improvement Center (CIMMYT) which was founded in Mexico in 1966 (Ammar *et al*., 2004). This organisation has the objective of developing improved maize and wheat germplasm, but it rapidly became an international base for the breeding of triticale in conjunction with its main mandate (Ammar *et al*., 2004). CIMMYT and the research centre at the University of Manitoba soon started working closely together by interbreeding their respective germplasm and primaries. They also made use of the contrasting climatic conditions at these two centres to develop

triticales that were adapted to a range of altitudes, soil types and environments (Ammar *et al*., 2004).

The gap between the yield of triticale and that of wheat was greatly reduced by the middle of the 1970's, and the adaptability of triticale around the globe was established a mere 15 years after the development and production of triticale had commenced in the 1960's (Ammar *et al*., 2004). Due to the efforts of CIMMYT and other breeding programs, 146 triticale cultivars were released for commercial production in 23 countries across five continents between the years 1975 and 2000 (Ammar *et al*., 2004). This successful spread of triticale throughout the world furthermore prompted local breeding initiatives in various countries, resulting in the production of their own primaries. Such initiatives in France, Ukraine, Romania, the Russian Federation (Ammar *et al*., 2004) and South Africa (Roux *et al*., 2006), amongst others, have resulted in very successful and widely-grown cultivars.

2.2 Genetics

Triticale is an allopolyploid (or amphiploid) plant, which means that its cells contain the combined genomes of two or more plant species, and thus contain more than the usual single pair of chromosomes per cell, as in the case of euploid plants (Kent & Evers, 1994; Ammar *et al*., 2004). Hexaploid triticale stably bares the genomes of durum wheat (A and B genomes) and rye (R genome) (Varughese *et al*., 1997), and contains the complete set of chromosomes of both these parent species (Ammar *et al*., 2004). As far as the appearance of the grain kernel is concerned, triticale resembles its wheat parent more than it does its rye parent in terms of grain shape, size and colour (Peña, 2004). Both hexaploid and octaploid triticale cultivars have been bred. The hexaploid plants (n=42) were produced from a durum wheat (AABB) (tetraploid, n=28) and diploid rye (n=14) (Wu *et al*., 1978; Kent & Evers, 1994; Briggs, 2001). The octaploid plants (n=56) contain chromosomes derived from a bread wheat (AABBDD) (hexaploid, n=42) and diploid rye (n=14) (Wu *et al*., 1978; Kent & Evers, 1994; Briggs, 2001). Rye is always the pollen parent (Kent & Evers, 1994). Most advanced triticale cultivars are hexaploid, as hexaploid lines are more vigorous and fertile than octoploid lines (Wu *et al*., 1978; Stallknecht *et al*., 1996). Most octoploid lines had poor seed development and were generally much more unstable than hexaploid lines, resulting in the conversion of many breeding programmes to hexaploid cultivars (Salmon *et al*., 2004).

The wheat parent of hexaploid triticale was bred from tetraploid wheat, which does not contain the D-genome (the genome responsible for some of the major breadmaking quality

attributes of hexaploid wheat) (Tohver *et al*., 2005). Furthermore, the secalins encoded by the rye chromosomes contained by triticale have an evident detrimental effect on its bread quality. The absence of the D-genome results in the elimination of one third of the storage protein loci which are responsible for the breadmaking quality of wheat, including *Glu-D1* (on 1DL), *Gli-D1* and *Glu-D3* (on 1DS) as well as *Gli-D2* (on 6DS) (Wos *et al*., 2006; Martinek *et al*., 2008). This absence, together with the presence of the rye secalin loci (*Sec-3* on 1RL, *Sec-1* on 1RS and *Sec-2* on 2RL), results in a considerable decrease in the rheological properties and gluten strength of the dough, as well as a significant increase in the stickiness of the dough (Tohver *et al*., 2005; Wos *et al*., 2006; Martinek *et al*., 2008). The absence of the D-genome furthermore results in a loss of hardness, as this genome is responsible for hardness in wheat (Budak *et al*., 2004). To improve triticale's breadmaking quality would require the incorporation of high molecular weight (HMW) subunits found on the 1D genome in order to introduce their positive effects (Martinek *et al*., 2008).

2.3 Cultivar improvements

The triticale cultivars originally developed did not seem to pose much promise for the baking industry. They had shrivelled kernels which did not mill well, and furthermore resembled their rye parent more than their wheat parent, in that they were prone to undesirably high α-amylase activity (Kent & Evers, 1994; Peña, 2004). They were also characterised by long weak straw, low yields, high susceptibility to ergot (*Claviceps purpurea*) (Stallknecht *et al*., 1996). A positive aspect was that they were also found to contain high levels of protein and the amino acid lysine (Stallknecht *et al*., 1996). After numerous efforts over the years by the triticale research community to improve the characteristics of triticale by crossing it with bread wheats, modern lines of triticale now have improved agronomic traits such as higher yields, even higher levels of lysine, resistance to ergot and lodging, plump kernels, as well as resistance to drought, cold and acidic soils (Stallknecht *et al*., 1996; Naeem *et al*., 2002). In countries where there has been a focus on the breeding and development of triticale, modern cultivars can compete with the best common wheats when conditions are favourable, and are found to be higher yielding than most wheats when grown under unfavourable conditions and in marginal soils (Wu *et al*., 1976; Wu *et al*., 1978; Ammar *et al*., 2004; Salmon *et al*., 2004; Tohver *et al*., 2005). Such adverse conditions include drought, extreme pH values, extreme temperatures, deficient or toxic levels of trace elements and salinity (Salmon *et al*., 2004). When comparing the yield of triticale cultivars developed by CIMMYT during the 1980's to

that developed during the 1990's, it was found that there was an average yield increase of 1.5% per year (Mergoum *et al*., 2004).

In terms of flour yield, triticale has been found to yield less flour upon milling than wheat with 58 – 68% flour for triticale compared to 71 – 75% for Canadian Western Red Spring (CWRS) wheat (Kent & Evers, 1994). This has, however, started to increase in recent years due to breeding efforts. A recent study carried out by Boros (2006) observed that some modern Polish cultivars had a 1000-kernel weight that was equal to or even exceeded that of wheat. Based on a relationship in wheat where increased 1000-kernel weight correlates to an increase in flour yield, it could be expected that an increased 1000-kernel weight could result in an increased flour yield in triticale.

Despite the fact that the breadmaking potential of triticale is known to be poor, recent advances and improvements have been made with chromosome manipulation by restoring the composition of storage protein genes (analogous to those in bread wheat) in triticale (Wos *et al*., 2006). An example of this is the program initiated by the Strzelce Plant Breeding Station in Poland in the year 2000 to improve the breadmaking quality of winter triticale by making use of the multi-breakpoint translocation chromosomes FC1 and Valdy (Wos *et al*., 2006). These chromosomes contain inserts from the 1D chromosome of wheat, and encode for, amongst other things, the important HMW glutenin subunits *5 + 10*. The result of the incorporation of these genes is an improved genetic stability, higher yields, dough characteristics (as expressed by the results of rheological tests) that are more comparable to what can be obtained from good quality bread wheat, and as a result, better breadmaking quality (Wos *et al*., 2006).

2.4 Nutritional composition

From early on in its development, it has been known that triticale has a high nutritive value (Hulse & Laing, 1974). Triticale contains the same chemical components as other cereals, i.e. protein, starch, fat, vitamins, minerals and fibre (Wu *et al*., 1976). The chemical composition of triticale is more similar to the composition of wheat than it is to that of rye, due to the fact that triticale received two genomes from its wheat parent, and only one genome from its rye parent (Varughese *et al*., 1997; Peña, 2004).

Triticale compares well with wheat in terms of nutritional composition, and is generally a good source of vitamins, minerals and essential amino acids (Lorenz *et al*., 1974; Roux *et al*., 2006). The total starch content of Canadian triticale cultivars was found to be equal to or to exceed that of Canadian wheat cultivars (Dyson, 2006). Furthermore, triticale has a high lipid content, a dietary fibre content that is usually higher than that of wheat and a

vitamin content that is more or less similar to that of wheat and rye (Dyson, 2006). Generally, triticale has a higher mineral ash content than wheat (Kent & Evers, 1994; Stallknecht *et al*., 1996). Triticale has also been found to have a soluble as well as total pentosan content that is similar or even slightly higher than that of wheat, yet a good deal lower than that of rye (Saini & Henry, 1989).

Early lines of triticale were found to have levels of protein and the amino acid lysine that were much higher than that of wheat or rye (Stallknecht *et al*., 1996; Peña, 2004). The high lysine levels are significant due to the fact that lysine is usually the limiting amino acid in cereal grains (Kies & Fox, 1970; Villegas *et al.*, 1970). However, the plumper kernels and higher yield potential of modern triticale lines that are the result of careful breeding, have lead to lower levels of protein that are similar to those of normal bread wheat (Stallknecht *et al*., 1996). Nonetheless, the lower protein content did not affect the levels of lysine. It has in fact been found that the lysine content is actually higher when the protein content of a grain is low (Mossé *et al*., 1988). Modern lines of triticale are generally found to have a protein content of between 10 to 16% (Leon *et al*., 1996; Martín *et al*., 1999; Doxastakis *et al.*, 2002; Alaru *et al*., 2003; Roux *et al*., 2006). Canadian triticale cultivars were found to have a slightly lower total protein content compared to that of CWRS wheat, but it was still higher than that of rye, barley, oat and maize (Dyson, 2006). Protein analyses performed on South African triticale cultivars in 2003 and 2004 revealed that it contained 12 – 14.5% protein compared to 14 – 15.5% for good quality bread wheats (Roux *et al*., 2006).

2.5 Major producers and yield performance

Since its development, winter and spring triticale cultivars have been grown in more than 30 countries, including Germany, Sweden, Estonia, Canada, the United States of America (USA), China, Poland, France, Australia, Spain, Switzerland, Italy, Portugal, Hungary and South Africa (Kent & Evers, 1994; Mergoum *et al*., 2004). In 1989, the total global area under triticale cultivation was estimated to be 1.6 million ha (Kent & Evers, 1994). Of this area, Poland and China each contributed 37.5%, while France contributed 8.8% and Australia 6.8% (Kent & Evers, 1994). By the year 2007, around 3.7 million ha was used worldwide to cultivate triticale (FAO, 2007). The worldwide production of triticale increased from 1.2 million tonnes in 1982 to 3.1 million tonnes in 1987, and subsequently to 4.2 million tonnes in 1989 (Kent & Evers, 1994). This figure increased to more than 12 million tonnes in the year 2007 (FAO, 2007). Since the mid-1980's the production of triticale (in

terms of weight) has increased by approximately 18% per year, while the area planted with triticale increased by 23.6% per year (FAO, 2003).

Whereas the production of triticale increases annually, the production of cereals such as rye, oat, sorghum and millet has been decreasing during the last 15 years (Salmon *et al*., 2004). The average yield of triticale worldwide matched the yield of rye in 1984, and has exceeded it thereafter (Kent & Evers, 1994). The ability of triticale to produce a higher yield and biomass than other cereals over a range of soil types as well as climatic conditions, has resulted in its cultivation worldwide (Mergoum *et al*., 2004). The fact that triticale production is increasing so steadily worldwide, seems to indicate that triticale could become valuable in serving as a source of food to the rapidly growing population of the earth.

3. Current uses of triticale

3.1 Triticale for human consumption

Presently, triticale is not used on a large scale in the baking industry (Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al*., 2004). Baked triticale products were available in Canada and the USA for a period of time in the 1980's. Although demand by consumers was high, crop production and product availability decreased due to changes in wheat marketing programs in Canada, and Government support programs of wheat and barley in the USA (Stallknecht *et al*., 1996).

Triticale produces bread with an inferior loaf volume due to a low, weak gluten content as well as inherently high levels of the enzyme α-amylase (Stallknecht *et al.,* 1996; Peña, 2004). Triticale gluten behaves very similarly to that of rye and is too weak to yield bread with quality comparable to that of bread made with wheat flour (Tohver *et al*., 2005). The triticale cultivars with the highest gluten content still contain 20 – 30% less gluten than average wheat cultivars, with wheat averaging around 70% and triticale between 45 – 50% (Peña, 1996). The poor gluten, high α-amylase activity, as well as the higher ash content of triticale, distract from the baking potential of triticale in the industry (Stallknecht *et al.,* 1996). There has, however, been considerable interest during the last decade to improve the nutritional and baking quality of triticale, especially in the area of gene transformation techniques (Stallknecht *et al*., 1996). This has led to the cultivation and production of triticale variants with medium dough strengths, which are suitable for use in a wider variety of baked products (Peña, 2004).

As consumers in general become more health-conscious, they are becoming aware of the health benefits of including a range of cereal grains in their diets (Stallknecht *et al*.,

1996). This increased consumption of grains, together with the current consumer trend of trying new and novel products, is leading to an increase in consumer interest in seeking baked products such as bread that are made using cereal grains other than wheat.

One very positive potential use of triticale is in the production of products that are usually made using soft wheat with weaker dough properties, such as layer cakes, biscuits and cookies (León *et al*., 1996; Pérez *et al*., 2003; Mergoum *et al*., 2004). It is also well-suited for use in health bars, as well as for malting and brewing due to its high α-amylase activity (Peña, 2004).

Thus, given the nutritional and agronomic advantages of triticale, the improvements that are taking place in terms of baking potential, as well as increasing levels of consumer interest in products made from alternative grain cereals, triticale is believed to have the necessary attributes and potential to become an important food cereal for humans in the future (Naeem *et al*., 2002).

3.2 Triticale as animal feed

Most of the triticale harvested around the world is used as livestock feed (Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al*., 2004). Triticale is used for the purpose of animal feed in the form of grain, forage, silage, hay or straw (Myer & Lozano del Rio, 2004). It is a good feed for pigs, poultry and ruminants and can be used for livestock grazing, cut forage, hay, silage, as well as for the dual purpose of forage/grain (Myer & Lozano del Rio, 2004; Anonymous, 2005).

Triticale has been shown in comparative studies to have a biomass yield which is equal to or higher than that of other cereal grains (Delogu *et al*., 2002). This renders it a very good crop for the production of animal feed. Some triticale breeding programs are developing cultivars which are specifically suited for use as animal feed (Myer & Lozano del Rio, 2004). The increased grain plumpness seen with modern cultivars results in higher starch content, and thus a more energy dense grain, compared to the shrivelled kernels of earlier strains (Myer & Lozano del Rio, 2004). Despite the lower protein content associated with the plumper kernels and higher starch content (when compared to older cultivars), the quality and content of protein is still higher than that found in most other cereal grains used as feed (Myer & Lozano del Rio, 2004). Modern triticale cultivars have a higher protein and essential amino acid content (especially lysine) than maize (Myer & Lozano del Rio, 2004). Lysine is typically the most limiting essential amino acid in the diet of pigs (Myer & Lozano del Rio, 2004). When compared to wheat, triticale usually has an overall protein content which is slightly lower than or similar to that of wheat, yet the

concentrations of lysine and threonine are generally higher (Boros, 2002). These higher levels of limiting essential amino acids, especially lysine and threonine, result in the fact that smaller amounts of a supplemental protein source are necessary when using triticale in the diets of poultry and pigs (Myer & Lozano del Rio, 2004). More importantly, it has been found that the digestibility of the protein and amino acids in triticale grain is similar to or better than that of wheat and maize (Hill, 1991; Van Barneveld, 2002). The energy content of modern variants of triticale is usually between 95 – 100% of what can be expected for maize and wheat for non-ruminant animals, and is equal to that of wheat, maize and barley for ruminants (Hill, 1991; Boros, 2002; Van Barneveld, 2002). It has a higher level and greater availability of phosphorus than maize (Van Barneveld, 2002). Triticale forage compares very well to other forage cereals in terms of nutritive values (Varughese *et al*., 1996).

3.3 The use of triticale for the production of biofuels

Modern cultivars of triticale have been found to be very competitive as a feedstock for bio-ethanol production (Eudes, 2006). It is a more vigorous and adaptable crop than either of its parent species as well as oats and barley, and it produces a greater biomass when receiving similar input to these crops (Eudes, 2006). Due to its high starch content, it has the ability to supply large quantities of carbohydrate polymers which can serve as a feedstock for bio-ethanol production (Eudes, 2006). Crops that have a high yield potential as well as high starch content, together with a low content of soluble polysaccharides and protein, are considered to be ideal for bio-ethanol production (Boros, 2006).

4. Near infrared spectroscopy

4.1 Background

Near infrared spectroscopy was discovered unintentionally in 1800 by Sir Fredrick William Herschel, an astronomer and musician (Butler, 1983; Davies, 1998; McClure, 2003; Pasquini, 2003). Herschel was looking for a colour of glass for a telescope that would allow the maximum amount of light and minimum amount of heat to pass through. Herschel used a blackened thermometer to measure the temperature in each region of the colour spectrum caused by sunlight passing through a prism, and noticed that the temperature continued to climb when the thermometer was left in the area beyond the end of the visible red light region (Butler, 1983; Davies, 1998; McClure, 2003; Pasquini, 2003). He came to the conclusion that there was energy in the region beyond the red light in waves not visible to the human eye (Butler, 1983), and called it “calorific rays” (Pasquini,

2003). It later became known as infrared, derived from the Greek prefix “infra”, meaning below (Pasquini, 2003).

Although the NIR region was the first invisible part of the electromagnetic spectrum to be discovered, it was a region neglected by spectroscopists for decades due to its broad, weak and overlapping absorption bands, thought to be unusable (Butler, 1983; Davies, 1998; McClure, 2003; Pasquini, 2003). Interest in the mid infrared (MIR) region saw an increase during the Second World War when the technology was used in the field, while the NIR region received virtually no attention (McClure, 2003). However, with the invention of the computer, it was found that the spectra produced by NIR spectroscopy could be interpreted (Davies, 1998), and so started the boom of NIR spectroscopy. Pioneers such as Karl Norris (generally regarded as the father of NIR spectroscopy), Phil Williams, Fred McClure, John Shenk and others opened the door to the potential of NIR spectroscopy (Davies, 1998; McClure, 2003). During this time, NIR spectroscopy went through a period of rapid development brought about mainly by the improvement of NIR instruments, the development of the computer, and the development of a new discipline named chemometrics, a tool for gathering and interpreting the spectral data obtained (Pasquini, 2003).

Today NIR spectroscopy has gained widespread acceptance as a fast, accurate and economical method of analysis that is non-destructive, requires minimal or no sample preparation, and is almost universally applicable (Butler, 1983; Osborne, 2000; Pasquini, 2003). NIR spectroscopy is mainly used for quality assessment, process control or for identification, and has hundreds of applications, including grains, forages, feeds, flour, baked products, dairy products, pharmaceuticals, petrochemicals, fine chemicals, radioactive materials, and more recently medical imaging and diagnostics (Osborne *et al.*, 1993; Workman, 2005).

4.2 Principles of NIR spectroscopy

Near infrared spectroscopy is a form of vibrational spectroscopy that makes use of photon energy in the range of 2.65×10^{-19} to 7.96×10^{-20} J, which corresponds to the wavelength range of 780 to 2500 nm (Pasquini, 2003; Workman, 2005). The method is based on scanning an object to obtain qualitative and/or quantitative information resulting from the interaction of the NIR electromagnetic waves with the constituents of the sample (Pasquini, 2003), and then exploiting the empirical relationship between spectral and reference analytical data (obtained by conventional analytical methods) (Osborne, 2000).

NIR spectra consist of overtones and combination bands of the fundamental molecular absorptions occurring in the MIR region (Workman, 2005), and they originate when radiant energy is transferred to the vibrational energy of atoms held together by chemical bonds (Osborne *et al.*, 1993). With the addition of energy the amplitudes of these vibrations increase, and similarly to resonance, only radiation of a certain frequency or wavelength can excite the vibrational level of molecules (depending on the fundamental vibrational energy level of the molecules) (Osborne *et al.*, 1993). Thus the radiation needs to have a frequency capable of supplying exactly the amount of energy between two vibrational levels (or of their overtones / combinations of two or more fundamental vibrations) so that it can be absorbed and produce excitation to a higher energy level (Osborne *et al.*, 1993). Thus, for a given molecule, some frequencies of radiation will be absorbed, others will not be absorbed, and some will only be partially absorbed (Osborne *et al.*, 1993). In the NIR region, bonds associated with hydrogen show good absorption, and certain bonds (such as O-H, C-H, N-H and S-H) have known wavelength regions where they absorb (Pasquini, 2003; Workman, 2005). Thus NIR spectroscopy operates by determining the presence of certain functional groups associated with molecules, which can be used either for identification or classification of the sample according to the spectra (qualitative analysis), or can be correlated with known compositional or physical parameters (determined by conventional analytical methods) by using multivariate calibration techniques or chemometrics (quantitative analysis) (Osborne *et al.*, 1993; Workman, 2005).

4.3 Instrumentation

Near infrared spectroscopy instruments have changed considerably since their initial development, and they still continue to change, with new features, uses and flexibilities being added with every new instrument (McClure, 2003). NIR spectroscopy instruments vary in terms of radiation sources, detectors, wavelength selection, and measurement modes (Pasquini, 2003).

Radiation sources are high powered, resulting in a high signal-to-noise ratio. The majority of manufacturers currently use halogen lamps or tungsten coils as radiation sources (Williams & Norris, 2001). The most frequently used detectors currently employed for the NIR region are made from lead sulphide (PbS), silicon or indium gallium arsenide (InGaAs) photoconductive materials (Williams & Norris, 2001).

Different types of instruments exists based on wavelength selection methods, such as Filter-based instruments (including narrow-band interference filters, tilting filters, liquid crystal tunable filters (LCTF) and acousto optical tunable filters (AOTF)), LED-based (light

emitting diodes) instruments, dispersive optics-based instruments (such as grating monochromators) and Fourier-transform based instruments (McClure, 2003; Pasquini, 2003).

The choice of an NIR instrument greatly depends on the nature of the substance to be scanned; be it liquid, solid, powder or slurry, as this influences the measurement mode. Depending on the sample, instrument and measurement mode, the radiation can be absorbed, reflected or transmitted, and the radiation is measured in the form of transmittance, transflectance, diffuse reflectance or interactance (Williams & Norris, 2001).

4.4 Calibration development

The development of NIR spectroscopy calibration models used for the quantitative analysis of a matrix, involves correlating the NIR spectra obtained with values determined by conventional analytical methods (Workman, 2005). Thus, the relationship between the absorbance values (log 1/R) corresponding to the amount of a component present in a sample (as determined by NIR spectroscopy), and the values obtained for the amount of that component present in the matrix (as determined by conventional analytical or reference methods) is expressed as an approximation by using a form of regression equation (Hruschka, 2001). Once the calibration is developed, it can be applied to independent samples to estimate the amount of the component present.

The sample set used for the development of a calibration (the calibration data set) is of great importance, and care must be taken to include both a large enough sample size and a large enough range, to account for all possible variation that may occur when evaluating future samples (Pasquini, 2003).

The reference data used is determined by conventional or traditional analytical methods, such as those accepted by the AACC International, and accuracy here is of great importance in order to obtain effective calibration models (Pasquini, 2003).

4.4.1 Chemometrics and multivariate calibration methods

Due to the large amounts of spectral data obtained from NIR spectroscopy, as well as the complex nature of the NIR region which seldomly permits the use of single wavelength models for quantitative analysis, techniques are needed to extract relevant information from the data (Pasquini, 2003). In the case of NIR spectroscopy, chemometrics is the mathematical and statistical tool of choice.

Chemometrics employs several methods of spectral pretreatment, used to minimise the effect of light / radiation scattering caused by different particle sizes, reduce instrument

noise, and to correct for other spectra baseline-affecting occurrences (Pasquini, 2003; Delwiche & Reeves, 2004). These methods include first and second derivatives of the spectra (Pasquini, 2003), as well as multiplicative scatter correction (MSC) (Geladi *et al.*, 1985) and standard normal variate (SNV) (Barnes *et al*., 1989).

For the quantitative analysis of samples, multivariate regression models such as partial least squares regression (PLS), multiple linear regression (MLR) or principal component regression (PCR) can be used. For MLR, the variables included are the original variables (wavelengths), whereas for PLS and PCR the variables are the principal components (Næs *et al.*, 2002). For PLS and PCR, it is imperative that the optimum number of factors / variables be chosen. Many software packages contain automatic optimisation algorithms which suggest the optimal number of variables, but the user should verify that they are indeed the best.

The predictive ability of a model developed by multivariate regression methods is evaluated by making use of either cross-validation or an independent test set. Cross-validation removes one sample or segment of samples at a time from the total sample set and then predicts their values according to the model, from which the calibration error is calculated (Pasquini, 2003). When using an independent test set, the calibration model is used to predict values for an external set of samples that did not form a part of the calibration data set (Pasquini, 2003). This is known as validation and is the true test of the accuracy of a model.

4.4.2 Statistical evaluation

Statistical evaluation is the last step in the development of a calibration model, and is used to evaluate the accuracy and efficiency of the model. In NIR spectroscopy calibration development, the statistical analyses normally used include the standard error of cross validation (*SECV*), standard error of prediction (*SEP*), coefficient of determination (R^2), the bias and the ratio of the standard error of prediction to the standard deviation of the test set (*RPD*) (Williams, 2001).

The *SECV* is a measure of the accuracy of the model determined from the calibration error when performing a cross-validation, whereas the *SEP* is in the same way a measure of accuracy of the model when a validation is performed using an independent test set. The *SEP* should be as close as possible to the standard error of laboratory (*SEL*). The bias is an indication of how much the results differ, and both the *SEP* and bias should be as close as possible to zero (Williams, 2001). The R^2 value is a measure of how well the spectral data correlates to the reference data, and gives an indication of whether or not the

model has potential for application. A R^2 of as close as possible to one is desired (Williams, 2001). The *RPD* is an important statistic for the evaluation of a model, as it gives an indication of the efficiency of a calibration model (Williams, 2001). Guidelines for the interpretation of the R^2 and *RPD* can be seen in Tables 1 and 2 respectively.

Table 2.1 Guidelines for the interpretation of R^2 (Williams, 2001)

Value of R^2	Interpretation
Up to 0.25	Not usable in near infrared calibrations
0.26-0.49	Poor correlation, reasons should be researched
0.50-0.64[a]	Acceptable for rough screening; more than 50% of variance in *y* accounted for by *x*
0.66-0.81	Acceptable for screening and some other approximate calibrations
0.83-0.90	Can be used with caution for most applications, including research
0.92-0.96	Can be used for most applications, including quality assurance
0.98+	Can be used for any application

[a] Due to rounding off, there are no values for 0.65, 0.82 etc in this table.

Table 2.2 Guidelines for the interpretation of the RPD (Williams, 2001)

RPD value	Classification	Application
0.0-2.3	Very poor	Not recommended
2.4-3.0	Poor	Very rough screening
3.1-4.9	Fair	Screening
5.0-6.4	Good	Quality control
6.5-8.0	Very good	Process control
8.1+	Excellent	Any application

5. Methods of quality analysis for grains

Widely accepted methods for the evaluation of grain quality are available from the AACC International. Methods for the determination of protein content in flour are available for the Kjeldahl method (AACC method 46-11A, AACC, 2008) and combustion method (AACC method 46-30, AACC, 2008). Moisture determination is described in AACC method 44-15A (AACC, 2008). Ash determination can be performed according to AACC method 08-02 (AACC, 2008) as well as AACC method 08-21, which is a near infrared (NIR) spectroscopy method (AACC, 2008). The determination of falling number, a measurement

based on the breakdown of starch gel by the α-amylase present in the sample, is described by AACC method 56-81B (AACC, 2008).

Various methods exist for the determination of hardness in grains, including AACC method 55-30 and adaptations of this method, such as the method described by Williams and Sobering (1986). An NIR method for the determination of grain hardness also exists (AACC method 39-70A, AACC, 2008). Hardness has an effect on the quality as well as the functionality of grains, and is largely genetically determined (Pomeranz & Williams, 1990).

An indication of gluten strength and potential baking quality of grains can be obtained by performing sodium dodecyl sulphate (SDS) sedimentation, where the height of the sediment formed correlates with the gluten strength or quality of a sample (Dick & Quick, 1983; Carter *et al.*, 1999). This results from the swelling of the glutenin strands under the influence of the lactic acid in the stock solution (AACC method 56-60, AACC 2008). It is a very useful preliminary test if only small amounts of sample are available or if time is limited (Dick & Quick, 1983). This method can be performed according to AACC method 56-70 (AACC, 2008), or adaptations thereof, such as the micro SDS sedimentation method described by Dick & Quick (1983).

6. Conclusion

When taking into consideration the high yield of triticale under both biotic and abiotic stress, the changing climatic conditions of the earth and its growing population, it is clear that triticale can make a contribution in future efforts for sustainable food production for the population of the earth. Furthermore it can also contribute in providing feed for animals and as a feedstock for biofuel production; both of which are necessary to support the growing human population. Triticale is thus a crop deserving of continued research and breeding efforts, and NIR spectroscopy can play a vital role in facilitating this.

7. References

AACC (2008). *Approved Methods of the American Association of Cereal Chemists*, 10th ed. St. Paul, Minnesota, USA: American Association of Cereal Chemists.

Alaru, M., Laur, Ü. & Jaama, E. (2003). Influence of nitrogen and weather conditions on the grain quality of winter triticale. *Agronomy Research*, **1**, 3-10.

Ammar, K., Mergoum, M. & Rajaram, S. (2004). The history and evolution of triticale. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp 1-9. Rome: Food and Agriculture Organisation of the United Nations.

Anonymous (2005). Spring and winter triticale for grain, forage and value-added. Triticale production manual. Alberta Agriculture, Food and Rural development [WWW document]. URL http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/fcd1053/$file/TriticaleManualIntroduction.pdf?OpenElement. 4 April 2008.

Arseniuk, E. & Oleksiak, T. (2004). Triticale in Poland. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp 131-134. Rome: Food and Agriculture Organisation of the United Nations.

Barnes, R.J., Dhanoa, M.S. & Lister, S.J. (1989). Standard normal variate transformation and de-trending of near–infrared diffuse reflectance spectra. *Applied Spectroscopy*, **43**(5), 772-777.

Boros, D. (2002). Physico-chemical quality indicators suitable in selection of triticale for high nutritive value. In: *Proceedings of the 5th International Triticale Symposium*, vol. I. Pp. 239-244. Radzików, Poland.

Boros, D. (2006). Triticale of high end-use quality enhances opportunities to increase its value in world cereal market. In: *Proceedings of the 6th International Triticale Symposium.* Pp. 119-125. Stellenbosch, South Africa.

Briggs, K.G. (2001). The growth potential of triticale in western Canada. Pp. 14, Government of Alberta, Canada: Alberta Agriculture, Food and Rural Development Publication.

Budak, H., Baenziger, P. S., Beecher, B.S., Graybosch, R.A., Campbell, B.T., Shipman, M.J., Erayman, M. & Eskridge, K.M. (2004). The effect of introgressions of wheat D-genome chromosomes into 'Presto' triticale. *Euphytica*, **137**, 261–270.

Butler, L.A. (1983). The history and background of NIR. *Cereal Foods World*, **28**(4), 238-240.

Carter, B.P., Morris, C.F. & Anderson, J.A. (1999). Optimizing the SDS Sedimentation test for end-use quality selection in a soft white and club wheat breeding program. *Cereal Chemistry*, **76**(6), 907-911.

Cooper, K.V., Jessop, R.S. & Darvey, N.L. (2004). Triticale in Australia. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp 87-92. Rome: Food and Agriculture Organisation of the United Nations.

Davies, T. (1998). The history near infrared spectroscopic analysis: Past, present and future – "From sleeping technique to the morning star of spectroscopy". *Analusis magazine*, **26**(4), 17-19.

Delogu, G., Faccini, N., Faccioli, P., Reggiani, F., Lendini, M., Berardo, N. & Odoardi, M. (2002). Dry matter yield and quality evaluation at two phenological stages of forage triticale grown in the Po valley and Sardinia, Italy. *Field Crops Research*, **74**, 207-215.

Delwiche, S.R. & Reeves, J.B. (2004). The effect of spectral pre-treatments on the partial least squares modelling of agricultural products. *Journal of Near Infrared Spectroscopy*, **12**, 177-182.

Dick, J.W. & Quick, J.S. (1983). A modified screening test for rapid estimation of gluten strength in early-generation durum wheat breeding lines. *Cereal Chemistry*, **60**(4), 315-318.

Doxastakis, G., Zafiriadis, I., Irakli, M., Marlani, H. & Tananaki, C. (2002). Lupin, soya and triticale addition to wheat flour doughs and their effect on rheological properties. *Food Chemistry*, **77**(2), 219-227.

Dyson, C., 2006. Triticale grain for feed - Nutritional information. Alberta Government Agriculture and Food [WWW document]. URL http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/fcd10575. 12 May 2007.

Eudes, F. (2006). Canadian triticale biorefinery initiative. In: *Proceedings of the 6th International Triticale Symposium*. Pp. 85-88. Stellenbosch, South Africa.

FAO (2003). FOASTAT, FAO statistical databases – agriculture [WWW document]. URL http://apps.fao.org. 14 May 2007.

FAO (2007). FOASTAT, FAO statistical databases – agriculture [WWW document]. URL http://faostat.fao.org/site/567/default.aspx#ancor. 1 December 2008.

Geladi, P., MacDougall, D. & Martens, H. (1985). Linearization and scatter-correction for near-infrared reflectance spectra of meat. *Applied Spectroscopy*, **39**(3), 491-500.

Hill, G.M. (1991). Triticale in animal nutrition. In: *Proceedings of the 2nd International Triticale Symposium*. Pp. 422-427. Passo Fundo, Rio Grande do Sul, Brazil.

Hruschka, W.E. (2001). Data analysis: wavelength selection methods. In: *Near-Infrared Technology in the Agricultural and Food Industries*, 2nd ed. (edited by P. Williams & K. Norris). Pp. 39-58. St. Paul, USA: American Association of Cereal Chemists.

Hulse, J.H. & Laing, E.M. (1974). Nutritive value of triticale protein. Ottawa, Canada: International Development Research Centre (as cited by Boros, 2006).

Kent, N.L. & Evers, A.D. (1994). Kent's Technology of Cereals: An Introduction for Students of Food Science and Agriculture. Pp. 17-18, 50, 96-97, 157. New York, USA: Elsevier Science Ltd.

Kies, C. & Fox, H.M. (1970). Protein nutritive value of wheat and triticale grain for humans, studied at two levels of protein intake. *Cereal Chemistry*, **47**, 671-678.

León, A.E., Rubiolo, A. & Añón, M.C. (1996). Use of triticale flours in cookies: quality factors. *Cereal Chemistry*, **73**, 779-784.

Lorenz, K., Reuter, F.W. & Sizer, C. (1974). The mineral composition of triticales and triticale milling fractions by X-ray fluorescence and atomic absorption. *Cereal Chemistry*, **51**, 534-541.

Martín, A., Alvarez, J.B., Martín, L.M., Barro, F. & Ballesteros, J. (1999). The development of Tritordeum: a novel cereal for food processing. *Journal of Cereal Science*, **30**, 85-95.

Martinek, P., Vinterová, M., Burešová, I. & Vyhnánek, T. (2008). Agronomic and quality characteristics of triticale (X *Triticosecale* Wittmack) with HMW glutenin subunits *5 + 10*. *Journal of Cereal Science*, **47**, 68-78.

McClure, W.F. (2003). 204 Years of near infrared technology: 1800 – 2003. *Journal of Near Infrared Spectroscopy*, **11**, 487-518.

Mergoum, M., Pfeiffer, W.H., Peña, R.J., Ammar, K., Rajaram, S., 2004. Triticale crop improvement: the CIMMYT programme. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 11-26. Rome: Food and Agriculture Organisation of the United Nations.

Mossé, J., Huet, J.C. & Baudet, J. (1988). The amino acid composition of triticale grain as a function of nitrogen content: comparison with wheat and rye. *Journal of Cereal Science*, **7**, 49-60.

Myer, R. & Lozano del Rio, A.J. (2004). Triticale as animal feed. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 49-58. Rome: Food and Agriculture Organisation of the United Nations.

Næs, T., Isaksson, T., Fearn, T. & Davies, T. (2002). A user-friendly guide to multivariate calibration and classification. Pp. 344. Chichester, UK: NIR Publications.

Naeem, H.A., Darvey, N.L., Gras, P.W. & MacRitchie, F. (2002). Mixing properties, baking potential, and functionality changes in storage proteins during dough development of triticale-wheat flour blends. *Cereal Chemistry*, **79**(3), 332-339.

Oettler, G. (2005). The fortune of a botanical curiosity – Triticale: past, present and future. *Journal of Agricultural Science*, **143**, 329-346.

Osborne, B.G. (2000). Recent developments in NIR analysis of grains and grain products. *Cereal Foods World*, **45**(1), 11-15.

Osborne, B.G., Fearn, T. & Hindle, P.H. (1993). *Practical NIR Spectroscopy with Practical Applications in Food and Beverage Analysis,* 2nd ed. Pp227. Harlow, UK: Longman Scientific and Technical.

Pasquini, C. (2003). Near infrared spectroscopy: fundamentals, practical aspects and analytical applications. *Journal of the Brazilian Chemical Society*, **14**(2), 198-219.

Peña, R.J. (1996). Factors affecting triticale as a food crop. In: *Triticale: today and tomorrow.* (edited by H. Guedes-Pinto, N. Darvey & V.P. Carnide). Pp. 753-761. Dordrecht, The Netherlands: Kluwer Academic Press.

Peña, R.J. (2004). Food uses of triticale. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 37-48. Rome: Food and Agriculture Organisation of the United Nations.

Pérez, G.T., León, A.E., Ribotta, P.D., Aguirre, A., Rubiolo, O.J. & Añón, M.C. (2003). Use of triticale flours in cracker-making. *European Food Research and Technology*, **217**, 134-137.

Pomeranz, Y. & Williams, P.C. (1990). Wheat hardness: its genetic, structural, and biochemical background, measurement, and significance. In: *Advances in Cereal Science and Technology*, *Vol. 10* (edited by Y. Pomeranz). Pp. 471-557. St Paul, Minnesota: American Association of Cereal Chemists.

Roux, H.S., Marais, G.F., Snyman, J.E. & Botes, W.C. (2006). The South African triticale breeding programme: current status. In: *Proceedings of the 6th International Triticale Symposium.* Pp. 80-84. Stellenbosch, South Africa.

Salmon, D.F., Mergoum, M. & Gómez-Macpherson, H. (2004). Triticale production and management. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 27-36. Rome: Food and Agriculture Organisation of the United Nations.

Saini, H.S. & Henry, R.J. (1989). Fractionation and evaluation of triticale pentosans: comparison with wheat and rye. *Cereal Chemistry*, **66**, 11-14.

Stallknecht, G.F., Gilbertson, K.M. & Ranney, J.E. (1996). Alternative wheat cereals as food grains: einkorn, emmer, spelt, kamut, and triticale. In: *Progress in new crops* (edited by J. Janick). Pp. 156-170. Alexandria, Virginia, USA: ASHS Press.

Tohver, M., Kann, A., Täht, R., Mihhalevski, A. & Hakman, J. (2005). Quality of triticale cultivars suitable for growing and bread-making in northern conditions. *Food Chemistry*, **89**, 125-132.

Van Barneveld, R.J. (2002). Triticale: a guide to the use of triticale in livestock feeds. Pp. 1-12. Kingston, Australia: Grains Research Development Corporation.

Varughese, G., Pfeiffer, W.H. & Peña, R.J. (1996). Triticale (Part 1): a successful alternative crop. *Cereal Foods World*, **41**(7), 474-482.

Varughese, G., Pfeiffer, W.H. & Peña, R.J. (1997). Triticale: a reappraisal. World Bank [WWW document]. URL http://www.worldbank.org/html/cgiar/newsletter/april97/8tritic.html. 8 March 2008.

Villegas, E., McDonald, C.E. & Gilles, K.A. (1970). Variability in the lysine content of wheat, rye, and triticale protein. *Cereal Chemistry*, **47**, 746-757.

Williams, P.C. (2001). Implementation of near-infrared technology. In: *Near-Infrared Technology in the Agricultural and Food Industries*, 2nd ed. (edited by P. Williams & K. Norris). Pp 145-169. St. Paul, USA: American Association of Cereal Chemists.

Williams, P.C. & Norris, K. (2001). Variables affecting near-infrared spectroscopic analysis. In: *Near-Infrared Technology in the Agricultural and Food Industries*, 2nd ed. (edited by P. Williams & K. Norris). Pp. 171-186. St. Paul, USA: American Association of Cereal Chemists.

Williams, P.C. & Sobering, D.C. (1986) Attempts at standardization of hardness testing of wheat. I. The grinding/sieving (paticle size index) method. *Cereal Foods World*, **31**(5), 359-364.

Workman, J. (2005). An introduction to near infrared spectroscopy. Infrared Spectroscopy. [WWW document]. URL http://www.spectroscopynow.com. 15 October 2008.

Wos, H., Arseniuk, E., Brzezinski, W. & Stachowicz, M. (2006). Incorporation of breadmaking quality to winter triticale breeding program. In: *Proceedings of the 6th International Triticale Symposium*. Pp. 182-183. Stellenbosch, South Africa.

Wu, Y.V., Sexson, K.R. & Wall, J.S. (1976). Triticale protein concentrate: preparation, composition, and properties. *Journal of Agricultural and Food Chemistry*, **24**(3), 511-517.

Wu, Y.V., Stringfellow, A.C., Anderson, R.A., Sexson, K.R. & Wall, J.S. (1978). Triticale for food uses. *Journal of Agricultural and Food Chemistry*, **26**(5), 1039-1048.

CHAPTER 3

Evaluation of the compositional and functional quality of South African triticale (X *Triticosecale* Wittmack) cultivars using conventional methods

CHAPTER 3

EVALUATION OF THE COMPOSITIONAL AND FUNCTIONAL QUALITY OF SOUTH AFRICAN TRITICALE (X *TRITICOSECALE* WITTMACK) CULTIVARS USING CONVENTIONAL METHODS

Abstract

Little is known regarding the compositional and functional quality of triticale (X *Triticosecale* Whittmack) cultivars developed and bred in South Africa, and a comprehensive study in this regard has not been performed. Different cultivars of South African triticale from various localities in the Western Cape obtained for two subsequent harvest season were evaluated in this study (using conventional methods) in order to evaluate the protein and ash contents, as well as the α-amylase activity (falling number), hardness (particle size index), 1000-kernel mass and baking potential (SDS sedimentation). It was found that the quality of South African cultivars compared well with what has been observed in worldwide studies, and that the cultivars fared similarly when compared to wheat as in these studies. Significant differences were observed between both cultivars and localities within years, illustrating the effect of genetic as well as environmental factors on the quality of the grain produced. It was observed in most cases that significant differences existed between localities when comparing the two harvest seasons, whereas differences between the cultivars for the two seasons were mostly not significant. From these results the effect of environment becomes evident. Interactions between cultivars and localities were found to be significant in all cases, and correlations were observed between protein content and PSI as well as protein content and SDS sedimentation results for both years.

Introduction

The original aim with the development of triticale (X *Triticosecale* Wittmack) was to obtain a crop that possesses the advantageous characteristics of both parent species, i.e. durum wheat (*Triticum* sp.) and rye (*Secale* sp.). The suitability of wheat for use in various baked products was desired in combination with the hardiness, adaptability to less than ideal environmental conditions and low input requirements of rye. The latter was achieved with great success, as triticale has proven itself as a hardy crop which can grow in marginal soils and unfavourable environmental conditions (Wu *et al*., 1976; Wu *et al*., 1978; Ammar *et al*., 2004; Salmon *et al*., 2004; Tohver *et al*., 2005). Due to its weak gluten and high α-

amylase activity triticale is, however, not yet comparable to wheat in terms of its potential for use in baked products and is therefore not used on a large scale in the baking industry (Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al.*, 2004).

The poor baking performance of triticale is confirmed when it is evaluated in terms of functional quality, as it does not compare well with wheat in terms of parameters that are indicative of baking quality (Kent & Evers, 1994; Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al.*, 2004). It does, however, compare well with wheat in terms of nutritional composition and is therefore widely used as animal feed (Stallknecht *et al*., 1996; Peña, 2004; Salmon *et al.*, 2004).

The advancements of breeding lines within a breeding program will depend on the intended use of cultivars, be it for baking purposes, animal feed or for use as raw material for biofuel production. The quality characteristics of a breeding line thus play a very important role in determining the future of a new cultivar, and it is of great importance to evaluate for these quality parameters.

Worldwide studies on the compositional quality of triticale have found it to have a protein content of between 10 and 16% (Bushuk & Larter, 1980; Peña & Bates, 1982; Johnson & Eason, 1988; Heger & Eggum, 1991; Kulshrestha & Usha, 1992; Leon *et al*., 1996; Alaru *et al.*, 2003; Roux *et al*., 2006). This is comparable to that of bread wheat, which usually has a protein content of between 9 and 14% (Hunter & Stanford, 1973; Chawla & Kapoor, 1982; Graybosch *et al.*, 1995; Martín *et al.*, 1999; Anon., 2001). It has been reported that the protein content of triticale depends more on cultivar (genetic predisposition) than on environment, although growing environment does have a strong influence (Alaru *et al.,* 2003).

Ash content values for triticale have been found to range from 0.44 to 3.0% with most values between 1 and 2% (Lorenz & Maga, 1972; Leon *et al*., 1996; Seguchi *et al*., 1999; Doxastakis *et al*., 2001). The ash content of triticale is generally higher than that of wheat (Kent & Evers, 1994; Leon *et al*., 1996; Stallknecht *et al*., 1996) and has also been reported to be higher than that of einkorn and durum (D'Egidio *et al*., 1993; Leon *et al*., 1996). This higher ash content can be detrimental to baking quality (Doxastakis *et al*., 2001).

Being one of the main negative influences on the dough properties and thus baking quality of triticale, the high α-amylase activity (as measured by the falling number) is a very important quality indicator to evaluate when considering breeding for the purpose of baking (Jestin & Bonhomme, 1996). The falling number for triticale is generally very low compared to that of wheat, and can even be lower than what is expected for rye (Erekul &

Köhn, 2006). Triticale flowers earlier and ripens later compared to wheat, which results in a longer period of seed-filling (Pfeiffer, 1994). This can be a disadvantage in years where rains are experienced during the time of seed-filling, due to the occurrence of lodging and pre-harvest sprouting (Pfeiffer, 1994; Alaru *et al*., 2003). Falling number values for triticale of between 62 and 180 seconds, with the average below 100 seconds, have been reported (Leon *et al*., 1996; Alaru *et al*., 2003; Erekul & Köhn, 2006; Jondreville *et al*., 2007). It was also found that falling numbers were much lower in years with higher rainfall, whereas it was higher in samples from semi-arid cultivation areas (Leon *et al*., 1996; Erekul & Köhn, 2006).

The hardness of a grain is an indication of its suitability for different end-uses or commercial purposes (Wiliams & Sobering, 1986). There are numerous methods to determine grain hardness, of which the particle size index (PSI) method is one of the most commonly used methods (Williams & Sobering, 1986). Varying PSI values are observed for wheat, depending on whether it is a hard, medium or soft wheat. Triticale hardness as determined by PSI generally ranges from soft to very soft (Alvarez *et al*., 1992; Martín *et al*., 1999; Ramírez *et al*., 2003; Barrera *et al*., 2007) when compared to wheat, although some harder triticale cultivars have been observed (Ramírez *et al*., 2003).

The average 1000-kernel mass for triticale has been found to range from 35 to 55 g (Alvarez *et al*., 1992; Erekul & Köhn, 2006; Jondreville *et al*., 2007; Kozak *et al*., 2007). Being indicative of kernel density and thus potential flour yield, the 1000-kernel mass is greatly determined by cultivar or genetic predisposition, and to a lesser extent by environmental conditions; with overly dry conditions having a negative effect on the 1000-kernel weight (Erekul & Köhn, 2006).

Sodium dodecyl sulphate (SDS) sedimentation is a rapid, small-scale test that predicts the baking potential of flours by giving an indication of gluten strength (Moonen *et al*., 1982; Dick & Quick, 1983). It has been shown that SDS-sedimentation results correlate positively with the bread-making quality of flour and are highly repeatable (Moonen *et al*., 1982; AACC method 56-60, AACC 2008). A study performed by Martín *et al.* (1999) on triticale obtained an average value of 41.2 mL for the SDS-sedimentation test. The weak gluten of triticale is demonstrated by the results of this test, as the values obtained for triticale are often not even half of that obtained for wheat (Erekul & Köhn, 2006).

Limited information is currently available regarding the compositional and functional quality of South African triticale cultivars and a comprehensive study has never been performed. The objective of this study was therefore to evaluate the compositional and functional quality of South African triticale cultivars, grown over two seasons, as expressed

by protein and ash contents as well as falling number, PSI, 1000-kernel mass and SDS-sedimentation values.

Materials and Methods

Triticale and wheat samples

Six triticale cultivars (US2007, USGen19, Bacchus, Tobie, Rex and Ibis) from each of six localities (Langgewens, Napier, Roodebloem, Mariendahl, Tygerhoek and Vredenburg), harvested during the 2006 season, were obtained (n = 36). Only one replicate was available for each location. In addition, six triticale cultivars by three replicates (US2007, AgBeacon, Bacchus, Tobie, Rex and Ibis), harvested during the 2007 season, from each of nine localities (Langgewens, Napier, Roodebloem, Mariendahl, Tygerhoek, Riversdal, Piketberg, Klipheuwel and Albertinia) were obtained (n = 162). Three replicates of Kariega, a bread wheat cultivar currently used as a South African baking standard, were obtained from all localities (except Mariendahl) for the 2007 season (n = 24). The triticale samples obtained were derived from a breeding program that was not focused on baking quality. All grain samples were kindly supplied by the Department of Genetics, Stellenbosch University, South Africa.

The whole grain samples were stored at 4°C until being milled. All samples were milled on a UDY Cyclone mill (UDY Corporation, Fort Collins, Colorado, USA) fitted with a 1 mm sieve. The flour samples were kept in airtight containers at room temperature until being analysed.

Chemical composition and functional quality of triticale

Moisture content

The moisture determination was performed according to AACC Approved Method 44-15A (AACC, 2008). The mass of each moisture dish with its lid was obtained to the nearest 0.001 g and recorded (W_1), after which 5 ± 0.001 g of the ground flour sample was weighed into the moisture dish and the new mass recorded (W_2). Each moisture dish was then placed uncovered (with the lid beneath the dish) in an air oven (Model EM 10, Chopin, Villeneuve-la-Garenne Cedex, France) at 130°C for an hour. After being removed from the oven, the lids were placed back on the dishes which were subsequently allowed to cool in a desiccator for 40 min. The mass of the covered dishes were then obtained to the nearest 0.001 g (W_3), and the moisture content (%) was determined according to equation 3.1. The results obtained for the moisture determination were used in subsequent analyses.

% moisture = $(W_2 – W_3 / W_2 – W_1) \times 100$ … equation 3.1

where: W_1 = mass of the moisture dish

W_2 = mass of the moisture dish + sample before being dried

W_3 = mass of the moisture dish + sample after being dried

Protein content

The protein content of the samples was determined according to the AACC Approved Method 46-30 (AACC, 2008) using the Dumas combustion nitrogen analyser (Model Truspec® N Elemental Determinator, supplied by Leco Africa, Kempton Park, South Africa). To ensure that the instrument was performing within specifications, a number of blank samples followed by a number of ethylenediaminetetraacetic acid (EDTA) (Leco Africa, Kempton Park, South Africa) samples were first analysed. EDTA is a chemical standard with a known nitrogen content (9.57%), and is thus used to calibrate the instrument. The EDTA standard (0.05 ± 0.001 g) was weighed into a tin foil sample cup (Leco Africa, Kempton Park, South Africa), twisted closed into a compact ball and placed on the carousel loading head of the instrument. The instrument determines the protein content of the sample from the amount of nitrogen gas produced during combustion, as described in the Truspec® User Manual (Anon., 1994). Similary 0.35 ± 0.001 g of the flour samples were weighed and loaded in the instrument. To convert nitrogen to protein content, a conversion factor of 5.7 was used for both the triticale and wheat flour (AACC method 46-30, AACC; Chawla & Kapoor, 1982; Kulshrestha & Usha, 1992). The protein content was expressed on a 12% moisture basis (mb).

Ash content

The ash content of the samples was determined according to an adapted version of AACC Approved Method 08-02 (AACC, 2008). Porcelain crucibles were dried for 30 minutes at 130°C in an air oven, after which they were allowed to cool in a desiccator. The mass of each crucible was obtained to the nearest 0.001 g and recorded (W_1). Subsequently 5 ± 0.001 g of the flour was weighed into each crucible and the new mass recorded (W_2). Five mL magnesium acetate alcohol (6 g MgAc in 30 mL dH_2O, made up to 400 mL with ethanol) was added to each crucible after which it was set alight and allowed to burn out. The crucibles were placed in a muffle furnace at 700°C for 3 hours where after they were allowed to cool for 45 min in a desiccator. The end product should be a greyish-white ash.

After 45 min, the crucibles were weighed to the nearest 0.001 g and the mass recorded (W_3). The ash content was determined according to equation 3.2.

$$\% \text{ ash} = (W_2 - W_3 / W_2 - W_1) \times 100 \quad \text{…equation 3.2}$$

Where: W_1 = mass of the crucible

W_2 = mass of the crucible + sample before ashing

W_3 = mass of the crucible + sample after ashing

Falling number (α-amylase activity)

The α-amylase activity in the samples was evaluated according to the AACC Approved Method 56-81B (AACC, 2008) using a Shakematic 1095 and a Falling number 1500 apparatus (Perten Instruments AB, Huddinge, Sweden). The amount of flour used for each sample was determined based on a 14% mb from a correction table supplied in the AACC method. The samples were placed in the standardised precision viscometer tube provided with the Falling Number apparatus, together with 25 mL distilled water (at room temperature) and mixed for approximately 5 s using the Shakematic. The viscometer-stirrer was used to scrape down the flour-water mixture from the sides of the tube, after which the tube was placed in the boiling water bath. The apparatus stirred the sample, after which the stirrer was dropped. An indication of the presence of α-amylase activity was obtained from the time in seconds from when stirring starts till the stirrer has dropped through the sample.

Particle size index (PSI)

Hardness determinations were carried out according to the method described by Williams and Sobering (1986). Stainless steel sieves with 75 µm openings, fitted with receiving pans, were used. The weight of each receiving pan was obtained to the nearest 0.001 g and recorded (W_1). Thereafter 10 ± 0.01 g of the ground sample was weighed onto the sieve (W_2), to which 50 g of whole kernels were added to facilitate efficient sieving. This was repeated for a series of four sieves which was stacked and sieved on a Ro-tap percussion sieve shaker (Retsch, Haan, Germany) for 10 min. The fine flour adhering to the bottom of each sieve was carefully brushed off into the respective receiving pans. The receiving pans together with the throughs were weighed correct to the nearest 0.001 g (W_3). The PSI values of the samples were then determined according to equation 3.3.

$$\% \text{ PSI} = [(W_3 - W_1) / W_2] \times 100 \quad \text{…equation 3.3}$$

Where: W_1 = mass of the receiving pan

W_2 = mass of the original sample

W_3 = mass of the receiving pan and throughs

1000-Kernel mass

The 1000-kernel mass was determined according to the industry-accepted method using a Seedburo Count-A-Pak 801 Seed Counter apparatus (Seedburo, Illinois, USA), which makes use of a laser to count the moving whole grain kernels. Every disturbance of the beam is counted as one kernel by the instrument, and the count is electronically displayed on a screen. All broken kernels were manually removed from a 50 g sample. The sample was placed in the instrument which aligns the kernels in a row on a spiral track by vibration. The vibration causes the kernels to move in a single file passed the laser beam. The counted kernels were collected in a receiving container and upon reaching 1000 kernels, the counter stops vibrating to prevent any further kernels from falling into the receiving container. The counted kernels were then weighed to obtain the 1000-kernel mass. The 1000-kernel mass could only be determined for the 2006 samples.

SDS sedimentation

A micro SDS sedimentation test was performed according to an adapted version of AACC Approved Method 56-70 (AACC, 2008) as described by Dick & Quick (1983). One gram of flour was weighed correct to the nearest 0.001 g and placed together with 4 mL distilled water in a glass test tube (150 mm long with an outer diameter of 16 mm and an inner diameter of 14 mm). The test tube was vortexed for 2 s (or until thoroughly mixed), allowed to soak for 5 min and vortexed again for 2 s. After soaking for another 5 min, 12 mL of a stock solution was added to the water and flour suspension. The stock solution was prepared fresh daily by making up a 1:48 ratio of 85% lactic acid (1:8 v/v with water, prepared and left to stand overnight before use) and sodium dodecyl sulphate (2% solution). After the addition of the stock solution, the tube was stoppered, inverted 10 times, placed upright for 10 min and the height of the sediment in the tube measured in mm. Due to the dimensions of the tube, 1 mm was equivalent to 1 cm^3 or 1 mL.

Statistical analysis

Statistical analyses were performed and graphs compiled using STATISTICA version 8.0 (StatSoft, Inc., Tulsa, OK, USA). Two-way analysis of variance (ANOVA) was performed to compare the localities and cultivars, as well as to determine the interactions between localities and cultivars within each year. The locality by cultivar interaction could however only be determined for the 2007 data, due to the fact that only one repetition from each locality could be obtained for the 2006 season. Error bars were used to indicate 0.95% confidence intervals, and least significant difference (LSD) *post-hoc* testing was used.

In addition to the analyses within each year, the results of the two respective harvest seasons were statistically evaluated and compared for all the quality evaluations that were performed, thereby determining the influence of yearly variability and the effect of varying environmental conditions. This was performed for cultivar and locality samples common to both seasons.

Correlation coefficients (r) for protein content with SDS sedimentation and PSI values, respectively, were determined. The wheat samples were excluded for this determination. All statistically significant differences ($P < 0.05$) are referred to as significant differences in the remainder of the chapter.

Results

Protein content

2006 Harvest season

(Detailed results in Appendix 1; Figs. 3.1 & 3.2 in Appendix 3)

Significant differences ($P < 0.05$) were observed between the average protein contents for the cultivars with US2007 and USGen19 having the highest values (Table 3.1). Differences between the averages at the various localities were, however, found not to be significant ($P > 0.05$), with Langgewens and Tygerhoek exhibiting slightly higher average values than the rest (Table 3.2).

2007 Harvest season

(Detailed results in Appendix 2; Figs. 3.3 & 3.4 in Appendix 3)

For the cultivars, the average protein value for the wheat samples was found to be significantly ($P < 0.05$) higher than the averages for the triticale samples (Table 3.3). Riversdal and Tygerhoek had significantly higher average values compared to the rest of the localities ($P < 0.05$) (Table 3.4). A significant interaction ($P < 0.05$) was observed

between the localities and cultivars, with the average for the wheat samples exhibiting the highest values at only half of the localities (Fig. 3.1).

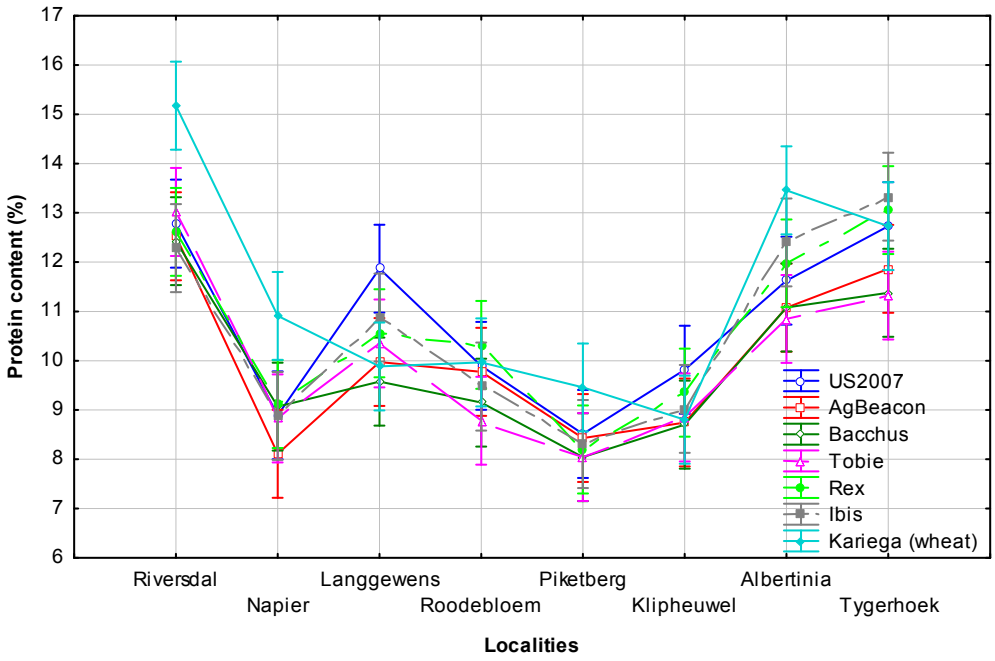


Figure 3.1 Results for locality by cultivar interaction for protein for the 2007 harvest season as obtained with ANOVA (P < 0.05). Error bars denote 0.95 confidence intervals.

Comparisons between the 2006 and 2007 harvest seasons

The average protein content for the 2006 season was significantly higher (*P* < 0.05) than that of the 2007 season, differing by more than 3.5% (results not shown). The differences between the cultivars for the two years did not prove to be significant (P > 0.05) (Fig. 3.2), whereas differences between the averages of localities for the two years was significant (Fig. 3.3).

Table 3.1 Mean values (± standard deviation) and ranges obtained for all parameters for the 2006 season cultivars.

Cultivars	Mean ± standard deviation					
	Protein (%)	Ash (%)	Falling number (seconds)	Particle Size Index	1000 kernel mass (g)	SDS Sedimentation (mm)
US2007	14.7 ± 0.7 **b**[a]	1.97 ± 0.06 **c**	87 ± 29 **b**	64.74 ± 1.22 **ac**	46.65 ± 2.71 **c**	38 ± 2 **b**
USGen19	14.7 ± 0.7 **b**	1.92 ± 0.06 **abc**	196 ± 29 **c**	62.86 ± 1.22 **b**	40.88 ± 2.71 **ab**	33 ± 2 **d**
Bacchus	13.4 ± 0.7 **a**	1.87 ± 0.06 **ab**	99 ± 29 **ab**	69.46 ± 1.22 **d**	39.13 ± 2.71 **a**	30 ± 2 **a**
Tobie	13.2 ± 0.7 **a**	1.96 ± 0.06 **c**	181 ± 29 **c**	66.02 ± 1.22 **a**	42.36 ± 2.71 **ab**	24 ± 2 **c**
Rex	14.0 ± 0.7 **ab**	1.94 ± 0.06 **bc**	161 ± 29 **cd**	64.21 ± 1.22 **bc**	44.37 ± 2.71 **bc**	30 ± 2 **a**
Ibis	14.0 ± 0.7 **ab**	1.85 ± 0.06 **a**	128 ± 29 **ad**	62.97 ± 1.22 **b**	46.64 ± 2.71 **c**	36 ± 2 **b**
Range[b]	12.7 – 16.0	1.53 – 2.19	62 – 285	58.99 – 71.82	34.82 – 55.12	21 – 43

[a] Different letters in each column indicate significant differences ($P < 0.05$)
[b] Range = max - min

Table 3.2 Mean values (± standard deviation) and ranges obtained for all parameters for the 2006 season localities.

Localities	Mean ± standard deviation					
	Protein (%)	Ash (%)	Falling number (seconds)	Particle Size Index	1000 kernel mass (g)	SDS Sedimentation (mm)
Napier	13.9 ± 0.7 **a**[a]	1.96 ± 0.06 **a**	143 ± 29 **ab**	66.18 ± 1.22 **a**	47.18 ± 2.71 **a**	27 ± 2 **a**
Langgewens	14.4 ± 0.7 **a**	1.68 ± 0.06 **b**	156 ± 29 **a**	64.90 ± 1.22 **ab**	44.80 ± 2.71 **ab**	36 ± 2 **b**
Roodebloem	13.7 ± 0.7 **a**	1.89 ± 0.06 **a**	108 ± 29 **bc**	63.53 ± 1.22 **b**	43.48 ± 2.71 **ab**	28 ± 2 **a**
Mariendahl	13.7 ± 0.7 **a**	2.14 ± 0.06 **c**	208 ± 29 **d**	63.76 ± 1.22 **b**	41.20 ± 2.71 **b**	34 ± 2 **bc**
Tygerhoek	14.3 ± 0.7 **a**	2.10 ± 0.06 **c**	71 ± 29 **c**	63.25 ± 1.22 **b**	41.51 ± 2.71 **b**	33 ± 2 **c**
Vredenburg	13.8 ± 0.7 **a**	1.72 ± 0.06 **b**	167 ± 29 **ad**	68.84 ± 1.22 **c**	41.86 ± 2.71 **b**	34 ± 2 **bc**
Range[b]	12.7 – 16.0	1.53 – 2.19	62 – 285	58.99 – 71.82	34.82 – 55.12	21 – 43

[a] Different letters in each column indicate significant differences ($P < 0.05$)
[b] Range = max - min

Table 3.3 Mean values (± standard deviation) and ranges obtained for all parameters for the 2007 season cultivars.

Cultivars	Mean ± standard deviation				
	Protein (%)	Ash (%)	Falling number (seconds)	Particle Size Index	SDS Sedimentation (mm)
US2007	10.8 ± 0.3 **a**[a]	1.81 ± 0.07 **ab**	93 ± 9 **b**	61.52 ± 0.56 **c**	38 ± 2 **b**
AgBeacon	10.1 ± 0.3 **b**	1.91 ± 0.07 **c**	155 ± 9 **a**	60.35 ± 0.56 **a**	28 ± 2 **a**
Bacchus	9.9 ± 0.3 **b**	1.71 ± 0.07 **de**	119 ± 9 **c**	66.64 ± 0.56 **d**	29 ± 2 **a**
Tobie	10.0 ± 0.3 **b**	1.86 ± 0.07 **ac**	234 ± 9 **d**	60.38 ± 0.56 **a**	27 ± 2 **a**
Rex	10.6 ± 0.3 **a**	1.74 ± 0.07 **bd**	162 ± 9 **a**	60.07 ± 0.56 **a**	29 ± 2 **a**
Ibis	10.6 ± 0.3 **a**	1.73 ± 0.07 **bd**	139 ± 9 **e**	58.17 ± 0.56 **b**	35 ± 2 **c**
Kariega (wheat)	11.3 ± 0.3 **c**	1.63 ± 0.07 **e**	439 ± 9 **f**	58.94 ± 0.56 **b**	86 ± 2 **d**
Range[b]	7.5 – 14.2	1.49 – 2.87	63 – 300	50.85 – 76.81	20 – 65

[a] Different letters in each column indicate significant differences ($P < 0.05$)
[b] Range = max - min

Table 3.4 Mean values (± standard deviation) and ranges obtained for all parameters for the 2007 season localities.

Localities	Mean ± standard deviation				
	Protein (%)	Ash (%)	Falling number (seconds)	Particle Size Index	SDS Sedimentation (mm)
Riversdal	12.6 ± 0.4 **a**	1.92 ± 0.08 **b**	135 ± 11 **ab**	60.17 ± 0.65 **ab**	36 ± 2 **a**
Napier	8.8 ± 0.4 **b**	1.69 ± 0.08 **c**	139 ± 11 **a**	63.80 ± 0.65 **d**	29 ± 2 **bc**
Langgewens	10.5 ± 0.4 **d**	1.82 ± 0.08 **b**	114 ± 11 **c**	59.49 ± 0.65 **a**	33 ± 2 **d**
Roodebloem	9.6 ± 0.4 **c**	1.84 ± 0.08 **b**	185 ± 11 **e**	60.75 ± 0.65 **b**	26 ± 2 **e**
Piketberg	8.3 ± 0.4 **e**	1.85 ± 0.08 **b**	121 ± 11 **bc**	70.88 ± 0.65 **c**	30 ± 2 **b**
Klipheuwel	9.1 ± 0.4 **bc**	1.90 ± 0.08 **b**	202 ± 11 **f**	61.92 ± 0.65 **e**	27 ± 2 **ce**
Mariendahl	9.2 ± 0.4 **bc**	2.08 ± 0.08 **a**	138 ± 11 **a**	71.24 ± 0.65 **c**	28 ± 2 **bce**
Albertinia	11.5 ± 0.4 **f**	1.65 ± 0.08 **c**	148 ± 11 **ad**	55.17 ± 0.65 **f**	34 ± 2 **ad**
Tygerhoek	12.3 ± 0.4 **a**	1.67 ± 0.08 **c**	161 ± 11 **a**	57.34 ± 0.65 **g**	34 ± 2 **ad**
Range[b]	7.5 – 14.2	1.49 – 2.87	63 – 300	50.85 – 76.81	20 – 65

[a] Different letters in each column indicate significant differences ($P < 0.05$)
[b] Range = max - min

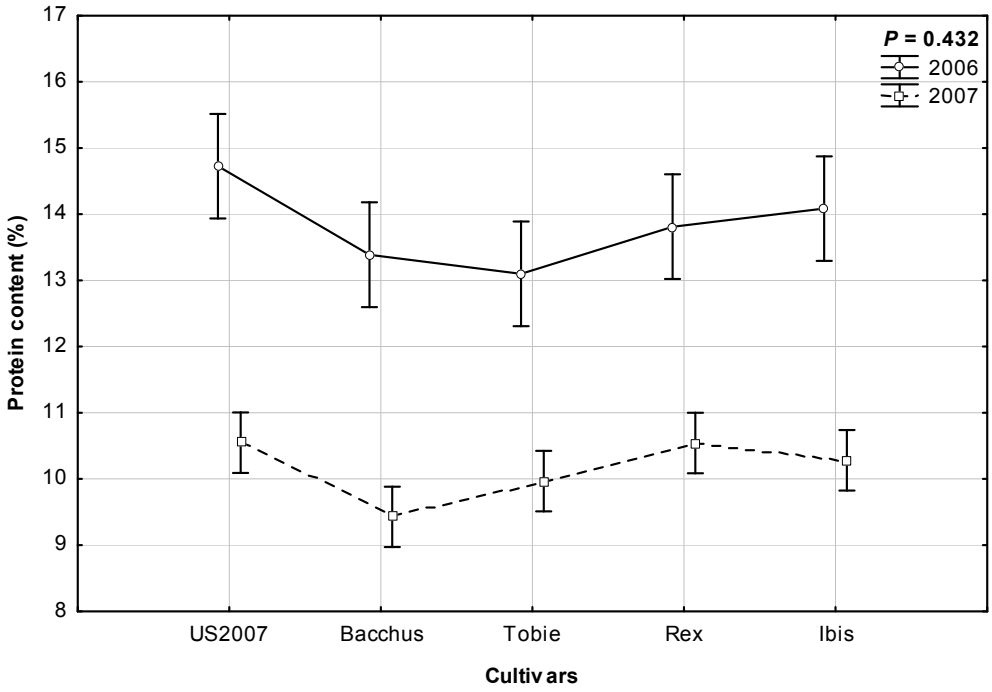


Figure 3.2 Differences between average protein contents obtained for cultivars over the two harvest seasons as determined by ANOVA ($P > 0.05$). Error bars denote 0.95 confidence intervals.

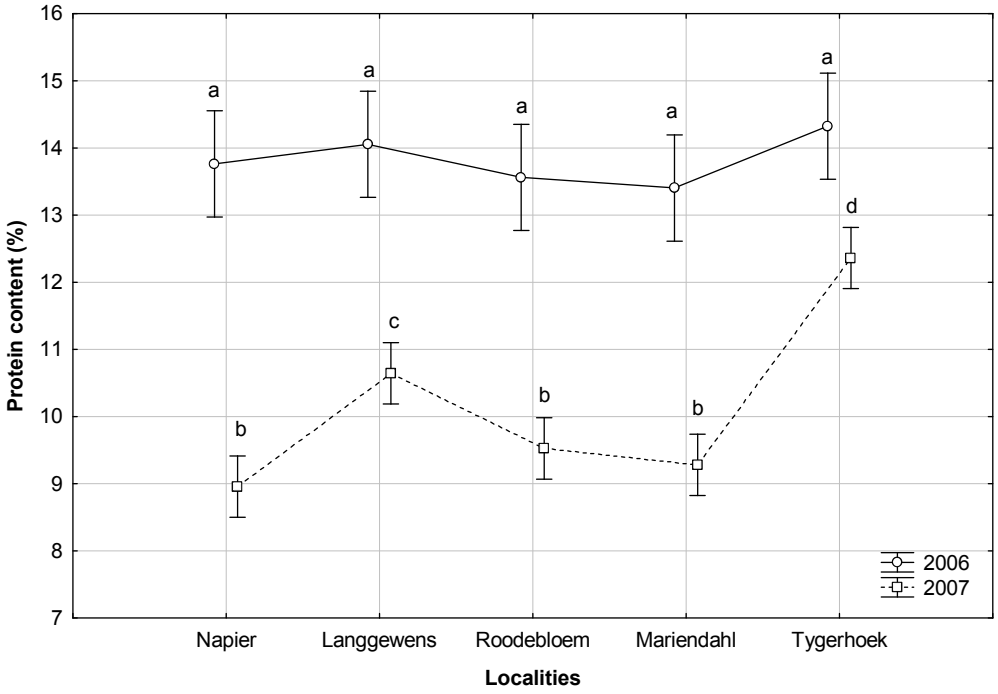


Figure 3.3 Differences between average protein contents obtained for localities over the two harvest seasons as determined by ANOVA ($P < 0.05$). Error bars denote 0.95 confidence intervals and different letters indicate significant differences.

Ash content

2006 harvest season

(Detailed results in Appendix 1; Figs. 3.5 & 3.6 in Appendix 3)

The cultivar averages for ash content differed significantly ($P < 0.05$), with Tobie and US2007 obtaining the highest values (Table 3.1). Significant differences ($P < 0.05$) were also observed between the averages of the localities, with the average ash contents for both Mariendahl and Tygerhoek obtaining significantly higher values (Table 3.2).

2007 harvest season

(Detailed results in Appendix 2; Figs. 3.7 & 3.8 in Appendix 3)

Significant differences ($P < 0.05$) were observed for the average ash contents of both localities and cultivars (Tables 3.3 & 3.4). The average for the wheat samples was found to be the lowest when compared to the triticale cultivars, while the average of the samples from Mariendahl was significantly higher than the average of the rest of the localities. The locality by cultivar interaction was found to be significant ($P < 0.05$) (Fig. 3.4). A similar trend was found to exist for the cultivars at the localities, apart from AgBeacon which differed from the trend for the localities of Roodebloem and Piketberg.

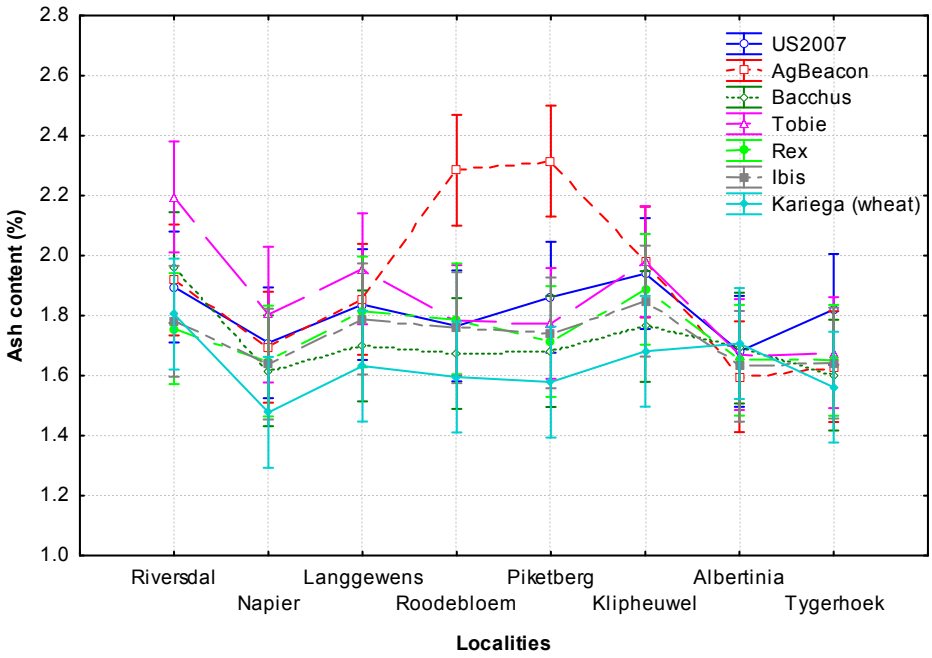


Figure 3.4 Results for locality by cultivar interaction for the 2007 harvest season as obtained with ANOVA (P < 0.05). Error bars denote 0.95 confidence intervals.

Comparisons between the 2006 and 2007 harvest seasons

The average values for the ash contents of the two years differed significantly from each other ($P < 0.05$) (results not shown). Cultivar averages were found not to be significantly different from each other over the two season ($P > 0.05$) (Fig. 3.5), whereas the differences between the averages for localities differed significantly ($P < 0.05$) (Fig 3.6). A similar trend was observed between the two seasons for the cultivar averages.

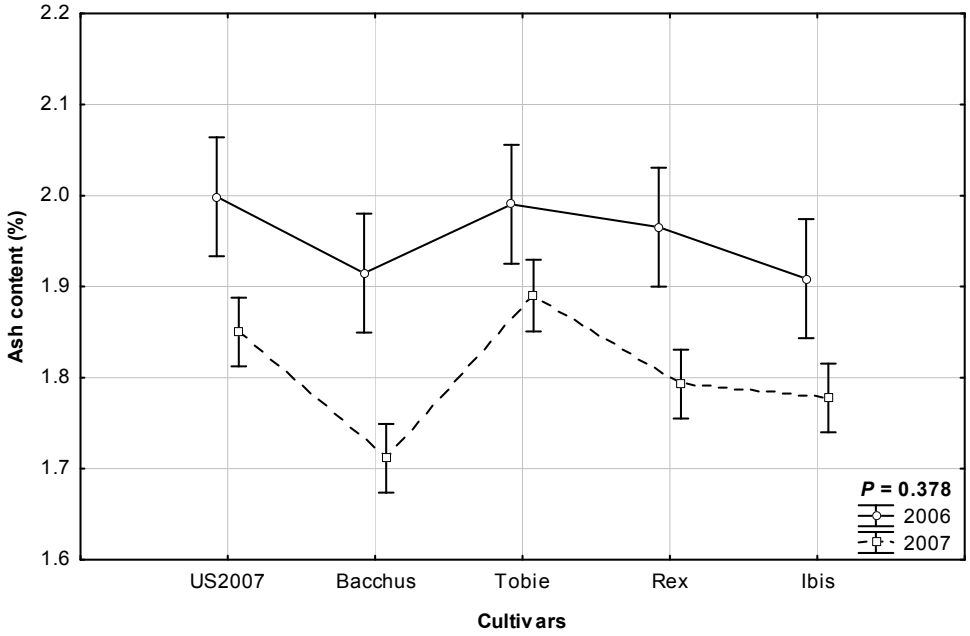


Figure 3.5 Differences between average ash contents obtained for cultivars over the two harvest seasons as determined by ANOVA ($P > 0.05$). Error bars denote 0.95 confidence intervals.

Falling number

2006 harvest season

(Detailed results in Appendix 1; Figs. 3.9 & 3.10 in Appendix 3)

As was expected, the falling number values for triticale were generally very low, with most values below 160 seconds. Significant differences ($P < 0.05$) were observed between the average falling number values for cultivars as well as between the averages for localities (Tables 3.1 & 3.2). In terms of cultivars, USGen19 obtained the best average value with 196 seconds, while US2007 had the lowest average at 87 seconds. For the localities, samples from Mariendahl had the highest falling number, with an average of 208 seconds, while the samples from Tygerhoek had the lowest with an average of 71 seconds.

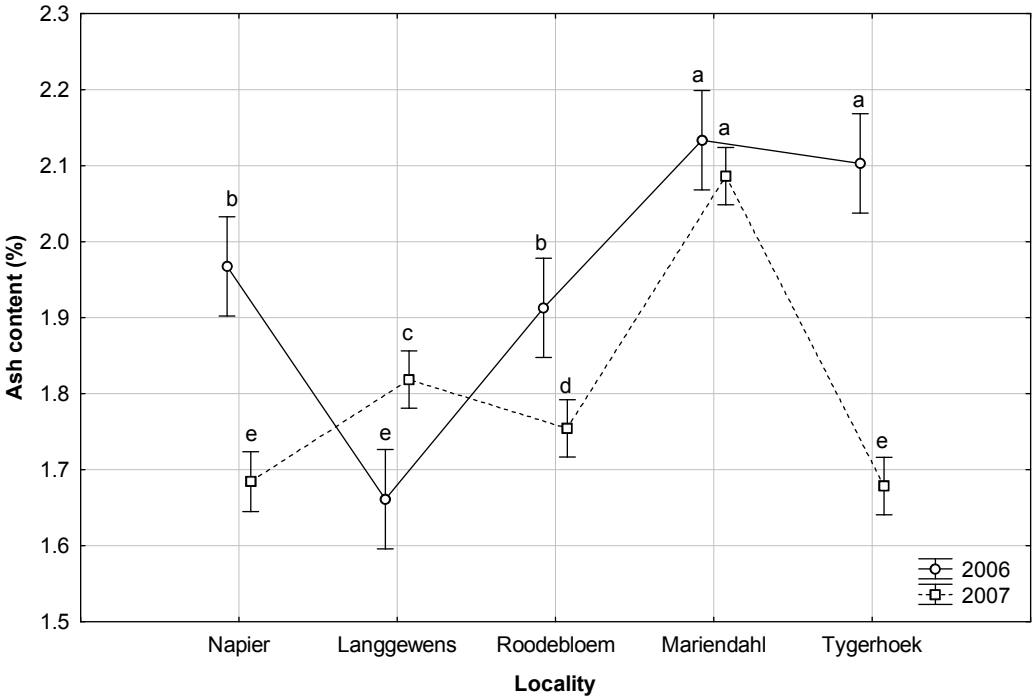


Figure 3.6 Differences between average ash contents obtained for localities over the two harvest seasons as determined by ANOVA ($P < 0.05$). Error bars denote 0.95 confidence intervals and different letters indicate significant differences.

2007 harvest season

(Detailed results in Appendix 2; Figs. 3.11 & 3.12 in Appendix 3)

Similar to the falling number results obtained for the triticale samples harvested during the 2006 harvest season, low falling numbers were generally observed for the 2007 samples. When comparing the averages for the triticale cultivars with the average observed for the wheat samples, it was clear that the triticale samples have very low falling number values (Table 3.3). The average value of the wheat samples differed significantly from that of Tobie (the triticale cultivar with the highest average value) by more than 200 seconds ($P < 0.05$). Tobie also differed significantly from the rest of the triticale samples, with a value of 234 seconds. The average falling number values for Roodebloem and Klipheuwel differed significantly from the other localities' averages ($P < 0.05$) (Table 3.4). The locality by cultivar interaction was significant, and from Fig. 3.7 it is again evident that wheat has a significantly higher falling number value than triticale ($P < 0.05$).

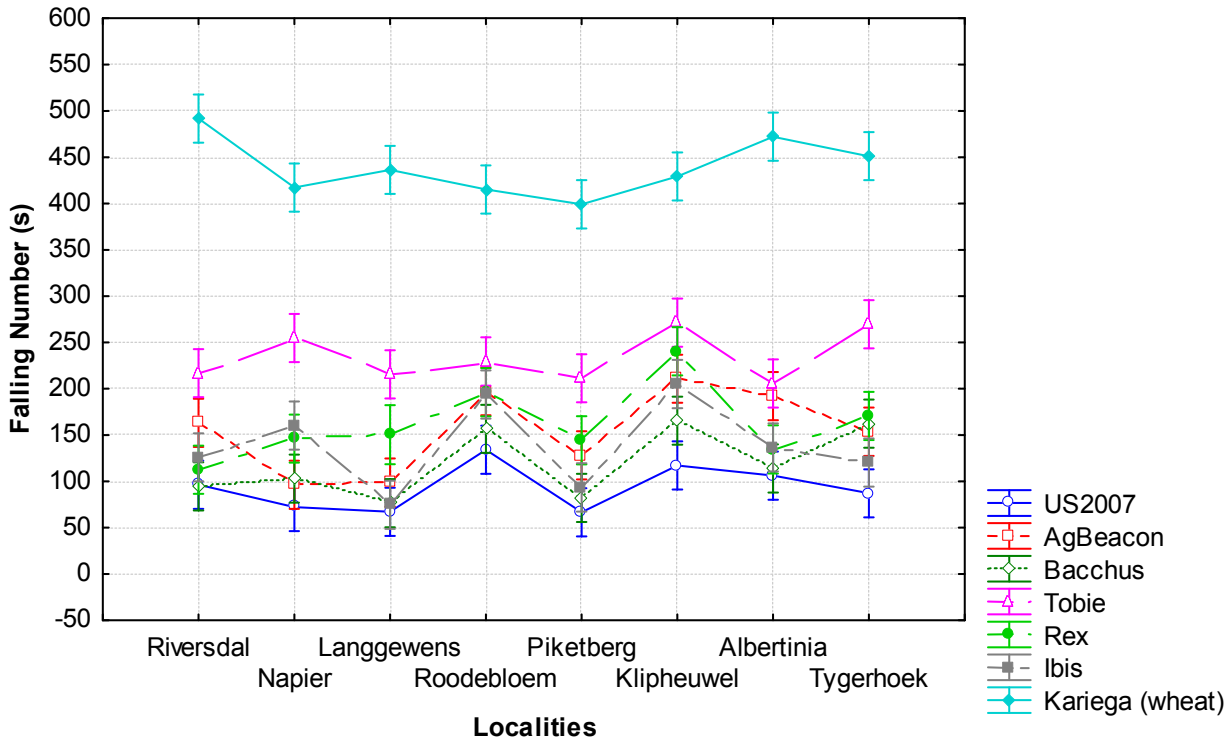


Figure 3.7 Results for locality by cultivar interaction for the 2007 harvest season as obtained with ANOVA (P < 0.05). Error bars denote 0.95 confidence intervals.

Comparisons between the 2006 and 2007 harvest seasons

Falling number values were significantly higher (P < 0.05) in 2007 compared to 2006 (results not shown). Significant differences were observed for both the cultivar and locality averages over the two seasons (P < 0.05) (Figs. 3.8 & 3.9). A similar trend was however observed for the averages of the cultivars over the two years (Fig. 3.8).

Particle size index (PSI)

2006 harvest season

(Detailed results in Appendix 1; Figs. 3.13 & 3.14 in Appendix 3)

The average PSI value for Bacchus was significantly higher than that of the rest of the samples (P < 0.05) with a value of 69.46 (Table 3.1). Significant differences also were found between the average PSI values for localities (P < 0.05) (Table 3.2). The average PSI value for Vredenburg was significantly higher (P < 0.05) than the rest with an average of 68.84.

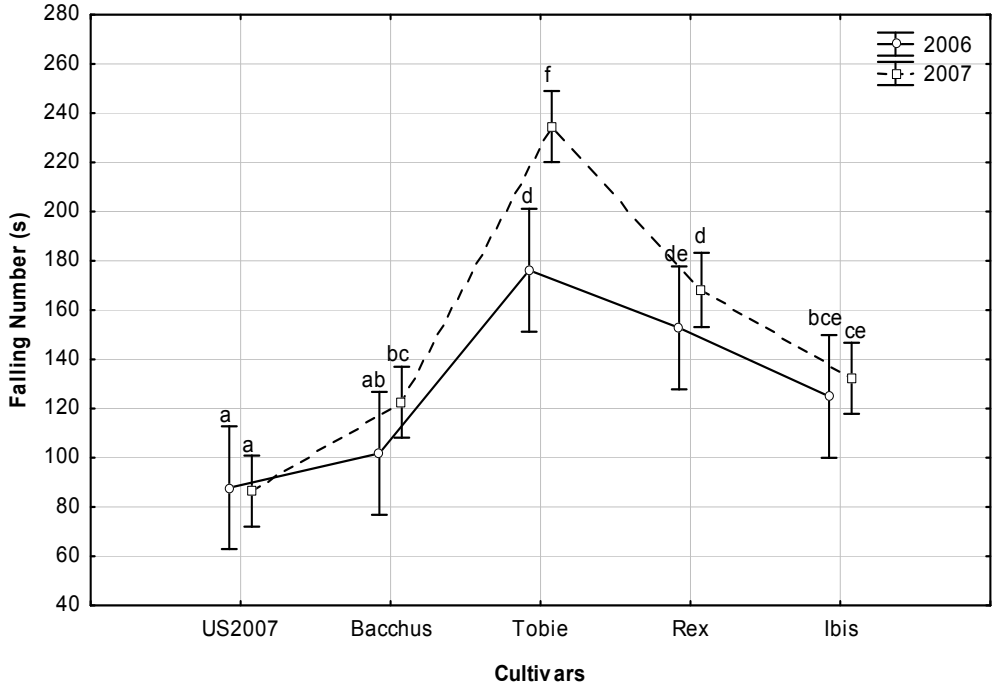


Figure 3.8 Differences between average falling number values obtained for cultivars over the two harvest seasons as determined by ANOVA ($P < 0.05$). Error bars denote 0.95 confidence intervals and different letters indicate significant differences.

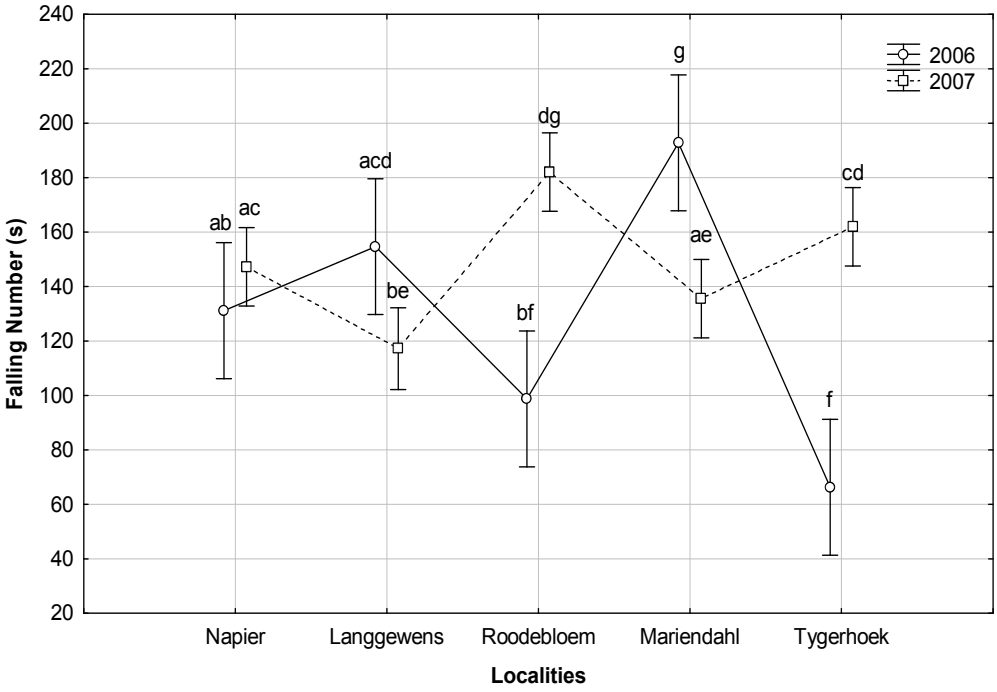


Figure 3.9 Differences between average falling number values obtained for localities over the two harvest seasons as determined by ANOVA ($P < 0.05$). Error bars denote 0.95 confidence intervals and different letters indicate significant differences.

2007 harvest season

(Detailed results in Appendix 2; Figs. 3.15 & 3.16 in Appendix 3)

Bacchus was found to have a significantly higher ($P < 0.05$) average value compared to the rest of the cultivars at 66.64 (Table 3.3). Differences between the averages for localities were found to be significant ($P < 0.05$) (Table 3.4). Five of the six triticale cultivars had significantly higher ($P < 0.05$) average PSI values than the average for the wheat samples, indicating that the triticale cultivars are generally softer than wheat. The locality by cultivar interaction was significant ($P < 0.05$) (Fig. 3.10).

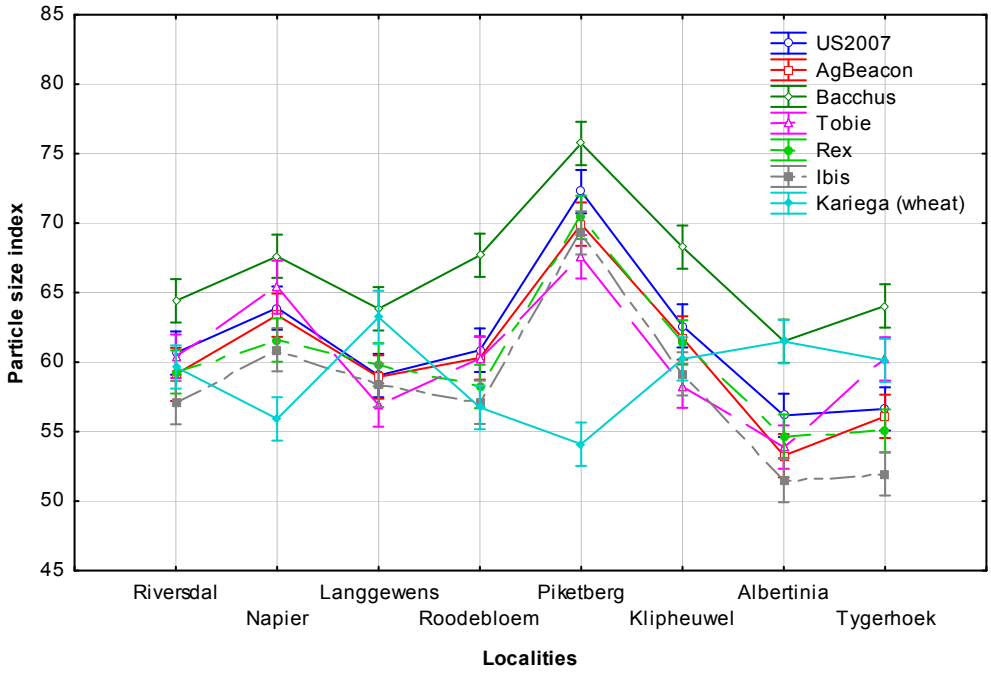


Figure 3.10 Results for locality by cultivar interaction for the 2007 harvest season as obtained with ANOVA. Error bars denote 0.95 confidence intervals.

Comparisons between the 2006 and 2007 harvest seasons

The average PSI values for 2006 were significantly higher than for 2007 ($P < 0.05$) (results not shown). The average values obtained for the cultivars were not significantly different from each other ($P > 0.05$), and very similar averages were observed for the cultivars between the two years (Fig. 3.11), whereas the averages for the localities differed significantly from each other ($P < 0.05$) (Fig. 3.12).

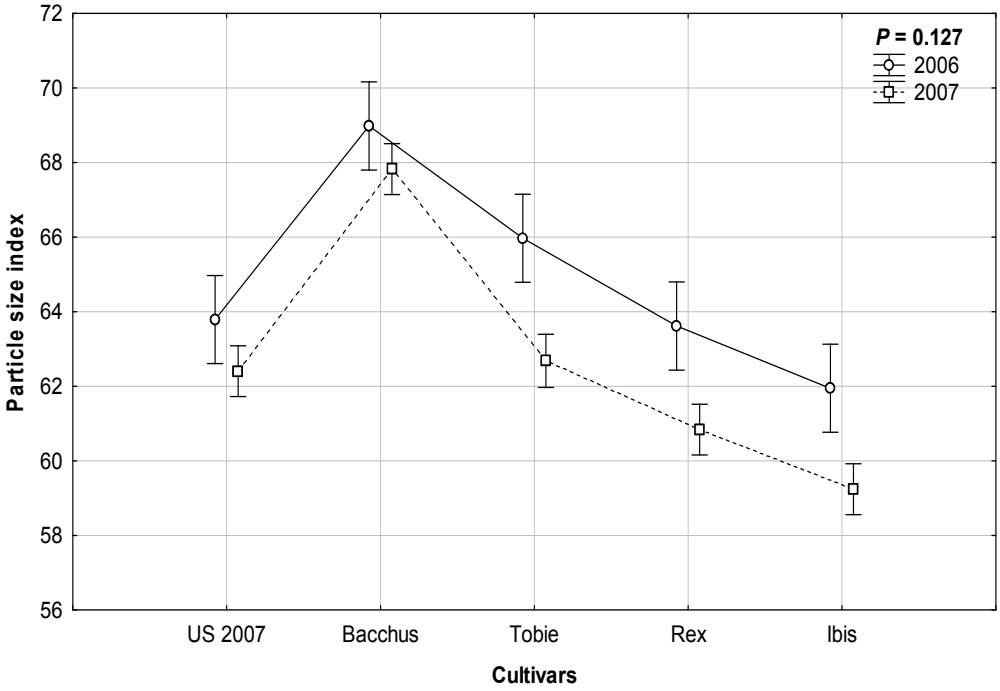


Figure 3.11 Differences between average PSI values obtained for cultivars over the two harvest seasons as determined by ANOVA ($P > 0.05$). Error bars denote 0.95 confidence intervals.

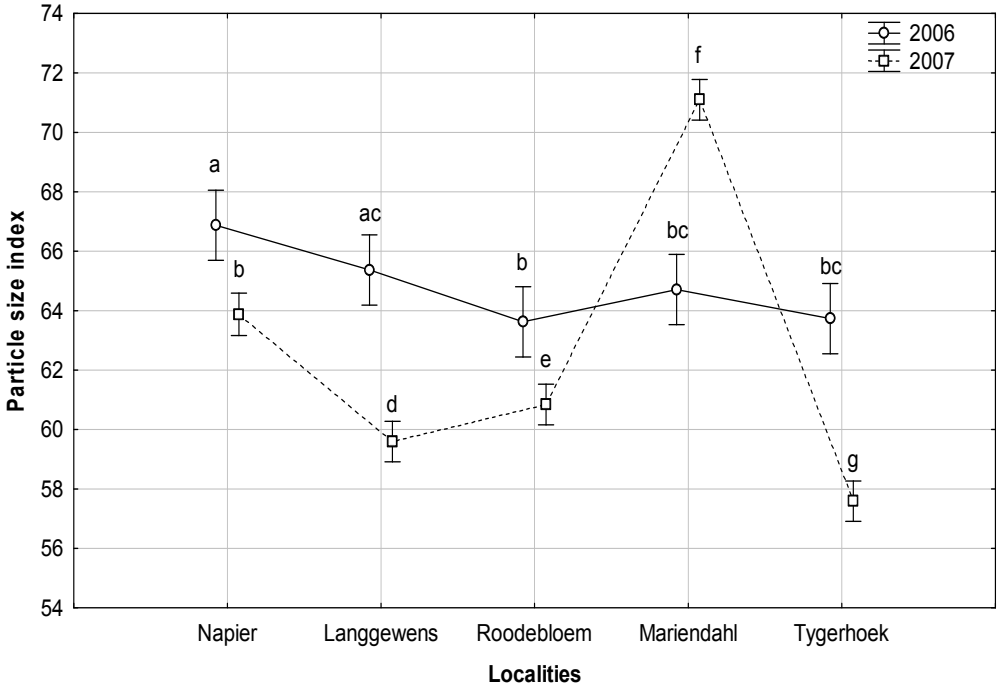


Figure 3.12 Differences between average PSI values obtained for localities over the two harvest seasons as determined by ANOVA ($P < 0.05$). Error bars denote 0.95 confidence intervals and different letters indicate significant differences.

1000-kernel mass

2006 harvest season (only)

(Detailed results in Appendix 1; Figs. 3.17 & 3.18 in Appendix 3)

Significant differences were observed between the averages for both the cultivars and localities ($P < 0.05$) (Tables 3.1 & 3.2), with Ibis and US2007 having the highest value for the cultivars and Napier for the localities.

SDS sedimentation

2006 harvest season

(Detailed results in Appendix 1; Figs. 3.19 & 3.20 in Appendix 3)

The average values observed for SDS sedimentation were very low in comparison to what can be expected for wheat, which is indicative of the weak gluten present in triticale. Significant differences ($P < 0.05$) were observed between the averages for cultivars as well as localities (Tables 3.1 & 3.2).

2007 harvest season

(Detailed results in Appendix 2; Figs 3.21 & 3.22 in Appendix 3)

Differences were observed to be statistically significant for the averages of localities as well as cultivars ($P < 0.05$) (Tables 3.3 & 3.4). From the results it is evident that triticale had significantly lower SDS sedimentation values ($P < 0.05$) compared to the wheat samples, due to wheat having much stronger gluten. The interaction between the cultivars and localities was statistically significant ($P < 0.05$), and Fig. 3.13 again confirms the fact that triticale does not compare well with wheat in terms of gluten content.

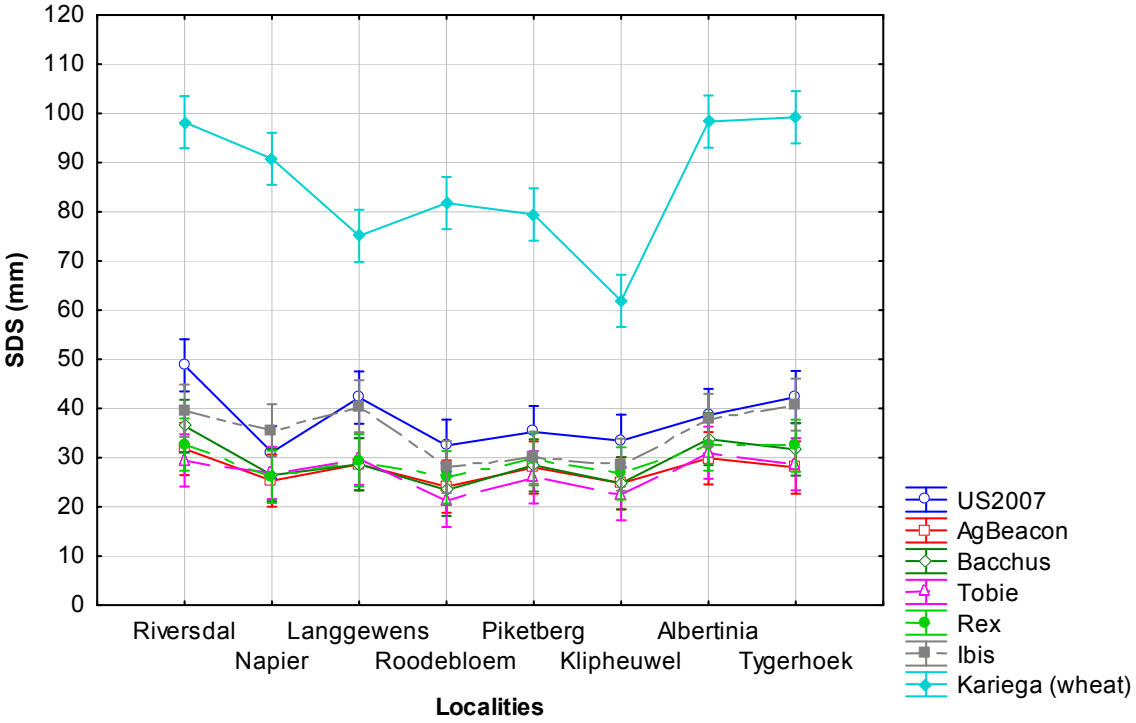


Figure 3.13 Results for locality by cultivar interaction for the 2007 harvest season as obtained with ANOVA (P < 0.05). Error bars denote 0.95 confidence intervals.

Comparisons between the 2006 and 2007 harvest seasons

The average SDS sedimentation values observed for the two seasons were found not to differ significantly from each other (*P* > 0.05). The cultivar averages for the two seasons were observed not to differ significantly (*P* > 0.05), with very similar SDS values being obtained over the two years (Fig. 3.14). No significant differences were found between the locality averages (*P* > 0.05) (Fig 3.15).

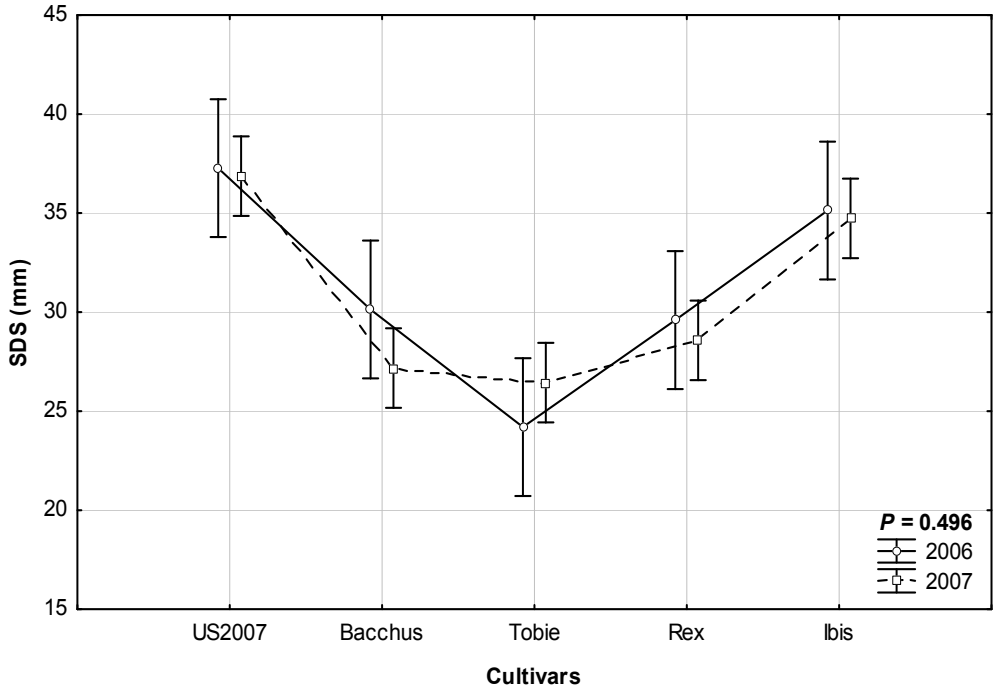


Figure 3.14 Differences between SDS sedimentation values obtained for cultivars over the two harvest seasons as determined by ANOVA ($P > 0.05$). Error bars denote 0.95 confidence intervals.

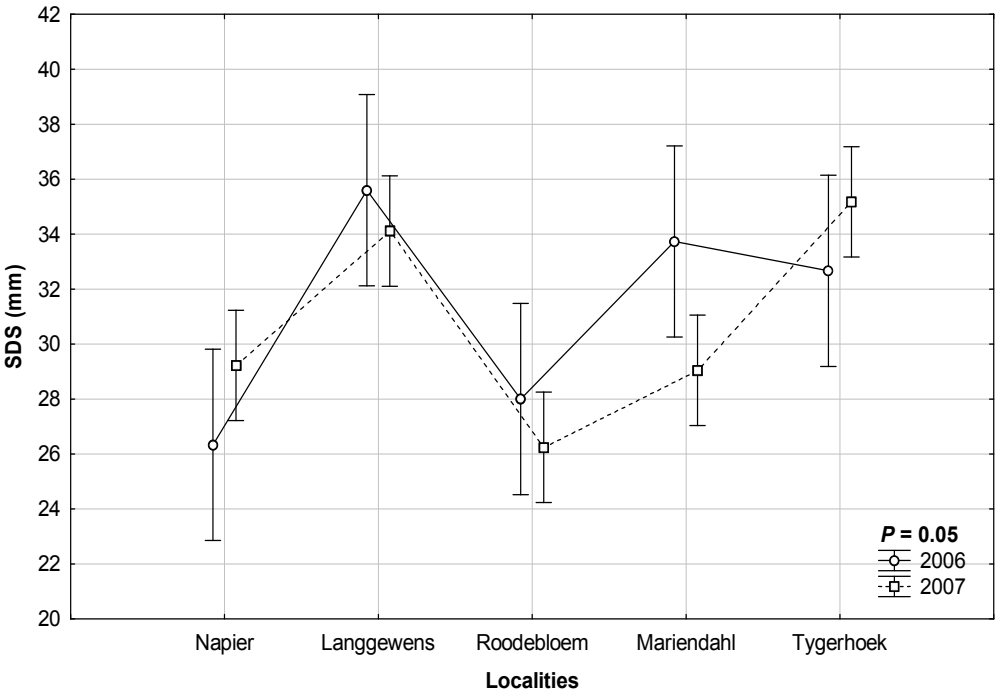


Figure 3.15 Differences between average SDS sedimentation values obtained for localities over the two harvest seasons as determined by ANOVA ($P > 0.05$). Error bars denote 0.95 confidence intervals.

Correlations between protein-SDS sedimentation and protein-PSI

2006 harvest season

A highly significant ($P < 0.001$) correlation ($r = 0.54$) was observed between protein content and SDS sedimentation values of the triticale samples (Fig. 3.16). A significantly ($P < 0.05$) negative correlation ($r = -0.38$) was observed between protein content and PSI values of the samples (Fig. 3.17).

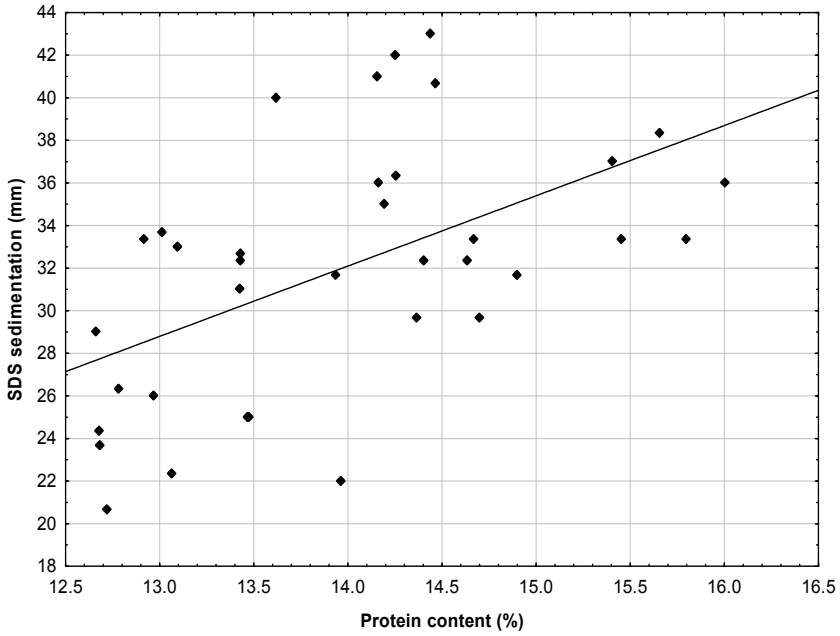


Figure 3.16 Correlation between protein contents and SDS sedimentation values for the 2006 harvest season (r = 0.54, $P < 0.001$).

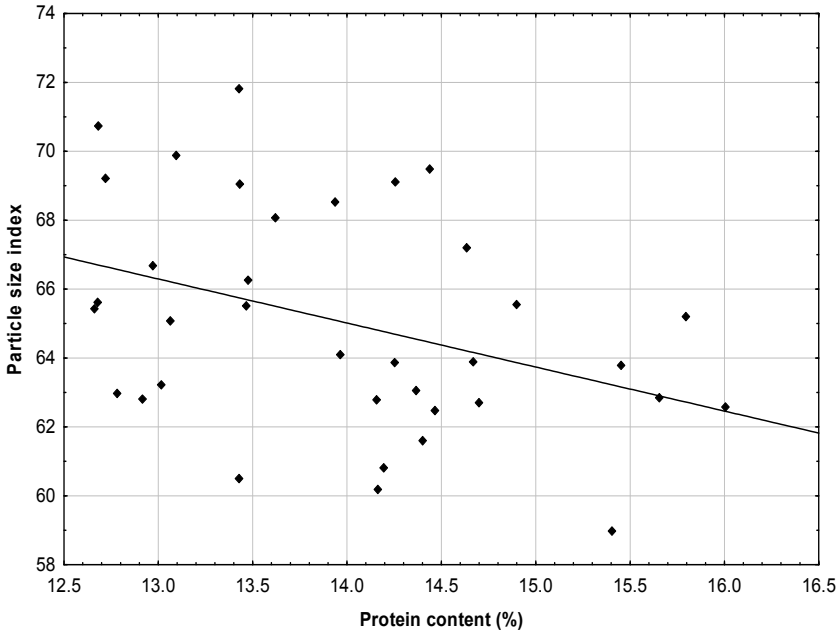


Figure 3.17 Correlation between protein contents and particle size index values for the 2006 harvest season ($r = -0.38$, $P < 0.05$).

2007 harvest season

A highly significant ($P < 0.001$) correlation ($r = 0.58$) (33.6%) was again observed between the protein content and SDS sedimentation values of the triticale samples (Fig. 3.18). A highly significant ($P < 0.001$) negative correlation ($r = -0.64$) (41.0%) between protein content and PSI values was also observed (Fig 3.19).

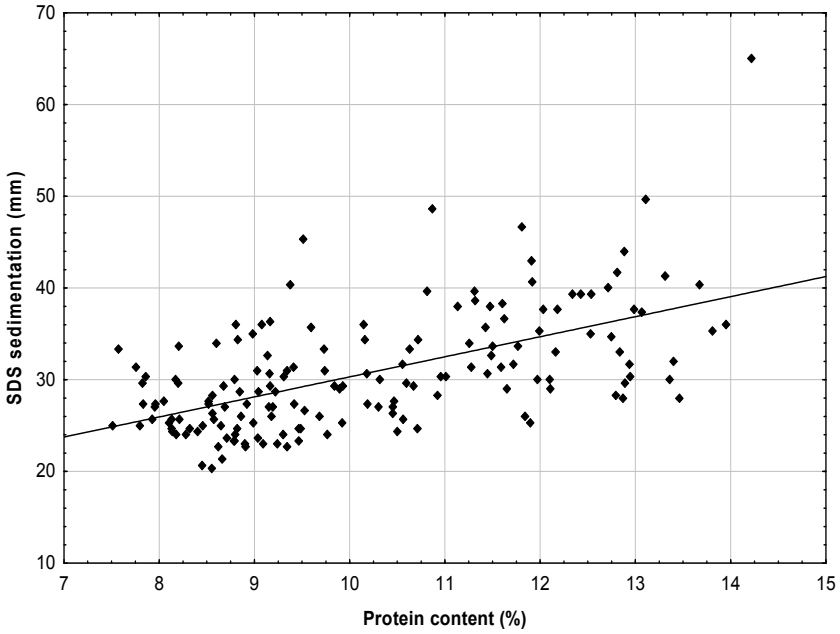


Figure 3.18 Correlation between protein contents and SDS sedimentation values for the 2007 harvest season ($r = 0.58$; $P < 0.001$).

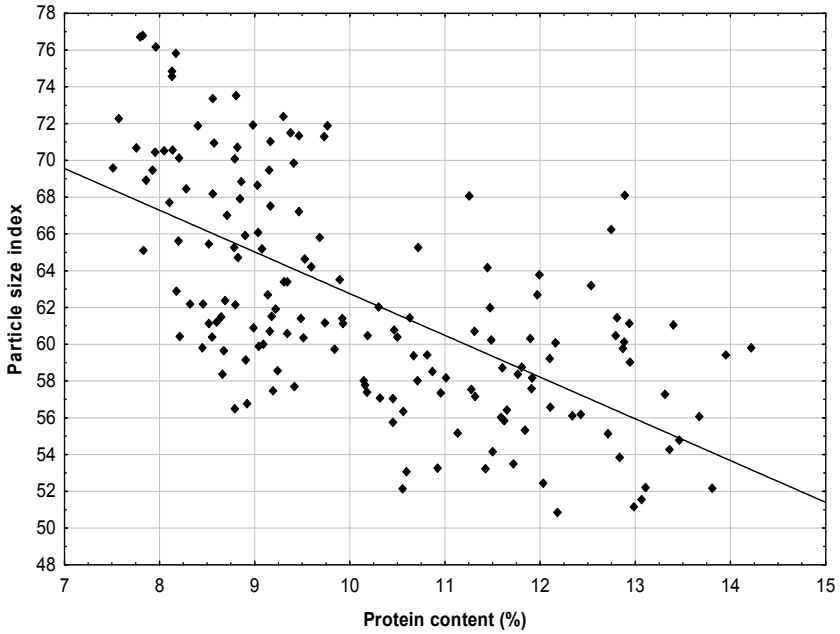


Figure 3.19 Correlation between protein contents and particle size index values for the 2007 harvest season ($r = -0.64$; $P < 0.001$).

Discussion

The compositional and functional quality results obtained for the South African triticale cultivars obtained in this study were similar to previous results in terms of protein content (Bushuk & Larter, 1980; Peña & Bates, 1982; Johnson & Eason, 1988; Heger & Eggum, 1991; Kulshrestha & Usha, 1992; Leon *et al*., 1996; Alaru *et al*., 2003; Roux *et al*., 2006), ash content (Lorenz & Maga, 1972; Leon *et al*., 1996; Seguchi *et al*., 1999; Doxastakis *et al*., 2001), falling number values (Leon *et al*., 1996; Alaru *et al*., 2003; Erekul & Köhn, 2006; Jondreville *et al*., 2007), kernel hardness (Alvarez *et al*., 1992; Martín *et al*., 1999; Ramírez *et al*., 2003; Barrera *et al*., 2007), 1000-kernel mass (Alvarez *et al*., 1992; Erekul & Köhn, 2006; Jondreville *et al*., 2007; Kozak *et al*., 2007) and SDS sedimentation values (Martín *et al*., 1999).

Generally, results obtained for quality determination are largely determined by cultivar and environmental conditions. Various studies have reported on the effect of the environment on the quality of grains (Miezan *et al*., 1977; Lukow & McVetty, 1991; Correll *et al*., 1994; Graybosch *et al*., 1995; Panozzo & Eagles, 1998; Uhlen *et al*., 1998; Dupont *et al*., 2001; Alaru *et al*., 2003). Although these studies did not all agree on whether genotype (cultivar) or environment played the more important role, they do agreed that both genotype and environmental conditions played pivotal roles in the end-quality of grains. The most important factors shown to have an influence on quality are the temperature during seed-filling, in-crop rainfall (winter rainfall for the Western Cape area) and rainfall in the period leading up to harvest (Correll *et al*., 1994; Panozzo & Eagles, 1998; Uhlen *et al*., 1998; Dupont *et al*., 2001).

For protein content, values between 7.5 and 16% were obtained for the triticale cultivars tested in this study; compared to values between 10 and 16% as reported in literature (Bushuk & Larter, 1980; Chawla & Kapoor, 1982; Peña & Bates, 1982; Johnson & Eason, 1988; Heger & Eggum, 1991; Kulshrestha & Usha, 1992; Leon *et al*., 1996; Alaru *et al*., 2003; Roux *et al*., 2006). The averages observed for the triticale cultivars also compared well to the wheat samples, for which an average of 11.3% was obtained. The significant differences observed between cultivars is due to the fact that the cultivars are bred for different purposes. This is illustrated by US2007, a cultivar bred for and used as whole grain animal feed for which a higher protein content is thus desired (Willem Botes, Department of Genetics, Stellenbosch University, Stellenbosch, 2008). For both seasons this cultivar was found to have a significantly higher protein content than the majority of the cultivars. Similarly, AgBeacon was bred for a higher starch (and thus lower protein) content, as it is used for dual feed (silage and whole grain) as well as for the production of

biofuels, for which the higher starch content is important (Willem Botes, Department of Genetics, Stellenbosch University, Stellenbosch, 2008). This cultivar was observed to have a significantly lower protein content than most of the cultivars.

A large and significant difference was observed between the average protein values for the two seasons, with 2006 values ranging from 12.7 – 16%, and 2007 values ranging from 7.5 – 14.2%. Though the averages obtained for the localities differed significantly between the two years ($P < 0.05$), the averages for the cultivars between the two years did not differ significantly ($P \geq 0.05$). This indicates that the reason for the difference could be due to different environmental conditions over the years. Rainfall and temperature data obtained from the ARC, Infruitec-Nietvoorbij (Appendix 4) for most of the localities indicated that 2007 generally received more rainfall than 2006, with higher rainfall figures being recorded especially for the period from May to September of 2007. Of specific interest is the fact that much higher rainfall figures were recorded in 2007 for the two months prior to harvest, which is the main seed-filling period. The average temperature for the two months prior to harvest was also slightly higher for 2006 than for 2007. The higher temperature and lower rainfall before harvest can be the cause of the higher protein contents observed in 2006 (Pfeiffer, 1994; Alaru, 2003).

The ash content values obtained for the triticale samples were also observed within the range described in literature (0.44 – 3.0%) (Lorenz & Maga, 1972; Leon *et al*., 1996; Seguchi *et al*., 1999; Doxastakis *et al*., 2001), with values between 1.49 – 2.87%. Consistent with what is reported in literature, the wheat samples had an average ash content which was in almost all cases significantly lower than that of the triticale samples (Kent & Evers, 1994; Leon *et al*., 1996; Stallknecht *et al*., 1996). Triticale's higher ash content can be detrimental for baking quality (Doxastakis *et al*., 2001), due largely to the negative effect on the colour of the flour obtained (Figoni, 2004).

Falling number values obtained were mostly consistent with, and in some cases higher than values observed in other studies (62 – 180 seconds) (Leon *et al*., 1996; Alaru *et al*., 2003; Erekul & Köhn, 2006; Jondreville *et al*., 2007). Values between 62 – 300 seconds were obtained, which was significantly lower than that obtained for the average of the wheat samples (*ca*. 440 seconds). This is indicative of the very high α-amylase activity of triticale caused by the presence of the rye chromosomes (Tohver *et al*., 2005). The main factor contributing to the falling number value obtained, apart from genetic predisposition, is rainfall during seed-filling and harvest time, with a high rainfall resulting in higher α-amylase activity and, as a consequence, a lower falling number (Leon *et al*., 1996; Alaru *et al*., 2003; Erekul & Köhn, 2006). It would thus be expected that the results for 2007, when

much higher rainfall values were recorded prior to and during harvest time than in 2006, would have much lower falling number values. The opposite was, however, found to be true, with 2007 having a significantly higher average falling number than 2006. The reason for this is not clear, but it is possible that this could be due to more samples from more localities being analysed in 2007 compared to 2006, resulting in a greater range. In addition the maturity of the grain at the time of rainfall could also have had an influence.

For PSI, values between 50.85 and 76.81 were obtained, with 2006 obtaining significantly higher averages than 2007. Five of the six cultivars evaluated for 2007 had average PSI values that were significantly higher than that of the wheat samples evaluated, indicating that the triticale samples were softer, as was expected (Alvarez *et al*., 1992; Martín *et al*., 1999; Ramírez *et al*., 2003; Barrera *et al*., 2007). The average PSI values for cultivars did not differ significantly over the two years, and very similar values were in fact obtained by the cultivars over the two seasons, showing that hardness is mostly influenced by genetic predisposition (Pomeranz & Williams, 1990). The significant differences observed between localities does, however, mean that hardness can, to a lesser extent, be influenced by environmental conditions (Pomeranz & Williams, 1990). Panozzo and Eagles (1998) found that grain tends to become harder with an increase in temperature during the grain-filling period. Due to the significant correlation observed in this study between PSI and protein content (based on the dependence of hardness on the protein composition of a kernel), the differences observed between localities can be expected, as protein content also showed a significant difference between years for localities, and not for cultivars.

According to literature, values for 1000-kernel mass for triticale can be expected to range from 35 – 55 g (Alvarez *et al*., 1992; Erekul & Köhn, 2006; Jondreville *et al*., 2007; Kozak *et al*., 2007), which is very similar to what has been reported for wheat (Alvarez *et al*., 1992; Anon., 2001; Erekul & Köhn, 2006). The results obtained for 1000-kernel mass in this study thus compared well, with values ranging from 34.82 – 55.12 g. Conditions of heat and drought during grain-filling have been found to decrease 1000-kernel mass (Panozzo & Eagles, 1998), whereas cool and moist weather during grain-filling has been found to increase 1000-kernel mass (Erekul & Köhn, 2006). As recorded weather conditions at the localities evaluated during the 2006 season (the only season for which 1000-kernel mass could be evaluated) were not overly dry or hot during seed-filling, the good 1000-kernel mass values which were obtained in this study could be expected. According to Erekul and Köhn (2006), genetic factors play the greatest role in determining 1000-kernel mass, which explains the significant differences observed between cultivars.

Average SDS sedimentation values observed for the localities and cultivars evaluated, ranged from 20 – 65 mm. This is in agreement with an average of 41.2 mL (equivalent to 41.2 mm) obtained for triticale by Martín *et al.* (1999). These values are, however, very low in comparison to the average obtained for the wheat samples (86 mm) in this study. Being indicative of gluten strength, these results are consistent with literature and confirm the known weak gluten quality of triticale (Erekul & Köhn, 2006). The averages for the two years did not differ significantly, implying that environmental conditions do not play such a large role in protein quality (as opposed to protein quantity). A study done by Graybosch *et al*. (1995), however, found the opposite to be true, with their results suggesting that protein quality (as measured by SDS sedimentation) is more sensitive to environmental conditions than protein content. The reason for the non-significant difference between the two years in this study could be the relatively small sample set for the 2006 season.

The significant correlation observed between the protein and SDS sedimentation results in this study, were illustrated by the fact that cultivars mostly obtained the same rank (from highest to lowest) for both protein content and SDS sedimentation values when compared. This is true for both seasons.

Conclusion

South African triticale cultivars are very similar in composition to cultivars from other areas, and generally have a good protein content; comparable to that of wheat. In accordance with what has been observed for triticale elsewhere, the South African cultivars do not perform well with regards to functional quality, though a few cultivars show acceptable results in terms of parameters that are indicative of baking quality (i.e. falling number, PSI and SDS sedimentation).

The genetic differences between cultivars are evident, with cultivars differing significantly for all parameters. The different intended uses for which some of these cultivars were bred can be clearly seen from the results obtained. Furthermore the effect of environment on the cultivars is evident, with significant differences being observed between localities within years as well as over the two harvest seasons. This environmental effect is also illustrated by the significant interaction observed in all cases between localities and cultivars.

References

AACC (2008). *Approved Methods of the American Association of Cereal Chemists*, 10th ed. St. Paul, Minnesota, USA: American Association of Cereal Chemists.

Alaru, M., Laur, Ü. & Jaama, E. (2003). Influence of nitrogen and weather conditions on the grain quality of winter triticale. *Agronomy Research*, **1**, 3-10.

Alvarez, J.B., Ballesteros, J., Sillero, J.A. & Martin, L.M. (1992). Tritordeum: a new crop of potential importance in the food industry. *Hereditas*, **116**, 193-197.

Ammar, K., Mergoum, M., Rajaram, S. (2004). The history and evolution of triticale. In: *Triticale Improvement and Production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 1-9. Rome: Food and Agriculture Organisation of the United Nations.

Anonymous (2001). Market reports – harvest report – September 26, 2001. US Wheat Associates. [WWW document]. URL http://www.uswheat.org/harvestReports/doc. 19 November 2008.

Anonymous (2004). TruSpec N Nitrogen Determinator, Instruction Manual. December, 2004.

Barrera, G.N., Pérez, G.T., Ribotta, P.D. & León, A.E. (2007). Influence of damaged starch on cookie and bread-making quality. *European Food Research and Technology*, **225**, 1-7.

Bushuk, W. & Larter, E.N. (1980). Triticale: production, chemistry, and technology. In: *Advances in cereal science and technology, Vol 3* (edited by Y. Pomeranz). Pp. 115-157. St. Paul, Minnesota, USA: American Association of Cereal Chemists. (as cited by Peña, 2004).

Chawla, V.K. & Kapoor, A.C. (1982). Chemical composition and protein quality of wheat-triticale chapatis. *Journal of Food Science*, **47**, 2015-2017.

Correll, R., Butler, J., Spouncer, L. & Wrigley, C. (1994). The relationship between grain-protein content of wheat and barley and temperatures during grain filling. *Australian Journal of Plant Physiology*, **21**, 869-873.

D'Egidio, M.G., Nardi, S. & Vallega, V. (1993). Grain, flour, and dough characteristics of selected strains of diploid wheat, *Triticum monococcum* L. *Cereal Chemistry*, **17**(3), 298-303.

Dick, J.W. & Quick, J.S. (1983). A modified screening test for rapid estimation of gluten strength in early-generation durum wheat breeding lines. *Cereal Chemistry*, **60**(4), 315-318.

Doxastakis, G., Zafiriadis, I., Irakli, M., Marlani, H. & Tananaki, C. (2002). Lupin, soya and triticale addition to wheat flour doughs and their effect on rheological properties. *Food Chemistry*, **77**(2), 219-227.

Dupont, F.M., Altenbach, S.B., Chung, O.K., Chan, R. & Lopez, R. (2001). Positive effects of growing environment on wheat protein content and breadmaking quality. 2001 AACC annual meeting. [WWW document]. URL http://www.aaccnet.org/meetings/2001/Abstracts/a01ma436.htm. 19 November 2008.

Erekul, O. & Köhn, W. (2006). Effect of weather and soil conditions on yield components and bread-making quality of winter wheat (*Triticum aestivum* L.) and winter triticale (*Triticosecale* Wittm.) varieties in north-east Germany. *Journal of Agronomy and Crop Science*, **192**, 452-464.

Figoni, P. (2004). *How baking works*. Pp 67, 75. Hoboken, New Jersey: John Wiley & Sons, Inc.

Graybosch, R.A., Peterson, C.J., Baenziger, P.S. & Shelton, D.R. (1995). Environmental modification of hard red winter wheat flour protein composition. *Journal of Cereal Science*, **22**, 45-51.

Heger, J. & Eggum, B.O. (1991). The nutritional values of some high-yielding cultivars of triticale. *Journal of Cereal Science*, **14**, 63-71.

Hunter, A.S., & Stanford, G. (1973). Protein content of winter wheat in relation to rate and time of nitrogen fertilizer application. *Agronomy Journal*, **65**, 772-774.

Jestin, L. & Bonhomme, H. (1996). Genetic variation, G*E interactions and selection response for Hagberg Falling Number in triticale. In: *Triticale: today and tomorrow* (edited by H. Guedes-Pinto, N. Darvey, V.P. Carnide). Pp. 609-613. Dordrecht, The Netherlands: Kluwer Academic Press.

Johnson, R. & Eason, P. (1988). Evaluation of triticale for use in diets for meat-type chickens. *Journal of the Science of Food and Agriculture*, **42**, 95-108.

Jondreville, C., Genthon, C., Bouguennec, A., Carre, B. & Nys, Y. (2007). Characterisation of European varieties of triticale with special emphasis on the ability of plant phytase to improve phytase phosphorus availability to chickens. *British Poultry Science*, **48**(6), 678-689.

Kent, N.L. & Evers, A.D. (1994). *Kent's Technology of Cereals: an introduction for students of Food Science and Agriculture*. Pp. 17-18, 50, 96-97, 157. New York, USA: Elsevier Science Ltd.

Kozak, M., Samborski, S., Rozbicki, J. & Mądry, W. (2007). Winter triticale grain yield, a comparitive study of 15 genotypes. *Soil and Plant Science*, **57**, 263-270.

Kulshrestha, K. & Usha, M.S. (1992). Biochemical composition and nutritional quality of triticale. *Journal of Food Science and Technology*, **29**(2), 109-110.

Leon, A.E., Rubiolo, A. & Anon, M.C. (1996). Use of triticale flours in cookies: quality factors. *Cereal Chemistry*, **71**(6), 779-784.

Lorenz, K. & Maga, J. (1972). Triticale and wheat flour studies: compositions of fatty acids, carbonyls, and hydrocarbons. *Journal of Agricultural and Food Chemistry*, **20**(4), 769-772.

Lukow, O.M. & McVetty, P.B.E. (1991). Effect of cultivar and environment on quality characteristics of spring wheat. *Cereal Chemistry*, **68**(6), 597-601.

Martín, A., Alvarez, J.B., Martín, L.M., Barro, F. & Ballesteros, J. (1999). The development of Tritordeum: a novel cereal for food processing. *Journal of Cereal Science*, **30**, 85-95.

Miezan, K., Heyne, E.G. & Finney, K.F. (1977). Genetic and environmental effects on the grain protein content in wheat. *Crop Science*, **17**, 591-593.

Moonen, J.H.E., Graveland, A. & Graveland, A. (1982). Use of the SDS-sedimentation test and SDS-polyacrylamidegel electrophoresis for screening breeder's samples of wheat for bread-making quality. *Euphytica*, **31**, 677-690.

Panozzo, J.F. & Eagles, H.A. (1998). Cultivar and environmental effects on quality characters in wheat. I. Starch. *Australian Journal of Agricultural Research*, **49**, 757-766.

Peña, R.J. (2004). Food uses of triticale. In: *Triticale Improvement and Production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 37-48. Rome: Food and Agriculture Organisation of the United Nations.

Peña, R.J. & Bates, L.S. (1982). Grain shrivelling in secondary hexaploid triticales. I. Alpha-amylase activity and carbohydrate content of mature and developing grains. *Cereal Chemistry*, **59**, 454-458.

Pfeiffer, W.H. (1994). Triticale improvement strategies at CIMMYT: Existing genetic variability and its implication to projected genetic advance. In: Proceedings of the 5th Portuguese Triticale Conference (as cited by Alaru *et al*., 2003).

Pomeranz, Y. & Williams, P.C. (1990). Wheat hardness: its genetic, structural, and biochemical background, measurement, and significance. In: *Advances in Cereal Science and Technology*, *Vol. 10* (edited by Y. Pomeranz). Pp. 471-557. St Paul, Minnesota: American Association of Cereal Chemists.

Ramírez, A., Pérez, G.T., Ribotta, P.D. & León, A.E. (2003). The occurrence of friabilins in triticale and their relationship with grain hardness and baking quality. *Journal of Agricultural and Food Chemistry*, **51**, 7176-7181.

Roux, H.S., Marais, G.F., Snyman, J.E. & Botes, W.C. (2006). The South African triticale breeding programme: current status. In: Proceedings of the 6th International Triticale Symposium, Stellenbosch, South Africa, pp. 80-84.

Salmon, D.F., Mergoum, M. & Gómez-Macpherson, H. (2004). Triticale production and management. In: *Triticale Improvement and Production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp 27-34. Rome: Food and Agriculture Organisation of the United Nations.

Seguchi, M., Ishihara, C., Yoshino, Y., Nakatsuka, K. & Yoshihira, T. (1999). Breadmaking properties of triticale flour with wheat flour and relationship to amylase activity. *Journal of Food Science*, **64**(4), 582-586.

Stallknecht, G.F., Gilbertson, K.M., Ranney, J.E. (1996). Alternative wheat cereals as food grains: einkorn, emmer, spelt, kamut, and triticale. In: *Progress in new crops* (edited by J. Janick). Pp. 156-170. Alexandria, Virginia, USA: ASHS Press.

Tohver, M., Kann, A., Täht, R., Mihhalevski, A. & Hakman, J. (2005). Quality of triticale cultivars suitable for growing and bread-making in northern conditions. *Food Chemistry*, **89**, 125-132.

Uhlen, A.K., Hafskjold, R., Kalhovd, A.-H., Sahlström, S., Longva, Å. & Magnus, E.M. (1998). Effects of cultivar and temperature during grain filling on wheat protein content, composition, and dough mixing properties. *Cereal Chemistry*, **75**(4), 460-465.

Williams, P.C. & Sobering, D.C. (1986) Attempts at standardization of hardness testing of wheat. I. The grinding/sieving (paticle size index) method. *Cereal Foods World*, **31**(5), 359-364.

Wu, Y.V., Sexson, K.R. & Wall, J.S. (1976). Triticale protein concentrate: preparation, composition, and properties. *Journal of Agricultural and Food Chemistry*, **24**(3), 511-517.

Wu, Y.V., Stringfellow, A.C., Anderson, R.A., Sexson, K.R. & Wall, J.S. (1978). Triticale for food uses. *Journal of Agricultural and Food Chemistry*, **26**(5), 1039-1048.

CHAPTER 4

Near infrared (NIR) spectroscopy calibration models for the prediction of moisture, protein and ash contents, kernel hardness and baking potential of South African triticale (X *Triticosecale* Whittmack) cultivars

CHAPTER 4

NEAR INFRARED (NIR) SPECTROSCOPY CALIBRATION MODELS FOR THE PREDICTION OF MOISTURE, PROTEIN AND ASH CONTENTS, KERNEL HARDNESS AND BAKING POTENTIAL OF SOUTH AFRICAN TRITICALE (X *TRITICOSECALE* WHITTMACK) CULTIVARS

Abstract

Near infrared (NIR) spectroscopy was applied to develop calibration models for the prediction of moisture, protein and ash contents, as well as kernel hardness and baking potential of South African triticale (X *Triticosecale* Whittmack) cultivars (n = 198). This was done in diffuse reflectance mode for both triticale flour and wholegrain using two different instruments and software packages. Spectra were obtained with the Büchi NIRFlex N-500 as well as the Bruker MPA Fourier transform NIR (FT–NIR) spectrophotometer, respectively, using The Unscrambler and OPUS software packages for model development. Full cross-validations were performed for all parameters, after which the best regression models obtained (R^2 > 0.66) were validated using an independent test set (n=50). Good prediction results were obtained with flour for moisture (Bruker: *SE*P = 0.08; R^2 = 0.95; *RPD* = 4.65) and protein (Büchi: *SEP* = 0.44; R^2 = 0.96; *RPD* = 5.23; Bruker: *SE*P = 0.32; R^2 = 0.96; *RPD* = 4.88). For whole grain, acceptable results were obtained for protein (Büchi: *SEP* = 0.55; R^2 = 0.94; *RPD* = 4.18; Bruker: *SE*P = 0.70; R^2 = 0.90; *RPD* = 3.23). Calibration models developed for ash content, kernel hardness and baking potential (SDS sedimentation) needs further investigation and improvements might be obtained by extending the range of the data sets, confirming the accuracy of the reference methods and/or applying variable selection methods.

Introduction

The evaluation of quality is of paramount importance in the agricultural industry, both during breeding and commercial production phases. In the cereal grain industry specifically, quality parameters such as moisture and protein contents need to be assessed and reported upon receival at the silos in order to assign deliveries to a grade, to innumerate producers and to determine ratios in which to blend grains to meet specific requirements (Osborne, 2000). When breeding new cereal cultivars, there is a need to determine the presence or level of key processing and quality characteristics during the screening of early generations, with the desired characteristics depending on the proposed

end-use of the cultivar (Osborne, 2000). Traditional methods for determining quality parameters such as protein, moisture and ash contents, as well as methods for obtaining indications of hardness and potential baking quality are time-consuming, expensive, destructive and cumbersome, and might require large amounts of sample. These tests are not suited for rapid analyses at receiving silos, nor for determining quality in early generation breeding lines, when little sample is usually available (Osborne, 2000). The need thus exists for methods of testing that are rapid and economical.

During the 1970's the Federal Grain Inspection Service (FGIS) of the United States of America recognised the potential of near infrared (NIR) spectroscopy in determining the quality of wheat shipments (Butler, 1983; Pasquini, 2003). NIR spectroscopy was found to pose the advantages of being an analytical method that was fast, cheap (lower cost per test), non-invasive, non-destructive, required minimal sample preparation, generated no hazardous remains, and was virtually universally applicable (Butler, 1983; Osborne, 2000; Pasquini, 2003). Furthermore, it performed well when being operated by relatively unskilled labourers working under extreme environments in terms of sample throughput and temperature (Osborne, 2000). The FGIS purchased NIR spectroscopy instruments for the quality evaluation of wheat at their export locations in January 1979 (Butler, 1983).

At the present time, NIR is still the only method of quality evaluation available that is rapid and adequately affordable for widespread implementation in breeding programs, crop management and at receival points. Negative aspects of this technology, however, include that there can be a high cost involved in generating the initial calibrations, as well as for maintaining the calibrations and for employing trained personnel to set up and update these calibrations.

NIR applications are based on the empirical relationship between reference analytical data, obtained with conventional analytical methods, and spectral data, obtained by scanning a sample, to acquire quantitative and/or qualitative information originating from the interaction between the NIR electromagnetic waves and the constituents of the sample (Osborne, 1987; Osborne et al., 1993). The NIR wavelength range covers 750 to 2500 nm (Butler, 1983).

Various studies have been carried out on the determination of grain quality characteristics by making use of NIR spectroscopy, in particular for protein (Osborne & Fearn, 1983; Shenk *et al*., 1985; Delwiche, 1998; Manley *et al*., 2002) and moisture content (Osborne & Fearn, 1983; Law & Tkachuk, 1977; Osborne, 1987; Manley *et al*., 2002), as well as for hardness determination (Osborne & Fearn, 1983; Williams &

Sobering, 1986; Norris *et al*., 1989; Osborne, 1991; Manley *et al*., 2002). Both whole grain and ground flour samples can be analysed (Osborne, 2000).

Triticale (X *Triticosecale* Wittmack), a cross between durum wheat and rye, has a very similar composition to that of common wheat, yet studies making use of wheat NIR spectroscopy models to make predictions for triticale have proven to be unsuccessful (Igne *et al*., 2007a). Only a handful of studies can be found in literature regarding the development of NIR calibration models specifically for triticale and only for moisture and protein (Viljoen *et al*., 2005; Igne *et al*, 2007a; Igne *et al*., 2007b) and ash (Viljoen *et al*., 2005) contents. With the increasing importance of this crop in the food and animal feed industries, it is becoming necessary to develop and make use of dedicated NIR calibration models for the evaluation and determination of triticale quality parameters.

The objective of this study was thus to develop NIR prediction models for South African triticale cultivars for the prediction of protein, moisture and ash contents, as well as for grain hardness and baking quality (as expressed by SDS sedimentation). This was done for both flour and whole grain samples using two different NIR instruments and software packages.

Materials and methods

Triticale samples and sample preparation

The samples analysed comprised the samples evaluated in Chapter 3. Thus six cultivars (US2007, USGen19, Bacchus, Tobie, Rex and Ibis) from each of six localities (Langgewens, Napier, Roodebloem, Mariendahl, Tygerhoek and Vredenburg, one replicate from each locality), harvested during the 2006 season (n = 36) were evaluated, as well as three replicates of six cultivars (US2007, AgBeacon, Bacchus, Tobie, Rex and Ibis), from each of nine localities (Langgewens, Napier, Roodebloem, Mariendahl, Tygerhoek, Riversdal, Piketberg, Klipheuwel and Albertinia) harvested during the 2007 season (n = 162). The wheat samples were excluded for the development of calibration models. All grain samples were kindly supplied by the Department of Genetics, Stellenbosch University, Stellenbosch. The whole grain samples were stored at 4°C until being milled. The samples were milled on a UDY Cyclone mill (UDY Corporation, Fort Collins, Colorado, USA) fitted with a 1 mm sieve. The flour samples were kept in airtight containers at room temperature until being analysed.

Triticale quality evaluation (reference data)

Results as obtained for the analyses performed in Chapter 3 for moisture, protein and ash contents, as well as for particle size index (PSI) and SDS sedimentation were used as the reference data for the NIR calibrations. Methods for obtaining the reference data were as described in Pp 34 – 37 of Chapter 3.

Near infrared spectroscopy measurements (spectral data)

Spectra were obtained for both whole grain and flour samples using a Büchi NIRFlex N-500 Fourier transform NIR (FT-NIR) spectrophometer (Büchi Labortechnik AG, Flawil, Switzerland) as well as a Bruker MPA FT-NIR spectrophometer (Bruker Optics GmbH, Germany).

Spectra were obtained with the Büchi NIRFlex N-500 in diffuse reflectance mode from 1000 to 2500 nm with a total of 1501 data points. A resolution of 16 cm^{-1} was used. Whole grain samples were presented to the instrument in glass petri dishes, and ground flour samples in clear sepcap glass vials (National Scientific, Rockwood, TN, USA).

The Bruker MPA was used to obtain diffuse reflectance spectra from 1000 – 2500 nm with a resolution of 2 cm^{-1} with 1501 data points. The whole grain samples were presented to the instrument in the instrument's solid cell, while the ground flour samples were scanned with a hand-held fibre optics probe.

NIR spectroscopy calibration model development

Partial least squares (PLS) regression models were developed for each quality parameter from the spectral data obtained with the Büchi NIRFlex N-500 using The Unscrambler (Version 9.2, CAMO, Oslo, Norway) software package. For the Bruker MPA spectral data, PLS regression models were developed with the OPUS (Version 6.5, Bruker Optics GmbH, Germany) software package. The software packages offered slightly differing pretreatment options, thus not all the same pretreatments could be applied to both sets of spectral data. The pretreatments tested for each instrument/software package combination are summarised in Table 4.1.

All calibration models developed were evaluated by means of full cross-validation, whereafter the models with the best results (only those with $R^2 > 0.66$) (Williams, 2001) for the different parameters were validated by means of an independent validation set. An identical independent validation set (n = 50) was chosen by selecting every third value from a list of ascending values for protein content. In order to facilitate comparisons, this

set was used for the other parameters as well. The remaining samples then made up the calibration set. Only spectral outliers were removed.

The accuracy of each calibration model was determined from the standard error of prediction (*SEP*), the coefficient of determination (R^2) and the ratio of the *SEP* to the standard deviation of the validation set (*RPD*). The *RPD* gives an indication of the efficiency of the calibration model. The aim is to obtain the lowest *SEP* with the highest R^2 and *RPD* values. Furthermore, the *SEP* value should be as close as possible to the standard error of laboratory (*SEL*).

Table 4.1 Pretreatments used for the development of calibration models for the Büchi NIRFlex N-500 and Bruker MPA spectral data

Büchi NIRFlex N-500 (The Unscrambler)	Bruker MPA (OPUS)
No spectral pretreatment	No spectral pretreatment
1st derivative[a], 5 points & MSC[b]	Vector normalisation (SNV[e])
2nd derivative[c], 5 points	Min-Max normalisation
2nd derivative, 5 points & MSC	MSC
2nd derivative[d], 9 points	1st derivative, 17 points
MSC	2nd derivative, 17 points
MSC & 2nd derivative, 5 points	1st derivative, 17 points & straight line subtraction
	1st derivative, 17 points & SNV
	1st derivative, 17 points & MSC

[a] 1st derivative Savitzky-Golay

[b] Multiplicative scatter correction

[c] 2nd derivative Savitzky-Golay

[d] 2nd derivative Savitzky-Golay

[e] Standard normal variate

Results

Reference data

A summary of the reference data for moisture, protein and ash contents, PSI and SDS sedimentation values are given in Table 4.2. The distribution of these reference values are depicted in Fig. 4.1. A Gaussian distribution was observed for all parameters. Typical raw (no pretreatment) spectra for triticale flour and wholegrain can be seen in Figs. 4.2 and 4.3.

NIR calibration development

Only the best pretreatment for each parameter as determined by full cross-validation for the two different NIR spectroscopy instruments and software packages, will be discussed. However, all calibration results are listed in Tables 4.3 – 4.6. Calibration models with $R^2 > 0.66$ were validated using an independent validation set for the Büchi as well as

Table 4.2 Summary of the reference data for the different parameters

Parameter	Total sample set				Calibration set					Validation set				
	n	**Range**	**Mean**	***SD*[a]**	***n***	**Range**	**Mean**	***SD***	***SEL*[b]**	***n***	**Range**	**Mean**	***SD***	***SEL***
Moisture (%)	198	10.2 - 12.4	11.2	0.43	148	10.2 - 12.4	11.2	0.44	0.03	50	10.2 - 11.9	11.2	0.43	0.07
Protein (%)	198	7.5 - 16.0	10.9	2.18	148	7.6 - 15.7	10.8	2.12	0.83	50	7.5 - 16.0	11.1	2.30	0.53
Ash (%)	197	1.49 - 2.87	1.83	0.19	147	1.49 - 2.17	1.84	0.24	0.13	50	1.55 - 2.20	1.85	0.16	0.05
Particle size index	196	50.85 - 76.81	62.81	5.79	146	51.2 - 76.8	62.2	5.66	3.27	50	50.8 - 75.8	62.9	5.72	1.85
SDS sedimentation (mm)	198	20 - 65	31	6.39	148	20 - 72	36	14.39	1.67	50	22 - 45	32	6.31	0.90

[a] Standard deviation

[b] Standard error of laboratory: $SEL = \sqrt{\frac{\sum (y_1 - y_2)^2}{2n}}$

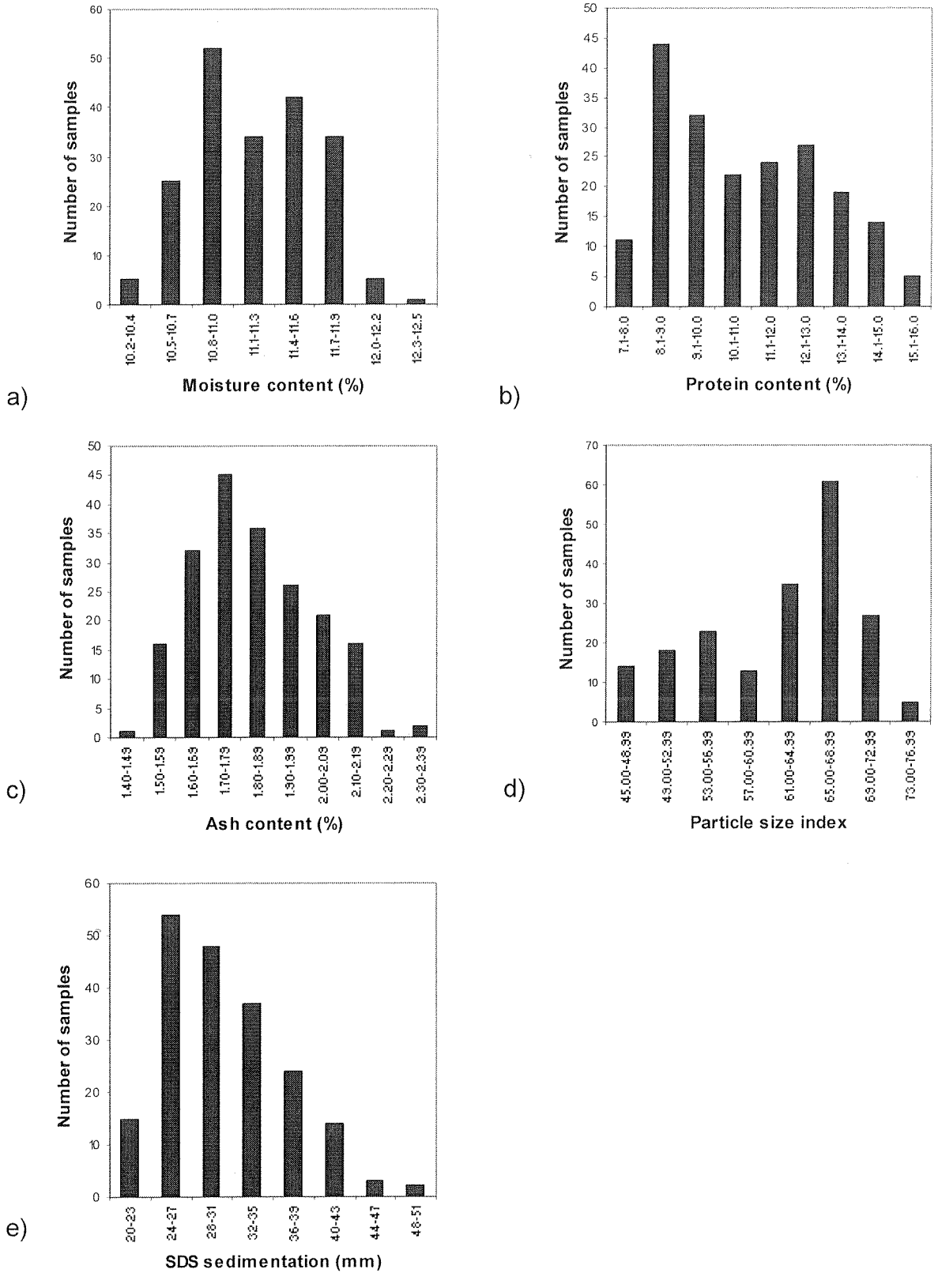
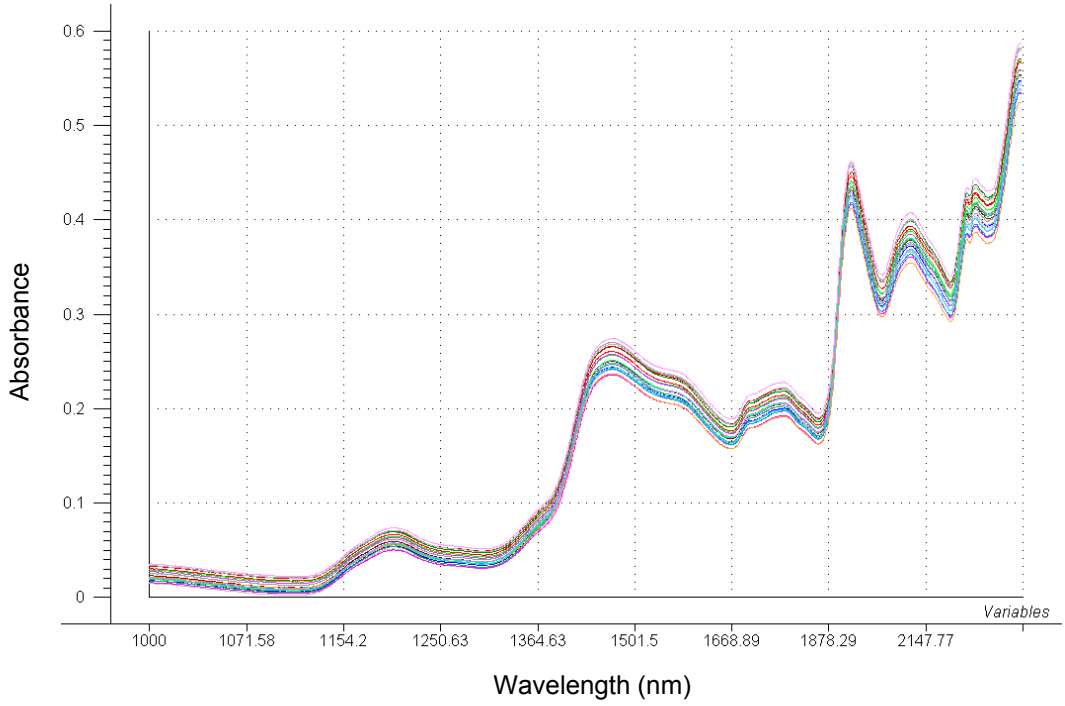


Figure 4.1 Histograms of reference value distributions for a) moisture, b) protein and c) ash contents, as well as d) PSI and e) SDS sedimentation values.

a)



b)

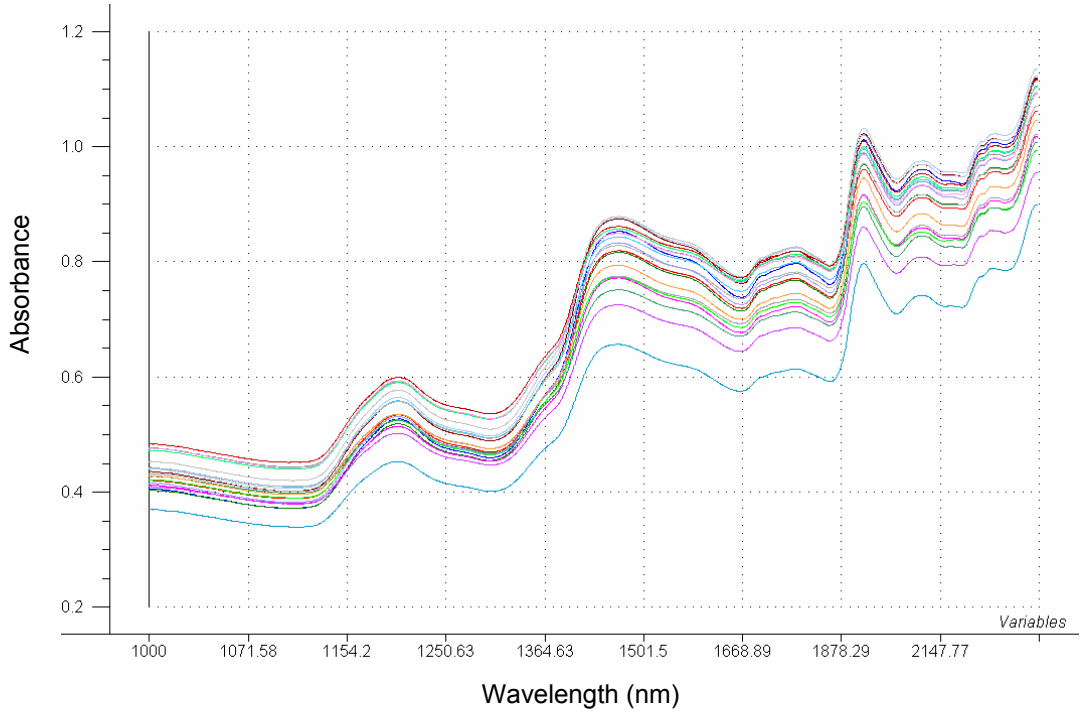
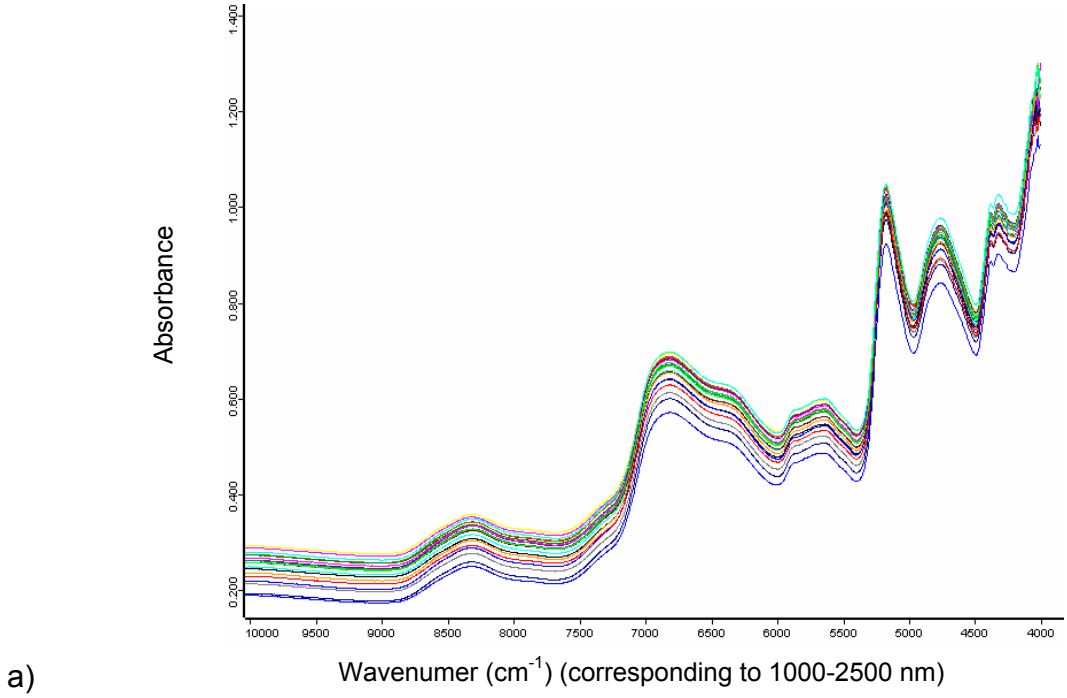
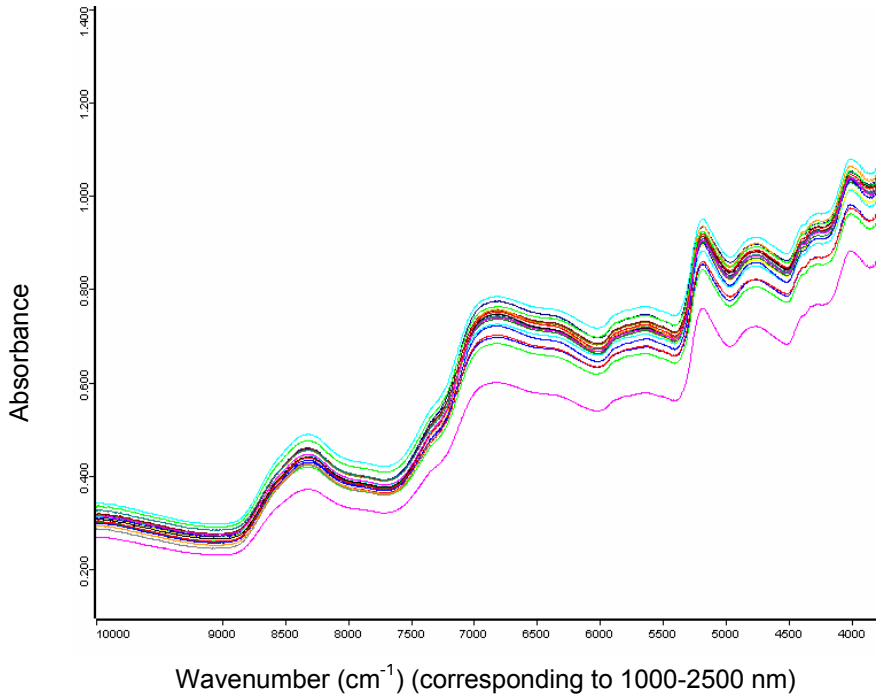


Figure 4.2 Typical NIR spectra of a) triticale flour and b) triticale whole grain for the Büchi intrument using The Unscrambler.



a)



b)

Figure 4.3 Typical NIR spectra of a) triticale flour and b) triticale whole grain for the Bruker instrument using OPUS.

Table 4.3 Summary of the NIR prediction results for flour as obtained from the Büchi spectra (The Unscrambler software) using full cross-validation

Parameter	Pretreatment	Full cross-validation			
		PLS factors	*SECV*[a]	R^2	Bias
Moisture	No spectral pretreatment	10	0.21	0.77	-0.0006
	1st derivative[b], 5 points	4	0.22	0.75	0.00008
	1st derivative, 5 points & MSC[c]	4	0.22	0.74	0.00052
	2nd derivative[d], 5 points	4	0.25	0.69	-0.00003
	2nd derivative, 5 points & MSC	4	0.25	0.68	0.00737
	2nd derivative, 9 points	3	0.24	0.71	-0.00015
	MSC	7	0.22	0.74	-0.00007
	MSC & 2nd derivative, 5 points	4	0.25	0.68	0.00851
Protein	No spectral pretreatment	4	0.56	0.94	0.00148
	1st derivative, 5 points	4	0.47	0.95	-0.0037
	1st derivative, 5 points 3rd & MSC	3	0.47	0.95	-0.00097
	2nd derivative, 5 points 3rd	4	0.76	0.88	0.00153
	2nd derivative, 5 points & MSC	3	0.80	0.87	-0.00664
	2nd derivative, 9 points	3	0.69	0.90	0.00523
	MSC	2	0.57	0.93	0.00065
	MSC & 2nd derivative, 5 points	4	0.82	0.86	-0.00226
Ash	No spectral pretreatment	10	0.11	0.61	-0.00167
	1st derivative, 5 points	6	0.14	0.42	-0.00066
	1st derivative, 5 points & MSC	3	0.14	0.39	0.00039
	2nd derivative, 5 points	3	0.15	0.29	-0.00133
	2nd derivative, 5 points & MSC	3	0.16	0.25	-0.00271
	2nd derivative, 9 points	3	0.15	0.36	-0.00041
	MSC	10	0.12	0.59	0.00102
	MSC & 2nd derivative, 5 points	4	0.16	0.25	-0.00271
PSI	No spectral pretreatment	10	3.08	0.71	0.01833
	1st derivative, 5 points	5	3.31	0.67	0.00436
	1st derivative, 5 points & MSC	5	3.39	0.66	-0.05856
	2nd derivative, 5 points	3	3.57	0.62	0.00727
	2nd derivative, 5 points & MSC	3	3.93	0.54	-0.02947
	2nd derivative, 9 points	3	3.59	0.62	-0.00357
	MSC	6	3.20	0.69	0.01191
	MSC & 2nd derivative, 5 points	3	3.89	0.55	-0.07662
SDS	No spectral pretreatment	13	4.72	0.39	0..06920
	1st derivative, 5 points	4	4.82	0.32	0.03606
	1st derivative, 5 points & MSC	2	5.05	0.27	-0.00514
	2nd derivative, 5 points	2	5.65	0.15	0.01408
	2nd derivative, 5 points & MSC	10	5.57	0.13	0.01385
	2nd derivative, 9 points	2	5.27	0.22	0.02012
	MSC	3	5.07	0.26	0.01105
	MSC & 2nd derivative, 5 points	10	5.60	0.12	0.02260

[a] Standard error of cross-validation

[b] 1st derivative Savitzky-Golay, 3rd polynomial order

[c] Multiplicative scatter correction

[d] 2nd derivative Savitzky-Golay, 3rd polynomial order

Table 4.4 Summary of the NIR prediction results for whole grain as obtained from the Büchi spectra (The Unscrambler software) using full cross-validation

Parameter	Pretreatment	Full cross-validation			
		PLS factors	*SECV*[a]	R^2	Bias
Moisture	No spectral pretreatment	7	0.32	0.46	0.0002
	1st derivative[b], 5 points	3	0.32	0.46	0.0006
	1st derivative, 5 points & MSC[c]	2	0.33	0.43	-0.0026
	2nd derivative[d], 5 points[e]	2	0.37	0.30	-0.0009
	2nd derivative, 5 points & MSC	2	0.36	0.31	-0.0054
	2nd derivative, 9 points[f]	3	0.35	0.35	-0.0031
	MSC	6	0.32	0.45	0.0022
	MSC & 2nd derivative, 5 points	2	0.37	0.29	-0.0044
Protein	No spectral pretreatment	11	0.64	0.91	0.0065
	1st derivative, 5 points	8	0.67	0.91	-0.0184
	1st derivative, 5 points & MSC	7	0.65	0.91	-0.0207
	2nd derivative, 5 points	2	1.15	0.72	-0.0028
	2nd derivative, 5 points & MSC	2	1.28	0.66	-0.0292
	2nd derivative, 9 points	3	1.09	0.75	-0.0106
	MSC	10	0.62	0.92	-0.0018
	MSC & 2nd derivative, 5 points	2	1.28	0.66	-0.0206
Ash	No spectral pretreatment	13	0.12	0.57	-0.0007
	1st derivative, 5 points	5	0.14	0.39	-0.0009
	1st derivative, 5 points & MSC	6	0.13	0.46	-0.0011
	2nd derivative, 5 points	2	0.16	0.22	-0.0014
	2nd derivative, 5 points & MSC	2	0.16	0.24	0.0011
	2nd derivative, 9 points	5	0.15	0.36	-0.0009
	MSC	14	0.11	0.59	-0.0005
	MSC & 2nd derivative, 5 points	4	0.15	0.27	-0.0012
PSI	No spectral pretreatment	9	4.02	0.52	0.0154
	1st derivative, 5 points	4	3.96	0.54	0.0281
	1st derivative, 5 points & MSC	3	4.11	0.50	-0.0414
	2nd derivative, 5 points	2	4.35	0.44	-0.0033
	2nd derivative, 5 points & MSC	2	4.52	0.41	0.0710
	2nd derivative, 9 points	2	4.22	0.47	0.0371
	MSC	10	4.01	0.52	0.0013
	MSC & 2nd derivative, 5 points	2	4.47	0.41	0.0524
SDS	No spectral pretreatment	9	4.40	0.46	0.0183
	1st derivative, 5 points	5	5.11	0.29	-0.0541
	1st derivative, 5 points & MSC	5	5.04	0.32	0.0581
	2nd derivative, 5 points	10	5.45	0.16	-0.0208
	2nd derivative, 5 points & MSC	10	5.62	0.13	-0.1180
	2nd derivative, 9 points	10	5.40	0.18	-0.0007
	MSC	7	4.72	0.37	-0.0361
	MSC & 2nd derivative, 5 points	10	5.57	0.14	-0.0603

[a] Standard error of cross-validation

[b] 1st derivative Savitzky-Golay, 3rd polynomial order

[c] Multiplicative scatter correction

[d] 2nd derivative Savitzky-Golay, 3rd polynomial order

Table 4.5 Summary of the NIR prediction results for flour as obtained from the Bruker spectra (Opus software) by full cross-validation

Parameter	Pretreatment	Full cross-validation			
		PLS factors	*SECV*[a]	R^2	Bias
Moisture	No spectral pretreatment	10	0.13	0.91	0.00017
	Vector normalisation (SNV[b])	9	0.13	0.90	0.00096
	Min-Max normalization	7	0.13	0.90	-0.00060
	MSC[c]	5	0.15	0.87	-0.00032
	1st derivative[d], 17 points	9	0.13	0.90	0.00059
	2nd derivative[e], 17 points	6	0.15	0.87	-0.00059
	1st derivative & straight line subtraction	7	0.13	0.90	-0.00261
	1st derivative, 17 points & SNV	9	0.16	0.86	0.00078
	1st derivative, 17 points & MSC	8	0.17	0.84	0.00069
Protein	No spectral pretreatment	8	0.57	0.93	0.00236
	Vector normalisation (SNV)	10	0.50	0.95	-0.00070
	Min-Max normalisation	10	0.51	0.94	-0.00246
	MSC	8	0.53	0.94	0.00180
	1st derivative, 17 points	9	0.55	0.94	0.00257
	2nd derivative, 17 points	5	0.62	0.92	0.00006
	1st derivative & straight line subtraction	9	0.57	0.93	0.00370
	1st derivative, 17 points & SNV	8	0.52	0.94	0.00251
	1st derivative, 17 points & MSC	9	0.53	0.94	0.00164
Ash	No spectral pretreatment	9	0.14	0.40	0.00077
	Vector normalisation (SNV)	7	0.15	0.30	-0.00227
	Min-Max normalisation	5	0.16	0.16	0.00106
	MSC	4	0.16	0.15	-0.00055
	1st derivative, 17 points	2	0.17	0.06	0.00006
	2nd derivative, 17 points	1	0.17	0.01	0.00009
	1st derivative & straight line subtraction	2	0.17	0.06	0.00005
	1st derivative, 17 points & SNV	1	0.17	0.02	0.00002
	1st derivative, 17 points & MSC	1	0.17	0.02	0.00002
PSI	No spectral pretreatment	6	3.60	0.60	-0.00265
	Vector normalisation (SNV)	4	3.77	0.56	0.00647
	Min-Max normalisation	5	3.81	0.55	-0.00153
	MSC	5	3.80	0.55	-0.00231
	1st derivative, 17 points	4	3.80	0.55	0.00232
	2nd derivative, 17 points	2	4.13	0.47	0.05780
	1st derivative & straight line subtraction	3	3.98	0.51	0.00149
	1st derivative, 17 points & SNV	3	4.02	0.50	-0.01070
	1st derivative, 17 points & MSC	2	4.07	0.48	0.00664
SDS	No spectral pretreatment	6	5.40	0.32	0.00035
	Vector normalisation (SNV)	5	5.40	0.32	-0.00717
	Min-Max normalisation	6	5.40	0.32	-0.00727
	MSC	5	5.39	0.32	0.01110
	1st derivative, 17 points	4	5.41	0.32	0.00022
	2nd derivative, 17 points	3	5.23	0.36	0.02050
	1st derivative & straight line subtraction	4	5.40	0.32	0.00148
	1st derivative, 17 points & SNV	3	5.44	0.31	0.00344
	1st derivative, 17 points & MSC	2	5.44	0.31	0.00228

[a] Standard error of cros-validation

[b] Standard normal variate

[c] Multiplicative scatter correction

[d] 1st derivative, 3rd polynomial order

[e] 2nd derivative, 3rd polynomial order

Table 4.6 Summary of the NIR prediction results for whole grain as obtained from the Bruker spectra (Opus software) using full cross-validation

Parameter	Pretreatment	Full cross-validation			
		PLS factors	*SECV*[a]	R^2	Bias
Moisture	No spectral pretreatment	4	0.33	0.46	0.00007
	Vector normalisation (SNV[b])	3	0.33	0.44	0.00020
	Min-Max normalisation	4	0.32	0.48	-0.00014
	MSC[c]	3	0.33	0.44	0.00019
	1st derivative[d], 17 points	4	0.32	0.49	0.00020
	2nd derivative[e], 17 points	4	0.33	0.46	0.00183
	1st derivative & straight line subtraction	4	0.32	0.49	0.00029
	1st derivative, 17 points & SNV	3	0.32	0.47	0.00076
	1st derivative, 17 points & MSC	3	0.32	0.49	0.00088
Protein	No spectral pretreatment	10	0.70	0.90	-0.00077
	Vector normalisation (SNV)	7	0.67	0.90	0.00071
	Min-Max normalisation	7	0.69	0.90	-0.00075
	MSC	7	0.67	0.90	0.00070
	1st derivative, 17 points	7	0.69	0.90	0.00037
	2nd derivative, 17 points	5	0.91	0.82	0.00253
	1st derivative & straight line subtraction	6	0.71	0.89	0.00259
	1st derivative, 17 points & SNV	6	0.69	0.90	0.00076
	1st derivative, 17 points & MSC	6	0.66	0.90	0.00197
Ash	No spectral pretreatment	6	0.16	0.35	0.00132
	Vector normalisation (SNV)	7	0.16	0.33	-0.00029
	Min-Max normalisation	8	0.16	0.33	-0.00043
	MSC	7	0.16	0.33	-0.00050
	1st derivative, 17 points	5	0.15	0.38	0.00027
	2nd derivative, 17 points	4	0.16	0.29	-0.00051
	1st derivative & straight line subtraction	5	0.14	0.44	0.00100
	1st derivative, 17 points & SNV	4	0.15	0.43	0.00136
	1st derivative, 17 points & MSC	3	0.15	0.36	-0.00017
PSI	No spectral pretreatment	6	3.60	0.62	0.01020
	Vector normalisation (SNV)	5	3.58	0.62	-0.00070
	Min-Max normalisation	4	1.60	0.61	0.00387
	MSC	5	3.58	0.62	-0.00018
	1st derivative, 17 points	4	3.46	0.65	-0.03160
	2nd derivative, 17 points	2	3.78	0.58	-0.00208
	1st derivative & straight line subtraction	4	3.51	0.64	-0.07080
	1st derivative, 17 points & SNV	5	3.63	0.61	0.00462
	1st derivative, 17 points & MSC	4	3.65	0.61	0.00621
SDS	No spectral pretreatment	7	4.80	0.44	-0.02030
	Vector normalisation (SNV)	7	4.91	0.41	0.01280
	Min-Max normalisation	8	4.89	0.42	0.00626
	MSC	7	4.90	0.42	0.01150
	1st derivative, 17 points	6	5.16	0.35	0.01710
	2nd derivative, 17 points	1	5.82	0.18	0.00935
	1st derivative & straight line subtraction	3	5.34	0.31	-0.01420
	1st derivative, 17 points & SNV	2	5.15	0.35	-0.01600
	1st derivative, 17 points & MSC	4	5.42	0.29	0.02500

[a] Standard error of cross-validation

[b] Standard normal variate

[c] Multiplicative scatter correction

[d] 1st derivative, 3rd polynomial order

[e] 2nd derivative, 3rd polynomial order

Table 4.7 Summary of calibration and validation results for the best full cross-validation models from the Büchi and Bruker data

Instrument	Parameter	Pretreatment	Calibration			Validation				
			PLS factors	*SEP*[a]	R^2	PLS factors	*SEP*	R^2	Bias	*RPD*[b]
Büchi	Moisture (%) (flour)	No spectral pretreatment	4	0.25	0.68	4	0.25	0.67	0.0451	1.76
	Protein (%) (flour)	1st derivative[c], 5 points	4	0.36	0.97	4	0.44	0.96	-0.1362	5.23
	PSI (flour)	No spectral pretreatment	6	3.43	0.66	6	3.78	0.57	0.1495	1.51
	Protein (%) (whole grain)	No spectral pretreatment	12	0.49	0.95	12	0.55	0.94	-0.0881	4.18
Bruker	Moisture (%) (flour)	No spectral pretreatment	7	0.07	0.98	7	0.08	0.95	-0.0045	4.65
	Protein (%) (flour)	Vector normalisation (SNV)	5	0.46	0.96	5	0.32	0.96	-0.035	4.88
	Protein (%) (whole grain)	MSC	7	0.52	0.94	7	0.70	0.90	0.0060	3.23

[a] Standard error of prediction

[b] Ratio of the *SEP* to the standard deviation of the validation set

[c] 1st derivative Savitzky-Golay, 3rd polynomial order

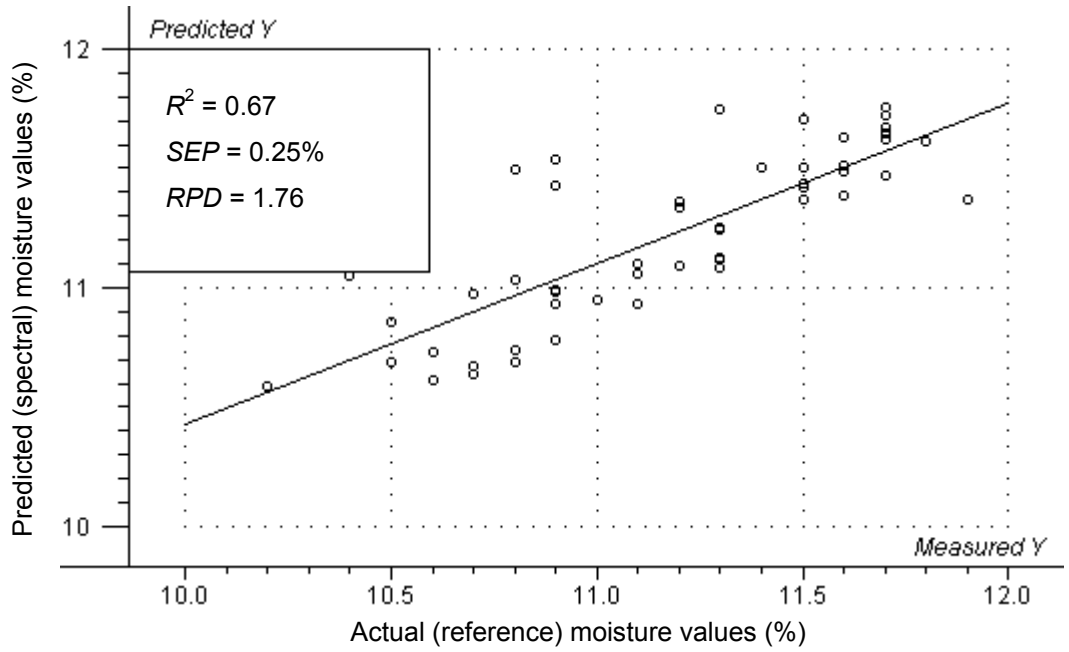


Figure 4.4 Validation plot for moisture with no spectral pretreatment for the Büchi data using The Unscrambler (flour).

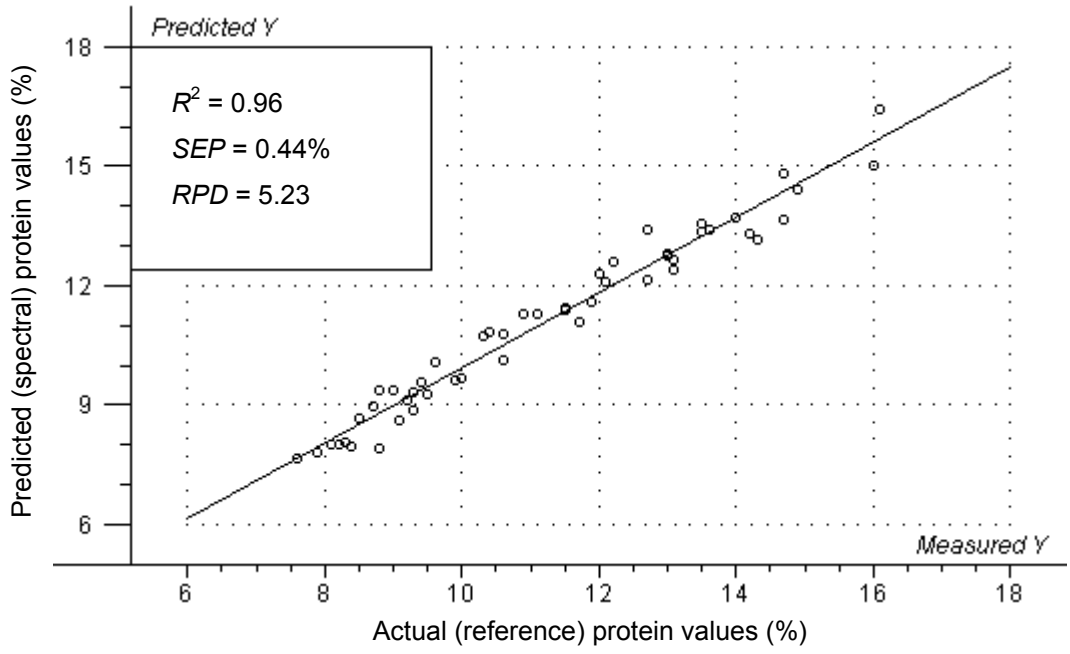


Figure 4.5 Validation plot for protein with 1st derivative (5 points, 3rd polynomial order) as spectral pretreatment for the Büchi data using The Unscrambler (flour).

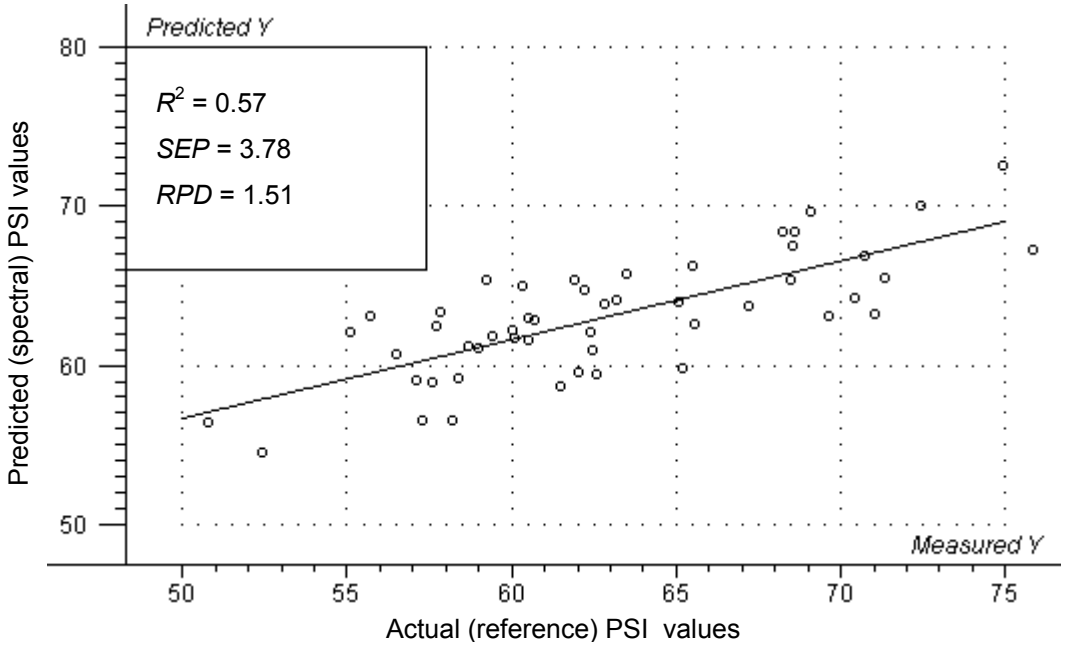


Figure 4.6 Validation plots for PSI with no spectral pretreatment for the Büchi data using The Unscrambler (flour).

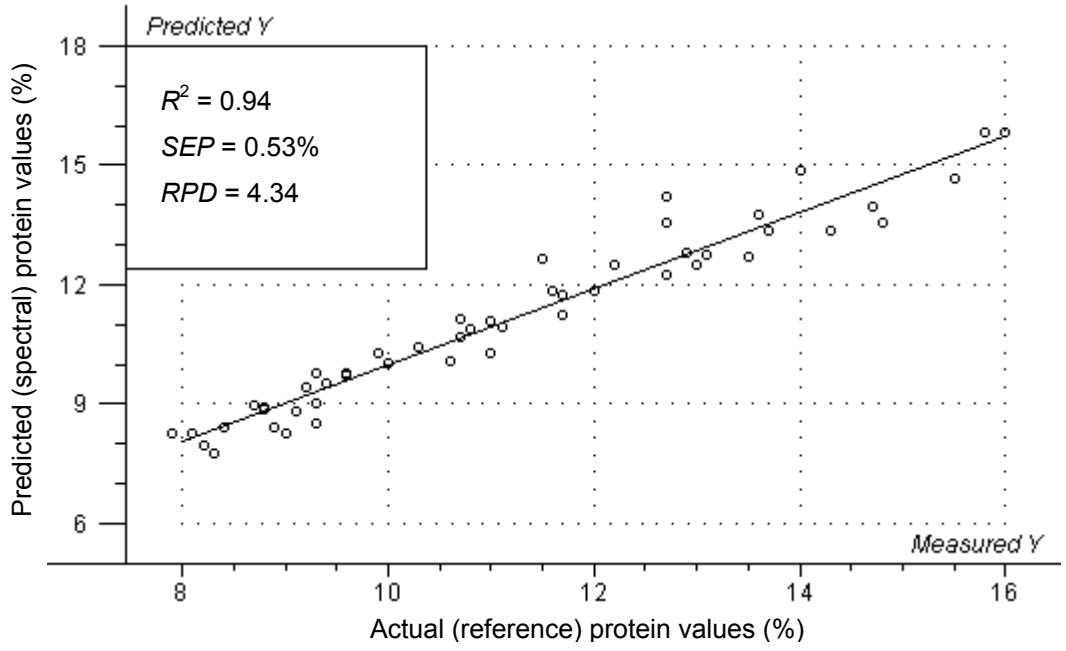


Figure 4.7 Validation plots for protein with MSC as spectral pretreatment for the Büchi data using The Unscrambler (whole grain).

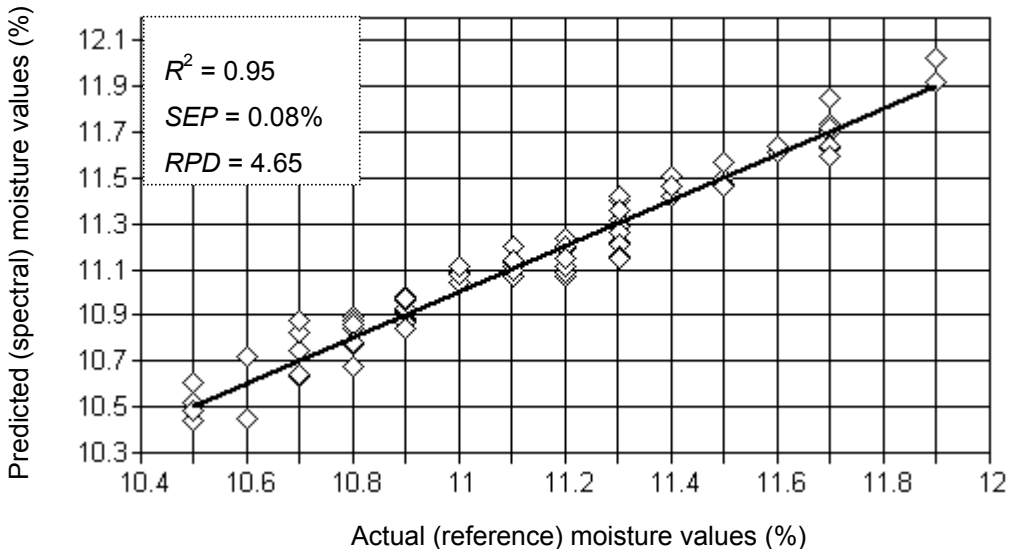


Figure 4.8 Validation plots for moisture with no spectral pretreatment for the Bruker data using OPUS (flour).

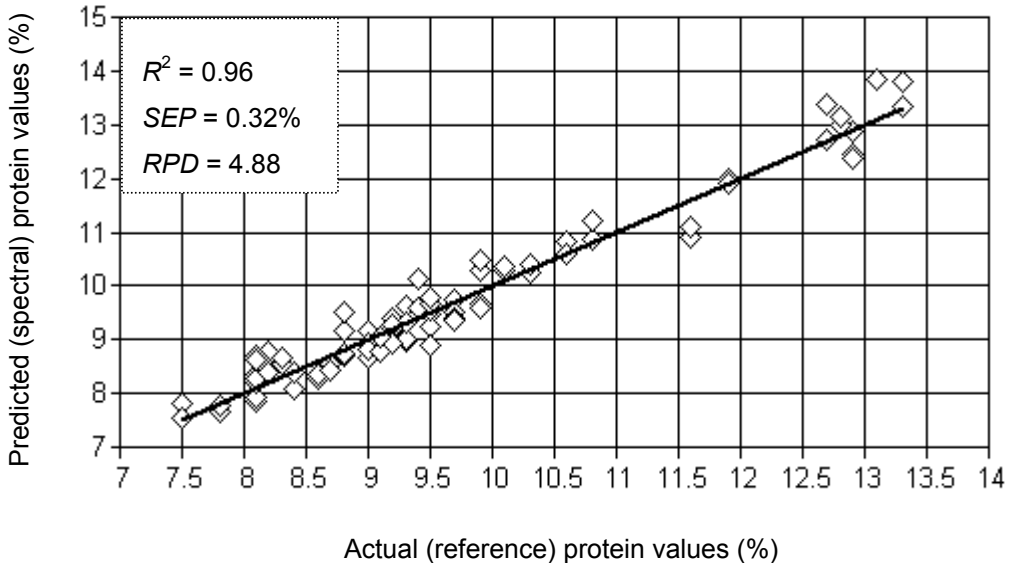


Figure 4.9 Validation plots for protein with Vector normalisation (SNV) as spectral pretreatment for the Bruker data using OPUS (flour)

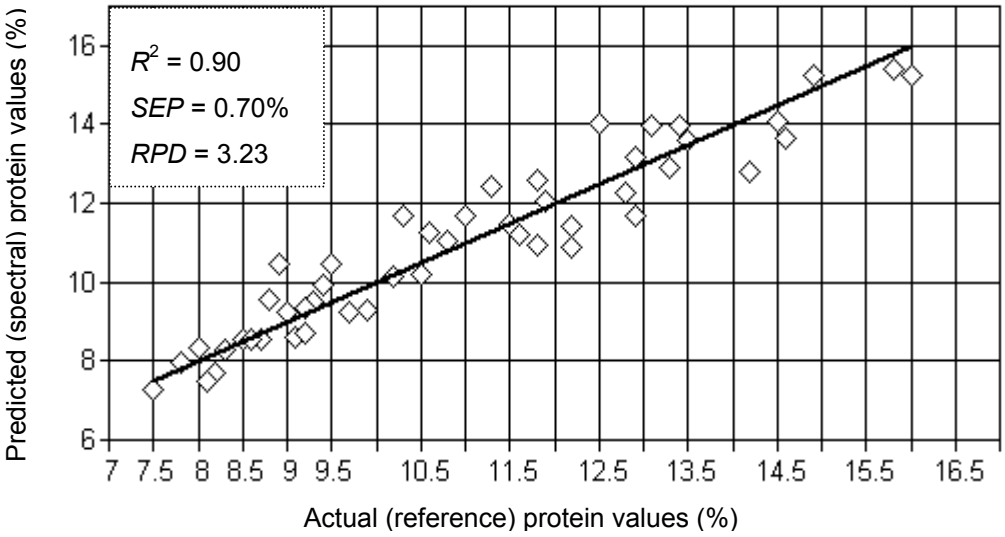


Figure 4.10 Validation plot for protein with MSC as spectral pretreatment for the Bruker data using OPUS (whole grain).

Bruker data and reported in Table 4.7 and Figs. 4.2 – 4.8. Extremely low biases were observed for all treatments (less than 0.1 in all cases). Very similar results were obtained for the two instruments and software packages.

Discussion

The Gaussian distribution observed for the reference data is indicative of the fact that samples were randomly obtained and not specifically chosen for a certain known composition. Such a distribution is likely to result in more accurate predictions for samples that have values close to the mean, and less accurate for samples near the extremes of the range (Williams, 2001). During the initial phases of the development of a calibration model, this is quite normal, often due to small initial sample sets, but as the model is expanded with the addition of subsequent harvest seasons, the range is expected to expand. This can also be facilitated by choosing samples in future with values that fall in the extremes of the range, in order to obtain a more robust calibration model.

Moisture content

Good results were obtained for flour with the Büchi instrument using full cross-validation with The Unscrambler software (Table 4.3). The best result was observed with no spectral pretreatment (R^2 = 0.77; *SECV* = 0.21%). This model was validated using the test set (n = 50), which resulted in a R^2 value of 0.67, a *SEP* of 0.25% and a *RPD* of 1.76 (Table 4.7; Fig. 4.4). The R^2 value implies that the model could be used for rough screening, but according to the *RPD* it is not recommended that the model be used for application without further research (Williams, 2001). Furthermore, the *SEP* (0.25%) was higher than the *SEL* (0.07) for the moisture reference values, indicating that the NIR calibration result was less accurate than the reference method. These results could improve if a test set is chosen specifically to cover the range of moisture values, instead of using a fixed test set for all parameters as was the case in this study. Furthermore, the high number of PLS factors (10) used when evaluating the model with full cross-validation, could be the reason for these results to be slightly optimistic. As can be seen only 4 PLS factors were adequate when evaluating the model by means of an independent test set which would have prevented overfitting.

The full cross-validation results obtained for the whole grain moisture prediction of the Büchi data using The Unscrambler did not yield any R^2 values over 0.46. It is thus not recommended that these calibrations be applied and further investigation into these models is necessary (Table 4.4).

Excellent full cross-validation results were obtained for the determination of moisture content in flour with the Bruker instrument using OPUS with the best result being observed with no spectral pretreatment (R^2 = 0.91; *SECV* = 0.13%) (Tabel 4.5). This model was validated using the test set, resulting in a R^2 value of 0.95, a *SEP* of 0.08% and a *RPD* of 4.65 (Table 4.7; Fig 4.8). The *RPD* implies that the model is good and can be used for screening purposes (Williams, 2001).

As with the Büchi results, the whole grain calibrations for moisture prediction with the Bruker yielded low R^2 values < 0.49 (Table 4.6). The correlations were thus poor (Williams, 2001) and more research is necessary for the moisture prediction of the whole grain samples. The contradicting results for the NIR moisture predictions obtained for the flour and whole grain samples as well as for the two different instruments could be due to the fact that the samples were not analysed by reference and NIR methods simultaneously. The moisture for the flour could have changed slightly after being analysed on the one NIR instrument (Bruker) until it was analysed on the second NIR instrument (Büchi). Slight changes could also have taken place from when the samples were analysed using the reference method until they were analysed by NIR. In the case of the whole grain samples, it is likely that some moisture loss could have taken place during the grinding process, resulting in a difference in the moisture content of the whole grain sample and that of the flour samples being analysed by the reference method.

The R^2 and *SEP/SECV* values obtained for the moisture calibrations for flour for both instruments, however, compared well that reported in literature for South African wheat cultivars (R^2 = 0.72; *SEP* = 0.15% (Manley *et al*., 2002) and the Bruker/OPUS calibration for flour compared well, with slightly improved SEP values, with results recently obtained for triticale (R^2 = 0.82 – 0.98; *SEP* = 0.30 – 0.34% (Igne *et al*., 2007a; b).

Protein content

Extremely good calibration models were obtained with full cross-validation for the prediction of protein content in flour using the Büchi together with The Unscrambler software (Table 4.3). Similar models were observed with 1st derivative (5 points, 3rd polynomial order) pretreatment as well as 1st derivative (5 points, 3rd polynomial order) followed by MSC pre-treatment (R^2 = 0.95; *SECV* = 0.47% for both models). These models can thus be used for most applications, including quality assurance (Williams, 2001). Equally good models were obtained using an independent test set with the 1st derivative pretreatment resulting in a R^2 value of 0.96, a *SEP* of 0.44% and a *RPD* of 5.23 (Table 4.7; Fig. 4.5). The high *RPD* confirms that the model can be used for quality control

(Williams, 2001). The *SEP* obtained (0.44%) is also lower than the *SEL* for the protein reference method (0.53), implying that the NIR spectroscopy method is very accurate compared to the reference method. The lower value observed for the *SEP* compared to the *SEL* could be due to less error being involved in collecting spectra compared to the wet chemistry method involved.

For the whole grain samples, the best model for the Büchi data as determined by full cross-validation was obtained with a MSC pre-treatment (R^2 = 0.92; *SECV* = 0.62%). Though these results are not as good as for the flour calibrations, it is still a very good result, and the method was validated using the test set. This resulted in a R^2 of 0.94, a *SEP* of 0.53% and a *RPD* of 4.34, indicating that the model can be applied for screening purposes (Williams, 2001) (Table 4.7; Fig. 4.7). Furthermore the *SEP* is equal to the *SEL* for the protein reference values, thus the NIR spectroscopy method is very accurate.

Good results were also obtained from the Bruker data (using OPUS) for the protein content prediction in flour, with the best results seen when using SNV as pretreatment method (Table 4.5). This pretreatment resulted in a R^2 value of 0.95 and an *SECV* of 0.50%. This method was validated with the test set, yielding a R^2 of 0.96, a *SEP* of 0.32% and a RPD of 4.88, indicating that the methods was good for screening (Table 4.7; Fig 4.9). Again the *SEP* which is lower than the reference *SEL* illustrates the accuracy of the model.

For the whole grain protein content as determined by the Bruker (OPUS), SNV and MSC both yielded the best results as pretreatment methods with R^2 values of 0.90 and *SECV* values of 0.67% (Table 4.6). These pretreatments were validated with the test set, with MSC yielding the best results (R^2 = 0.90; *SEP* = 0.70%; *RPD* = 3.23) (Table 4.7; Fig. 4.10). Again, this implies that the model is adequate for application in screening (Williams, 2001).

The results obtained for the protein prediction models compared very well with (and in some cases showed an improvement compared to) results obtained in literature for wheat as observed by Delwiche (1998) (R^2 = 0.90 – 0.98; *SEP* = 0.47 – 0.59%) and Manley *et al*. (2002) for South African wheat (R^2 = 0.65; *SEP* = 0.51%). The results also compared well with results observed for triticale by Viljoen *et al*. (2005) (R^2 = 0.86; *SEP* = 0.60%) and Igne *et al*. (2007a) (R^2 = 0.92 – 0.96; *SEP* = 0.30 – 0.34%).

Ash content

For the ash calibration models, no R^2 values higher than 0.66 were obtained, thus no validations were carried out, as the models need to be improved. The best cross-validation

result for the Büchi data (The Unscrambler) for flour was obtained with no spectral pretreatment (Table 4.3). With a R^2 of 0.61, more than 50% of the variance in NIR data is accounted for by the reference data, but the model is only adequate for rough screening and more research is needed (Williams, 2001) to improve these calibrations. A *SECV* of 0.11% was obtained. The best pretreatment for the whole grain data obtained from the Büchi with full cross-validation was MSC (R^2 = 0.59; *SECV* = 0.11% (Table 4.4). As for the flour model, the R^2 implies that the model is adequate for rough screening and that more than 50% of the variance in NIR data is accounted for by the reference data, but that the model needs to be improved before it can be applied (Williams, 2001).

The prediction results for ash content for the Bruker data as analysed with the OPUS software, were found to be very poor. For flour, the best result was observed with no spectral pretreatment (R^2 = 0.40; *SECV* = 0.14% (Table 4.5). This indicates that there is a poor correlation between the reference and spectral data. The same was found to be true for the whole grain Bruker data analysed with Opus, where the best pretreatment (1st derivative followed by straight line subtraction) yielded a R^2 value of 0.44 and a *SECV* of 0.14% (Table 4.6). The results obtained were thus not as good as those obtained by Viljoen *et al*. (2005) (R^2 = 0.86; *SECV* = 0.17%), but could be improved in future by selecting a sample set that covers a wider range and has more values towards the extremes of the range. The sample set used in this study has a very narrow range (1.49 – 2.87%), compared to a range of 0.93 – 3.41% as was used in the study done by Viljoen *et al*. (2005).

Particle size index (PSI)

The results obtained from the Büchi data for flour as analysed by The Unscrambler were generally acceptable; the best result for full cross-validation being obtained for no spectral pretreatment (R^2 = 0.71; *SECV* = 3.08) (Table 4.3). This is to be expected, as the determination of the particle size index with NIR spectroscopy is dependant on the spectral data indicating differences in particle size; the effect of which would have been removed by the smoothing effect of the other pretreatments. Validation with the independent test set yielded disappointing results (R^2 = 0.57; *SEP* = 3.78; *RPD* = 1.51) (Table 4.7; Fig. 4.6). With an *RPD* lower than 2.3, it is not recommended that the model be used for quality control or screening purposes. For the Büchi whole grain data as analysed with The Unscrambler, the best pretreatment (1st derivative, 5 points, 3rd polynomial order) resulted in a R^2 of 0.54 and a *SECV* of 3.96 with full cross-validation (Table 4.4). The R^2 value

indicates that more than 50% of the variance in NIR data is accounted for by the reference data, and that the model could be used for rough screening only (Williams, 2001).

Slightly improved results were obtained with the Bruker results as determined by OPUS using full cross-validation, with the best R^2 and *SECV* values obtained for flour (0.60 and 3.60 respectively) with no pretreatment applied (Table 4.5) and the best R^2 (0.65) and *SECV* (3.46) for whole grain being obtained with 1st derivative as pretreatment (Table 4.6).

Results for hardness determination by NIR spectroscopy for triticale could not be found in literature, but the R^2 values obtained in this study with both instruments and software packages were an improvement on results obtained previously on South African wheat (Manley *et al.* (2002) (R^2 = 0.18).

SDS sedimentation

Results for SDS sedimentation prediction of triticale flour by NIR spectroscopy were found to be poor for both instruments and software packages, and no independent validations were carried out. With the Büchi instrument and The Unscrambler software, the best full cross-validation results for flour were obtained with no spectral pretreatment (R^2 = 0.39; *SECV* = 4.72 mm) (Table 4.3), and for the whole grain the best results were also seen when applying no pretreatment (R^2 = 0.46; *SECV* = 4.40 mm) (Table 4.4). The correlation between the spectral and reference data was thus poor in both cases, and improvement of these models should be attempted by adding data from subsequent seasons and by increasing the range (Williams, 2001).

SDS sedimentation prediction results obtained for the Bruker data (OPUS software) were very similar, with the best R^2 and *SECV* values (0.36 and 5.23 mm respectively) being observed for the 2nd derivative pretreatment for flour with full cross-validation (Table 4.5). For the whole grain, the best results were observed with no spectral pre-treatment (R^2 = 0.44; *SECV* = 4.80 mm) (Table 4.6). Both models thus also showed poor correlations between reference and spectral data, and should not be applied without further investigation (Williams, 2001).

Conclusion

NIR spectroscopy shows promise for the rapid and accurate estimation of the moisture and protein contents of triticale. While a calibration model only suitable for screening purposes was obtained for moisture content, excellent calibration models, adequate for application in quality control were obtained for protein content. More work needs to be done, however, to improve models to predict ash content, hardness and baking potential.

The poor results for SDS sedimentation can probably be expected, as indicators of baking quality are generally harder to estimate using NIR spectroscopy. The models developed, however, pose a great deal of promise. Depending on the desired end use, be it early generation screening for breeding purposes or for quality control in the food industry, the moisture, protein, ash and hardness prediction models can be applied, and can be further improved with the addition of data from subsequent harvest seasons. In addition improvement of the various models can be investigated by means of applying different variable selection methods with the result that only the variables that really contribute to the model would be included.

References

Butler, L.A. (1983). The history and background of NIR. *Cereal Foods World*, **28**(4), 238-240.

Delwiche, S.R. (1998). Protein content of single kernels of wheat by near-infrared reflectance spectroscopy. *Journal of Cereal Science*, **27**, 241-254.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007a). Triticale moisture and protein content prediction by near-infrared spectroscopy (NIRS). *Cereal Chemistry*, **84**(4), 328-330.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007b). Influence of yearly variability of agricultural products on calibration process: a triticale example. *Cereal Chemistry*, **84**(6), 576-581.

Law, D.P. & Tkachuk, R. (1977). Determination of moisture content in wheat by near infrared diffuse reflectance spectrophotometry. *Cereal Chemistry*, **54**(4), 874-881.

Manley, M., Van Zyl, L. & Osborne, B.G. (2002). Using Fourier transform near infrared spectroscopy in determining kernel hardness, protein content and moisture content of whole wheat flour. *Journal of Near Infrared Spectroscopy*, **10**, 71-76.

Norris, K.H., Hruschka, W.R., Bean, M.M. & Slaughter, D.C. (1989). A definition of wheat hardness using near infrared reflectance spectroscopy. *Cereal Foods World*, **34**(9), 696-705.

Osborne, B.G. (1987). Determination of moisture in white flour, ground wheat and whole wheat by near infrared reflectance using a single calibration. *Journal of the Science of Food and Agriculture*, **38**, 341-436.

Osborne, B.G. (1991). Measurement of the hardness of wheat endosperm by near-infrared spectroscopy. *Postharvest News and Information*, **2**, 331-334.

Osborne, B.G. (2000). Recent developments in NIR analysis of grains and grain products. *Cereal Foods World*, **45**(1), 11-15.

Osborne, B.G. & Fearn, T. (1983). Collaborative evaluation of near infrared reflectance analysis for the determination of protein, moisture and hardness in wheat. *Journal of the Science of Food and Agriculture*, **34**, 1011-1017.

Osborne, B.G., Fearn, T. & Hindle, P.H. (1993). *Practical NIR Spectroscopy with Practical Applications in Food and Beverage Analysis*, 2nd ed, p. 227. Harlow, UK: Longman Scientific and Technical.

Pasquini, C. (2003). Near infrared spectroscopy: fundamentals, practical aspects and analytical applications. *Journal of the Brazilian Chemical Society*, **14**(2), 198-219.

Shenk, J.S. & Westerhaus, M.O. (1985). Accuracy of NIRS instruments to analyze forage and grain. *Crop Science*, **25**, 1120-1122.

Viljoen, M., Brand, T.S., Brandt, D.A. & Hoffman, L.C. (2005). Prediction of the chemical composition of winter grain and maize with near infrared reflectance spectroscopy. *South African Journal of Plant and Soil*, **22**(2), 89-93.

Williams, P.C. (2001). Implementation of near-infrared technology. In: *Near-Infrared Technology in the Agricultural and Food Industries*, 2nd ed. (edited by P. Williams & K. Norris). Pp 145-169. St. Paul, USA: American Association of Cereal Chemists.

Williams, P.C. & Sobering, D.C. (1986). Attempts at standardization of hardness testing of wheat. II. The near-infrared reflectance method. *Cereal Foods World*, **31**(6), 417-420.

CHAPTER 5

General discussion and conclusion

CHAPTER 5

GENERAL DISCUSSION AND CONCLUSION

Triticale is a crop with a great deal of agronomic and economic potential (Mergoum *et al*., 2004). It is therefore valuable to have a compositional and functional profile for this crop. Although various studies in this regard have been carried out in other parts of the world, no comprehensive study has to date been performed to determine the composition and functionality of South African triticale cultivars. Six triticale cultivars from each of six localities in the Western Cape (one repetition from each) obtained for the 2006 harvest season, as well as six cultivars from each of nine localities (three repetitions from each) obtained for the 2007 harvest season, were evaluated in this study. In addition three repetitions of a strong bread wheat cultivar (Kariega) were obtained from eight of the nine localities for 2007. The moisture, protein and ash contents, falling number (as an indication of α-amylase activity), hardness (as determined by particle size index) and potential baking quality (measured in terms of gluten strength as determined by SDS sedimentation) were determined for all these samples.

The composition and functionality of the South African triticale cultivars were found to be similar to results obtained in worldwide studies. The protein content of the South African cultivars was found to range from 7.5 to 16.0%; also comparing well with the wheat cultivar, Kariega, which resulted in an average protein content of 11.3%. Cultivars within years differed significantly ($P < 0.05$), and from these results the purpose for which the respective cultivars were bred became evident. The cultivar US2007 is an example of this; it was bred for high protein content in order to be used as whole grain animal feed and as expected resulted in the highest protein content when analysed during this study. Significant differences were observed between localities over the two seasons, with a lower average protein content observed for 2007. During 2007, higher rainfall and lower temperatures were experienced, which illustrates the influence of environmental conditions, specifically the lowering effect that a high rainfall combined with lower temperatures during seed-filling can have on protein content (Pfeiffer, 1994; Alaru, 2003).

The relatively high ash content observed for triticale (1.49 – 2.87%) was in almost all cases significantly higher than that observed for wheat, which is consistent with literature (Kent & Evers, 1994; Leon *et al*., 1996; Stallknecht *et al*., 1996) and is known to have a detrimental effect on milling and baking quality.

Falling number values for the triticale samples (62 – 300 s) were significantly lower than the average for the wheat samples (440 s), which confirmed the known inherently high α-amylase activity of triticale. It was expected that values for the 2007 samples would be lower due to a higher rainfall during the seed-filling period (Leon *et al*., 1996; Alaru *et al.*, 2003; Erekul & Köhn, 2006), yet the 2007 samples had significantly ($P < 0.05$) higher falling number values compared to the 2006 samples. This occurrence could possibly be explained in terms of grain maturity, as the level of maturity also affects the degree to which rainfall influences α-amylase activity. It would thus be advisable to consult grain maturity data when evaluating triticale α-amylase activity in future. The inherently high α-amylase activity of triticale will, however, continue to be a disadvantage if triticale cultivars are to be considered for baked products.

A range of 50.85 to 76.81 was observed for particle size index (PSI), and the average values obtained for the triticale cultivars were in almost all cases significantly higher than the average for the wheat samples, implying that the triticale samples were generally softer. This is in agreement with previous studies (Alvarez *et al*., 1992; Martín *et al*., 1999; Ramírez *et al*., 2003; Barrera *et al*., 2007). The cultivars were found not to differ significantly between the two years, illustrating the effect of genetic predisposition in this regard. The significant differences observed between localities over the two seasons, however, imply that environment does, to a lesser extent, play a role in kernel hardness (Pomeranz & Williams, 1990). Ideally a harder kernel would be more beneficial for improved baking quality.

Values for 1000-kernel mass ranged from 34.82 to 55.12 g, which compared well with values reported in literature for wheat (Alvarez *et al.*, 1992; Erekul & Köhn, 2006). Due to the positive relationship that exists for wheat between 1000-kernel mass and flour yield, it is possible that this could relate to a good flour yield. Triticale is not known to produce high flour yields and these high 1000-kernel mass values could be an indication of improvement in the new cultivars in this regards.

SDS sedimentation results obtained for the triticale samples (20 – 65 mm) were similar to those reported in literature (Martín *et al.*, 1999), but were much lower than for the wheat samples (average of 86 mm). The latter illustrated the weak gluten that triticale possesses compared to wheat (Erekul & Köhn, 2006). The non-significant difference between localities over the two seasons implies that the environmental effect on gluten strength is not as important as the genetic effect. The low values obtained compared to wheat confirm the known poor baking quality characteristics of triticale for the production of products such as bread. The use of triticale for baked products such as biscuits and short

crust pastries where weaker gluten is desirable, however, still remains an option and triticale could be ideal for such products.

Significant correlations were observed between the protein content and the PSI values obtained, implying that hardness is dependant on protein composition, whereas the significant correlation between the protein content and the SDS sedimentation values observed, illustrate that there is a relationship between the gluten strength and the protein content of a sample.

From the results in this study a good indication of the compositional and functional profile of South African triticale cultivars was obtained in addition to the possible effects of genetic and environmental factors. The small sample set (due to only one repetition from each locality being analysed) for the 2006 season made comparison between seasons slightly difficult, as it was not clear whether differences observed were due to differences between seasons, or due to the number of samples being too few to be truly representative. It is thus advisable to expand this study by analysing and including samples from subsequent harvest seasons, not only to extend current results regarding the effect of environmental and genetic factors, but also to increase the knowledge on the compositional and functional profile of South African triticale cultivars. In order to quantify the effects of environment and genetic factors, it is advisable in future to calculate values for the heritability of the parameters being studied.

The results obtained from the conventional analysis methods were used in the development of near infrared (NIR) spectroscopy calibration methods to predict the moisture, protein and ash contents, as well as the hardness and baking potential of South African triticale cultivars (both flour and whole grain) using two NIR instruments (Büchi and Bruker). Full cross-validations were performed for all parameters, and the best prediction models obtained were validated using an independent test set (n = 50). Models with R^2 values >0.66 were validated, as models only become useful for application when R^2 values >0.66 are attained (Williams, 2001). Accurate models for the prediction of protein and moisture content were obtained.

Protein could be predicted accurately (*SEP* = 0.44%; R^2 = 0.96; *RPD* = 5.23) for flour with the Büchi as well as the Bruker (*SEP* = 0.32%; R^2 = 0.96; *RPD* = 4.88). The accuracy of the predictions models were slightly reduced for whole grain analysed on the Büchi (*SEP* = 0.55%; R^2 = 0.94; *RPD* = 4.18) as well as the Bruker (*SEP* = 0.70%; R^2 = 0.90; *RPD* = 3.23). These results imply that the models for flour can be used for quality control and those obtained for whole grain for screening. The results obtained for the protein prediction models compared very well to, and were in some cases an improvement on,

results recently reported by Igne *et al*. (2007a) for triticale (R^2 = 0.92 – 0.96 and *SEP* = 0.30 – 0.34%).

The prediction model obtained for moisture content on flour analysed on the Bruker was rendered suitable for screening purposes (*SEP* = 0.08%; R^2 = 0.95; *RPD* = 4.65). Much poorer results were obtained for moisture prediction with the Büchi instrument compared to the Bruker, potentially due to the fact that spectra obtained using the Büchi instrument could only be acquired a few weeks after spectra were obtained with the Bruker instrument, and the moisture content of the samples could possibly have changed during that time. In addition, the samples were not analysed with the reference method at the same time as when the spectra were collected. Thus the moisture content of the samples, when analysed by NIR, could have differed slightly from the reference values obtained with the AACC method, resulting in a lower correlation between reference and spectral data. Results obtained from the Bruker data compared well with results obtained by Igne *et al.* (2007 a; b) (R^2 = 0.82 – 0.98; *SEP* = 0.30 – 0.34%).

Results for the prediction of PSI, ash content and SDS sedimentation did not yield acceptable models, but this can be improved with the addition of data from subsequent harvest seasons, so as to expand the range. The sample set in this study exhibited a Gaussian distribution for all parameters. Such a distribution is not desired, and sample sets with larger and more evenly spread ranges will possibly yield better models in future. In addition variable (wavelength) selection methods could also be applied and tested. This would result in only the variables that mainly contribute to the model for each parameter being included.

This study provides a comprehensive insight into the compositional (protein and ash content) and functional (1000-kernel mass, falling number, kernel hardness and SDS sedimentation) quality of South African triticale cultivars. These results could be valuable for decision-makers during the development of new cultivars in breeding programmes. This could especially be of value when cultivars are developed for specific purposes, such as animal feed, improved baking quality or for the production of biofuels. The effect of environment and genetic predisposition on these parameters was also reported in this study. Although these results need to be confirmed during subsequent seasons, this information has not been reported before for South African cultivars under local conditions. Apart from the usual NIR calibration models, i.e. protein, moisture and ash contents, NIR calibration models were also developed to predict functional quality parameters, i.e. kernel hardness and SDS sedimentation. Though the latter calibrations were not of adequate accuracy for use in quality control purposes, it could already be valuable for screening of

early generation breeding material when only rough estimations are required. These models also form a good basis for further calibration model development.

References

Alaru, M., Laur, Ü. & Jaama, E. (2003). Influence of nitrogen and weather conditions on the grain quality of winter triticale. *Agronomy Research*, **1**, 3-10.

Alvarez, J.B., Ballesteros, J., Sillero, J.A. & Martin, L.M. (1992). Tritordeum: a new crop of potential importance in the food industry. *Hereditas*, **116**, 193-197.

Barrera, G.N., Pérez, G.T., Ribotta, P.D. & León, A.E. (2007). Influence of damaged starch on cookie and bread-making quality. *European Food Research and Technology*, **225**, 1-7.

Erekul, O. & Köhn, W. (2006). Effect of weather and soil conditions on yield components and bread-making quality of winter wheat (*Triticum aestivum* L.) and winter triticale (*Triticosecale* Wittm.) varieties in north-east Germany. *Journal of Agronomy and Crop Science*, **192**, 452-464.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007a). Triticale moisture and protein content prediction by near-infrared spectroscopy (NIRS). *Cereal Chemistry*, **84**(4), 328-330.

Igne, B., Gibson, L.R., Rippke, G.R., Schwarte, A. & Hurburgh, C.R. (2007b). Influence of yearly variability of agricultural products on calibration process: a triticale example. *Cereal Chemistry*, **84**(6), 576-581.

Kent, N.L. & Evers, A.D. (1994). *Kent's Technology of Cereals: an introduction for students of Food Science and Agriculture*. Pp. 17-18, 50, 96-97, 157. New York, USA: Elsevier Science Ltd.

Leon, A.E., Rubiolo, A. & Anon, M.C. (1996). Use of triticale flours in cookies: quality factors. *Cereal Chemistry*, **71**(6), 779-784.

Martín, A., Alvarez, J.B., Martín, L.M., Barro, F. & Ballesteros, J. (1999). The development of Tritordeum: a novel cereal for food processing. *Journal of Cereal Science*, **30**, 85-95.

Mergoum, M., Pfeiffer, W.H., Peña, R.J., Ammar, K. & Rajaram, S. (2004). Triticale crop improvement: the CIMMYT programme. In: *Triticale improvement and production* (FAO plant production and protection paper 179) (edited by M. Mergoum & H. Gómez-Macpherson). Pp. 11-26. Rome: Food and Agriculture Organisation of the United Nations.

Pfeiffer, W.H. (1994). Triticale improvement strategies at CIMMYT: Existing genetic variability and its implication to projected genetic advance. In: Proceedings of the 5th Portuguese Triticale Conference (as cited by Alaru *et al*., 2003).

Pomeranz, Y. & Williams, P.C. (1990). Wheat hardness: its genetic, structural, and biochemical background, measurement, and significance. In: *Advances in Cereal Science and Technology*, *Vol. 10* (edited by Y. Pomeranz). Pp. 471-557. St Paul, Minnesota: American Association of Cereal Chemists.

Ramírez, A., Pérez, G.T., Ribotta, P.D. & León, A.E. (2003). The occurrence of friabilins in triticale and their relationship with grain hardness and baking quality. *Journal of Agricultural and Food Chemistry*, **51**, 7176-7181.

Stallknecht, G.F., Gilbertson, K.M. & Ranney, J.E. (1996). Alternative wheat cereals as food grains: einkorn, emmer, spelt, kamut, and triticale. In: *Progress in new crops* (edited by J. Janick). Pp. 156-170. Alexandria, Virginia, USA: ASHS Press.

Williams, P.C. (2001). Implementation of near-infrared technology. In: *Near-Infrared Technology in the Agricultural and Food Industries*, 2nd ed. (edited by P. Williams & K. Norris). Pp 145-169. St. Paul, USA: American Association of Cereal Chemists.

APPENDICES

Appendix 1

Table 1.1 Moisture content (%) values for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Napier	Bacchus	11.9	11.9	11.9	11.9
Napier	Tobie	11.9	11.9	11.8	11.9
Napier	Ibis	11.5	11.4	11.5	11.5
Napier	Rex	11.7	11.7	11.7	11.7
Napier	US2007	11.7	11.7	11.7	11.7
Napier	USGen19	11.7	11.7	11.7	11.7
Langewens	Bacchus	11.1	11.1	11.2	11.1
Langewens	Tobie	11.2	11.1	11.1	11.1
Langewens	Ibis	10.9	10.8	10.9	10.9
Langewens	Rex	10.8	10.8	10.8	10.8
Langewens	US2007	11.0	11.0	11.0	11.0
Langewens	USGen19	11.3	11.3	11.3	11.3
Roodebloem	Bacchus	11.9	11.9	11.9	11.9
Roodebloem	Tobie	11.6	11.6	11.5	11.6
Roodebloem	Ibis	11.5	11.5	11.6	11.5
Roodebloem	Rex	11.4	11.4	11.5	11.4
Roodebloem	US2007	11.0	11.0	11.0	11.0
Roodebloem	USGen19	11.7	11.7	11.6	11.7
Mariendahl	Bacchus	11.7	11.7	11.7	11.7
Mariendahl	Tobie	11.8	11.7	11.8	11.8
Mariendahl	Ibis	11.5	11.5	11.5	11.5
Mariendahl	Rex	11.4	11.4	11.4	11.4
Mariendahl	US2007	11.7	11.7	11.7	11.7
Mariendahl	USGen19	11.4	11.4	11.4	11.4
Tygerhoek	Bacchus	11.6	11.5	11.6	11.6
Tygerhoek	Tobie	11.4	11.5	11.4	11.4
Tygerhoek	Ibis	11.0	11.0	11.0	11.0
Tygerhoek	Rex	11.4	11.4	11.3	11.4
Tygerhoek	US2007	11.4	11.4	11.4	11.4
Tygerhoek	USGen19	11.2	11.2	11.2	11.2
Vredenburg	Bacchus	11.9	11.8	11.9	11.9
Vredenburg	Tobie	11.7	11.7	11.7	11.7
Vredenburg	Ibis	11.7	11.6	11.6	11.6
Vredenburg	Rex	11.6	11.6	11.6	11.6
Vredenburg	US2007	11.7	11.7	11.7	11.7
Vredenburg	USGen19	11.7	11.6	11.7	11.7

Table 1.2 Protein content (%) values for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Average
Napier	Bacchus	12.6	12.7	12.7
Napier	Tobie	12.7	12.8	12.7
Napier	Ibis	15.0	15.9	15.5
Napier	Rex	12.8	13.3	13.1
Napier	US2007	14.7	15.1	14.9
Napier	USGen19	14.5	14.9	14.7
Langgewens	Bacchus	13.0	13.2	13.1
Langgewens	Tobie	12.8	12.5	12.7
Langgewens	Ibis	14.6	14.4	14.5
Langgewens	Rex	16.0	15.6	15.8
Langgewens	US2007	14.1	14.4	14.3
Langgewens	USGen19	15.9	16.1	16.0
Roodebloem	Bacchus	12.8	13.2	13.0
Roodebloem	Tobie	13.6	14.3	14.0
Roodebloem	Ibis	13.4	13.5	13.4
Roodebloem	Rex	12.8	12.8	12.8
Roodebloem	US2007	14.8	14.5	14.7
Roodebloem	USGen19	14.3	14.4	14.4
Mariendahl	Bacchus	14.7	13.8	14.3
Mariendahl	Tobie	12.4	12.9	12.7
Mariendahl	Ibis	13.0	12.9	12.9
Mariendahl	Rex	12.9	13.1	13.0
Mariendahl	US2007	14.0	14.3	14.2
Mariendahl	USGen19	15.3	15.5	15.4
Tygerhoek	Bacchus	13.9	14.0	13.9
Tygerhoek	Tobie	14.0	12.9	13.5
Tygerhoek	Ibis	14.0	14.3	14.2
Tygerhoek	Rex	14.8	14.0	14.4
Tygerhoek	US2007	15.5	15.8	15.7
Tygerhoek	USGen19	14.4	14.0	14.2
Vredenburg	Bacchus	13.3	13.6	13.4
Vredenburg	Tobie	13.1	13.8	13.5
Vredenburg	Ibis	14.4	12.9	13.6
Vredenburg	Rex	14.5	14.8	14.6
Vredenburg	US2007	14.3	14.6	14.4
Vredenburg	USGen19	13.6	13.2	13.4

Table 1.3 Ash content (%) values for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Napier	Bacchus	2.02	2.00	1.99	2.00
Napier	Tobie	2.09	2.07	2.08	2.08
Napier	Ibis	1.86	1.87	1.86	1.86
Napier	Rex	1.95	1.97	1.96	1.96
Napier	US2007	1.93	1.93	1.92	1.93
Napier	USGen19	1.95	1.94	1.94	1.94
Langgewens	Bacchus	1.66	1.63	1.66	1.65
Langgewens	Tobie	1.68	1.65	1.69	1.67
Langgewens	Ibis	1.55	1.56	1.55	1.55
Langgewens	Rex	1.63	1.63	1.65	1.64
Langgewens	US2007	1.79	1.79	1.79	1.79
Langgewens	USGen19	1.80	1.80	1.77	1.79
Roodebloem	Bacchus	1.80	1.80	1.78	1.79
Roodebloem	Tobie	1.95	1.98	1.95	1.96
Roodebloem	Ibis	1.87	1.89	1.87	1.87
Roodebloem	Rex	1.92	1.94	1.94	1.93
Roodebloem	US2007	2.00	1.99	2.03	2.01
Roodebloem	USGen19	1.80	1.78	1.78	1.79
Mariendahl	Bacchus	2.07	2.11	2.09	2.09
Mariendahl	Tobie	2.18	2.16	2.14	2.16
Mariendahl	Ibis	2.09	2.11	2.10	2.10
Mariendahl	Rex	2.14	2.12	2.11	2.12
Mariendahl	US2007	2.22	2.16	2.18	2.19
Mariendahl	USGen19	2.15	2.16	2.15	2.15
Tygerhoek	Bacchus	2.04	2.04	2.02	2.04
Tygerhoek	Tobie	2.07	2.08	2.06	2.07
Tygerhoek	Ibis	2.15	2.14	2.16	2.15
Tygerhoek	Rex	2.18	2.19	2.15	2.17
Tygerhoek	US2007	2.07	2.09	2.08	2.08
Tygerhoek	USGen19	2.11	2.11	2.10	2.11
Vredenburg	Bacchus	1.62	1.62	1.61	1.62
Vredenburg	Tobie	1.78	7.19	1.78	1.78
Vredenburg	Ibis	1.52	1.54	1.53	1.53
Vredenburg	Rex	1.82	1.84	1.86	1.84
Vredenburg	US2007	1.83	1.84	1.85	1.84
Vredenburg	USGen19	1.73	1.72	1.70	1.72

Table 1.4 Falling number (s) values for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Napier	Bacchus	105	110	102	106
Napier	Tobie	217	223	226	222
Napier	Ibis	109	113	106	109
Napier	Rex	146	147	130	141
Napier	US2007	73	80	80	78
Napier	USGen19	200	205	202	202
Langewens	Bacchus	91	88	93	91
Langewens	Tobie	215	227	179	207
Langewens	Ibis	167	174	177	173
Langewens	Rex	208	211	212	210
Langewens	US2007	92	92	94	93
Langewens	USGen19	166	163	158	162
Roodebloem	Bacchus	65	66	65	65
Roodebloem	Tobie	192	192	191	192
Roodebloem	Ibis	66	66	66	66
Roodebloem	Rex	109	106	110	108
Roodebloem	US2007	63	62	62	62
Roodebloem	USGen19	159	148	153	153
Mariendahl	Bacchus	180	190	175	182
Mariendahl	Tobie	186	187	184	186
Mariendahl	Ibis	213	216	215	215
Mariendahl	Rex	234	237	241	237
Mariendahl	US2007	138	147	149	145
Mariendahl	USGen19	280	286	286	284
Tygerhoek	Bacchus	66	66	65	66
Tygerhoek	Tobie	77	75	72	75
Tygerhoek	Ibis	62	62	62	62
Tygerhoek	Rex	66	68	67	67
Tygerhoek	US2007	62	62	62	62
Tygerhoek	USGen19	93	95	87	92
Vredenburg	Bacchus	87	80	81	83
Vredenburg	Tobie	202	208	209	206
Vredenburg	Ibis	150	151	136	146
Vredenburg	Rex	203	205	199	202
Vredenburg	US2007	77	80	84	80
Vredenburg	USGen19	284	278	293	285

Table 1.5 Particle size index values for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Average
Napier	Bacchus	70.55	70.91	70.73
Napier	Tobie	68.99	69.44	69.22
Napier	Ibis	63.75	63.82	63.79
Napier	Rex	65.70	64.47	65.09
Napier	US2007	65.40	65.71	65.56
Napier	USGen19	62.64	62.77	62.71
Langgewens	Bacchus	69.55	70.21	69.88
Langgewens	Tobie	65.37	65.48	65.43
Langgewens	Ibis	62.49	62.45	62.47
Langgewens	Rex	65.43	64.97	65.20
Langgewens	US2007	63.93	63.81	63.87
Langgewens	USGen19	62.77	62.38	62.58
Roodebloem	Bacchus	66.68	66.66	66.67
Roodebloem	Tobie	64.08	64.10	64.09
Roodebloem	Ibis	60.60	60.38	60.49
Roodebloem	Rex	63.65	62.31	62.98
Roodebloem	US2007	63.56	64.23	63.90
Roodebloem	USGen19	63.26	62.83	63.05
Mariendahl	Bacchus	69.39	68.83	69.11
Mariendahl	Tobie	65.69	65.55	65.62
Mariendahl	Ibis	62.83	62.77	62.80
Mariendahl	Rex	63.62	62.83	63.23
Mariendahl	US2007	62.66	62.92	62.79
Mariendahl	USGen19	59.28	58.69	58.99
Tygerhoek	Bacchus	68.52	68.53	68.53
Tygerhoek	Tobie	65.89	65.12	65.51
Tygerhoek	Ibis	60.26	60.12	60.19
Tygerhoek	Rex	61.60	61.60	61.60
Tygerhoek	US2007	63.16	62.53	62.85
Tygerhoek	USGen19	60.43	61.19	60.81
Vredenburg	Bacchus	71.99	71.65	71.82
Vredenburg	Tobie	66.56	65.96	66.26
Vredenburg	Ibis	67.95	68.17	68.06
Vredenburg	Rex	67.70	66.69	67.20
Vredenburg	US2007	69.49	69.47	69.48
Vredenburg	USGen19	69.04	69.04	69.04

Table 1.6 Values for 1000-kernel mass (g) for 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Average
Napier	Bacchus	43.32	43.27	43.30
Napier	Tobie	36.80	36.25	36.53
Napier	Ibis	55.36	54.88	55.12
Napier	Rex	50.58	49.98	50.28
Napier	US2007	54.09	53.59	53.84
Napier	USGen19	44.16	43.83	44.00
Langgewens	Bacchus	41.18	41.18	41.18
Langgewens	Tobie	47.69	46.39	47.04
Langgewens	Ibis	45.76	47.23	46.50
Langgewens	Rex	44.40	42.93	43.67
Langgewens	US2007	49.72	50.28	50.00
Langgewens	USGen19	39.89	40.93	40.41
Roodebloem	Bacchus	39.40	41.13	40.27
Roodebloem	Tobie	42.85	42.36	42.61
Roodebloem	Ibis	46.12	46.89	46.51
Roodebloem	Rex	41.41	43.11	42.26
Roodebloem	US2007	44.37	44.77	44.57
Roodebloem	USGen19	43.79	45.56	44.68
Mariendahl	Bacchus	35.95	35.64	35.80
Mariendahl	Tobie	44.85	43.74	44.30
Mariendahl	Ibis	43.76	42.77	43.27
Mariendahl	Rex	43.94	43.70	43.82
Mariendahl	US2007	44.58	45.28	44.93
Mariendahl	USGen19	35.44	34.76	35.10
Tygerhoek	Bacchus	34.56	35.07	34.82
Tygerhoek	Tobie	39.09	41.89	40.49
Tygerhoek	Ibis	43.56	46.37	44.97
Tygerhoek	Rex	44.78	46.84	45.81
Tygerhoek	US2007	42.92	42.77	42.85
Tygerhoek	USGen19	40.20	40.09	40.15
Vredenburg	Bacchus	39.76	39.09	39.43
Vredenburg	Tobie	43.35	43.05	43.20
Vredenburg	Ibis	42.81	44.22	43.52
Vredenburg	Rex	40.61	40.14	40.38
Vredenburg	US2007	43.58	43.85	43.72
Vredenburg	USGen19	41.08	40.79	40.94

Table 1.7 SDS sedimentation (mm) values for the 2006 harvest season

Locality	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Napier	Bacchus	23	24	24	24
Napier	Tobie	21	21	20	21
Napier	Ibis	32	35	33	33
Napier	Rex	22	22	23	22
Napier	US2007	31	32	32	32
Napier	USGen19	30	30	29	30
Langgewens	Bacchus	33	32	33	33
Langgewens	Tobie	29	29	29	29
Langgewens	Ibis	40	42	40	41
Langgewens	Rex	33	34	33	33
Langgewens	US2007	42	42	41	42
Langgewens	USGen19	35	37	37	36
Roodebloem	Bacchus	26	27	25	26
Roodebloem	Tobie	23	21	22	22
Roodebloem	Ibis	33	32	32	32
Roodebloem	Rex	26	27	26	26
Roodebloem	US2007	33	34	33	33
Roodebloem	USGen19	30	30	29	30
Mariendahl	Bacchus	35	37	37	36
Mariendahl	Tobie	23	25	25	24
Mariendahl	Ibis	33	34	33	33
Mariendahl	Rex	35	33	33	34
Mariendahl	US2007	40	41	42	41
Mariendahl	USGen19	37	38	36	37
Tygerhoek	Bacchus	32	31	32	32
Tygerhoek	Tobie	25	25	25	25
Tygerhoek	Ibis	36	36	36	36
Tygerhoek	Rex	33	32	32	32
Tygerhoek	US2007	38	38	39	38
Tygerhoek	USGen19	35	36	34	35
Vredenburg	Bacchus	31	32	30	31
Vredenburg	Tobie	26	24	25	25
Vredenburg	Ibis	41	40	39	40
Vredenburg	Rex	33	32	32	32
Vredenburg	US2007	42	43	44	43
Vredenburg	USGen19	33	32	33	33

Appendix 2

Table 2.1 Moisture content (%) values for 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	10.6	10.6	10.6	10.6
Riversdal	1	AgBeacon	10.8	10.8	nd[#]	10.8
Riversdal	1	Bacchus	10.6	10.7	nd	10.6
Riversdal	1	Tobie	10.9	10.9	nd	10.9
Riversdal	1	Rex	10.8	10.8	10.8	10.8
Riversdal	1	Ibis	10.8	10.8	10.8	10.8
Riversdal	2	US2007	10.8	10.8	10.8	10.8
Riversdal	2	AgBeacon	10.9	10.9	10.9	10.9
Riversdal	2	Bacchus	10.8	10.7	nd	10.7
Riversdal	2	Tobie	10.7	10.7	10.7	10.7
Riversdal	2	Rex	10.6	10.6	10.6	10.6
Riversdal	2	Ibis	10.8	10.8	nd	10.8
Riversdal	3	US2007	10.2	10.2	nd	10.2
Riversdal	3	AgBeacon	10.9	10.9	10.9	10.9
Riversdal	3	Bacchus	10.8	10.9	nd	10.8
Riversdal	3	Tobie	10.6	10.6	nd	10.6
Riversdal	3	Rex	10.2	10.2	10.2	10.2
Riversdal	3	Ibis	10.7	10.7	nd	10.7
Riversdal	1	Wheat (Kariega)	11.4	11.4	11.4	11.4
Riversdal	2	Wheat (Kariega)	11.3	11.4	11.4	11.4
Riversdal	3	Wheat (Kariega)	11.2	11.2	11.2	11.2
Napier	1	US2007	10.7	10.7	10.7	10.7
Napier	1	AgBeacon	10.7	10.7	nd	10.7
Napier	1	Bacchus	10.7	10.7	10.7	10.7
Napier	1	Tobie	10.6	10.6	10.7	10.6
Napier	1	Rex	10.6	10.6	10.6	10.6
Napier	1	Ibis	11.0	11.0	11.0	11.0
Napier	2	US2007	10.8	10.9	nd	10.8
Napier	2	AgBeacon	10.7	10.7	10.7	10.7
Napier	2	Bacchus	10.9	10.9	nd	10.9
Napier	2	Tobie	11.3	nd	nd	11.3
Napier	2	Rex	10.8	10.8	10.8	10.8
Napier	2	Ibis	10.9	10.9	nd	10.9
Napier	3	US2007	11.3	11.3	11.2	11.3
Napier	3	AgBeacon	10.9	10.9	10.8	10.9
Napier	3	Bacchus	11.0	11.0	11.0	11.0
Napier	3	Tobie	11.0	10.9	nd	11.0
Napier	3	Rex	10.8	10.7	10.7	10.7
Napier	3	Ibis	10.9	10.9	10.8	10.9
Napier	1	Wheat (Kariega)	11.5	11.4	11.4	11.4
Napier	2	Wheat (Kariega)	11.4	11.5	11.5	11.5
Napier	3	Wheat (Kariega)	11.4	11.5	11.5	11.5
Langgewens	1	US2007	11.1	11.1	11.1	11.1
Langgewens	1	AgBeacon	10.8	10.8	10.8	10.8
Langgewens	1	Bacchus	11.2	11.2	nd	11.2
Langgewens	1	Tobie	11.1	11.0	11.1	11.1
Langgewens	1	Rex	10.8	10.8	nd	10.8
Langgewens	1	Ibis	11.0	10.9	10.9	10.9

Table 2.1 continued

Langgewens	2	US2007	11.1	11.1	11.0	11.0
Langgewens	2	AgBeacon	10.9	11.0	nd	10.9
Langgewens	2	Bacchus	11.3	11.3	11.3	11.3
Langgewens	2	Tobie	11.3	11.3	nd	11.3
Langgewens	2	Rex	11.0	11.0	11.0	11.0
Langgewens	2	Ibis	11.0	11.0	nd	11.0
Langgewens	3	US2007	11.3	11.3	nd	11.3
Langgewens	3	AgBeacon	11.3	11.2	11.2	11.3
Langgewens	3	Bacchus	11.4	11.4	11.4	11.4
Langgewens	3	Tobie	11.4	11.4	11.4	11.4
Langgewens	3	Rex	11.0	11.0	11.0	11.0
Langgewens	3	Ibis	11.2	11.2	nd	11.2
Langgewens	1	Wheat (Kariega)	11.8	11.8	11.9	11.9
Langgewens	2	Wheat (Kariega)	11.8	11.8	11.8	11.8
Langgewens	3	Wheat (Kariega)	11.3	11.3	nd	11.3
Roodebloem	1	US2007	12.0	12.0	12.0	12.0
Roodebloem	1	AgBeacon	11.1	11.1	11.1	11.1
Roodebloem	1	Bacchus	11.2	11.2	11.1	11.2
Roodebloem	1	Tobie	11.0	11.0	10.9	11.0
Roodebloem	1	Rex	11.1	11.1	11.1	11.1
Roodebloem	1	Ibis	11.3	11.4	11.4	11.4
Roodebloem	2	US2007	11.8	11.6	11.8	11.7
Roodebloem	2	AgBeacon	11.3	11.3	11.3	11.3
Roodebloem	2	Bacchus	11.7	11.7	11.7	11.7
Roodebloem	2	Tobie	11.7	11.7	11.7	11.7
Roodebloem	2	Rex	11.7	11.7	11.7	11.7
Roodebloem	2	Ibis	11.6	11.6	11.6	11.6
Roodebloem	3	US2007	11.7	11.7	11.7	11.7
Roodebloem	3	AgBeacon	11.6	11.6	11.6	11.6
Roodebloem	3	Bacchus	11.7	11.8	11.8	11.8
Roodebloem	3	Tobie	11.7	11.7	11.8	11.7
Roodebloem	3	Rex	11.5	11.5	11.5	11.5
Roodebloem	3	Ibis	11.3	11.2	11.3	11.3
Roodebloem	1	Wheat (Kariega)	11.2	11.2	11.1	11.2
Roodebloem	2	Wheat (Kariega)	11.2	11.2	11.2	11.2
Roodebloem	3	Wheat (Kariega)	11.5	11.5	11.5	11.5
Piketberg	1	US2007	11.0	11.0	11.0	11.0
Piketberg	1	AgBeacon	10.8	10.8	10.8	10.8
Piketberg	1	Bacchus	10.6	10.7	10.6	10.6
Piketberg	1	Tobie	10.9	10.9	10.9	10.9
Piketberg	1	Rex	10.9	10.9	11.1	11.0
Piketberg	1	Ibis	11.1	11.1	11.1	11.1
Piketberg	2	US2007	10.8	10.8	10.8	10.8
Piketberg	2	AgBeacon	10.5	10.5	10.5	10.5
Piketberg	2	Bacchus	10.5	10.5	10.5	10.5
Piketberg	2	Tobie	10.8	10.8	10.8	10.8
Piketberg	2	Rex	10.7	10.7	10.7	10.7
Piketberg	2	Ibis	10.6	10.6	10.6	10.6
Piketberg	3	US2007	10.6	10.6	10.7	10.6
Piketberg	3	AgBeacon	10.4	10.4	10.5	10.4
Piketberg	3	Bacchus	11.2	11.2	11.2	11.2
Piketberg	3	Tobie	10.7	10.8	10.8	10.8
Piketberg	3	Rex	10.6	10.6	10.6	10.6
Piketberg	3	Ibis	10.5	10.5	10.5	10.5

Table 2.1 continued

Piketberg	1	Wheat (Kariega)	10.9	10.9	10.9	10.9
Piketberg	2	Wheat (Kariega)	11.0	11.0	11.0	11.0
Piketberg	3	Wheat (Kariega)	11.0	11.0	10.9	11.0
Klipheuwel	1	US2007	11.3	11.3	11.3	11.3
Klipheuwel	1	AgBeacon	11.3	11.3	11.3	11.3
Klipheuwel	1	Bacchus	11.2	11.2	11.2	11.2
Klipheuwel	1	Tobie	11.5	11.5	11.5	11.5
Klipheuwel	1	Rex	11.3	11.3	11.4	11.3
Klipheuwel	1	Ibis	11.3	11.3	11.3	11.3
Klipheuwel	2	US2007	11.3	11.3	11.2	11.2
Klipheuwel	2	AgBeacon	11.6	11.6	11.6	11.6
Klipheuwel	2	Bacchus	11.4	11.4	11.4	11.4
Klipheuwel	2	Tobie	11.6	11.5	11.5	11.5
Klipheuwel	2	Rex	11.0	11.0	11.0	11.0
Klipheuwel	2	Ibis	11.3	11.3	11.3	11.3
Klipheuwel	3	US2007	11.6	11.6	11.9	11.7
Klipheuwel	3	AgBeacon	11.4	11.4	11.4	11.4
Klipheuwel	3	Bacchus	11.2	11.2	11.2	11.2
Klipheuwel	3	Tobie	nd	11.4	11.4	11.4
Klipheuwel	3	Rex	11.2	11.2	11.2	11.2
Klipheuwel	3	Ibis	11.2	11.2	11.2	11.2
Klipheuwel	1	Wheat (Kariega)	11.8	11.8	11.8	11.8
Klipheuwel	2	Wheat (Kariega)	11.6	11.5	11.6	11.6
Klipheuwel	3	Wheat (Kariega)	11.6	11.6	11.6	11.6
Mariendahl	1	US2007	10.5	10.5	10.5	10.5
Mariendahl	1	AgBeacon	11.0	11.1	11.1	11.0
Mariendahl	1	Bacchus	10.9	10.9	10.9	10.9
Mariendahl	1	Tobie	10.9	10.9	11.0	10.9
Mariendahl	1	Rex	11.0	11.0	11.0	11.0
Mariendahl	1	Ibis	11.0	11.0	11.0	11.0
Mariendahl	2	US2007	10.8	10.8	10.8	10.8
Mariendahl	2	AgBeacon	10.8	10.7	10.7	10.7
Mariendahl	2	Bacchus	10.9	10.9	11.0	10.9
Mariendahl	2	Tobie	10.8	10.8	10.8	10.8
Mariendahl	2	Rex	11.0	11.0	11.0	11.0
Mariendahl	2	Ibis	11.1	11.1	11.2	11.1
Mariendahl	3	US2007	10.4	10.4	10.4	10.4
Mariendahl	3	AgBeacon	10.9	10.9	10.9	10.9
Mariendahl	3	Bacchus	10.8	10.8	10.8	10.8
Mariendahl	3	Tobie	10.7	10.7	10.7	10.7
Mariendahl	3	Rex	10.8	10.8	10.7	10.8
Mariendahl	3	Ibis	10.4	10.4	10.4	10.4
Albertinia	1	US2007	12.0	12.0	12.0	12.0
Albertinia	1	AgBeacon	11.6	11.5	11.6	11.6
Albertinia	1	Bacchus	11.6	11.6	11.6	11.6
Albertinia	1	Tobie	11.6	11.6	11.6	11.6
Albertinia	1	Rex	11.1	11.1	11.1	11.1
Albertinia	1	Ibis	11.5	11.5	11.5	11.5
Albertinia	2	US2007	11.5	11.5	11.5	11.5
Albertinia	2	AgBeacon	11.7	11.6	11.7	11.7
Albertinia	2	Bacchus	12.0	12.0	12.0	12.0
Albertinia	2	Tobie	11.3	11.3	11.3	11.3
Albertinia	2	Rex	11.8	11.8	11.8	11.8
Albertinia	2	Ibis	11.8	11.8	11.8	11.8

Table 2.1 continued

Albertinia	3	US2007	12.0	12.1	12.1	12.1
Albertinia	3	AgBeacon	12.4	12.4	12.4	12.4
Albertinia	3	Bacchus	11.8	11.8	11.9	11.9
Albertinia	3	Tobie	12.2	12.2	12.2	12.2
Albertinia	3	Rex	11.4	11.4	11.4	11.4
Albertinia	3	Ibis	11.4	11.4	11.4	11.4
Albertinia	1	Wheat (Kariega)	11.4	11.4	11.4	11.4
Albertinia	2	Wheat (Kariega)	11.8	11.8	11.8	11.8
Albertinia	3	Wheat (Kariega)	11.7	11.7	11.7	11.7
Tygerhoek	1	US2007	11.7	11.7	11.7	11.7
Tygerhoek	1	AgBeacon	11.8	11.8	nd	11.8
Tygerhoek	1	Bacchus	11.8	11.8	11.8	11.8
Tygerhoek	1	Tobie	11.6	11.6	11.6	11.6
Tygerhoek	1	Rex	11.6	11.5	11.6	11.6
Tygerhoek	1	Ibis	11.5	11.5	11.5	11.5
Tygerhoek	2	US2007	12.0	11.9	11.9	11.9
Tygerhoek	2	AgBeacon	11.7	11.7	11.8	11.7
Tygerhoek	2	Bacchus	11.4	11.4	11.4	11.4
Tygerhoek	2	Tobie	11.6	11.6	11.6	11.6
Tygerhoek	2	Rex	11.5	11.5	11.5	11.5
Tygerhoek	2	Ibis	11.6	11.6	11.6	11.6
Tygerhoek	3	US2007	11.7	11.8	11.8	11.8
Tygerhoek	3	AgBeacon	11.8	11.7	11.7	11.8
Tygerhoek	3	Bacchus	11.8	11.8	11.8	11.8
Tygerhoek	3	Tobie	11.5	11.5	11.5	11.5
Tygerhoek	3	Rex	11.6	11.6	11.6	11.6
Tygerhoek	3	Ibis	11.5	11.5	11.6	11.5
Tygerhoek	1	Koring	11.5	11.5	11.5	11.5
Tygerhoek	2	Koring	11.6	11.6	11.6	11.6
Tygerhoek	3	Koring	nd	11.4	11.4	11.4

[#]not determined due to insufficient sample

Table 2.2 Protein content (%) values for 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	14.3	14.2	14.2	14.2
Riversdal	1	AgBeacon	12.2	12.0	12.1	12.1
Riversdal	1	Bacchus	12.6	12.5	12.5	12.5
Riversdal	1	Tobie	12.9	12.8	12.8	12.9
Riversdal	1	Rex	13.9	14.0	14.4	13.9
Riversdal	1	Ibis	13.3	13.3	13.3	13.3
Riversdal	2	US2007	12.8	12.9	12.7	12.8
Riversdal	2	AgBeacon	13.0	13.0	12.9	12.9
Riversdal	2	Bacchus	12.0	12.1	11.9	12.0
Riversdal	2	Tobie	12.8	12.8	12.8	12.8
Riversdal	2	Rex	10.9	11.1	10.9	11.0
Riversdal	2	Ibis	11.6	11.6	11.6	11.6
Riversdal	3	US2007	11.3	11.3	11.3	11.3
Riversdal	3	AgBeacon	12.5	12.5	12.5	12.5
Riversdal	3	Bacchus	12.8	12.7	12.8	12.7
Riversdal	3	Tobie	13.3	13.5	13.1	13.4
Riversdal	3	Rex	13.0	12.9	12.8	12.9
Riversdal	3	Ibis	11.9	11.9	11.9	11.9
Riversdal	1	Wheat (Kariega)	17.0	17.0	16.7	16.9
Riversdal	2	Wheat (Kariega)	14.8	14.9	14.7	14.8
Riversdal	3	Wheat (Kariega)	13.9	13.8	13.8	13.8
Napier	1	US2007	8.2	8.2	8.2	8.2
Napier	1	AgBeacon	8.2	8.4	8.5	8.3
Napier	1	Bacchus	9.2	9.0	9.0	9.0
Napier	1	Tobie	9.0	9.1	9.0	9.0
Napier	1	Rex	9.3	9.2	9.2	9.2
Napier	1	Ibis	8.6	8.6	8.6	8.6
Napier	2	US2007	9.1	9.1	9.2	9.1
Napier	2	AgBeacon	7.8	7.8	7.9	7.8
Napier	2	Bacchus	9.5	9.2	9.4	9.5
Napier	2	Tobie	8.7	8.5	8.6	8.6
Napier	2	Rex	9.4	9.5	9.6	9.5
Napier	2	Ibis	9.5	9.5	9.5	9.5
Napier	3	US2007	9.5	9.2	9.4	9.3
Napier	3	AgBeacon	8.2	8.1	8.3	8.2
Napier	3	Bacchus	8.8	8.7	8.7	8.7
Napier	3	Tobie	8.8	8.9	8.8	8.8
Napier	3	Rex	8.7	8.6	8.7	8.7
Napier	3	Ibis	8.6	8.5	8.5	8.5
Napier	1	Wheat (Kariega)	9.6	9.6	9.6	9.6
Napier	2	Wheat (Kariega)	11.3	10.9	11.3	11.3
Napier	3	Wheat (Kariega)	11.8	11.8	11.8	11.8
Langgewens	1	US2007	12.9	12.8	12.9	12.9
Langgewens	1	AgBeacon	10.7	10.6	10.7	10.7
Langgewens	1	Bacchus	9.6	9.5	9.5	9.5
Langgewens	1	Tobie	10.2	10.2	10.1	10.1
Langgewens	1	Rex	10.3	10.5	10.6	10.5
Langgewens	1	Ibis	11.5	11.7	11.6	11.6
Langgewens	2	US2007	10.8	10.9	10.8	10.8
Langgewens	2	AgBeacon	9.8	9.9	9.8	9.8
Langgewens	2	Bacchus	9.9	9.9	9.9	9.9
Langgewens	2	Tobie	10.4	10.6	10.4	10.5
Langgewens	2	Rex	9.9	9.9	9.9	9.9

Table 2.2 continued

Langgewens	2	Ibis	11.0	10.7	10.9	10.9
Langgewens	3	US2007	12.0	11.9	11.6	11.9
Langgewens	3	AgBeacon	9.5	9.4	9.3	9.4
Langgewens	3	Bacchus	9.3	9.3	9.3	9.3
Langgewens	3	Tobie	10.3	10.4	10.6	10.4
Langgewens	3	Rex	11.3	11.3	11.3	11.3
Langgewens	3	Ibis	10.2	10.2	10.1	10.2
Langgewens	1	Wheat (Kariega)	11.5	11.5	11.6	11.5
Langgewens	2	Wheat (Kariega)	8.1	8.2	8.1	8.1
Langgewens	3	Wheat (Kariega)	10.0	10.1	10.0	10.0
Roodebloem	1	US2007	9.1	9.0	9.1	9.0
Roodebloem	1	AgBeacon	nd	8.9	8.9	8.9
Roodebloem	1	Bacchus	8.8	8.7	8.8	8.8
Roodebloem	1	Tobie	8.6	8.6	8.5	8.6
Roodebloem	1	Rex	9.1	9.3	9.3	9.2
Roodebloem	1	Ibis	8.9	8.9	8.9	8.9
Roodebloem	2	US2007	9.1	9.3	9.0	9.2
Roodebloem	2	AgBeacon	10.5	10.5	10.5	10.5
Roodebloem	2	Bacchus	9.9	9.9	9.6	9.8
Roodebloem	2	Tobie	8.5	9.2	8.4	8.5
Roodebloem	2	Rex	10.7	10.7	10.7	10.7
Roodebloem	2	Ibis	9.3	9.2	9.2	9.2
Roodebloem	3	US2007	11.4	11.5	11.5	11.5
Roodebloem	3	AgBeacon	9.9	9.9	9.9	9.9
Roodebloem	3	Bacchus	8.9	8.9	8.9	8.9
Roodebloem	3	Tobie	9.5	9.2	9.4	9.3
Roodebloem	3	Rex	10.9	11.1	11.0	11.0
Roodebloem	3	Ibis	10.4	10.1	10.4	10.3
Roodebloem	1	Wheat (Kariega)	9.8	9.8	9.8	9.8
Roodebloem	2	Wheat (Kariega)	10.7	10.4	10.5	10.5
Roodebloem	3	Wheat (Kariega)	9.5	9.6	nd	9.6
Piketberg	1	US2007	7.5	7.6	7.6	7.6
Piketberg	1	AgBeacon	7.9	7.9	7.9	7.9
Piketberg	1	Bacchus	7.9	nd	7.8	7.8
Piketberg	1	Tobie	7.5	7.5	7.5	7.5
Piketberg	1	Rex	8.8	8.7	8.9	8.8
Piketberg	1	Ibis	7.8	7.8	8.0	7.9
Piketberg	2	US2007	9.1	9.0	9.4	9.2
Piketberg	2	AgBeacon	9.4	9.5	9.3	9.4
Piketberg	2	Bacchus	8.2	8.2	8.1	8.2
Piketberg	2	Tobie	8.5	8.5	8.5	8.5
Piketberg	2	Rex	7.7	7.7	7.8	7.8
Piketberg	2	Ibis	8.2	8.2	8.2	8.2
Piketberg	3	US2007	8.8	8.8	8.8	8.8
Piketberg	3	AgBeacon	7.9	8.0	8.0	8.0
Piketberg	3	Bacchus	8.2	8.1	8.1	8.1
Piketberg	3	Tobie	8.2	8.1	8.1	8.1
Piketberg	3	Rex	8.1	8.0	8.1	8.0
Piketberg	3	Ibis	8.9	8.9	8.7	8.9
Piketberg	1	Wheat (Kariega)	9.1	9.0	9.2	9.1
Piketberg	2	Wheat (Kariega)	8.7	8.8	8.7	8.7
Piketberg	3	Wheat (Kariega)	10.4	10.6	10.6	10.5
Klipheuwel	1	US2007	9.7	9.9	9.7	9.7
Klipheuwel	1	AgBeacon	9.0	9.0	9.0	9.0

Table 2.2 continued

Klipheuwel	1	Bacchus	8.1	8.1	8.2	8.1
Klipheuwel	1	Tobie	9.2	9.1	9.0	9.1
Klipheuwel	1	Rex	10.1	10.2	10.2	10.2
Klipheuwel	1	Ibis	nd	10.2	10.2	10.2
Klipheuwel	2	US2007	9.1	9.0	9.1	9.1
Klipheuwel	2	AgBeacon	8.4	8.5	8.4	8.5
Klipheuwel	2	Bacchus	9.6	9.6	9.8	9.7
Klipheuwel	2	Tobie	8.8	8.8	8.8	8.8
Klipheuwel	2	Rex	9.2	9.1	9.3	9.2
Klipheuwel	2	Ibis	8.7	8.7	8.6	8.7
Klipheuwel	3	US2007	10.6	10.7	10.6	10.6
Klipheuwel	3	AgBeacon	8.7	8.7	9.0	8.8
Klipheuwel	3	Bacchus	8.3	8.3	8.3	8.3
Klipheuwel	3	Tobie	8.6	8.6	8.7	8.7
Klipheuwel	3	Rex	8.7	8.7	8.7	8.7
Klipheuwel	3	Ibis	8.3	8.2	8.1	8.2
Klipheuwel	1	Wheat (Kariega)	8.7	8.8	8.9	8.8
Klipheuwel	2	Wheat (Kariega)	8.2	8.2	8.3	8.2
Klipheuwel	3	Wheat (Kariega)	9.4	9.2	9.4	9.4
Mariendahl	1	US2007	9.7	9.7	9.7	9.7
Mariendahl	1	AgBeacon	8.5	8.4	8.4	8.4
Mariendahl	1	Bacchus	8.2	8.1	8.1	8.1
Mariendahl	1	Tobie	9.4	9.6	9.4	9.5
Mariendahl	1	Rex	8.6	8.5	8.6	8.6
Mariendahl	1	Ibis	9.0	9.3	9.2	9.2
Mariendahl	2	US2007	9.4	9.4	9.4	9.4
Mariendahl	2	AgBeacon	8.6	8.5	8.5	8.6
Mariendahl	2	Bacchus	7.9	7.9	8.0	8.0
Mariendahl	2	Tobie	9.4	9.2	9.3	9.3
Mariendahl	2	Rex	9.2	9.2	9.1	9.2
Mariendahl	2	Ibis	8.6	8.5	8.6	8.6
Mariendahl	3	US2007	9.0	9.0	9.0	9.0
Mariendahl	3	AgBeacon	8.8	8.8	8.9	8.8
Mariendahl	3	Bacchus	7.8	7.8	7.8	7.8
Mariendahl	3	Tobie	13.0	12.9	12.8	12.9
Mariendahl	3	Rex	11.3	11.3	11.2	11.3
Mariendahl	3	Ibis	9.0	8.7	8.8	8.8
Albertinia	1	US2007	12.4	12.5	12.3	12.4
Albertinia	1	AgBeacon	10.8	10.9	11.0	10.9
Albertinia	1	Bacchus	9.6	9.6	9.6	9.6
Albertinia	1	Tobie	10.5	10.6	10.6	10.6
Albertinia	1	Rex	11.6	11.6	11.6	11.6
Albertinia	1	Ibis	12.0	12.0	12.1	12.0
Albertinia	2	US2007	11.4	11.3	11.3	11.3
Albertinia	2	AgBeacon	11.7	11.8	11.6	11.7
Albertinia	2	Bacchus	11.3	11.5	11.6	11.5
Albertinia	2	Tobie	11.5	11.4	11.4	11.4
Albertinia	2	Rex	12.9	12.8	12.8	12.8
Albertinia	2	Ibis	12.1	12.1	12.3	12.2
Albertinia	3	US2007	11.2	11.2	11.0	11.1
Albertinia	3	AgBeacon	10.6	10.5	10.6	10.6
Albertinia	3	Bacchus	12.2	12.2	12.1	12.2
Albertinia	3	Tobie	10.6	10.2	10.5	10.6
Albertinia	3	Rex	11.5	11.4	11.6	11.5

Table 2.2 continued

Albertinia	3	Ibis	13.0	13.0	13.0	13.0
Albertinia	1	Wheat (Kariega)	13.2	13.2	13.3	13.2
Albertinia	2	Wheat (Kariega)	13.3	13.2	13.2	13.3
Albertinia	3	Wheat (Kariega)	14.0	13.8	13.9	13.9
Tygerhoek	1	US2007	13.7	13.6	13.7	13.7
Tygerhoek	1	AgBeacon	12.1	12.1	12.0	12.1
Tygerhoek	1	Bacchus	11.9	12.0	12.0	12.0
Tygerhoek	1	Tobie	11.9	12.0	11.8	11.9
Tygerhoek	1	Rex	13.4	13.4	nd	13.4
Tygerhoek	1	Ibis	13.8	13.8	13.8	13.8
Tygerhoek	2	US2007	12.7	12.7	12.7	12.7
Tygerhoek	2	AgBeacon	11.8	11.8	11.9	11.8
Tygerhoek	2	Bacchus	11.4	11.5	11.4	11.4
Tygerhoek	2	Tobie	11.8	11.8	11.8	11.8
Tygerhoek	2	Rex	13.5	13.4	13.5	13.5
Tygerhoek	2	Ibis	13.2	nd	12.9	13.1
Tygerhoek	3	US2007	11.7	11.8	11.9	11.8
Tygerhoek	3	AgBeacon	11.7	11.6	11.7	11.6
Tygerhoek	3	Bacchus	10.7	10.7	10.7	10.7
Tygerhoek	3	Tobie	10.4	10.2	10.3	10.3
Tygerhoek	3	Rex	12.3	12.3	12.4	12.3
Tygerhoek	3	Ibis	13.1	13.2	13.1	13.1
Tygerhoek	1	Wheat (Kariega)	13.4	13.3	13.3	13.3
Tygerhoek	2	Wheat (Kariega)	12.4	12.5	12.3	12.4
Tygerhoek	3	Wheat (Kariega)	12.5	12.5	12.4	12.4

[#]not determined due to insufficient sample

Table 2.3 Ash content (%) values for the 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	1.85	1.92	1.92	1.89
Riversdal	1	AgBeacon	2.03	2.04	2.05	2.04
Riversdal	1	Bacchus	1.83	1.87	1.86	1.85
Riversdal	1	Tobie	2.10	2.16	2.12	2.13
Riversdal	1	Rex	1.78	1.77	1.77	1.77
Riversdal	1	Ibis	1.82	1.77	1.77	1.79
Riversdal	2	US2007	1.84	1.85	1.83	1.84
Riversdal	2	AgBeacon	1.84	1.80	1.83	1.82
Riversdal	2	Bacchus	1.91	1.92	1.91	1.91
Riversdal	2	Tobie	2.10	2.16	2.10	2.12
Riversdal	2	Rex	1.73	1.66	1.71	1.70
Riversdal	2	Ibis	2.87	3.47	1.85	2.73
Riversdal	3	US2007	1.92	1.94	1.93	1.93
Riversdal	3	AgBeacon	1.89	1.90	1.90	1.89
Riversdal	3	Bacchus	2.10	2.03	2.12	2.09
Riversdal	3	Tobie	2.35	2.33	2.35	2.34
Riversdal	3	Rex	1.76	1.79	1.64	1.73
Riversdal	3	Ibis	1.71	1.74	1.72	1.72
Riversdal	1	Wheat (Kariega)	1.71	1.76	1.69	1.72
Riversdal	2	Wheat (Kariega)	1.77	1.83	1.84	1.82
Riversdal	3	Wheat (Kariega)	1.80	1.84	1.85	1.83
Napier	1	US2007	1.66	1.69	1.64	1.66
Napier	1	AgBeacon	1.83	1.83	1.83	1.83
Napier	1	Bacchus	1.60	1.56	1.57	1.58
Napier	1	Tobie	1.84	1.78	1.78	1.80
Napier	1	Rex	1.58	1.59	1.60	1.59
Napier	1	Ibis	1.59	1.56	1.58	1.58
Napier	2	US2007	1.79	1.80	1.82	1.81
Napier	2	AgBeacon	1.70	1.70	1.73	1.71
Napier	2	Bacchus	1.79	1.50	1.63	1.64
Napier	2	Tobie	nd[#]	nd	nd	nd
Napier	2	Rex	1.69	1.69	1.71	1.70
Napier	2	Ibis	1.67	1.68	1.66	1.67
Napier	3	US2007	1.68	1.65	1.64	1.66
Napier	3	AgBeacon	1.56	1.53	1.54	1.54
Napier	3	Bacchus	1.63	1.60	1.63	1.62
Napier	3	Tobie	1.85	1.82	1.82	1.83
Napier	3	Rex	1.66	1.66	1.68	1.67
Napier	3	Ibis	1.69	1.65	1.66	1.67
Napier	1	Wheat (Kariega)	1.46	1.45	1.49	1.46
Napier	2	Wheat (Kariega)	1.50	1.44	1.45	1.46
Napier	3	Wheat (Kariega)	1.49	1.50	1.53	1.51
Langgewens	1	US2007	1.88	1.92	1.89	1.90
Langgewens	1	AgBeacon	1.81	1.81	1.82	1.81
Langgewens	1	Bacchus	1.70	1.71	1.67	1.69
Langgewens	1	Tobie	1.93	1.88	1.91	1.91
Langgewens	1	Rex	1.83	1.85	1.85	1.85
Langgewens	1	Ibis	1.82	1.81	1.82	1.82
Langgewens	2	US2007	1.74	1.74	1.76	1.75
Langgewens	2	AgBeacon	1.92	1.94	1.96	1.94
Langgewens	2	Bacchus	1.73	1.73	1.73	1.73
Langgewens	2	Tobie	1.94	1.95	1.96	1.95
Langgewens	2	Rex	1.77	1.78	1.76	1.77

Table 2.3 continued

Langgewens	2	Ibis	1.79	1.80	1.78	1.79
Langgewens	3	US2007	1.62	1.90	1.86	1.79
Langgewens	3	AgBeacon	1.82	1.80	1.80	1.81
Langgewens	3	Bacchus	1.65	1.70	1.67	1.68
Langgewens	3	Tobie	2.00	1.99	2.00	2.00
Langgewens	3	Rex	1.84	1.80	1.82	1.82
Langgewens	3	Ibis	2.50	1.78	1.74	2.01
Langgewens	1	Wheat (Kariega)	1.65	1.59	1.63	1.62
Langgewens	2	Wheat (Kariega)	1.61	1.63	1.66	1.63
Langgewens	3	Wheat (Kariega)	1.62	1.63	nd	1.62
Roodebloem	1	US2007	1.89	1.77	1.81	1.82
Roodebloem	1	AgBeacon	1.95	1.94	1.95	1.95
Roodebloem	1	Bacchus	1.66	1.64	1.66	1.65
Roodebloem	1	Tobie	1.73	1.76	1.76	1.75
Roodebloem	1	Rex	1.74	1.74	1.81	1.76
Roodebloem	1	Ibis	1.68	1.69	1.58	1.65
Roodebloem	2	US2007	1.71	1.75	1.79	1.75
Roodebloem	2	AgBeacon	3.18	2.87	3.11	3.05
Roodebloem	2	Bacchus	1.71	1.70	1.73	1.71
Roodebloem	2	Tobie	1.81	1.78	1.76	1.78
Roodebloem	2	Rex	1.83	1.91	1.85	1.86
Roodebloem	2	Ibis	1.78	1.77	1.81	1.78
Roodebloem	3	US2007	1.73	1.72	1.71	1.72
Roodebloem	3	AgBeacon	1.79	1.74	1.77	1.77
Roodebloem	3	Bacchus	1.67	1.66	1.64	1.66
Roodebloem	3	Tobie	1.82	1.84	1.82	1.83
Roodebloem	3	Rex	1.82	1.78	1.79	1.80
Roodebloem	3	Ibis	1.82	1.76	1.82	1.80
Roodebloem	1	Wheat (Kariega)	1.59	1.60	1.59	1.59
Roodebloem	2	Wheat (Kariega)	1.56	1.55	1.56	1.56
Roodebloem	3	Wheat (Kariega)	1.61	1.67	1.63	1.63
Piketberg	1	US2007	2.20	1.91	1.91	2.01
Piketberg	1	AgBeacon	1.90	1.87	1.89	1.89
Piketberg	1	Bacchus	1.84	1.83	1.83	1.83
Piketberg	1	Tobie	1.69	1.71	1.69	1.70
Piketberg	1	Rex	1.70	1.69	1.69	1.69
Piketberg	1	Ibis	1.67	1.68	1.67	1.67
Piketberg	2	US2007	1.87	1.88	1.86	1.87
Piketberg	2	AgBeacon	3.14	3.18	2.01	2.78
Piketberg	2	Bacchus	1.56	1.55	1.61	1.57
Piketberg	2	Tobie	1.84	1.80	1.84	1.83
Piketberg	2	Rex	1.75	1.71	1.74	1.73
Piketberg	2	Ibis	1.74	1.77	1.76	1.76
Piketberg	3	US2007	1.81	1.77	1.82	1.80
Piketberg	3	AgBeacon	1.90	1.91	1.90	1.90
Piketberg	3	Bacchus	1.62	1.63	1.66	1.64
Piketberg	3	Tobie	1.77	1.79	1.82	1.79
Piketberg	3	Rex	1.71	1.73	1.71	1.72
Piketberg	3	Ibis	1.79	1.79	1.81	1.80
Piketberg	1	Wheat (Kariega)	1.63	1.62	1.69	1.65
Piketberg	2	Wheat (Kariega)	1.41	1.59	1.59	1.53
Piketberg	3	Wheat (Kariega)	1.48	1.49	1.51	1.50
Klipheuwel	1	US2007	1.94	1.96	1.97	1.96
Klipheuwel	1	AgBeacon	1.95	1.93	1.98	1.95

Table 2.3 continued

Klipheuwel	1	Bacchus	1.72	1.70	1.69	1.71
Klipheuwel	1	Tobie	1.99	2.00	2.06	2.02
Klipheuwel	1	Rex	1.88	1.88	1.85	1.87
Klipheuwel	1	Ibis	1.84	1.84	1.85	1.84
Klipheuwel	2	US2007	1.98	1.95	1.95	1.96
Klipheuwel	2	AgBeacon	2.03	2.05	2.03	2.04
Klipheuwel	2	Bacchus	1.83	1.82	1.84	1.83
Klipheuwel	2	Tobie	1.93	1.95	1.91	1.93
Klipheuwel	2	Rex	1.93	1.89	1.94	1.92
Klipheuwel	2	Ibis	1.88	1.92	1.89	1.90
Klipheuwel	3	US2007	1.90	1.91	1.90	1.90
Klipheuwel	3	AgBeacon	1.95	1.94	1.94	1.94
Klipheuwel	3	Bacchus	1.78	1.75	1.73	1.75
Klipheuwel	3	Tobie	1.99	1.99	2.00	1.99
Klipheuwel	3	Rex	1.86	1.86	1.85	1.85
Klipheuwel	3	Ibis	1.81	1.80	1.81	1.81
Klipheuwel	1	Wheat (Kariega)	1.63	1.63	1.65	1.64
Klipheuwel	2	Wheat (Kariega)	1.78	1.76	1.77	1.77
Klipheuwel	3	Wheat (Kariega)	1.63	1.65	1.62	1.63
Mariendahl	1	US2007	2.18	3.08	2.12	2.46
Mariendahl	1	AgBeacon	2.01	2.05	1.73	1.93
Mariendahl	1	Bacchus	1.93	1.97	2.01	1.97
Mariendahl	1	Tobie	2.15	2.17	2.16	2.16
Mariendahl	1	Rex	2.01	2.03	2.00	2.01
Mariendahl	1	Ibis	2.03	2.02	2.04	2.03
Mariendahl	2	US2007	2.08	2.08	2.10	2.09
Mariendahl	2	AgBeacon	2.13	2.12	2.15	2.14
Mariendahl	2	Bacchus	1.98	2.45	1.96	2.13
Mariendahl	2	Tobie	2.20	2.20	2.20	2.20
Mariendahl	2	Rex	2.06	2.06	2.02	2.05
Mariendahl	2	Ibis	2.07	2.04	2.07	2.06
Mariendahl	3	US2007	2.13	2.13	2.10	2.12
Mariendahl	3	AgBeacon	2.03	2.03	2.04	2.03
Mariendahl	3	Bacchus	1.94	1.97	1.98	1.96
Mariendahl	3	Tobie	2.30	2.33	2.31	2.31
Mariendahl	3	Rex	2.13	2.12	2.13	2.13
Mariendahl	3	Ibis	2.12	nd	2.05	2.09
Albertinia	1	US2007	1.77	1.75	1.76	1.76
Albertinia	1	AgBeacon	1.71	1.70	1.68	1.70
Albertinia	1	Bacchus	1.58	1.63	1.69	1.63
Albertinia	1	Tobie	1.80	1.74	1.77	1.77
Albertinia	1	Rex	1.74	1.76	1.71	1.74
Albertinia	1	Ibis	1.67	1.67	1.69	1.68
Albertinia	2	US2007	1.73	1.73	1.74	1.73
Albertinia	2	AgBeacon	1.72	1.61	1.60	1.64
Albertinia	2	Bacchus	1.70	1.76	1.73	1.73
Albertinia	2	Tobie	1.66	1.68	1.69	1.67
Albertinia	2	Rex	1.73	1.70	1.41	1.61
Albertinia	2	Ibis	1.56	1.54	1.59	1.57
Albertinia	3	US2007	1.54	1.57	1.53	1.55
Albertinia	3	AgBeacon	1.48	1.50	1.30	1.42
Albertinia	3	Bacchus	1.70	1.72	1.20	1.54
Albertinia	3	Tobie	1.56	1.54	1.60	1.57
Albertinia	3	Rex	1.49	1.50	1.51	1.50

Table 2.3 continued

Albertinia	3	Ibis	1.63	1.66	1.65	1.65
Albertinia	1	Wheat (Kariega)	1.75	1.67	1.69	1.70
Albertinia	2	Wheat (Kariega)	1.64	3.18	1.67	2.16
Albertinia	3	Wheat (Kariega)	1.76	1.79	1.79	1.78
Tygerhoek	1	US2007	1.82	1.78	1.81	1.80
Tygerhoek	1	AgBeacon	1.58	1.58	1.58	1.58
Tygerhoek	1	Bacchus	1.63	1.63	1.68	1.64
Tygerhoek	1	Tobie	1.85	1.83	1.84	1.84
Tygerhoek	1	Rex	1.65	1.67	1.69	1.67
Tygerhoek	1	Ibis	1.70	1.71	1.72	1.71
Tygerhoek	2	US2007	1.97	2.02	2.01	2.00
Tygerhoek	2	AgBeacon	1.66	1.69	1.67	1.67
Tygerhoek	2	Bacchus	1.57	1.56	1.66	1.60
Tygerhoek	2	Tobie	1.66	1.72	1.66	1.68
Tygerhoek	2	Rex	1.78	1.76	1.79	1.78
Tygerhoek	2	Ibis	1.68	1.68	1.69	1.68
Tygerhoek	3	US2007	2.45	1.66	1.66	1.92
Tygerhoek	3	AgBeacon	1.64	1.65	1.61	1.63
Tygerhoek	3	Bacchus	1.59	1.60	1.60	1.60
Tygerhoek	3	Tobie	1.51	1.53	1.54	1.53
Tygerhoek	3	Rex	1.50	1.51	1.50	1.50
Tygerhoek	3	Ibis	1.54	1.52	1.55	1.54
Tygerhoek	1	Wheat (Kariega)	1.71	1.74	1.74	1.73
Tygerhoek	2	Wheat (Kariega)	1.43	1.45	1.44	1.44
Tygerhoek	3	Wheat (Kariega)	1.51	1.18	1.51	1.40

[#]not determined due to insufficient sample

Table 2.4 Falling number (s) values for the 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	94	118	116	117
Riversdal	1	AgBeacon	156	162	158	159
Riversdal	1	Bacchus	76	80	78	78
Riversdal	1	Tobie	217	197	210	208
Riversdal	1	Rex	98	104	99	100
Riversdal	1	Ibis	113	110	114	112
Riversdal	2	US2007	89	86	86	87
Riversdal	2	AgBeacon	169	170	165	168
Riversdal	2	Bacchus	70	70	70	70
Riversdal	2	Tobie	180	181	179	180
Riversdal	2	Rex	98	112	112	107
Riversdal	2	Ibis	151	144	145	147
Riversdal	3	US2007	86	85	84	85
Riversdal	3	AgBeacon	161	165	163	163
Riversdal	3	Bacchus	139	132	136	136
Riversdal	3	Tobie	263	256	268	262
Riversdal	3	Rex	nd[#]	128	132	130
Riversdal	3	Ibis	120	118	119	119
Riversdal	1	Wheat (Kariega)	508	490	483	494
Riversdal	2	Wheat (Kariega)	503	495	504	501
Riversdal	3	Wheat (Kariega)	480	479	483	481
Napier	1	US2007	70	70	70	70
Napier	1	AgBeacon	95	96	96	96
Napier	1	Bacchus	78	79	78	78
Napier	1	Tobie	255	256	244	252
Napier	1	Rex	120	122	114	119
Napier	1	Ibis	113	114	115	114
Napier	2	US2007	67	67	67	67
Napier	2	AgBeacon	95	92	93	93
Napier	2	Bacchus	111	114	110	112
Napier	2	Tobie	305	295	299	300
Napier	2	Rex	202	203	204	203
Napier	2	Ibis	178	180	181	180
Napier	3	US2007	80	79	80	80
Napier	3	AgBeacon	101	99	100	100
Napier	3	Bacchus	118	123	116	119
Napier	3	Tobie	210	214	215	213
Napier	3	Rex	116	117	117	117
Napier	3	Ibis	186	187	188	187
Napier	1	Wheat (Kariega)	401	406	409	405
Napier	2	Wheat (Kariega)	412	420	422	418
Napier	3	Wheat (Kariega)	nd	428	429	429
Langgewens	1	US2007	70	70	70	70
Langgewens	1	AgBeacon	117	118	111	115
Langgewens	1	Bacchus	74	76	78	76
Langgewens	1	Tobie	232	238	233	234
Langgewens	1	Rex	173	171	174	173
Langgewens	1	Ibis	75	75	75	75
Langgewens	2	US2007	66	65	65	65
Langgewens	2	AgBeacon	99	98	108	102
Langgewens	2	Bacchus	79	79	78	79
Langgewens	2	Tobie	179	183	180	181
Langgewens	2	Rex	125	130	129	128

Table 2.4 continued

Langgewens	2	Ibis	69	68	68	68
Langgewens	3	US2007	66	66	66	66
Langgewens	3	AgBeacon	76	83	78	79
Langgewens	3	Bacchus	74	75	75	75
Langgewens	3	Tobie	231	230	233	231
Langgewens	3	Rex	nd	nd	nd	nd
Langgewens	3	Ibis	80	81	81	81
Langgewens	1	Wheat (Kariega)	425	422	444	430
Langgewens	2	Wheat (Kariega)	454	428	437	440
Langgewens	3	Wheat (Kariega)	432	448	437	439
Roodebloem	1	US2007	186	190	189	188
Roodebloem	1	AgBeacon	189	188	183	187
Roodebloem	1	Bacchus	153	154	159	155
Roodebloem	1	Tobie	nd	211	216	214
Roodebloem	1	Rex	200	202	199	200
Roodebloem	1	Ibis	185	185	186	185
Roodebloem	2	US2007	129	125	130	128
Roodebloem	2	AgBeacon	197	202	195	198
Roodebloem	2	Bacchus	161	161	157	160
Roodebloem	2	Tobie	227	227	225	226
Roodebloem	2	Rex	187	184	188	186
Roodebloem	2	Ibis	229	226	228	228
Roodebloem	3	US2007	93	80	85	86
Roodebloem	3	AgBeacon	207	209	206	207
Roodebloem	3	Bacchus	155	156	154	155
Roodebloem	3	Tobie	246	251	249	249
Roodebloem	3	Rex	201	203	202	202
Roodebloem	3	Ibis	168	172	165	168
Roodebloem	1	Wheat (Kariega)	nd	423	417	420
Roodebloem	2	Wheat (Kariega)	411	405	406	407
Roodebloem	3	Wheat (Kariega)	423	409	422	418
Piketberg	1	US2007	63	63	63	63
Piketberg	1	AgBeacon	117	117	121	118
Piketberg	1	Bacchus	102	102	102	102
Piketberg	1	Tobie	202	203	205	203
Piketberg	1	Rex	126	120	120	122
Piketberg	1	Ibis	90	93	89	91
Piketberg	2	US2007	71	73	72	72
Piketberg	2	AgBeacon	161	168	166	165
Piketberg	2	Bacchus	71	72	71	71
Piketberg	2	Tobie	207	205	209	207
Piketberg	2	Rex	153	150	143	149
Piketberg	2	Ibis	85	86	87	86
Piketberg	3	US2007	65	65	65	65
Piketberg	3	AgBeacon	99	103	101	101
Piketberg	3	Bacchus	73	73	73	73
Piketberg	3	Tobie	222	223	226	224
Piketberg	3	Rex	167	165	155	162
Piketberg	3	Ibis	106	103	102	104
Piketberg	1	Wheat (Kariega)	356	353	353	354
Piketberg	2	Wheat (Kariega)	424	409	406	413
Piketberg	3	Wheat (Kariega)	440	428	423	430
Klipheuwel	1	US2007	126	119	123	123
Klipheuwel	1	AgBeacon	210	208	216	211

Table 2.4 continued

Klipheuwel	1	Bacchus	159	158	158	158
Klipheuwel	1	Tobie	285	275	283	281
Klipheuwel	1	Rex	266	265	271	267
Klipheuwel	1	Ibis	222	226	226	225
Klipheuwel	2	US2007	127	125	122	125
Klipheuwel	2	AgBeacon	216	217	211	215
Klipheuwel	2	Bacchus	155	157	158	157
Klipheuwel	2	Tobie	263	263	263	263
Klipheuwel	2	Rex	240	237	250	242
Klipheuwel	2	Ibis	180	176	177	178
Klipheuwel	3	US2007	107	103	103	104
Klipheuwel	3	AgBeacon	207	206	206	206
Klipheuwel	3	Bacchus	183	183	179	182
Klipheuwel	3	Tobie	274	266	270	270
Klipheuwel	3	Rex	211	209	215	212
Klipheuwel	3	Ibis	212	214	213	213
Klipheuwel	1	Wheat (Kariega)	444	443	431	439
Klipheuwel	2	Wheat (Kariega)	431	433	445	436
Klipheuwel	3	Wheat (Kariega)	412	nd	412	412
Mariendahl	1	US2007	63	63	63	63
Mariendahl	1	AgBeacon	139	143	139	140
Mariendahl	1	Bacchus	96	98	95	96
Mariendahl	1	Tobie	178	181	179	179
Mariendahl	1	Rex	186	183	189	186
Mariendahl	1	Ibis	100	98	100	99
Mariendahl	2	US2007	79	78	81	79
Mariendahl	2	AgBeacon	159	162	160	160
Mariendahl	2	Bacchus	141	140	139	140
Mariendahl	2	Tobie	213	211	213	212
Mariendahl	2	Rex	nd	164	171	168
Mariendahl	2	Ibis	147	141	144	144
Mariendahl	3	US2007	72	72	74	73
Mariendahl	3	AgBeacon	155	156	157	156
Mariendahl	3	Bacchus	105	108	107	107
Mariendahl	3	Tobie	217	223	218	219
Mariendahl	3	Rex	173	177	172	174
Mariendahl	3	Ibis	95	94	93	94
Albertinia	1	US2007	117	117	117	117
Albertinia	1	AgBeacon	177	183	181	180
Albertinia	1	Bacchus	125	134	131	130
Albertinia	1	Tobie	192	191	192	192
Albertinia	1	Rex	163	164	165	164
Albertinia	1	Ibis	197	194	198	196
Albertinia	2	US2007	99	100	101	100
Albertinia	2	AgBeacon	202	197	199	199
Albertinia	2	Bacchus	110	113	110	111
Albertinia	2	Tobie	203	203	207	204
Albertinia	2	Rex	145	143	147	145
Albertinia	2	Ibis	106	111	108	108
Albertinia	3	US2007	105	100	100	102
Albertinia	3	AgBeacon	200	194	196	197
Albertinia	3	Bacchus	106	93	103	101
Albertinia	3	Tobie	222	220	222	221
Albertinia	3	Rex	95	97	92	95

Table 2.4 continued

Albertinia	3	Ibis	104	105	107	105
Albertinia	1	Wheat (Kariega)	485	468	457	470
Albertinia	2	Wheat (Kariega)	486	458	466	470
Albertinia	3	Wheat (Kariega)	486	478	465	476
Tygerhoek	1	US2007	85	84	86	85
Tygerhoek	1	AgBeacon	161	168	165	165
Tygerhoek	1	Bacchus	186	181	183	183
Tygerhoek	1	Tobie	266	268	257	264
Tygerhoek	1	Rex	157	141	157	152
Tygerhoek	1	Ibis	118	113	112	114
Tygerhoek	2	US2007	88	82	81	84
Tygerhoek	2	AgBeacon	139	140	143	141
Tygerhoek	2	Bacchus	161	158	158	159
Tygerhoek	2	Tobie	279	279	280	279
Tygerhoek	2	Rex	172	179	179	177
Tygerhoek	2	Ibis	102	98	98	99
Tygerhoek	3	US2007	94	93	91	93
Tygerhoek	3	AgBeacon	158	155	153	155
Tygerhoek	3	Bacchus	144	145	145	145
Tygerhoek	3	Tobie	265	263	269	266
Tygerhoek	3	Rex	184	184	182	183
Tygerhoek	3	Ibis	147	146	149	147
Tygerhoek	1	Wheat (Kariega)	458	453	462	458
Tygerhoek	2	Wheat (Kariega)	468	462	463	464
Tygerhoek	3	Wheat (Kariega)	434	437	423	431

[#]not determined due to insufficient sample

Table 2.5 Particle size index values for the 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	67.05	67.22	67.14	59.82
Riversdal	1	AgBeacon	66.57	66.53	66.55	59.22
Riversdal	1	Bacchus	70.35	70.55	70.45	63.21
Riversdal	1	Tobie	67.49	66.65	67.07	59.76
Riversdal	1	Rex	67.09	66.39	66.74	59.42
Riversdal	1	Ibis	64.84	64.46	64.65	57.28
Riversdal	2	US2007	68.55	68.90	68.73	61.45
Riversdal	2	AgBeacon	66.15	66.58	66.37	59.03
Riversdal	2	Bacchus	71.22	70.78	71.00	63.78
Riversdal	2	Tobie	67.57	67.99	67.78	60.48
Riversdal	2	Rex	64.70	64.73	64.72	57.35
Riversdal	2	Ibis	63.05	63.40	63.23	55.82
Riversdal	3	US2007	67.74	68.26	68.00	60.71
Riversdal	3	AgBeacon	nd[#]	nd	nd	nd
Riversdal	3	Bacchus	73.62	73.20	73.41	66.24
Riversdal	3	Tobie	68.53	68.16	68.35	61.06
Riversdal	3	Rex	68.40	68.43	68.42	61.13
Riversdal	3	Ibis	65.81	65.20	65.51	58.15
Riversdal	1	Wheat (Kariega)	54.88	54.62	54.75	60.04
Riversdal	2	Wheat (Kariega)	53.86	54.13	54.00	59.30
Riversdal	3	Wheat (Kariega)	54.24	54.42	54.33	59.63
Napier	1	US2007	72.82	72.73	72.78	65.59
Napier	1	AgBeacon	69.66	69.20	69.43	62.17
Napier	1	Bacchus	75.61	75.88	75.75	68.63
Napier	1	Tobie	73.35	73.15	73.25	66.08
Napier	1	Rex	69.48	68.86	69.17	61.91
Napier	1	Ibis	68.41	68.57	68.49	61.21
Napier	2	US2007	70.05	69.82	69.94	62.69
Napier	2	AgBeacon	72.44	72.16	72.30	65.11
Napier	2	Bacchus	74.42	74.31	74.37	67.22
Napier	2	Tobie	nd	nd	nd	nd
Napier	2	Rex	69.08	68.28	68.68	61.40
Napier	2	Ibis	67.62	67.67	67.65	60.34
Napier	3	US2007	70.48	70.79	70.64	63.40
Napier	3	AgBeacon	70.28	69.95	70.12	62.87
Napier	3	Bacchus	73.96	74.34	74.15	67.00
Napier	3	Tobie	72.19	71.65	71.92	64.72
Napier	3	Rex	68.67	68.84	68.76	61.48
Napier	3	Ibis	68.24	68.60	68.42	61.14
Napier	1	Wheat (Kariega)	50.13	49.48	49.81	55.22
Napier	2	Wheat (Kariega)	52.01	51.93	51.97	57.33
Napier	3	Wheat (Kariega)	49.92	49.70	49.81	55.22
Langgewens	1	US2007	67.64	67.21	67.43	60.12
Langgewens	1	AgBeacon	66.67	66.73	66.70	59.38
Langgewens	1	Bacchus	72.07	71.59	71.83	64.63
Langgewens	1	Tobie	65.54	65.17	65.36	58.00
Langgewens	1	Rex	68.19	67.94	68.07	60.77
Langgewens	1	Ibis	65.86	66.25	66.06	58.72
Langgewens	2	US2007	67.12	66.36	66.74	59.42
Langgewens	2	AgBeacon	67.04	67.06	67.05	59.74
Langgewens	2	Bacchus	70.83	70.64	70.74	63.51
Langgewens	2	Tobie	63.49	62.79	63.14	55.73
Langgewens	2	Rex	68.80	68.07	68.44	61.15

Table 2.5 continued

Langgewens	2	Ibis	65.96	65.75	65.86	58.51
Langgewens	3	US2007	65.37	64.54	64.96	57.59
Langgewens	3	AgBeacon	64.89	65.23	65.06	57.70
Langgewens	3	Bacchus	70.48	70.76	70.62	63.39
Langgewens	3	Tobie	64.57	64.25	64.41	57.03
Langgewens	3	Rex	64.63	65.19	64.91	57.55
Langgewens	3	Ibis	65.31	65.00	65.16	57.80
Langgewens	1	Wheat (Kariega)	58.92	59.28	59.10	64.28
Langgewens	2	Wheat (Kariega)	56.67	57.23	56.95	62.18
Langgewens	3	Wheat (Kariega)	nd	nd	nd	nd
Roodebloem	1	US2007	67.01	67.41	67.21	59.90
Roodebloem	1	AgBeacon	66.49	66.47	66.48	59.15
Roodebloem	1	Bacchus	72.75	72.14	72.45	65.26
Roodebloem	1	Tobie	67.93	67.46	67.70	60.40
Roodebloem	1	Rex	66.30	65.51	65.91	58.56
Roodebloem	1	Ibis	63.93	64.38	64.16	56.77
Roodebloem	2	US2007	55.31	55.59	55.45	60.72
Roodebloem	2	AgBeacon	55.00	55.19	55.10	60.38
Roodebloem	2	Bacchus	66.54	67.29	66.92	71.90
Roodebloem	2	Tobie	54.66	54.39	54.53	59.82
Roodebloem	2	Rex	52.65	52.72	52.69	58.03
Roodebloem	2	Ibis	52.29	51.97	52.13	57.49
Roodebloem	3	US2007	56.66	56.81	56.74	61.97
Roodebloem	3	AgBeacon	56.62	55.66	56.14	61.39
Roodebloem	3	Bacchus	60.76	60.82	60.79	65.93
Roodebloem	3	Tobie	55.39	55.21	55.30	60.58
Roodebloem	3	Rex	52.93	52.76	52.85	58.18
Roodebloem	3	Ibis	51.96	51.49	51.73	57.09
Roodebloem	1	Wheat (Kariega)	51.62	51.20	51.41	56.78
Roodebloem	2	Wheat (Kariega)	51.97	52.05	52.01	57.37
Roodebloem	3	Wheat (Kariega)	50.87	50.51	50.69	56.08
Piketberg	1	US2007	67.33	67.29	67.31	72.29
Piketberg	1	AgBeacon	64.18	64.70	64.44	69.49
Piketberg	1	Bacchus	72.01	71.89	71.95	76.81
Piketberg	1	Tobie	64.43	64.69	64.56	69.60
Piketberg	1	Rex	64.97	65.15	65.06	70.09
Piketberg	1	Ibis	63.44	64.29	63.87	68.93
Piketberg	2	US2007	65.69	66.31	66.00	71.01
Piketberg	2	AgBeacon	64.96	64.68	64.82	69.86
Piketberg	2	Bacchus	71.05	70.83	70.94	75.82
Piketberg	2	Tobie	60.46	60.16	60.31	65.46
Piketberg	2	Rex	65.61	65.68	65.65	70.66
Piketberg	2	Ibis	64.75	65.43	65.09	70.12
Piketberg	3	US2007	68.48	68.70	68.59	73.53
Piketberg	3	AgBeacon	65.30	65.54	65.42	70.44
Piketberg	3	Bacchus	69.69	69.63	69.66	74.58
Piketberg	3	Tobie	62.34	62.88	62.61	67.70
Piketberg	3	Rex	65.45	65.53	65.49	70.51
Piketberg	3	Ibis	64.00	63.58	63.79	68.85
Piketberg	1	Wheat (Kariega)	49.88	49.61	49.75	55.16
Piketberg	2	Wheat (Kariega)	49.69	49.80	49.75	55.16
Piketberg	3	Wheat (Kariega)	46.67	46.30	46.49	51.98
Klipheuwel	1	US2007	55.82	56.03	55.93	61.19
Klipheuwel	1	AgBeacon	55.86	55.44	55.65	60.92

Table 2.5 continued

Klipheuwel	1	Bacchus	65.42	65.67	65.55	70.56
Klipheuwel	1	Tobie	54.91	54.49	54.70	59.99
Klipheuwel	1	Rex	55.04	55.31	55.18	60.45
Klipheuwel	1	Ibis	51.90	52.20	52.05	57.41
Klipheuwel	2	US2007	60.08	60.01	60.05	65.20
Klipheuwel	2	AgBeacon	56.98	56.89	56.94	62.17
Klipheuwel	2	Bacchus	60.95	60.40	60.68	65.82
Klipheuwel	2	Tobie	51.34	50.87	51.11	56.49
Klipheuwel	2	Rex	56.41	56.14	56.28	61.53
Klipheuwel	2	Ibis	54.45	54.22	54.34	59.63
Klipheuwel	3	US2007	56.02	56.39	56.21	61.46
Klipheuwel	3	AgBeacon	57.26	56.55	56.91	62.14
Klipheuwel	3	Bacchus	63.37	63.41	63.39	68.46
Klipheuwel	3	Tobie	53.29	52.75	53.02	58.35
Klipheuwel	3	Rex	57.08	57.20	57.14	62.37
Klipheuwel	3	Ibis	55.31	55.00	55.16	60.43
Klipheuwel	1	Wheat (Kariega)	56.70	56.35	56.53	61.77
Klipheuwel	2	Wheat (Kariega)	55.18	55.80	55.49	60.76
Klipheuwel	3	Wheat (Kariega)	52.79	52.91	52.85	58.19
Mariendahl	1	US2007	66.63	65.97	66.30	71.30
Mariendahl	1	AgBeacon	67.12	66.68	66.90	71.89
Mariendahl	1	Bacchus	69.74	70.15	69.95	74.85
Mariendahl	1	Tobie	66.64	66.08	66.36	71.36
Mariendahl	1	Rex	65.84	66.01	65.93	70.94
Mariendahl	1	Ibis	62.39	62.49	62.44	67.54
Mariendahl	2	US2007	66.75	66.25	66.50	71.50
Mariendahl	2	AgBeacon	68.20	68.66	68.43	73.38
Mariendahl	2	Bacchus	71.23	71.38	71.31	76.18
Mariendahl	2	Tobie	67.43	67.40	67.42	72.39
Mariendahl	2	Rex	64.46	64.34	64.40	69.45
Mariendahl	2	Ibis	63.17	63.04	63.11	68.19
Mariendahl	3	US2007	67.19	66.73	66.96	71.94
Mariendahl	3	AgBeacon	65.82	65.55	65.69	70.70
Mariendahl	3	Bacchus	71.71	72.02	71.87	76.73
Mariendahl	3	Tobie	64.66	61.38	63.02	68.10
Mariendahl	3	Rex	63.07	62.86	62.97	68.05
Mariendahl	3	Ibis	62.94	62.69	62.82	67.90
Albertinia	1	US2007	50.92	50.67	50.80	56.18
Albertinia	1	AgBeacon	47.57	48.02	47.80	53.26
Albertinia	1	Bacchus	59.25	58.84	59.05	64.23
Albertinia	1	Tobie	50.86	51.03	50.95	56.33
Albertinia	1	Rex	50.74	50.52	50.63	56.02
Albertinia	1	Ibis	47.11	46.82	46.97	52.45
Albertinia	2	US2007	51.61	51.95	51.78	57.14
Albertinia	2	AgBeacon	48.34	47.74	48.04	53.50
Albertinia	2	Bacchus	55.18	54.73	54.96	60.24
Albertinia	2	Tobie	47.85	47.65	47.75	53.21
Albertinia	2	Rex	48.28	48.51	48.40	53.84
Albertinia	2	Ibis	45.32	45.32	45.32	50.85
Albertinia	3	US2007	49.92	49.63	49.78	55.19
Albertinia	3	AgBeacon	47.57	47.61	47.59	53.06
Albertinia	3	Bacchus	55.23	54.34	54.79	60.07
Albertinia	3	Tobie	46.60	46.65	46.63	52.12
Albertinia	3	Rex	48.87	48.55	48.71	54.15

Table 2.5 continued

Albertinia	3	Ibis	45.32	45.98	45.65	51.17
Albertinia	1	Wheat (Kariega)	56.25	56.06	56.16	61.41
Albertinia	2	Wheat (Kariega)	55.43	55.75	55.59	60.86
Albertinia	3	Wheat (Kariega)	56.82	57.08	56.95	62.18
Tygerhoek	1	US2007	50.79	50.53	50.66	56.05
Tygerhoek	1	AgBeacon	50.99	51.38	51.19	56.56
Tygerhoek	1	Bacchus	57.78	57.20	57.49	62.71
Tygerhoek	1	Tobie	55.35	54.73	55.04	60.32
Tygerhoek	1	Rex	48.77	48.88	48.83	54.26
Tygerhoek	1	Ibis	46.91	46.44	46.68	52.17
Tygerhoek	2	US2007	49.96	49.46	49.71	55.13
Tygerhoek	2	AgBeacon	49.73	50.06	49.90	55.31
Tygerhoek	2	Bacchus	59.12	58.88	59.00	64.18
Tygerhoek	2	Tobie	52.77	53.27	53.02	58.35
Tygerhoek	2	Rex	49.13	49.59	49.36	54.78
Tygerhoek	2	Ibis	45.85	46.22	46.04	51.54
Tygerhoek	3	US2007	53.38	53.45	53.42	58.74
Tygerhoek	3	AgBeacon	50.76	51.32	51.04	56.42
Tygerhoek	3	Bacchus	59.93	60.28	60.11	65.26
Tygerhoek	3	Tobie	57.05	56.53	56.79	62.03
Tygerhoek	3	Rex	50.75	50.66	50.71	56.10
Tygerhoek	3	Ibis	46.82	46.62	46.72	52.21
Tygerhoek	1	Wheat (Kariega)	56.55	56.11	56.33	61.58
Tygerhoek	2	Wheat (Kariega)	53.75	53.35	53.55	58.87
Tygerhoek	3	Wheat (Kariega)	54.80	54.50	54.65	59.94

[#]not determined due to insufficient sample

Table 2.6 SDS sedimentation (mm) values for the 2007 harvest season

Locality	Repetition	Cultivar	Replicate 1	Replicate 2	Replicate 3	Average
Riversdal	1	US2007	65	65	65	65
Riversdal	1	AgBeacon	30	30	30	30
Riversdal	1	Bacchus	38	40	40	39
Riversdal	1	Tobie	27	29	28	28
Riversdal	1	Rex	36	37	35	36
Riversdal	1	Ibis	42	41	41	41
Riversdal	2	US2007	42	41	42	42
Riversdal	2	AgBeacon	30	30	31	30
Riversdal	2	Bacchus	35	36	35	35
Riversdal	2	Tobie	29	28	28	28
Riversdal	2	Rex	31	30	30	30
Riversdal	2	Ibis	37	37	36	37
Riversdal	3	US2007	40	39	40	40
Riversdal	3	AgBeacon	34	35	36	35
Riversdal	3	Bacchus	35	34	35	35
Riversdal	3	Tobie	31	33	32	32
Riversdal	3	Rex	32	32	31	32
Riversdal	3	Ibis	40	40	42	41
Riversdal	1	Wheat (Kariega)	100	99	99	99
Riversdal	2	Wheat (Kariega)	95	98	95	96
Riversdal	3	Wheat (Kariega)	99	99	100	99
Napier	1	US2007	30	30	29	30
Napier	1	AgBeacon	25	25	24	25
Napier	1	Bacchus	32	30	31	31
Napier	1	Tobie	24	23	24	24
Napier	1	Rex	28	29	29	29
Napier	1	Ibis	34	35	33	34
Napier	2	US2007	34	32	32	33
Napier	2	AgBeacon	28	27	27	27
Napier	2	Bacchus	24	25	25	25
Napier	2	Tobie	23	23	22	23
Napier	2	Rex	24	25	25	25
Napier	2	Ibis	45	46	45	45
Napier	3	US2007	31	31	31	31
Napier	3	AgBeacon	23	25	24	24
Napier	3	Bacchus	24	23	24	24
Napier	3	Tobie	34	34	35	34
Napier	3	Rex	25	25	25	25
Napier	3	Ibis	28	27	27	27
Napier	1	Wheat (Kariega)	80	80	80	80
Napier	2	Wheat (Kariega)	93	93	93	93
Napier	3	Wheat (Kariega)	98	100	100	99
Langgewens	1	US2007	44	44	44	44
Langgewens	1	AgBeacon	29	30	29	29
Langgewens	1	Bacchus	27	26	27	27
Langgewens	1	Tobie	36	37	35	36
Langgewens	1	Rex	28	27	28	28
Langgewens	1	Ibis	38	39	38	38
Langgewens	2	US2007	40	39	40	40
Langgewens	2	AgBeacon	30	29	29	29
Langgewens	2	Bacchus	29	29	29	29
Langgewens	2	Tobie	27	26	28	27
Langgewens	2	Rex	28	30	30	29

Table 2.6 continued

Langgewens	2	Ibis	48	50	48	49
Langgewens	3	US2007	42	43	44	43
Langgewens	3	AgBeacon	27	27	28	27
Langgewens	3	Bacchus	30	30	31	30
Langgewens	3	Tobie	27	26	26	26
Langgewens	3	Rex	32	31	31	31
Langgewens	3	Ibis	34	35	34	34
Langgewens	1	Wheat (Kariega)	80	80	80	80
Langgewens	2	Wheat (Kariega)	84	85	82	84
Langgewens	3	Wheat (Kariega)	62	61	62	62
Roodebloem	1	US2007	29	28	29	29
Roodebloem	1	AgBeacon	23	22	23	23
Roodebloem	1	Bacchus	24	23	23	23
Roodebloem	1	Tobie	20	20	21	20
Roodebloem	1	Rex	23	23	23	23
Roodebloem	1	Ibis	28	27	27	27
Roodebloem	2	US2007	30	31	31	31
Roodebloem	2	AgBeacon	24	25	24	24
Roodebloem	2	Bacchus	24	23	25	24
Roodebloem	2	Tobie	21	20	21	21
Roodebloem	2	Rex	25	25	24	25
Roodebloem	2	Ibis	26	28	27	27
Roodebloem	3	US2007	39	37	38	38
Roodebloem	3	AgBeacon	25	26	25	25
Roodebloem	3	Bacchus	22	24	23	23
Roodebloem	3	Tobie	23	23	22	23
Roodebloem	3	Rex	30	31	30	30
Roodebloem	3	Ibis	30	30	30	30
Roodebloem	1	Wheat (Kariega)	80	80	80	80
Roodebloem	2	Wheat (Kariega)	90	90	90	90
Roodebloem	3	Wheat (Kariega)	75	75	76	75
Piketberg	1	US2007	34	34	32	33
Piketberg	1	AgBeacon	26	25	26	26
Piketberg	1	Bacchus	30	29	30	30
Piketberg	1	Tobie	25	25	25	25
Piketberg	1	Rex	31	30	29	30
Piketberg	1	Ibis	30	31	30	30
Piketberg	2	US2007	35	38	36	36
Piketberg	2	AgBeacon	31	32	31	31
Piketberg	2	Bacchus	30	30	30	30
Piketberg	2	Tobie	28	27	28	28
Piketberg	2	Rex	32	31	31	31
Piketberg	2	Ibis	32	35	34	34
Piketberg	3	US2007	36	35	37	36
Piketberg	3	AgBeacon	27	27	27	27
Piketberg	3	Bacchus	25	25	27	26
Piketberg	3	Tobie	24	26	26	25
Piketberg	3	Rex	27	29	27	28
Piketberg	3	Ibis	25	27	26	26
Piketberg	1	Wheat (Kariega)	72	71	71	71
Piketberg	2	Wheat (Kariega)	75	75	75	75
Piketberg	3	Wheat (Kariega)	93	92	91	92
Klipheuwel	1	US2007	31	31	31	31
Klipheuwel	1	AgBeacon	26	25	25	25

Table 2.6 continued

Klipheuwel	1	Bacchus	25	24	24	24
Klipheuwel	1	Tobie	23	23	23	23
Klipheuwel	1	Rex	27	27	28	27
Klipheuwel	1	Ibis	30	30	32	31
Klipheuwel	2	US2007	35	37	36	36
Klipheuwel	2	AgBeacon	25	25	25	25
Klipheuwel	2	Bacchus	26	26	26	26
Klipheuwel	2	Tobie	23	24	23	23
Klipheuwel	2	Rex	25	26	27	26
Klipheuwel	2	Ibis	29	30	29	29
Klipheuwel	3	US2007	32	34	34	33
Klipheuwel	3	AgBeacon	24	24	24	24
Klipheuwel	3	Bacchus	23	25	24	24
Klipheuwel	3	Tobie	22	21	21	21
Klipheuwel	3	Rex	27	28	26	27
Klipheuwel	3	Ibis	25	26	26	26
Klipheuwel	1	Wheat (Kariega)	58	59	61	59
Klipheuwel	2	Wheat (Kariega)	60	59	58	59
Klipheuwel	3	Wheat (Kariega)	66	68	68	67
Mariendahl	1	US2007	34	32	34	33
Mariendahl	1	AgBeacon	24	25	24	24
Mariendahl	1	Bacchus	24	25	25	25
Mariendahl	1	Tobie	23	24	23	23
Mariendahl	1	Rex	25	26	26	26
Mariendahl	1	Ibis	30	29	29	29
Mariendahl	2	US2007	40	41	40	40
Mariendahl	2	AgBeacon	27	26	26	26
Mariendahl	2	Bacchus	27	28	27	27
Mariendahl	2	Tobie	24	25	23	24
Mariendahl	2	Rex	27	27	27	27
Mariendahl	2	Ibis	29	28	28	28
Mariendahl	3	US2007	35	35	35	35
Mariendahl	3	AgBeacon	25	24	25	25
Mariendahl	3	Bacchus	25	25	25	25
Mariendahl	3	Tobie	29	30	30	30
Mariendahl	3	Rex	34	34	34	34
Mariendahl	3	Ibis	29	29	28	29
Albertinia	1	US2007	38	40	40	39
Albertinia	1	AgBeacon	28	29	28	28
Albertinia	1	Bacchus	35	35	37	36
Albertinia	1	Tobie	26	26	25	26
Albertinia	1	Rex	32	31	31	31
Albertinia	1	Ibis	39	37	37	38
Albertinia	2	US2007	39	39	38	39
Albertinia	2	AgBeacon	33	31	31	32
Albertinia	2	Bacchus	32	33	33	33
Albertinia	2	Tobie	36	35	36	36
Albertinia	2	Rex	32	33	34	33
Albertinia	2	Ibis	38	38	37	38
Albertinia	3	US2007	37	38	39	38
Albertinia	3	AgBeacon	30	30	29	30
Albertinia	3	Bacchus	32	33	34	33
Albertinia	3	Tobie	33	31	31	32
Albertinia	3	Rex	34	33	34	34

Table 2.6 continued

Albertinia	3	Ibis	37	38	38	38
Albertinia	1	Wheat (Kariega)	100	100	100	100
Albertinia	2	Wheat (Kariega)	100	98	98	99
Albertinia	3	Wheat (Kariega)	95	97	97	96
Tygerhoek	1	US2007	41	40	40	40
Tygerhoek	1	AgBeacon	28	30	29	29
Tygerhoek	1	Bacchus	30	30	30	30
Tygerhoek	1	Tobie	25	25	26	25
Tygerhoek	1	Rex	30	30	30	30
Tygerhoek	1	Ibis	36	35	35	35
Tygerhoek	2	US2007	40	40	40	40
Tygerhoek	2	AgBeacon	26	26	26	26
Tygerhoek	2	Bacchus	31	31	30	31
Tygerhoek	2	Tobie	34	34	33	34
Tygerhoek	2	Rex	28	27	29	28
Tygerhoek	2	Ibis	38	37	37	37
Tygerhoek	3	US2007	47	46	47	47
Tygerhoek	3	AgBeacon	29	29	29	29
Tygerhoek	3	Bacchus	35	34	34	34
Tygerhoek	3	Tobie	27	27	27	27
Tygerhoek	3	Rex	38	40	40	39
Tygerhoek	3	Ibis	50	50	49	50
Tygerhoek	1	Wheat (Kariega)	96	96	95	96
Tygerhoek	2	Wheat (Kariega)	100	100	99	100
Tygerhoek	3	Wheat (Kariega)	102	103	102	102

Appendix 3

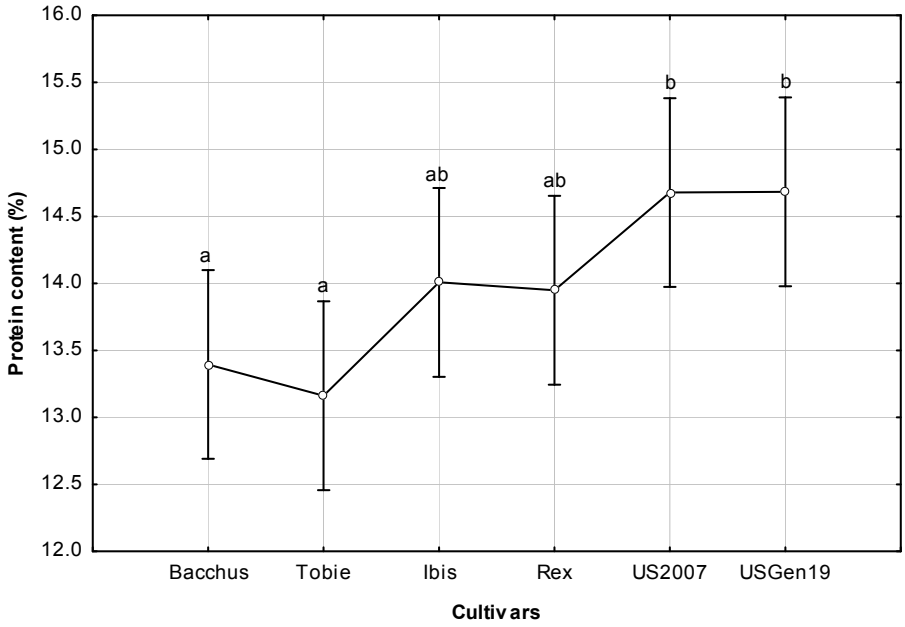


Figure 3.1 Differences between average protein content (12% mb) obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

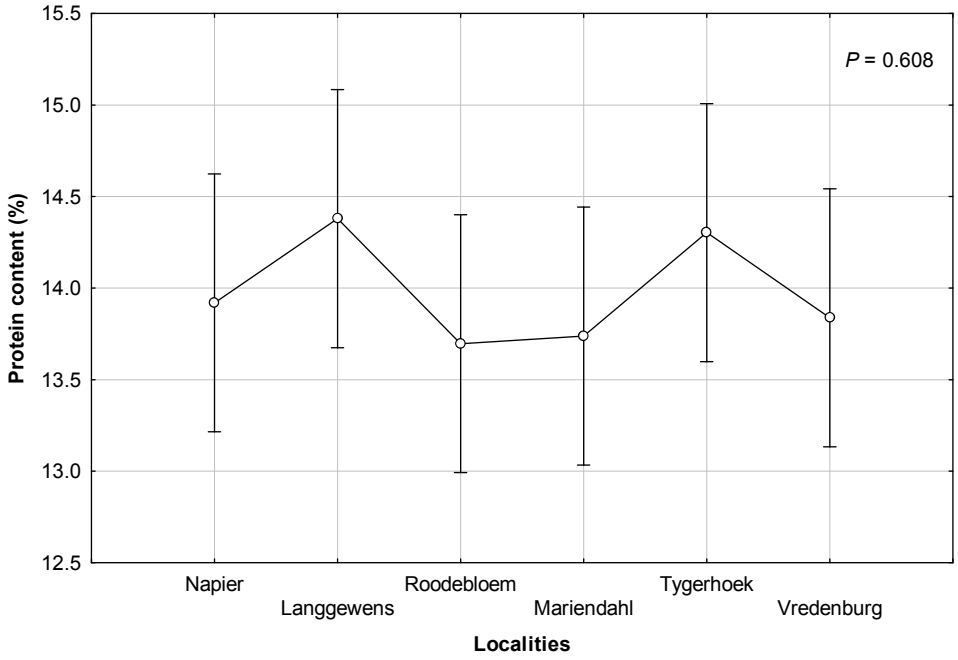


Figure 3.2 Differences between average protein content (12% mb) obtained for localities as determined by ANOVA ($P \geq 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

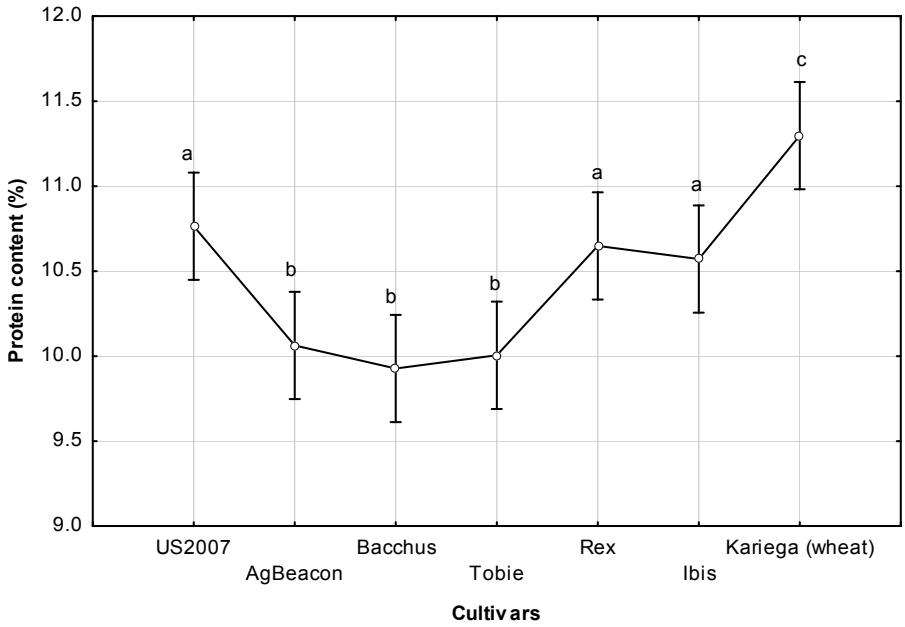


Figure 3.3 Differences between average protein contents (12% mb) obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

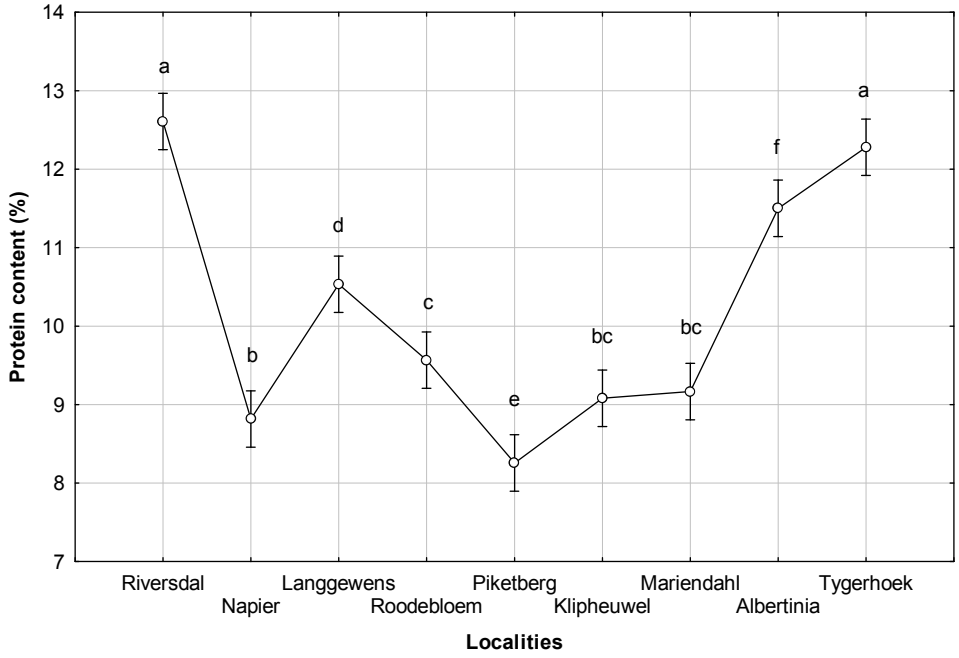


Figure 3.4 Differences between average protein contents (12% mb) obtained for localities as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

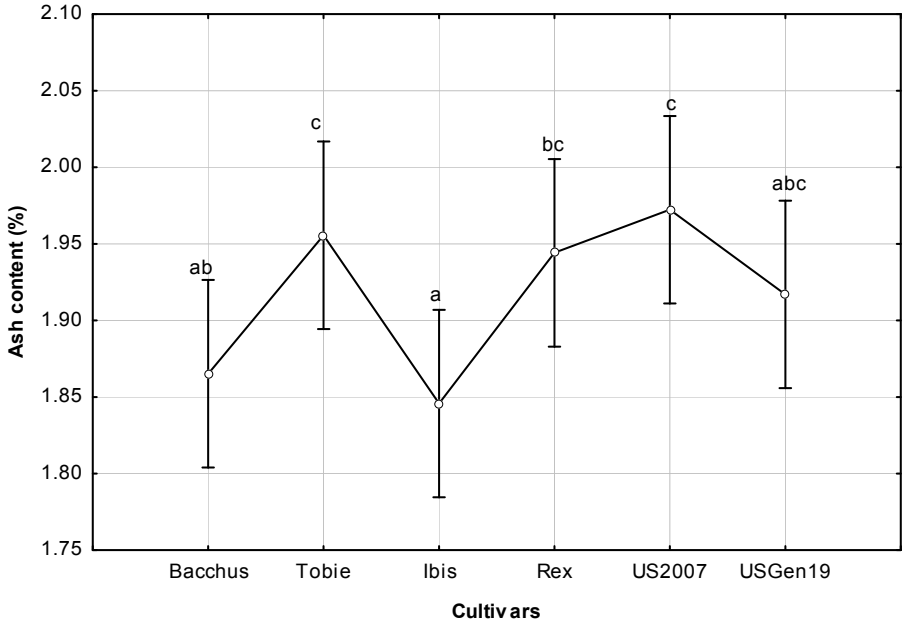


Figure 3.5 Differences between average ash contents obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

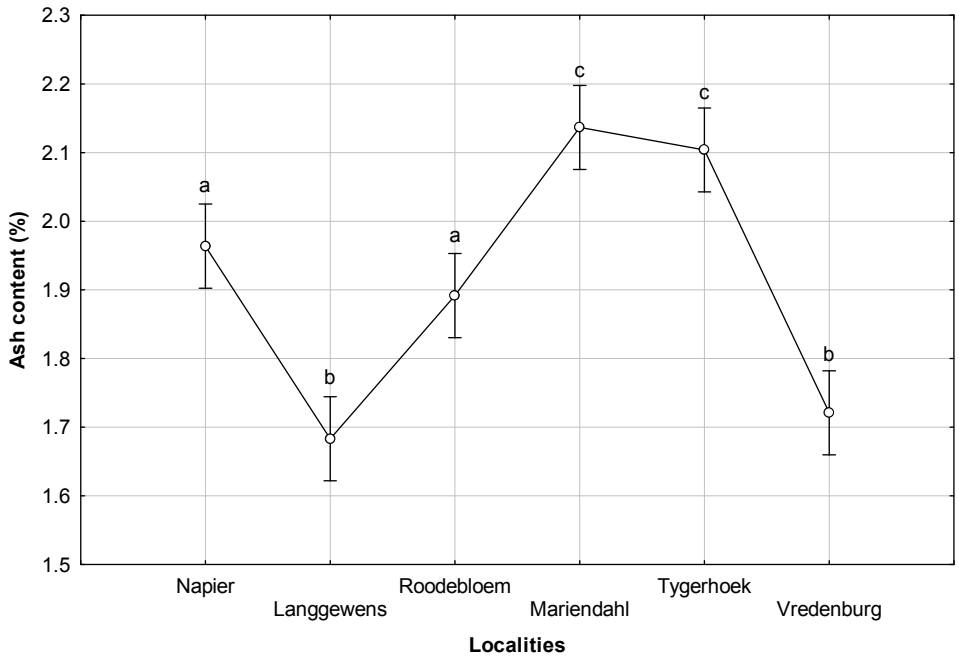


Figure 3.6 Differences between average ash contents obtained for localities as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

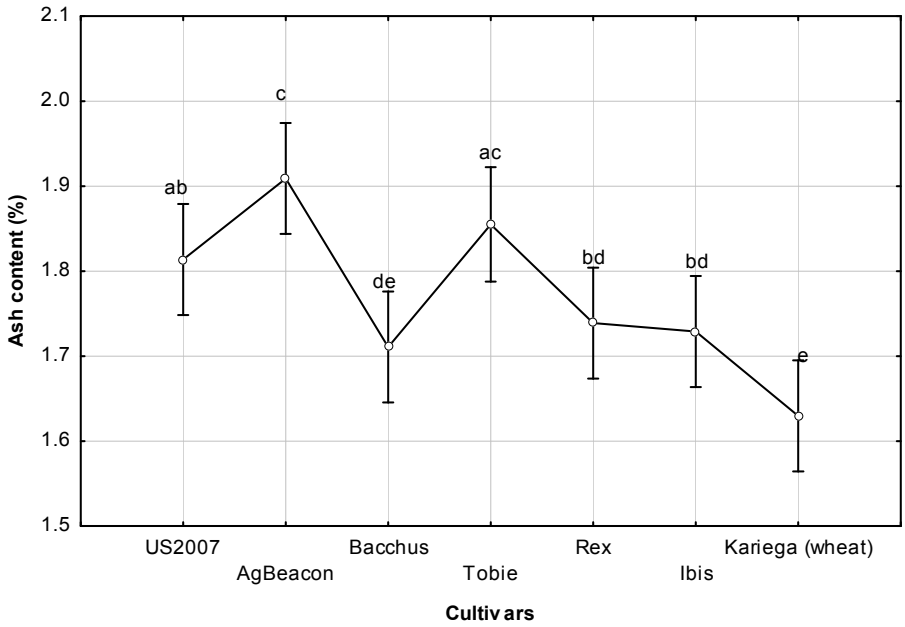


Figure 3.7 Differences between average ash contents obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

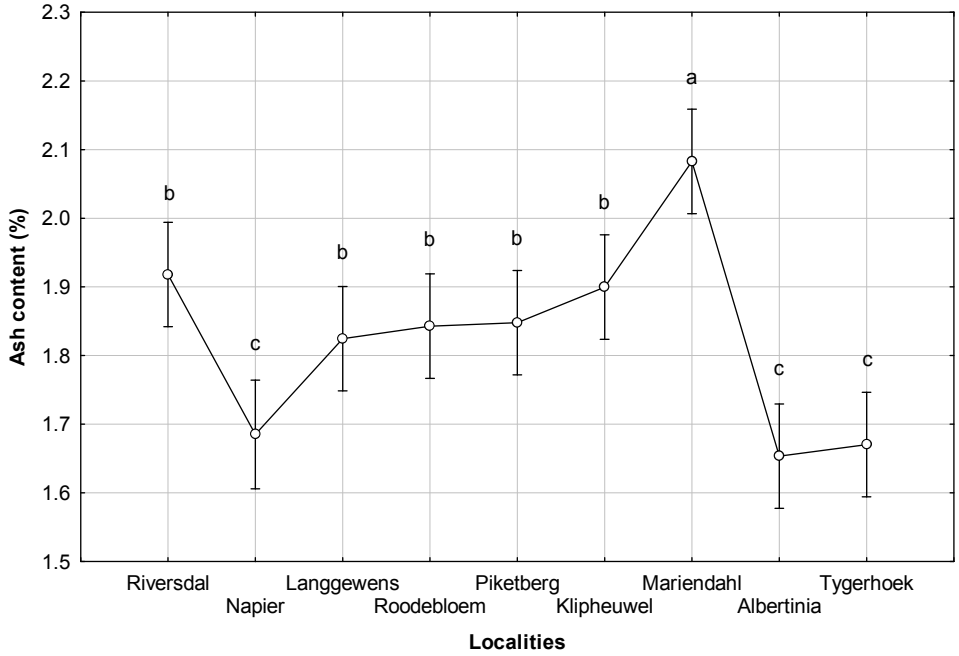


Figure 3.8 Differences between average ash content obtained for localities as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

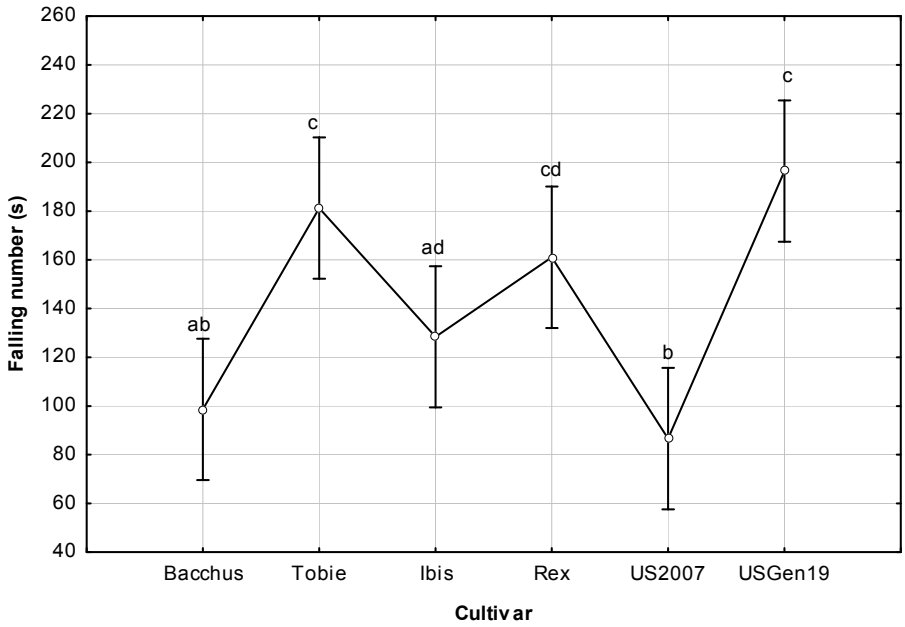


Figure 3.9 Differences between average falling number values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

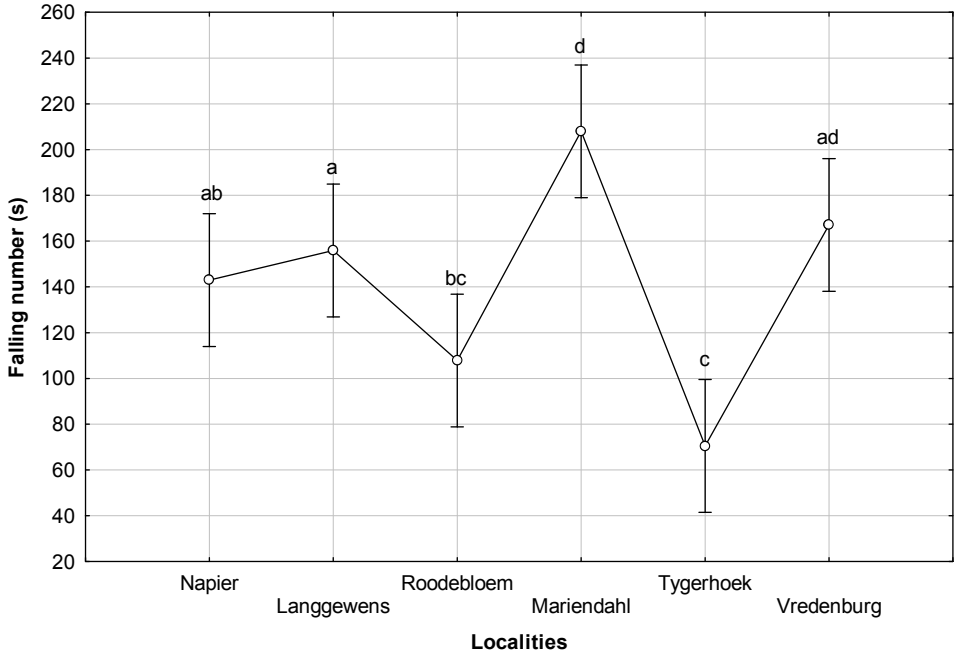


Figure 3.10 Differences between average falling number values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

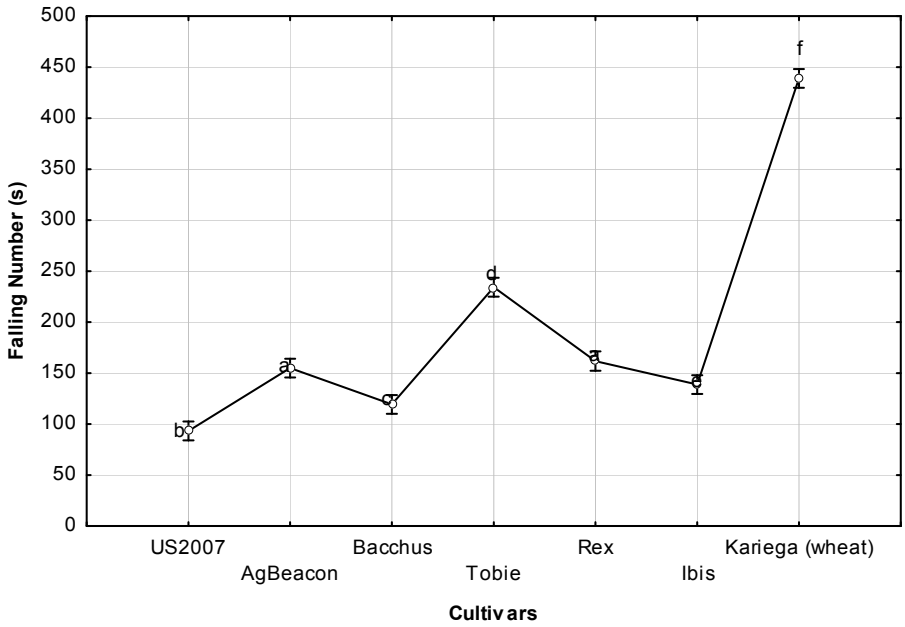


Figure 3.11 Differences between average falling number values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

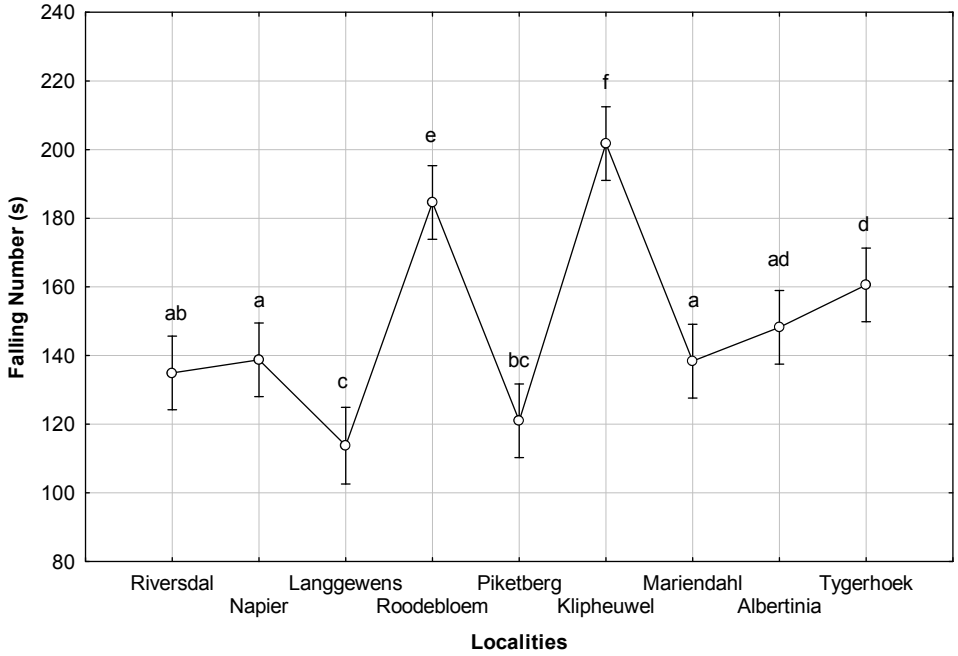


Figure 3.12 Differences between average falling number values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

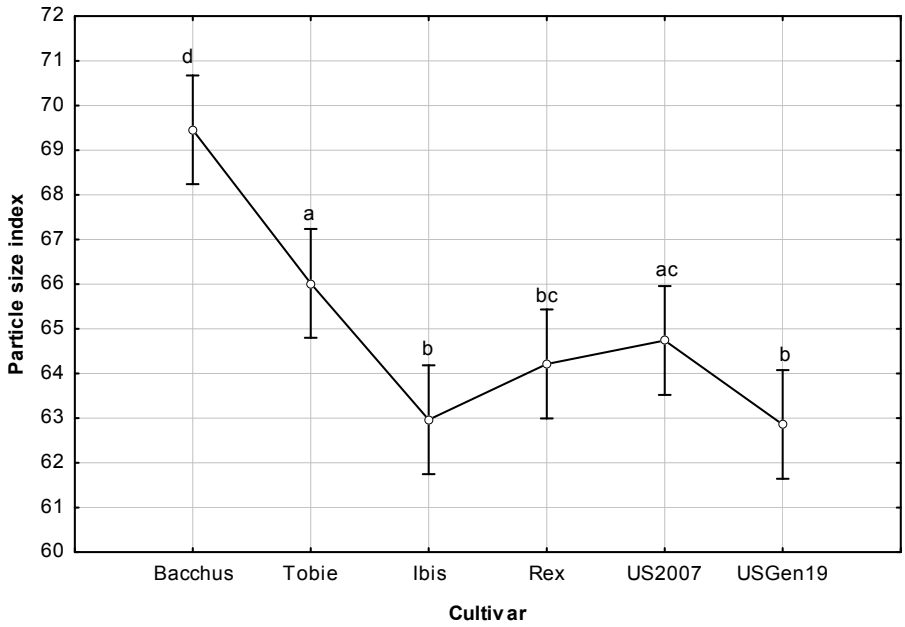


Figure 3.13 Differences between average PSI values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

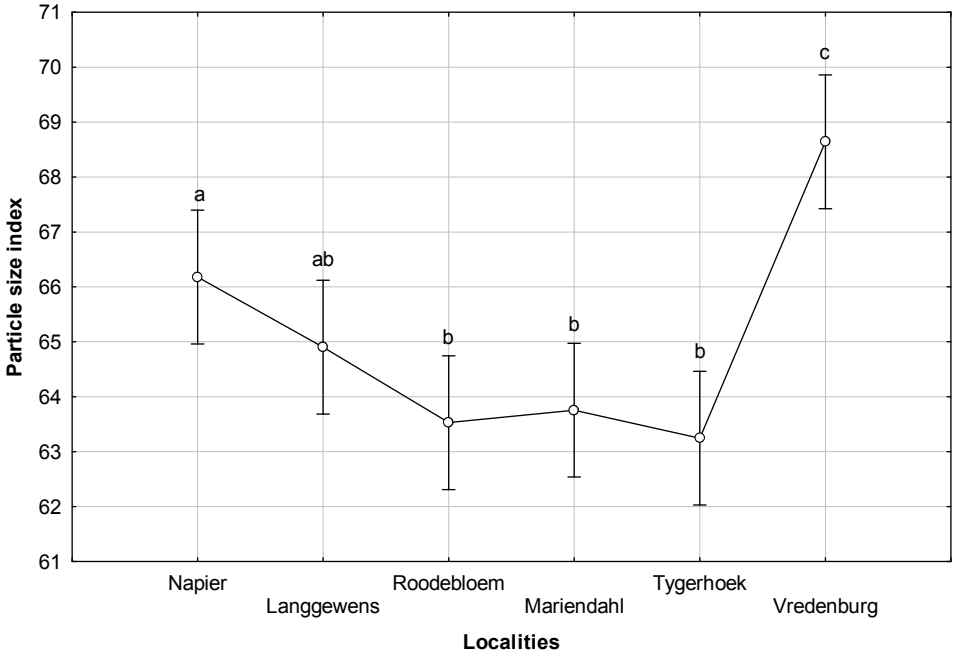


Figure 3.14 Differences between average PSI values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

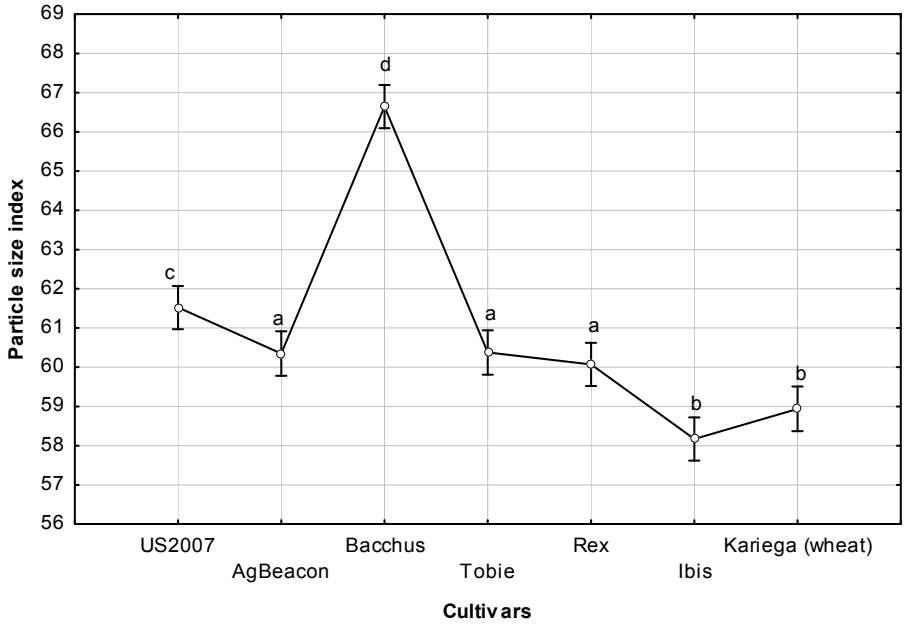


Figure 3.15 Differences between average PSI values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2007 harvest season . Error bars denote 0.95 confidence intervals.

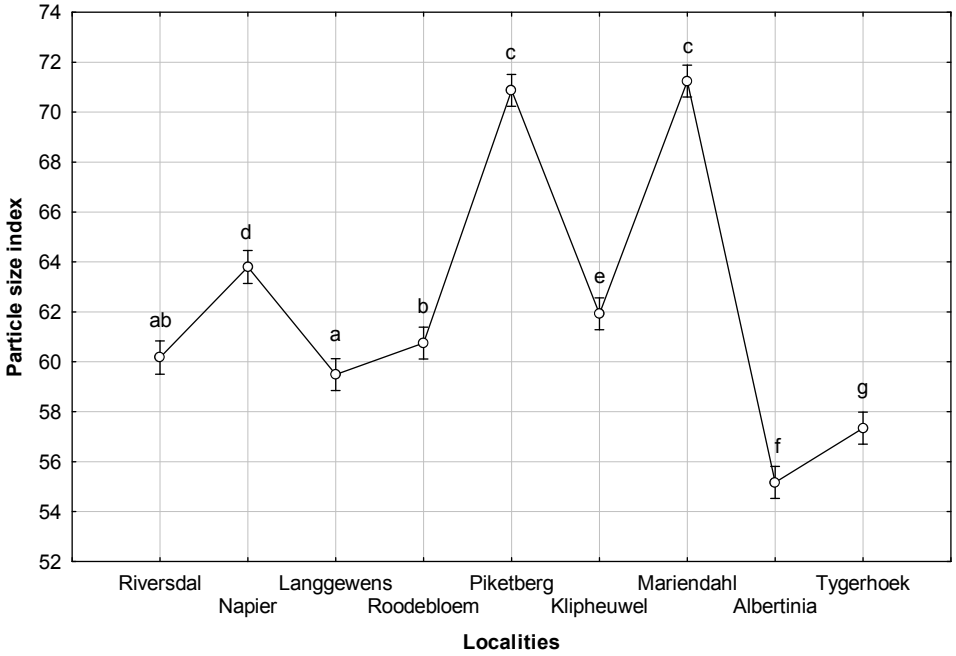


Figure 3.16 Differences between average PSI values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

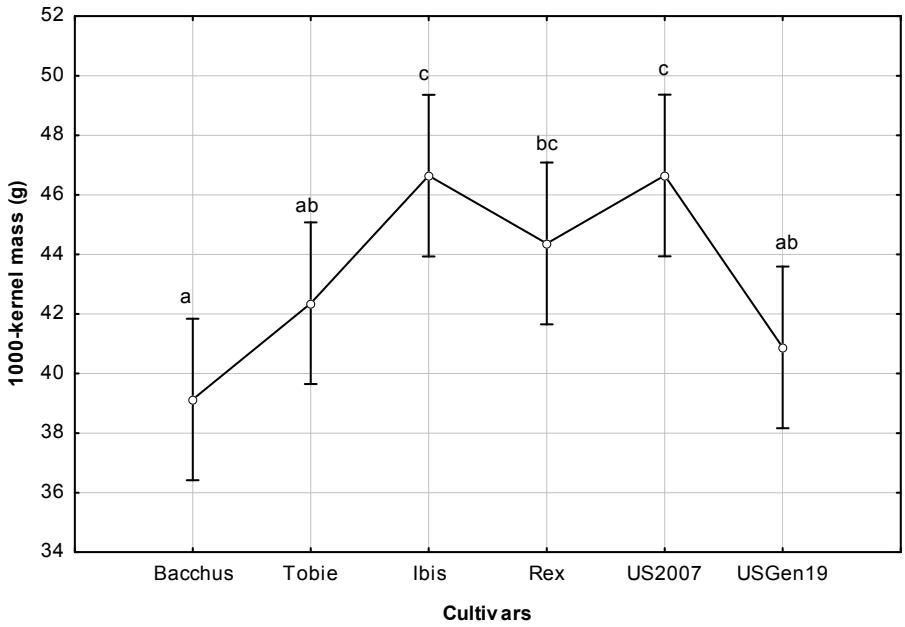


Figure 3.17 Differences between average 1000-kernel mass values obtained for cultivars as determined by ANOVA (P < 0.05) for the 2006 harvest season (only). Error bars denote 0.95 confidence intervals.

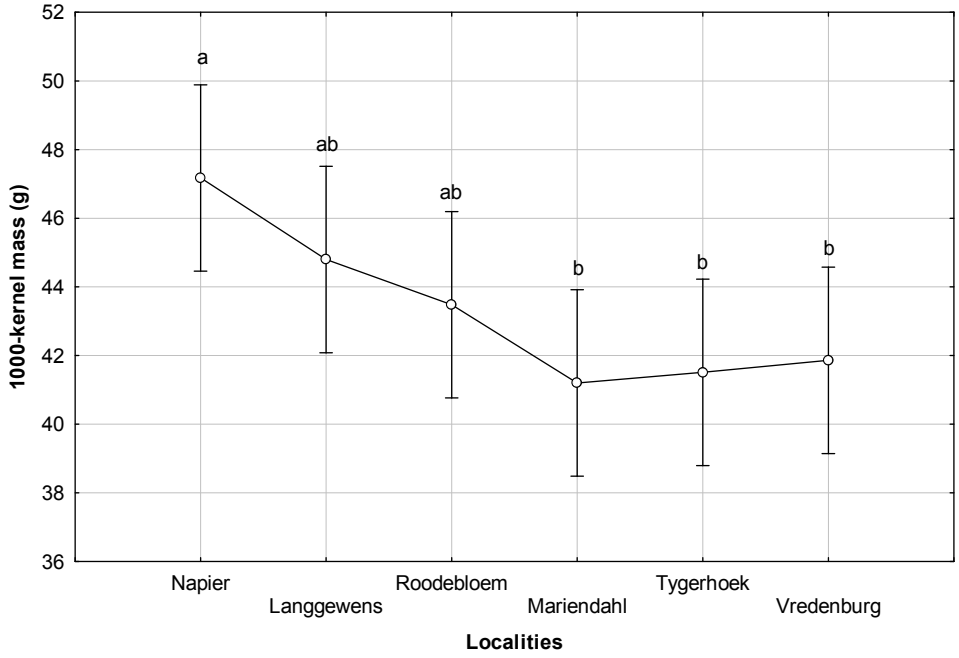


Figure 3.18 Differences between average 1000-kernel mass values obtained for localities as determined by ANOVA (P < 0.05) for the 2006 harvest season (only). Error bars denote 0.95 confidence intervals.

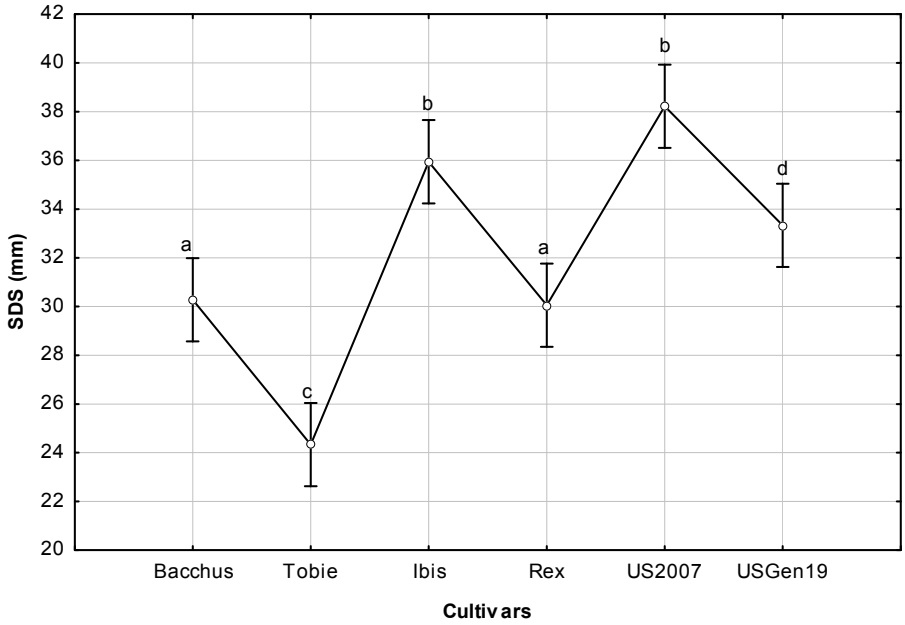


Figure 3.19 Differences between average SDS sedimentation values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

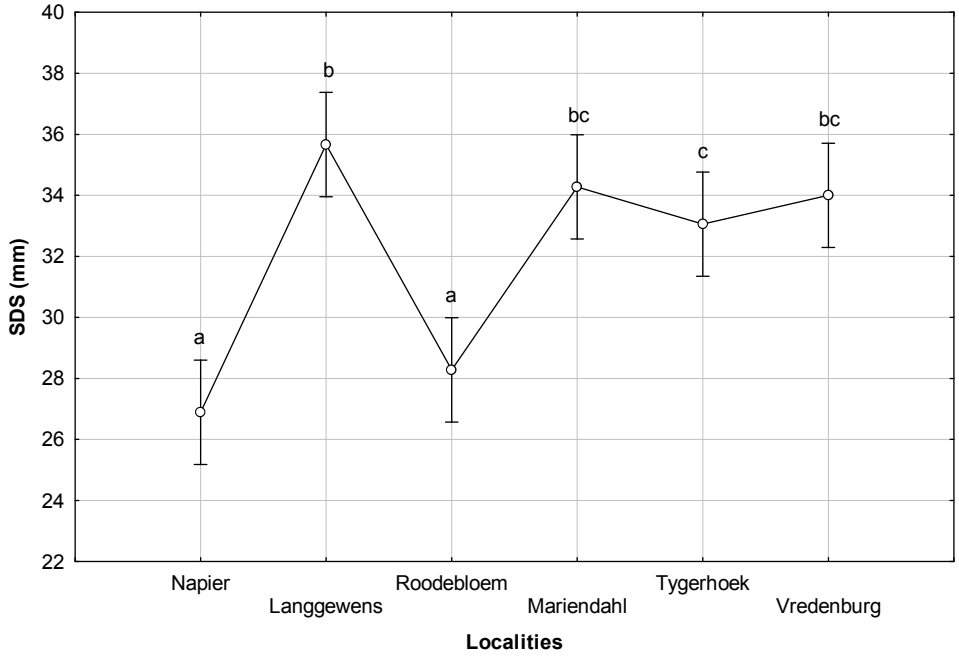


Figure 3.20 Differences between average SDS sedimentation values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2006 harvest season. Error bars denote 0.95 confidence intervals.

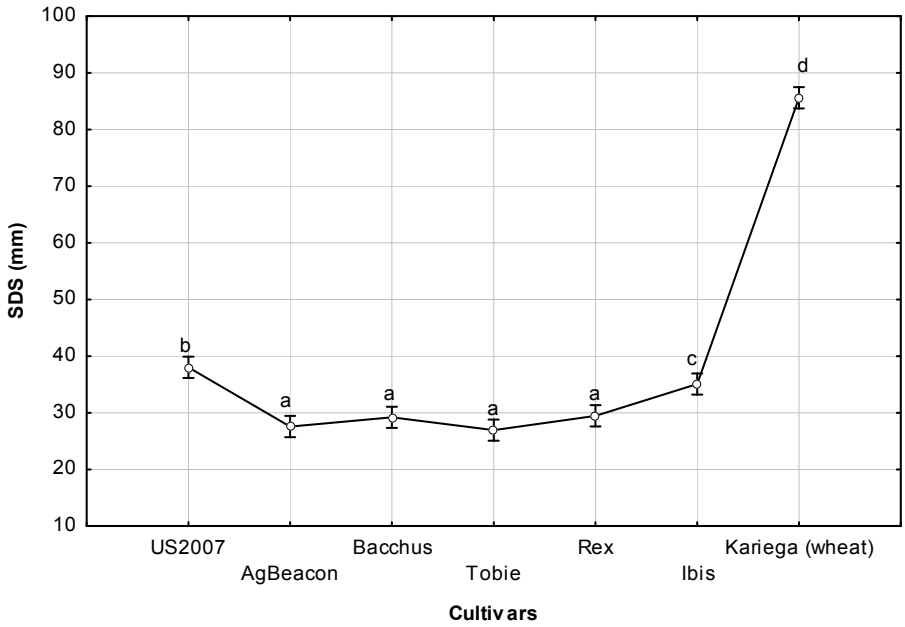


Figure 3.21 Differences between average SDS sedimentation values obtained for cultivars as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

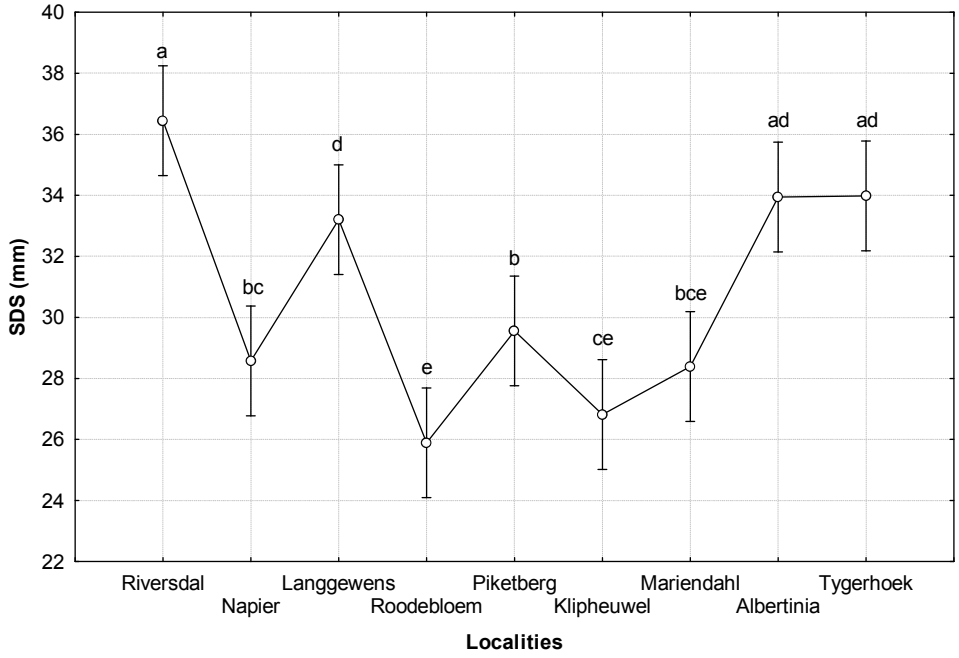


Figure 3.22 Differences between average SDS sedimentation values obtained for localities as determined by ANOVA ($P < 0.05$) for the 2007 harvest season. Error bars denote 0.95 confidence intervals.

Appendix 4

Table 4.1 Rainfall and temperature data for the 2006 and 2007 harvest seasons

Locality	2006 harvest season				2007 harvest season			
	Month	Rainfall (mm)	Max Temp (°C)	Min Temp (°C)	Month	Rainfall (mm)	Max Temp (°C)	Min Temp (°C)
Piketberg	Jan	0	33.9	17.1	Jan	11.8	34.5	16.6
	Feb	1.9	34.1	17.7	Feb	12.3	31.3	15.2
	March	0.9	30.8	13.8	March	16.5	31.6	14.9
	April	25.1	26.7	13.2	April	45.3	26.9	12.8
	May	119.1	19.9	10.4	May	41.4	22.8	10.7
	June	44.2	20.3	8.4	June	120.4	18.1	8.6
	July	31.9	17.2	7.5	July	57.0	17.6	6.3
	August	36.7	18.2	6.5	August	36.0	18.2	6.6
	September	12.3	23.3	7.5	September	2.9	21.6	6.6
	October	8.1	27.5	10	October	19.8	29.1	10.2
	November	23	30.1	12.6	November	12.1	29.0	11.6
	December	1.9	30.2	13.2	December	38.1	33.2	15.5
Napier	Jan	15.6	28	16	Jan	2.2	29.7	15.8
	Feb	5.5	29	17.4	Feb	22.4	28.5	15.3
	March	12.2	26.8	11.7	March	13.3	27.5	13.0
	April	37.7	24	11.3	April	34.2	25.6	11.4
	May	46.2	20.8	8	May	33.6	22.2	9.0
	June	17.8	20.1	5.9	June	34.0	19.7	4.9
	July	131.8	18	6.2	July	76.3	18.0	3.8
	August	74	17.9	6.7	August	31.7	18.6	4.0
	September	22.4	20.5	8.7	September	16.2	21.5	5.6
	October	15.8	23	9.2	October	31.6	23.2	9.5
	November	19.4	25.3	11.7	November	116.4	24.2	10.8
	December	7.9	26.7	12.6	December	40.1	26.1	14.9
Roodebloem	Jan	0			Jan	5.5	29.8	16.8
	Feb	8.9	29.3	18.1	Feb	49.2	28.0	16.5
	March	6.7	27	12.7	March	22.7	27.2	15.0
	April	28.5	23.7	12.3	April	55.9	25.0	12.7
	May	60.9	20.1	8.9	May	87.7	21.6	10.4
	June	43.5	19.3	8.1	June	110.7	18.4	7.9

Table 4.1 continued

	July	87.1	17.6	8.5	July	120.2	17.2	6.2
	August	111	17.3	7.9	August	85.2	17.4	6.7
	September	27.7	20.1	9.9	September	64.6	20.4	8.4
	October	63	22.5	10.8	October	77.7	22.6	10.9
	November	42.9	25	13.3	November	127.4	23.6	12.1
	December	29	26	14.5	December	35.9	26.6	15.4
Tygerhoek	Jan	5.1	26.6	15.1	Jan	3.0	28.5	15.2
	Feb	2.9	27.4	15.4	Feb	28.5	26.7	14.7
	March	3.3	25.1	12.1	March	18.2	25.8	13.3
	April	11.6	21.8	11.5	April	52.1	23.5	12.0
	May	18.9	18.5	9.7	May	73.7	20.0	9.8
	June	19.6	18.1	8.1	June	46.8	17.3	8.2
	July	109.9	15.6	7.5	July	50.8	15.7	6.9
	August	96	15.3	6.9	August	53.1	16.0	6.8
	September	26.7	18.2	9.4	September	13.4	19.1	7.8
	October	31.3	20.9	9.8	October	13.7	21.2	9.5
	November	23	23.6	12	November		23.5	10.1
	December	18.6	24.7	13.1	December	43.0	24.2	13.8
Mariendahl	Jan	3.2	29.9	15	Jan	2.4	30.1	15.3
	Feb	15.6	30.6	15.2	Feb	38.8	28.0	14.2
	March	10	27.7	11.8	March	32.6	28.2	12.8
	April	51.4	24.7	11.5	April	87.2	24.9	11.5
	May	161.7	19.6	9.6	May	137.4	21.7	9.6
	June	87.6	20	8	June	116.8	17.7	7.9
	July	102.6	16.8	8.1	July	131.6	17.1	7.1
	August	88.4	17.8	7.5	August	114.4	17.4	7.6
	September	24	21.8	9	September	39.8	19.8	8.0
	October	39.2	23.8	9.8	October	40.2	24.2	9.9
	November	63.4	26	11.8	November	45.0	24.0	10.7
	December	21.6	25.9	13.3	December	26.6	28.2	14.7
Riversdal	Jan	17	27.3	16.6	Jan	19.8	29.3	16.4
	Feb	19.6	28.7	17.3	Feb	25.5	27.7	16.1
	March	16.2	26.2	13	March	49.2	26.2	13.9
	April	52	23.5	12.3	April	62.8	24.4	12.3
	May	53.8	20.1	8.4	May	91.6	22.2	9.5

Table 4.1 continued

	June	37.7	19.5	6.9	June	29.6	19.9	7.1
	July	83.5	17.8	6.1	July	48.1	17.8	5.1
	August	152.3	17.4	7	August	20.2	18.5	6.1
	September	27.2	19.8	9.3	September	10.6	21.9	7.5
	October	38	22.1	10.7	October	22.7	22.9	10.4
	November	21.3	24.7	12.5	November	129.7	24.0	11.7
	December	23.9	26.1	14.2	December	59.4	25.3	15.2
Langgewens	Jan	0.2	31.6	17.1	Jan	0	31.6	17.2
	Feb	5.2	32	17.4	Feb	37.9	29.2	16.0
	March	4.2	28.9	14.6	March	33.6	29.2	15.6
	April	24.4	25.6	13.5	April	86.1	25.7	13.0
	May	121.6	19.3	11	May	59.3	22.2	11.3
	June	42.7	20	10	June	220.1	17.3	9.5
	July	47.5	16.6	8.3	July	89.8	17.3	7.8
	August	53.3	17.4	7.7	August	79.1	17.1	7.7
	September	30.2	22	10.2	September	32.6	20.1	8.2
	October	27.2	25.1	11.1	October	18.5	25.0	11.0
	November	47.3	27.6	13.6	November	40.0	25.0	11.9
	December	8.4	27.3	14.3	December	26.5	29.6	16.0
Klipheuwel	Jan	1.8	27.7	16.5	Jan	2.1	28.2	17.0
	Feb	1.8	28.7	16.7	Feb	23.8	26.4	15.9
	March	3.7	26.1	14.4	March	19.8	26.8	15.3
	April	10.7	23.6	12.9	April	60.9	23.7	12.9
	May	44.8	19	10.7	May	71.1	20.6	11.2
	June	22.3	19.3	9.7	June	84.8	17.0	9.4
	July	75.6	16.2	8.4	July	65.4	16.7	8.0
	August	65	17	8.1	August	89.2	16.8	8.1
	September	19.8	21	10.9	September	27.7	19.1	9.1
	October	29.6	23	11.7	October	19.1	23.4	11.8
	November	28.8	25.2	13.8	November	31.0	22.7	12.2
	December	20.3	24.7	14.7	December	37.9	26.8	15.5