Improving the agronomic management and utilisation of organic bread making wheat

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Abstract

Yield of wheat produced under organic standards has been repeatedly shown to be 20-40% lower than achieved in conventional farming systems, with reduced protein levels. This has mainly been linked to the inability to (a) control certain diseases and (b) use foliar applications of mineral N-fertilisers late in the growing season. The organic bread making sector in the UK has therefore been unable to meet the requirements of the market without utilising imported high protein organic wheat to augment the lower protein content of the UK crop.

A large scale field trial carried out between 2003 and 2005 at Gilchesters Organic Farm in Northumberland was used to investigate the effect of applying a *Rhizobium* inoculant and green waste compost (GWC) amendments to two-year clover swards. The trial also evaluated the effect of the agronomic management of the pre-crop fertility building on the performance and quality of subsequent wheat varieties. This consisted of three winter and four spring bread making wheat varieties, selected from a range of European wheat breeding programmes. Results identified that the addition of *Rhizobium* inoculant to red clover seeds improved the establishment of the clover crop, total rhizobia number per plant and rhizobia volume. The establishment of clover plants was significantly higher with inoculum (190 plants m^{-2}) than without (152 plants m^{-2}). The numbers of nodules per plant in the year of crop establishment were also significantly higher with inoculum than without. The nodule volume was also significantly higher with the use of inoculum than without, with a 50% increase in the size of nodules. In the second year of growth the established clover swards in the absence of inoculum had a mean plant count of 150 m^{-2}, with a maximum predicted nodule number being achieved from 425 plants m^{-2}. Clover swards established with the inoculant had a mean plant count of 200 m^{-2}, with the maximum predicted nodule number achieved with 350 plants m^{-2}.

Results from the subsequent wheat variety trial showed that variety choice had a clear effect on both winter and spring wheat yields, but the improvements to fertility management practices also significantly affected yield and protein quality. For the winter wheat varieties use of *Rhizobium* inoculant significantly increased grain yield by 0.64 t ha^{-1}, while use of GWC improved yield by 0.35 t ha^{-1} . For the spring wheats, grain yield was higher in the presence of clover inoculum (7.04 t ha^{-1}) than in the absence (6.77 t ha^{-1}) by 0.27 t ha^{-1}, but the use of GWC had no effect on yield. This demonstrates clearly that yields in organic wheat production can be significantly increased by improved variety choice and fertility management regimes.

Protein content for the winter wheat varieties was significantly higher in the absence of the inoculum (12.5 %) than with inoculum (11.6 %) but from grains with a smaller specific weight. The addition of GWC also significantly increased the protein content (from 11.82 % without GWC to 12.38 %). In the absence of the inoculum, grain specific weights were improved by the addition of GWC for all three winter wheat varieties. For the spring wheat varieties *Rhizobium* inoculum and GWC amendment had no significant effects on any of the grain quality parameters.

An additional variety trial at Gilchesters Organic Farm evaluated the field performance of six spring wheat varieties from a range of European breeding programmes for their performance and grain quality in both 2006 and 2007. Results identified that variety choice had a considerable impact on yield and grain quality with large differences between seasons. The year 2006 was the best year for agronomic performance with favourable growing conditions, but yields in 2007 were significantly lower because of heavy rainfall combined with lower solar radiation levels during grain fill. Paragon and Fasan were the top yielding varieties in both years but Tybalt and Fasan produced grains of the highest grain quality. The baking performance of the varieties was in contrast to the grain quality results, i.e. Zebra, Fasan and Paragon all produced high volume loaves with high bulk fermentation after proving despite the grain analysis suggesting they were below the NABIM (National Association of British and Irish Millers) standard required for bread making. Tybalt, however, failed to hold the bulk fermentation after proving, collapsing prior to baking despite the grain quality results identifying good baking quality characteristics. Varieties Paragon and Fasan produced the best overall yield and baking results.

A fertility trial was also used to evaluate the potential for additional amendments of organic manures and fertilisers, i.e. Farm Yard Manure (FYM), Green Waste Compost (GWC), Chicken Manure Pellets (CMP) and a combination of FYM + CMP applied at rates of 250 and 125 kg ha^{-1}, on the performance of the spring wheat variety Paragon in 2006 and 2007. Results showed no significant benefits from the rate or type of fertiliser to the yield or grain quality. Although responses to fertility inputs were small in the current season of application, there are likely to be cumulative benefits later in the rotation as organic N becomes mineralised. Fertility management in an organic system is a long-term strategy based on progressively building fertility by growing N-fixing crops and supplementing this with available organic manures, as appropriate for any given organic rotation.

This thesis is dedicated to my wife Sybille who has walked this path with me every step of the way, from that first question: "Why are we not organic?" To my children Rosie, Harry and Flo who continue to inspire me, to my mother Marianne for her continued love and support and to my father John Wilkinson whose beautiful mathematical mind has expanded our ability to interpret the science, allowing it to speak to us in an entirely new language. R is your legacy to a whole generation of scientific researchers at University.

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CHAPTER 1: General Introduction

1.1 UK Organic Market

The UK organic market grew by 6% in 2017, worth £2.2 billion, accounting for 1.6% of the total UK food and drink market. In this period sales of organics in independent retail grew by 9.7%, home delivery by 9.5% and in supermarkets by 4.2% (Anon., 2018).

The rise in consumption of organic produce has been driven by a combination of factors, amongst which are a desire by consumers to purchase foods which promote local production with short supply chains, where the provenance of their food is clearly identifiable and with benefits to their health. Produce saw the biggest single growth within the sector of 6.5%, from a base of over 6000 registered producers in the UK.

1.2 Production Comparisons

Consumers expect organic produce to have higher environmental, health and sensory related qualities when compared to conventional. Comparisons between dairy products (Palupi *et al.*, 2012) identified higher protein, omega-3 and -6 and conjugated linoleic acid levels in organic dairy produce. In the meat industry meta-analysis on sixty-seven published studies (Średnicka-Tober *et al.*, 2016) identified significant differences in fatty acid profiles. Meta-analysis on 343 peer-reviewed publications (Baranski *et al.*, 2014) indicated statistically significant and meaningful differences between organic and non-organic crops and crop-based foods, particularly higher in a range of antioxidants and lower in Cd and pesticide residues in organic crops. However, the UK Food Standards Agency report (FSA 2009) concluded that conventionally grown foods were not nutritionally different to organic. Comparisons in crop rotations (Barbieri *et al.*, 2017) show organic rotations are 15% longer with a higher crop diversity which is more widely distributed across the agricultural landscape. Wheat specific meta-analysis from European and North American peer-reviewed publications (Hossard *et al.*, 2016b) confirmed significant reductions in mineral N and pesticide use for low-input and organic wheat crops, as well as significantly lower yield (20-25%).

The differences in environmental impacts by meta-analysis of 109 cross referenced publications (Mondelaers *et al.*, 2009) indicate that organic farming has soils with a higher organic matter content and contributes positively to both agro-biodiversity and natural biodiversity. It also scored better than conventional farming with lower nitrate and phosphorus leaching and lower greenhouse gas emissions.

1.3 UK Wheat Production

Between 1998 and 2003 the retail value of the organic bread and cereals markets rose to £75 million p.a. (an increase of 250% over the five years.) The development of organic bakery products was seen as a way to add value and drive greater product diversity and innovation. Since that time the organic sector has grown to meet the demands of consumers purchasing for both a health conscious option and a positive environmental impact through their consumer choices.

In 2003 organic wheat production in the UK did not meet the functional/quality requirements of industrial baking and was normally used in combination with imported wheats of higher functional quality to make good industrial bread. Consequently the majority of organic wheat for bread making was imported and the maximum proportion of UK organic wheat that was used stood at approximately 25%. It was not unrealistic at the time to believe that improvements in the functional quality of UK organic wheat could allow the level of its incorporation to rise to approximately 50% (closer to the organic action plan target of 70%, launched by DEFRA in 2005). This was calculated to increase revenue to UK growers of around £1.9 million p.a.

When reviewing the factors affecting wheat quality from both conventional and organic cereal production systems (Bilsborrow *et al.*, 2013) variety is the dominant driver. Even where differing pedo-climatic conditions are evaluated in variety and fertility trials (Borghi *et al.*, 1995), the same cultivar grown at different locations or even in the same location but under different crop management regimes may produce grains of similar protein concentrations but different bread making protein quality.

1.4 Limitations of organic cereal production

The availability of volume milling quality wheat from UK organic production is limited by the current yield gap between conventional and organic. A recent systematic review (Hossard *et al.*, 2016b) demonstrates differences of 20-30% across Europe and northern America. This gap in production can be attributed to nutrient availability through the fertility management practices available to organic producers (Bilsborrow *et al.*, 2013), combined with the limited pest and disease control and the weed management options available (GutiÉRrez-Alamo *et al.*, 2008). The influence of these three major factors on final grain yield and quality in organic cereal production is then exaggerated when the greatest single component in any interaction is wheat variety (Carcea *et al.*, 2006). Wheat production is dominated by genetically uniform varieties with high yields and adaptability to a wide geographic area (Murphy *et al.*, 2005). However, once synthetic fertilisers, fungicides and herbicides become unavailable these varieties become less yield stable, as they are less adaptable to low soil fertility and less resistant to pathogens. Comparative studies into performances for spring (Osman *et al.*, 2016) and winter (Buchi *et al.*, 2016) wheats, disease resistance (Finckh *et al.*, 2000; Bilsborrow *et al.*, 2013), weed control (Costanzo and Barberi, 2016), fertility management (Przystalski *et al.*, 2008) and environmental impact all attest to the impact of variety on the harvest data from each of these interactions. These and many other studies have led to a wide review of what constitutes the right variety for organic production (Feil, 1992a; Miko *et al.*, 2014; Migliorini *et al.*, 2016) and the need for breeding programmes to develop cereals with more suitable traits for organic farming (Murphy *et al.*, 2005; Wolfe *et al.*, 2008).

1.5 Aims and Objectives

The overall aim of this thesis is to improve bread making quality of UK organic wheat by addressing deficiencies at different steps in the production chain. This will be achieved by:

- Evaluating the role of grass-clover as a fertility building pre-crop on the yield and quality of organic spring and winter wheat.
- Evaluating the effects of variety on yield and quality of organic spring wheat.
- Optimising fertility management practices to provide grain with functional attributes best suited to organic bread making practices.
- Evaluating the effects of site and season on the performance of organic spring wheat.
- Evaluating how grain quality can influence processability for bread-making.

CHAPTER 2: Literature Review

2.1 European Organic Food Trends

In 2016 the total harvested production of all cereals in the EU-28 was 301 million tonnes, representing 11.6% of global cereal production. Common wheat accounted for 44.7% of output, barley 19.9% and rye (plus winter cereal mixtures) 2.6% (Fig. 2.1) (Eurostat, 2017). France (18%), Germany (15%) and Poland (9.9%) together contributed 43% of the total EU-28 output. Spain was the next largest with 8% (Fig. 2.1).

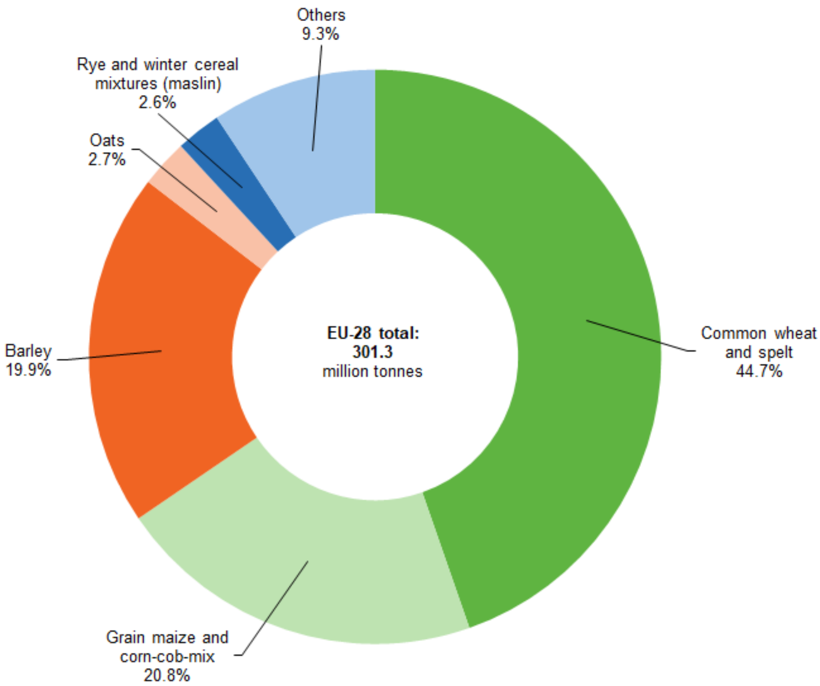


Fig. 2.1 EU-28 Cereal production 2016 (Eurostat, 2017)

France was the largest producer of common wheat, Germany was the largest producer of barley (Fig. 2.2).

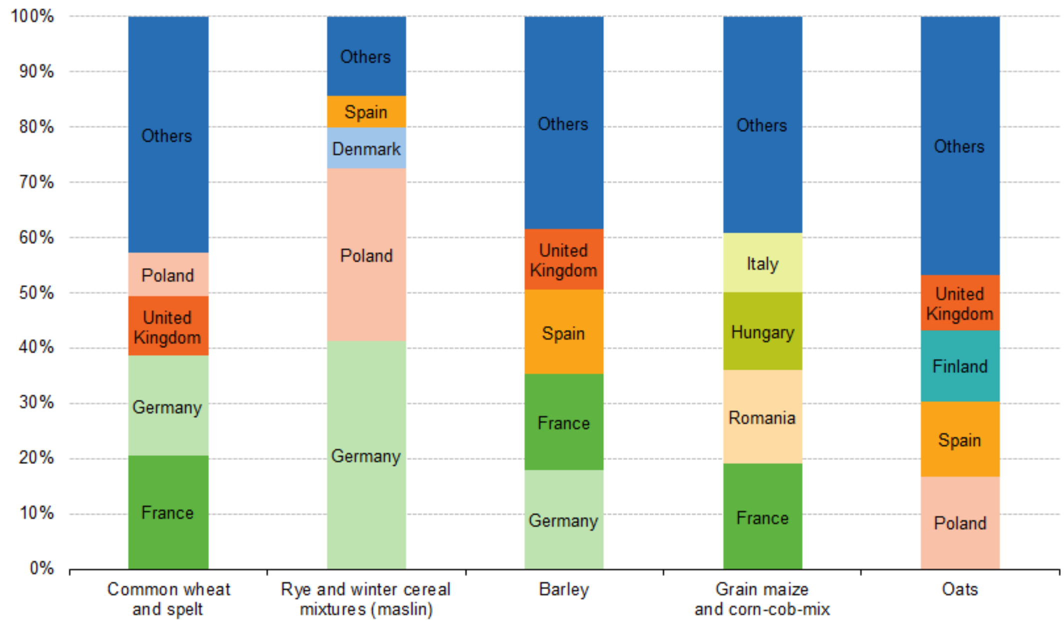


Fig. 2.2 EU-28 cereal production percentage by country (Eurostat, 2017)

Within this organic cereal production in 2016 was 1.9 million tonnes, or 0.006% of EU-28 total production.

The total organic area made up 6.7% of the total EU-28 Utilised Agricultural Area (UAA). Of this 11.9 million hectares in organic land, 8.9% was in the production of cereals excluding rice (Eurostat, 2017).

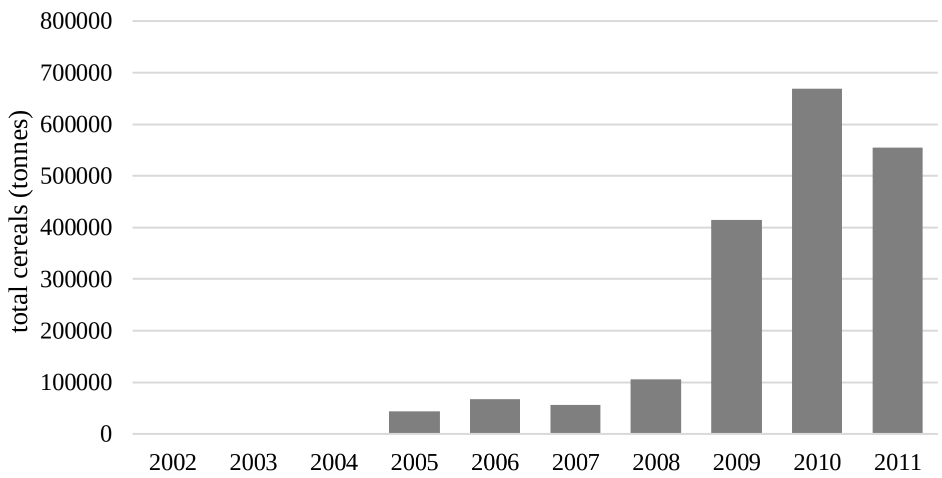


Fig. 2.3 EU-28 organic cereal production 2002-2011 (Eurostat, 2017)

EU organic cereal production levels have risen sharply from their modest levels at the beginning of the 21st century to meet consumer demand over the last fifteen years. A major driver for the increasing consumer demand for organic foods is the scientific evidence for potential nutritional and health benefits of organic crop consumption (Brandt, 2013; Baranski *et al.*, 2014), especially wholegrains (Kirwan *et al.*, 2016; Karl *et al.*, 2017).

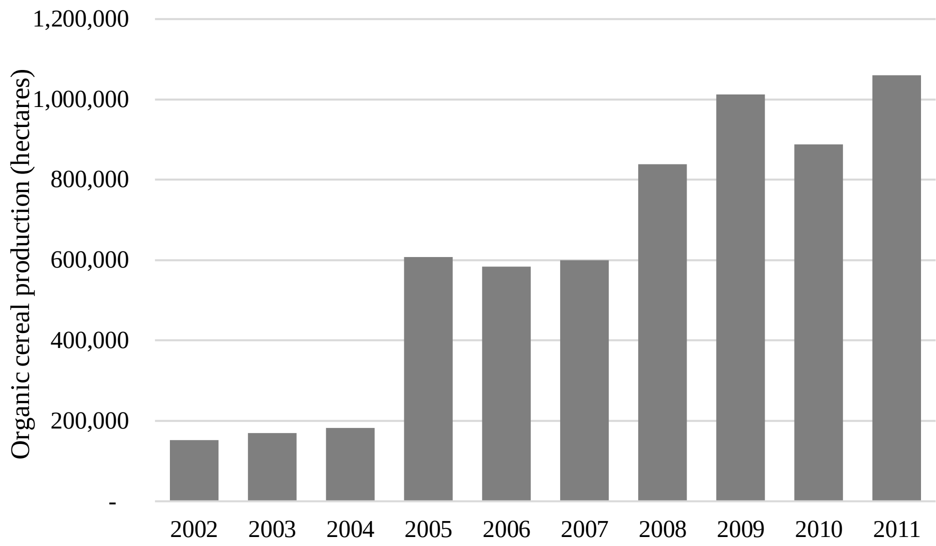


Fig. 2.4 EU-28 land area in organic cereal production (hectares) 2001-2011

From 2012 to 2016 total EU organic cereal production increased from 1.5 million tonnes to over 2 million, with wheat as the dominant commodity which peaked at 1.07 million tonnes in 2015, representing 50% of all organic cereals. Barley and oats represent the next largest group with 900,000 tonnes in 2016, a combined share of 37% of total organic cereal production (Fig. 2.5) (Eurostat, 2017).

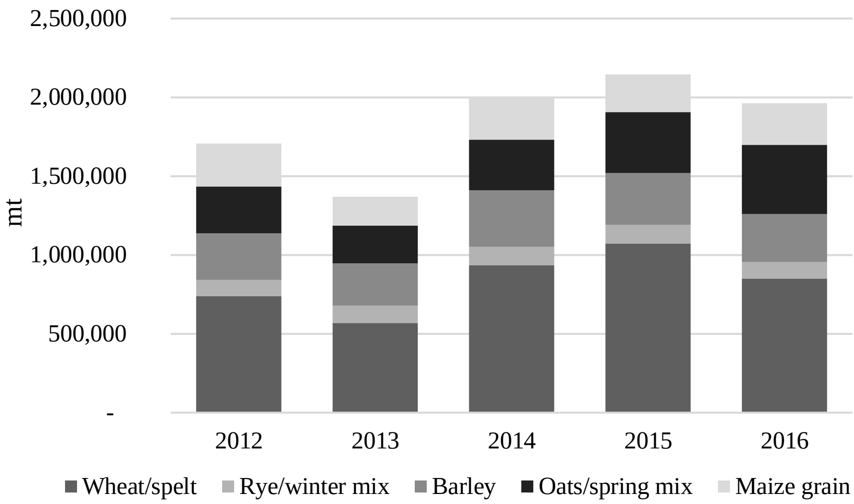


Fig. 2.5 EU-28 organic cereal and grain production 2012-2016 (Eurostat, 2017)

2.2 UK Organic Production Trends

Based on the farm-gate value of unprocessed food in 2015, the UK supplied over half (52%) of the food consumed in the UK. The leading foreign suppliers of food consumed in the UK were countries from the EU (29%) and three countries globally (USA, Canada and France) accounted for 54% of all un-milled grain imports.

Food exports (at 2016 prices) rose 23% between 2009 and 2011, due largely to increases in the existing markets. Cereals is the second largest export group with a value of £2.3 billion, but cereal imports still remain higher at £3.3 billion, the fourth largest import group (after fruit, meat and beverages.) The UK cereal processors, organic and conventional, are therefore still highly dependent upon large volumes of imported cereals, mostly high grain quality and high protein wheat for a blended high quality grist for baking, from a very narrow production base.

Production and consumption figures of organic food over the period 2002-2016 show UK volumes have plateaued. The total UK organic agricultural area, consisting of land certified as organic and land in conversion to organic, peaked in 2008/09 at between 730 and 740,000 ha and declined thereafter to a six year low in 2014 of 296,683 ha - a reduction of 2.2% on 2013. The area in conversion peaked in 2007/08 at 160,000 ha and has since decreased to just 19,675 hectares in 2014 (Scott, 2015). UK production alone has thus struggled to fully meet the demands of the consumer, peaking in 2009 with just over 60,000 hectares of organic cereal production (Fig. 2.6). UK production of organic cereals stood at 146,578 tonnes in 2013, fluctuating between 129 and 136,000 tonnes from 2014 to 2016 (Eurostat, 2017). This is a small percentage (0.001%) of the total UK production of 14.8 million tonnes (2017/18) of cereals from all agricultural production.

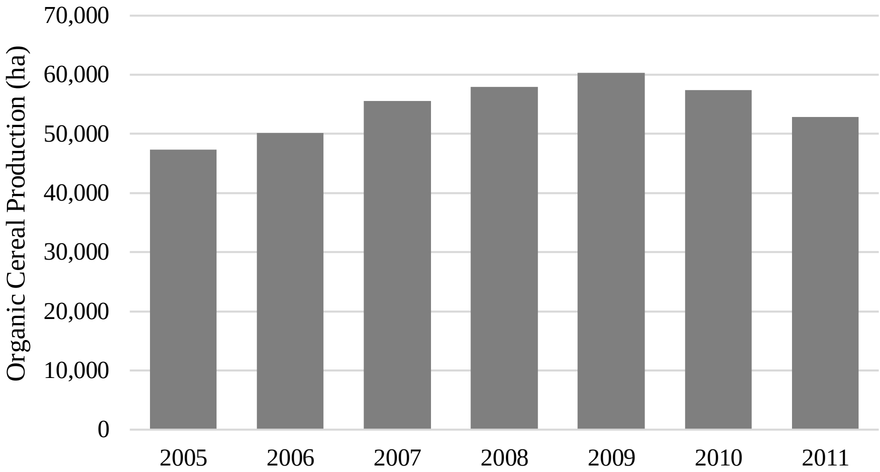


Fig. 2.6 UK land area (hectares) in organic cereal production 2002-2011

Wheat and spelt accounted for 31% of the UK organic cereal harvest, a little less than barley which accounted for 33%. Oats represented 27% and rye 2% (Fig. 2.7) (Eurostat, 2017).

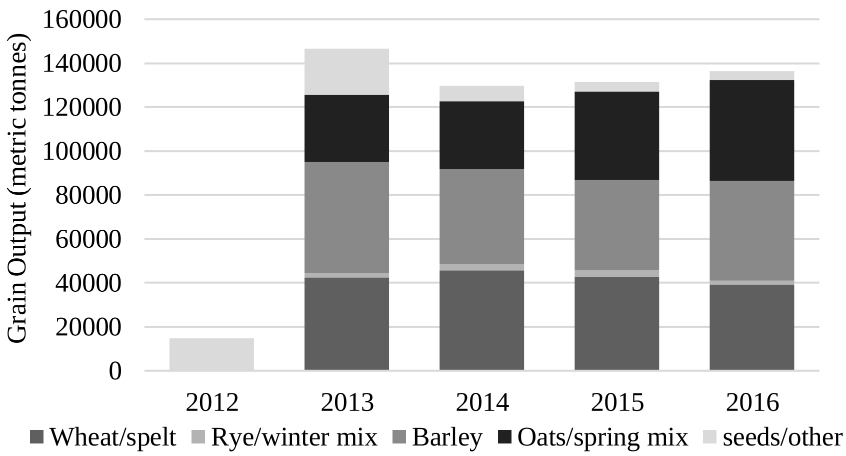


Fig. 2.7 UK cereal and grain output 2012-2016

The number of organic producers and processors peaked in 2007/08 at around 7650, the same time as the peak in area of organic land in the UK. Organic producer and processor numbers have been steadily declining throughout the UK over the past six years to around 6200 (Scott, 2015).

2.3 UK Organic Food Trends

The peak in UK production of organic food coincided with the start of the recession and the subsequent fall in household incomes and the rise in food prices ranging from 14% to 45% (DEFRA, 2016). The shift away from the more expensive organic foods did, however, see a continued rise in the demand for bread and bakery products within the food sector as a whole, despite price increases in cereals, bread and flour of 27% (Statista, 2017). In 2015, compared to 2007, the lowest income households purchased 32% less carcass meat, 18% less fish, and 12% less fresh and processed potatoes (DEFRA, 2016), whereas purchases of cakes, buns and pastries increased 6% between 2007 and 2015 and purchases of confectionery increased 6%. Between 2007 and 2015, average households (av. income of £31,000) traded down to cheaper products to save 4.8% in food purchases (DEFRA, 2016). That notwithstanding, the largest single segment of the UK food market remains bread and bakery products, with a market volume of £16,233 million, expected to continue growing annually by 2.9%. The total revenue of the UK food market is currently £96,823 million (Statista, 2017).

The recession also saw the emergence of the artisan and small craft bakery at this time, with first time entrants into the bakery business forming start-up businesses as consumers moved away from highly processed products with questionable nutritional quality to locally sourced bread with short chain supplies of UK cereals. Government policy to encourage and promote wholegrain in the diet (Kyrø and Tjønneland, 2016) has simultaneously improved consumer awareness for wholemeal breads. The customer base at Gilchesters Organics Mill, Northumberland changed from 85% home/domestic users in 2006 to 2009 to 70% artisan bakeries in 2017-18. This represented a five-fold increase in bakery customers with a 350% increase in sales (Wilkinson, 2018). In 2016 there were 6600 micro, small and medium sized enterprises (SMEs) in the food and drink sector (96% of all businesses) with turnover of £19 billion and 115,000 employees; one third (2250) are manufacturers of bakery products with turnover of £4 billion.

Sales of ethical produce have increased year on year since 2007 in the UK, despite the economic downturn. Rainforest Alliance made up the largest single share in 2014, accounting for 24% of the total ethical food sector at £2.0 billion; an increase of 3.6% on 2013. Fairtrade and organic products are the next largest contributors at 19% each (£1.6 bn and £1.7 bn respectively). Sales of organic food and drink have been steady in the last few years, although still 16% down on their peak in 2008 (ECRA, 2016).

2016 recorded the fifth year in a row of strong growth in the organic sector. Organic sales topped £2.09 billion, an increase of 7.1% in 2016, compared to the 12.5% drop in 2008. The organic food sector represented 1.5% of the total UK food and drink market (Anon, 2016) and 4% of global organic sales, which is currently valued at $81 billion. By comparison organic sales in Denmark are 10% of their domestic market share.

The Soil Association organic market report breakdown of consumer trends in the UK market reveals 39% of shoppers buy organic on a weekly basis (Anon, 2016). Supermarkets account for 69% of all purchases, showing a growth of 6% in organic purchases in 2016, independent retailer sales grew by 6.3% and sales of organic through home delivery grew by 10.5%.

2.4 Agronomic management of organic wheat

Recent systematic reviews have concluded that organic arable yields are mainly lower, with an estimated mean yield loss ranging between 11 and 13% of conventional production (Hossard *et al.*, 2016b). However, the yield gap varies between crop species, with tuber crops having a greater yield gap than cereals. There are a number of factors contributing to this yield difference, which will be discussed below.

2.4.1 Variety Selection

It is well known that varieties differ in their ability to produce the characteristics desired by millers/processors. Both spring and winter wheats are suited to organic production methods with winter wheats producing the highest yields under organic conditions (up to 7.5 t/ha) and spring wheats achieving higher quality in terms of protein content for a lower yielding crop. In the north of England, with fewer machinery workdays, spring wheats will generally not mature until September, with the possibility of exposure to adverse weather conditions. A winter wheat variety with improved grain quality would therefore be of benefit to organic growers in the north. The environment where selection is carried out has a great influence on the traits of bread wheat lines (Miko *et al.*, 2014). There is therefore an urgent need to introduce genetic variation (incorporating traits from landraces and older varieties which have generally become adapted to low-input /organic production) within the breeding programmes in Europe. This would stabilise organic production through varieties already adapted to specific regional and local pedo-climatic conditions (Migliorini *et al.*, 2016), which can give clear benefits across the entire cropping system.

Varieties that are adapted to specific regional and local pedo-climatic conditions would also become important in building nutrient and food security into our long term sustainable food production strategy. Where modern composite cross populations have been created and tested against pure lines, a greater degree of stability was recorded in the composite populations for grain quality characteristics (Wolfe *et al.*, 2008; Doring *et al.*, 2015). Future food security and food safety are threatened by an over-reliance on high external inputs and a narrow genetic base, because (a) traditional crops, varieties and landraces have been replaced by high yielding genetically uniform genotypes and (b) a small number of crops suitable for large-scale cultivation have replaced the much greater diversity of crop species and landraces that were grown until the early 20th century. Many of these, now minor or underutilised species/landraces, were grown in the past because they were associated with certain agronomic/rotational and/or nutritional/health benefits (Finckh *et al.*, 2000). For example there is evidence that local cereal landraces, through continuous cropping exposure and selection in diverse environments, are more resilient than modern cultivars to biotic and abiotic stresses (Migliorini *et al.*, 2016). There has been a clear shift in recent years towards heritage wheats and in particular spelt, emmer and einkorn with their potential nutritional and health benefits (Antognoni *et al.*, 2017). This has been helped by the significant increase in the number of artisan bakers where the use of heritage and older grains has created a new and distinct market from the mass produced supermarket loaf which has dominated the market over the last few decades.

Traditional conventional breeding programmes are very effective at developing varieties to be used in fairly homogeneous farming systems, but are less effective in organic production systems where agronomic practices are more variable due to the greater need to consider local pedo-climatic, agronomic (e.g. type of organic fertilisers available) and market conditions needed in organic production. Most varieties, therefore, currently available to organic producers are from breeding programmes in which selection was carried out in the background of fertility management practices developed for intensive conventional production (in particular high split-dose mineral nitrogen fertilisation regimes). However, in organic production fertility management practices and nutrient availability patterns to wheat crops are significantly different and more viable (Bilsborrow *et al.*, 2013). It has therefore been hypothesised that the current varieties may not be best suited to achieve satisfactory baking quality characteristics under organic fertility management regimes (Przystalski *et al.*, 2008). So while organic production is already limited by some agronomic and management requirements it may be particularly limited by the lack of organic seeds and varieties bred for organic conditions.

The Council Regulation (EC) No 834/2007 for organic production and the labelling of organic products stipulates that for the production of products other than seed and vegetative propagating material only organically produced seed and propagating material shall be used. However production varies by country with France, Spain, Germany and Italy accounting for 53% of the organically farmed area, with pasture and meadow representing 45.4% of this. However, in other countries, including the Czech Republic, Austria and Slovakia, more than 65% of the organic area used is in permanent grassland (Eurostat, 2016). This implies that a lot of organic farmers use permitted derogations for using untreated conventionally bred and selected seed material for their production.

The number of plant breeding companies in the EU carrying out organic breeding is very low and organic seed production represents a small proportion of the total seed market, 12.9% for wheat in Austria (BAES, 2016), a country where the sector is comparatively well developed. There is therefore an urgent need to expand the availability and range of organic seed. It is estimated that more than 95% of organic agriculture is based on crop varieties that were bred for the high-input conventional sector with selection taking place under these conditions (Lammerts van Bueren *et al.*, 2011).

Modern plant breeding programmes outside the EU use methods which are not compatible with the organic principles e.g. genetic modification (IFOAM, 2017). On the other hand, traditional plant breeding involving quantitative genetic/phenotypic selection approaches, which compare genotypes for a wide range of traits over a range of contrasting environments, is extremely time consuming and expensive and is therefore difficult to fully exploit especially for the smaller organic and low-input market sectors. Much organic production is realised by using varieties selected under conventional high-input agricultural conditions but these modern cultivars have been found to be less robust under conditions of greater field heterogeneity such as those often found in organic/low-input systems (Wolfe *et al.*, 2008).

Organic wheat, produced without the use of foliar feeding methods (e.g. foliar urea) to enhance grain protein content, may result in a low protein grain, resulting in a negative effect on bread making quality. There appears to be a positive correlation between straw length and protein content/baking quality (Eisele and Köpke, 1997; Jones, 1998). These trials addressed the suitability of varieties from conventional wheat breeding programmes for organic production and allowed early evaluation of new varieties being bred for low input and organic fertility management systems to be assessed concurrently. Norwegian Graminor (cv Zebra), German Lochow Petkus (cv Bussard, Fasan) and the Swiss GZPK (cv Asita, Wiwa) breeders have introduced varieties with greater straw height and improved grain quality characteristics, with perceived better nitrogen scavenging root systems for both winter and spring crops for organic cereal production since 2006 (Haberle *et al.*, 1995). This requires varieties with a genetic predisposition to stimulate mineralisation of nitrogen from organic matter in the rhizosphere and for this to be expressed with higher yields and/or quality characteristics in compost and pre-crop fertility management regimes (Murphy *et al.*, 2005).

These changes in root growth, according to the old concept of morphogenetic equilibrium between roots and shoots (Vannoordwijk and Dewilligen, 1987), attest that modern short-stemmed genotypes possess a shallower root system than the old long-straw cultivars. Supporting evidence for this hypothesis (Haberle *et al.*, 1995) also found a statistically significant, positive correlation between plant height and rooting depth in wheat cultivars of different geographic origin. Comparative studies on the root growth of tall and short wheat genotypes, however, have clearly disproved the hypothesis of morphogenetic equilibrium (Pommer and Oppitz, 1986; Siddique *et al.*, 1990). While older cultivars and landrace varieties continue to be a source of material for comparisons and assessments with modern varieties, new populations have been developed and are entering into mainstream organic production. Skagit 1109, a new landrace population grown in Washington State (Bread Lab, USA), Wakelyns population (Wakelyns Agroforestry, UK) and Liocharls/Brandex populations (Dottenfelderhof, DE) are all commercially available organic wheat populations in production and under evaluation.

2.4.2 Fertility Management

Inadequate nutrient supply is a major factor contributing to the yield gap between conventional and organic crop production systems. This is largely attributed to a reduced N availability especially during the critical late seed/grain filling phase in organic systems. The exclusion of soluble mineral fertilisers from organic agriculture means that it is reliant on biological processes for supplying nutrients whereby an active soil microbial community is vital for effective functioning. For organic management systems to produce grains with good baking quality characteristics there needs to be an interaction between variety choice and the fertility management system both pre-cropping and during the cropping season (Carcea *et al.*, 2006).

Vaisanen and Pihala (1999) studied the effect of pre-crop and fertilisation on baking quality of organic spring wheat on 20 organic farms in southern Finland. Three spring wheat varieties were grown after (1) peas/cereals or monocrop peas, (2) clover/grass leys (1- or 2-years), (3) grass/clover leys + manure (low clover seed percentage) or (4) cereals + manure. Soil structure had a greater effect than previous crop in a very wet year. However, the best soil structure and strongest wheat crops were found after clover leys and poorest after peas. The most important factor for flour baking quality was cultivar.

Loges *et al.* (1997) identified that N-uptake in winter wheat crops was highest after 100% clover. Loshakov *et al.* (1997) determined the effect of long-term application of green manure on the yield and grain quality characteristics of winter wheat at Moscow and Berlin. Green manures increased wheat yield in rotations by 8.9-13.6% at the Moscow sites and by 1.7-29.9% at the Berlin ones. The best preceding crops for wheat were clover and grass/clover leys. Increasing the percentage of cereals in the rotations at both sites from 50% to 83% led to reductions in grain quality and wet gluten content in winter wheats.

Nitrogen availability and acquisition is one of the most important factors for plant development (Pandey *et al.*, 2017) and the N contribution from biological N_2 fixation enables organic production to function in the absence of industrial N fertilisers (Carlsson *et al.*, 2009). Whilst most organic certification bodies advocate the use of mixed farm enterprises to ensure manure availability, this is not always economically viable or possible. Therefore the use of pre-crop green manures in a stockless system is vital for quality cereal production (Millington *et al.*, 1990). A study of N fixation in above-ground plant tissue of both red clover (*Trifolium pratense*) and white clover (*Trifolium repens*) (Carlsson and Huss-Danell, 2003) demonstrated that fixation in both crops was significantly ($P<0.001$) correlated with legume dry matter (DM; kg per ha and year). Their valuable estimations of Nfix in legumes (roots not considered) are given by:

$$Nfix = 0.026.DM+7 \text{ for } \textit{T. pratense}$$

$$Nfix = 0.031.DM+24 \text{ for } \textit{T. repens}$$

This typically gives values in the range of 373 kg N ha^{-1} year^{-1} for above-ground plant tissue for red clover and 545 kg N ha^{-1} year^{-1} for white clover. Comparative UK studies by Stopes *et al*. (1996) provide similar Nfix figures of 371 kg N ha^{-1} year^{-1} for red clover after 13 months and 450 kg N ha^{-1} after 18 months. White clover Nfix was 432 kg N ha^{-1} after 18 months.

Time of cutting of the green cover crop was also found to affect crude protein yield in red clover (Bender and Tamm, 2018). Improvements in protein yields produced improvements in quality in subsequent cereal crops. Cereal crops with the highest yields occurred more frequently from clover cut for the first time at anthesis. Jamriska (1988) also established the date of the first cut as having the greatest influence on dry matter yield and subsequent cereal crop yields.

Given the importance to successful wheat production, for both yield and quality characteristics, the availability of biological N_2 from pre-crop clover leys combined with agronomic practices should aim to minimise any losses during incorporation and maximise their availability during the following wheat growing season.

Leaching losses of the total N accumulated by red clover above-ground were recorded in winter and spring wheats grown after 1 and 2 year clover leys (Stopes *et al.*, 1991). Losses of one third by leaching (measured using porous ceramic cup samplers) following cultivation of the 1-year green manure in September prior to establishing a winter wheat crop was recorded. Delaying cultivation until the spring substantially reduced leaching due to uncultivated soil over winter.

The mineralisation of C and N at a range of low temperatures, characteristic of autumn green manure incorporation and its decomposition (Lahti and Kuikman, 2003), demonstrated the susceptibility of crop derived N to losses and how the pre-crop value of a green manuring crop can be manipulated by proper timing of incorporation. N supply from soil to spring crops was higher in late and delayed autumn incorporation than for early or spring incorporation due to lower temperature which had the potential to reduce the susceptibility of mineral N to leaching, yielding more N to subsequent crops.

2.4.3 Weed management

The ability to restrict and reduce the growth and multiplication of weeds in organic crop production systems, where the use of herbicides is not permitted, requires a combination of mechanical and cultural agronomic practices over time. Any approach must be seen in this context from a system perspective. Crop: weed interactions usually manifest themselves more slowly, so that weed management should be tackled in an extended time domain, needing deep integration with the other cultural practices, aiming to optimise the whole cropping system rather than annual in-crop weed control (Bàrberi, 2002).

Pre-crop: Green manure crops which have a broad leaf top covering (smother crops) and which offer multiple harvests or require multiple cuts during the growing season have the ability to both shade out and compete aggressively with weeds (Dorn *et al.*, 2015), but also to mechanically destroy growing weeds at the point of harvesting during the growing season. Additionally, the mulch layer of cover crop residues on the soil surface was shown to reduce weed germination, weed emergence and establishment (Peachey *et al.*, 2004).

In crop: Weeds compete well in organic wheat crops and reduce the specific weight of wheat grains when they are allowed to proliferate (Stopes *et al.*, 1991). Their control in organic winter wheats, which offer the higher yield, becomes imperative as they will adversely affect the quality characteristics when not kept in check. A clean top soil, through good management of the pre-crop, must be maintained during the following growing season. Reductions in weed numbers and biomass can be higher from post-emergence harrowing than pre-emergence weeding in spring wheat (Stenerud *et al.*, 2015). Under sowing wheat crops with white clover in the same study further reduced weed emergence in wheat crops.

Timing of sowing of winter wheats in chemically dependent systems has become earlier in the autumn, indeed into late summer in the UK, in order to promote rapid growth and leaf development before the winter. This has allowed weed seeds to germinate concurrently, which are then controlled with a late application residual herbicide, not the growing wheat crop. Under organic systems winter wheats are generally sown later in order to reduce the growth and competition of potential weed species. This delayed sowing date also aids the retention of soil nutrients as discussed earlier but leads to a lower potential grain yield (Green and Ivins, 1985).

Cultivar competitiveness is another important consideration when implementing an integrated organic management system. This competitive ability can be defined as 'suppressive', with the ability for taller crops as competitive cultivars to outgrow, overshadow, reduce the PAR and thus limit the growth of competing weeds (Andrew *et al.*, 2015), or a tolerance ability. Variations between varieties in their canopy architecture during the growing season can influence this suppressive success which is further influenced by soil nutrient availability and leaf health/longevity.

A strongly suppressive cultivar will reduce seed production in weed species, whereas a tolerant one will maintain yield under weed pressure but not indefinitely. The need, therefore, for taller wheat varieties promotes both grain quality characteristics and weed suppression, as a weapon in the organic producer's armoury (Cosser *et al.*, 1997). This cultural tool in combination with the management practices discussed and mechanical tine weeding provide a broad spectrum of control and suppression techniques which can effectively reduce the impact of weeds in the crop to a negligible level.

Mechanical weed control (MWC) by sprung tine harrows is widely practised throughout the organic industry in field grown crops. Soils should be dry for MWC to be effective as wet soils clump and ball onto the tine ends reducing their ability to up-root annual weeds or loosen the top soil. This affects the time available to undertake such weeding and the choice of cereal crop in the rotation when weed management becomes an important decision. For soils with a light weed burden, winter wheats may still be sown. Late sowing in the autumn, for soil nutrient retention, results in soils that are unlikely to be dry enough to weed harrow until February or March the following year. Pre-emergence weeding, to up-root and bury the most competitive annual weeds, is therefore unlikely and reliance is on a post-emergence treatment. This can and should be aggressive (Brandsæter *et al.*, 2012), as ineffectual MWC may stimulate further weed emergence or allow recovery from damage (Bàrberi, 2002). The wheat plants will be well enough established at this growth stage to withstand hard MWC. A second post-emergence weeding may be considered if the weather in early spring allows. Melander *et al.* (2005) found the first pass in winter wheat to stimulate the emergence of weed seedlings which were then destroyed in the second pass. The effect of the speed of the MWC pass (Cirujeda *et al.*, 2003) has been found to be more pronounced in heavier soils with the highest speed of 8 km h^{-1} being the most effective.

Spring sown wheat crops allow a much better weeding programme, as drier soils at sowing and early crop development allow for multiple strikes into dry soils in both the pre-emergence and post-emergence development stages of annual and perennial weeds. A combination of pre- and post-emergence weed harrowing can reduce weed density by 59% and weed biomass by 67% compared to untreated spring wheat (Stenerud *et al.*, 2015). Spring wheat yields can increase by 6.2% with pre-emergence harrowing and by 4% with post –emergence. However, when combined they can give yield increases of 10% (Brandsæter *et al.*, 2012). Harrowing treatments, Gilbert *et al.* (2009) concluded, had no effect on above-ground biomass N uptake of wheat and no effect on yield. The study suggested an immobilisation of soil N, through increased aeration and soil microbial activity.

Management of allelopathy, defined as "Any process that involves secondary metabolites produced by plants, algae, bacteria, and fungi that influence the growth and development of biological systems" (Bàrberi, 2002), is being considered with increasing interest (Schulz, 2004), as the idea of exploiting plants within the organic system that release allelopathic compounds as natural herbicides is very appealing (Inderjit, 2009). Allelochemicals identified in the phenolics and terpenoids from different wheat accessions have been shown to inhibit the growth of the weed species *Bromus japonicus, Chenopodium album* (Singh *et al.*, 2003) and *Lolium rigidum* (Weih *et al.*, 2008), in the latter where higher levels of phenolics in the shoots of certain wheat accessions inhibited growth. A clear potential exists for the transfer of these characteristics into wheat breeding programmes.

2.5 Grain quality and functionality

Wheat production systems are generally based on the premise of producing the highest yield achievable for the minimum costs. Therefore growing for quality first is at odds with the breeding programmes and production systems currently being employed if the achievable quality comes with a yield penalty, particularly in organic systems where the quality potential of the variety cannot be achieved with additional inorganic fertilisers, such as foliar N applications, later in the growing season.

2.5.1 Grain specific weights

The greatest requirement for wheat within the milling industry is for the production of white flour. High extraction rates of white flour are critical to the economic efficiency of all milling businesses that offer white flour. Semi-dwarf genes have made a major contribution to grain yield gains in wheat during recent decades. These genes have frequently been reported to increase grain number (ca. 40% in grains m^{-2}) but decrease grain size (ca. 20% in specific weight) (Slafer and Miralles, 1993).

Increasing sink size (number of grains m^{-2}) in order to achieve greater grain yield of wheat was studied in three bread wheat cultivars from three different eras (1920s, 1940s and 1980s) (Slafer and Andrade, 1993). The modern, higher yielding cultivar had a lower individual grain weight than the old, low yielding cultivar. The number of fertile florets at anthesis was greater in the modern cultivar than in the other two. The number of fertile florets plus the ability of the cultivar to set grains in those florets were responsible for the differences in grain number per ear between cultivars.

Milling grists (blends of varieties or grains with varying quality specifications) attempt to balance the effect of kernel size during milling on the baking performance of the end flour. Larger grains allow for a higher extraction of flour and for some varieties an increase in protein quality (Borghi *et al.*, 1998), whilst for other cultivars a reduction in grain size increased the quality characteristics (Gaines *et al.*, 1997). The effect of climate, soil nutrient availability and variety can all influence wheat quality in any given season (Borghi *et al.*, 1995).

2.5.2 Grain quality characteristics

A comparison of old and modern cultivars, made in order to review the breeding progress in small grain cereals (Feil, 1992a), identified an initial decline in quality within UK wheat crops with the introduction of the semi-dwarf gene. The early increase in grain yield of winter wheat seems to have been much greater in the UK (Austin *et al.*, 1989) than in Scandinavia (Ledent and Stoy, 1985; Ledent and Stoy, 1988) and Germany (Karpensteinmachan and Scheffer, 1989), where it is suggested that breeding for high baking quality was probably responsible for the relatively small increases in grain yield in Germany over the same period. Recent wheat varieties from all European breeders have increased both yield and quality traits (Osman *et al.*, 2016), but selection for quality slows the progress in grain yield (Mesdag, 1985).

The continuity of traditional baking practices in post-war Germany and the long held bread heritage maintained throughout the decades of the last century have greatly influenced the wheat breeding programmes (Feil, 1992b), in stark contrast to the UK where post-war Britain focussed on yield and quality to satisfy the Chorleywood Baking Process and the transfer from small scale bakeries to the large scale commercial bakeries. This transition has been mirrored by the decline in that period of the traditional artisan baker of wholemeal breads and the rise of industrial processed white flour loaves. This has built a production platform of UK cereals with a reliance on a limited numbers of pure lines to deliver a country's entire needs.

A study of old and new cultivars in Finland (Slafer and Peltonen-Sainio, 2001) also identified an increase in yield over time, an increase in N fertiliser use and an increase in baking quality with increased N fertiliser application rates.

The effect of breeding for yield and the changes in wheat characteristics highlight the conflict between quality and yield. The direct relationship between increasing sink and quality characteristics of bread wheat was assessed (Borghi *et al.*, 1986) by modification of the sink/source relationship and its influence on grain yield and grain protein content. Artificial manipulation of sink and source was carried out in several bread wheat varieties in order to study the variations in the pattern of storage product accumulation in the grain. A halving of the number of spikes led to a 14% increase in the size of the kernels, while the protein content per spike showed a 20% drop in comparison to the control. Reduction of sink size affected nitrogen accumulation to a lesser extent than carbohydrate storage in the grain.

Reduction of sink induced remarkable increases in the protein fractions: gliadins +59%, glutenins +44% (both of which form the basis for wheat flour strength and stretch during bulk fermentation of the dough), insoluble residue +30%, non-protein nitrogen +28%, albumins and globulins + 16%, all of which greatly influence the performance and outcome of wheat flour breads.

This demonstrates a popular misconception amongst new bakers outside the scientific community where grain protein percentage is assessed as the de facto indicator of grain protein fraction (gluten) quality. The study from Borghi *et al.* (1986) demonstrates the relationship between kernel size and protein %, and importantly how this decrease in protein % does not at all reflect the quality of the protein fractions. Indeed the reverse was observed where a reduced sink and lower yield improved the baking quality and improved the protein fractions.

Added N fertiliser applications to boost grain protein do not necessarily lead to an improvement in the baking quality characteristics of that grain. A review of gluten properties

(Dellavalle *et al.*, 2006) raises the possibility of compensating for low protein content by altering protein quality. Although this hypothesis was not made with a view to organic wheat production, it is nonetheless a very pertinent observation that within the organic bread sector the underlying protein quality characteristics of organically produced wheat appear to perform better for artisan bakers than those from wheat grown under conventional methods. Also, conversely, organic bread wheat with its lower protein content is not suited to the highly mechanised commercial Chorleywood bread-making process.

2.5.3 Wheat baking Functionality and Processing

Wheat is unique in its ability to retain gas in a dough system and thus to generate highly expanded textures characteristic of bakery goods. There are, as already identified, a number of flour quality attributes which need to be considered when assessing the suitability of a given grain sample for a given process or product, but the majority of the variation found in bread-making ability is as a result of differences in the protein fraction of the wheat; in particular, the portion of the protein (gliadins and glutenins) which forms the gluten on the addition of water is of critical importance (Macritchie, 2016). The foremost determinants of wheat quality are therefore the endosperm texture (grain hardness), protein content and gluten strength. The varieties of wheat will differ from region to region and from season to season, as highlighted, in their protein quality and thus their ability to perform in the bread making process (Bhatta *et al.*, 2017). In long-flow experimental milling (Souza *et al.*, 2008) that closely approximates commercial flour yields, variety and seasonal variation affected flour yields.

Flour milling in its basic form is a process of reducing the particle size of the starting grain (wheat) to that required for subsequent (baking) processing. For white flour production the correct degree of particulation is achieved by separating the wheat grain exterior (the bran) from the interior (the endosperm). The fracturing of the endosperm to produce the white flour is either roller milled or stone milled. The reduction milling of grains by roller milling (successive passes of grains through steel rollers) has pressures on the fractured starch which are different from the single pass process of stone milling (Osborne, 2007). Hard or soft grains will therefore be fractured differently, creating different baking characteristics in the resulting white flours (Choy *et al.*, 2015). Baking characteristics are further affected by the absence or presence of bran, as the gas retention properties of doughs are compromised by the inclusion of bran. UK roller milling is designed for an 80% extraction of white flour from wheat grain, whereas most traditional stone mills are doing well to extract 50%. More modern stone milling operations are able to improve the extraction to around 70%. The quality characteristics of the protein are therefore of increased importance in these traditional milling settings, as well as the grain kernel size for high extraction. Bran content and its impact in the bread making process increases with lower white flour extraction (Gaines *et al.*, 1997). Grain hardness and water absorption in flour also increase with increasing kernel size (Yinian *et al.*, 2008), both of which will improve the flour quality from a stone milling process.

Grain hardness (Ha) is a measure of endosperm texture and in wheat is possibly the single most important and defining quality characteristic, as it helps determine wheat classification and affects milling, baking and end use quality (Pasha *et al.*, 2010). Ha can be measured in a number of ways, but the most extensively used for determining texture are NIR hardness (Near-infrared reflectance), SKCS (Single Kernel Characterisation System) a one crush resistance test, and a pearling index (Fox *et al.*, 2007). Starch granules associated with friabilin, an endosperm specific protein, formed by the interaction of different polypeptides (primarily puroindolines), are directly related to grain softness (Pasha *et al.*, 2010). Fox *et al.* (2007) identified a significant (p<0.05) genetic effect on grain hardness but also environmental effects, possibly linked to the effect of protein on hardness with increasing protein resulting in harder grain. Hard grain wheat produces a higher yield of flour, flour with more damaged starch granules, flour with a higher water absorption and flour with a higher lipid content, all of which are targeted at the production of leavened bread products.

Selecting varieties for baking quality is further complicated now by the fact that the main baking quality parameters (e.g. protein content, dough strength, loaf volume) are directly or indirectly correlated with grain nitrogen content, which in turn is negatively correlated with yield. Conventional farmers look for higher yielding varieties. Therefore conventional breeders aim at increasing yield while maintaining the level of baking quality traits similar to their standard modern variety. In Austria, where organic production has a high value in agriculture, the use of ‘value for cultivation and use’ (VCU) assessments of cultivars from conventional and biodynamic breeders has led to the successful development of breeding lines specifically for organic agriculture to meet the quality needs of their winter cereals (Loschenberger *et al.*, 2008).

2.5.4 Grain Nutrient Density

Significant genetic variation exists in minerals, vitamins, antioxidants and other compounds linked to human nutrition and health in many crop species, although environment and management systems also have an effect. The gradual transition from landrace to elite cultivars over the last two centuries to the rapid development and use of pure line varieties in the latter stages of the last century (Newton *et al.*, 2010) coincides with a deterioration of grain mineral density and general baking quality within the UK grain harvest (Fan *et al.*, 2008).

Modern, high yielding wheat cultivars tend to have lower phytonutrient levels, protein content/gluten quality and mineral concentrations (Fe and Zn) than traditional varieties (Murphy *et al.*, 2007). This is compounded by the fact that many phytochemicals and minerals such as Fe and Zn are not evenly distributed in the grain of cereals but are highest in the bran, leading to loss of grain nutrients in the production of white flour.

Anti-nutritional factors: Cereals and legumes are rich in minerals, but the bioavailability of these minerals is usually low due to the presence of anti-nutritional factors, such as phytate (a naturally occurring organic complex and a simple ringed carbohydrate with a phosphate group attached to each carbon), trypsin inhibitors, and tannin. Phytate in particular has a pronounced effect on nutrient availability (Guttieri *et al.*, 2006), particularly as it is stored in the aleurone layer of the grain between the pericarp (bran layers) and the endosperm.

Studies have proved that phytate is an anti-nutritional factor because of its strong metal-binding or chelation ability (Guo *et al.*, 2015). Its affinity to essential micronutrients, such as Zn, Ca, Fe, Mg and Mn, forms insoluble composites that significantly reduce physiological utilisation of them under intestinal conditions (Kratzer, 1953; Rodriguez-Ramiro *et al.*, 2017). Given the health problems associated with Fe and Zn deficiencies, studies using the phytic acid to zinc molar ratio (Gargari *et al.*, 2007) and phytic acid to Fe molar ratio (Marguerite *et al.*, 2006) are used to assess the bio-availability of these elements in the diet in relation to the phytate content in flour, breads and other foods. Phytate is a significant cause of deficiency in Ca, Zn, and Fe, particularly among people whose staple food is wheat-based. The molar ratios of phytate to zinc or iron are deemed to be an effective measurement in cereals for the bio-availability of essential micronutrients from cereals.

Moreover, phytate could impair some enzymatic activity, such as protease, amylase and trypsin, acidic phosphatase and tyrosinase (Urbano *et al.*, 2000), and consequently influences the utilisation of protein, starch and fatty acids. Undoubtedly, this anti-nutritional factor is a significant problem (Guo *et al.*, 2015).

Reduction of anti-nutritional factors: Reductions in the influence of these factors is necessary to derive the full nutritive value of cereals and grains (Dwivedi *et al.*, 2015), otherwise the drive to switch consumers from white flour products, with a low nutritional value (Fardet *et al.*, 2006), to wholemeal ones will be achieved without an actual increase in nutritional density. Improvements in grain quality and nutrient density from organic breeding programmes and cereal production systems should therefore be assessed in conjunction with their milling and baking performance for them to be of benefit to human health (Levrat-Verny *et al.*, 1999).

Levels of phytate in the aleurone layer have been found to vary between hard and soft grains (Greffeuille *et al.*, 2005). Hard grains were found to store more phytate in the aleurone layer than soft wheats, and the phytic acid in the subsequent flour streams was also higher from hard wheats. As the separation of white flour occurs at the aleurone layer interface, and the hard grains' resistance to milling gave a more complete destruction of the bran and aleurone layer, then the choice of grain pre-milling can influence the subsequent phytic acid content of the flour. The ash content of the flour is a key indicator of bran content and therefore phytic acid levels in any given flour sample. High flour ash is a sign of higher aleurone and bran fragments. When the phytic acid levels and ash content were compared between wild wheats and modern cultivars (Guttieri *et al.*, 2006), the wild types had a higher ash content in the flours but had significantly lower phytic acid levels. The lower phytic acid genotypes demonstrated a significant redistribution of minerals from the bran to the endosperm, suggesting this redistribution as the most likely cause for the increase in the ash content rather than higher levels of bran in the flour.

Improvements within the milling process have been identified to more effectively remove the aleurone layer from the bran (Guo *et al.*, 2015) and therefore reduce phytic acid levels in flour. However, this can only improve the content within white flour and does not adequately address the need to improve the nutritional density through greater use of wholemeal flour.

To improve the nutritional density of available minerals of wholemeal bread products a manipulation of phytic acid during baking would assist in reducing the anti-nutritional impact of phytate in wholemeal flour (Rodriguez-Ramiro *et al.*, 2017). Use of the phytase-active bakers' yeast, *S. cerevisiae* L1.12, was found to effectively reduce the levels of phytic acid (IP6) in wholemeal dough during a short fermentation time (Izzreen *et al.*, 2017).

Optimum conditions for phytase activity are a pH range of 5.0-5.5 and a temperature range from 50 to 55^{o}C. Phytic acid was significantly reduced in low pH doughs (pH 4.3-4.6) adjusted with citric or lactic acid (Fretzdorff and Brummer, 1992), and completely hydrolysed in wholemeal flour doughs at pH 4.5 and 30^{o}C. For traditional, long fermentation breads the most effective method to reduce the phytic acid content is to use a sourdough starter. Fermentation tests on other cereals and legumes (Dwivedi *et al.*, 2015) reduced phytic acid levels, tannin and trypsin inhibitor levels. Further research into the influence of fermentation (Buddrick *et al.*, 2014) identified time as the most significant factor in reducing phytate levels in wholemeal flour and fermentation temperature less so. Levels were significantly lower after a 7 hour process compared with 3 hours. So modern intensively processed short-duration processing, e.g. the Chorleywood Process, is likely to lead to high phytate levels with consequent negative effects on human health.

All wholemeal flours, with high levels of both beneficial and anti-nutritional minerals (Levrat-Verny *et al.*, 1999), can be used to improve the bioavailability of minerals from consuming whole grain products in the diet where those flours are baked using slow fermentation sourdough preparations.

2.5.5 Hypothesis

From the absence of evidence for the potential of growing milling quality organic cereals in the north east of England, the first hypothesis this study aims to test is that a long term organic soil fertility management strategy does exist to optimise nutrient availability sufficient for growing high protein bread wheat in the north east of England. The second is that milling quality wheat can be grown in the NE of England from both winter and spring tall straw wheat varieties currently available from European wheat breeding programmes not presently on the UK national recommended variety list. This would test the validity of the VCU parameters used in wheat experiments on the continent and hitherto untested in organic or conventional wheat variety evaluations in the UK, where only the modern varieties from the recommended list are assessed for British agriculture.

CHAPTER 3: Effect of clover and variety choice on yield and baking quality of organic wheat

3.1 Introduction

Previous studies have shown that yields and protein content of wheat produced under organic standards are often between 20 and 40% lower than those achieved in conventional farming systems (Taylor and Cormack, 2002). This is thought to be at least partially due to insufficient nitrogen supply during later growth stages (Dubois and Fossati, 1981; Miceli *et al.*, 1992; Ayoub *et al.*, 1994; Sowers *et al.*, 1994; Pechaneck *et al.*, 1997; Mittler, 2000; Mäder *et al.*, 2002). Nitrogen contribution from biological N_2 fixation can improve the N supply in organic systems, where the use of industrial N fertilisers is not permitted (Huss-Danell *et al.*, 2007). Perennial forages are widespread in temperate areas, where much of the agriculture is based on livestock production. However, due to the symbiosis with N_2-fixing rhizobia, perennial forage legumes also have great potential for increasing N availability in stockless organic farming systems. This alternative approach to manage leguminous green manure crops by repeatedly cutting and mulching them directly in the field has been shown to improve N availability from above-ground plant parts (Millington *et al.*, 1990). However, this green manuring practice has been shown to incur gaseous N losses of up to 40% when the cut clover is left on the field to decompose during the summer months (Aulakh *et al.*, 1982). Gaseous N losses from clover swards after the removal of a hay crop range from 1.3 to 4.7 kg N ha^{-1}, whereas this can be 8 times higher from cut and mulched clover in summer months with high air temperatures and after rainfall which increases soil moisture (Aulakh *et al.*, 1983). The use of clovers as green manure crops can combine N supply and weed suppression within an organic arable rotation (Koehler-Cole *et al.*, 2017). The inability to control certain diseases (especially *Septoria* spp.) along with higher levels of competition from weeds may also contribute to lower grain yields and processing quality in organic wheat production systems (Mittler, 2000). It has been suggested by Sprent (1990) that it is root exudate and sloughed off cells that are the source of nutrients for rhizobia during multiplication. The flavone luteolin (5,7,3,4-tetrahydroxyflavone) has been identified as a component of plant cell exudates responsible for the switching on of the nodulation genes during this period of multiplication (Peters *et al.*, 1986). This helps to confirm that the trigger for nodulation is not host specific and can influence management decisions in field preparations to benefit green legume crop establishment with improved rhizobia activity. Although a positive correlation between straw length and protein content has long been recognised in continental Europe (e.g. most of the A and E classification high baking quality/protein wheat varieties in the German variety list are longer straw varieties), the higher lodging risk from longer straw varieties under UK conditions has resulted in the disappearance of these varieties from the UK national listing (Mittler, 2000; Lochow Petkus, 2003; CPB Twyford, 2003). Initial results from the Nafferton Factorial Systems Comparison (NFSC) Experiments indicated that lodging risk is significantly lower in organic production systems (Schmidt *et al.*, 2000; Lueck *et al.*, 2006). It was therefore decided to re-evaluate a range of longer-straw varieties from European breeding programmes, which had not been included previously in UK variety trials (Carver and Taylor, 2002; Taylor *et al.*, 2006) under commercial organic farming conditions in Northern Britain.

The **Aims and Objectives** of the experiment were therefore to evaluate the effects of:

(i) Inoculation of clover seed (grown as clover ley prior to wheat crops) with specific *Rhizobium* seed inocula and

(ii) Applications of high C:N ratio green waste based compost to cut clover green manure to increase N availability for the following wheat crop.

3.2 Materials and methods

3.2.1 Trial Site

The site at Gilchesters Organic Farm, Northumberland UK (55º2'28.89N 1º53'28.73W) comprised 24.3 ha of a 60 ha block of arable land, having been in continuous arable production for 15 years up to the harvest of 2002. The 24.3 ha comprised four fields (A, B, C and D) of roughly 6 ha each, managed as one complete arable block with the same conventional rotation across all 4 fields, of winter feed wheat, winter milling wheat, barley and oilseed rape (Appendix 2). The crops were managed for high yields with ADAS recommended levels of N, P and K. All straw except for oilseed rape was removed. No grass or clover leys had been sown in the 15 years prior to the trial. The complete absence of manures, composts or green organic matter from the previous farming fertility management plan combined with the heavy machinery employed in the arable farming activities left the soils compacted and with minimal aerobic decomposition activity, identified by a low colloidal organic matter percentage and high iron levels in the soil analysis (Table 3.1). Trial site soil samples (top soil) were collected on 15th January 2003 and sent to Glenside Fertility Farming Systems, Throsk, Stirling, for analysis.

The site along with the rest of the farm entered into an organic conversion scheme in September 2002. The trial site was registered as "in-conversion" until September 2004, after which time it became fully certified for organic production.

3.2.2 Seedbed preparation

To stimulate rhizobia activity and numbers prior to the establishment of clover, the 24 ha site was aerated by subsoiling (Kverneland CTC cultivator) post-harvest (September 2002) to a depth of 30-35 cm. The barley stubble across the whole site was left over winter in order to increase root cell exudates through aerobic decomposition of the exposed top soil.

3.2.3 Experimental design and agronomic management

The site was selected in order to study the impact of clover management on N fixation during the 2 year conversion period, a necessary transition for the land to qualify for certification in organic production. Therefore, a factorial experimental design with **(i)** *Rhizobium* inoculation (+/-), **(ii)** high C:N ratio green waste compost amendment to clover swards (+/-) and **(iii)** wheat variety (4 spring and 3 winter wheat varieties) as factors was proposed. This resulted in a 2x2x7x 4 factorial experiment with 112 plots (Fig. 3.1) to assess the impact of *Rhizobium* inoculum on the establishment and *Rhizobium* activity of the clover crop in year 1 (2003) and of green waste compost (GWC) amendments after years 1 and 2 (2003-04). The impact of both these treatments on the production and retention of N was assessed in the subsequent wheat variety trials (2005), which would be the first organic wheat crop post conversion.

The experiment was established on 26 March 2003, with each 6 ha field acting as a replicate. A pure red clover (*Trifolium pratense*) mix of 60% late flowering diploid (cultivar Britta) and 40% early tetraploid (cultivar Rotra) organic seed was purchased from McCreath Simpson & Prentice, Berwick-upon-Tweed. Each field was subdivided into 2 main plots each of 3 ha in which pure red-clover swards were established using either seed inoculated with a commercial *Rhizobium* (Becker-Underwood, Saskatoon, Canada) preparation or untreated seed. The site was prepared with HE-VE discs, the clover sown by direct drill (Vaderstad Rapid A400S) at a seed rate of 9.8 kg ha^{-1} and rolled. All the untreated plots were sown first.

3.2.4 Clover seed treatment

2 kg (5 units of 400 g) of *Rhizobium* inoculum in the form of a high-stick® paste were applied to 120 kg of clover seed, once all the untreated blocks had been sown. The treated clover plots were also sown on 26 March 2003. The *Rhizobium* is one of the strains isolated at Rothamsted Research Centre from those naturally occurring in UK soils, and was multiplied and cultured by Microbio Group Ltd (a wholly owned subsidiary of Becker Underwood). The strain and multiplication process comply with the UK Register of Organic Farming Standards protocols.

Table 3.1 Trial site soil analysis for fields, January 2003

		Field			
		A	**B**	**C**	**D**
Cation Exchange Capacity		16.99	18.12	16.78	16.95
pH		6	6.3	6.7	6.9
Organic matter %		3.3	3.4	5	4.5
N	kg ha^{-1}	92	93	112	106
S	ppm	10	9	12	16
P	kg ha^{-1}	97	147	164	196
Ca: Mg	ratio	64.14	66.16	72.15	63.21
Ca	kg ha^{-1}	4879	5396	5505	4871
Mg	kg ha^{-1}	628	776	679	948
K	kg ha^{-1}	133	175	250	557
Na	kg ha^{-1}	75	83	72	56
B	ppm	0.5	0.53	0.8	1.08
Fe	ppm	533	665	722	908
Mn	ppm	64	83	61	87
Cu	ppm	1.7	2	1.9	1.3
Zn	ppm	7.4	8	7.4	7.2
Mo	ppm	1.36	1.84	1.6	1.72

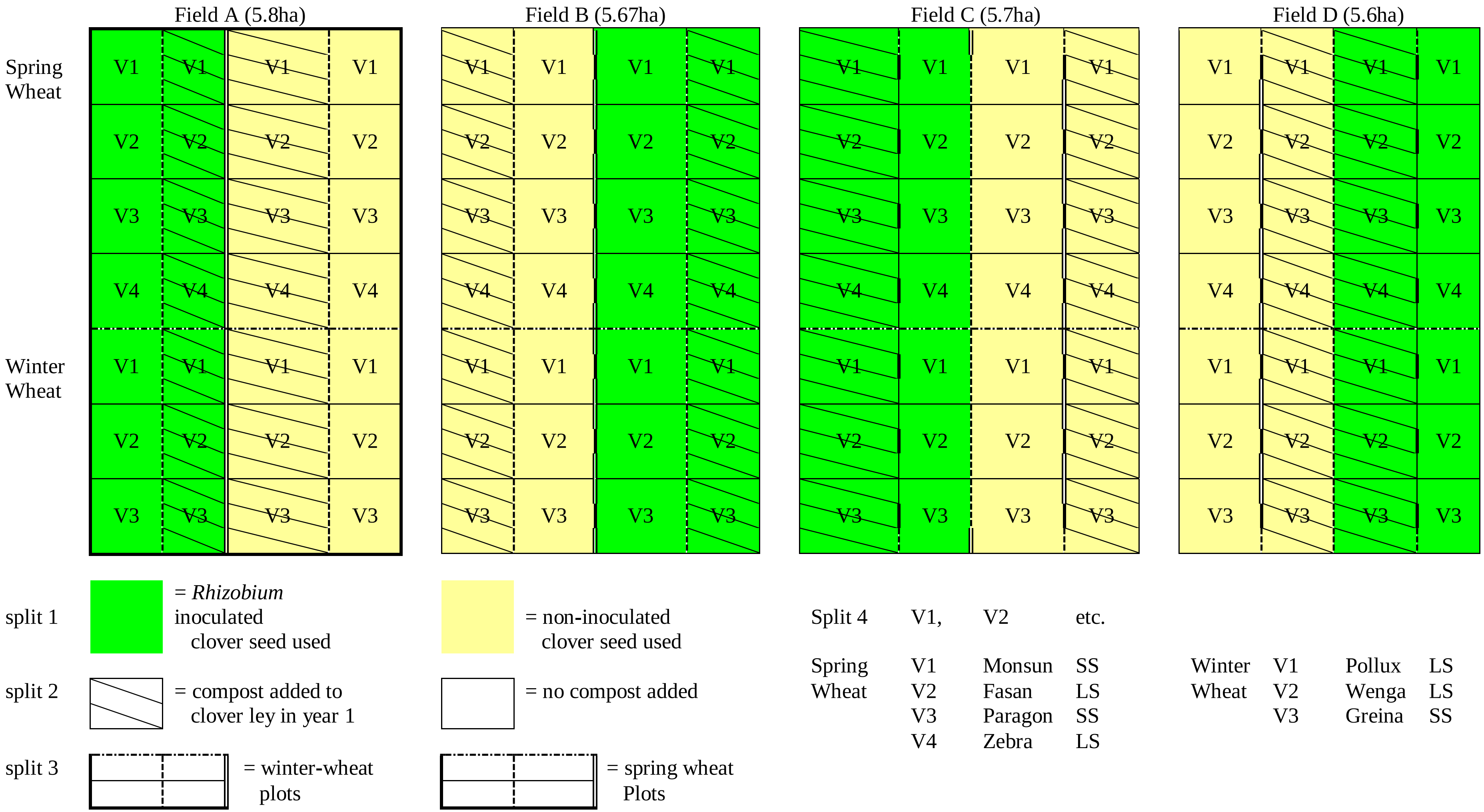


Fig. 3.1 Distribution of main plots, sub-plots and sub-sub-plots of each replicate field (For layout see Appendix 2: trial site map)

3.2.5 Clover crop management

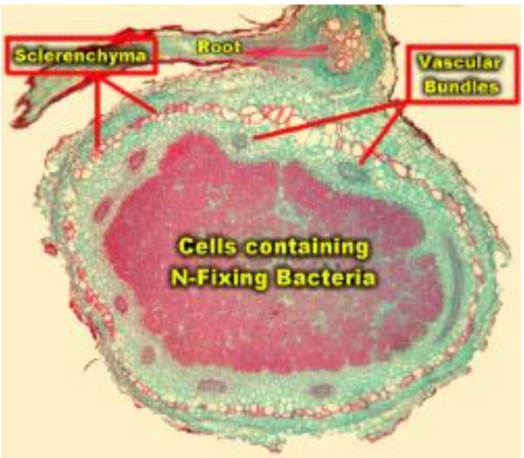
Each main plot was then subdivided into 2 sub-plots of 1.4 to 1.7 ha, one of which was to receive no inputs and the other which would receive 15 t ha^{-1} of a high C:N green-waste compost (COMVERT, Morpeth, UK composted from county council green waste collection). All plots were topped at the end of May/beginning June and again on 28-29th August 2003. The green-waste compost (GWC) was applied to each half sub-plot immediately after each cutting of the clover. Compost composition was analysed at NRM Laboratories (Natural Resource Management Ltd, Braziers Lane, Bracknell, Berks) (Table 3.2). The same procedure for topping and GWC application was repeated in 2004.

Table 3.2 Green Waste Compost analytical results on dry matter basis

	Value	**units**
Dry Matter	40	%
Conductivity 1:6	812	µS cm
Total Nitrogen	1.54	% w/w
Total Carbon	20.3	% w/w
C:N ratio	13:1	
Nitrate Nitrogen	159	mg kg^{-1}
Ammonium Nitrogen	126	mg kg^{-1}
Total Phosphorus (P)	2583	mg kg^{-1}
Total Potassium (K)	5850	mg kg^{-1}
Total Copper (Cu)	23.1	mg kg^{-1}
Total Zinc (Zn)	143	mg kg^{-1}
pH	7.8	

3.2.6 Clover and Rhizobium sampling techniques

Each of the 112 plots and sub-plots was assessed above-ground for clover plant numbers (to determine % establishment) and below ground for *Rhizobium* development. Assessments were made and samples taken during each of the growing seasons, in 2003 (Year 1) and in 2004 (Year 2). In Year 1 the first samples were taken between 15 and 25 May 2003, prior to the first clover cutting and the first GWC application. In year 2 sampling was done after cutting and GWC applications. Due to the scale of the site and the size of each plot and sub-plot, the location of sampling stations was determined by a combination of random and stratified sampling techniques. The stratified technique required calculating the length of the diagonal of each plot and dividing by 6. This gave 5 sampling station locations per plot along the diagonal of each plot excluding the margins. The randomisation technique was to select the distance of the sample stations from 1 to 5 m off the diagonal by a simple roll of a dice and then alternately left or right of the diagonal. Sample stations measured 400 cm^2, recording the number of clover plants within the quadrat to determine the level of establishment (plants per m^2). For *Rhizobium* nodule assessments, the root system from one plant, chosen at random from within each sample station was laid out over a grid of 12 x 16 cm^2 squares and analysed for: number of *Rhizobium* nodules, location of nodules on the root, active or inactive nodules and nodule volume/size. Nodule volume was calculated by measuring nodule length, width and height using a standard digital calliper (Oxford Precision OXD-331-2060K) applied to the formula for an oblate ellipsoid, given by $\frac{\pi}{6}.L.W.H$ cubic units (mm^3). Each nodule was then dissected to establish whether it was active or not. Active nodules are recognisable by their mottled pinkish interior when bisected, visible to the naked eye (Fig. 3.2).



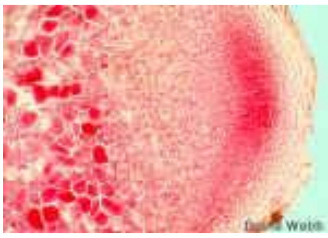


Fig. 3.2. Active *Rhizobium* nodules (photo library images http://www1.biologie.uni-hamburg.de)

From the two clover cultivars sown (Britta and Rotra) two different root systems were observed: one a dominant single tap root with a supporting lateral root system, the other a multiple root system with shorter main tap roots and supporting lateral roots (Fig. 3.3). The German plant breeders of both cultivars confirmed that neither would normally have multiple tap roots, suggesting the occurrence may be due to the lack of rainfall in the winter and spring of 2003. However, the diploid variety Britta was considered the one most inclined to develop in this way under such conditions.

Fig. 3.3 Clover with single tap root (left) and multiple roots (right)

3.2.7 Wheat varieties

On 28th October 2004 three bread-making winter wheat varieties and on 9th March 2005 four bread-making spring wheat varieties were planted/superimposed on the fertility management plots (sub-sub-plots). Treatments were randomised at main plot, sub-plot level (Fig. 3.1). Crops were sown using an Accord 3 m combination drill consisting of a Rabe power-harrow (Rabe Werk, Bad Essen, Germany) and an Accord 24 row precision drill (H. Weiste and Co., Germany). Winter wheats were sown to achieve 400 plants m^{-2} according to thousand grain weight (equivalent rate of 185 kg ha^{-1}) and spring wheats to achieve 350 plants m^{-2} according to thousand grain weight (equivalent rate of 210 kg ha^{-1}.)

The varieties used were: **(a)** 1 short straw winter variety (Greina) from the Swiss national breeding programme, Agroscope/DSP and 2 long-straw winter wheat varieties (Wenga and Pollux) from an organic farming focused breeding programme developed by Peter Kunz for Sativa (Reinau, Switzerland), **(b)** 2 short straw spring wheat varieties bred for UK (Paragon; CPB-Twyford, Cambridge, United Kingdom) and German (Monsun; Lochow Petkus, Bergen, Germany) intensive farming systems and **(c)** 2 long straw varieties bred specifically for high baking quality wheat production in Germany (Fasan, Lochow Petkus) and Norway (Zebra, Swallof-Weibull, Sweden) (photographs: Appendix 3).

Table 3.3 Wheat variety and plot sizes (ha)

	Straw length	Field A	Field B	Field C	Field D
Winter Wheat		2.8 ha	2.99 ha	2.99 ha	2.94 ha
Greina	short straw (ss)	1.22	0.86	1.1	0.9
Wenga	long straw (ls)	0.76	1.03	0.86	1.04
Pollux	ls	0.82	1.1	1.03	1
Spring Wheat		3 ha	1.68 ha	2.71	1.46 ha
Paragon	ss	0.72	0.42	0.68	0.37
Monsun	ss	0.8	0.4	0.68	0.37
Fasan	ls	0.7	0.45	0.68	0.37
Zebra	ls	0.78	0.41	0.67	0.38

(Site plan: Appendix 2)

3.2.8 Wheat assessments

The following crop assessments (Table 3.4) were made during the growing season:

Soil samples were taken across two winter wheat variety (Greina, Wenga) x treatment plots and two spring wheat variety (Paragon, Fasan) x treatment plots, as well as from the remaining clover plots not ploughed out in each replicate field. Samples were removed from three depths: 0-30, 30-60 and 60-90 cm and analysed at Sabanci University, Turkey (QLIF Project partner) for mineral N and C by Dumas Combustion and all other nutrients by ICP-AES.

leaf greenness (relative leaf chlorophyll content) in all varieties across all treatments was recorded by single-photon avalanche diode detection (SPAD) assessed with a Minolta chlorophyll meter model SPAD 502 (Spectrum Technologies Inc., Plainfield, Illinois, USA) with 4 readings taken for each variety/treatment. Each reading had 10 sample points. All readings were taken on 27th June 2005, at GS 55 for the winter wheat varieties and at GS 37 for the spring wheat varieties (Zadoks *et al.*, 1974).

Grain tocopherol concentrations (α-tocopherol, α-tocotrienol, β-tocopherol and β-tocotrienol) were measured for all varieties against all treatments by freeze drying grain in the Food Analysis Department of the Central Food Research Institute (CFRI, Herman Otto Vt 15, Budapest, Hungary). For freeze drying, all grain samples were dried with a freeze dryer at −50 °C under vacuum (1.6 mmHg) with a pressure of 1.1×10^{-2} mbar for 72 h and then ground with a dry grinder to obtain fine powdered flour. The crude extracts were stored at −20 °C during analysis.

Crop yields were assessed by combining a 320 m^2 section from each treatment using a plot-combine harvester (CLAAS Dominator 38, Germany). Grain fresh weights were determined by weighing grain harvested in each plot immediately after harvest. A sample of harvested grain was dried at 80 °C for two days using a drying oven (Genlab Ltd, Widnes,UK) and grain yields are presented at 15% moisture content. Protein analyses were carried out by the wheat breeding/seed production company Lochow Petkus GmbH (Bergen, Germany) using standard protocols (ICC Standard No. 159; ICC 2006). Grain hardness was also determined by them with a Single Kernel Characterisation System (SKCS), a one crush resistance test.

Table 3.4 Key winter and spring wheat trial assessment dates

	Winter Wheat trial	**Spring Wheat trial**
Soil Analysis	19.01.2005	19.01.2005
Weeding	4.04.2005	25.05.2005
Weeding		3.06.2005
Chlorophyll SPAD	27.06.2005	27.06.2005
Harvest	17.08.2005	7.9.2005

(Photographs: Appendix 3)

3.2.9 Statistical analysis

For the 2003 and 2004 clover trial analyses of variance were derived from linear mixed-effects models (Pinheiro, 2000) from data with a negative binomial distribution (non-symmetrical – due to the clumped nature of the samples recorded). Fixed effects were season, clover seed treatment, and fertility management, with fields as random effect, given the nested structure of the trial (Crawley, 2007), and applied where necessary. Regression analysis and extrapolation of the clover trial data was derived from General Linear Hypothesis Tests (GLHT) (Everitt and Hothorn, 2011). For the 2004-05 wheat trial, spring and winter wheat varieties were added as fixed factors. The effects of the fixed factors on chlorophyll content, yield, yield components, protein, hectolitre weight, hardness and vitamin E levels were determined. Analyses of variance were derived from general linear models and linear mixed-effects models. The analyses were carried out in the R statistical environment (R Development Core Team, 2012) and residual normality was assessed using the qqnorm function in R. The combined data for main effects of treatments and varieties were analysed first, and where interaction terms were significant, further analyses were conducted at each level of the interacting factor. Differences between significant main effect and interaction means were determined using Tukey's Honest Significant Difference (HSD) tests, based on mixed-effects models.

3.3 Results

3.3.1 Effect of clover management (Rhizobium seed inoculation and green-waste compost amendments) on clover

The use of *Rhizobium* inoculum improved the establishment of the clover crop, the total rhizobia nodule numbers for individual plants and the nodule volume (Table 3.5). The establishment of clover plants after sowing was significantly higher with the inoculum (190 plants m^{-2}) than without (152.5 plants m^{-2}). The nodules per plant in the year of crop establishment were also significantly higher with the inoculum (149.1 nodules $plant^{-1}$) than without (115.25 nodules $plant^{-1}$). The nodule volume also was significantly higher with the use of inoculum (3.8 mm^3) than for nodules without (2.5 mm^3), which represents a 47% increase in the size of the nodules. Determining the variance components of all factors (Table 3.6) to establish the factor proportions of the experiment shows overall 62.2% of the total main effect factor variance in the data population can be accounted for by the ±inoculum treatment.

Analysis of the clover plots and treatments in the second year showed no significant effect of inoculum or GWC amendment on nodules per plant or nodule volume (Table 3.7). However, more in depth analysis of the nodule population data did identify a correlation of plant density with both nodule numbers per plant and nodule volume (Table 3.8). Nodule volume appeared to decrease with increased numbers of nodules per plant (estimate -0.0037, P=0.0537, Table 3.8) Furthermore, the difference in the negative estimates (-0.0639 and -0.0386) also suggests that nodule volume in the absence of *Rhizobium* decrease more than with *Rhizobium* inoculum with increasing numbers of plants and nodules per m^2.

There was some indication that higher plant densities, from better establishment with more plants per m^2, contributed to more nodules per m^2, but with fewer nodules per plant (Fig.3.4). The best fit curve in Fig.3.4 identified a decrease in nodule numbers per plant with increased numbers of plants per m^2 occurring at a constant rate although the trends were not statistically significant. Specifically, from +rhizobium inoculum with an intercept (average) nodule count per plant of 208 a predicted loss of 7.7 nodules occurs with every additional plant per sample station (400 cm^{-2}), and from -rhizobium inoculum with an intercept (average) nodule count per plant of 139 a predicted loss of 4.0 nodules occurs with every additional plant per sample station (Table 3.9). Using regression models from the general linear model analysis with the data scaled up to m^2 of the determining factor (Table 3.10 also Appendix 4 in full), a clear pattern for optimum clover establishment can be seen (Fig. 3.5 & 3.6). The model suggests that past the optimum clover plant density (±*Rhizobium*), nodules per m^2 will eventually decline due to the law of diminishing returns.

The use of the inoculum results in a much greater colonisation of nodules, seen by the higher intercept value when compared to nodule numbers in the absence of the inoculum. The steepness of the curve also explains the greater decline of nodules per plant with increasing plant density that is associated with the use of inoculum.

Table 3.5 Main effect means ±SE and *p* values for clover plant numbers and nodules numbers and volumes in Year 1 (2003, year of establishment).

	Clover plants m^{-2}	**Nodules $plant^{-1}$**	**Nodule activity %**	**Nodule volume mm^3**
Rhizobium				
-inoculum	152.5±0.14	115.2±0.06	0.7±0.10	2.5±0.10
+inoculum	190±0.13	149.1±0.21	0.7±0.08	3.8±0.15
ANOVA				
Main effects				
Rhizobium (rh)	**0.0217**	**0.0319**	ns	**0.001115**
Field	ns	ns	ns	ns
GWC	ns	ns	ns	ns

ns: not significant

Table 3.6 Differences in the influence of experimental factors on nodule numbers per plant for Year 1 and Year 2 results as expressed by factor variance as a % of total variance

	Factor variance	**Proportion Factor variance/ total variance**	**Variance component** (% of total variance)
Yr 1 (2003)			
Field	1.54	0.027	2.7
±rhizobium	35.70	0.62	62
±GWC	19.77	0.34	34
Total Factor Variance (sum)	57.08		
Yr 2 (2004)			
Field	1.65	0.88	88.6
±rhizobium	0.04	0.02	2.2
±GWC	0.17	0.09	9.2
Total Factor Variance (sum)	1.87		

Table 3.7 Main effect means ±SE and *P* values for clover plant growth and nodule number and volume in Year 2 (2004).

	Clover root FW g	**Nodules plant^{-1}**	**Nodule volume mm^3**
Rhizobium			
-inoculum	38.2±5.97	356.3±20.43	1.07±0.33
+inoculum	32.4±3.34	393.3±31.20	1.10±0.55
Fertility			
+GWC	37.7±4.94	332.8±60.59	1.10±0.32
-GWC	32.9±2.49	416.8±35.26	1.07±0.53
ANOVA			
Main effects			
Rhizobium (rh)	ns	ns	ns
Fertility (ft)	ns	ns	ns
Interactions			
rh:ft	ns	ns	ns

ns: not significant

Table 3.8 Nodule volume (mm^3) linear model analysis for Year 1 as a function of nodule number, inoculation and plant numbers per sample station.

Coefficients:	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	2.146	0.4437	4.84	2.62e-05***
Nodules plant^{-1} (nod)	**-0.0037**	0.0018	-1.99	0.0537
-rhz:Plants	**-0.0639**	0.0519	-1.23	0.2265
+rhiz:Plants	**-0.0386**	0.0531	-0.72	0.4720

Signif. codes: ***= < 0.001, ** = < 0.01, `* = < 0.

Table 3.9 Nodule numbers per plant linear model analysis for Year 1 as a function of inoculum and plant numbers per sample station.

Coefficients:	Estimate	Std. Error	t value	Pr (>|t|)
intercept	139.72	24.51	5.699	2.1e-0.5***
-rhz:Plants	**-3.98**	3.50	-1.136	0.271
intercept	207.63	45.83	4.530	0.000259
+rhz:Plants	**-7.652**	5.48	-1.394	0.180203

Signif. codes: ***= < 0.001, ** = < 0.01, `* = < 0.

Table 3.10 Predicted nodules numbers per plant and per m^2 calculated for differing clover establishments from the regression equation in Table 3.9

Clover Plants m^{-2}	Total nodules m^{-2}		Nodules per plant	
	+Rhizobium	*-Rhizobium*	*+Rhizobium*	*-Rhizobium*
25	5,200	3,400	200	136
50	9,635	6,602	193	132
100	17,739	12,408	177	124
150	24,313	17,418	162	116
200	29,357	21,632	147	108
300	34,853	27,672	116	92
350	35,305	29,498	101	84
425	33,114	30,745	86	72
600	14,611	26,688	24	44

Highlighted figures: blue = mean clover plants for +rhizobium/-rhizobium, red = maximum nodules for +rhizobium/-rhizobium (Appendix 4 for full data sets)

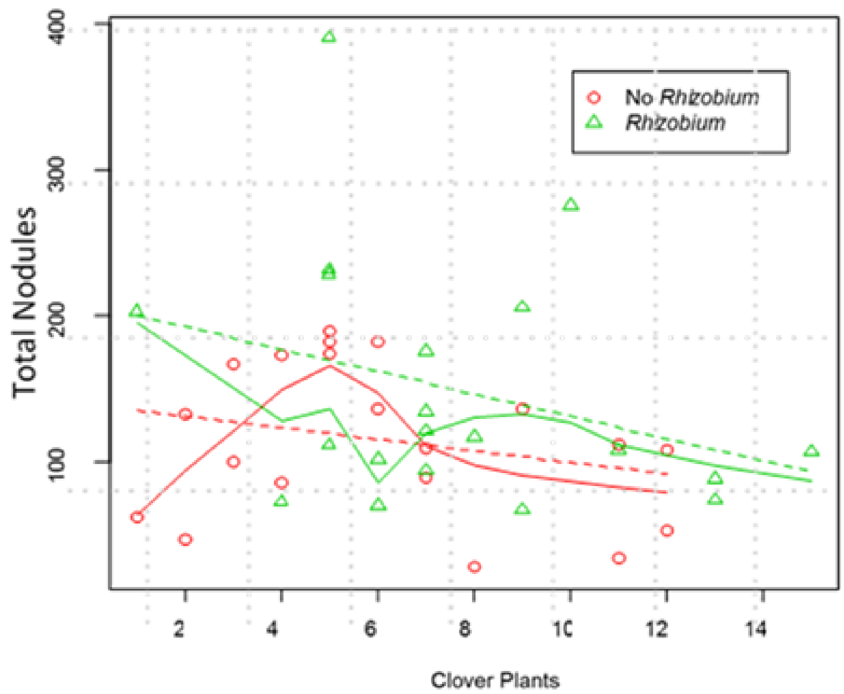


Fig. 3.4 The relationship between nodule numbers and clover plants/sample station with and without inoculum

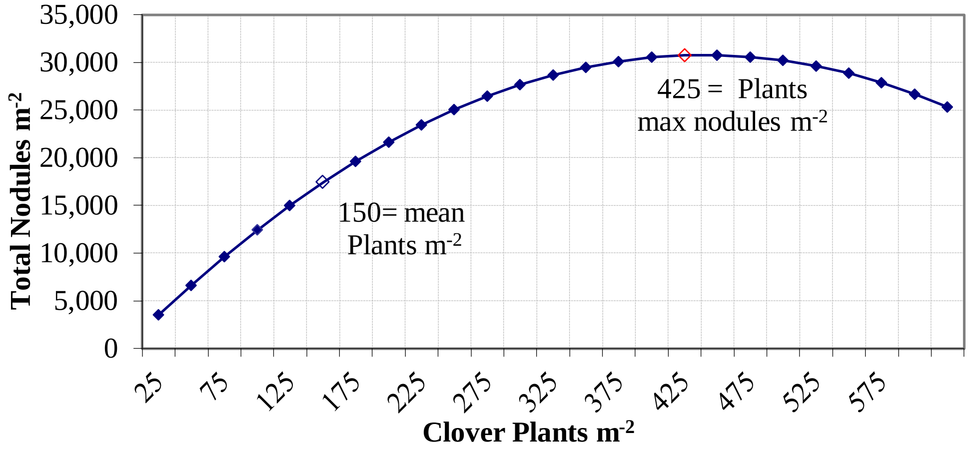


Fig. 3.5 The modelled relationship between nodule density (m^2) and the number of clover plants in the absence of inoculation

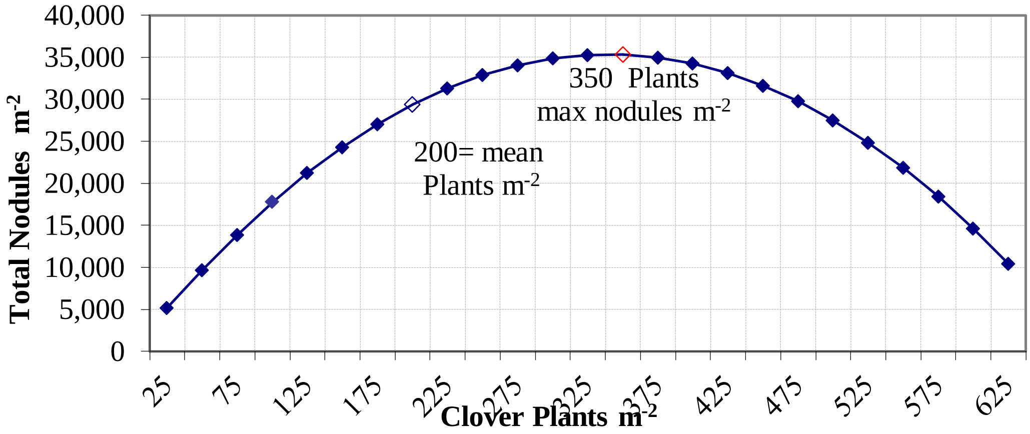


Fig. 3.6 The modelled relationship between nodule density (m^2) and clover plant number in the presence of Rhizobium inoculum.

3.3.2 Effect of clover management (Rhizobium seed inoculation and green-waste compost amendments) on soil macro-nutrient, nitrate and ammonium levels

Soil macro-nutrient content: The sampling, on 19th January 2005, of the soils from the remaining sections of unploughed clover in each field measured the soil macro and micro nutrient concentrations across the plots and sub-plots. Total available N (nitrate and ammonia) varied widely across the site and between the plots, but the trend was for more available N with +inoculum and with +GWC. Only soil N % and nitrate were significantly affected by soil depth, while *Rhizobium* inoculation and GWC amendments treatments had no significant effect (Table 3.11). There were greater levels of soil N and nitrate in the 0-30 cm horizon when compared with the other two soil depths.

Full analysis of the soil macro and micro nutrients revealed no main effects for *Rhizobium* inoculum or GWC amendments. Soil C, P and K concentrations are presented (Table 3.12), otherwise results are not shown.

Table 3.11 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum and fertility treatment on soil %N, nitrate and ammonium levels and total available N

	Soil N %	**Soil NO_3** mg kg^{-1}	**Soil NH_4^+** mg kg^{-1}	**Total available N** kg ha^{-1}
Soil depth				
0-30	0.26±0.005**a**	7.81±1.9**a**	10.2±3.19	814.5±189.00
30-60	0.18±0.007**b**	3.0±0.68**b**	7.5±1.70	477±94.05
60-90	0.14±0.007**c**	2.6±0.55**b**	5.3±1.20	355.5±63.90
Rhizobium				
-inoculum	0.19±0.01	5.6±1.44	5.5±0.91	463.5±65.70
+inoculum	0.19±0.01	4.3±0.94	9.7±2.79	625.5±127.35
Fertility				
+GWC	0.19±0.01	5.8±1.33	8.9±2.47	639.0±178.65
-GWC	0.20±0.01	4.0±0.72	6.7±1.03	472.5±54.45
ANOVA				
Main effects				
Depth (dp)	**0.0004**	**0.003**	ns	ns
Rhizobium (rh)	ns	ns	ns	ns
Fertility (ft)	ns	ns	ns	ns
Interactions				
dp:rh	ns	ns	ns	ns
dp:ft	ns	ns	ns	ns
rh:ft	ns	ns	ns	ns
dp:rh:ft	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). ns: not significant.

Table 3.12 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum and fertility treatment on soil % C, P and K concentrations

	Soil C %	**Soil P** mg kg^{-1}	**Soil K** mg kg^{-1}
Soil depth			
0-30	2.5±0.05**a**	22.3±0.87**a**	66.3±3.05**a**
30-60	1.6±0.09**b**	9.6±1.03**b**	47.3±2.81**b**
60-90	1.2±0.16**c**	4.6±0.68**c**	41.8±4.03**b**
Rhizobium			
-inoculum	1.9±0.14	12.7±1.44	47.4±3.14
+inoculum	1.7±0.06	11.7±0.73	52.3±3.26
Fertility			
+GWC	1.7±0.11	12.3±0.97	48.0±2.81
-GWC	1.9±0.10	12.0±1.19	51.8±3.52
ANOVA			
Main effects			
Depth (dp)	**0.0017**	**0.0001**	**0.0027**
Rhizobium (rh)	ns	ns	ns
Fertility (ft)	ns	ns	ns
Interactions			
dp:rh	ns	ns	ns
dp:ft	ns	ns	ns
rh:ft	ns	ns	ns
dp:rh:ft	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Spring wheat

Soil nutrient content: Sampling, on 5th March 2005, of the soils from the remaining sections of clover not ploughed in each field (clover) measured the soil macro and micro nutrient concentrations before drilling the spring wheat. Soil analysis for total available N (nitrate and ammonia) detected no significant effects for either rhizobium inoculation or GWC amendments to the soil (Table 3.14).

Full analysis of the soil macro and micro nutrients revealed no main effects for rhizobium inoculum or GWC amendments. Soil C, P and K concentrations are presented (Table 3.13), otherwise results are not shown. Soil potassium (K) concentration analysis detected a significant interaction ($p < 0.05$) between rhizobium inoculation and GWC amendments, (Table 3.13). Potassium concentration was higher with +inoculum/+compost (69.4 mg kg^{-1}) than with either +inoculum/-GWC (59.7mg kg^{-1}) or –inoculum/± GWC (55-62 mg kg^{-1}), (Table 3.15).

Table 3.13 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum and fertility treatment on soil % C, P and K concentrations

	Soil C %	**Soil P** mg kg^{-1}	**Soil K** mg kg^{-1}
Soil depth			
0-30	2.6±0.08**a**	19.6±1.14**a**	77.0±3.80**a**
30-60	1.8±0.09**b**	8.9±0.90**b**	57.3±2.80**b**
60-90	1.3±0.11**c**	3.4±0.50**c**	51.9±3.80**c**
Rhizobium			
-inoculum	1.9±0.10	10.4±0.91	59.2±4.44
+inoculum	1.9±0.08	10.9±0.87	64.6±4.69
Fertility			
+GWC	1.9±0.09	10.4±0.87	63.5±5.44
-GWC	1.9±0.09	10.9±0.90	60.8±3.91
ANOVA			
Main effects			
Depth (dp)	**0.0002**	**0.0005**	**0.0020**
Rhizobium (rh)	0.7332	0.7971	0.1530
Fertility (ft)	0.6815	0.7802	0.4117
Variety (vr)	0.8409	0.3112	0.6737
Interactions			
dp:rh	ns	ns	ns
dp:ft	ns	ns	ns
rh:ft	ns	ns	**0.0204**[1]
dp:rh:ft	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1] See table 3.15 for interaction means

Table 3.14 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum and fertility treatment on soil %N, nitrate and ammonia levels and total available N

	Soil N %	**Soil NO_3^-** mg kg^{-1}	**Soil NH_4^+** mg kg^{-1}	**Total available N** kg ha^{-1}
Soil depth				
0-30	0.20±0.01**a**	5.6±1.45	2.7±0.6	378.0±63.00
30-60	0.10±0.01**b**	3.5±0.76	0.9±0.58	202.5±45.90
60-90	0.10±0.01**c**	2.2±0.94	1.8±0.70	180.0±55.35
Rhizobium				
-inoculum	0.20±0.01	4.1±0.92	1.6±0.38	247.5±43.65
+inoculum	0.20±0.01	3.5±0.71	2.4±0.64	261.0±46.85
Fertility				
+GWC	0.20±0.01	3.9±0.57	2.4±0.76	270.0±47.25
-GWC	0.20±0.01	3.8±0.96	1.7±0.37	243.0±43.65
ANOVA				
Main effects				
Depth (dp)	**0.0004**	ns	ns	ns
Rhizobium (rh)	ns	ns	ns	ns
Fertility (ft)	ns	ns	ns	ns
Interactions				
dp:rh	ns	ns	ns	ns
dp:ft	ns	ns	ns	ns
rh:ft	ns	ns	ns	ns
dp:rh:ft	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 3.15 Interaction means for effect of *Rhizobium* inoculum and fertility treatment on soil K of spring wheat

Rhizobium	**-compost**	**+ compost**
-inoculum	61.8±4.40**a**	55.7±4.40**a**
+inoculum	59.7±3.50**b**	69.4±5.50**a**

Means with the same letter within the same column are not significantly different according to Tukey's Honest Significant Difference test ($P<0.05$).

3.3.3 Effect of clover management (Rhizobium seed inoculation and green-waste compost amendments) and variety choice on organic wheat yield and grain quality

Winter Wheat: Pre-harvest assessments of winter wheat varieties for leaf greenness were made on 27 July 2005 by SPAD at GS55. Analysis detected a significant main effect for variety but not for rhizobium inoculum or GWC amendment. The SPAD level was higher for Wenga (48.9) than for either Greina or Pollux (40.6 & 42) (Table 3.16). The use of *Rhizobium* inoculum was found to have increased the numbers of tillers per m^2 recorded post-harvest and increased grain yield. The use of GWC amendment also increased grain yield but not tiller numbers. A significant main effect for varieties was also detected, where the tillers per m^2 were lower for Wenga than Greina and Pollux, and the grain yield was lower for Wenga compared to Pollux. The yield of Greina was not different to either.

Overall grain yields were higher with +inoculum (6.2 t ha^{-1}) than –inoculum (5.63 t ha^{-1}) by 0.64 t ha^{-1}, higher with +GWC amendment (6.14 t ha^{-1}) than –GWC (5.7 t ha^{-1}) by 0.35 t ha^{-1}. Grain yield for Pollux (6.3 t ha^{-1}) was significantly higher than Wenga (5.6 t ha^{-1}) by 0.7 t ha^{-1} (Table 3.16).

All three winter varieties achieved baking or milling grade quality sufficient for the requirements of current UK organic millers and for export (HGCA classification UKp, bread making, 11-13% protein, >250 s Hagberg Falling Number and >76 kg hl^{-1} (specific weight) with grain quality parameters being indistinguishable between them. All had specific weights of approximately 81 kg hl^{-1}, for high flour extraction rates, with high HFN for good starch content (low α-amylase activity) and with high protein percentages at or around 12%. Grain hardness for all three was hard (mid-range), suitable for fine flour particle size and efficient bran separation in artisan or stone milling flour production systems (Table 3.17). Analysis detected a significant interaction ($p = 0.005$) between fertility and variety for specific weight (Table 3.17). In the absence of *Rhizobium* inoculum there was no difference in the specific weight of varieties in the absence of compost, whereas in the presence of compost it was highest for Greina (86.1 kg hl^{-1}), intermediate for Pollux (84.2 kg hl^{-1}) and lowest for Wenga (82.5 kg hl^{-1}), with Greina being significantly higher than Wenga. In the presence of rhizobium Greina had the highest specific weight both with and without compost treatments and Wenga had the lowest (Table 3.18).

Rhizobium inoculum and the GWC amendment also significantly affected NIR protein percentages (Table 3.17). The grain protein % levels were higher in the absence of the inoculum and higher with the use of the GWC amendment.

Table 3.16 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum, fertility treatment and variety choice on winter wheat yield parameters

	Yield t ha^{-1}	**Tillers m^{-2} Post-harvest**	**SPAD GS 55**
Rhizobium			
-inoculum	5.6±0.35	267.9±18.49	43.3±1.33
+inoculum	6.2±0.24	302.1±14.61	44.3±1.02
Fertility			
+GWC	6.1±0.35	296.6±16.86	44.3±1.16
-GWC	5.7±0.24	275.8±16.65	43.3±1.19
Variety			
Greina	5.8±0.49**ab**	319.0±24.89**a**	40.6±1.48**b**
Pollux	6.3±0.24**a**	305.1±13.70**a**	42.0±0.91**b**
Wenga	5.6±0.33**b**	231.9±15.13**b**	48.9±0.87**a**
ANOVA			
Main effects			
Rhizobium (rh)	**0.0329**	**0.00726**	ns
Fertility (ft)	**0.046**	ns	ns
Variety (vr)	0.0505	**0.0099**	**<0.0001**
Interactions			
rh:ft	ns	ns	ns
rh:vr	ns	ns	ns
ft:vr	ns	ns	ns
rh:ft:vr	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 3.17 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum, fertility treatment and variety choice on winter wheat grain quality parameters

	NIR protein %	Hagberg Falling Number (S)	Specific Weight kg hl^{-1}	Grain hardness h
Rhizobium				
-inoculum	12.5±0.16	324.0±11.9	80.5±0.61	69.2±1.96
+inoculum	11.6±0.20	309.1±11.32	82.1±0.74	59.0±2.54
Fertility				
+GWC	12.3±0.18	318.0±12.87	81.3±0.78	65.4±2.86
-GWC	11.8±0.21	314.5±10.60	81.4±0.64	62.4±2.19
Variety				
Greina	11.9±0.26	312.5±17.43	81.9±1.03	63.5±3.67
Pollux	12.2±0.23	319.6±12.76	81.1±0.82	64.3±3.01
Wenga	12.0±0.26	316.4±12.99	81.0±0.73	63.7±2.70
ANOVA				
Main effects				
Rhizobium (rh)	**0.0179**	ns	ns	ns
Fertility (ft)	**0.014**	ns	ns	ns
Variety (vr)	ns	ns	ns	ns
Interactions				
rh:ft	ns	ns	ns	ns
rh:vr	ns	ns	ns	ns
ft:vr	ns	ns	**0.0052**[1]	ns
rh:ft:vr	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 3.18

Table 3.18 Interaction means for effects of *Rhizobium* inoculum and fertility treatment on specific weight of winter wheat varieties (kg hl^{-1})

	without *Rhizobium* inoculum		with *Rhizobium* inoculum	
	-compost	+ compost	-compost	+ compost
Variety				
Greina	83.9±1.17**a**	86.1±0.42**a**	87.4±0.87**a**	86.1±0.77**a**
Pollux	83.8±0.71**a**	84.2±1.38**ab**	83.8±0.33**b**	84.2±0.58**b**
Wenga	81.2±0.86**a**	82.5±0.88**b**	81.6±0.642**b**	82.0±0.40**c**

Means with the same letter within columns are not significantly different according to Tukey's Honest Significant Difference test ($p < 0.05$).

Spring Wheat: Pre-harvest assessments of the spring wheat varieties for leaf greenness were made on 27 July 2005 by SPAD at GS37. Analysis detected a significant main effect for variety but not for inoculum or GWC amendment. The SPAD level was higher for both Paragon and Monsun (short straw varieties) than for both Fasan and Zebra (tall straw varieties) (Table 3.19).

The use of the *Rhizobium* inoculum was found to have increased grain yield but not tillers per m^2, recorded post-harvest. The use of the GWC amendment had no effect on yield or tiller numbers. A significant main effect for variety was detected for both tiller number and yield, where the tillers per m^2 were lower for Zebra than the other three varieties, but Zebra had the highest grain yield. Grain yield was higher after clover +inoculum (7.04 t ha^{-1}) than –inoculum (6.7 t ha^{-1}) by 0.27 t ha^{-1}, and was highest for Zebra (8.0 t ha^{-1}), intermediate for Fasan (7.0 t ha^{-1}) and Monsun (6.8 t ha^{-1}) and lowest for Paragon (5.7 t ha^{-1}) (Table 3.19).

For grain quality assessments, all four spring varieties achieved baking or milling grade sufficient for the requirements of current UK organic millers and for export (HGCA, 2017). However, there were significant differences within the grain quality parameters between them (Table 3.20). The specific weight for high flour extraction rates was better for the tall straw varieties, highest for Zebra (82 kg hl^{-1}), intermediate for Fasan (81 kg hl^{-1}) and lowest for Paragon and Monsun (78 kg hl^{-1}). High HFN was achieved by all but was lowest for Paragon (330 s) and also all varieties had high protein percentages. The highest protein percentages of 12% were found in Paragon and Monsun, the two varieties with the lowest grain (test) weights. For grain hardness Zebra and Monsun were hardest (upper range), Fasan was mid-range and Paragon the least hard (lower range) (Table 3.20). This placed them all in the upper tiers of grain hardness suitable for fine flour particulation and efficient bran separation in flour production systems (Wheat Hardness Index – Greenaway, 1969) with Monsun and Zebra being the most favourable.

Overall, Zebra had the lowest tiller count, grains with the highest specific weight with the lowest grain protein content and the highest yield.

Rhizobium inoculum and the GWC amendment had no significant effects on any of the grain quality parameters. However, for grain hardness a significant interaction (p = 0.005) between fertility and inoculum treatments was identified (Table 3.20). In the absence of compost there was no significant difference between inoculum treatments but in the presence of compost the presence of inoculum gave a significantly higher grain hardness (table 3.21)

For NIR protein a significant interaction (p = 0.005) was detected between *rhizobium* inoculum and variety (Table 3.20). The varieties Zebra, Paragon and Monsun showed no significant response to inoculum treatment while Fasan had a lower protein level in the presence of inoculum (Table 3.22). Protein levels in Zebra and Fasan were lower than Paragon and Monsun with their lower grain densities (Table 3.22).

Table 3.19 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum, fertility treatment and variety choice on spring wheat yield parameters

	Yield **t ha^{-1}**	**Tillers m^{-2}** **at harvest**	**SPAD** **GS37**
Rhizobium			
-inoculum	6.7±0.17	339.3±9.50	47.0±0.45
+inoculum	7.0±0.23	368.3±11.95	45.6±0.49
Fertility			
+GWC	6.8±0.23	352.0±12.58	46.1±0.52
-GWC	6.9±0.18	355.7±9.4	46.5±0.46
Variety			
Paragon SS	5.7±0.11**c**	388.4±15.59**a**	47.4±0.60**a**
Monsun SS	6.8±0.19**b**	355.0±11.95**a**	47.6±0.63**a**
Fasan LS	7.0±0.27**b**	354.7±17.37**a**	45.0±0.59**b**
Zebra LS	8.0±0.25**a**	317.2±12.75**b**	45.3±0.72**b**
ANOVA			
Main effects			
Rhizobium (rh)	**0.0532**	ns	ns
Fertility (ft)	ns	ns	ns
Variety (vr)	**<0.0001**	**0.0083**	**0.0008**
Interactions			
rh:ft	ns	ns	ns
rh:vr	ns	ns	ns
ft:vr	ns	ns	ns
rh:ft:vr	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Table 3.20 Main effect means ± SE and *p* values for the effects of *Rhizobium* inoculum, fertility treatment and variety choice on spring wheat grain quality

	NIR protein %	**Hagberg Falling Number (s)**	**Specific Weight** kg hl^{-1}	**Grain hardness** h
Rhizobium				
-inoculum	11.7±0.13	361.1±5.26	79.7±0.48	66.8±2.02
+inoculum	11.8±0.13	354.0±5.15	79.7±0.42	68.2±1.84
Fertility				
+GWC	11.9±0.13	361.1±4.82	79.7±0.48	68.6±1.98
-GWC	11.6±0.13	354.0±5.56	79.7±0.43	66.4±1.87
Variety				
Paragon SS	11.9±0.15**ab**	329.6±5.08**b**	78.1±0.43**c**	56.8±2.23**c**
Monsun SS	12.1±0.19**a**	370.8±6.58**a**	77.9±0.53**c**	73.3±2.78**a**
Fasan LS	11.6±0.20**bc**	368.1±4.44**a**	80.8±0.28**b**	68.3±2.29**b**
Zebra LS	11.4±0.15**c**	361.6±8.14**a**	82.1±0.55**a**	71.5±1.45**a**
ANOVA				
Main effects				
Rhizobium (rh)	ns	ns	ns	ns
Fertility (ft)	ns	ns	ns	ns
Variety (vr)	**0.004**	**<0.0001**	**<0.0001**	**<0.0001**
Interactions				
rh:ft	ns	ns	ns	**0.0253**[1]
rh:vr	**0.0426**[2]	ns	ns	ns
ft:vr	ns	ns	ns	ns
rh:ft:vr	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$). [1] See table 3.21 for interaction means. [2]See table 3.22 for interaction means.

Table 3.21 Interaction means for effect of *Rhizobium* inoculum and fertility management practices on grain hardness (h) of spring wheat

Rhizobium	**-compost**	**+ compost**
-inoculum	68.0±3.03**a**	65.5±3.03**b**
+inoculum	64.7±2.58**a**	71.6±2.38**a**

Means with the same letter within columns are not significantly different according to Tukey's Honest Significant Difference test (p <0.05).

Table 3.22 Interaction means for effect of *Rhizobium* inoculum on NIR protein % of spring wheat

Rhizobium	**Fasan**	**Monsun**	**Paragon**	**Zebra**
-inoculum	11.9±0.32**a**	12.0±0.25**a**	11.9±0.23**a**	11.2±0.19**b**
+inoculum	11.3±0.18**b**	12.2±0.29**a**	12.0±0.19**a**	11.6±0.22**b**

Means with the same letter within rows are not significantly different according to Tukey's Honest Significant Difference test (p<0.05).

3.3.4 Effects of clover management (Rhizobium seed inoculation and green-waste compost amendments) on vitamin E concentrations in organic wheat

Winter wheat: Vitamin E with its chemical antioxidant properties comprises four tocopherols (alpha, beta, gamma and delta) and four tocotrienols (alpha, beta, gamma and delta). Vitamin E in the form of α-tocopherol has the highest biological activity and is preferentially absorbed and accumulated in the human body. Grains were analysed for α- and β-tocopherols and α- and β- tocotrienols only.

The use of rhizobium inoculum and the GWC amendment had no significant effect on any vitamin E groups. A significant effect for variety was detected in all four groups (Table 3.23). The α-tocopherol was highest in Pollux, 9.8% higher than Greina. β-Tocopherol was highest in Greina, 21% higher than Wenga. α- and β- Tocotrienols were highest in Wenga, 15% (α-) and 38% (β-) higher than Greina. Analysis detected no significant interactions.

Table 3.23 Main effect means ± SE and *p* values for the effects of fertility treatment and variety choice on winter wheat tocopherol concentration (μg g^{-1} DW)

	α-tocopherol	**α-tocotrienol**	**β-tocopherol**	**β-tocotrienol**
	μg g^{-1}	μg g^{-1}	μg g^{-1}	μg g^{-1}
Rhizobium				
-inoculum	12.6±0.30	2.9±0.09	6.3±0.22	21.9±0.80
+inoculum	12.1±0.29	2.7±0.09	6.2±0.19	21.5±0.78
Fertility				
+GWC	12.4±0.32	2.8±0.09	6.2±0.19	21.5±0.91
-GWC	12.3±0.28	2.9±0.09	6.3±0.21	21.9±0.67
Variety				
Greina	11.5±0.33**b**	2.6±0.09**b**	6.9±0.17**a**	17.9±0.50**c**
Pollux	13.0±0.32**a**	2.8±0.09**b**	6.2±0.16**b**	22.4±0.52**b**
Wenga	12.5±0.36**ab**	3.0±0.13**a**	5.7±0.30**b**	24.7±0.86**a**
ANOVA				
Main effects				
Rhizobium (rh)	ns	ns	ns	ns
Fertility (ft)	ns	ns	ns	ns
Variety (vr)	**0.0299**	**0.0159**	**0.0009**	**<0.0001**
Interactions				
rh:ft	ns	ns	ns	ns
rh:vr	ns	ns	ns	ns
ft:vr	ns	ns	ns	ns
rh:ft:vr	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Spring wheat: The use of *Rhizobium* inoculum and the GWC amendment had no significant effect on either vitamin E group. For the spring wheats a significant effect for variety was detected in all four groups (Table 3.24). The α- and β-Tocopherols were highest in Paragon and Zebra, α- tocotrienol was highest in Monsun, intermediate in Zebra and Paragon and lowest in Fasan. β- Tocotrienol was highest in Paragon, intermediate in Monsun and Zebra and lowest in Fasan. Analysis detected no significant interactions.

Table 3.24 Main effect means ± SE and *p* values for the effects of fertility treatment and variety choice on spring wheat tocopherol concentration (µg/g DW)

	α-tocopherol	**α-tocotrienol**	**β-tocopherol**	**β-tocotrienol**
	µg g^{-1}	µg g^{-1}	µg g^{-1}	µg g^{-1}
Rhizobium				
-inoculum	11.6±0.51	2.9±0.28	4.3±0.21	18.7±0.77
+inoculum	11.6±0.53	2.6±0.15	4.1±0.21	19±0.85
Fertility				
+GWC	11.9±0.46	2.6±0.14	4.2±0.21	18.5±0.82
-GWC	11.3±0.57	2.9±0.29	4.2±0.21	19.3±0.79
Variety				
Paragon	12.7±1.21**a**	2.4±0.15**bc**	5.14±0.19**a**	22.3±1.22**a**
Monsun	10.6±0.45**b**	3.6±0.52**a**	2.99±0.16**c**	18.3±1.12**b**
Fasan	10.2±0.21**b**	2.0±0.06**c**	3.50±0.11**b**	14.8±0.48**c**
Zebra	12.7±0.41**a**	2.8±0.17**b**	5.08±0.24**a**	19.9±0.68**b**
ANOVA				
Main effects				
Rhizobium (rh)	ns	ns	ns	ns
Fertility (ft)	ns	ns	ns	ns
Variety (vr)	**0.0199**	**0.0026**	**<0.0001**	**<0.0001**
Interactions				
rh:ft	ns	ns	ns	ns
rh:vr	ns	ns	ns	ns
ft:vr	ns	ns	ns	ns
rh:ft:vr	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

3.4 Discussion

Most referenced earlier trial work on clover pre-crop studied the effects of N fixation in above ground plant tissue only (Carlsson *et al.*, 2009; Borgen *et al.*, 2012). A systematic literature review of N_2 fixation by legume forages (Anglade *et al.*, 2015) confirmed that the total N calculations by Carlsson and Huss-Danell (2003) of 371 kg N per ha per year from red clover were robust but limited due to the lack of an accurate value for the contribution of the below ground components. The review by Anglade *et al.* (2015) established a significant correlation between N accumulation in the clover shoot matter (above-ground) and fixed N in the forage, but conceded that the contribution of the below ground components was more difficult to establish due to the lack of relevant studies. This lack of root based N calculations, therefore, underestimates the total N_2 fixed as the values are not including the below ground contribution comprising the nitrogen derived from roots (Ndfr). What Anglade *et al*. (2015) were able to establish was that for every kg of N found in the shoot, symbiotic *Rhizobium* was responsible for 0.75-0.8 kg of N. The results from this thesis (see also Wilkinson *et al.* 2007) identified overall that the use of *Rhizobium* inoculation of red clover seed (but not compost amendment) significantly increased clover establishment, nodule number and the mean size of nodules. So with Millington *et al.* (1990) having identified the importance of green crop manures for stockless arable farm enterprises from only a one year clover ley, which supports the findings of this study, these results further identified the development of clover plants and associated *Rhizobium* populations after a second year of growth.

Given the sparse values of below ground N (BGN) contribution to total plant N reported in the literature it is hard to calculate and quantify the contribution of these clover root and nodule figures (Anglade *et al.*, 2015) further than a calculated 40% contribution of BGN to the total. However, by studying the nodule count and volume data for the two years a clear pattern emerges. In 2003 the treated clover had more *Rhizobium* root colonisation than the untreated blocks and the nodules were 50% larger. By May 2004 the nodule count for both the untreated and treated clover plants had increased to means ranging from 350 to 420 nodules per plant compared with the means in 2003 of 100 to 170 nodules per plant, whilst conversely the volumes had decreased in 2004 to a grand mean of 1.09 mm^3 from a grand mean of 3.2 mm^3 in 2003.

Despite linear model analysis identifying an increase in nodule volume with increasing root volume, this is relative only to the activity in 2003. When comparing the relative changes in mean nodule numbers per plant and mean volumes of those nodules from 2003 to 2004 the data shows an overall decline in mean volumes of nodules as the overall number of nodules per plant increased. This decrease in the mean volume of nodules per plant in 2004 is off-set by the 3 fold increase in nodule numbers per plant. For larger nodules, where the *Rhizobium* inoculation had been applied, the decrease in volume was less pronounced with the increasing numbers of nodules per plant. The significance of the ability of the inoculum to increase nodule size by 50% and overshadow the 2.5% loss in nodule number per plant would become increasingly noticeable in N production clover crops where only one year of production in the rotation is possible.

Direct comparisons of the total factor variance in the data highlighted the dominance of the rhizobia treatment in the clover and nodule data in 2003 (62.6%) and its virtual disappearance as an effect on clover nodule numbers and volume the following year (2.2%). The field effect by contrast accounted for only 37% of the variance in 2003 but 88.6% in 2004. The change in impact of the factor variance from treatment to field effect can be attributed to the change from initial colonisation by *Rhizobium* of the small clover root structure in Year 1 (where the use of the inoculum increased *Rhizobium* activity) to the increased colonisation by the spring in Year 2 on all of the larger mature clover root structures by all rhizobia, regardless of initial colonisation numbers. The extremely low levels of naturally occurring *Rhizobium* in the absence of clover from the previous non-organic rotation for several decades prior to this trial meant that where the *Rhizobium* inoculum was introduced this dominated nodule production within the fields (given that the natural soil levels were equally low in all fields and hence the low field effect) initially but by Year 2 the slower rate of colonisation on the untreated clover had achieved the same level of colonisation as the treated for any given clover plant density. The initial impact of the inoculum was no longer apparent once the rhizobia activity in both treatment plots had reached the same natural plateau.

The relatively low numbers of nodules in Year 1 compared to Year 2 also allowed the nodules to become larger. Once the clover sward was established, *Rhizobium* activity increased to levels in both treated and untreated plots where the natural laws of diminishing returns set in. The soil conditions in fields and specific plots within them differed sufficiently for different clover root system development. It is the overall differences in the root systems between fields and within fields which are sustaining greater or lesser natural plateaus of rhizobia activity.

The examination of the use of a high C:N compost on the clover, in an attempt to enhance the capture of the accumulated plant derived nitrogen which is lost as gaseous N when clover is cut and mulched, identified no positive results from the soil analysis. This is similar to the report by Duong *et al.* (2012), which concluded that compost N availability was not a good indicator of soil N availability. From the study of the harvest and grain quality parameters of the subsequent wheat crops the addition of the green waste compost to enhance available N, as suggested by Aulakh *et al.* (1983), significantly increased both the grain yield and protein levels of winter wheat. It had a cumulative impact on the both the grain yield and specific weight of the Greina (short straw) winter variety, increasing the yield both in the absence and in the presence of *Rhizobium* inoculum. It had a similar effect on the specific weight of Greina which was significantly higher with the compost amendment both in the absence and in the presence of rhizobium inoculum than Wenga or Pollux. For spring wheat the addition of the green waste compost to enhance soil N levels had no effect on improving grain yield or grain protein level. As detailed studies with differing compost types on differing soils (Duong *et al.*, 2012) concluded that composts can provide a 6 fold increase in N concentrations to subsequent wheat crops regardless of soil type, it is not possible from this study to assess the direct contribution of the compost in reducing gaseous N losses from the clover as a contribution to soil N. Duong *et al.* (2012) also concluded that compost N availability was not a good indicator of soil N availability, which was being determined more, possibly, by nutrient mobilisation after incorporation than at the time of application. Wright *et al.* (2007) concluded that dissolvable organic C and macronutrients from composts increased in availability over time whereas NO_3^- declined, and that availability of all was subject to considerable seasonal variation. Both of these studies would suggest that the positive response of winter wheats to the compost amendment in this study, against the lack of response by the spring wheats, can be accounted for by the benefits of earlier incorporation of compost resulting in a longer mineralisation period in the winter wheat soils than for the spring ones six months later, with the seasonal conditions over winter being sufficiently favourable to reduce potential losses of NO_3 over time to a minimum.

Grain hardness has been identified as having a genetic (G) as well as an environmental (E) component where the proteins which determine grain hardness can be influenced both by grain protein quality and availability to the growing crop (G x E) (Pasha *et al.*, 2010). Comparative studies on the effect of farming practices on grain hardness (Carcea *et al*., 2006) identified organic farming practices as having a negative impact on grain hardness when compared to higher grain hardness levels in conventional high fertility systems, highlighting the impact of protein availability and quality on overall grain hardness. The improvement of spring wheat grain hardness from compost amendments in combination with the *Rhizobium* inoculum from this study does support these findings and demonstrates that the potential impact of lower fertility organic farming on grain hardness can thus be addressed and overcome with improved N availability.

The stabilisation of the *Rhizobium* population across the fields of the trial site over the 2 years might explain the lack of significant differences in the soil analysis between the treatments, although the trend was for more available N from +*Rhizobium* and +GWC. However, it is recommended that further studies to identify the benefit of *Rhizobium* inoculum and compost amendments in shorter cropping periods in an organic rotation would be beneficial, particularly once past the 2 year conversion stage. This study was able to confirm *Rhizobia* colonisation does occur, even if at a much slower rate, on farms where no clover has been grown for prolonged periods of time and also suggesting that the switching on of the nodulation genes identified by Peters *et al.* (1986) is not host specific. Therefore, assessing the benefit of continued repeated inoculation of clover seeds when sowing legume forages in a rotation, where previous clover leys previously had this treatment in the rotation, could help identify the ability for clover *Rhizobium* to remain present in greater numbers in the absence of the host plant during the organic rotation and thus improve nutrient availability during longer organic rotations.

The recorded general observations from recent systematic reviews of improved yield with organic winter wheats and improved quality from spring wheats (Hossard *et al.*, 2016a) were not upheld by this study (Wilkinson *et al.*, 2006). Here the selected winter wheat varieties produced high yields plus baking quality equal to the milling quality of spring varieties chosen for the trial. Increased N fixation and retention through both inoculation and green waste compost treatments, the late incorporation of the clover in the autumn as recommended by Lahti and Kuikman (2003) for winter wheats and the use of tall straw varieties as recommended by Eisele and Köpke (1997), or different wheats for differing nutrient patterns all combined in this study to produce grain yields and grain quality parameters for both the winter and spring wheat crops that were very similar and of good quality (Bilsborrow *et al.*, 2013). Previous studies of wheat performance after clover (Lahti and Kuikman, 2003) demonstrated spring wheat grain yields of 4.03 t ha^{-1} from only 1 year clover leys, whilst from both 1 and 2 year red clover leys (Stopes *et al.*, 1996) for spring and winter wheats yields of 6.0 t per ha for winter wheat were not improved by a second year of clover, but were significantly higher than after white clover (5.2 t ha^{-1}) or for non-legume rye grass (3.3 t ha^{-1}). Nitrogen availability, long held to be the driver for grain yield and quality (Murphy *et al.*, 2005) was sufficient for all winter wheat cultivars and both tall and short spring ones to produce grains of equal density, hardness and protein content from comparable grain yields, despite differences in plant tiller numbers. This supports the study by Slafer and Miralles (1993) which shows the plant's ability to compensate for a low ear population particularly via an increased number of grains per ear and TGW.

The grain yield range in this study of 5.6 -7.0 t ha^{-1} was comparable between the winter and spring wheats, with the exception of the very high yielding Zebra at 8.0 t ha^{-1} (Wilkinson *et al*., 2006). Specifically grain yields of wheat crops planted after inoculum treated clover crops increased by 0.66 t ha^{-1} or 11.8%. These compare favourably with the winter wheat yields recorded after one year of red clover by Stopes *et al.* (1996) of 6.0 t ha^{-1} and are better than yields after two years of red clover of 5.2 t ha^{-1}, despite their results identifying no benefit to yield from a second year of red clover. Protein content, on the other hand, was significantly increased by compost amendment of red clover swards, but only when used in conjunction with *Rhizobium* inoculation. Our results identified significant interactions between fertility management practices and variety for both winter and spring wheats, which was also identified by Miko *et al.* (2014). They identified strong interactions between management practices and wheat genotypes from 37 bread wheat varieties in organic and low input systems. These interactions from this study further support the observation by Osman *et al.* (2016) and Rakszegi *et al.* (2016) on variety trait differences that the common Value for Cultivation and Use test (VCU) trialled on conventional fields needs to be extended in order for a wider range of wheat traits to be examined for organic agriculture and included on the checklist. ["At the end of the breeding process, breeders need to submit candidate varieties for official registration. In the European Union (EU), variety tests, so called Value for Cultivation and Use (VCU) tests, are part of this procedure. These tests favour the selection of certain types of varieties through the choice of testing environment and evaluation criteria. EU seed legislation provides national authorities flexibility to adjust criteria and testing protocols to local needs and new demands. As a result procedures and criteria differ among EU countries. However, in most EU countries (including The Netherlands and Germany) candidate varieties have to reach a minimum yield level to pass the regular VCU tests. The German VCU procedure includes extensive evaluation of baking quality. In contrast, in the Netherlands, since 2006 baking quality tests are no longer part of conventional spring wheat VCU." (Osman *et al.*, 2016)]

In the winter wheat trial *Rhizobium* inoculation and compost amendment had an additive effect on yield for Greina, while for Wenga only the combined use of both treatments resulted in a significant increase in yield, and for Pollux neither of the treatments had a significant effect. Also for Greina, the protein content was lowest when *Rhizobium* inoculum was used without compost amendment, and increased by compost amendment to the clover ley grown before wheat crops. In contrast, for Pollux, only the reduction in protein content associated with *Rhizobium* inoculation was significant and there was no significant effect of clover management practices on protein content for the variety Wenga. Bilsborrow *et al.* (2013) have subsequently shown that previous crop is important with respect to wheat performance and the use of grass/clover leys is key in providing fertility to the crop in an organic management system where mineral fertilisers are not permitted. This is consistent with the results from Miko *et al.* (2014), whose study on the 15 traits of 37 winter bread wheat varieties found that performance of genotypes was strongly influenced by environment and also by Maghirang *et al.* (2006), whose study of 15 winter and 15 spring hard bread wheats failed to identify definitive differences but concluded also that quantitative differences were primarily genetic *and* environmental.

The effect of clover management practices on the yield of spring wheat was less pronounced than that observed with winter wheat, but again the effect differed among varieties. The addition of the green waste compost to enhance soil N levels had no effect on improving yield or grain protein level for the spring wheat. The yield of short straw varieties was not significantly affected by clover management. Comparisons of landraces and modern spring bread varieties by Konvalina *et al.* (2014) and by Migliorini *et al.* (2016) observed marked differences in yields, as expected. Yields of both groups (landrace/tall straw – 1.9 t ha^{-1} and modern/short straw – 3.4 t ha^{-1}) were much lower than for either group here (short straw – 6 t ha^{-1}, tall straw – 7.5 t ha^{-1}). In this study no significant interactions were observed either between varieties and treatments or between varieties themselves for yield. However, for the short straw varieties the use of either *Rhizobium* inoculum or compost amendment alone reduced protein contents, which could not be accounted for by the usual dilution effect associated with yield. In contrast, for the long straw varieties the use of *Rhizobium* inoculation alone reduced protein content for Fasan, in line with the usual dilution effect from higher yield, while for Zebra the combined use of *Rhizobium* inoculum and compost amendment increased protein contents along with yield. In both these varieties the compost amendment in the presence of the inoculum increased protein levels along with yield increases thereby off-setting the dilution effect.

The range of different variety traits between the winter and spring varieties suggests that there are current varieties suited to organic systems for bread making quality despite the hypothesis to the contrary by Przystalski *et al.* (2008). That the experiment was able to produce bread quality wheat from winter wheats in the north of England as well as from spring wheats under organic conditions opens up the possibility for quality cereal production to be achieved in greater quantity across a wider range of locations within the UK for the needs of both local as well as national milling and baking enterprises.

The results re-affirm the positive observations from earlier pre-crop wheat trials that the strongest wheat crops, both spring and winter, can be produced after 100% clover leys (Loshakov *et al.*, 1997; Vaisanen and Pihala, 1999; Loges *et al.*, 2006). However, Vaisanen and Pihala (1999) concluded the most important factor for baking quality was the cultivar, which from this study might suggest that pedo-climatic and fertility input factors can be more influential, when one considers the origins and growing conditions of the varieties selected for this trial. One of the spring varieties, Zebra, is grown for organic bread wheat in Norway and all three of the winter wheats are grown for organic wheat production in Switzerland with harsher winters and a shorter drier summer growing period until harvest being especially conducive to higher protein and HFN levels. Greina is marketed as a dual purpose (spring or winter sowing) variety and it performed well as a winter wheat for the north of England trial. It performed as well as the two tall straw dedicated winter varieties.

For the grain quality parameters for milling quality the experiment in this study produced a high specific weight for all varieties across the yield range. The specific weight did not decrease significantly with yield increases as was reported by Slafer and Andrade (1993) in their work on older taller and modern shorter varieties. Where differences occurred between treatments for a variety the presence of compost and inoculum treatment improved yield and grain specific weight, as was the case with Zebra. All varieties could be milled as single or milling grists, as the range for high white flour extraction was similar for all (Borghi *et al.*, 1995), although more recent studies (Hook, 1984) conclude that the correlations between grain specific weight and flour yield are poor. The pre-crop treatment of GWC in the absence of rhizobium inoculum did affect grain weight and protein content and the GWC treatment in the presence of the inoculum improved the protein content for varieties with higher grain weight, thus overcoming the milling quality problems identified by Gaines *et al.* (1997). The experiment does concur with the one performed by Borghi *et al.* (1995) that the effect of climate, soil nutrient availability and cultivar can all influence wheat quality in any given season.

Grain hardness of the varieties identified all of them as being generally hard wheats, with all the winter varieties being of mid-range hardness (60s), with the spring varieties Zebra and Monsun recording the highest hardness (70s) and Paragon the lowest (50s). The interaction detected between grain hardness and fertility treatment lends support to the evidence provided by Fox *et al.* (2007) of an environmental influence on hardness of increased proteins resulting in harder grains. Green waste compost in the presence of *Rhizobium* inoculum increased grain hardness significantly over GWC in the absence of *Rhizobium* inoculum for spring wheat. Given the negative influence of organic farming practices on grain hardness (Carcea *et al.*, 2006) identifying the means to improve N retention and availability for the most critical of all milling quality components (Hrušková *et al.*, 2012) is essential for organic production. The importance of grain hardness in baking performance has also been identified with phytate levels in flour for nutrient availability (Greffeuille *et al.*, 2005) with higher phytate levels occuring in flours from harder grains. The concept of grain hardness increasing with grain size could not be confirmed from the trial (Yinian *et al.*, 2008).

For antioxidants in the grains, Vitamin E was not found to be affected by the fertility or clover treatments. There were significant differences in levels between the varieties and the levels of α-tocopherol and β-tocotrienol were higher in the winter wheat varieties than spring, which was expected given the strong correlation found by Lv *et al.* (2012) between wheat genotypes and α- and β-tocopherols α- and β-tocotrienols.

However, the levels in both the winter and spring varieties were either higher than or equal to the values of other wheat groups from other studies. For each of the tocopherols and tocotrienols tested by Hussain *et al.* (2012b), recorded means for landraces, primitive wheats, old and modern cultivars showed the highest α-tocopherol levels of 10.3 $\mu g\ g^{-1}$ in landrace wheats compared with 8.88 $\mu g\ g^{-1}$ in modern cultivars, which were lower than the 11-12 $\mu g\ g^{-1}$ in the spring and 12-13 $\mu g\ g^{-1}$ in the winter wheat varieties of this study. The levels of α-tocotrienol for the winter and spring wheats in this study were similar to the Hussain study wheat groups (3-4 $\mu g\ g^{-1}$). For β-tocopherol, levels were lowest in the primitive wheats (1.87 $\mu g\ g^{-1}$) and highest in the old cultivars and landraces (4.08 $\mu g\ g^{-1}$). These were similar to the spring wheats (4-5 $\mu g\ g^{-1}$) in this study but lower than the winter wheats (6-7 $\mu g\ g^{-1}$). All of the groups in the Hussain *et al.* (2012) study were found to have similar levels of β-tocotrienol (15-16 μg g^{-1}) but these were all lower than those of the spring wheats (18-19 μg g^{-1}) and the winter wheats (21-24 μg g^{-1}) in this study.

3.4 Conclusions

The results presented indicate that two of the main problems relating to the sustainability of current organic wheat production methods (lower yields and protein contents) can be addressed by changes in fertility management practices and variety choice. Yields of 6 to 8 t ha^{-1} recorded for the long-straw spring wheat varieties under improved fertility management were similar or only slightly below the average (7.5 t ha^{-1}) obtained with short straw winter wheat varieties (e.g. Malacca) under high input conventional conditions used in Northumberland. Suitable grain quality for organic bread flour can be achieved from winter wheat as well as spring wheat with changes in fertility management practices and the use of suitable varieties for organic production in the north of England.

CHAPTER 4: Effect of variety of organic spring wheat on grain yield and baking quality

4.1 Introduction

The use of varieties adapted to specific regional and local pedo-climatic conditions as a means of stabilising UK organic quality cereal production requires the assessment of varieties already recognised for their quality traits across a range of different conditions (Migliorini *et al.*, 2016; Osman *et al.*, 2016). The stable production platform also requires these grains to be consistent when exposed to seasonal variability in growing conditions and for them to be approved by industrial processors and millers. Previous studies have recorded how the effect of climate, soil nutrient availability and variety can all influence wheat quality in any given season (Borghi *et al.*, 1995) and even more so in organic production systems. Because breeding for modern bread wheat varieties has been predominantly conducted in the background of conventional, high input industrial chemical dependent agriculture (Murphy *et al.*, 2005) it is becoming increasingly important for UK organic cereal production to assess the performance of these varieties in as many different organic production systems/environments as possible (Mesdag, 1985; Loschenberger *et al.*, 2008; Lammerts van Bueren *et al.*, 2011). Comparison of bread wheat varieties from different breeding origins has identified the strong influence of genotype across a range of environmental conditions (Miko *et al.*, 2014) and how growing conditions can further influence the quality of those bread wheats (Carcea *et al.*, 2006), even from year to year (Švec *et al.*, 2006). Aside from regional and local pedo-climatic conditions, organic farms display considerable variation in management practices according to the options available from their wide range of business enterprises (Millington *et al.*, 1990; Stopes *et al.*, 1996). Rotations and fertility programmes will vary from farm to farm even in a relatively local area, further influencing the technical performance and stability of bread wheat for organic baking. Direct comparisons of wheat varieties between conventional and organic systems demonstrated that the best performing varieties from within a conventional cereal production system are often not the best when grown organically (Murphy *et al.*, 2007) and that varieties can adapt to different conditions.

The aims and objectives of this chapter were therefore:

(i) To identify organic spring wheat varieties which provide a suitable balance of protein content and quality through their ability to utilise organic N supply regimes and which provide a raw material of good value to the bread-making industry.

(ii) To determine how different varietal types respond to organic cultivation techniques and the effects/impacts on key quality traits.

4.2 Materials and methods

4.2.1 Trial site characteristics

Identical trials were established on three sites in England with contrasting pedo-climatic conditions (Table 4.1) in two wheat growing seasons, 2006 and 2007. The three farms were certified according to organic production standards. They contrasted in terms of soil type, local climate and disease pressure and hence yield and quality potential. The materials, methods and results of the experiment conducted at Gilchesters Farm are presented here. Selected results from the other two identical experiments are included at a later stage (Chapter 4 Discussion and in full in Appendix 6) to enable direct comparisons of the performance of the selected bread wheat varieties across the contrasting pedo-climatic conditions and seasons.

At Gilchesters in Northumberland, north eastern England, soils are of the Brickfield and Dunkeswick series (Jarvis *et al.*, 1984b). These tend to be heavy, moisture retentive, clay loams. Risk of damaging soil structure when the soil is wet limits opportunities to cultivate and establish cereal crops. Sowing in spring can be delayed, leading to a late harvest, which may compromise grain quality as weather conditions deteriorate. Temperatures are lower than at southerly sites but water stress is rare, leading to a higher yield potential. The experiments from the previous chapter, examining the effects of fertility management and variety choice on yield and baking quality of organic spring and winter wheat, were conducted at this site.

At Courtyard Farm in Norfolk, eastern England, the soil type is Newmarket 2 series (Hodge *et al.*, 1984). Coarse calcareous loams formed in chalk are light and very well drained, and can be cultivated soon after rainfall. Early sowing of cereals is possible in spring and harvest is early. Shallow rooting coupled with drought frequently restricts growth and yield. Some fields are low in fertility but others have benefited from manure inputs from outdoor pig enterprises.

At Sheepdrove Farm, Berkshire, southern England, the silty clay loam soils belong to the Andover 1 series formed over chalk (Jarvis *et al.*, 1984a). Well-drained, they dry rapidly after rain, facilitating early cultivation and sowing in spring. In drought conditions, effects of water stress on crop growth and yield may be alleviated by available water held in the chalk.

Soil samples were taken from each site prior to spring sowing in each season (Table 4.2) from the top soil (0-30 cm) and sent to Field Science Ltd, Bristol for analysis. Soil nutrient analyses showed differences in inherent fertility particularly in terms of C and N between the sites, with Courtyard being the lowest, Sheepdrove the highest and Gilchesters intermediate (Table 4.2).

Table 4.1 UK trial sites

		Courtyard	Gilchesters	Sheepdrove
Location		Hunstanton, Norfolk	Stamfordham, Northumberland	Wantage, Hampshire
Coordinates		52° 55' 46.54" N 0° 34' 17.88" E	55° 2' 15.162" N 1° 54' 7.0488" W	51° 32' 4.5852" N 1° 29' 5.64" W
Soil type		Coarse calcareous loam	Clay loam	Silty clay loam
pre-crop	**2002/03**	Triticale (no FYM)	Winter wheat	Spring barley
	2003/04	Spring barley	Winter barley	White clover
	2004/05	White clover	Red clover +rhizobium inoculum	White clover
	2005/06	White clover	Red clover Topped x4	White clover

Table 4.2 Soil analysis pre-drilling in 2006 and 2007

Year:	**2006**	**2007**
Farm:	**Gilchesters**	**Gilchesters**
N kg ha^{-1}	109.2±0.03	148.2±0.30
C %	2.6±0.05	3.8±0.50
P mg kg^{-1}	30.9±2.19	20.5±1.60
K mg kg^{-1}	204.0±15.12	127.5±5.81
Ca mg kg^{-1}	1299.2±48.44	4425.0±1099.09
S mg kg^{-1}	16.4±0.66	22.2±1.43
Mg mg kg^{-1}	192.7±6.31	125.5±18.49
Na mg kg^{-1}	18.4±2.54	31.1±4.76
Fe mg kg^{-1}	455.2±14.84	246.1±32.73
Mn mg kg^{-1}	58.7±3.17	159.8±14.84
Zn mg kg^{-1}	2.8±0.11	4.6±0.87
Cu mg kg^{-1}	2.26±0.037	3.8±0.25
B mg kg^{-1}	0.21±0.018	1.8±0.47
Al mg kg^{-1}	630.7±21.59	627.6±118.89
Pb mg kg^{-1}	6.20±0.34	13.0±1.32
Ni mg kg^{-1}	1.20±0.04	2.3±0.15
Cd µg kg^{-1}	103.80±2.82	209.0±32.34
Mo µg kg^{-1}	10.20±0.82	19.0±3.93
pH	6.9	6.7

4.2.2 Experimental design and agronomic methods

Six spring wheat varieties were evaluated at each farm site. At each site a 1.5 ha experimental area was selected sufficient for four 48 m x 120 m blocks in which the varieties were randomised with four replicates, for an 8 m x 30 m individual plot size per variety.

4.2.3 Wheat varieties

The spring varieties selected represented those with known track records for milling quality from both the conventional and organic production sectors in Europe. Varieties also spanned a range of agronomic characteristics:

Paragon: RAGT Seeds Ltd, Cambridge. First listed 1999, this NABIM Group 1 variety (NABIM, 2010) is the most popular spring variety grown for milling and bread making in the organic sector, accounting for about 16% of the total spring wheat supplies from the 2009 UK national harvest (HGCA, 2010). Although yields are generally lower than other spring varieties, they are compensated by very good bread making characteristics. The straw is relatively long but stiff (resistance to lodging score = 6) and the variety shows good all-round disease resistance (powdery mildew = 7 or moderately resistant; yellow rust = 9 or resistant; brown rust = 7 or moderately resistant; *Zymoseptoria* blotch = 6 or moderately resistant) (HGCA, 2010).

Tybalt: Zelder, Netherlands. First listed in 2003, Tybalt is a NABIM Group 2 variety (NABIM, 2010) with high yield potential relative to other spring varieties when sown in spring rather than late autumn. Tybalt accounted for about 24% of spring wheat supplies from the 2009 UK national harvest (HGCA 2010). It has weak straw and good disease resistance (especially to mildew and brown rust and average for yellow rust). Although weak strawed with a tendency to lodge, in organic systems because fertility is generally lower than in conventional systems, lodging is less of a risk. As a Group 2 variety, Tybalt's protein content and specific weight are usually lower than for Group 1 varieties such as Paragon, but baking performance is fair.

Fasan: Lochow Petkus, Germany. Fasan is a long straw variety bred specifically for high baking quality wheat production in Germany. Long strawed varieties such as Fasan and Zebra are more competitive against weeds in organic systems, may have a more vigorous root system and are able to exploit a greater volume of the soil profile to extract nutrients to greater depth than with less vigorously rooting varieties. In both Fasan and Zebra, the length of the peduncle (stalk bearing the ear) between the flag leaf's ligule and base of the ear is longer than in most varieties. This characteristic should afford some protection for the ear against foliar diseases infecting the flag leaf, such as *Zymoseptoria tritici*, transmitted upwards by rain splash, and also against fusarium-based ear diseases.

Monsun: a short-straw German variety bred for intensive farming systems.

Amaretto: Saatzuchtgesellschaft Streng's Amaretto is not listed in the UK but is on other European lists. Similar quality and yield characteristics to Paragon with good disease resistance.

Zebra: Swallof-Weibull, Sweden. Zebra is a long-straw Scandinavian variety bred specifically for high baking quality wheat production. The variety is popular in Norwegian organic systems for its good yield and is in high demand by Scandinavian mills and bakeries.

4.2.4 Compost and soil analysis

Prior to the trials in 2006 and 2007, 170 kg N ha^{-1} was applied in the form of composted farm yard manure (FYM) over the whole trial area, prior to ploughing out the grass/clover leys and drilling the plots. For this 45 tonnes of organic FYM compost was produced in each year from a single source (FYM removed from beef cattle sheds) at Sheepdrove Farm and turned at least once. Composts were analysed (Table 4.3) by Natural Resource Management Ltd, Berkshire. Correct quantities per plot were determined at Sheepdrove using a Keenan 'complete diet' feeder and delivered to the other two sites. A Millcreek model 57 type spreader was used and with a tractor forward speed of 8 km per h (5.0 mph) the correct application rate was achieved for an even distribution across the site. Trials were sown alongside 1st year wheat crops at all sites, according to the farm rotation (Table 4.1). All spring wheat varieties were grown in otherwise untreated plots.

Table 4.3 FYM compost analysis

		Amount / fresh t
Dry matter	53.2%	53.2 %
Total Nitrogen	1.7%	18.09 kg N
Total Phosphorus	0.315%	3.84 kg P_2O_5
Total Potassium	0.704%	4.5 kg K_2O
Total Copper	38.1 mg kg^{-1}	0.02 kg Cu
Total Zinc	178 mg kg^{-1}	0.09 kg Zn
Ammonium Nitrogen	63.3 mg kg^{-1}	0.03 kg NH_4-N
Nitrate Nitrogen	379 mg kg^{-1}	0.2 kg NO_3-N
Total Carbon	17.2%	
pH	7.35	

Soil nutrient levels for the beginning and end of each growing season are recorded in full for all three sites in the appendix 5 (Tables A4 and A5).

4.2.5 Crop establishment

Certified organic seed was sown at rates of 350 seeds m^{-2}, according to Thousand Grain Weight (TGW). Plots at Gilchesters were sown using an Accord 4 m combination drill with a Rabe power-harrow (Rabe Werk, Bad Essen, Germany). Plot weeding was carried out twice, 14 days apart weather permitting, with an Einbock comb-harrow (Table 4.5). Operation dates of trials in both years are given in Table 4.4.

Table 4.4 Field trial operation dates

	Site	
	Gilchesters	
Year	**2006**	**2007**
Drilling	10/04	02/4
1st weeding	12/05	26/04
2nd weeding	26/05	12/05
Harvest	30/8	12/9

4.2.6 Disease and leaf chlorophyll assessment protocols

Disease assessments for powdery mildew (*Blumeria (Erysiphe) graminis)*, Septoria (*Parastagonospora nodorum, Zymoseptoria tritici*) and wheat yellow rust (*Puccinia striiformis*) were made following the keys distributed by the Agricultural Development and Advisory Service (Anonymous, 1976) on all varieties at all three sites at GS 37 and 65 (Zadoks *et al.*, 1974). Disease levels were assessed as a percentage of leaf area affected firstly at GS37 according to a whole plant disease assessment and secondly at GS65 where assessments were made on the three youngest leaves: L1 (flag leaf), L2 (second leaf) and L3 (third leaf). For disease monitoring 20 tillers were assessed in each variety plot, by dividing plots into 4 50 x 50 cm sub-plots, within which 5 tillers were chosen randomly. Leaf greenness of upper leaves in all varieties was recorded by single-photon avalanche diode detection (SPAD) using a Minolta chlorophyll meter model SPAD 502 (Spectrum Technologies Inc., Plainfield, Illinois, USA). Ten measurements per plot were made on randomly selected flag leaves just after the start of flowering (GS65).

4.2.7 Harvest and grain sampling procedures

Crop yields were assessed using a plot-combine harvester (CLAAS Dominator 38, Germany) with a 2.4 m wide cutter bar. Grain moisture content was determined by drying grain at 80 °C for two days using a forced-draught drying oven (Genlab Ltd, Widnes,UK), with yield data presented at 15% moisture content. From each plot 60 kg of grain was retained for the various quality and baking tests. All grain analyses (protein content, HFN, gluten content and protein composition/quality) were carried out at Campden & Chorleywood Food Research Association (CCFRA).

4.2.8 Wheat quality assessments

Grain analysis for the characterisation of protein content of wheat samples was determined by NIR reflectance using a spectro-computer (Neotec model 6100 scanning monochromator interfaced to a PDP-11 computer). Hagberg Falling Number (HFN), to identify α-amylase activity in wheat, was determined by forming an aqueous suspension of flour and water followed by measurement in a Perten 1700 Falling Number instrument.

Protein composition/quality was undertaken by size exclusion high performance liquid chromatography (SE-HPLC). This separates and identifies the proportion of protein components which contribute to the total protein content of the wheat. The proportion of the large glutenin polymers is an important determinant of wheat end use quality as it is related to functional properties such as dough elasticity and strength (Singh, 2005). Size-exclusion (SE)-HPLC was used to determine changes in the glutenin polymer size distribution in the harvested grain samples (Dachkevitch and Autran, 1989; Singh *et al.*, 1990). This method uses sonication in SDS solution to solubilise all glutenin polymeric proteins and resolve them into five major fractions (F1-F5) according to their molecular size. F1 corresponds mainly to high molecular mass polymers, enriched in HMW subunits of glutenin; F2 corresponds mainly to low mass glutenin polymers, F3 and F4 mainly to the various classes of monomeric gliadin proteins, and F5 mainly to non-gluten proteins (albumins and globulins). The relative proportions of these can then be related to protein quality. Improved baking performance is generally associated with a higher proportion of the F1 fraction, high molecular weight glutenins.

This form of protein quality assessment was made following the Profilblé® method that was developed in France jointly by ARVALIS and INRA (Labuschagne and Aucamp, 2004). Flour (160 mg) was combined with 20 ml of 1% SDS in phosphate buffer, pH 6.9, to extract soluble wheat proteins. Following controlled sonication (Misonix Microson XL2000) and centrifugation, the supernatant was resolved by SE-HPLC analysis. This was performed using a Jasco HPLC system with a TSK gel G4000SW analytical column in conjunction with a TSK gel SW guard column. The chromatograms were integrated using methods provided by ARVALIS. All values are quoted as a percentage of the total response of the column, i.e. as a percentage of total protein. The response for each sample was normalised using the results from a control flour. Both daily and column specific checks were undertaken to ensure the linearity of response.

Gluten content and quality was also determined by Zeleny Sedimentation Value, where the degree of sedimentation of a flour suspended in a lactic acid solution during a standard time interval is taken as a measure of baking quality. Higher gluten content and better gluten quality both give rise to slower sedimentation and higher values.

Grain tocopherol concentrations (α-tocopherol, α-tocotrienol, β-tocopherol and β-tocotrienol) were measured for all varieties following freeze drying of grain in the Food Analysis Department of the Central Food Research Institute (CFRI, Herman Otto Vt 15, Budapest, Hungary). Grain macro- and micro-nutrient, heavy metals and phytate analysis for all varieties was undertaken at Sabbanci University, Turkey.

4.2.9 Baking quality assessments

Variety samples of wheat from the field trials were milled and baked by W&H Marriage using replicate samples of all six varieties to produce both white and wholemeal flour samples using a Chopin laboratory mill. The breads (wholemeal and white) were prepared and baked according to the standard Campden Baking Process (CBP). The dough was prepared by the company's technical baker using the following baker's formula: Flour 100%, Yeast 4%, Salt 1.5%, fungal alpha amylase 0.04% and water as required. Formulae are expressed in percentages, referred to as Baker's Percentage Method. The formula is then converted to kg and g, accordingly. When expressing formulae in the percentage system, the total flour requirement (100 g of flour) always represents 100 percent. Percentages of all other ingredients are based on the flour and are therefore relative to the amount of flour.

A planetary mixer was used for 3 minutes at speed 1 and 8 minutes at speed 2. The dough was given a standard 30 minute bulk fermentation following moulding into tins, with a 60 minute proving time. The dough was baked at 230 °C for 20-23 minutes. Further test bakes of stoneground and roller milled wholemeal and white Amaretto, Paragon and Tybalt flour were undertaken to compare their performance in these two milling processes.

4.2.10 Statistical analysis

Trial data analyses of variance from the 72 trial plots were derived from general linear models and linear mixed-effects models (Pinheiro, 2000). Fixed effects were season and variety. The effects of the fixed factors on yield, yield components, protein fractions, disease severity, grain macro and micro-nutrients and heavy metals and grain phytic acid content were determined. The analyses were carried out in the R statistical environment (R Development Core Team, 2012). The combined data for main effects were analysed first, and where interaction terms were significant, further analyses were conducted at each level of the interacting factor. Secondly main effects of season and variety were analysed and where interaction terms were significant, further analyses were conducted at each level of the interacting factor. Differences between significant main effect and interaction means were determined using Tukey's Honest Significant Difference (HSD) tests, based on mixed-effects models.

The relationships between wheat varieties and their grain yield, quality and nutrient contents with soil nutrient levels, environmental, and agronomic factors and site (geographical location) were investigated using redundancy analysis (RDA). In all cases the RDAs were carried out using the CANOCO 5 package. Automatic forward selection of the environment, site, soil nutrients and wheat variety within the RDAs was used and their significance in explaining additional variance calculated using Monte Carlo permutation tests.

4.3 Results

There was a marked difference in climatic conditions between the two growing seasons in 2006 and 2007. In 2006, May was wet and warm, June was warmer than normal, and July exceptionally so; both these months were dry and brighter than normal. This was followed by poor weather in August with wet conditions that were potentially detrimental to yield and grain quality, particularly when harvesting was delayed. In 2007, a very warm (record temperatures and sunshine hours in April) dry spring was followed by unsettled, wet weather from May until August. There were exceptional conditions in June and July 2007 which were extremely dull and wet. Rainfall exceeded 150% of average, leading to flooding in many parts of the country. Even in the absence of flooding, performance of cereal crops was adversely affected by increased levels of disease and harvest delays despite much improved conditions in August and September. Consequently, grain quality was very variable and lower than in 2006, resulting in severe restrictions on the availability of UK milling wheat (Defra, 2007). See Appendix 1 for full meteorological data.

4.3.1 Effect of year and variety of spring wheat on disease severity

The three main foliar diseases identified were Septoria (*Parastagonospora nodorum Zymoseptoria tritici*) and yellow rust (*Puccinia striiformis*) with only very low levels of powdery mildew (*Blumeria* (*Erysiphe*) *graminis*) (powdery mildew results not shown). Of the two Septoria diseases only results for *Z. tritici* are shown. For *Z. tritici* and yellow rust the disease levels in both years were low at GS 37 (mostly <1% infected leaf area). There was no significant effect of variety on *Z. tritici* severity but a significant difference in yellow rust disease levels (Table 4.5). Zebra had the highest yellow rust recorded at GS37 with an absence of disease on both Paragon and Tybalt. When the crops were again inspected at GS 65 for infection on the top three leaves (L1 flag leaf, L2 and L3) there was a significant difference between years for *Z. tritici* on L1 and for yellow rust on both L2 and L3. For *Z. tritici* the level of infection increased with leaf age, and for L3 was higher in 2007 (39.21%

leaf infection) than 2006 (7.9%) when averaged across all varieties. The varieties Fasan and Monsun had the highest levels of infection on L3 followed by Amaretto and Zebra (Table 4.5) but differences were not significant. The varieties Tybalt and Paragon had low levels of infection of *Z. tritici* on L3 when compared with the other varieties. For yellow rust disease at GS 65, Zebra showed the highest disease levels. The other 5 varieties showed much lower levels of yellow rust but in particular Paragon and Tybalt where very low levels of disease were detected on leaves L2 and L3 (Table 4.5 and 4.7 & 4.8).

For *Zymoseptoria tritici* and yellow rust significant interactions were detected between year and variety (Table 4.5). When interaction means were compared, levels of *Z. tritici* infection on L2 were generally low across both years with the exception of Monsun in 2007 at 28.7% (Table 4.6). Interestingly, although Zebra had the highest disease levels in 2006 it had zero *Septoria tritici* recorded in 2007 which was generally a much higher disease year than 2006.

For yellow rust at GS65, when interaction means were compared for leaf L2, the levels for Fasan were moderate in both years but highest for Zebra (30.0 and 83.7% in 2006 and 2007 respectively) (Table 4.7). On leaf L3 levels of yellow rust for Zebra (97%) were highest in 2007 (Table 4.8). Tybalt had no incidence of yellow rust recorded on L2 or L3 in either 2006 or 2007 with only very low levels recorded on the variety Paragon in 2007. So from the data recorded across both years the varieties Tybalt and Paragon showed the highest resistance to *Z. tritici* and yellow rust foliar disease.

Table 4.5 Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat disease severity (% infection) at GS 37 and GS65 for Gilchesters Farm

	Zymoseptoria tritici				**Yellow rust**			
	GS37 %	**GS65 Leaf L1 %**	**GS65 Leaf L2 %**	**GS65 Leaf L3 %**	**GS37 %**	**GS65 Leaf L1 %**	**GS65 Leaf L2 %**	**GS65 Leaf L3 %**
Year (YR)								
2006	0.0±0.00	0.0±0.00	2.3±0.75	7.9±1.56	0.0±0.01	0.0±0.03	7.3±2.72	3.5±1.68
2007	0.7±0.26	1.0±0.33	7.2±3.00	39.2±6.46	2.9±0.81	0.0±0.00	18.2±6.3	17.4±7.43
Variety (VR)								
Amaretto	0.5±0.38	0.0±0.00**b**	2.2±0.84**b**	26.2±9.81	1.0±0.63**bc**	0.1±0.08	4.0±1.04**c**	2.2±0.64**b**
Fasan	0.7±0.62	0.3±0.18**b**	4.8±1.27**b**	34.3±11.97	2.3±1.27**ab**	0.0±0.00	10.6±2.2**b**	3.7±1.31**b**
Monsun	0.5±0.37	2.1±0.88**a**	16±8.43**a**	31.4±11.79	1.1±0.61**bc**	0.0±0.00	5.0±1.55**c**	2.0±0.68**b**
Paragon	0.3±0.25	0.1±0.13**b**	1.1±0.64**c**	10.8±4.67	0.0±0.00**c**	0.0±0.00	0.1±0.13**d**	0.1±0.13**c**
Tybalt	0.0±0.06	0.5±0.19**b**	1.2±0.62**c**	7.4±3.51	0.0±0.00**c**	0.0±0.00	0.0±0.00**d**	0.0±0.00**c**
Zebra	0.0±0.01	0.0±0.00**b**	3.3±1.57**b**	21.2±3.15	4.4±1.97**a**	0.0±0.00	56.8±11.7**a**	55±16.42**a**
ANOVA								
Main Effects								
YR	ns	**0.0048**	ns	ns	0.06	ns	**0.0176**	**0.0034**
VR	ns	**<0.001**	**0.0251**	ns	**<0.001**	ns	**<0.001**	**<0.001**
Interactions								
YR:VR	ns	**<0.001**	**0.0164**[1]	ns	**<0.001**	ns	**<0.001**[2]	**<0.001**[3]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 4.6. [2]For interaction means see Table 4.7. [3]For interaction means see Table 4.8

Table 4.6 Interaction means for effects of year and wheat variety on Zymoseptoria % (leaf L2) at GS 65

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	0.7±0.40**c**	3.±2.1**b**	3.2±2.20**b**	0.0±0.00**d**	0.0±0.00**d**	6.7±1.90**a**
2007	3.7±1.25**c**	6.2±1.25**b**	28.7±14.70**a**	2.2±1.03**c**	2.5±0.80**c**	0.0±0.00**d**

Table 4.7 Interaction means for effects of year and wheat variety on yellow rust % (Leaf L2) at GS 65

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	3.1±1.10**c**	7.5±1.40**b**	3.2±1.03**c**	0.0±0.00**d**	0.0±0.00**d**	30.0±10.80**a**
2007	5.0±1.70**c**	13.7±3.70**b**	6.7±2.80**c**	0.2±0.20**d**	0.0±0.00**d**	83.7±6.50**a**

Table 4.8 Interaction means for effects of year and wheat variety on yellow rust % (Leaf L3) at GS 65

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	2.0±1.06**b**	4.2±2.60**b**	2.2±0.90**b**	0.0±0.00**c**	0.0±0.00**c**	13±9.02**a**
2007	2.5±0.80**b**	3.2±1.03**b**	1.7±1.10**b**	0.2±0.20**c**	0.0±0.00**c**	97±1.10**a**

For each table, means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

4.3.2 Effects of year and variety of spring wheat on crop performance and grain yield

For **grain yield**, analysis detected significant main effects of year and variety. Grain yields were significantly higher in 2006 at 6.29 t ha^{-1} compared to 2.13 t ha^{-1} in the following year, highest with Paragon (4.7 t ha^{-1}) and lowest with Zebra (3.2 t ha^{-1}) (Table 4.9). The highest recorded yields of the whole trial were in 2006 for Amaretto with 6.7 t ha^{-1}. No significant interactions were detected between year and variety due to the consistent reduction in yield for all varieties from 2006 to 2007 (Table 4.13).

For **plant height** significant differences were detected for year and variety. All varieties were significantly taller in 2006 (average of 97 cm across all varieties). Fasan and Zebra were the tallest varieties (102 cm), Amaretto (94 cm) and Paragon (92 cm) were intermediate, Monsun (89 cm) with Tybalt the shortest (81 cm) when averaged across both seasons (Table 4.9). There was a significant year x variety interaction for plant height ($p = 0.0274$) in that Zebra was the tallest variety in 2006 while Fasan was the tallest variety in 2007. Tybalt was the shortest variety across both seasons (Table 4.10).

For **leaf greenness/chlorophyll**, SPAD values at GS65 detected significant main effects for year and variety. Chlorophyll content was much higher in 2006, highest in Paragon and lowest in Fasan and Zebra (Table 4.9). Significant interactions were detected between year and variety (Table 4.11). When interaction means between year and variety were compared, levels were significantly lower in Zebra in 2007. Amaretto had the highest SPAD levels in 2006 followed by Paragon, Tybalt and Monsun, whereas in 2007 the highest SPAD levels were recorded in Paragon and Tybalt. Zebra had the lowest SPAD levels in both years, most likely due to the higher levels of disease recorded on this variety in both years (Table 4.11).

For **Thousand Grain Weight** analysis also detected significant main effects for year and variety. Grain weights were higher in 2006 with 43.3 g compared to 39.53 g in 2007 (Table 4.9), highest for Tybalt (43.63 g) and Monsun (43.44 g), intermediate with Fasan and Paragon (42.38 and 41.98 g respectively) and lowest with Amaretto and Zebra (38.5 and 39.22 g respectively). The varieties displayed a consistency in grain weight despite the strong seasonal variation in growing conditions each year, with the exception of Zebra and Amaretto which produced the smallest grains (TGW < 40 g) when compared with the other varieties (Table 4.9). There was a significant year x variety interaction on TGW (p=0.0059) with no significant difference between varieties in 2006 but in 2007 the TGW of Amaretto and Zebra was significantly lower than the other 4 varieties (Table 4.12).

Table 4.9 Effect means ± SE and *p* values for the effects of harvest year and variety on wheat growth and yield parameters from Gilchesters Farm

	Grain yield	**Plant Height**	**SPAD**	**TGW**
	t ha^{-1}	cm	GS65	g
Year (YR)				
2006	6.3±0.16	97.0±1.07	52.7±0.52	43.3±0.07
2007	2.1±0.18	88.9±2.09	38.5±1.68	39.5±1.36
Variety (VR)				
Amaretto	4.3±0.93**ab**	94.4±3.06**b**	48.0±2.84**a**	38.5±1.63**c**
Fasan	4.4±0.73**ab**	102±2.92**a**	43.3±2.99**b**	42.3±0.63**abc**
Monsun	4.1±0.78**b**	92.4±2.45**b**	46.8±2.32**ab**	43.4±1.63**ab**
Paragon	4.7±0.83**a**	92.4±3.05**b**	49.8±1.96**a**	41.9±1.06**abc**
Tybalt	4.3±0.77**ab**	81.1±2.91**c**	49.1±1.93**a**	43.6±1.18**a**
Zebra	3.2±0.88 **c**	102.7±2.76**a**	36.4±5.05**c**	39.2±3.66**bc**
ANOVA				
Main Effects				
YR	**0.002**	**0.0078**	**0.0004**	**0.0002**
VR	**<0.001**	**<0.001**	**<0.001**	**0.0159**
Interactions				
YR:VR	ns	**0.0274**[1]	**<0.001**[2]	**0.0059**[3]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 4.10. [2]For interaction means see Table 4.11. [3]For interaction means see Table 4.12.

Table 4.10 Interaction means for effects of year and wheat variety on plant height.

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	101.4±2.52**b**	102.6±5.70**ab**	97.1±1.73**b**	99.7±0.60**b**	88.0±2.00**c**	108.1±1.58**a**
2007	87.4±2.20**b**	101.4±2.66**a**	87.8±3.28**b**	85.2±2.85**c**	74.2±1.97**b**	97.2±3.6**a**

Table 4.11 Interaction means for effects of year and wheat variety on SPAD at GS65.

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	55.2±0.60**a**	50.8±1.20**bc**	52.8±0.62**abc**	54.1±0.67**ab**	53.7±1.09**ab**	49.4±0.80**c**
2007	40.9±1.77**b**	35.8±1.66**c**	40.7±0.73**b**	45.4±2.20**a**	44.5±1.50**a**	23.4±2.33**d**

Table 4.12 Interaction means for effects of year and wheat variety on thousand grain weight.

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	41.2±1.18**a**	42.8±0.89**a**	44.1±2.18**a**	42.3±1.90**a**	44.9±1.04**a**	44.4±2.30**a**
2007	34.8±2.01**a**	41.9±0.98**b**	42.4±2.88**b**	41.5±0.78**b**	42.3±2.09**b**	32.3±6.43**a**

Table 4.13 Means and SE for effects of year and wheat variety on grain yield (t ha^{-1}).

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	6.7±0.44**a**	6.3±0.35**a**	6.0±0.40**a**	6.8±0.15**a**	6.2±0.40**a**	5.5±0.23**b**
2007	1.9±0.32**b**	2.6±0.26**a**	2.1±0.36**ab**	2.6±0.57**a**	2.4±0.31**ab**	0.8±0.08**c**

For each table means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

4.3.3 Effects on year and spring wheat variety on grain quality

For **protein quality** there were significant effects of both year and variety on protein quality determined by SE-HPLC (Table 4.14). Protein quality, associated with a higher proportion of the F1 fraction i.e. high molecular weight glutenins, was higher in 2007 (3.76%) than in 2006 (3.42%) and significantly higher in the varieties Fasan and Tybalt than the other 4 varieties when averaged across both seasons.

For **grain gluten quality** by sedimentation value (Zeleny) quality was much higher in 2006 (65.13 ml) than in 2007 (49.91 ml). The highest gluten quality was found in Paragon, with the lowest quality in Amaretto (Table 4.14). There was no significant year x variety interaction on gluten quality.

The **Hagberg falling number** (HFN) was much higher (better) in 2007 (380.9 s) than in 2006 (310.9 s) (Table 4.14) despite the adverse weather conditions, i.e. increased rainfall during crop maturation, in the second growing season. The HFN of all varieties was over 300, with Fasan, Paragon and Tybalt being significantly higher than Amaretto, Monsun and Zebra (Table 4.14). There was a significant year x variety interaction (p<0.001) on HFN. Zebra had the highest HFN in 2006 (347 s) but had the lowest level in 2007 (291.6 s), although this was well above the UK milling wheat standard of >250 s (NABIM 2018). Amaretto had the lowest HFN in 2006 (266.5 s) but the fourth highest in 2007 (356 s) emphasising the clear differences between the varieties in HFN across the two seasons (Table 4.16).

For **grain crude protein** there were significant differences for variety only: highest with Zebra (15.06%) and Tybalt (14.98%), intermediate with Paragon (14.65%) and Fasan (14.45%) and lowest with Amaretto (13.75%) and Monsun (14.08%) (Table 4.14). Significant interactions were detected between year and variety (Table 4.15). Fasan had the highest protein content in 2006 (15.17%) whereas Zebra had the highest level in 2007 (15.94%) which was significantly higher than all other varieties. The variety Amaretto had the lowest recorded protein content in both seasons.

For **specific weight** analysis detected significant main effects for year and variety. Specific weight was higher in 2006 (80.38 kg hl^{-1}) than in 2007 (74.45 kg hl^{-1}). Fasan had grains with the highest level (80.35 kg hl^{-1}), Amaretto, Monsun and Paragon were intermediate (77-78 kg hl^{-1}) and the lowest were Zebra and Tybalt (76.2 and 74.8 kg hl^{-1} respectively) when averaged across both years (Table 4.14). All varieties produced consistently high specific weights when averaged across both seasons and higher than the UK milling standard for bread wheat of >76 kg hl^{-1} (NABIM 2018) with the exception of the variety Tybalt (74.79 kg hl^{-1}). There was a significant year x variety interaction on specific weight (Table 4.17). Fasan had the highest specific weight in both years but in 2007 it was significantly higher than all other varieties whereas in 2006 it was not significantly different from both Paragon and Amaretto. In both years Tybalt and Zebra had significantly lower specific weights than the remaining four varieties with the specific weight of Zebra in 2007 being particularly low at 69.5 kg hl^{-1}.

Table 4.14 Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat quality at Gilchesters Farm

	Protein Quality SE-HPLC	**Crude Protein**	**Gluten Quality (Zeleny)**	**Hagberg Falling Number**	**Specific weight**
	Fractions F1/(F3+F4) **%**	**%**	ml	s	kg hl^{-1}
Year (YR)					
2006	3.4±0.04	14.4±0.12	65.1±1.35	310.9±8.4	80.3±0.24
2007	3.7±0.05	14.4±0.23	49.9±1.82	380.8±17.3	74.4±0.86
Variety (VR)					
Amaretto	3.4±0.06**b**	13.7±0.17**c**	49.1±4.83**b**	311.2±18.92**b**	77.9±1.46**b**
Fasan	3.6±0.13**a**	14.4±0.31**abc**	58.1±3.07**ab**	381.8±16.84**a**	80.3±0.54**a**
Monsun	3.5±0.05**a**	14.0±0.3**bc**	59.3±3.45**a**	302.6±21.32**b**	77.3±1.32**b**
Paragon	3.4±0.09**b**	14.6±0.1**ab**	64.8±2.26**a**	376.5±29.25**a**	78.9±0.98**b**
Tybalt	3.5±0.06**a**	14.9±0.27**a**	61.5±2.74**a**	371.8±33.27**a**	74.8±1.77**c**
Zebra	3.4±0.07**b**	15.0±0.36**a**	54.5±4.63**b**	323.4±23.76**b**	76.1±2.13**c**
ANOVA					
Main Effects					
YR	**0.0013**	0.975	**0.0029**	**0.0244**	**0.0143**
VR	**<0.001**	**<0.001**	**0.001**	**0.0004**	**<0.001**
Interactions					
YR:VR	ns	**<0.001**[1]	ns	**0.0001**[2]	**0.001**[3]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1]For interaction means see Table 4.15. [2]For interaction means see Table 4.16. [3]For interaction means see Table 4.17.

Table 4.15 Interaction means for effects of year and wheat variety on crude protein %.

Year	**AMA**	**FAS**	**MON**	**PAR**	**TYB**	**ZEB**
2006	14.1±0.16**b**	15.1±0.22**a**	14.3±0.45**b**	14.4±0.08**b**	14.4±0.24**b**	14.3±0.28**b**
2007	13.4±0.15**d**	13.7±0.20**c**	13.8±0.42**c**	14.9±0.03**c**	15.5±0.25**b**	15.9±0.18**a**

Table 4.16 Interaction means for effects of year and wheat variety on grain Hagberg Falling Number.

Year	**AMA**	**FAS**	**MON**	**PAR**	**TYB**	**ZEB**
2006	266.5±10.83**c**	344±12.40**a**	301±12.70**b**	321±22.70**ab**	285±9.43**b**	347±21.24**a**
2007	356±14.76**c**	419.7±14.65**b**	304.2±44.20**d**	450±15.30**a**	458.5±8.65**a**	291.6±46.56**d**

Table 4.17 Interaction means for effects of year and wheat variety on grain specific weight.

Year	**AMA**	**FAS**	**MON**	**PAR**	**TYB**	**ZEB**
2006	81.4±0.10**a**	81.6±0.34**ab**	80.2±0.37**b**	80.9±0.16**ab**	78.7±0.34**c**	79.5±0.34**c**
2007	74.4±1.34**b**	79.1±0.47**a**	74.4±1.58**b**	76.3±0.83**b**	70.8±2.06**c**	69.5±1.10**c**

For each table, means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

From the **vitamin E groups** grains were analysed for α- and β-tocopherols and α- and β-tocotrienols only. For α-tocopherols there was no significant difference in concentrations between the growing seasons although levels were higher in 2006 than 2007 (Table 4.18). The varieties Zebra, Amaretto and Paragon had the highest grain concentrations and Fasan and Monsun the lowest. There was a significant year x variety interaction (p=0.038) for α-tocopherol concentration. Zebra had the highest concentration in 2006 (14.3 μg g^{-1}) while

Paragon had the highest concentration in 2007 (11.67 µg g^{-1}). Monsun α-tocopherol concentration was significantly lower than all other varieties in 2007 (8.36 µg g^{-1}) but in 2006 Fasan was significantly lower than all other varieties (10.02 µg g^{-1}) (Table 4.19). There were no significant differences in season or variety for α- tocotrienol. For both β-tocopherols and β-tocotrienols concentrations there were significant effects of both year and variety. β-Tocopherol concentrations in 2006 were double the level in 2007, with Amaretto producing the highest concentration and Fasan and Monsun the lowest. There was a significant year x variety interaction (p=0.0016) for β-tocopherol concentration (Table 4.20). Amaretto had the highest concentration in 2006 (10.56 µg g^{-1}) while Zebra and Paragon had the highest concentrations in 2007 (8.35 and 8.36 µg g^{-1} respectively). Fasan had the lowest concentration in 2006 (5.61 µg g^{-1}) while Monsun had the lowest concentration in 2007 (2.93 µg g^{-1}). β- Tocotrienol concentrations were significantly, higher in 2006 than 2007 and were higher in Paragon and Tybalt than all other varieties. β- Tocotrienols were lowest in Monsun (19.43 µg g^{-1}).

Table 4.18 Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat quality at Gilchesters Farm

	α-tocopherol	**α-tocotrienol**	**β-tocopherol**	**β-tocotrienol**
	µg g^{-1}	µg g^{-1}	µg g^{-1}	µg g^{-1}
Year (YR)				
2006	12.4±0.36	3.0±0.11	7.8±0.37	26.3±0.59
2007	10.5±0.41	3.1±0.17	3.5±0.22	15.2±0.66
Variety (VR)				
Amaretto	12.7±0.56**a**	3.1±0.18	7.3±1.35**a**	19.6±1.98**cd**
Fasan	10.3±0.65**c**	3.0±0.35	4.4±0.50**c**	19.9±1.72**cd**
Monsun	9.7±0.69**c**	3.1±0.24	4.7±0.68**c**	19.4±2.34**d**
Paragon	12.5±0.40**ab**	3.2±0.17	6.6±0.88**ab**	23±2.71**abc**
Tybalt	11.2±0.48**bc**	2.7±0.11	5.3±0.83**bc**	23.2±2.44**a**
Zebra	12.9±0.85**a**	3.3±0.30	6.2±1.07**b**	20.9±2.91**bc**
ANOVA				
Main Effects				
YR	0.0566	ns	**0.0004**	**0.0025**
VR	**<0.0001**	ns	**<0.0001**	**0.0225**
Interactions				
YR:VR	**0.0348**[1]	ns	**0.0016**[2]	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 4.19. [2]For interaction means see Table 4.20.

Table 4.19 Interaction means for effects of year and wheat variety on α-tocopherol.

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	13.9±0.47**ab**	10.0±0.1**d**	11.0±0.56**c**	13.1±0.46**b**	12.2±0.40**c**	14.3±0.60**a**
2007	11.5±0.55**a**	10.6±1.37**bc**	8.3±0.81**d**	11.6±0.35**a**	10.1±0.44**c**	11.2±1.3**ab**

Table 4.20 Interaction means for effects of year and wheat variety on β-tocopherol.

Year	AMA	FAS	MON	PAR	TYB	ZEB
2006	10.5±0.55**a**	5.6±0.42**e**	6.4±0.1**d**	8.3±0.57**b**	7.4±0.32**c**	8.3±0.57**b**
2007	4.0±1.04**c**	3.2±0.22**c**	2.9±0.19**c**	8.3±0.57**a**	7.4±0.32**b**	8.3±0.57**a**

For each table, means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Grain macro nutrient concentrations tested for phosphorus (P), potassium (K), sulphur (S) and calcium (Ca) only showed significant differences between years for K and Ca. Grain K ad Ca concentrations were both significantly higher in 2007 than 2006 (Table 4.21). There was no significant difference between varieties in grain macronutrient concentrations and also no significant year x variety interaction.

For concentrations of micro nutrients **zinc (Zn) and iron (Fe)** in wheat grain there were significant differences between varieties (Table 4.22). Although both Fe and Zn concentrations were higher in 2006 than 2007 it was only for Fe that the difference between years was statistically significant. Both Fe and Zn concentrations were highest in the variety Tybalt.

Significant interactions were detected between year and variety for Zn ($p=0.0079$) and Fe ($p<0.001$) (Table 4.22). Interaction means for year and variety for Zn (Table 4.23) identified that Tybalt and Zebra had significantly higher concentrations than all other varieties in 2007 with Fasan having the lowest concentration while in 2006 Tybalt had the highest concentration. For Fe concentrations Zebra was significantly lower than all other varieties in 2006 but in 2007 Zebra and Tybalt had significantly higher Fe concentrations than all other varieties (Table 4.24).

Table 4.21 Main effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat grain macro nutrient content.

	Grain P	**Grain K**	**Grain S**	**Grain Ca**
	mg g^{-1}	mg g^{-1}	mg g^{-1}	mg g^{-1}
Year (YR)				
2006	3.2±0.03	4.0±0.04	1.5±0.02	0.2±0.01
2007	3.5±0.17	5.3±0.30	1.3±0.07	0.4±0.03
Variety (VR)				
Amaretto	3.3±0.12	4.7±0.33	1.3±0.03	0.3±0.04
Fasan	3.4±0.07	4.4±0.23	1.4±0.06	0.4±0.04
Monsun	3.4±0.07	4.7±0.38	1.3±0.03	0.3±0.04
Paragon	3.0±0.13	4.7±0.27	1.4±0.02	0.3±0.03
Tybalt	3.5±0.15	4.9±0.37	1.5±0.02	0.3±0.03
Zebra	3.1±0.45	4.2±0.80	1.4±0.20	0.3±0.07
ANOVA				
Main Effects				
YR	ns	**0.0239**	ns	**0.0118**
VR	ns	ns	ns	ns
Interactions				
YR:VR	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 4.22 Main effect means ± SE and *p* values for the effects of harvest year and variety on wheat grain micro nutrient content

	Zn	**Fe**
	mg kg^{-1}	mg kg^{-1}
Year		
2006	35.3±0.89	41.5±0.74
2007	31.2±0.73	34.3±0.91
Variety (VR)		
Amaretto	31.9±1.29**b**	37.4±1.91**b**
Fasan	31.8±1.69**b**	35.7±2.27**b**
Monsun	33.5±1.73**ab**	37.9±2.46**b**
Paragon	31.7±1.01**b**	37.6±1.50**b**
Tybalt	36.2±1.89**a**	41.5±1.65**a**
Zebra	34.9±1.45**a**	38.1±1.02**b**
ANOVA		
Main Effects		
YR	ns	**0.0495**
VR	**0.0045**	**0.0001**
Interactions		
YR:VR	**0.0079**	**<0.0001**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$).

Table 4.23 Interaction means for effects of year and wheat variety on grain zinc (Zn) concentrations

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	34.0±0.16**b**	35.5±1.74**ab**	36.9±2.23**a**	32.7±1.61**c**	38.6±3.18**a**	34.1±2.32**b**
2007	29.7±1.37**bc**	28.0±1.03**c**	30.1±1.06**b**	30.4±0.72**b**	33.7±1.61**a**	35.9±1.72**a**

Table 4.24 Interaction means for effects of year and wheat variety on iron (Fe) concentrations

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	41.5±1.35**a**	41.3±0.80**a**	43.0±2.50**a**	40.4±0.70**a**	44.5±2.00**a**	38.0±1.80**b**
2007	33.4±2.08**b**	30.1±1.50**c**	32.8±2.00**bc**	33.7±1.36**b**	38.5±1.68**a**	38.2±0.40**a**

For each table, means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Grain **heavy metal concentrations** analysis detected significant main effects of year on all heavy metal concentrations, with grain concentrations of molybdenum, aluminium and lead being higher in 2007 and cadmium and nickel higher in 2006. For varieties there were significant effects for both molybdenum and cadmium. Concentrations of Mo were highest in Paragon and Fasan, and for Cd highest in Zebra and Monsun (Table 4.25). Significant interactions were detected between year and variety (Table 4.26), which identified that for Mo concentrations were higher in 2007 for all varieties.

Table 4.25 Main effect means ± SE and *p* values for the effects of harvest year and variety on wheat grain heavy metal concentrations

	Mo	Al	Cd	Ni	Pb
	µg kg^{-1}	µg kg^{-1}	µg kg^{-1}	µg kg^{-1}	µg kg^{-1}
Year (YR)					
2006	0.0±0.01	2.2±0.10	57.0±1.99	244.2±16	50.1±6.93
2007	0.3±0.03	3.3±0.27	30.1±1.39	155.7±6.97	134.7±12.91
Variety (VR)					
Amaretto	0.1±0.05**c**	2.5±0.20	39.2±6.08**c**	201.8±19.50	87.0±26.58
Fasan	0.2±0.06**a**	2.9±0.40	41.2±4.45**bc**	181.2±22.52	109.8±20.57
Monsun	0.1±0.05**bc**	2.5±0.33	51.3±5.99**a**	191.3±21.53	93.0±22.06
Paragon	0.2±0.08**a**	2.2±0.31	36.6±5.06**c**	171.4±22.7	79.6±18.94
Tybalt	0.1±0.06**ab**	2.4±0.20	46.4±6.35**ab**	229.9±37.03	89.5±27.30
Zebra	0.1±0.07**ab**	3.5±0.70	50.0±6.09**a**	235.9±36.01	82.0±28.40
ANOVA					
Main Effects					
YR	**0.0437**	**0.0223**	**0.0008**	**0.0305**	**0.0217**
VR	**0.0008**	ns	**0.0004**	ns	ns
Interactions					
YR:VR	**0.0143**	**0.0168**	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1]See Table 4.26 for interaction means SE.

Table 4.26 Interaction means for effects of year and wheat variety on grain molybdenum (Mo) concentrations.

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	0.0±0.01**c**	0.0±0.01**ab**	0.0±0.00**a**	0.0±0.02**a**	0.0±0.01**ab**	0.0±0.00**c**
2007	0.2±0.06**b**	0.3±0.07**a**	0.2±0.06**b**	0.3±0.10**a**	0.3±0.07**ab**	0.3±0.07**a**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

For the assessment of **phytic acid levels** in the grain along with the concentrations of phosphorus (P) and the molar ratios of zinc (Zn) and iron (Fe) there were significant effects of variety only (Table 4.27). Fasan was the variety with the highest levels of phytic acid and phytic acid P concentrations and with the highest phytic acid Zn and Fe molar ratios. Tybalt had the lowest levels of phytic acid and phytic acid P together with phytic acid Zn and Fe molar ratios (Table 4.27).

For year and variety interactions (Tables 4.28 and 4.29) significant interactions were detected for both Zn and Fe molar ratios. Fasan was the variety with the highest levels of each in 2007 and Tybalt the lowest of each in 2006.

Table 4.27 Main effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat grain phytic acid concentrations and molar ratios.

	Phytic acid	Phytic acid P	Phytic acid Zn molar ratio	Phytic acid Fe molar ratio
	mg g^{-1}	mg g^{-1}	%	%
Year (YR)				
2006	9.6±0.17	2.7±0.05	27.4±0.91	19.8±0.58
2007	9.5±0.22	2.7±0.06	30.6±0.97	23.9±1.04
Variety (VR)				
Amaretto	8.9±0.17**c**	2.5±0.05**c**	28.1±0.87**b**	20.6±1.03**bc**
Fasan	10.3±0.28**a**	2.9±0.08**a**	32.9±1.92**a**	25.4±2.05**a**
Monsun	9.3±0.28**c**	2.6±0.08**c**	27.9±1.47**b**	21.5±1.76**bc**
Paragon	10.1±0.26**ab**	2.8±0.07**ab**	31.8±1.02**a**	23.1±1.38**ab**
Tybalt	9.2±0.20**c**	2.6±0.06**c**	25.8±1.52**b**	19.1±1.08**c**
Zebra	9.5±0.50**bc**	2.6±0.14**bc**	27.3±1.98**b**	21.3±1.42**bc**
ANOVA				
Main Effects				
YR	ns	ns	ns	ns
VR	**0.0008**	**0.0008**	**0.0001**	**<0.0001**
Interactions				
YR:VR	ns	ns	**0.0064**[1]	**0.0004**[2]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1]For interaction means see Table 4.28. [2]For interaction means see Table 4.29.

Table 4.28 Interaction means for effects of year and wheat variety on phytic acid Zn molar ratios.

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	26.4±1.3**b**	29.3±1.2**a**	25.5±2.26**b**	30.3±0.91**a**	23.9±2.27**c**	29.2±3.3**a**
2007	29.7±0.21**b**	36.5±2.2**a**	30.3±1.11**b**	33.9±1.35**a**	27.7±1.78**c**	24.9±0.77**d**

Table 4.29 Interaction means for effects of year and wheat variety on phytic acid Fe molar ratios.

Year	Amaretto	Fasan	Monsun	Paragon	Tybalt	Zebra
2006	18.4±0.5**b**	21.4±0.7**a**	18.7±1.6**b**	20.8±0.1**a**	17.4±1.1**b**	22.2±2.2**a**
2007	22.7±1.2**c**	29.4±2.9**a**	24.2±2.5**bcd**	26.3±2.1**ab**	20.7±1.5**d**	20.0±1.6**d**

For each table, means labelled with the same letter within the same row for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

4.3.4 Effects of variety and flour processing on baking quality

For the **variety baking tests** single loaves were produced. All samples produced doughs which handled well and had good proof times, but none of the wholemeal loaves were found to be outstanding. There were inconsistencies in performance in the batches but reasonable volume and crumb structure were produced from the varieties Amaretto, Paragon and Fasan. There was a clear difference in the performance of these varieties in wholemeal bread loaf volume from the much poorer performance of Monsun, Tybalt and Zebra (Fig. 4.1). For the loaves baked from white flour, samples with the best loaf volumes were produced from the varieties Paragon and Zebra (Fig. 4.2).

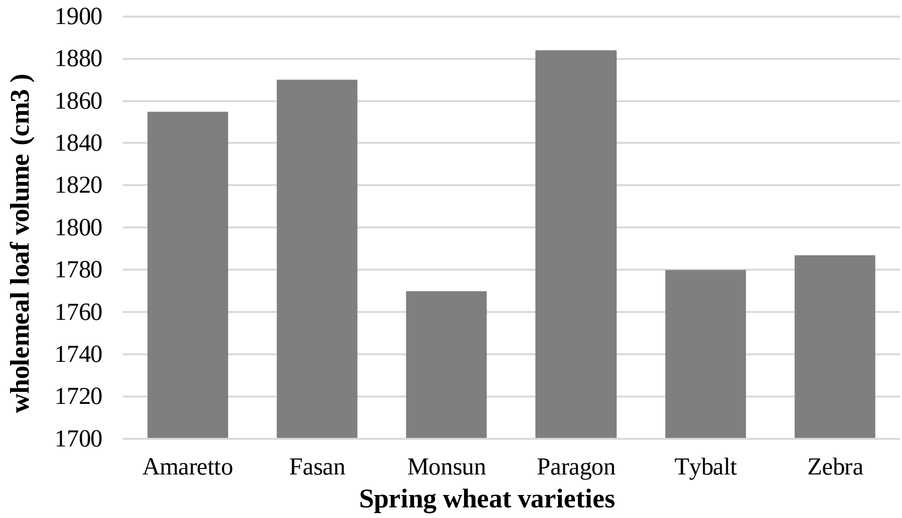


Fig. 4.1 Effect of spring wheat variety on wholemeal loaf volume 2006, single loaf

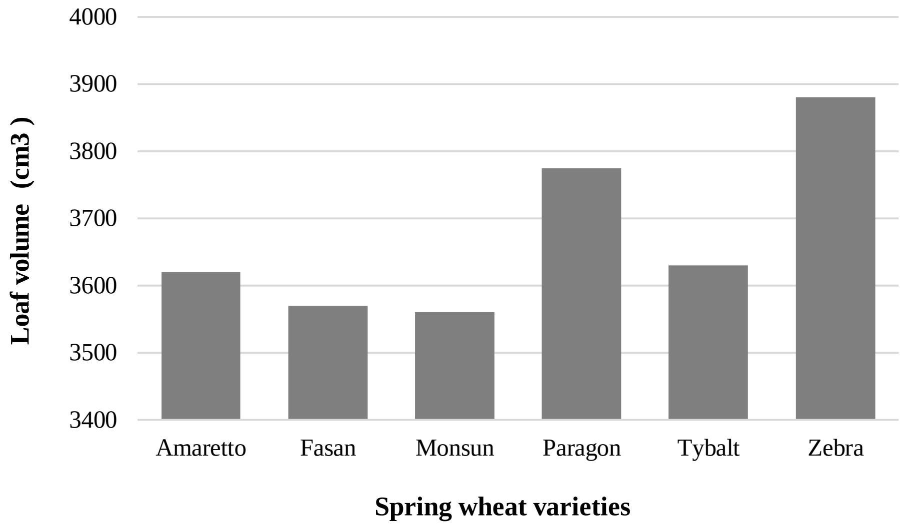


Fig. 4.2 Effect of spring wheat variety on white loaf volume 2006, single loaf

When the results of protein analysis by SE-HPLC (Table 4.14) were compared to the final loaves, protein quality data represented as a function of F1/(F3+F4) showed that an increase in the proportion of the F1 high molecular weight glutenins was only weakly associated with an increase in loaf volume.

For stoneground versus roller milled flour, test bakes were conducted on Amaretto, Paragon and Tybalt and are presented in Table 4.30. For **roller milled flour** both Amaretto and Tybalt produced good doughs, experiencing no problems with good water absorption of 64%. The Paragon had a good feel, also taking water at 64%, but after 30 minutes bulk fermentation the dough went soft and sticky. The Paragon dough felt very weak and started to feel as though it was breaking whilst moulding, whereas the Amaretto and Tybalt did not experience such problems. The proof was poor on the Paragon with the dough only growing to the height of 10.5 cm. The Tybalt was also disappointing as this only achieved a height of 11.5cm and collapsed as soon as it was lifted from the prover prior to being placed in the oven. The Amaretto seemed the best dough, reaching 12.0 cm after proving, and held no problems with transporting to the oven. The Amaretto did finish with a baked height of 9.5 cm, a drop back of 2.5 cm, which was disappointing. The Tybalt was very poor with a height of 7.2 cm, but this was to be expected due to collapsing. The Paragon finished with a baked height of 9.4 cm, from a back drop of 1-1.5 cm from proof height, which was the best of all the varieties. The appearance of the crumb texture and crust colour was difficult to judge on all the roller milled breads due to the poor volume. The crust was pale due to the loaf height being lower than the tin height, which is as expected.

For the **stoneground** samples, the flours had the same water absorption as the roller milled. Again the Paragon had the same problem with the dough after 30 minutes bulk fermentation. Both the Amaretto and Tybalt proved to 12.0 cm with no problems, whilst the Paragon could only attain 10.5 cm and appeared to be running out of gas as it was starting to break with holes appearing at the top of the dough. The bake was slightly better on the Amaretto and Tybalt than their equivalent roller milled flours, as these tended to hold a better finished baked height of 10.0 cm, with a loss of 2.0 cm, whereas the Paragon finished bake height was 9.0 cm, a loss of 1.5 cm. As with the roller milled the judging of the crumb and crust was difficult due to the lack of volume.

Table 4.30 Flour analysis, dough characteristics and final loaf test bake results from roller milled and stone ground samples of Amaretto, Paragon and Tybalt from 2006 harvest

		Amaretto		**Tybalt**		**Paragon**	
Flour		**Roller**	**Stoneground**	**Roller**	**Stoneground**	**Roller**	**Stoneground**
Protein	%	10.3	10.8	10.4	11.4	9.9	11.1
Moisture	%	13.7	13	14.4	13	13.6	13.6
Water Abs.	%	64	64	64	64	64	64
Dough Temp.	^{o}C	27.2	27.6	27.6	27.8	26.9	26.7
Proof time	min	50	45	50	45	40	40
Proof height	cm	12	12	11.5*	12	10.5	10.5
Baked height		9.5	10	7.2	10	9.4	9
Loaf volume	cm^3	1830	1868	1495	1830	1740	1680
Crust colour		Pale	OK	Pale	OK	Poor	Poor
Texture		Open	Open	Close & crumbly	Close	Close	Close

*Tybalt roller milled loaves collapsed as soon as they were moved from the prover whilst being placed in the oven

Amaretto **Tybalt** **Amaretto** **Tybalt**

30 min bake roller milled **30 min bake stone ground**

Fig. 4.3 Test baked loaves of white (top) and wholemeal (bottom) bread made with roller milled and stoneground Amaretto and Tybalt flour

Paragon white **wholemeal** **Paragon wholemeal** **white**

30 min bake from roller milled **30 min bake from stoneground**

Fig. 4.4 Test baked loaves of white (outer) and wholemeal (inner) bread made with roller milled and stoneground Paragon flour

4.4 Discussion

A primary requirement of the artisan bakery industry is to secure locally cultivated organic wheat flour of consistently good baking quality. Variations in wheat quality arising from the effects of the weather on different varieties in different growing regions is therefore a major problem (Johansson and Svensson, 1999). Results from this study identified that from the considerably different weather conditions between the two years with more sunshine, higher temperatures and less rainfall in 2006, all six spring wheat varieties in 2006 had taller plants with greener leaves, lower disease pressure, higher vitamin E levels, a higher yield, bigger grains, better protein quality, lower protein concentrations, lower grain macro nutrient concentrations but with no effect on phytic acid levels.

Comparisons between varieties and years identified Paragon as being one of the highest yielding in both years and Zebra the lowest yielding. Fasan, Tybalt and Monsun were consistent in their ranking but Amaretto fell from one of the highest yielding varieties in 2006 to one of the lowest in 2007 (Table 4.31). Further analysis of the results between the varieties and the seasons showed how Paragon and Tybalt maintained the highest levels of disease resistance to both *Zymoseptoria tritici* and yellow rust by GS65, also with the highest SPAD readings for leaf greenness at the same growth stage. Amaretto, whilst showing good disease resistance in 2006, recorded the highest % changes from 2006 to 2007 in both increased disease levels and reduced SPAD leaf greenness readings, along with Zebra. In 2006 Amaretto yield results were the most inconsistent in the trial, with the greatest variability. This made it higher yielding than Paragon in some plots in 2006 but as low as Monsun in others. In the poorer growing season in 2007 the variance of the yield increased for all varieties but Amaretto was unable to achieve yields comparable with Fasan.

Table 4.31 Rank order of the 6 varieties for grain yield in both seasons

Rank order	**2006**	**Yield t ha^{-1}**	**2007**	**Yield t ha^{-1}**
1	**Paragon**	6.8±0.15**a**	**Paragon**	2.6±0.57**ab**
2	**Amaretto**	6.7±0.44**ab**	**Fasan**	2.6±0.26**a**
3	**Fasan**	6.3±0.36**b**	**Tybalt**	2.4±0.31**ab**
4	**Tybalt**	6.2±0.40**b**	**Monsun**	2.1±0.36**ab**
5	**Monsun**	6.0±0.41**bc**	**Amaretto**	1.9±0.32**b**
6	**Zebra**	5.5±0.23**c**	**Zebra**	0.8±0.08**c**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

For varieties that showed the highest disease levels there was a significant reduction in yield and in 2007, where the disease levels were significantly higher throughout the season, the yields of all varieties were also significantly reduced. Zebra with its high susceptibility to yellow rust was the lowest yielding variety in both trial years, in stark contrast to its yield performance in 2005 in the previous trial in Chapter 3. The significance of variety as the main driver in wheat yield has been recognised and is well established (Przystalski *et al.*, 2008; Miko *et al.*, 2014) with Paragon being the highest yielding variety in both years.

Resistance to foliar diseases in wheat is influenced by both genetics and the environment (Bell, 1981). For example, wheat varieties with the *Sr6* allele with resistance to *Puccinia graminis* are found to have high resistance at 20ºC but are susceptible at 25 ºC and resistance decreases with increases in N availability (Mayama *et al.*, 1975). Studies into the effect of increased N supply on wheat leaf rust caused by *Puccinia triticina* identified an increase in spore production and lesion size in wheat plants receiving 214-274 kg N ha^{-1} compared with those at a lower rate of 160-210 kg N ha^{-1} (Robert *et al.*, 2005). Increasing other nutrient concentrations, such as potassium and calcium, is known most often to increase resistance in wheat to both of these diseases (Bell, 1981). In this study, the two year red clover fertility building ensured wheat plants were receiving high levels of N per hectare and Zebra and Fasan had high disease levels of yellow rust but also high protein concentrations with high protein quality. Monsun was most susceptible to *Zymoseptoria tritici*. The yield of Zebra was adversely affected by this whereas the yields of Fasan and Monsun were not. The protein concentrations and quality of Monsun were also comparable with the other varieties despite their better disease resistance performance.

The lower solar radiation levels, lower temperatures and higher rainfall recorded over the 2007 growing season had a clear impact on the yield. The subsequent disease levels, although higher in 2007, were not high enough to be detrimental to overall grain quality in the varieties, including Zebra which recorded the lowest green leaf area by SPAD and the lowest yields from its high infection of yellow rust.

For varieties to meet the criteria of both the growers and the end users of organic wheat they must have both a high yield and good grain and baking quality. In this study Tybalt, the shortest variety with a mean plant height of 81 cm, produced grains with the best quality levels, for the quality parameters tested, along with Fasan, one of the two tallest varieties (100 cm), whilst the other of the two tallest varieties Zebra (97 cm) had the highest crude protein and also the lowest yield. However, the baking tests identified Tybalt as the variety with the weakest dough and the most unstable after proving, producing the poorest loaves in all baking tests. The relationship between plant height, yield and protein quality identified in earlier studies (Eisele and Köpke, 1997; Calderini *et al.*, 2006) was based on varieties with greater variation in plant height than in this study, but Casebow *et al.* (2016) working on similar wheat heights to those in this study identified a very clear inflection point in the data at 80 cm. They identified no significant yield penalty with height from 80 to 100 cm from their organic trials, but specific weight was positively related to height with the penalty becoming more exaggerated as height declined. Trends in protein concentrations were found to be the opposite of those for yield but without the dilution of N as heights declined to 80 cm. SDS-sedimentation volumes also declined with plant height from 80 to 100 cm. These increases in specific weight and mean grain weight from 80 to 100cm along with reductions in N and S concentrations and sedimentation volumes did not, however, have any negative effect on loaf quality, leading the authors to conclude that negative effects on quality can be mitigated by stability or improvements in other relevant criteria, which they were unable to identify. This study, from its elite varieties selected, produced similar conflicting results. Therefore conclusions on quality differences from the narrow range of plant heights in the varieties from this study and other studies suggest that other criteria are at play in the final packing density of grains, from 80 cm to 100 and 120 cm.

Fasan, Zebra and Paragon were the tallest varieties grown in the trial with baking performances of these organically grown wheats that the industrial bakers found acceptable from their tests, suggesting that there might be other factors involved in the creation and deposition of the storage proteins (gliadins and glutenins) for baking quality which are not being tested for by the current industrial standards (Khan *et al.*, 2010). Fasan and Tybalt produced the highest protein quality results by SE-HPLC, with higher ratios of larger F1 gliadin to the smaller F3 and F4 glutenin fractions, yet only Fasan performed adequately in one of the baking tests whilst Tybalt did not. This raises questions over the quality assessment. Paragon produced the highest protein quality by Zeleny sedimentation value and Amaretto the lowest, yet both produced high-volume loaves from their respective wholemeal flours. The highest recorded protein percentages (for dough water absorption) were in varieties Zebra (15%) and Tybalt (14.8%) yet Tybalt doughs failed to prove adequately with poor water retention in the standard industrial test from the roller milled flour, whilst Zebra produced one of the highest loaf volumes from its white flour.

Milling of cereal grains to produce flour is a process which damages cereal starch granules. This results in changes to both the starch structure and properties, the degree and extent of which is determined by the milling process and grain variety (Li *et al.*, 2014). Comparative baking tests between stoneground and roller milled flours of Tybalt, Amaretto and Paragon identified that Paragon, although it performed poorly, was unaffected by the milling process, with a consistent bake from both flour streams. Amaretto had the best results of the three varieties and better from the stoneground flour than the roller milled. Tybalt flour performed poorly from the roller milling and showed an improvement in baking from the stoneground flour. Grain hardness is known to contribute significantly to the quality of the damaged starch (Choy *et al.*, 2015) and particularly when stone grinding wheat, where the resistance to milling is key to the way the starch fractures along natural stress lines (Greffeuille *et al.*, 2005). Grain hardness may have contributed to the differences observed both within and between varieties in the milling and baking evaluation but it is seldom if ever tested, as varieties are generally recognised as being either hard or soft (Pasha *et al.*, 2010). Studies have established significant differences in grain hardness between conventional and organic systems and where wheat varieties are being selected and grown with quality in mind, it is recommended that grain hardness tests be used alongside other recognised laboratory quality tests, particularly where stone milling has been identified as a potential end use (Carcea *et al.*, 2006).

The importance of variety in baking performance has also been identified with phytate levels in flour for nutrient availability (Greffeuille *et al.*, 2005) with higher phytate levels occuring in flours from harder grains. In this study the tallest varieties, Fasan and Zebra, had the highest phytic acid levels (9.8-10.9 mg g^{-1}) and Tybalt, the shortest variety, the lowest (8.8 mg g^{-1}). Comparisons of these values with values from other studies reviewed by Schlemmer *et al.* (2009), where ranges of 3.9-13.5 mg g^{-1} were identified from 25 *Triticum* species, places the phytic acid levels of all the varieties in this study in the upper levels for phosphorus storage. Comparisons between organic and conventionally grown wheat (Langenkamper *et al.*, 2006) found no significant differences in phytic acid levels from the differing production methods, ranging also between 10.8 and 11.2 mg g^{-1} for both groups. The choice of variety for flour milling should be taken into consideration by millers and bakers as variety, milling process and bread preparation choice can all influence the nutrient availability of breads for consumers. For UK stone milling purposes the phytate levels would be highest in Fasan and Zebra, and the phytic acid in the subsequent flour streams would also be higher from these hard wheats. Tybalt with the lowest phytic acid levels would therefore be better suited for the roller milling and industrial baking enterprises where otherwise high phytic acid levels without sourdough baking would reduce the nutrient delivery of the breads with these flours (Guo *et al.*, 2015). Given that the most concentrated sources of phytic acid tend to be in whole grains and that cereals form a high percentage of the human diet globally, this raises the issue of zinc and iron deficiencies in high phytate foods due to the phytic acid response during baking which impairs the bioavailability of iron and zinc (Ma *et al.*, 2005). The phytate/zinc and phytate/iron molar ratios are a useful index of nutrient bioavailability where it was observed that the Zn balance in adult men was higher in foods with a phytate/Zn ratio of up to 15 than it was in foods with a ratio > 16 (Harland and Morris, 1995). High dietary calcium was found to exacerbate the effect of phytic acid on zinc utilisation. Fasan not only had significantly higher levels of phytic acid but also the highest phytate/zinc and phytate/iron molar ratios with the highest levels of grain calcium, whereas Tybalt had the lowest molar ratios for both zinc and iron. Where plant breeders of organic wheat varieties are striving to reintroduce plant height for agronomic and quality improvements, consideration must be given to end use recommendations if the nutritional benefits are to be realised (Loschenberger *et al.*, 2008).

Lignin in the outer cell walls of plants consists of phenolic polymers which are known to increase as part of a plant’s defence against fungal and pathogen attack (lignification) (Bell, 1981). Lignification of the outer wall appears particularly important for resistance of species in the Poaceac to leaf-spotting fungi. By a systematic meta-analysis of the data from literature on plant phenolics and associated antioxidant levels in food from organic and conventional sources, Brandt *et al.* (2011) identified a higher level of phenolic compounds in organic foods possibly due to heightened defence mechanisms in the absence of fungicides. In their systematic literature review on the impact of organically grown crops on human health Johansson *et al.* (2014) concluded that no clear differences in the content of beneficial compounds could be found between organic and conventionally grown foods, with the one possible exception of phenolic compounds. These were subject to the influence of other parameters of much higher importance, such as selection of genotype, location, weather/ year, and harvest time. All of these have a pronounced effect on organic wheat production (Gomiero, 2017) and wheat is one of the food groups with the highest concentrations of vitamin E and total phenolics (Johansson *et al.*, 2014). In this study the analysis of vitamin E concentrations in the varieties at Gilchesters over the two years demonstrated how vitamin E levels were higher in 2006 when the crops had greener leaves (higher SPAD readings) and lower disease stress. However, there was no clear linkage between variety, disease levels and vitamin E in that varieties with the greatest disease resistance and lowest stress, Paragon and Amaretto, had similar concentrations of α- and β-tocopherols to the variety with the greatest disease infection, Zebra.

In both years and for all varieties the levels of both α- and β-tocopherols and α- and β-tocotrienols were higher than in wheats examined in other studies (Hussain *et al.*, 2012), which supports the studies by Johansson *et al.* (2014) and Hussain *et al.* (2012) that identified varietal importance in the delivery of antioxidants for human nutrition, and for benefits to human health (Baranski *et al.*, 2014). What is more difficult to determine is the impact on phenolic levels derived from greater natural disease resistance as suggested by Brandt (2013), as Paragon and Amaretto with a high disease resistance and potentially low level of lignification had the same high grain antioxidant levels as Zebra, which was subject to high disease stress and possibly higher levels of lignification. Research data is limited but from the most recent study by Rempelos *et al.* (2018) the level of constitutive phenolics in wheat, as an expression of genetic control, was reduced with increased levels of mineral N fertiliser and increased under compost applications, with the biggest differences in the effect being found in a tall straw variety from an organic breeding programme. The study further identified that where phenolics are induced due to pathogen attack, the rate of subsequent infection is determined to a lesser or greater degree by differences in varietal resistance.

Grain macro nutrients are positively correlated to soil macro nutrient levels (Wright *et al.*, 2007). The pre-sowing soil analysis in this study identified low calcium levels and high iron and aluminium levels at Gilchesters farm. Grain analysis on the subsequent wheat crops showed the lowest levels of calcium in the grains and also the ones with the highest aluminium and iron concentrations compared to the grains from the other two trial sites. This is in line with the known buffering effect of calcium in the soil on limiting aluminium uptake in plants and the interaction between iron availability and aluminium toxicity (Illmer and Buttinger, 2006). As a result wheat at Gilchesters from low calcium availability with a reduced uptake by the crop had an increased availability of aluminium and a greater uptake due to the presence of high levels of available iron. This demonstrates the importance of pH and liming on crop production and the subsequent nutritional density of grains for human health. Variety differences in grain nutrients in this study were identified with differences in plant height, where Fasan, the tallest variety, had the highest levels of calcium and Tybalt, the shortest, had the lowest levels. The highest levels of grain sulphur were found in Zebra, again one of the tallest varieties. A further link in the nutrient deposition in wheat grains was also identified by Murphy *et al*. (2007) where a significant Grain nutrient x Yield interaction was identified, with yield being negatively correlated to mineral concentrations, i.e. the dilution effect. Zebra, the lowest yielding variety in the trial, also had the highest levels of zinc, molybdenum, aluminium, cadmium and nickel, whilst Tybalt with higher yields had the lowest levels. Differences identified in other studies found that the modern varieties invest a smaller proportion of total biomass in roots than shoots, when measured at maturity (anthesis), than older, taller varieties, which had significantly higher root dry matter and higher root length density (Siddique *et al.*, 1990). This negative correlation since the introduction of the semi-dwarfing gene may be providing the driver for differences in yield, quality and nutrient density between the taller and shorter varieties and their sink: source relationship, as also identified by Borghi *et al.*, (1986).

4.4.1 Three farm trial site comparisons

When the performance of the varieties was compared from the three trial sites for yield and grain quality significant differences were observed between growing seasons, varieties and sites. For grain yield, analysis detected significant main effects of year, site, and variety. Grain yields were significantly higher in 2006 at 4.9 tonnes per hectare compared to 2.8 t per ha in the following year, higher at Gilchesters and Courtyard with 4.3 t ha^{-1} than at Sheepdrove farm (2.9 t ha^{-1}), highest with Tybalt (4.6 t ha^{-1}), intermediate with Fasan and Paragon (4.1 t ha^{-1}) and lowest with Zebra (2.8 t ha^{-1}) (Table 4.32). Significant interactions were detected between year and site, year and variety, and also site and variety (Table 4.32). The highest recorded yields of the whole trial were at Gilchesters in 2006 of 6.3 t ha^{-1} following two years of inoculated red clover, compared with Courtyard (4.9 t ha^{-1}) and Sheepdrove (3.5 t ha^{-1}) both after two years of white clover (Table 4.33). Ranking of varieties for yield by site and year are in Table 4.32a. When interaction means between variety and site were compared, yields were significantly lower at Sheepdrove for all varieties than at the other two sites (Table 4.34). Yields from Tybalt were significantly higher at Courtyard and at Gilchesters grain yield from Paragon was significantly higher than from Monsun and Zebra (Table 4.33).

For plant height significant differences were detected for season, site and variety. Varieties were on average significantly taller in 2006 (97 cm), tallest at Gilchesters (94 cm), intermediate at Courtyard (92 cm) and shortest at Sheepdrove (89 cm). Fasan was the tallest variety (100 cm), followed by Zebra (98 cm), Paragon, Amaretto and Monsun were intermediate (92-90 cm) with Tybalt the shortest (81 cm) (Table 4.32). Significant interactions were detected between year and site, year and variety, and also site and variety (Table 4.32). When interaction means between year and site were compared the greatest variation in height was at Sheepdrove with both Sheepdrove and Gilchesters having taller plants in 2006. At Courtyard the higher rainfall in 2007 on the site with the lightest soils produced taller plants in 2007 than in 2006 (Table 4.34). For site and variety interactions, Fasan was significantly taller at Courtyard than Sheepdrove, whilst Monsun and Zebra were significantly taller at Gilchesters than at Sheepdrove and Courtyard (Table 4.35).

For grain protein percentage contents were significantly higher in 2007 at 13.7% than in 2006 at 12.8% and higher at Gilchesters (14.5%) and Sheepdrove (13.9%) than at Courtyard farm (11.5%). They were highest with Zebra (14.1%) and intermediate with Paragon (13.6%) compared to all others (12.9%) (Table 4.32). Significant interactions were detected between season/year and site, year and variety, site and variety, and between year, site and variety (Table 4.32). When interaction means for the interaction between variety and site were compared, protein concentrations were lower at Courtyard for all varieties. Protein concentrations for Tybalt were significantly higher at Gilchesters, whilst for Zebra they were significantly lower at Courtyard (Table 4.33).

For thousand grain weight analysis detected significant main effects for season/year and variety but not site. Grain weights were higher in 2006 with 40.0 g compared to 38.9 g the following year, highest for Tybalt and Monsun (42 g), intermediate with Fasan and Paragon (40 g) and lowest with Amaretto and Zebra (37-36 g) (Table 4.32). With no main or interaction effect of site the varieties displayed a consistency in grain weight despite the strong seasonal variation in growing conditions each year, with the exception of Zebra which produced the smallest grains and recorded the largest degree of variation in TGWs at each farm (Table 4.33).

Table 4.32 Main effect means ± SE and *p* values for the effects of harvest year, site and variety on wheat yield parameters.

	Grain Yield	Plant height	Protein	TGW
	t ha^{-1}	cm	%	g
Year (YR)				
2006	4.9±0.16	97.1±1.07	12.8±0.22	40.0±0.05**a**
2007	2.8±0.14	86.7±1.36	13.7±0.17	38.9±0.63**b**
Site (ST)				
Courtyard	4.4±0.14**a**	92.4±1.25**ab**	11.5±0.19**c**	37.8±0.53
Gilchesters	4.2±0.33**a**	94.2±1.53**a**	14.5±0.12**a**	41.6±0.76
Sheepdrove	2.9±0.14**b**	89.0±2.07**b**	13.9±0.16**b**	39.7±0.63
Variety (VR)				
Amaretto	3.8±0.33**cd**	92.1±2.05**c**	12.9±0.28**c**	37.1±0.59**c**
Fasan	4.1±0.31**b**	100.2±2.16**a**	13.2±0.34**c**	39.7±0.7**b**
Monsun	3.7±0.31**d**	89.7±1.62**d**	12.9±0.38**c**	42.5±0.64**a**
Paragon	4.1±0.32**bc**	90.3±2.17**cd**	13.6±0.31**b**	39.9±0.7**b**
Tybalt	4.6±0.31**a**	81.2±1.78**e**	13.0±0.38**c**	41.9±0.69**a**
Zebra	2.8±0.37**e**	97.6±2.36**b**	14.1±0.35**a**	36.0±1.44**c**
ANOVA				
Main Effects				
YR	**0.0019**	**0.0017**	**0.0211**	**<.0001**
ST	**<0.0001**	**0.0026**	**<0.0001**	ns
VR	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**
Interactions				
YR:ST	**<0.0001**	**<0.0001**	**0.0339**	ns
YR:VR	**0.002**	**0.0028**	**0.0018**	**<0.0001**
ST:VR	**<0.0001**[1]	**0.0004**[2]	**<0.0001**	ns
YR:ST:VR	ns	ns	**0.0002**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 4.34. [2]For interaction means see Table 4.35.

Table 4.32a. The ranking order of grain yield of spring wheat varieties grown at Courtyard, Sheepdrove and Gilchesters Farms 2006 – 2007

	2006			2007		
Rank order	**Courtyard**	**Sheepdrove**	**Gilchesters**	**Courtyard**	**Sheepdrove**	**Gilchesters**
1	Tybalt	Tybalt	Paragon	Tybalt	Tybalt	Paragon
2	Fasan	Amaretto	Amaretto	Fasan	Paragon	Fasan
3	Paragon/Monsun	Paragon	Fasan/Tybalt	Monsun	Amaretto	Tybalt
4	-	Fasan	-	Paragon	Fasan	Monsun
5	Zebra	Monsun	Monsun	Amaretto	Monsun	Amaretto
6	Amaretto	Zebra	Zebra	Zebra	Zebra	Zebra

Table 4.33 Site effect means ± SE and *p* values for the effects of harvest year and variety in wheat yield parameters from each farm

	Grain yield			Plant height		
	t ha^{-1}			cm		
Year (YR)	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	4.9±0.17	6.3±0.16	3.5±0.13	89.4±1.59	99.5±1.64	102.2±1.17
2007	3.9±0.18	2.1±0.18	2.3±0.18	95.5±1.73	88.9±2.09	75.8±1.03
Variety (VR)						
Amaretto	3.8±0.19**de**	4.3±0.93**ab**	3.2±0.28**b**	92.7±1.26**c**	94.4±3.06**b**	89.2±5.34**bc**
Fasan	4.8±0.27**b**	4.5±0.73**ab**	2.9±0.23**bc**	104.3±2.27**a**	102.0±2.92**a**	94.2±4.94**a**
Monsun	4.3±0.20**c**	4.1±0.78**b**	2.7±0.26**c**	88.7±1.52**d**	92.5±2.45**b**	87.9±4.01**c**
Paragon	4.2±0.30**cd**	4.7±0.83**a**	3.2±0.17**b**	90.6±1.41**cd**	92.5±3.05**b**	87.9±5.82**c**
Tybalt	5.6±0.25**a**	4.3±0.77**ab**	3.8±0.24**a**	80.7±1.42**e**	81.1±2.91**c**	81.9±4.53**d**
Zebra	3.7±0.40**e**	3.2±0.88 **c**	1.6±0.3**d**	97.5±1.96**b**	102.7±2.76**a**	92.7±6.05**ab**
ANOVA						
Main Effects						
YR	**0.0022**	**0.002**	**0.0064**	**0.0388**	**0.0078**	**0.0001**
VR	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**	**<0.0001**
Interactions						
YR:VR	**0.0461**	ns	**0.0079**	ns	**0.0274**	**0.0188**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$).

Table 4.33 (cont.) Site effect means ± SE and *p* values for the effects of harvest year and variety in wheat yield parameters from each farm

	Protein			TGW		
	%			g		
Year (YR)	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	10.8±0.28	14.5±0.12	13.3±0.21	37.1±0.05	43.3±0.07	39.9±0.07
2007	12.2±0.16	14.5±0.23	14.6±0.14	38.6±0.96	39.5±1.36	38.8±1.03
Variety (VR)						
Amaretto	11.5±0.43**b**	13.7±0.17**c**	13.5±0.34**cd**	36.3±0.6**b**	38.5±1.63**c**	36.5±0.66**b**
Fasan	11.2±0.25**bc**	14.4±0.31**abc**	13.8±0.36**bc**	37.4±1.25**b**	42.4±0.63**abc**	39.4±1.06**ab**
Monsun	10.6±0.39**c**	14.1±0.3**bc**	13.9±0.31**b**	42.1±0.72**a**	43.4±1.63**ab**	42.4±1.02**a**
Paragon	12.3±0.56**a**	14.6±0.1**ab**	14.1±0.35**b**	37.2±0.36**b**	41.9±1.06**abc**	40.7±1.32**a**
Tybalt	11.0±0.27b**c**	14.9±0.27**a**	13.1±0.41**d**	40.3±1.01**a**	43.6±1.18**a**	41.7±1.23**a**
Zebra	12.3±0.53**a**	15.0±0.36**a**	15.2±0.16**a**	33.6±1.2**c**	39.2±3.66**bc**	35.7±2.30**b**
ANOVA						
Main Effects						
YR	0.0623	0.975	**0.0023**	**<0.0001**	**0.0002**	**<0.0001**
VR	**0.0002**	**<0.0001**	**<0.0001**	**<0.0001**	**0.0159**	**<0.0001**
Interactions						
YR:VR	0.2695	**<0.0001**	0.0936	**<0.0001**	**0.0059**	**<0.0001**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$).

Table 4.34 Interaction means for effects of wheat variety and site on grain yield.

	Grain Yield t ha^{-1}					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	3.8±0.19**b**	4.8±0.27**a**	4.3±0.20**a**	4.2±0.30**b**	5.6±0.25**a**	3.7±0.40**a**
GIL	4.3±0.93**a**	4.5±0.73**b**	4.1±0.78**b**	4.7±0.83**a**	4.3±0.77**b**	3.2±0.88**b**
SHE	3.2±0.28**c**	2.9±0.23**c**	2.7±0.26**c**	3.2±0.17**c**	3.8±0.24**c**	1.6±0.30**c**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, p < 0.05)

Table 4.35 Interaction means for effects of wheat variety and site on straw height.

	Straw height cm					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	92.7±1.26**a**	104.3±2.27**a**	88.7±1.52**b**	90.6±1.41**a**	80.7±1.42**a**	97.5±1.96**b**
GIL	94.4±3.06**a**	102.0±2.92**a**	92.5±2.45**a**	92.5±3.05**a**	81.1±2.91**a**	102.7±2.76**a**
SHE	89.2±5.34**a**	94.2±4.94**b**	87.9±4.01**b**	87.9±5.82**a**	81.9±4.53**a**	92.7±6.05**b**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, p <0.05)

Grain quality assessments on the varieties from the three sites by SE-HPLC, to determine the glutenin and gliadin fractions (F-1 being the large high molecular weight glutenins, F-3 low molecular weight glutenins and F-4 low molecular weight gliadins) for overall gluten quality found that the quality was higher in 2007, and highest for the tallest variety, Zebra at all three farms. The gluten quality was lowest for Paragon and Monsun at Courtyard and lowest for Paragon at Sheepdrove (Table 4.36). There was less variation among varieties at Gilchesters.

Table 4.36 Effect means ± SE and *p* values for the effects of harvest year and variety in spring wheat on gluten quality from each farm

	Protein Quality by SE-HPLC		
	Fractions F1/(F3+F4) **%**		
	COU	**SHE**	**GIL**
Year (YR)			
2006	3.1±0.04	3.4±0.04	3.4±0.042
2007	3.6±0.04	3.8±0.05	3.7±0.04
Variety (VR)			
Amaretto	3.4±0.1**b**	3.6±0.06**bc**	3.4±0.04**b**
Fasan	3.6±0.11**a**	3.8±0.13**a**	3.6±0.10**a**
Monsun	3.2±0.10**c**	3.6±0.05**b**	3.5±0.03**b**
Paragon	3.2±0.11**c**	3.3±0.09**d**	3.4±0.10**b**
Tybalt	3.3±0.08**bc**	3.5±0.06**c**	3.5±0.06**ab**
Zebra	3.4±0.11**b**	3.7±0.07**b**	3.4±0.12**b**
ANOVA			
Main Effects			
YR	**0.0024**	**0.0013**	**0.0015**
VR	**<0.0001**	**<0.0001**	**<0.0001**
Interactions			
YR:VR	ns	**0.0048**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, p < 0.05)

4.4.2 Three farm trial site redundancy analysis (RDA)

Correlations between farm sites, agronomic and environmental factors and varieties and their respective yield and quality levels.

In the biplot shown in Fig. 4.5, correlations between organic agronomic practices at each site, their macronutrient supply (from individual soil analysis) and climatic (radiation, precipitation and air temperature) factors, the choice of wheat variety and their effect on grain yield and quality were observed. Weather data from each site taken from sowing date to harvest date, along with soil macro nutrient data and wheat variety were used as drivers. Most variation (43%) is explained by axis 1 and a further 21% by axis 2. For individual drivers 32% of the variation is explained by Courtyard Farm, 20.9% by soil N and 10.15 from solar radiation (Table 4.38)

The tight cluster of Amaretto, Paragon and Monsun in the centre of the axes (zero correlation) indicate varieties with the least correlation to any of the other drivers or other response variables. This suggests that these three varieties are less susceptible to the pedo-climatic variations set up in this trial. Tybalt was more positively associated with gluten quality and at Sheepdrove farm than the other five varieties, but as demonstrated in Chapter 4 this did not equate to a good baking performance in the variety baking tests. Fasan and Zebra were more positively correlated with yield and straw height.

Sheepdrove Farm, a mixed farming enterprise with livestock and their manures incorporated in the organic rotation since 1995, has a strong, positive correlation (indicated by the length of the arrows from the centre of the axes) with high Soil N and C and grain gluten quality (gliadin:glutenin ratio). Gilchesters, a stockless arable farm with red clover and green waste compost amendments within the rotation since 2002, had a strong, positive correlation with grain protein levels (by NIR), leaf greenness (by SPAD), yield and TGW. Both farms had a strong positive correlation with high grain nutrient levels. The strong positive correlations of these two sites to the other response variables can be measured by only 0.5% of the variation being attributed to each of these two drivers.

Courtyard was negatively correlated to the other two trial sites, with yield and grain quality and grain nutrient content, especially Zn and Fe. The negative correlation of this trial site accounted for 20.9% of the total variation (Table 4.38). It had a strong correlation with soil P and phytic acid: Zn and Fe molar ratios. Whereas grains from Gilchesters and Sheepdrove which were positively correlated with Zn and Fe levels had a strong negative correlation with phytic acid:Zn and Fe molar ratios.

Grain yield and straw height were positively correlated with solar radiation and negatively correlated with rainfall. Grain quality characteristics (F3+F4) were negatively correlated with solar radiation, plant establishment numbers and Courtyard Farm but positively correlated with soil nutrient levels and Gilchesters and Sheepdrove. From this study the RDA suggests that under organic agronomic practices soil nutrient levels, specifically N, have a greater influence on grain quality than either variety choices or the weather conditions.

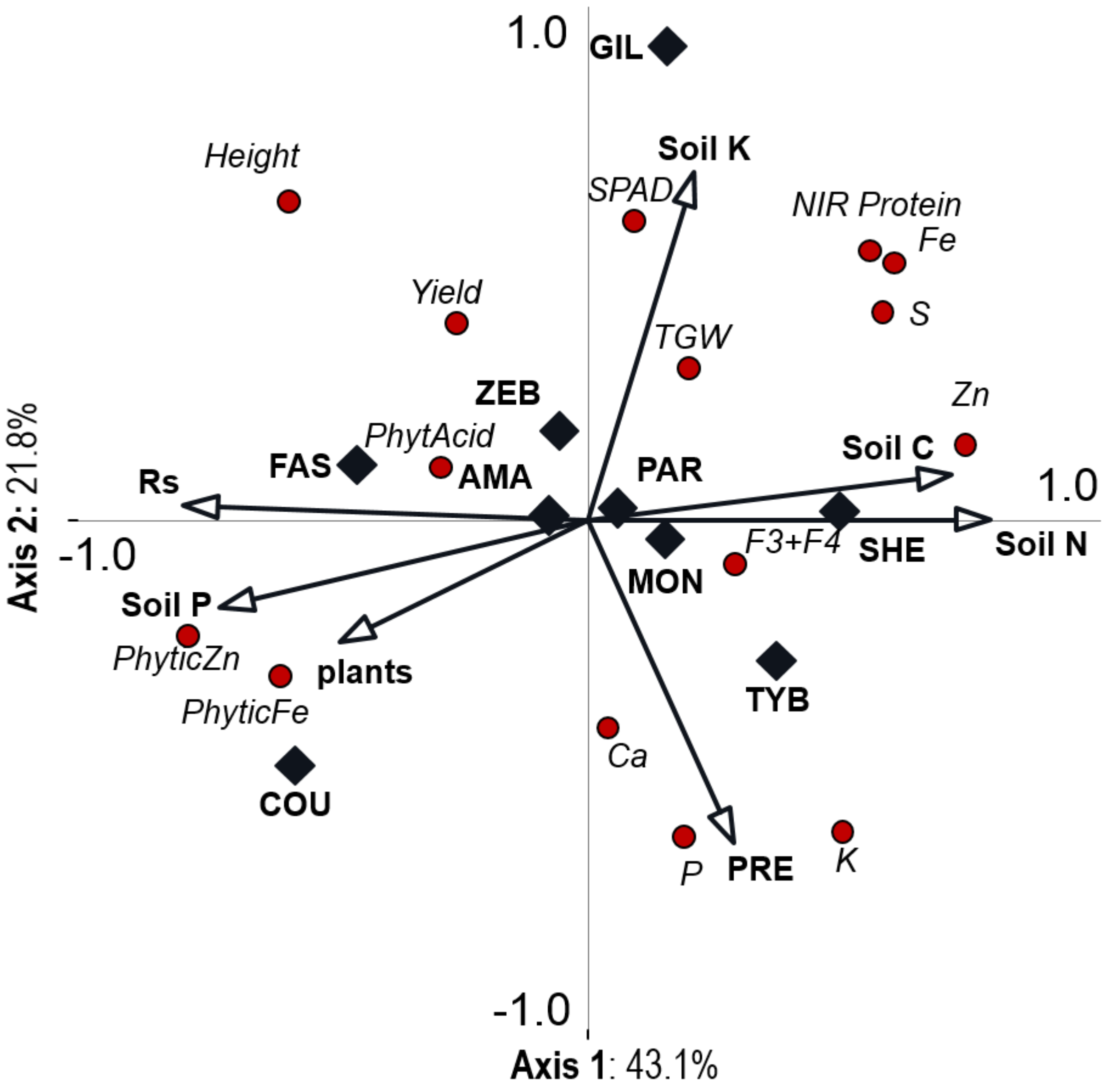


Fig. 4.5 Biplot derived from redundancy analysis (RDA) showing the relationship between weather conditions, soil nutrients, site (with associated organic agronomic management) and wheat variety drivers, with grain harvest, quality and nutrient levels as the response variables. (Drivers: For climate Rs=solar radiation, Pre=precipitation, plants=plants/m^2. For site COU=Courtyard Farm, GIL=Gilchesters Farm, SHE=Sheepdrove Farm. For varieties AMA=Amaretto, FAS=Fasan, MON=Monsun, PAR=Paragon, TYB=Tybalt, ZEB=Zebra. Response variables: NIR Protein=grain protein %, F3+F4=grain protein quality by SE-HPLC, PhytAcid=grain phytic acid content, PhyticFe=grain phytic acid:Fe molar ratio, PhyticZn=grain phytic acid:Zn molar ratio, Height=straw height, Yield=grain yield t/ha, TGW=thousand grain weight). Conditional Term Effects and P values are shown in Table 4.37

Table 4.37 Variety trial RDA conditional term effects for site, soil nutrients weather and variety

RDA Driver	Explained variation %	Pseudo-F	P value
Courtyard	32.0	55.0	0.002
Soil N	20.9	51.6	0.002
Radiation	10.1	31.3	0.002
Precipitation	7.1	27.2	0.002
Tybalt	5.9	28.1	0.002
Fasan	3.8	21.5	0.002
Zebra	2.3	14.5	0.002
Monsun	0.8	5.4	0.002
Gilchesters	0.5	3.2	0.014
Sheepdrove	0.5	3.2	ns
Amaretto	0.3	1.8	ns
Paragon	0.3	1.8	ns
Soil P	0.2	1.4	ns
Soil K	0.2	1.3	ns
Plants m^{-2}	0.1	1.0	ns

4.5 Conclusion

Although variety choice is influential on yield and quality and is the primary consideration for growing spring wheat in conventional systems, under organic systems for bread-making the variety choice is influenced more strongly by the fertility practices employed on individual farms as highlighted in the RDA analysis. Results in this study indicate that variety choice is a pre-requisite for achieving milling grade flour and that spring varieties Paragon, Tybalt, Fasan and Amaretto can produce adequate protein levels in growing seasons of considerable variation. However, further studies into the baking performance of these and other varieties from organic production systems is required to more fully understand the protein complexities that are delivering different results in baking performance than the standard industrial tests would indicate.

CHAPTER 5: Effect of fertiliser input type and rate on the grain yield and baking quality of organic spring wheat in the UK

5.1 Introduction

Protein content of wheat in organic systems is often 20 to 40% below that grown in conventional systems (Taylor and Cormack, 2002). In both systems, variety choice sets the potential grain quality for milling and baking, but fertility management and the environment affect the extent to which this potential is achieved. Manufactured fertilisers are widely used in conventional production to influence grain yield and quality. In both winter and spring milling wheats, late application of nitrogen is often used to increase grain protein content: 40 kg ha^{-1} N applied as solid fertiliser prills at the end of May (flag leaf emergence) may increase protein by 0.5 to 0.7% and yield by 0.1 to 0.2 t ha^{-1}. Foliar urea applied at 40 kg N ha^{-1} may increase protein by 0.75 to 1% but is unlikely to affect yield (MAFF, 2000; HGCA, 2009). These inputs allow a late application of nitrogen to be utilised by the grain during grain fill. As these inputs of mineral N fertiliser are not permitted according to organic standards, fertility management depends on building fertility in the rotation with grass/clover leys, green manures, grain legumes and animal manures and composts, e.g. composted green waste, farm yard manure (FYM) from cattle, pigs and poultry, in some cases subject to certain restrictions or requirements (Anon, 2009). The amount of N that can be applied in the form of 'organic' manures is restricted to 170 kg N ha^{-1} averaged over the whole farm area and a maximum of 250 kg N ha^{-1} on any one field.

Although the total quantity of N applied to spring wheat may be the same irrespective of fertility input type, e.g. 170 kg N ha^{-1}, different manures provide different concentrations of total and readily available N for rapid crop uptake. For example, poultry manure contains 16 – 30 kg N t^{-1} (30-45% of which is readily available) compared with FYM (cattle) of 6 kg N t^{-1} (20-25% readily available) (MAFF, 2000). However, potential losses of N by volatilisation and leaching are greater for poultry manure than other forms of FYM and composts. Compared with mineral fertilisers such as ammonium nitrate, organic N in manures is much less available to the crop to which it is applied and becomes potentially available as a result of mineralisation over months and years. The use of such fertilisers can therefore result in a limited supply of available nitrogen at times when the crop has high demands e.g. during stem extension and grain fill.

The **aims and objectives** of this experiment were therefore:

To identify the impact of differing nitrogen application regimes on grain yield and quality of organic spring wheat

To quantify the effect of supplementary nitrogen inputs from manures applied later in the growing season on grain yield and quality

5.2 Materials and Methods

5.2.1 Trial site characteristics

Identical trials were established on the same fields at the same three sites in England used in the variety trial described in Chapter 4 (Table 4.1) in two wheat growing seasons (2006 and 2007). In each year the fertility management trial was situated adjacent to the variety trial. The three farms were certified according to organic production standards. The materials, methods and results of the experiments conducted at Gilchesters Farm are presented here. The results from the other two identical experiments are included at a later stage (General Discussion Chapter 6) to enable direct comparisons of the performance of the selected bread wheat variety across the contrasting pedo-climatic conditions and seasons.

Soil samples were taken from each site prior to spring sowing in each season (Table 5.1) from the top soil (0-30 cm) and sent to Field Science Ltd, Bristol for analysis. The results are presented in Chapter 4 and indicate a sufficient P index of 2 in 2007 and 3 in 2006, with a sufficient K index of 2- in 2006 and 2+ in 2007 indicating that P and soil K were both adequate and likely to be non-limiting for crop growth. The pH at just below 7 was also ideal and non-limiting for crop production.

5.2.2 Experimental design and agronomic methods

The spring wheat crop was preceded by a fertility building grass/clover ley of two years duration. At Gilchesters, winter wheat then winter barley was followed by a two-year red clover ley that was cut frequently and mulched (Table 4.1).

The design was a randomised complete block with four replicates. Plots were 8 m long by 30 m wide. Experimental treatments were four fertility input types (FYM = farm yard manure, CMP = chicken manure pellets, FYM + CMP = 50:50 for the appropriate N rate and GWC = green waste compost) at two fertility input levels, 125 and 250 kg N ha^{-1}, and were accompanied by an untreated control. There was a total of 108 plots in each year. In both years, each of the fertility input types applied at the three sites was obtained from a single, homogenous batch, and so was identical in composition. FYM and green waste compost were supplied by Sheepdrove Farm. The chicken manure pellets were a proprietary brand (Greenvale Ltd, Yorkshire) made from composted chicken manure formed into small, 3 mm diameter pellets and approved for restricted use by organic farming standards. Analytical composition of the three organic manures (determined by Natural Resource Management Ltd, Bracknell, Berkshire) ensured the different levels of nitrogen inputs were applied in spring shortly before the leys were ploughed out for the following wheat crop. Analytical composition and quantities applied to achieve the different N-input levels are shown in Table 5.2. FYM and composts were applied mechanically with a Millcreek model 57 type manure spreader in February or March, but chicken manure pellets were batch weighed and applied by hand post-emergence between the plant rows in early to mid-May. An untreated 'control' plot was included in the experimental design. The spring variety Paragon was selected due to its known track record for milling quality from both the conventional and organic production sectors in Europe.

5.2.3 Crop establishment

FYM compost and GWC were both ploughed in before drilling. Certified organic seed was sown at rates of 500 seeds m^{-2} (200 kg ha^{-1}, according to TGW of 40.39g). Plots at Gilchesters were sown using an Accord 4 m combination drill consisting of a Rabe power-harrow (Rabe Werk, Bad Essen, Germany). Plot weeding was carried out twice, 14 days apart weather permitting, with an Einbock comb-harrow (Table 5.1). Operation dates for each trial are given in Table 5.1.

Table 5.1 Field trial operation dates for both 2006 and 2007

	Gilchesters	
	2006	**2007**
Drilling	10/04	02/4
1st weeding	12/05	26/04
Chicken pellet application	17/05	12/05
2nd weeding	26/05	18/5
Harvest	30/8	12/09

Table 5.2 Analytical composition of farm yard manure (FYM), green waste compost (GWC) and chicken manure pellets (CMP) on a dry matter basis in 2006 and 2007

	2006			2007		
	FYM	**GWC**	**CMP**	**FYM**	**GWC**	**CMP**
Dry matter (%)	22.9	40.0	92.0	53.2	40.7	86.2
Total nitrogen (% w/w)	3.18	1.54	3.82	1.70	2.94	4.24
Total carbon (% w/w)	38.1	20.3	38.6	17.2	35.5	39.7
C:N ratio	12:1	13.1	10:1	10.1	12.1	9.5:1
Nitrate nitrogen (mg/kg)	305	159	404	63	1508	313
Ammonium nitrogen (mg/kg)	298	126	5383	502	357	5579
Total phosphorus (P) (mg/kg)	6727	2583	10200	3150	10600	13600
Total potassium (K) (mg/kg)	23675	5850	13560	7040	17100	36900
Total copper (Cu) (mg/kg)	14.0	23.1	22.6	38.1	60.1	133
Total zinc (Zn) (mg/kg)	90	143	71	178	300	885
pH	8.69	7.80	na	7.35	7.31	na
Applications (t ha^{-1} fresh weight) to achieve:						
125 kg ha^{-1} N	23.4	27.6	4.5	18.8	14.1	4.0
250 kg ha^{-1} N	34.4	40.6	6.5	27.6	20.8	5.9

5.2.4 Disease and leaf chlorophyll assessment protocols

Disease assessments for powdery mildew (*Blumeria* (*Erysiphe*) *graminis*), Septoria (*Parastagonospora nodorum, Zymoseptoria tritici*) and wheat yellow rust (*Puccinia striiformis*) were carried out according to Section 4.2.6 at GS 65 (Zadoks *et al.*, 1974). Disease levels were assessed as a percentage of leaf area affected at GS65 with assessments carried out on the two youngest leaves: L1 (flag leaf), L2 (second leaf). For disease monitoring 20 tillers were assessed in each plot, by dividing plots into 4 50 x 50 cm sub-plots, within which 5 tillers were chosen randomly. Leaf greenness of upper leaves was recorded by single-photon avalanche diode detection (SPAD) using a Minolta chlorophyll meter model SPAD 502 (Spectrum Technologies Inc., Plainfield, Illinois, USA). Ten measurements per plot were made on randomly selected flag leaves just after the start of flowering (GS65).

5.2.5 Harvest and grain sampling procedures

Crop yields were assessed using a plot-combine harvester (CLAAS Dominator 38, Germany) with a 2.4 m wide cutter bar. Grain moisture content was determined by drying grain at 80°C for two days using a forced-draught drying oven (Genlab Ltd, Widnes, UK), with yield data presented at 15% moisture content. All grain analyses (protein content, HFN, gluten content and protein composition/quality) were carried out at Campden & Chorleywood Food Research Association (CCFRA).

5.2.6 Wheat quality assessments

Grain analysis for the characterisation of protein content of wheat samples was determined by NIR reflectance by DUMAS. Hagberg Falling Number (HFN), to identify α-amylase activity in wheat, was determined by forming an aqueous suspension of flour and water followed by measurement in a Perten 1700 Falling Number instrument. Gluten content and quality was determined by Zeleny Sedimentation Value, where the degree of sedimentation of a flour suspended in a lactic acid solution during a standard time interval is taken as a measure of baking quality. Higher gluten content and better gluten quality both give rise to slower sedimentation and higher values. Grain macro-nutrient and phytate analysis was undertaken at Sabanci University, Turkey.

5.2.7 Statistical analysis

Trial data analyses of variance from the 108 trial plots were derived from general linear models and linear mixed-effects models (Pinheiro, 2000). The data collected from the untreated control is included in the tables but was not included in these analyses. Fixed effects were season, fertility treatment and rate. The effects of the fixed factors on yield, yield components, protein fractions, disease severity, grain macro and micro-nutrients and heavy metals and grain phytic acid content were determined. The analyses were carried out in the R statistical environment (R Development Core Team, 2012). The combined data for main effects were analysed first, and where interaction terms were significant, further analyses were conducted at each level of the interacting factor. Secondly main effects of season and variety were analysed and where interaction terms were significant, further analyses were conducted at each level of the interacting factor. Differences between significant main effect and interaction means were determined using Tukey's Honest Significant Difference (HSD) tests, based on mixed-effects models. The relationships between wheat varieties and their grain yield, quality and nutrient contents with soil nutrient levels, environmental, and agronomic factors and site (geographical location) were investigated using redundancy analysis (RDA). In all cases the RDAs were carried out using the CANOCO 5 package. Automatic forward selection of the environmental variables, site, soil nutrients and wheat variety within the RDAs was used and their significance in explaining additional variance calculated using Monte Carlo permutation tests.

5.3 Results

5.3.1 Effect of fertiliser input type and rate on disease severity

Disease levels for powdery mildew (*Blumeria* (*Erysiphe*) *graminis*) and Septoria (*Parastagonospora nodorum, Zymoseptoria tritici*) were very low in both seasons and especially for yellow rust (data not presented). Assessments for powdery mildew found no significant differences for year, fertility type or fertility rate. No interactions were detected (Table 5.5). For *Z. tritici* the disease severity was significantly higher on L2 in 2007 than 2006 (1.1 vs 0%). No significant interactions were detected for *Z. tritici* between experimental factors (Table 5.3).

Table 5.3 Main effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat disease severity for Leaves 1 and 2 (L1 and L2) at GS65 Gilchesters Farm

	Mildew L1[1]	**Mildew L2**	***Z. tritici* L1**[2]	***Z. tritici* L2**
	%	%	%	%
Year				
2006	0±0	0.01±0.005	0±0	0±0
2007	0±0	0.12±0.073	0±0	1.1±0.13
Fertility rate				
0 N	0±0	0.1±0.009	0±0	1.1±0.47
125 kg N ha^{-1}	0±0	0.09±0.064	0±0	0.6±0.17
250 kg N ha^{-1}	0±0	0.04±0.031	0±0	0.5±0.09
Fertility Type				
CMP	0±0	0.13±0.125	0±0	0.7±0.31
FYM	0±0	0.08±0.062	0±0	0.5±0.13
FYM+CMP	0±0	0.01±0.006	0±0	0.5±0.13
GWC	0±0	0.04±0.031	0±0	0.5±0.12
ANOVA				
Main Effects				
YR	ns	ns	ns	**0.0053**
FR	ns	ns	ns	ns
FT	ns	ns	ns	ns
Interactions				
YR:FR	ns	ns	ns	ns
YR:FT	ns	ns	ns	ns
FR:FT	ns	ns	n	ns
YR:FR:FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

5.3.2 Effect of fertiliser input type and rate on crop performance

For **grain yield**, analysis detected significant main effects of both year and fertility type. Grain yields were significantly higher in 2006 (5.5 t ha^{-1}) than 2007 (2.5 t ha^{-1}), and higher with CMP (4.4 t ha^{-1}) than with FYM+CMP (3.8 t ha^{-1}) (Table 5.4). There was no significant difference in grain yield between the CMP, FYM and GWC treatments. Both 125 and 250 kg N ha^{-1} resulted in a >0.4 t ha^{-1} yield increase above the control treatment. No significant interactions were detected for grain yield (Table 5.4).

For **plant height** analysis detected a significant main effect for year only, with Paragon achieving a taller plant height in 2006 of 93.4 cm compared to 85.3 cm in 2007 (Table 5.4). There was no effect of fertiliser rate or type on plant height.

At **Growth Stage 65 SPAD** (single-photon avalanche diode) analysis registered a significant main effect for year only. Levels were significantly higher in 2006 at 53.8 compared with 45.8 the following year (Table 5.4). Higher SPAD values were obtained at 250 kg N ha^{-1} when compared with 125 kg N ha^{-1} although this was not significant. There was no significant effect of fertiliser type on SPAD suggesting limited differences in N availability between the different fertiliser treatments used.

Thousand Grain Weight analysis detected significant main effects for both year and fertiliser type (Table 5.4). TGW was significantly higher in 2007 (43.1 g) than 2006 (39.3 g), highest with FYM+CMP (41.6 g) and GWC (41.5 g) and lowest with CMP (39.7 g). Significant interactions were detected between fertiliser type and fertiliser input rate and between year and fertiliser type for TGW. At 125 kg N ha^{-1} CMP had the lowest TGW but at 250 kg N ha^{-1} it was not significantly different from all other treatments (Table 5.5). In 2006 there was no effect of fertiliser type on TGW. In 2007 TGW from CMP (40.7 g) was significantly lower than FYM (44.8 g) and GWC (44.4 g) but not FYM+CMP (43.2 g) (Table 5.6).

Table 5.4 Main **e**ffects means ± SE and *p* values of harvest year, fertiliser rate and type on spring wheat grain yield parameters

	Grain Yield	**Plant height**	**SPAD**	**TGW**
	t ha^{-1}	cm	GS65	g
Year				
2006	5.5±0.1	93.4±0.6	53.8±0.33	39.3±0.3
2007	2.5±0.15	85.3±1.28	45.8±0.55	43.1±0.73
Fertility rate				
0 N	3.8±0.2	87.2±2.04	48±1.3	40±0.3
125 kg N ha^{-1}	4.3±0.3	90.3±1.13	49.3±0.82	41.2±0.56
250 kg N ha^{-1}	4.2±0.32	88.4±1.32	50.3±0.87	40.4±0.65
Fertility type				
CMP	4.4±0.42**a**	90.2±1.81	50.2±1.04	39.7±0.77**b**
FYM	4.5±0.44**a**	90±1.68	49.2±1.05	40.9±1.0**a**
FYM+CMP	3.8±0.45**b**	87.6±1.9	50.5±1.29	41.6±0.76**a**
GWC	4.3±0.44**ab**	89.6±1.67	49.4±1.41	41.5±0.88**a**
ANOVA				
Main Effects				
YR	**0.0005**	**0.0265**	**0.0048**	**0.0003**
FR	ns	ns	ns	ns
FT	**0.0479**	ns	ns	**0.0137**
Interactions				
YR:FR	ns	ns	ns	ns
YR:FT	ns	ns	ns	**0.0136**[2]
FR:FT	ns	ns	ns	**0.0025**[1]
YR:FR:FT	ns	ns	ns	**0.0265**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 5.5. [2]For interaction means see Table 5.6

Table 5.5 Interaction means for effects of fertiliser rate and type on TGW at Gilchesters Farm

	Fertility type			
Fertility rate	**CMP**	**FYM**	**FYM+CMP**	**GWC**
125 kg N ha^{-1}	38.9±0.85**b**	41.6±1.27**a**	41.8±0.87**a**	42.9±1.20**a**
250 kg N ha^{-1}	40.6±1.32**a**	40.3±1.57**a**	41.2±1.46**a**	39.8±0.96**a**

Means labelled with the same letter within the same rows are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 5.6 Interaction means for effects of year and fertiliser type on TGW at Gilchesters Farm

	TGW			
Year	**CMP**	**FYM**	**FYM+CMP**	**GWC**
2006	38.8±0.50**a**	38.4±0.50**a**	40.3±0.60**a**	39.6±0.60**a**
2007	40.7±1.50**b**	44.8±0.90**a**	43.2±1.30**ab**	44.4±1.20**a**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

5.3.3 Effect of fertiliser input type and rate on quality parameters

For **grain protein** % **concentrations**, analysis detected a significant main effect for fertiliser type only. Protein content was significantly higher with CMP (13.2 %) and with a combination of FYM+CMP (13.3 %) and lowest with FYM (12.4 %) and GWC (12.6%) (Table 5.7). Protein content was not significantly different between the two fertility rates.

For **grain specific weight** analysis detected significant main effects for year and fertility type only. Grain specific weights were higher in 2006 (78.8 kg hl^{-1}) than 2007 (74.5 kg hl^{-1}) and highest with GWC (77.6 kg hl^{-1}) and FYM (77.9 kg hl^{-1}), intermediate with CMP (76.8 kg hl^{-1}) and lowest with FYM+CMP (75.4 kg hl^{-1}) (Table 5.7).

Significant interactions were detected between fertility type and rate for grain specific weight and between year and fertility type. At the lower rate of 125 kg N ha^{-1}, grain specific weight was highest using GWC (78.5 kg hl^{-1}) and lowest from FYM+CMP (75.2 kg hl^{-1}). CMP (76.2 kg hl^{-1}) and FYM (77.4 kg hl^{-1}) were not significantly different (Table 5.8). At the higher rate of 250 kg N ha^{-1}, the use of FYM+CMP resulted in a significantly lower specific weight than with FYM alone (Table 5.8). The effect of fertiliser type was significant only in one of the two years (Table 5.9). There was no difference between treatments in 2006, but in 2007 the specific weight was highest with FYM (76.8 kg hl^{-1}) and GWC (75.95 kg hl^{-1}), intermediate with CMP (74.3 kg hl^{-1}) and lowest with FYM + CMP combined (71.5 kg hl^{-1}).

For **Hagberg Falling Number** analysis detected no significant main effects and no significant interactions (Table 5.7). Hagberg Falling Number was much higher (433.8 s) in 2007 than in 2006 (349.5 s) with little differences between fertiliser rate and types.

For gluten quality **Zeleny sedimentation** analysis detected a significant main effect for year only. Gluten quality was higher in 2006 (70 ml) than in 2007 (64.7 ml).

Table 5.7 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain quality parameters

	Specific weight	**HFN**	**Protein**	**Zeleny sediment**
	kg hl^{-1}	s	%	ml
Year				
2006	78.8±0.10	349.7±5.60	12.9±0.10	70.0±0.25
2007	74.5±0.67	433.8±18.83	13.0±0.22	64.7±0.64
Fertility rate				
0 N	76.9±0.58	386.2±15.02	13.2±1.93	67.6±0.69
125 kg N ha^{-1}	76.9±0.57	389.4±15.28	12.8±0.13	67.6±0.65
250 kg N ha^{-1}	76.9±0.60	383.7±14.23	13.0±0.18	67.7±0.67
Fertility type				
CMP	76.8±0.83**ab**	386.6±20.44	13.2±0.16**a**	68.7±0.65
FYM	77.9±0.37**a**	395±19.44	12.4±0.22**b**	66.7±1.2
FYM+CMP	75.4±1.11**b**	371.5±23.8	13.3±0.21**a**	68.2±0.68
GWC	77.6±0.64**a**	394.6±20.20	12.6±0.21**b**	67.1±1.10
ANOVA				
Main Effects				
YR	**0.002**	ns	ns	**0.0095**
FR	ns	ns	ns	ns
FT	**0.0004**	ns	**0.0302**	ns
Interactions				
YR:FR	ns	ns	ns	ns
YR:FT	**0.0001**[2]	n	ns	ns
FR:FT	**0.0303**[1]	ns	ns	ns
YR:FR:FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see table 5.8. [2]For interaction means see table 5.9

Table 5.8 Interaction means for effects of fertiliser rate and type on specific grain weight at Gilchesters Farm

	Specific weight			
Fertility rate	**CMP**	**FYM**	**FYM+CMP**	**GWC**
125 kg N ha^{-1}	76.2±1.23**bc**	77.7±0.51**ab**	75.2±1.51**c**	78.5±0.48**a**
250 kg N ha^{-1}	77.4±1.15**ab**	78.1±0.57**a**	75.6±1.76**b**	76.6±1.11**ab**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 5.9 Interaction means for effects of year and fertiliser type on specific grain weight at Gilchesters Farm

	Specific weight			
Year	**CMP**	**FYM**	**FYM+CMP**	**GWC**
2006	78.7±0.27**a**	78.8±0.16**a**	78.8±0.16**a**	78.8±0.28**a**
2007	74.3±1.37**b**	76.8±0.58**a**	71.5±1.186**c**	75.9±1.17**ab**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

5.3.4 Effect of fertiliser input type and rate on grain macro nutrient concentrations

For **Grain Sulphur (S) concentrations**, analysis detected a significant main effect for fertiliser type only. Levels with CMP, FYM+CMP and GWC (0.15%) were higher than with FYM (0.14%) (Table 5.10). Significant interactions were detected between year and fertiliser type, and between year and fertiliser rate and type (Table 5.10). The effect of fertiliser type was significant only in one of the two years. There was no difference between treatments in 2006, but in 2007 the S concentration was increased by the addition of CMP (0.148%) and FYM+CMP (0.154%) (Table 5.11).

For **Grain Phosphorus (P) concentrations** analysis detected significant main effects for year only. Levels were higher in 2007 (0.39%) than in 2006 (0.33%). No significant interactions were detected (Table 5.10). For **Grain Potassium (K) concentrations** analysis detected a significant main effect for year only. Levels were higher in 2007 (0.55%) than in 2006 (0.45%). No significant interactions were detected (Table 5.10).

Table 5.10 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on spring wheat grain N, P and K content

	Grain S	**Grain P**	**Grain K**
	%	%	%
Year			
2006	0.15±0.001	0.33±0.003	0.45±0.004
2007	0.15±0.002	0.39±0.003	0.55±0.007
Fertility rate			
0 N	0.15±0.00	0.40±0.01	0.50±0.01
125 kg N ha^{-1}	0.15±0.001	0.36±0.006	0.50±0.01
250 kg N ha^{-1}	0.15±0.002	0.36±0.007	0.50±0.01
Fertility type			
CMP	0.15±0.001**a**	0.35±0.008	0.49±0.014
FYM	0.14±0.002**b**	0.30±0.01	0.48±0.013
FYM+CMP	0.15±0.002**a**	0.30±0.01	0.50±0.01
GWC	0.15±0.002**a**	0.36±0.009	0.49±0.012
ANOVA			
Main Effects			
YR	ns	**0.0008**	**0.0039**
FR	ns	ns	ns
FT	**0.0009**	ns	ns
Interactions			
YR:FR	ns	ns	ns
YR:FT	**0.0001**	ns	ns
FR:FT	ns	ns	ns
YR:FR:FT	**0.0015**	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $P < 0.05$)

Table 5.11 Interaction means for effects of year and fertiliser type on grain S % at Gilchesters farm

	Grain S%			
Fertility rate	**CMP**	**FYM**	**FYM+CMP**	**GWC**
125 kg N ha^{-1}	0.14±0.001**a**	0.14±0.002**a**	0.14±0.001**a**	0.14±0.001**a**
250 kg N ha^{-1}	0.14±0.002**b**	0.13±0.001**c**	0.15±0.003**a**	0.14±0.005**b**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, $P < 0.05$)

5.3.5 Effect of fertiliser input type and rate on grain micro nutrient concentrations

For **Grain Calcium (Ca) concentrations,** analysis detected a significant main effect for year only (Table 5.12). Levels were higher in 2007 (0.04%) than in 2006 (0.03%). No significant interactions were detected.

For **Grain Iron (Fe) concentrations** analysis detected a significant main effect for fertiliser type only. Levels were highest with FYM+CMP (38.5 mg kg^{-1}) and lowest with both FYM (35.8 mg kg^{-1}) and CMP (36.7 mg kg^{-1}). GWC was not significantly different to any of the other treatments (Table 5.12). Significant interactions were detected between year and fertiliser type, fertiliser type and rate, and year and rate and type. When interaction means between fertiliser input type and rate were compared, the FYM+CMP treatment had a significantly higher grain Fe concentration than all other treatments at 125 kg N ha^{-1} but at the high N treatment it was only significantly higher than the FYM treatment (Table 5.13). The effect of fertiliser type was significant in each of the two years. In 2006 Fe concentrations were higher with GWC (38.1 mg kg^{-1}) than FYM (36.7 mg kg^{-1}) at 250 kg N ha^{-1}. In 2007 at the lower fertility rate the addition of CMP to FYM significantly increased Fe concentrations with FYM (32.8 mg kg^{-1}) and CMP (36.1 mg kg^{-1}) alone to 39.7 mg kg^{-1}. At the higher rate of 250 kg N ha^{-1} the lowest concentrations were found in grains from FYM (Table 5.14).

For **Grain Zinc (Zn) concentrations,** analysis detected no significant main effects or interactions (Table 5.12).

Table 5.12 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on spring wheat grain Ca, Fe and Zn concentrations

	Ca	Fe	Zn
	%	mg kg^{-1}	mg kg^{-1}
Year			
2006	0.03±0.002	37.8±0.35	30.3±0.45
2007	0.04±0.001	36.1±0.75	31±0.41
Fertility rate			
0 N	0±0	37.2±1.21	31.1±1.13
125 kg N ha^{-1}	0.04±0.002	37.1±0.52	30.7±0.5
250 kg N ha^{-1}	0.04±0.001	37±0.6	30.5±0.36
Fertility type			
CMP	0.04±0.001	36.7±0.52**b**	30.7±0.33
FYM	0.03±0.001	35.8±0.75**b**	29.4±0.36
FYM+CMP	0.04±0.003	38.5±0.84**a**	31.7±0.88
GWC	0.03±0.002	37±0.9**ab**	30.4±0.61
ANOVA			
Main Effects			
YR	**0.041**	ns	ns
FR	ns	ns	ns
FT	ns	**0.0041**	ns
Interactions			
YR:FR	ns	ns	ns
YR:FT	ns	**0.0082**	ns
FR:FT	ns	**0.0234**[1]	ns
YR:FR:FT	ns	**0.0261**[2]	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 5.13. [2]For interaction means see Table 5.14

Table 5.13 Interaction means for effects of fertiliser type and fertiliser rate on wheat grain Fe content

	Grain Fe			
Fertility rate	**CMP**	**FYM**	**FYM+CMP**	**GWC**
125 kg N ha^{-1}	36.8±0.71**b**	36.6±1.22**b**	38.8±0.93**a**	35.7±1.18**b**
250 kg N ha^{-1}	36.6±0.83**ab**	35.0±0.9**b**	38.1±1.53**a**	38.3±1.25**a**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, p < 0.05)

Table 5.14 Interaction means for effects of year and fertiliser type and rate on grain Fe at Gilchesters farm

Year	**CMP**		**FYM**		**FYM+CMP**		**GWC**	
	125 kg N ha^{-1}	250 kg N ha^{-1}	125 kg N ha^{-1}	250 kg N ha^{-1}	125 kg N ha^{-1}	250 kg N ha^{-1}	125 kg N ha^{-1}	250 kg N ha^{-1}
2006	37.5±1.17 **a**	37.1±0.46**ab**	38.5±0.42**a**	36.7±0.61**b**	37.8±1.27**ab**	38.5±1.60**ab**	37.6±0.71**ab**	38.1±1.53**a**
2007	36.1±0.79**b**	35.8±1.89**bc**	32.8±0.40**d**	32.8±0.89**d**	39.7±1.35**a**	37.6±3.31a**bc**	33.3±1.87**cd**	38.9±3.01**ab**

Means labelled with the same letter within the same row are not significantly different (Tukey's honestly significant difference test, p < 0.05)

5.3.6 Effect of fertiliser input type and rate on grain phytic acid

For **Grain Phytic acid content**, analysis detected no significant main effects and no significant interactions (Table 5.15). For **grain Phytic acid P content**, analysis detected significant main effects for year only. Levels were higher in 2006 (83.1%) than in 2007 (72.7%) (Table 5.16). No significant interactions were detected. For **grain Phytic acid/ Zinc (Zn) molar ratio,** analysis detected no significant main effects or interactions (Table 5.15). For **grain Phytic acid/ Iron (Fe) molar ratio**, analysis detected no significant main effects and no significant interactions (Table 5.15).

Table 5.15 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain phytic acid concentration, phytic acid P % and phytic acid Zn and Fe molar ratios

	Phytic acid	**Phytic acid P**	**Phytic acid Zn**	**Phytic acid Fe**
	mg g^{-1}	%	molar ratio	molar ratio
Year				
2006	9.8±0.13	83.1±0.98	32.1±0.63	21.9±0.3
2007	10.1±0.18	72.7±1.21	32.4±0.7	23.9±0.6
Fertility rate				
0 N	9.7±0.32	75.8±1.14	31.2±1.96	22.2±1.15
125 kg N ha^{-1}	9.7±0.15	77.5±1.53	31.7±0.7	22.3±0.41
250 kg N ha^{-1}	10.1±0.16	79.8±1.35	32.8±0.58	23.2±0.53
Fertility type				
CMP	9.7±0.2	77.4±1.78	31.3±0.72	22.3±0.52
FM	9.9±0.19	78±1.65	33.3±0.78	23.5±0.8
FM+CMP	10±0.24	79.2±2.08	31.5±1.1	22.1±0.79
GWC	10.1±0.23	80.1±2.78	33.1±1	23.2±0.47
ANOVA				
Main Effects				
YR	ns	**0.0101**	ns	ns
FR	ns	ns	ns	ns
FT	ns	ns	ns	ns
Interactions				
YR:FR	ns	ns	ns	ns
YR:FT	ns	ns	ns	ns
FR:FT	ns	ns	ns	ns
YR:FR:FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

5.3.6 Effect of fertiliser input type and rate on grain heavy metal concentrations

For **Grain Molybdenum (Mo) concentrations**, analysis detected significant main effects for year and fertiliser type but not rate. Levels were significantly higher in 2007 (0.4%) than 2006 (0.1%), and highest with applications of FYM (0.3%) (Table 5.16). Significant interactions were detected between year and fertiliser type, between fertiliser type and rate, and between year, fertiliser type and rate (Table 5.16). When interaction means between fertiliser type and rate were compared, the FYM treatment had the highest Mo concentrations at both N rates. At high N it was significantly higher than with all other fertiliser types but at low N (125 kg N ha^{-1}) it was only significantly higher than the CMP and FYM +CMP treatments (Table 5.17).

For **Grain Aluminium (Al) concentrations,** analysis detected no significant main effects or significant interactions (Table 5.16).

For **Grain Cadmium (Cd) concentrations** analysis detected significant main effects for year and fertiliser type but not rate. Levels were significantly higher in 2006 (44.5 μg kg^{-1}) than 2007 (26.2 μg kg^{-1}), highest with applications of GWC (39.7 μg kg^{-1}) and lowest with FYM (32.8 μg kg^{-1}) (Table 5.16). No significant interactions were detected. For **Grain Nickel (Ni) concentrations** analysis detected significant main effects for year only. Concentrations were higher in 2007 (142.8 μg kg^{-1}) than in 2006 (105.6 μg kg^{-1}). No significant interactions were detected (Table 5.16). There was also a large difference in Ni concentrations between fertiliser rates but this was not significant. For **Grain Lead (Pb) concentrations** analysis detected significant main effects for year only. Concentrations were much higher in 2007 (116.1 μg kg^{-1}) than in 2006 (39.2 μg kg^{-1}). Significant interactions were detected between fertiliser type and fertiliser rate (Table 5.16). When interaction means were compared Pb concentrations at low N were significantly higher for the FYM + CMP than both the FYM and GWC treatments, but at high N there was no significant difference between treatments (Table 5.17).

Table 5.16 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain heavy metal concentrations

	Mo	Al	Cd	Ni	Pb
	mg kg^{-1}	mg kg^{-1}	µg kg^{-1}	µg kg^{-1}	µg kg^{-1}
Year					
2006	0.1±0.01	2.4±0.32	44.5±1.16	105.6±7.55	39.2±4.92
2007	0.4±0.04	3±0.27	26.2±1.67	142.8±4.54	116.1±9.87
Fertility rate					
0 N	0.3±0.11	2.3±0.40	32.7±4.70	118.5±12.75	59.5±17.47
125 kg N ha^{-1}	0.2±0.03	2.7±0.21	36.7±2.18	130.8±8.50	71.9±10.37
250 kg N ha^{-1}	0.2±0.04	2.7±0.4	36.5±2.26	111.6±5.74	72.4±10.10
Fertility type					
CMP	0.2±0.02**b**	3.1±0.49	36.7±2.84**b**	127.5±7.24	81.7±14.38
FYM	0.3±0.09**a**	2.2±0.11	32.8±3.74**c**	114.4±7.35	72.3±14.45
FYM+CMP	0.2±0.03**b**	2.9±0.5	37.1±2.9**b**	129.2±15.27	83.7±14.7
GWC	0.2±0.05**b**	2.5±0.49	39.7±3.11**a**	113.1±9.95	47.8±13.02
ANOVA					
Main Effects					
YR	**0.0082**	ns	**0.0151**	**0.0286**	**0.0035**
FR	ns	ns	ns	ns	ns
FT	**<0.001**	ns	**0.0245**	ns	ns
Interactions					
YR:FR	ns	ns	ns	ns	ns
YR:FT	**<0.001**	ns	ns	ns	ns
FR:FT	**0.0232**[1]	ns	ns	ns	**0.0157**[1]
YR:FR:FT	**0.0095**	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1] see Table 5.17 for interaction means.

Table 5.17 Interaction means for effects of fertiliser type and fertiliser rate on wheat grain molybdenum and lead heavy metal concentrations

		Mo		
Fertility rate	**CMP**	**FYM**	**FYM+CMP**	**GWC**
125 kg N ha^{-1}	0.5±0.06**b**	0.8±0.09**a**	0.6±0.07**b**	0.7±0.08**a**
250 kg N ha^{-1}	0.4±0.04**d**	0.8±0.09**a**	0.5±0.06**c**	0.7±0.09**b**
		Pb		
125 kg N ha^{-1}	75.4±9.86**ab**	64.9±11.81**b**	94.9±12.23**a**	51.4±9.77**b**
250 kg N ha^{-1}	78.1±12.3**a**	77.6±9.94**a**	80.6±14.9**a**	85.7±10.82**a**

Means labelled with the same letter within the same rows are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

5.4 Discussion

In both years at Gilchesters, according to the fundamental principle of building fertility in organic farming systems, spring wheat crops followed 2 to 3 year grass/clover leys ploughed out in spring. The site had a history of long term organic management alongside the inoculated red clover treatment. This results in high SNS (Soil Nitrogen Supply) index (equivalent to ~3 to 5 according to MAFF, 2000) and high residual fertility, which may influence the effectiveness of additional organic fertility inputs, e.g. manures and composts applied prior to ploughing out the leys (Nicholson *et al.*, 2017).

The supplementary organic manure inputs in this experiment were made with materials permitted under organic farming standards (Anon, 2009): green waste compost, FYM (cattle) compost and poultry manure compost (note: applied as chicken manure pellets for practical reasons, i.e. availability, uniformity and ease of application). Each fertiliser provides different concentrations of N and amounts of readily available N, potentially available for rapid crop uptake and in addition to the fertility levels of the inoculated clover leys (Loges *et al.*, 2006; Zhu-Barker *et al.*, 2015; Pandey *et al.*, 2017). From the analysis conducted all the materials are low in available N with the exception of CMP, which is much higher in ammonium nitrogen and therefore the most likely of the materials to provide a readily available source of nitrogen fertiliser during the growing season. This ammonium N may also be liable to higher levels of losses from the soil plant system following volatilisation as the pellets were applied to the soil surface during early vegetative growth.

The results of this study found that fertility rate and type had no significant effect on the disease levels or on yield when compared with the control. What is noteworthy is the variety Paragon, selected for this experiment, had lower disease levels across the trial plots than for the adjacent plots in the Chapter 4 variety trial, particularly in 2007 where the disease levels increased due to the adverse/less favourable environmental conditions. The fertility treatments did result in significantly higher SPAD readings in 2007 than the control, but not in 2006. The much lower SPAD values in 2007 are likely in response to a) higher disease levels and b) lower uptake of available N, both of which are likely to have been key factors contributing to the much lower yields in 2007 when compared with 2006.

In 2006 and 2007, yield of Paragon did not invariably respond to the applied fertility inputs (type or level). Positive fertility building over several years of organic management with the pre-crop inoculated red clover ley and the availability of mineralised N from a variety of sources and inputs (residual + supplementary from applied manures) was sufficient to support crop growth and yield, as has been observed in a number of other long-term studies. A Waste and Resources Action Programme (WRAP UK) experiment conducted in Scotland concluded that the benefit of compost (GWC) amendments to crop yields became apparent only after 3 years and then was sustained over the 7 years of the trial through continued applications (Nicholson *et al.*, 2017). The 30 year bio-Dynamic, bio-Organic, Konventionell (DOK) trial at FiBL in Switzerland found first year wheats in an organic rotation received no benefit from addition of composted fertilisers, but they did make an impact on second year wheat quality, subsequently in the rotation. They also concluded that grass/clover leys in particular supplied substantial amounts of nutrients over a prolonged period following incorporation (Mäder *et al.*, 2007), which was in line with soil N figures reported in section 2.4.2 of this study.

Nitrogen availability is affected by factors such as soil temperature and rainfall and in particular Excess Winter Rainfall (EWR) where N mineralisation from organic fertilisers is slower when conditions are cool and/or dry (L-Baeckström *et al.*, 2004). In 2006, which was generally a very much drier season than 2007, there were no significant yield differences between any of the different types and levels of fertility inputs, or against the control. The lower temperatures in 2007 may have contributed to lower N mineralisation with lower N availability and uptake. The yields were much higher in 2006 at 5.5 t ha^{-1} than in 2007 at only 2.5 t ha^{-1} but were similar to the adjacent trial plots of Paragon from the previous experiment in Chapter 4 conducted concurrently (6.5 & 2.6 t ha^{-1}.) Of the four fertility treatments the use of CMP resulted in higher yields (4.4 t ha^{-1}) than FYM with the CMP amendment (3.8 t ha^{-1}). Although the CMP gave the highest yield, the higher availability of NH_4-N in this treatment was generally not reflected in a more readily available source of N for crop uptake as reflected in little difference in SPAD values between the different fertiliser types.

Neither the fertility rate nor type had any effect on plant height and when the crops were taller in 2006 than 2007 there was no yield advantage from the taller plants in 2006.

Thousand grain weights were higher in 2007 than in the previous year and the use of the CMP resulted in the highest TGW values. The addition of CMP to the FYM also improved the overall grain weights when compared to the FYM treatment alone. At the lower application rate of 125 kg ha^{-1} the TGW was lowest from just FYM, whilst at the higher rate of 250 kg ha^{-1} TGW was higher with the single application of CMP or FYM than with FYM+CMP combined or with the GWC. Both the yield and TGW increase associated in this study with the additional application of CMP are in line with other similar studies which identified improvements in both the conditions for fertilisation and a stimulating effect on soil microbial activity from poultry manure applications (Delgado *et al.*, 2011).

The specific weights of grains were higher in 2006 than in 2007 and the effects of the treatments were significant for both fertiliser rate and type. The specific weights were higher with either the GWC or the FYM and lowest with the CMP and FYM plus CMP. At the lower fertility rate grain specific weight was highest from the GWC (after amended red clover) and lowest from the FYM +CMP (after amended red clover). At the higher application rate the FYM+CMP grains still had the lowest specific weights. The impact of GWC on agricultural soils was found to be less pronounced on clay loam soils than on sandy soils, but GWC amendments in this study (on clay loam soils) were still able to positively influence grain quality (Zhu-Barker *et al.*, 2015). The addition of GWC after red clover produced grains with the highest specific weight whilst the addition of the FYM with the CMP, or the CMP alone did not have the same effect, producing grains with the lowest specific weights. Conversely the GWC and the FYM alone were producing grains with the lowest protein percentages, whilst the CMP or FYM+CMP grains had the highest. This suggests that the dilution effect on protein levels from the larger grains produced with the GWC and FYM is what is being recorded, with the higher protein levels for the FYM+CMP being achieved from the higher levels of available N being provided by the CMP application. However results from Halle's 'Eternal Rye Cropping' experiment reported by Schmidt *et al.* (2000) found no effect of fertilisation treatments on specific weight and thousand grain weight or on processing quality of the flour as determined by HFN, which they concluded was influenced mainly by environmental conditions. Comparative studies in the US also found no significant effect of fertility management on wheat specific weights, observing that where yield increases did occur associated with fertility-building green manures in the cropping rotation then so too did specific weights (Murphy *et al.*, 2007). A significant genotype x management system interaction was established in comparative organic and conventional trials, lending weight to the concept of organic cereal production benefitting from varieties better adapted to low input systems. Results from the Swiss DOK trial could also find no effect of input on TGW or grain specific weights (Mäder *et al.*, 2007). ADAS/DC-Agri trials on composts and manures identified the synchronisation of N availability with crop nutrient demand to be a major challenge in organic crop production systems compounded by the fact, from their trials, that modern wheat cultivars have a relatively poor ability to take up nitrogen in organic compared to non-organic conditions (Nicholson *et al.*, 2017).

For organic management systems to produce grains with good baking quality characteristics there needs to be a positive interaction between variety choice and the fertility management system both before and during the cropping season (Carcea *et al.*, 2006; Mäder *et al.*, 2007). The protein content of wheat is an indicator of bread making performance. The field trials in this study included several factors which would be expected to have an influence on wheat protein content. From the two years of field trials (2006 and 2007) analysis detected a significant effect of the type of fertiliser on wheat protein content between treatments only, but not against the control. The late application of CMP on its own and when added to FYM produced the highest grain protein percentages, although this was not reflected in higher SPAD values at GS65. The later supply and uptake of nitrogen from the CMP treatment for utilisation during grain filling may be a potential reason for these differences. There was, however, no significant effect of fertiliser application rate. Results from the 30 year DOK experiment concluded grain protein from organic systems was generally high (compared to conventional), which they attributed to better selection of varieties better adapted to low N input (Mäder *et al.*, 2007). The study into organic and conventional spring wheats between 1995 and 2002 identified weather conditions as the most important factor in determining baking quality (Hanell *et al.*, 2004). Grain quality in both years was good with proteins around 13%, HFN levels well in excess of 250 s and specific weights generally above 76 kg hl^{-1}, which shows the clear potential for growing organic spring wheat for bread making in north east England.

Spring wheat had a relatively short growth cycle of 4-5 months in this study. The limited response in terms of SPAD to the different fertiliser types supports the evidence that there was limited mineralisation and uptake of N during crop growth, which is reflected in the marginal treatment effects observed here. Evaluation of the same treatments applied prior to drilling but using winter wheat would have allowed greater time for mineralisation and uptake, potentially resulting in a greater treatment response than was observed in the current study.

This dilution effect observed on protein levels was also observed in the grain macro nutrient levels where the concentrations of P and K were higher in 2007, the year which resulted in smaller grains and lower yield than the previous year. This also applied to the concentration of the micro nutrient Ca, which was also higher in 2007. Only fertility type affected grain S concentrations, which were lower with the application of FYM than with the other fertilisers. Bhogal *et al.* (2016) identified repeated applications of GWC and FYM as a valuable source of P, K and S, without increasing soil or crop metal concentrations. Fe concentrations in this study increased where CMP was added to the FYM at both the lower and higher rates of application, which was also observed in other fertiliser rate studies which identified similar increases in Fe, Mg and Ca concentrations with increased fertility rates (Zebarth *et al.*, 1992).

5.4.1 Three farm trial site comparisons

When the type and rate of fertilisers were compared across the three trial sites for their impact on the wheat yield and grain quality significant differences were observed between growing seasons, varieties and sites. In 2006 and 2007, yield of Paragon did not invariably respond to the applied fertility inputs (type or level) (Table 5.18). This was probably because fertility previously built up over several years of 'organic' management and also the pre-crop grass/clover ley and the availability of N available from all sources (residual + supplementary from applied manures) varied. The latter is affected by factors such as soil temperature and water supply (N mineralisation from organic to 'inorganic' forms is slower when conditions are cool and/or dry, whilst soluble, inorganic N may be leached in wet conditions), ability of the crop to take up N, and weed competition (although competition was generally low). In 2006, which was generally a very much drier season than 2007, there were no significant yield differences between any of the different types and levels of fertility inputs at Gilchesters or Sheepdrove, but there were significant differences at Courtyard. Here yields were lower for FYM (3.8 t ha^{-1}) and FYM+CMP (3.9 t ha^{-1}) at the lower fertility rate of 125 kg ha^{-1} than at 250 kg ha^{-1} (4.1 and 4.5 t ha^{-1} respectively). On the other hand, in 2007 there were significant differences at Sheepdrove but not at Courtyard or Gilchesters (Table 5.18). At Sheepdrove, the zero control treatment, which received no supplementary manure or compost, yielded least (1.5 t ha^{-1}) whilst other treatments yielded approximately 3 t ha^{-1} with the exception of treatments which included chicken manure pellets at either 125 or 250 kg ha^{-1} N. These treatments yielded significantly less at about 2 t ha^{-1} but the reasons for the lower yields are difficult to explain as there was no lodging and there were no differences in weed competition or disease infection between treatments. Phytotoxicity might be a possible explanation, but is unlikely because the yield of the treatment combination of 125 kg ha^{-1} N FYM + 125 kg ha^{-1} N Chicken Manure Pellets was 3.1 t ha^{-1} and amongst the highest yields. (For all assessments see tables in Appendix 7).

Table 5.18 Effects means ± SE and *p* values of site, harvest year, fertiliser rate and type on spring wheat grain yield t ha^{-1}

		2006			2007		
Fertility		**Courtyard**	**Sheepdrove**	**Gilchesters**	**Courtyard**	**Sheepdrove**	**Gilchesters**
Type	**Rate**						
FYM	250	4.1±0.70	3.7±0.20	5.9±0.70	3.2±0.00	3.0±0.20**bcd**	2.6±0.40
FYM/CMP	250	4.5±0.40	3.7±0.20	5.0±0.40	2.9±0.50	3.1±0.20**d**	2.3±0.50
CMP	250	4.5±0.60	3.5±0.20	5.7±0.40	2.2±0.20	2.1±0.30**abc**	2.7±0.50
GWC	250	4.5±0.30	3.7±0.40	5.1±0.60	3.2±0.20	3.2±0.10**d**	1.9±0.90
FYM	125	3.8±0.60	3.8±0.30	5.4±0.50	3.3±0.30	3.0±0.30**cd**	3.0±0.80
FYM/CMP	125	3.9±0.90	3.8±0.40	5.6±0.50	3.1±0.20	2.6±0.20**bcd**	1.9±0.20
CP	125	4.1±0.90	3.6±0.30	5.9±0.80	3.2±0.10	2.0±0.20**ab**	2.9±0.20
GWC	125	4.3±0.20	3.7±0.30	5.7±0.40	3.3±0.20	3.0±0.10**bcd**	3.1±0.40
CTRL	0	4.2±0.30	3.7±0.20	5.7±0.60	3.1±0.30	1.5±0.20**a**	1.9±0.10
ANOVA							
Main Effect							
FT		ns	ns	ns	ns	**0.0001**	ns
FR		**0.0106**	ns	ns	**0.0527**	ns	ns
Interaction							
FR:FT		ns	ns	ns	ns	ns	ns

Means labelled with the same letter within the same rows are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table 5.19 Protein content (%) of spring wheat from different fertiliser rates and types grown at Courtyard, Sheepdrove and Gilchesters. Data is an average of 2006 and 2007

	250 kg N ha^{-1}			125 kg N ha^{-1}		
Fertility type	**Courtyard**	**Sheepdrove**	**Gilchesters**	**Courtyard**	**Sheepdrove**	**Gilchesters**
CMP	14.3±0.50**a**	15.7±0.10**a**	13.6±2.00**a**	12.6±0.30**a**	15.5±0.10**a**	15.2±0.10**a**
FYM	12.1±0.40**a**	14.3±0.10**c**	12.6±1.80**a**	12.2±0.60**a**	14.4±0.20**c**	11.0±2.40**b**
FYM+CMP	13.2±0.40**a**	15.2±0.10**b**	13.6±2.00**a**	12.2±0.40**a**	14.9±0.20**b**	15.4±0.10**a**
GWC	12.0±0.40**a**	13.9±0.20**d**	11.4±2.50**a**	11.8±0.40**a**	14.4±0.10**c**	12.7±1.80**b**

Within columns, means with the same letter are not significantly different ($p<0.05$) according to Tukey's HSD test

5.4.2 Three farm trial site redundancy analysis (RDA)

Correlations between farm sites, agronomic management and environmental factors with Paragon spring wheat yield and quality levels.

In the biplot shown in Fig. 5.1 correlations between organic agronomic practices at each site, their macronutrient supply and climatic (radiation, precipitation and air temperature) factors with their effect on grain yield and quality were observed. Weather data from each site taken from sowing date to harvest date, along with fertiliser types and rates with the total levels of applied N, P and K, were used as drivers. Most variation (34.18%) is explained by axis 1 and a further 31.08% by axis 2. For individual drivers 29.6% of the variation is explained by precipitation, 23.5% by Courtyard Farm, 16.1% by solar radiation and 6% each by temperature, Gilchesters and Sheepdrove (Table 5.20).

Yield and TGW were more closely positively correlated with solar radiation along the negative axis 2 and negatively associated with temperature and rainfall. Plant height, leaf greenness (by SPAD), thousand grain weight and yield were positively correlated with Gilchesters along the positive Axis 2, whilst grain protein (NIR protein) and gluten quality (F3+F4) were positively correlated with Sheepdrove along the positive axis 1 and negatively correlated with yield and thousand grain weight. Courtyard was negatively correlated with grain Zn and Fe concentrations but showed a strong positive correlation with phytic acid:Fe and Zn molar ratios along the positive Axes 1 and 2.

Fertility type chicken pellets at the higher rate of 250 kg N ha^{-1} was positively correlated with gluten quality at Sheepdrove farm. The GWC at both rates were more positively correlated to Courtyard Farm. The applied level of N (kg ha^{-1}) from spring fertiliser applications had a weak positive correlation to grain gluten quality, whilst the applied levels of P and K (kg ha^{-1}) from the fertiliser applications had a weak positive correlation to grain yield and thousand grain weight.

The climate drivers indicated a strong negative correlation between solar radiation and rainfall, as could be expected, which accounted for 29.6% and 16.1% of the variation (Table 5.20). Yield and TGW were strongly, positively correlated to solar radiation, whilst grain P, K and Ca concentrations were positively correlated to rainfall and temperature for the Paragon wheat grown in the trial.

The differences in the trial sites were shown by no positive correlation. Sheepdrove and Gilchesters had a strong negative correlation to Courtyard with the grain gluten quality being positively correlated to Sheepdrove and the yield parameters positively correlated to Gilchesters. Courtyard again was negatively correlated to grain nutrient levels, particularly Zn, with a positive correlation to Phytic acid:Zn molar ratio.

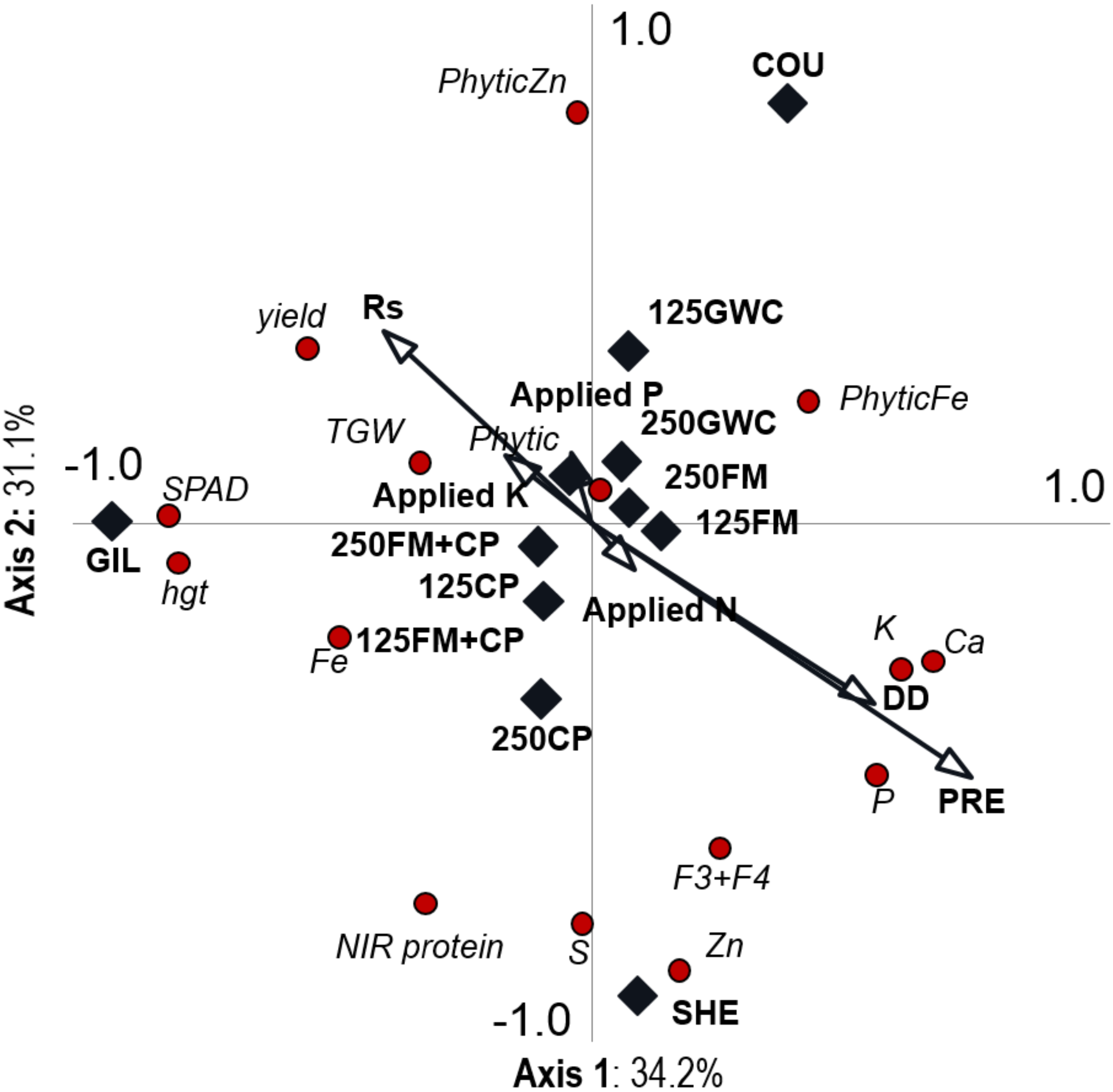


Fig. 5.1 Biplot derived from redundancy analysis (RDA) showing the relationship between weather conditions, agronomic management (fertility treatment and rate) applied nutrients and site drivers, with wheat grain harvest, quality and nutrient levels as the response variables. (Drivers: For climate Rs=solar radiation, Pre=precipitation, DD=temperature. For site COU=Courtyard Farm, GIL=Gilchesters Farm, SHE=Sheepdrove Farm. For fertility type FM= Farmyard Manure, GWC=Green Waste Compost, CP=Chicken Pellets and FM+CP=Farmyard Manure plus Chicken Pellets applied 50:50. For fertility rate 125=125 kg N ha^{-1}, 250=250 kg N ha^{-1}. Response variables: NIR protein=grain protein %, F3+F4=grain protein quality by SE-HPLC, Phytic=grain phytic acid content, PhyticFe=grain phytic acid:Fe molar ratio, PhyticZn=grain phytic acid:Zn molar ratio, hgt=straw height, yield=grain yield t/ha, TGW=thousand grain weight). Conditional Term Effects and P values in Table 5.21

Table 5.20 Variety trial RDA conditional term effects for site, weather and agronomic management Paragon spring wheat

RDA Driver	Explained variation %	Pseudo-F	P value
Precipitation	29.6	63.0	0.002
Courtyard	23.5	74.5	0.002
Radiation	16.1	77.4	0.002
Temperature	6.1	35.9	0.002
Gilchesters	6.1	35.9	0.002
Sheepdrove	6.1	35.9	0.002
250 CP	1.8	11.5	0.002
125 GWC	0.6	4.2	0.006
Applied P	0.7	4.8	0.002
Applied K	0.5	3.7	0.01
250 FM+CP	0.4	3.0	0.028
125 CP	0.3	2.0	ns
250 GWC	0.3	2.0	ns
Applied N	0.3	2.3	0.048
125 FM+CP	<0.1	0.6	ns
250 FM	0.1	0.9	ns

When the total fertiliser levels for applied N, P and K but not fertiliser type were used as drivers, alongside climatic conditions, site and soil nutrient levels, the results are represented in the biplot in Figure 5.2. Most variation (34.25%) is explained by axis 1 and a further 31.38% by axis 2. For individual drivers 29.6% of the variation is explained by precipitation, 23.5% by Courtyard Farm, 16.1% by solar radiation and 6% each from temperature, Gilchesters and Sheepdrove (Table 5.22).

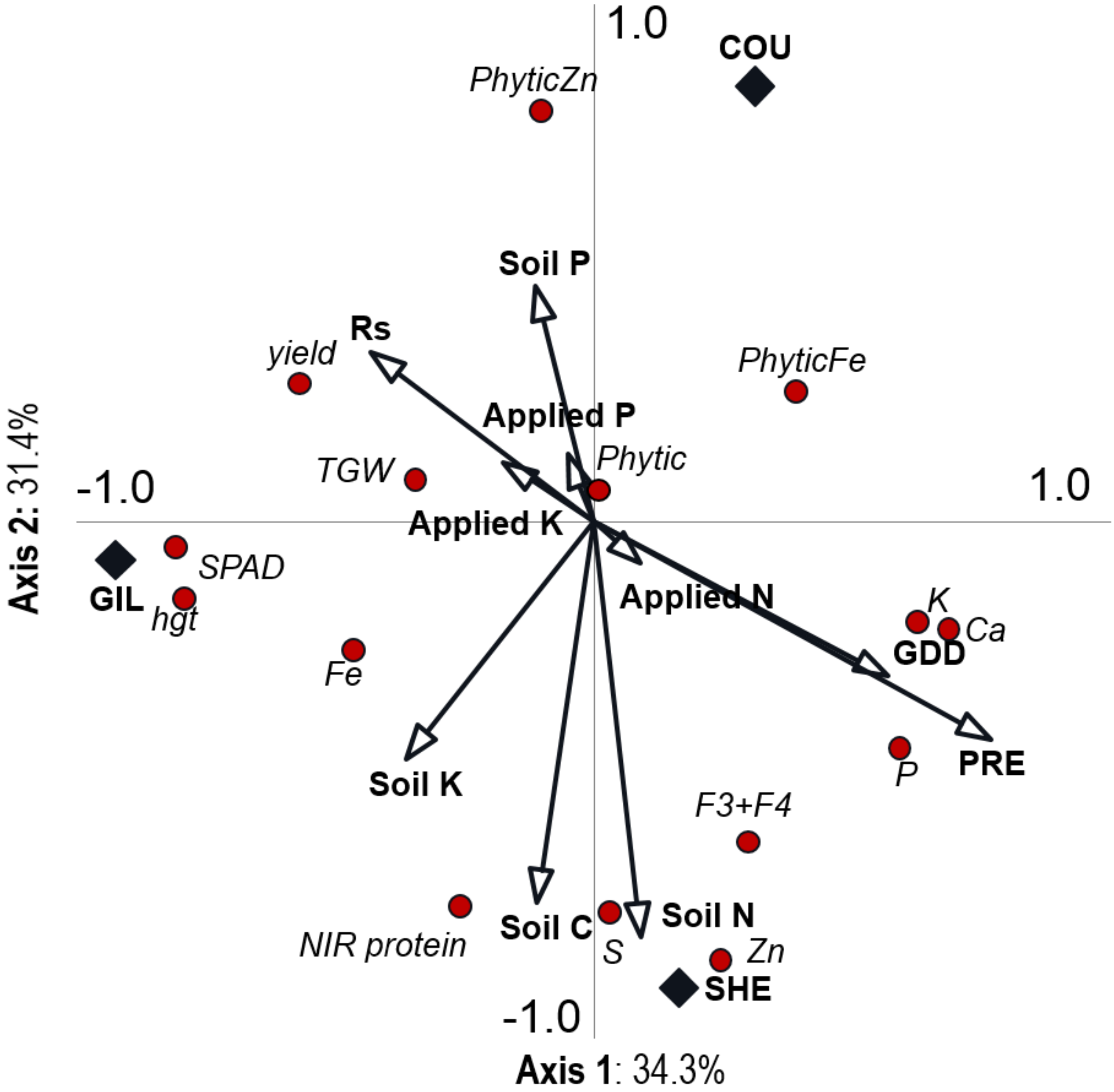


Fig. 5.2 Biplot derived from redundancy analysis (RDA) showing the relationship between weather conditions, agronomic management (fertility treatment applied nutrients) and site drivers, with wheat grain harvest, quality and nutrient levels as the response variables. (Drivers: For climate Rs=solar radiation, Pre=precipitation, GDD=temperature. For site COU=Courtyard Farm, GIL=Gilchesters Farm, SHE=Sheepdrove Farm. Response variables: NIR protein=grain protein %, F3+F4=grain protein quality by SE-HPLC, Phytic=grain phytic acid content, PhyticFe=grain phytic acid:Fe molar ratio, PhyticZn=grain phytic acid:Zn molar ratio, hgt=straw height, yield=grain yield t/ha, TGW=thousand grain weight). Conditional Term Effects and P values in Table 5.22.

Table 5.21 Variety trial RDA conditional term effects for site, weather and agronomic management Paragon spring wheat

RDA Driver	Explained variation %	Pseudo-F	P value
Precipitation	29.6	63.0	0.002
Courtyard	23.5	74.5	0.002
Solar Radiation	16.1	77.4	0.002
temperature	6.1	35.9	0.002
Gilchesters	6.1	35.9	0.002
Sheepdrove	6.1	35.9	0.002
Applied P	1.1	7.0	0.002
Applied N	1.0	6.2	0.004
Applied K	1.4	9.7	0.002
Soil K	0.6	4.4	0.004
Soil P	0.5	3.6	0.008
Soil C	0.2	1.3	ns
Soil N	<0.1	0.6	ns

5.5 Conclusion

In seasons favourable for the synchronisation of N mineralisation and crop nutrient uptake (such as in 2006) short term fertiliser applications provided no yield benefits. In years at the sites with less favourable environmental conditions yield increases of 0.27 t ha^{-1} were achieved. Applications of composted chicken manure did not improve yield or protein content at Gilchesters. Major seasonal differences between trial years resulted in high variability in yield and SPAD values demonstrating major differences in N uptake and availability.

Environmental conditions notwithstanding, short term applications intending to improve N from either rate had no effect on spring wheat yield or grain quality, nor were there any clear differences due to the type of fertiliser applied. As spring wheat has a shorter growth cycle than winter cereals, this may have reduced the synchronisation of N mineralisation and nutrient uptake of these organic fertilisers, thus compounding their ineffectiveness in this study still further.

CHAPTER 6: General Discussion

6.1 Organic wheat production and the UK market

The latest organic farming statistics (DEFRA, 2018) show an increase in organic farmland, the first since 2008, and an increase of land in conversion for the third consecutive year. There however continues to be a shortage of production of arable crops, in particular cereals, which remains an area of concern. UK organic wheat production stood at 40,000 Mt in 2016 (Eurostat, 2017). Seasonal variation resulted in a noticeable drop in yields along the eastern counties in 2017 and the delay in spring sowing combined with very dry conditions in the south of England in 2018 are signalling another reduction in yield with a potential impact on grain specific weights (AHDB, 2018). Demand for organic products continues to rise in the UK with strong growth in the bread sector being recorded. However, UK grain traders (Gleadell, 2018; Saxon, 2018) have reported a slowing of UK organic wheat purchases by the main UK organic millers over the last ten years. Saxon Grains particularly have seen a decline from 5000 t/month of organic cereals into mills during this period to zero in 2018, as mills become increasingly dependent on increasing volumes of imported Nabim classification Group 1 (breadmaking/milling) wheats. This in turn is leading to a decline in production of UK organic bread making wheat. Full wheat classifications are shown in Appendix 8 (Tables 8.1 to 8.5).

The organic bread making sector in the UK has been unable to meet the requirements of the market without utilising imported high protein wheat to augment the lower protein content of the home grown crop. The DEFRA Arable Link funded project Better Organic Bread (BOB) was established in 2005 to identify ways to improve wheat protein and bread making quality from UK organic wheat production. With the uncertainty of current trading freedoms and known pricing structures being held in the balance through Brexit, the need for consistent quality from UK grown organic cereals has never been greater.

6.2 Organic Production Methods

The use of clover and other nitrogen fixing legumes within the organic rotation has now become commonplace and has become well understood in the years since this was assessed in Chapter 3 prior to 2004. Manipulation of clover crops by mulching, removal for silage additions, grazing or complemented municipal green waste compost additions are practised extensively throughout the UK (Scott, 2015). Weeding regimes and the use of rotations have also been well researched and are implemented in a similar fashion in the diverse and complex husbandry practices employed by organic farmers and producers (Stenerud *et al*., 2015). However, the selection of wheat varieties for use within the organic rotation, especially where milling quality is a goal, is still heavily dependent upon varieties from the UK Recommended List where trials are carried out under a high input conventional management system. A small number of trials are carried out each year in the absence of fungicides and these offer the best potential for growers to identify suitable varieties for an organic production system. Organic production is limited by some agronomic and management requirements but particularly by the lack of organic seeds and varieties selected for organic conditions.

6.3 Pre-crop treatment and compost input

The first hypothesis, that a long term organic soil fertility management strategy can be found for the optimisation of nutrient availability for growing high protein, high gluten bread wheats in the north east of England, is confirmed by the results of this study. *Rhizobium* inoculation of red clover seed significantly increased establishment of clover and the number and size of root nodules on clover plants: yields of the subsequent wheat crops were also increased. On average, the yield increase was greater for winter wheat (+0.65 t/ha) than spring wheat (+0.26 t/ha). There were significant interactions between fertility management practices *(Rhizobium* inoculation x green waste compost) in both winter and spring wheat. Given the standard recommendations and general understanding prior to this trial of yield from winter wheat and quality from spring, this study clearly demonstrated the ability of the right fertility management to deliver high yielding organic breadmaking quality wheats from both winter and spring crops where the right varieties are selected. Gilchesters Farm's commercial use/exploitation of the 2003-2005 trial data continued with cereal production of the tall straw winter wheat varieties Pollux and Wenga sown after inoculated red clover with compost amendments as a source of baking quality wheat to supply the Gilchesters Organics flour mill, built in 2006. The 2006 and 2007 spring wheat trials (both variety and fertility management) in this study were sown alongside these commercial crops of winter wheat, which continued to provide high yields of milling quality grains from 2006 to 2009 ranging from 6 to 8.6 t ha^{-1}. The soil fertility after inoculated clover plus compost amendments was sufficient for Pollux to be grown as a first year wheat, followed by Wenga as a second year wheat. In 2007, the year of low yield and quality for the spring wheat trials, milling quality grains were produced from both Pollux in the surrounding fields and Wenga in the fields from the previous year's high yielding plots in 2006 (Table 6.1).

Table 6.1 Grain quality of commercial winter wheat varieties at Gilchesters 2007

Variety (Winter wheat)	Specific weight	Protein	HFN	Gluten Classification	Milling Classification
	kg hl^{-1}	%	s		NABIM Group
Pollux (1st wheat)	78.3	13.4	361	A	1
Wenga (2nd wheat)	77.3	11.6	317	B	2

Results from Coastal Grains, commercial organic grain store, Belford, Northumberland

The yield gap of 20-30% between organic and conventional wheat production (Hossard *et al.*, 2016) can be addressed with improved long term fertility management practices alongside varieties better suited for organic production systems. Continental European breeders (e.g. Saatzucht: PKGZ, Edelhoff, IG, Dottenfelderhof and Probstdorfer) now have a range of Group 1 and 2 hard winter wheat varieties akin to the earlier Pollux and Wenga selected for this study in 2003, which reinforces the importance of variety (Bilsborrow *et al.*, 2013) for organic quality wheat production. Full wheat classifications for Austrian and Germany are shown in Appendix 8 (Tables 8.2 to 8.5).

6.4 Variety choice

The second hypothesis, which proposed that milling quality wheat can be grown in the north east of England from winter wheat varieties as well as spring from tall straw varieties not presently on the UK national recommended variety list, is also confirmed by the results of this study. It has established the ability of selected winter wheats to produce yield and quality equal to organic spring varieties. In spring wheat grown according to organic standards for bread making, variety (genotype) also sets the potential for grain yield and quality. However, the proportion of this potential that is achieved depends on environment (weather, site effects including soil fertility and its management, water availability, disease profiles and overall standards of agronomic management by the grower). Yield and quality are therefore a function of the genotype x environment interaction. This applies equally to spring and winter varieties. The results of the trials in this study confirm that variety choice is the primary consideration for growing spring wheat in organic systems for bread-making as it is in conventional systems. Varieties to be grown should be chosen on the basis of their known characteristics, including yield, quality, agronomic characteristics and disease resistance.

However, organic growers, producers and grain processors (particularly those looking for niche markets) should look further afield than simply those varieties described in publications such as the HGCA Recommended List for cereals and oilseeds as their suitability to meet the requirements of the large scale millers and bakers might not be the same as for more local, artisan bakers who are often the outlet for organic wheat production. This could result in the possibility of missing an opportunity to evaluate grains better suited to their own requirements. Currently available varieties such as the NABIM Group 1 variety Paragon (HGCA, 2010, released in 1999) continue to meet these requirements in organic production systems. Although yield is significantly lower than most other varieties such as Tybalt and Granary (NABIM Group 2 released in 2003 and 2009 respectively), it still performs well against the newer variety Mulika (NABIM Group 1 released in 2011). Recent evaluations co-ordinated by UK organic seed merchants at three UK sites of tall straw varieties Ehogold (BQ 8 released 2014) and Edelmann (BQ 7 released 2017) from Saatzucht Edelhoff (Austria) along with Senaturo (Group A released 2017) (Saatzucht IG, Germany) continue to assist in the search for high quality varieties suitable for the organic sector. A continued process of evaluation for such varieties which maintain their position over a wide range of weather conditions, geographic locations and fertility management strategies is vital for the long term success of the organic sector.

In the variety evaluation trial (Chapter 4), although 2006 and 2007 were very different years in terms of weather, disease levels and grain yield there was a high degree of consistency between the years in terms of grain quality. When grain yield was much lower in 2007 this was not reflected in a reduction of grain quality. Protein levels ranged between 13 and 15 %, protein quality was higher than in 2006 as were HFNs and where a reduction in specific weights and gluten quality did occur these were still within the tolerances required for milling quality. This consistency in grain quality offers clear potential for organic growers in Northeast England to produce high quality organic spring wheats as long as they can get good fertility management in their rotation. The northeast is generally an area which only has a small amount of conventional breadmaking wheat grown, which is mostly due to the weather and risk of not achieving sufficient grain quality, in particular HFN.

6.5 Organic manure inputs (type and rate)

Yields in organic wheat production systems are dependent on the availability of N mineralised from soil organic matter, animal manures and other organic matter-based inputs such as legume crops in the rotation or green waste compost amendments. Doltra *et al.* (2011) and Mader *et al.* (2007) showed that the yield of wheat was 14% lower in an organic versus conventional production system in a 21 year agro-system comparison in central Europe and largely attributed this to a 71% reduction in soluble nitrogen input to the organic system. Insufficient N supply in organic farming systems is primarily due to (a) insufficient organic fertiliser inputs and (b) a mismatch between crop demand and supply (Pang and Letey, 2000), especially during key periods like grain filling which can affect grain yield and quality (Bilsborrow *et al.*, 2013).

The synchronisation of nitrogen availability with crop nutrient demand has been shown to be a major challenge in organic production systems. Most inputs of organic N are dependent on a high level of microbial activity to release available N through mineralisation. However biogas digestate is creating considerable interest among commercial organic growers (vegetable growers G's Fresh of Ely, Cambridgeshire) with the ability to supply a source of readily available N to a standing crop and thereby increase both grain yield and quality. The use of biogas digestate is currently permitted in the UK under a Derogation from the Certification Bodies.

Just as in conventional systems, significant increases in yield in response to fertiliser inputs are not guaranteed. However, in the trials that assessed the effects of type and level of fertility inputs that may be used in organic systems of production, yields of spring wheat were improved in some cases. Where high fertility levels exist and are maintained within an organic rotation grain quality is determined more by variety choice and environmental factors. Although fertility treatments may not show responses or only small responses in the current season of application – because of inherent fertility built up previously over the rotation – these will have cumulative effects which will benefit crops later in the rotation as organic N becomes mineralised. The fundamental principle of fertility management in organic systems is to build soil fertility within the rotation over the long term. This would include, for example, grass/clover leys, forage and grain legumes and manure/compost inputs, preferably generated within the system rather than 'brought-in'. This would maintain a closed nutrient cycling system as far as possible and at strategic points, support more exploitative crops in the rotation such as wheat (HGCA, 2008). Therefore, the cumulative effects of the fertility management strategies are productivity drivers: crops later in the rotation should benefit from current fertility inputs. For example, a spring wheat crop immediately following a grass/clover ley may not respond significantly to current inputs of manures or composts at rates of 170 kg N ha^{-1} (or up to a maximum of 250 kg N ha^{-1} on a single field) because of inherent fertility built up previously over the rotation, but cumulative effects should benefit later crops in the rotation as organic N becomes mineralised. Furthermore, long-term studies have identified that where rotations maintain adequate fertility and a two year grass/clover ley precedes spring wheat, significant yield benefits from short-term compost applications are unlikely in the first year after incorporation of the ley. This represents a long-term strategy rather than a short-term one aimed specifically at a cash crop such as milling wheat for example, which appears only once every 5 or more years in the rotation.

6.6 Baking and milling quality

Baking trials from the different spring wheat varieties milled as either stoneground or roller milled flours produced interesting results for both wholemeal and white bread. Although grain hardness was not measured (and seldom is) when assessing milling quality, the fractured starch produced results that were inconsistent with the grain quality assessments. Tybalt and Fasan had the best grain quality according to the industrial standards set by NABIM in the UK, yet Tybalt protein structure and gluten quality were insufficient to produce commercial loaves of wholemeal flour and only an adequate loaf volume with white flour from roller milled flour. The performance of both flour streams improved the bread volumes when stoneground. Paragon produced consistent loaf volumes from roller milled and stone ground flour, whilst Amaretto produced larger loaf volumes with wholemeal and white flour from the stoneground samples, compared to roller milled.

These results suggest that the industrial benchmarks for quality accepted over many years for the commercial requirements of industrial roller milling and high speed baking processes are possibly resulting in the mis-allocation of UK organic wheats, which are capable of producing grains with good gluten quality that perform well after stone-grinding for the needs of smaller artisan bakery businesses.

6.7 Nutritional and mineral delivery

The levels of phenolics and tocopherols assessed from the winter and spring varieties identified a higher delivery of tocopherols from the winter varieties over the spring, but neither had levels that were outside the range observed in wheats from other studies. The variety and fertility management options in this study confirm the maintenance of these anti-oxidants in our diet through these organic production systems. Phytic acid levels were not significantly affected by the fertility management or variety choices, but whilst their inclusion in wholemeal flours from stone milling remains present, their inclusion in white flour streams from both roller and stone milling can be affected by grain hardness at the point in the milling process where bran and aleurone layers become separated from the endosperm. Grain hardness is a measure of protein packing and density and therefore quality. This resistance to milling, which affects not only the quality of fractured starch in the white flour but also determines the cleanness of the separation of the aleurone layer, can be affected by nutrient availability to the growing crop. Organic soils with high N levels will improve the grain hardness of hard wheats grown for milling and should be a consideration when the end use is for stone milling or alternatively roller milled white flour production.

Fe and Zn are important nutrients with respect to human nutrition and health with considerable research effort in recent years to evaluate the potential for increasing the uptake of these in cereal based diets. Wheat has relatively low concentrations of these important nutrients compared with other crops and cereals (Cakmak, 2018) which has therefore attracted particular attention for bio-fortification. From this study the different fertilisers used had little effect on the concentrations of Fe and Zn in wheat grain and there was little variation evident between the different varieties, except for Fe in one. But what is of considerable interest is how the processing of flour (roller milled vs stone ground, wholemeal vs white) can potentially influence the bioavailability of these elements in the diet; this is an area for further research..

6.8 UK Trial Site Comparisons

When the results from the Gilchesters site were compared with the other two UK trial sites (Sheepdrove and Courtyard) overall, site specific differences were recorded within the varieties for yield and grain quality. Paragon was the top yielding variety at Gilchesters in both years followed by Amaretto in 2006 and Fasan in 2007. However, at both Courtyard and

Sheepdrove, Tybalt was the highest yielding variety in both years, with Paragon second highest at Sheepdrove in 2007. Zebra was the lowest yielding variety at all sites in both seasons with the exception of Courtyard in 2006 where Amaretto was the lowest. So the overall results show clear differences between sites and seasons in ranking of the different varieties.

However, the RDA analysis results for the comparative data from all three sites demonstrated that the biggest drivers for grain quality were associated with the soil fertility at each farm and less so with the varieties at and between the sites. The effect of management practices to improve and maintain soil quality within organic farming systems produces long term benefits to crop production that are more closely associated with grain and soil nutrient levels than immediate seasonal crop fertilisation regimes.

Grain protein concentrations were higher at Gilchesters than the other two sites in 2006, with Fasan having the highest concentrations in 2006 and Tybalt in 2007, followed by Zebra and Paragon. At Sheepdrove Zebra had the highest protein concentrations in both years, with Monsun second in 2006 and Paragon and Monsun second in 2007. At Courtyard, which had the lowest protein levels in 2006, Paragon had the highest in 2006 with Zebra second. In 2007 Tybalt had the highest with Zebra and Paragon second. There were no consistent trends in protein across the sites but Zebra, which had low yields across the sites with good protein levels, does confirm our general understanding of the relationship between yield and quality.

For the fertility trial increases of the order of 0.5 to 1.0 t ha^{-1} were obtained by increasing N rate, in particular by using FYM based compost and chicken manure pellets. At Sheepdrove in 2007 a yield response of ~1.0 t ha^{-1} to 125 and 250 kg ha^{-1} N was recorded. These yield increases from the treatments on the lighter soils at the southern sites were in contrast to the Gilchesters results. Thus whether a response is recorded, and the magnitude of the response, will depend on several factors: the inherent or residual fertility of the soil to which the fertility input is applied; the amount of N present in the soil which is readily available (NO_3^-, NH_4^+) for crop uptake in the current season; and the potential for mineralisation of soil N, which will depend upon soil microbiological activity and be influenced by soil temperature, water supply, soil health etc. Indeed, the responsiveness to applied fertility inputs is likely to be influenced by the general growing conditions and expected to be better when they promote vigorous growth (although a counter-argument is that when growth is suppressed by adverse conditions, supplementary fertiliser should stimulate it).

Analysis of wheat protein content data for all sites and all years of the variety Paragon, to which fertility treatments had been applied, indicated that there was a significant effect of site. Although mean differences between sites could vary from 14.3% (Gilchesters) to 11.5% (Courtyard) the strongest association by RDA analysis was between the high soil N at Sheepdrove and wheat protein and gluten quality. The analysis found no strong associations at all between fertility treatments and grain protein or gluten quality. With only weak evidence (p=0.02) of a site and treatment interaction at Courtyard, it suggested that the increase in protein content recorded when the CMP treatment is applied would not be seen at a higher fertility site such as Gilchesters or Sheepdrove.

CHAPTER 7: Conclusions

7.1 Conclusions

Organic farming systems are based on maintaining soil and crop productivity by selecting management approaches which maintain adequate soil fertility, encourage biodiversity and minimise external inputs. In order to supply the crop with sufficient nitrogen to enable a milling wheat crop to realise its optimum protein content, long term fertility building strategies are required. In this project selected strategies were examined to quantify the potential for increasing available nitrogen to winter and spring sown wheat on organic farms.

The results indicate that inoculated red clover swards with and without composted green waste amendments can improve both yield and quality of wheat crops and that variety choice offers potential for achieving milling grade quality. The winter varieties Pollux, Wenga and Greina along with the spring varieties Paragon, Tybalt, Fasan and Amaretto are capable of producing adequate protein levels when grown in different regions.

The use of composted materials based on farm yard manure and green waste can provide an increase of up to 0.5 t ha^{-1} in yield where lower fertility conditions exist, but where adequate background fertility has been maintained by a two year grass/clover ley, additional yield benefits from compost applications are generally small. The use of supplementary organic fertility inputs based on chicken manure pellets resulted in a yield benefit of up to 0.5 t ha^{-1} only at sites with lower soil fertility. In addition to this yield effect, protein content was increased by up to 1% in chicken manure treated plots. As expected, site and season had a significant effect on yield and wheat protein content. Large yield differences were the result of changes in spring and summer weather conditions, with 2006 providing better yields compared to 2007 when conditions were cool, dull and wet. Yield loss resulting from foliar disease was greater at the site with lighter soil. Differences in wheat protein content between seasons contributed up to 2% and between sites up to 2.5%.

The understanding that good baking quality of wheat will only be attained if the grain meets the criteria of adequate protein content (13%), hectolitre weight (76 kg/hl) and Hagberg Falling Number (250 s) is dependent on the baking process used. Fertility management and variety choice can enable protein content targets to be attained but baking quality is also affected by radiation levels (affecting grain size) or wet weather at harvest (increasing *alpha*-amylase levels in the grain). The results of the milling and baking trials from this study showed that the choice of wheat variety did not consistently increase loaf volume. Large differences in loaf volume arose from the effect of variety and season (up to 30% difference between seasons) which cannot be explained entirely by variation in hectolitre weight, HFN or protein % as environmental factors in organic production affect the packing density and quality of grain proteins differently for each variety.

7.2 Further work

Gilchesters Organics continues to produce organic stoneground flours principally from a range of organic tall straw winter wheat varieties grown in the North East. We have a particular interest in growing older varieties/landraces (e.g. the spelt variety Oberkulmer Rotkorn) which have become adapted to their own environment. Demand is now growing for specialist cereals where both flavour and functionality are of primary importance to the customer, but without a recognised premium. Without this premium many organic farmers are reluctant to grow cereals if yields are considered low or if milling quality cereals are in low demand due to downward market pressure from the larger UK mills or with a perception that quality is difficult to achieve. This negative perception amongst organic farmers requires a change in the way organic cereals are assessed for quality and subsequently processed and marketed. This requires a better understanding of the potential baking quality of organic wheat from their protein and gluten structures and for artisan bakers and chefs to recognise the value of these grains beyond their current pure commodity status and value.

Further research of interest would be:

An evaluation and comparison of landraces, populations, heritage and new organic breeding lines vs modern cultivars, particularly for winter cereals, as the basis for improved milling quality.

An evaluation of grain quality in relation to grain hardness in organic cereals and its impact on baking quality from stoneground flours used in artisan bread production.

An evaluation of the effects of variation in grain quality on baking quality from an artisan and stone milling perspective.

A marketing study to assess the value and potential of specialised UK wheat products similar to the way high value spelt, emmer and einkorn grains are sold as specialist cereals in the market.

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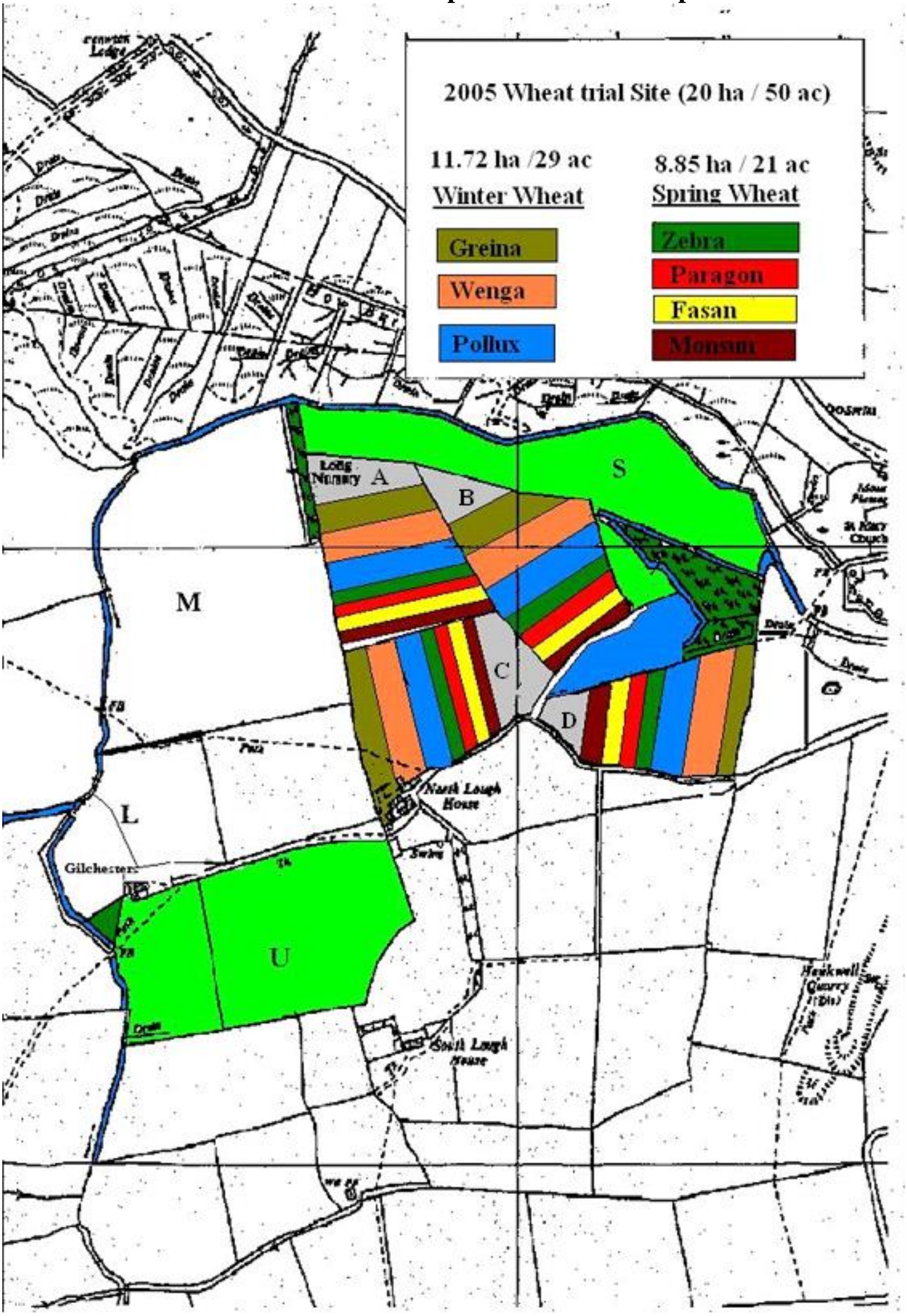
APPENDIX 1: UK Meteorological Data 2006 and 2007

Table A1. Meteorological data for Courtyard (COU), Sheepdrove (SHE) and Gilchesters (GIL) farms, 2006 and 2007

	Mean Temperature			**Total Rainfall**			**Solar Radiation**		
	°C			mm			kW m^{-2}		
	COU	**SHE**	**GIL**	**COU**	**SHE**	**GIL**	**COU**	**SHE**	**GIL**
2006									
April	8.7	8.2	6.9	38.0	32.0	21.8	113.5	107.1	44.0
May	12.7	11.9	9.7	89.0	112.8	74.4	135.1	132.6	53.4
June	16.1	15.9	14.0	9.6	8.0	27.8	177.6	182.2	57.3
July	19.3	19.7	17.4	32.4	30.0	13.2	192.6	186.3	64.9
August	16.3	15.9	14.4	93.4	16.2	60.0	112.6	124.4	39.5
September	17.6	16.2	14.8	71.0	96.8	71.4	96.5	92.5	29.7
2007									
April	11.0	10.9	9.3	2.8	4.8	13.4	131.5	91.5	43.6
May	12.2	11.5	10.0	111.2	130.8	50.6	118.7	85.4	52.8
June	15.0	15.0	12.9	129.0	119.6	84.9	146.4	79.7	19.2
July	16.2	15.1	13.9	113.2	189.4	10.8	145.0	142.9	19.6
August	16.4	15.7	14.0	53.4	31.8	35.8	134.1	122.6	44.3
September	14.8	13.7	12.3	37.6	24.8	23.2	101.0	95.1	29.5

Data supplied by UK Met office

APPENDIX 2: Chapter 3 Trial site map



APPENDIX 3: Pictures from field trials

Winter and Spring Wheat trials 2005

Photo.1 21.04.2005 Field A spring wheat establishment

Photo.2 21.04.2005 Field B winter wheat

Greina Wenga

Photo.3 21.04.2005 Field B spring and winter wheat

Zebra Pollux

Photo.4 21.04.2005 Field C winter wheat

Wenga Greina

Photo.5 21.04.2005 Field C winter and spring wheat

Pollux foreground, spring wheat plots establishment

Photo.6 21.04.2005 Field D winter wheat

Pollux Field D

Photo.7 21.04.2005 Field D winter and spring wheat

Pollux Zebra Paragon

Photo.8 21.04.2005 Field D spring wheat

Fasan Monsun

Photo.9 8.5.2005 Field A winter wheat

Wenga Pollux

Photo.10 8.5.2005 Field A winter and spring wheat

WW Pollux SW Zebra

Photo.11 8.05.2005 Field B winter wheat

Greina Wenga

Photo.12 8.05.2005 Field C winter wheat

Wenga Greina

Photo.13 8.05.2005 Field C spring wheat

Monsun Fasan

Photo.14 8.05.2005 Field C spring wheat

Paragon Monsun

Photo.15 10.06.2005 Field C spring and winter wheat

Zebra canopy Pollux canopy

Photo.16 27.06.2005 Field C winter wheat

Greina Wenga

Photo.17&18 27.06.2005 Field A spring wheat

Fasan (1m marker) Monsun

Photo.19 27.06.2005 Field C winter wheat

Greina (1m marker) Wenga (1m marker)

Photo.20 27.06.2005 Field C spring and winter wheat

Zebra Pollux

Photo.21 27.06.2005 Field C spring wheat

Fasan Monsun

Photo.22 08.08.2005 Field B winter wheat

Greina (1m marker) Wenga

Photo.23 08.08.2005 Field D spring wheat

Zebra Paragon

Photo.24 08.08.2005 Field C spring wheat

Zebra Paragon

Photo.25 07.09.2005 Field C spring wheat

Zebra

Photo.26 08.08.2005 Field D winter wheat

Pollux (1m marker)

APPENDIX 4: Chapter 3 Predictions for clover plants and nodules, Year 2

Table A2. Year 2 Clover plant numbers, nodules per plant and total *Rhizobium* nodules for sample station and m^2 in the absence of *Rhizobium* inoculation.

noRhyznods_Plants

"NO Rhyzobium Treated Nodules ~Plants" linear model in R
"intercept =139.7 ; Plants -3.98

	20 cm Station			M^2 Scale		
	Plants	Nods/plant	total nods	Plants	Nods/plant	total nods
	1	140	140	25	140	3500
	2	132	264	50	132	6602
	3	128	384	75	128	9605
	4	124	496	100	124	12408
	5	120	601	125	120	15013
mean plants	**6**	**116**	**697**	**150**	**116**	**17418**
	7	112	785	175	112	19625
	8	108	865	200	108	21632
	9	104	938	225	104	23441
	10	100	1002	250	100	25050
	11	96	1058	275	96	26461
	12	92	1107	300	92	27672
	13	88	1147	325	88	28685
	14	84	1180	350	84	29498
	15	80	1205	375	80	30113
	16	76	1221	400	76	30528
max nods	**17**	**72**	**1230**	**425**	**72**	**30745**
	18	68	1230	450	68	30762
	19	64	1223	475	64	30581
	20	60	1208	500	60	30200
	21	56	1185	525	56	29621
	22	52	1154	550	52	28842
	23	48	1115	575	48	27865
	24	44	1068	600	44	26688
	25	41	1013	625	41	25313

one ha =10,000 m^2 , 1000 clover seed wieight = 10gms , **nodules (max) = 425 plants.**

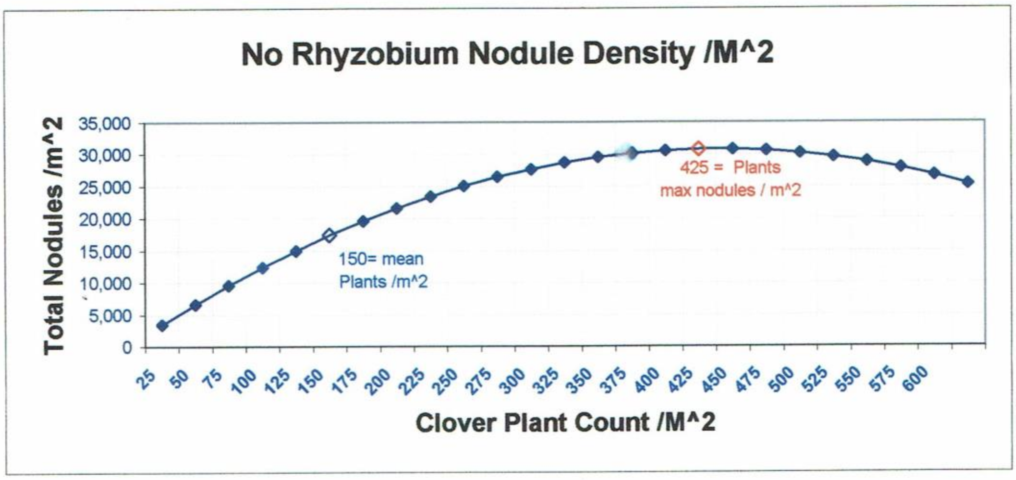
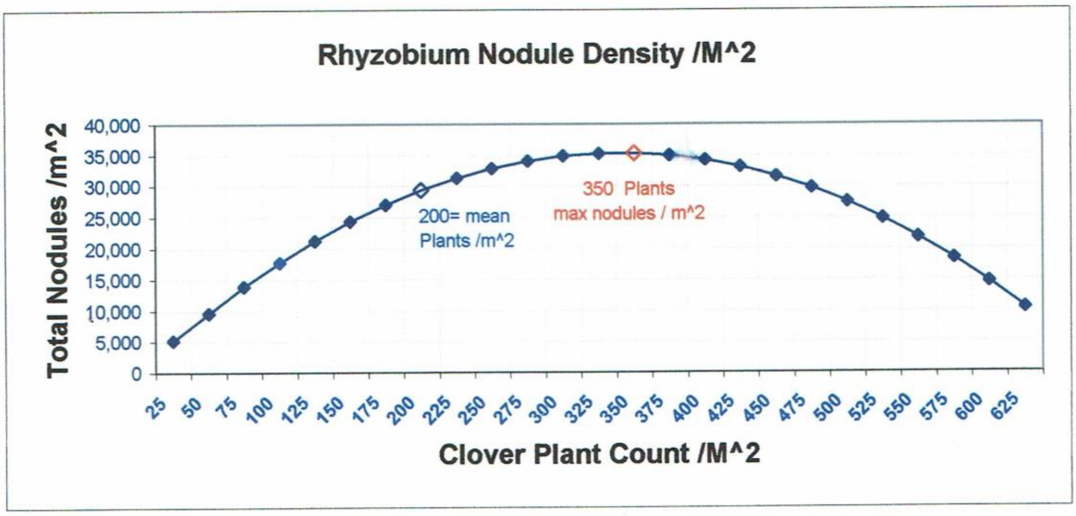


Table A3. Year 2 Clover plant numbers, nodules per plant and total *Rhizobium* nodules for sample station and m^2 with *Rhizobium* inoculation.

"Rhyzobium treated Nodules ~Plants" linear model in R
"intercept =207.6 ; Plants -7.652

	20 cm Station			M^2 Scale		
	Plants	Nods/plant	total nods	Plants	Nods/plant	total nods
	1	208	208	25	208	5200
	2	193	385	50	193	9635
	3	185	555	75	185	13878
	4	177	710	100	177	17739
	5	170	849	125	170	21218
	6	162	973	150	162	24313
	7	154	1081	175	154	27026
mean Plants	8	147	1174	200	147	29357
	9	139	1252	225	139	31305
	10	131	1315	250	131	32870
	11	124	1362	275	124	34053
	12	116	1394	300	116	34853
	13	109	1411	325	109	35270
max nods	14	101	1412	350	101	35305
	15	93	1398	375	93	34958
	16	86	1369	400	86	34227
	17	78	1325	425	78	33114
	18	70	1265	450	70	31619
	19	63	1190	475	63	29741
	20	55	1099	500	55	27480
	21	47	993	525	47	24837
	22	40	872	550	40	21811
	23	32	736	575	32	18402
	24	24	584	600	24	14611
	25	17	418	625	17	10438

one ha =10,000 m^2 , 1000 clover seed wieight = 10gms , **nodules (max) = 350 plants.**



APPENDIX 5: Chapter 4 Soil analysis pre- and post-trial for all three sites for both years

Table A5.1 Pre drilling soil analysis for variety trial at Gilchesters, Courtyard and Sheepdrove farm trial sites

Year	Farm	N kg ha^{-1}	C %	P mg kg^{-1}	K mg kg^{-1}	Ca mg kg^{-1}	S mg kg^{-1}	Mg mg kg^{-1}
2006	Courtyard	64.3±2.57	1.1±0.18	53.0±5.99	115.2±21.5	3400.5±354.45	11.4±0.36	44.7±1.95
2006	Gilchesters	126.0±1.35	2.6±0.05	30.9±2.19	204.0±15.1	1299.2±48.44	16.4±0.66	192.7±6.31
2006	Sheepdrove	222.3±4.29	8.4±0.57	44.1±11.86	337.8±18.39	15606.8±1087.36	20.8±1.85	160.0±14.13
2007	Courtyard	56.1±1.65	0.9±0.06	60.2±6.87	77.1±3.37	2784.1±362.07	15.4±0.28	34.1±2.24
2007	Gilchesters	171.0±13.50	3.8±0.50	20.5±1.60	127.5±5.81	4425.0±1099.09	22.2±1.43	125.5±18.49
2007	Sheepdrove	315.9±5.85	9.8±0.66	15.9±1.71	144.5±7.42	15970.5±992.46	21.2±0.70	146.6±7.06

Year	Farm	Na mg kg^{-1}	Fe mg kg^{-1}	Mn mg kg^{-1}	Zn mg kg^{-1}	Cu mg kg^{-1}	B mg kg^{-1}
2006	Courtyard	13.5±0.81	94.4±8.49	155.5±4.94	2.3±0.10	0.9±0.02	0.6±0.10
2006	Gilchesters	18.4±2.50	455.2±14.84	58.7±3.17	2.8±0.11	2.2±0.03	0.1±0.02
2006	Sheepdrove	49.7±5.74	59.3±7.35	156.1±14.3	7.5±0.84	2.7±0.33	2.2±0.04
2007	Courtyard	28±3.64	152.7±17.26	196.5±7.60	2.7±0.21	1.2±0.07	1.1±0.06
2007	Gilchesters	31.1±4.76	246.1±32.73	159.8±14.84	4.6±0.87	3.81±0.25	1.8±0.47
2007	Sheepdrove	55.6±1.49	40.8±6.77	125.6±18.85	5.8±0.10	1.71±0.11	3.5±0.21

Year	Farm	Al mg kg^{-1}	Pb mg kg^{-1}	Ni mg kg^{-1}	Cd µg kg^{-1}	Mo µg kg^{-1}
2006	Courtyard	370.7±24.94	4.3±0.13	0.7±0.02	138.0±5.02	8.0±0.00
2006	Gilchesters	630.7±21.59	6.2±0.34	1.2±0.04	103.8±2.82	10.2±0.82
2006	Sheepdrove	148.5±26.20	4.9±0.06	1.4±0.12	346.9±8.97	8.0±0.00
2007	Courtyard	321.9±119.27	5.2±0.52	0.8±0.02	158.5±6.70	13.3±3.57
2007	Gilchesters	627.6±118.89	13.0±1.32	2.3±0.15	209.0±32.34	19±3.93
2007	Sheepdrove	182.9±46.25	5.3±0.22	0.7±0.07	537.5±14.89	9.0±0.00

Table A5.2 Post harvest soil analysis for variety trial at Gilchesters, Courtyard and Sheepdrove farm trial sites

Year	Farm	N kg ha^{-1}	C %	P mg kg^{-1}	K mg kg^{-1}	Ca mg kg^{-1}	S mg kg^{-1}	Mg mg kg^{-1}
2006	Courtyard	50.7±1.28	1.0±0.22	73.6±11.73	170.7±47.78	2366.7±589.13	15.5±0.74	50.8±1.34
2006	Gilchesters	181.3±4.09	2.4±0.07	27.7±4.94	133.9±30.47	1385.9±54.71	17.1±0.58	182.2±6.64
2006	Sheepdrove	212.9±3.04	6.9±0.54	11.0±1.49	168.3±6.11	17038.6±1073.36	17.2±0.38	75.0±3.79
2007	Courtyard	52.8±5.94	0.8±0.09	60.5±8.80	69.5±7.81	3000.9±401.46	11.6±0.49	32.0±3.46
2007	Gilchesters	189.0±10.35	3.5±0.47	19.0±1.40	100.2±6.8	4821.8±1241.47	17.1±1.35	136.1±15.37
2007	Sheepdrove	210.6±11.31	6.7±0.81	13.2±1.77	111.5±3.8	12979.0±731.99	14.8±0.47	83.9±2.74

Year	Farm	Na mg kg^{-1}	Fe mg kg^{-1}	Mn mg kg^{-1}	Zn mg kg^{-1}	Cu mg kg^{-1}	B mg kg^{-1}
2006	Courtyard	23.1±2.22	175.2±18.11	203.6±10.25	2.2±0.13	1.1±0.07	1.2±0.21
2006	Gilchesters	26.0±0.61	452.0±11.26	73.7±3.09	2.9±0.29	2.3±0.03	0.5±0.02
2006	Sheepdrove	68.8±2.91	40.0±6.90	129.4±14.41	4.3±0.32	2.2±0.39	2.5±0.14
2007	Courtyard	12.8±1.34	104.0±16.44	177.0±16.44	2.5±0.25	1.2±0.09	1.2±0.12
2007	Gilchesters	18.2±0.38	244.0±39.39	145.4±4.93	4.9±0.62	4.0±0.21	2.3±0.56
2007	Sheepdrove	30.3±1.28	59.3±6.78	160.0±24.03	4.3±0.26	2.6±0.17	3.1±0.25

Year	Farm	Al mg kg^{-1}	Pb mg kg^{-1}	Ni mg kg^{-1}	Cd µg kg^{-1}	Mo µg kg^{-1}
2006	Courtyard	383.2±140.43	5.3±0.13	0.9±0.04	153.5±2.92	12.0±2.07
2006	Gilchesters	796.6±13.95	5.8±0.36	1.6±0.08	113.8±2.68	19.7±1.68
2006	Sheepdrove	135.0±20.28	3.7±0.33	1.0±0.14	341.5±16.66	9.0±0.00
2007	Courtyard	282.6±138.76	4.7±0.54	0.7±0.03	155.9±8.56	12.1±3.15
2007	Gilchesters	569.3±164.22	13.6±0.68	2.2±0.16	217.8±28.65	22.1±5.11
2007	Sheepdrove	297.5±76.69	4.2±0.23	1.3±0.12	447.3±11.06	10.5±0.91

APPENDIX 6: Chapter 4 Results tables for all three sites for both years
Disease assessments

Table A6.1 Main effect means ± SE and *p* values for the effects of harvest year and variety in wheat disease severity (AUDPC) at GS 37 and 65.

	Septoria triticii			**Yellow rust**		
	GS37 %	**GS65 Leaf F-1 %**	**GS65 Leaf F-2 %**	**GS37 %**	**GS65 Leaf F-1 %**	**GS65 Leaf F-2 %**
Year (YR)						
2006	1.2±0.22	0.2±0.08	6.9±1.18	0.1±0.03	6.1±1.63	4.3±1.49
2007	0.8±0.14	0.3±0.12	7.7±1.46	2.9±0.81	10.8±2.56	9.2±2.83
Site (ST)						
Courtyard	2.1±0.27	0.2±0.06**b**	6.0±1.26**b**	0.0±0.00	1.6±0.54**b**	0.2±0.13**b**
Gilchesters	0.3±0.14	0.5±0.18**a**	4.8±1.57**b**	1.5±0.45	12.7±3.49**a**	10.5±3.9**a**
Sheepdrove	0.3±0.07	0.1±0.11**b**	11.1±1.86**a**	0.2±0.08	11.1±2.7**a**	9.4±2.63**a**
Variety (VR)						
Amaretto	1.1±0.31	0.1±0.05**b**	3.7±0.84**c**	0.5±0.33	2.9±0.6**bc**	1.1±0.3**b**
Fasan	0.9±0.3	0.4±0.22**ab**	12.2±2.11**a**	1.2±0.68	7.9±1.79**b**	2.9±1.00**b**
Monsun	1.1±0.35	0.8±0.34**a**	11.1±3.77**a**	0.5±0.33	3.1±0.72**bc**	2.9±0.75**b**
Paragon	1.3±0.4	0.1±0.06**b**	4.6±1.01**b**	0.0±0.00	0.1±0.04**c**	0.1±0.04**b**
Tybalt	0.5±0.18	0.2±0.09**b**	1.4±0.27**c**	0.0±0.00	0.0±0.00**c**	0.0±0.00**b**
Zebra	1.3±0.51	0.0±0.08**b**	10.9±2.77**a**	2.4±1.08	37.04±6.21**a**	33.4±7.54**a**
ANOVA						
Main Effects						
YR	ns	ns	ns	ns	**0.0283**	**0.0334**
ST	ns	**0.0158**	**0.0083**	ns	**<.0001**	**<.0001**
VR	ns	**<.0001**	**<.0001**	ns	**<.0001**	**<.0001**
Interactions						
YR:ST	ns	**<.0001**	**0.0023**	ns	**0.0078**	**0.0005**
YR:VR	ns	**<.0001**	0.5563	ns	**<.0001**	**<.0001**
ST:VR	ns	**<.0001**[1]	**0.0053**[2]	ns	**<.0001**[3]	**<.0001**[4]
YR:ST:VR	ns	**<.0001**[5]	**0.0002**[6]	ns	**<.0001**[7]	**<.0001**[8]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1] see Table 6.3 for interaction means SE. [2] see Table 6.4 for interaction means SE. [3] see Table 6.5 for interaction means SE. [4] see Table 6.6 for interaction means SE. [5] see Table 6.7 for interaction means SE. [6] see Table 6.8 for interaction means SE. [7] see Table 6.9 for interaction means SE. [8] see Table 6.10 for interaction means SE.

Table A6.2 Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat disease severity (AUDPC) at growth stages (GS) 37 and 65 for each Farm

	Septoria tritici								
	GS37 %			**GS65 Leaf F-1 %**			**GS65 Leaf F-2 %**		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year (YR)									
2006	3.4±0.37	0.0±0.00	0.3±0.07	0.4±0.10	0.0±0.00	0.33±0.22	3.1±0.41	2.3±0.75	15.2±2.78
2007	0.8±0.13	0.7±0.26	0.0±0.00	0.0±0.02	1.1±0.33	0.0±0.00	9.1±2.36	7.5±3.00	6.9±2.23
Variety (VR)									
Amaretto	2.1±0.54**b**	0.5±0.38	0.4±0.19	0.3±0.13	0.0±0.00**b**	0.0±0.00	3.6±0.68**b**	2.2±0.84	5.2±2.25**bc**
Fasan	1.3±0.42**c**	0.7±0.62	0.5±0.26	0.2±0.09	0.3±0.18**b**	0.88±0.64	8.6±2.76**ab**	4.8±1.27	23.1±2.82**a**
Monsun	2.2±0.65**b**	0.5±0.37	0.2±0.10	0.3±0.16	2.1±0.88**a**	0.0±0.00	4.1±0.69**b**	16.0±8.43	13.3±7.56**ab**
Paragon	2.7±0.72**b**	0.3±0.25	0.4±0.20	0.2±0.13	0.1±0.13**b**	0.0±0.00	4.2±0.53**b**	1.1±0.64	8.5±2.32**bc**
Tybalt	1.0±0.36**c**	0.1±0.06	0.4±0.19	0.1±0.06	0.5±0.19**b**	0.1±0.13	1.5±0.54**b**	1.2±0.62	1.3±0.18**c**
Zebra	3.3±0.91**a**	0.1±0.01	0.1±0.03	0.2±0.25	0.0±0.00**b**	0.0±0.00	14.3±6.28**a**	3.3±1.57	15.0±4.63**ab**
ANOVA									
Main Effects									
YR	**0.0041**	ns	ns	**0.0354**	**0.0048**	ns	**0.0372**	ns	0.0638
VR	**0.0007**	ns	ns	ns	**<.0001**	ns	**0.0015**	**0.0164**	**0.0024**
Interactions									
YR:VR	ns	ns	ns	ns	**<.0001**	ns	**0.0005**	**0.0251**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.2 (cont.) Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat disease severity (AUDPC) at growth stages (GS) 37 and 65 for each Farm

	Yellow rust								
	GS37 %			**GS65 Leaf F-1 %**			**GS65 Leaf F-2 %**		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year (YR)									
2006	0.0±0.00	0.1±0.01	0.2±0.08	0.4±0.22	7.3±2.72	10.8±3.86	0.0±0.00	3.5±1.68	9.3±3.96
2007	0.0±0.00	2.9±0.81	0.0±0.00	2.9±1.01	18.2±6.30	11.3±3.87	0.5±0.25	17.4±7.43	9.6±3.55
Variety (VR)									
Amaretto	0.0±0.00	1.0±0.63**bc**	0.0±0.00	1.3±0.80**b**	4.0±1.04**b**	3.3±1.12**c**	0.5±0.38	2.2±0.64**b**	0.7±0.25**b**
Fasan	0.0±0.00	2.3±1.27**ab**	0.1±0.03	0.8±0.29**b**	10.6±2.2**b**	12.2±3.97**b**	0.0±0.00	3.7±1.31**b**	5.1±2.47**b**
Monsun	0.0±0.00	1.1±0.61**bc**	0.1±0.03	0.4±0.25**b**	5.0±1.55**b**	4.0±1.04**bc**	0.2±0.13	2.0±0.68**b**	6.6±1.36**b**
Paragon	0.0±0.00	0.0±0.00**c**	0.0±0.00	0.0±0.00**b**	0.1±0.13**b**	0.0±0.00**c**	0.0±0.00	0.1±0.13**b**	0.0±0.00**b**
Tybalt	0.0±0.00	0.0±0.00**c**	0.0±0.00	0.0±0.00**b**	0.0±0.00**b**	0.0±0.00**c**	0.0±0.00	0.0±0.00**b**	0.0±0.00**b**
Zebra	0.0±0.00	4.4±1.97**a**	1.0±0.00	7.3±2.30**a**	56.8±11.70**a**	46.8±6.47**a**	1.0±0.63	55.±16.42**a**	44.3±7.53**a**
ANOVA									
Main Effects									
YR	ns	0.06	ns	**0.0152**	**0.0176**	ns	0.0825	**0.0034**	ns
VR	ns	**<.0001**	ns	**<.0001**	**<.0001**	**<.0001**	0.0742	**<.0001**	**<.0001**
Interactions									
YR:VR	ns	**<.0001**	ns	**<.0001**	**<.0001**	ns	0.0742	**<.0001**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.3 Interaction means for effects of wheat variety and site on Septoria % (leaf F-1) at GS 65.

	Septoria % (Leaf F-1)					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	0.3±0.13**A**	0.2±0.09**A**	0.3±0.16**B**	0.2±0.13**A**	0.1±0.06**A**	0.2±0.25**A**
GIL	0.0±0.00**Ab**	0.3±0.18**Ab**	2.1±0.88**Aa**	0.1±0.13**Ab**	0.5±0.19**Ab**	0.0±0.00**Ab**
SHE	0.0±0.00**A**	0.8±0.64**A**	0.0±0.00**B**	0.0±0.00**A**	0.1±0.10**A**	0.0±0.00**A**

Means labelled with the same capital letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.4 Interaction means for effects of wheat variety and site on Septoria % (leaf F-2) at GS 65.

	Septoria % (Leaf F-2)					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	3.6±0.68**Ab**	8.6±2.76**Bab**	4.1±0.69**Bb**	4.2±0.53**Ab**	1.5±0.54**Ab**	14.3±6.28**Aa**
GIL	2.2±0.84**A**	4.8±1.27**B**	16.0±8.43**A**	1.1±0.64**A**	1.2±0.62A	3.3±1.57**B**
SHE	5.2±2.25**Abc**	23.1±2.82**Aa**	13.3±7.56**ABab**	8.5±2.32**Abc**	1.3±0.18Ac	15.0±4.63**Aab**

Means labelled with the same capital letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.5 Interaction means for effects of wheat variety and site on yellow rust % (Leaf F-1) at GS 65.

	Yellow Rust % (Leaf F-1)					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	1.3±0.8**Ab**	0.8±0.29**Bb**	0.4±0.25**Ab**	0.0±0.00**Ab**	0.0±0.00**Ab**	7.3±2.3**Ca**
GIL	4.0±1.04**Ab**	10.6±2.2**Ab**	5.0±1.55**Ab**	0.13±0.13**Ab**	0.0±0.00**Ab**	56.8±11.7**Aa**
SHE	3.3±1.12**Ac**	12.2±3.97**Ab**	4.0±1.04**Abc**	0.0±0.00**Ac**	0.0±0.00**Ac**	46.8±6.47**Ba**

Means labelled with the same capital letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.6 Interaction means for effects of wheat variety and site on yellow rust % (Leaf F-2) at GS 65.

	Yellow Rust % (Leaf 2)					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	0.5±0.38**A**	0±0**A**	0.19±0.13**A**	0±0**A**	0±0A	1±0.63**B**
GIL	2.25±0.64**Ab**	3.75±1.31**Ab**	2±0.68**Ab**	0.13±0.13**Ab**	0±0Ab	55±16.42**Aa**
SHE	0.75±0.25**Ab**	5.13±2.47**Ab**	6.63±1.36**Ab**	0±0**Ab**	0± 0Ab	44.38±7.53**Aa**

Means labelled with the same capital letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.7 Interaction means for effects of year, wheat variety and site on Septoria % (leaf 1) at GS 65.

Septoria % (Leaf F-1)

Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	0.6±0.12**a**	0.2±0.14**c**	0.7±0.14**b**	0.5±0.2**a**	0.1±0.12**a**	0.5±0.5**a**
	GIL	0.0±0.00**b**	0.0±0.00**d**	0.0±0.00**c**	0.0±0.00**b**	0.0±0.00**b**	0.0±0.00**a**
	SHE	0.0±0.00**b**	1.7±1.18**a**	0.0±0.00**c**	0.0±0.00**b**	0.2±0.25**a**	0.0±0.00**a**
2007	**COU**	0.0±0.00**b**	0.2±0.12**c**	0.0±0.00**c**	0.0±0.00**b**	0.0±0.00**b**	0.0±0.00**a**
	GIL	0.0±0.00**b**	0.7±0.25**b**	4.2±0.75**a**	0.2±0.25**a**	1.0±0.00**a**	0.0±0.00**a**
	SHE	0.0±0.00**b**	0.0±0.00**d**	0.0±0.00**c**	0.0±0.00**b**	0.0±0.00**b**	0.0±0.00**a**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.8 Interaction means for effects of year, wheat variety and site on Septoria % (leaf F-2) at GS 65.

				Septoria % (Leaf F-2)			
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	3.5±1.20**a**	2.2±0.40**d**	5.0±0.80**b**	4.5±0.50**b**	0.8±0.30**b**	2.5±1.04**c**
	GIL	0.7±0.40**a**	3.5±2.10**cd**	3.2±2.20**b**	0.0±0.00**d**	0.0±0.00**c**	6.7±1.9**b**
	SHE	8.0±4.00**a**	21.2±3.01**a**	24.2±13.40**a**	13.7±2.40**a**	1.7±0.25**a**	22.5±2.50**a**
2007	**COU**	3.7±0.70**a**	15.0±2.80**b**	3.2±1.03**b**	4.0±1.00b**c**	2.2±0.90**a**	26.2±9.40**a**
	GIL	3.7±1.25**a**	6.2±1.25**c**	28.7±14.70**a**	2.2±1.03**c**	2.5±0.80**a**	0.0±0.00**d**
	SHE	2.5±1.40**a**	25.0±5.00**a**	2.5±2.50**b**	3.2±1.03**bc**	1.0±0.00**b**	7.5±7.50**bc**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.9 Interaction means for effects of year, wheat variety and site on yellow rust % (Leaf F-1) at GS 65.

				Yellow Rust % (Leaf F-1)			
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	0.0±0.00**c**	0.1±0.10**d**	0.0±0.00**d**	0.0±0.00	0.0±0.00	2.2±0.90**d**
	GIL	3.1±1.10**b**	7.5±1.40**b**	3.2±1.03**b**	0.0±0.00	0.0±0.00	30.0±10.80**b**
	SHE	4.7±1.80**ab**	5.7±1.50**b**	4.5±2.02**ab**	0.0±0.00	0.0±0.00	50.0±7.00**b**
2007	**COU**	2.7±1.30**b**	1.5±0.20**c**	0.9±0.40**c**	0.0±0.00	0.0±0.00	12.5±2.50**c**
	GIL	5.0±1.70**a**	13.7±3.70**a**	6.7±2.80**a**	0.2±0.20	0.0±0.00	83.7±6.50**a**
	SHE	2.0±1.00**b**	18.7±6.50**a**	3.5±0.80**b**	0.0±0.00	0.0±0.00	43.7±11.80**b**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.10 Interaction means for effects of year, wheat variety and site on yellow rust % (Leaf F-2) at GS 65.

				Yellow Rust % (Leaf F-2)			
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	0.0±0.00**c**	0.0±0.00**c**	0.0±0.00**d**	0.0±0.00	0.0±0.00	0.0±0.00**d**
	GIL	2.0±1.06**a**	4.2±2.60**b**	2.2±0.90**b**	0.0±0.00	0.0±0.00	13.0±9.02**c**
	SHE	0.5±0.20**b**	0.2±0.20**c**	5.2±1.80**a**	0.0±0.00	0.0±0.00	50.0±7.07**b**
2007	**COU**	1.0±0.70**a**	0.0±0.00**c**	0.3±0.20**c**	0.0±0.00	0.0±0.00	2.0±1.08**d**
	GIL	2.5±0.80**a**	3.2±1.03**b**	1.7±1.10**b**	0.2±0.20	0.0±0.00	97.0±1.10**a**
	SHE	1.0±0.40**ab**	10.0±3.50**a**	8.0±2.00**a**	0.0±0.00	0.0±0.00	38.7±13.90**b**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Yield Assessments

Table A6.11 Main effect means ± SE and *p* values for the effects of harvest year, site and variety on wheat yield parameters.

	Grain Yield t ha^{-1}	**Plant height** cm	**SPAD** GS65	**TGW** g
Year (YR)				
2006	4.9±0.16	97.0±1.07	46.9±0.67	40.0±0.05**a**
2007	2.7±0.14	86.7±1.36	39.6±0.65	38.9±0.63**b**
Site (ST)				
Courtyard	4.4±0.14**a**	92.4±1.25**ab**	41.9±0.47**b**	37.8±0.53
Gilchesters	4.2±0.33**a**	94.2±1.53**a**	45.6±1.35**a**	41.5±0.76
Sheepdrove	2.9±0.14**b**	89.0±2.07**b**	42.3±0.78**b**	39.3±0.63
Variety (VR)				
Amaretto	3.8±0.33**cd**	92.1±2.05**c**	44.7±1.25**ab**	37.0±0.59**c**
Fasan	4.1±0.31**b**	100.1±2.16**a**	41.3±1.1**c**	39.7±0.7**b**
Monsun	3.7±0.31**d**	89.7±1.62**d**	44.0±1.01**b**	42.4±0.64**a**
Paragon	4.0±0.32**bc**	90.3±2.17**cd**	46.2±1.03**a**	39.9±0.7**b**
Tybalt	4.6±0.31**a**	81.2±1.78**e**	45.8±0.91**ab**	41.8±0.69**a**
Zebra	2.8±0.37**e**	97.6±2.36**b**	37.5±1.86**d**	36.0±1.44**c**
ANOVA				
Main Effects				
YR	**0.0019**	**0.0017**	**0.001**	**<.0001**
ST	**<.0001**	**0.0026**	**0.0003**	ns
VR	**<.0001**	**<.0001**	**<.0001**	**<.0001**
Interactions				
YR:ST	**<.0001**	**<.0001**	**<.0001**	ns
YR:VR	**0.002**	**0.0028**	**0.0135**	**<.0001**
ST:VR	**<.0001**[1]	**0.0004**[2]	**<.0001**[3]	ns
YR:ST:VR	ns	ns	**<.0001**[4]	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1]For interaction means see Table 6.13. [2]For interaction means see Table 6.14. [3]For interaction means see Table 6.15. [4]For interaction means see Table 6.16

Table A6.11a. The ranking order of grain yield of spring wheat varieties grown at Courtyard, Sheepdrove and Gilchesters Farms 2006 – 2007

	2006			**2007**		
Rank order	**Courtyard**	**Sheepdrove**	**Gilchesters**	**Courtyard**	**Sheepdrove**	**Gilchesters**
1	Tybalt	Tybalt	Paragon	Tybalt	Tybalt	Paragon
2	Fasan	Amaretto	Amaretto	Fasan	Paragon	Fasan
3	Paragon/Monsun	Paragon	Fasan/Tybalt	Monsun	Amaretto	Tybalt
4	-	Fasan	-	Paragon	Fasan	Monsun
5	Zebra	Monsun	Monsun	Amaretto	Monsun	Amaretto
6	Amaretto	Zebra	Zebra	Zebra	Zebra	Zebra

Table A6.12 Site effect means ± SE and *p* values for the effects of harvest year and variety in wheat yield parameters from each Farm

	Grain yield t ha^{-1}			**Plant height** cm		
Year (YR)	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	4.9±0.17**a**	6.2±0.16	3.5±0.13a	89.3±1.59	99.5±1.64	102.2±1.17
2007	3.8±0.18**b**	2.1±0.18	2.3±0.18b	95.5±1.73	88.9±2.09	75.8±1.03
Variety (VR)						
Amaretto	3.8±0.19**de**	4.3±0.93**ab**	3.2±0.28**b**	92.7±1.26**c**	94.4±3.06**b**	89.2±5.34**bc**
Fasan	4.8±0.27**b**	4.4±0.73**ab**	2.9±0.23**bc**	104.3±2.27**a**	102.0±2.92**a**	94.2±4.94**a**
Monsun	4.3±0.20**c**	4.1±0.78**b**	2.7±0.26**c**	88.7±1.52**d**	92.4±2.45**b**	87.9±4.01**c**
Paragon	4.2±0.30**cd**	4.7±0.83**a**	3.2±0.17**b**	90.6±1.41**cd**	92.4±3.05**b**	87.8±5.82**c**
Tybalt	5.6±0.25**a**	4.3±0.77**ab**	3.8±0.24**a**	80.7±1.42**e**	81.1±2.91**c**	81.9±4.53**d**
Zebra	3.7±0.40**e**	3.2±0.88 **c**	1.6±0.30**d**	97.5±1.96**b**	102.7±2.76**a**	92.7±6.05**ab**
ANOVA						
Main Effects						
YR	**0.0022**	**0.002**	**0.0064**	**0.0388**	**0.0078**	**0.0001**
VR	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**
Interactions						
YR:VR	**0.0461**	ns	**0.0079**	ns	**0.0274**	**0.0188**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.12 (cont.) Site effect means ± SE and *p* values for the effects of harvest year and variety in wheat yield parameters from each Farm

	SPAD GS65			**TGW** g		
Year (YR)	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	43.57±0.67	52.71±0.52	44.58±1.11	37.1±0.05	43.3±0.07	39.9±0.07
2007	40.37±0.48	38.5±1.68	40.1±0.89	38.6±0.96	39.53±1.36	38.82±1.03
Variety (VR)						
Amaretto	41.6±1.31**ab**	48.06±2.84**a**	44.56±1.59**a**	36.36±0.6**b**	38.5±1.63**c**	36.56±0.66**b**
Fasan	39.91±1.08**b**	43.35±2.99**b**	40.9±0.97**b**	37.45±1.25**b**	42.38±0.63**abc**	39.39±1.06**ab**
Monsun	41.95±0.55**ab**	46.81±2.32**ab**	43.28±1.61**a**	42.10±0.72**a**	43.44±1.63**ab**	42.04±1.02**a**
Paragon	43.39±1.34**a**	49.83±1.96**a**	45.6±1.36**a**	37.20±0.36**b**	41.98±1.06**abc**	40.78±1.32**a**
Tybalt	43.13±0.86**a**	49.13±1.93**a**	45.33±1**a**	40.32±1.01**a**	43.63±1.18**a**	41.71±1.23**a**
Zebra	41.83±1.44**ab**	36.45±5.05**c**	34.38±1.54**c**	33.62±1.2**c**	39.22±3.66**bc**	35.73±2.30**b**
ANOVA						
Main Effects						
YR	**0.0349**	**0.0004**	**0.0129**	**<.0001**	**0.0002**	**<.0001**
VR	**0.0363**	**<.0001**	**<.0001**	**<.0001**	**0.0159**	**<.0001**
Interactions						
YR:VR	ns	**<.0001**[1]	Ns	**<.0001**	**0.0059**	**<.0001**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.13 Interaction means for effects of wheat variety and site on grain yield

	Grain Yield t ha^{-1}					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	3.82±0.19**ABa**	4.88±0.27**Ab**	4.32±0.2**Ac**	4.23±0.3**Acd**	5.63±0.25**Aa**	3.72±0.4**Ae**
GIL	4.37±0.93**Aa**	4.48±0.73**ABa**	4.11±0.78**Aa**	4.76±0.83**Ba**	4.34±0.77**Ba**	3.2±0.88**Ab**
SHE	3.23±0.28**Bb**	2.98±0.23**Bbc**	2.72±0.26**Bc**	3.24±0.17**Cb**	3.88±0.24**Ba**	1.62±0.3**Bd**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.14 Interaction means for effects of wheat variety and site on straw height

	Straw height cm					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	92.75±1.26**Ac**	104.33±2.27**Aa**	88.78±1.52**Ad**	90.66±1.41**Acd**	80.74±1.42**Ae**	97.5±1.96**ABb**
GIL	94.44±3.06**Ab**	102±2.92**ABa**	92.48±2.45**Ab**	92.48±3.05**Ab**	81.1±2.91**Ac**	102.71±2.76**Aa**
SHE	89.24±5.34**Abc**	94.23±4.94**Ba**	87.98±4.01**Ac**	87.89±5.82**Ac**	81.93±4.53**Ad**	92.76±6.05**Bab**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.15 Interaction means for effects of wheat variety and site on SPAD at GS 65.

	GS 65 SPAD					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	41.6±1.31**Bab**	39.91±1.08**Ab**	41.95±0.55**Bab**	43.39±1.34**Ba**	43.13±0.86**Ba**	41.83±1.44**Aab**
GIL	48.06±2.84**Aa**	43.35±2.99**Ab**	46.81±2.32**Aab**	49.83±1.96**Aa**	49.13±1.93**Aa**	36.45±5.05**Bc**
SHE	44.56±1.59**ABa**	40.9±0.97**Ab**	43.28±1.61**Aba**	45.6±1.36**ABa**	45.33±1A**Ba**	34.38±1.54**Bc**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.16 Interaction means for effects of year, wheat variety and site on SPAD at GS65

		GS 65 SPAD					
Year	**Site**	**AMA**	**FAS**	**MON**	**PAR**	**TYB**	**ZEB**
2006	**COU**	43.47±2.08**c**	40.37±2.08**bc**	42.77±0.67**c**	46±1.32**c**	44±1.23**c**	44.77±1.47**b**
	GIL	55.22±0.6**a**	50.85±1.2**a**	52.85±0.62**a**	54.17±0.67**a**	53.7±1.09**a**	49.47±0.8**a**
	SHE	47.8±0.4**b**	42.57±0.96**b**	46.8±0.74**b**	49.05±0.68**b**	47.07±1.15**b**	34.17±1.5d
2007	**COU**	39.72±1.17**d**	39.45±0.97**c**	41.12±0.7**d**	40.77±1.43**d**	42.25±1.17**c**	38.87±1.3**c**
	GIL	40.9±1.77**cd**	35.85±1.66**d**	40.77±0.73**de**	45.47±2.2**c**	44.55±1.5**c**	23.42±2.33**e**
	SHE	41.32±2.15**cd**	39.22±1.27**c**	39.75±1.8**e**	42.15±0.41**d**	43.57±1.13**c**	34.57±2.95**d**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Grain Quality and macro-nutrient Assessments

For **grain crude protein contents** were significantly higher in 2007 at 13.7% than in 2006 at 12.8%, higher at Gilchesters (14.5%) and Sheepdrove (14%) than at Courtyard farm (11.5%). Highest with Zebra (14.16%), intermediate with Paragon (13.6%) and lowest with Amaretto and Monsun (12.9%) (Table A6.18). Significant interactions were detected between season/year and site, year and variety, site and variety, and between year, site and variety (Table A6.18). When interaction means for the interaction between variety and site were compared, protein concentrations were significantly lower at Courtyard for all varieties. Protein concentrations for Tybalt were significantly higher at Gilchesters, whilst for Zebra they were significantly lower at Courtyard (Table A6.18).

Table A6.17 Effect means ± SE and *p* values for the effects of harvest year and variety in spring wheat on gluten quality from for Farm

	Protein Quality by SE-HPLC Fractions (F3+F4)/F1 %		
	COU	SHE	GIL
Year (YR)			
2006	3.1±0.04	3.4±0.04	3.4±0.04
2007	3.6±0.04	3.7±0.05	3.7±0.04
Variety (VR)			
Amaretto	3.4±0.10**b**	3.6±0.06**bc**	3.4±0.04**cd**
Fasan	3.5±0.11**a**	3.8±0.13**a**	3.6±0.01**a**
Monsun	3.2±0.1**c**	3.6±0.05**b**	3.4±0.10**b**
Paragon	3.1±0.11**c**	3.3±0.09**d**	3.4±0.03**d**
Tybalt	3.3±0.08**bc**	3.5±0.06**c**	3.5±0.06**b**
Zebra	3.4±0.11**b**	3.6±0.07**b**	3.4±0.01**c**
ANOVA			
Main Effects			
YR	**0.0024**	**0.0013**	**0.0015**
VR	**<.0001**	**<.0001**	**<.0001**
Interactions			
YR:VR	ns	**0.0048**	

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

For **Grain Macro Nutrient concentrations** tested for phosphorous (P), potassium (K), sulphur (S) and calcium (Ca), concentrations were all higher in 2007 than in 2006. All concentrations were highest in varieties grown at Sheepdrove, except for grain K which was highest in wheat grown at Gilchesters. The lowest concentrations of P and Ca were in wheat grown at Gilchesters (Table A6.18). Only in the Ca concentrations could differences be found between varieties, with the highest concentrations in Fasan, intermediate in Monsun and the lowest in Tybalt. Concentrations in Amaretto, Paragon and Zebra were not significantly different to Fasan or Monsun (Table A6.18).

When interaction means for the interaction between variety and site were compared for the individual nutrients (Table A6.19), grain P concentrations were similar for all varieties grown at Gilchesters. At Courtyard and Sheepdrove the highest P concentrations were to be found in Zebra and the lowest in Amaretto and Monsun. For grain K, again there were no differences in the high concentrations between the varieties at Gilchesters, the highest concentrations at Courtyard were found in Tybalt and at Sheepdrove in Paragon. For grain S, the high concentrations at Gilchesters were similar for all varieties whilst Zebra and Paragon had the highest S concentrations at Courtyard and at Sheepdrove only Zebra was higher. For grain Ca there were no significant differences between varieties at either Gilchesters or Sheepdrove and at Courtyard only Tybalt had lower concentrations than the other varieties (Table A6.19).

Table A6.18 Main effect means ± SE and p values for the effects of harvest year and variety on spring wheat grain macro nutrient content.

	Protein %	**Grain P** mg g^{-1}	**Grain K** mg g^{-1}	**Grain S** mg g^{-1}	**Grain Ca** mg g^{-1}
Year (YR)					
2006	12.8±0.22	3.5±0.04	4.0±0.03	1.3±0.02	0.3±0.01
2007	13.7±0.17	4.0±0.07	4.7±0.11	1.3±0.03	0.4±0.01
Site (ST)					
Courtyard	11.5±0.19**c**	3.8±0.04**b**	4.2±0.03**b**	1.2±0.01**b**	0.4±0.01**b**
Gilchesters	14.4±0.12**a**	3.3±0.09**c**	4.6±0.17**a**	1.4±0.04**a**	0.3±0.02**c**
Sheepdrove	13.9±0.16**b**	4.0±0.05**a**	4.3±0.06**b**	1.4±0.02**a**	0.4±0.02**a**
Variety (VR)					
Amaretto	12.9±0.28**c**	3.7±0.08	4.3±0.13	1.3±0.03	0.4±0.02**ab**
Fasan	13.1±0.34**c**	3.7±0.08	4.2±0.08	1.3±0.03	0.4±0.02**a**
Monsun	12.9±0.38**c**	3.7±0.07	4.4±0.14	1.3±0.03	0.4±0.02**b**
Paragon	13.6±0.31**b**	3.8±0.09	4.5±0.1	1.3±0.02	0.4±0.03**ab**
Tybalt	13.0±0.38**c**	3.7±0.08	4.5±0.14	1.3±0.03	0.3±0.01**c**
Zebra	14.1±0.35**a**	3.7±0.18	4.1±0.26	1.4±0.07	0.4±0.03**ab**
ANOVA					
Main Effects					
YR	**0.0211**	**0.0011**	**0.0071**	**0.0428**	**0.0025**
ST	**<.0001**	**<.0001**	**0.0021**	**<.0001**	**<.0001**
VR	**<.0001**	**<.0001**	**0.0001**	**<.0001**	**<.0001**
Interactions					
YR:ST	**0.0339**	ns	**0.0005**	**0.0004**	**0.0056**
YR:VR	**0.0018**	**<.0001**	**0.018**	**<.0001**	**0.0019**
ST:VR	**<.0001**	**0.003**	ns	**<.0001**	ns
YR:ST:VR	**0.0002**	**<.0001**	0.0984	**<.0001**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$).

Table A6.19 Effect means ± SE and p values for the effects of harvest year and variety in spring wheat grain macro nutrient content from each Farm

	Protein %			**Grain P** mg g^{-1}		
	COU	GIL	SHE	**COU**	**GIL**	**SHE**
Year (YR)						
2006	10.8±0.28	14.4±0.12	13.2±0.21	3.6±0.03	3.2±0.03	3.7±0.06
2007	12.1±0.16	14.4±0.23	14.6±0.14	4.1±0.03	3.5±0.17	4.3±0.02
Variety (VR)						
Amaretto	11.5±0.43**b**	13.7±0.17**c**	13.4±0.34**cd**	3.8±0.08**c**	3.3±0.12	3.9±0.12**c**
Fasan	11.2±0.25**bc**	14.4±0.31**abc**	13.8±0.36**bc**	3.8±0.13**bc**	3.4±0.07	4.0±0.13**b**
Monsun	10.6±0.39**c**	14.0±0.3**bc**	13.9±0.31**b**	3.7±0.08**c**	3.4±0.07	4.0±0.1b**c**
Paragon	12.2±0.56**a**	14.6±0.1**ab**	14.0±0.35**b**	3.9±0.12**ab**	3.5±0.13	4.0±0.15**bc**
Tybalt	11.0±0.27b**c**	14.9±0.27**a**	13.1±0.41**d**	3.8±0.11**c**	3.5±0.15	3.9±0.14**bc**
Zebra	12.3±0.53**a**	15.0±0.36**a**	15.2±0.16**a**	3.9±0.1**a**	3.1±0.45	4.3±0.04**a**
ANOVA						
Main Effects						
YR	0.0623	0.975	**0.0023**	**0.0008**	ns	**0.0055**
VR	**0.0002**	**<.0001**	**<.0001**	**0.0086**	ns	**<.0001**
Interactions						
YR:VR	0.2695	**<.0001**	0.0936	ns	ns	**<.0001**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)
COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.19 (cont.) Effect means ± SE and *p* values for the effects of harvest year and variety in spring wheat grain macro nutrient content from each Farm

	Grain K mg g^{-1}			**Grain S** mg g^{-1}			**Grain Ca** mg g^{-1}		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year (YR)									
2006	4.1±0.04	4.0±0.04	3.9±0.04	1.1±0.02	1.5±0.02	1.3±0.02	0.3±0.01	0.2±0.01	0.4±0.03
2007	4.2±0.03	5.3±0.3	4.7±0.05	1.2±0.02	1.3±0.07	1.5±0.01	0.5±0.01	0.4±0.03	0.5±0.01
Variety (VR)									
Amaretto	4.1±0.06**b**	4.7±0.33	4.1±0.18**d**	1.1±0.03**b**	1.3±0.03	1.3±0.03**b**	0.4±0.02**a**	0.3±0.04	0.±0.02
Fasan	4.1±0.05**b**	4.4±0.23	4.2±0.08**cd**	1.1±0.02**b**	1.4±0.06	1.4±0.03**b**	0.4±0.03**a**	0.4±0.04	0.5±0.02
Monsun	4.2±0.04**b**	4.7±0.38	4.3±0.14**bc**	1.1±0.02**b**	1.3±0.03	1.4±0.02**b**	0.4±0.02**a**	0.3±0.04	0.4±0.02
Paragon	4.3±0.06**a**	4.7±0.27	4.5±0.14**a**	1.2±0.04**a**	1.4±0.02	1.4±0.04**b**	0.4±0.03**a**	0.3±0.03	0.5±0.07
Tybalt	4.3±0.05**a**	4.9±0.37	4.4±0.18**ab**	1.1±0.03**b**	1.5±0.02	1.3±0.04**b**	0.3±0.02**b**	0.3±0.03	0.4±0.01
Zebra	4.0±0.03**c**	4.2±0.8	4.2±0.18**cd**	1.2±0.04**a**	1.4±0.20	1.6±0.02**a**	0.4±0.04**a**	0.3±0.07	0.5±0.04
ANOVA									
Main Effects									
YR	**0.0481**	**0.0239**	**0.0007**	0.0523	ns	**0.0053**	**0.0014**	**0.0118**	ns
VR	**<.0001**	ns	**<.0001**	**<.0001**	ns	**<.0001**	**<.0001**	ns	ns
Interactions									
YR:VR	ns	ns	**0.0005**	**0.011**	ns	**0.0038**	**0.0118**	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Grain Zinc and Iron concentration Assessments

For **zinc (Zn) and iron (Fe) concentrations** in the wheat grain, concentrations of Zn were higher in 2007 and highest in wheat at Sheepdrove and lowest at Courtyard. For Fe wheat concentrations were higher in 2006 and highest at Gilchesters and lowest at Sheepdrove. Zn concentrations were highest in Zebra, intermediate in Tybalt and Monsun and lowest in Amaretto. Fe concentrations were highest in Tybalt and lowest in Fasan (Table A6.20)

Table A6.20 Main effect means ± SE and *p* values for the effects of harvest year, site and variety on wheat grain micro nutrient content

	Zn mg kg^{-1}	**Fe** mg kg^{-1}
Year		
2006	32.2±0.97	34.7±0.68
2007	36.1±1.1	31.0±0.75
Site		
Courtyard	25.5±0.64**c**	27.1±0.67**b**
Gilchesters	33.3±0.65**b**	38.0±0.78**a**
Sheepdrove	43.5±0.82**a**	33.8±0.42**c**
Variety (VR)		
Amaretto	32.2±1.67**c**	32.4±1.28**b**
Fasan	32.4±1.75**bc**	30.5±1.32**c**
Monsun	35.4±1.93**b**	32.7±1.44**b**
Paragon	33.8±1.51**bc**	33.0±1.11**b**
Tybalt	33.9±1.91**b**	35.7±1.31**a**
Zebra	37.1±2.11**a**	33.3±1.11**b**
ANOVA		
Main Effects		
YR	**0.0436**	**0.0263**
ST	**<.0001**	**<.0001**
VR	**<.0001**	**<.0001**
Interactions		
YR:ST	**<.0001**	**0.0005**
YR:VR	**0.0001**	**<.0001**
ST:VR	**<.0001**[1]	**0.0145**[1]
YR:ST:VR	**0.0097**[2]	**0.0002**[2]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). [1] see Table 6.22 for interaction means SE. [2] see Table 6.23 and 6.24 for interaction means SE.

When interaction means for the interaction between variety and site were compared for the individual nutrients (Table A6.20) all varieties had the highest grain concentrations of Zn at Sheepdrove and the lowest at Courtyard (Table A6.20). For Fe concentrations all varieties had the highest concentration at Gilchesters and lowest at Courtyard, with the exception of

Monsun and Zebra where concentrations between Gilchesters and Sheepdrove were not significantly different (Table A6.20)

When interaction means for the interaction between year, variety and site were compared for individual nutrients (Table A6.21) grain concentrations of Zn at Sheepdrove were significantly higher in all varieties in 2007 than in 2006 (Table A6.21). Fe grain concentrations at Gilchesters and Courtyard were significantly higher in all varieties in 2006 compared to the following season. At Sheepdrove, with the exception of Fasan all the varieties had higher grain concentrations in 2007 than 2006 (Table A6.21)

Table A6.21 Effect means ± SE and *p* values for the effects of harvest year and variety in wheat grain micro nutrient content from each Farm

	Zn mg kg^{-1}			**Fe** mg kg^{-1}		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year						
2006	22.2±0.56	35.3±0.89	39.2±0.79	30.6±0.73	41.4±0.74	32.2±0.41
2007	28.9±0.6	31.1±0.73	47.7±0.74	23.6±0.47	34.3±0.91	35.5±0.56
Variety (VR)						
Amaretto	23.5±1.29**b**	31.9±1.29**b**	41.2±1.41**b**	26.5±1.81b**c**	37.4±1.91**b**	33.3±0.78**bc**
Fasan	23.8±1.01**b**	31.8±1.69**b**	41.6±1.95**b**	25.2±1.96**c**	35.7±2.27**b**	30.5±0.51**d**
Monsun	26.3±1.45**a**	33.5±1.73**ab**	46.2±1.56**a**	25.3±1.18**c**	37.9±2.46**b**	34.9±0.81**ab**
Paragon	27.6±1.07**a**	31.7±1.01**b**	42.0±1.75**b**	28.7±1.90**a**	37.6±1.50**b**	33.2±0.82**c**
Tybalt	23.9±1.50**b**	36.2±1.89**a**	41.8±2.46**b**	29.4±1.32**a**	41.5±1.65**a**	36.1±1.36**a**
Zebra	28.0±2.27**a**	34.9±1.45**a**	48.1±1.94**a**	27.4±1.41**ab**	38.1±1.02**b**	35.0±0.53**a**
ANOVA						
Main Effects						
YR	**0.0069**	ns	**0.0013**	**0.017**	**0.0495**	**0.0039**
VR	**<.0001**	**0.0045**	**<.0001**	**0.0005**	**0.0001**	**<.0001**
Interactions						
YR:VR	**0.0016**[1]	**0.0079**[1]	**0.0429**[1]	ns	**<.0001**[2]	**0.006**[2]

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1] see Table A6.22 for interaction means SE. [2] see Table A6.23 for interaction means SE. COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.22 Interaction means for effects of wheat variety and site on grain Zinc (Zn) concentrations

	Zn mg kg^{-1}					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	23.59±1.29**Cb**	23.89±1.01**Cb**	26.36±1.45**Ca**	27.63±1.07**Ca**	23.94±1.5**Cb**	28.01±2.27**Ca**
GIL	31.92±1.29**Bb**	31.83±1.69**Bb**	33.55±1.73**Bab**	31.74±1.01**Bb**	36.2±1.89**Ba**	34.94±1.45**Ba**
SHE	41.2±1.41**Ab**	41.6±1.95**Ab**	46.28±1.56**Aa**	42.03±1.75**Ab**	41.82±2.46**Ab**	48.16±1.94**Aa**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.23 Interaction means for effects of wheat variety and site on grain Iron (Fe) concentrations

	Fe mg kg^{-1}					
	Amaretto	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
COU	26.5±1.81**Cbc**	25.2±1.96**Cc**	25.3±1.18**Bc**	28.7±1.9**Ca**	29.4±1.32**Ca**	27.4±1.41**Bab**
GIL	37.4±1.91**Ab**	35.7±2.27**Ab**	37.9±2.46**Ab**	37.6±1.5**Ab**	41.5±1.65**Aa**	38.1±1.02**Ab**
SHE	33.3±0.78**Bbc**	30.5±0.51**Bd**	34.9±0.81**Aab**	33.2±0.82**Bc**	36.1±1.36**Ba**	35.0±0.53**Aa**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.24 Interaction means for effects of year, wheat variety and site on grain Zinc (Zn) concentrations

		Zn mg kg^{-1}					
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	20.4±0.54	21.8±1.12	22.8±1.27	25.4±1.19	20.4±1.27	22.4±1.58
	GIL	34.0±0.16	35.5±1.74	36.9±2.23	32.7±1.61	38.6±3.18	34.1±2.32
	SHE	37.5±0.42	36.6±0.97	42.7±0.75	39.8±2.9	35.5±1.28	43.1±0.37
2007	**COU**	26.7±0.92	25.9±0.77	29.8±0.27	29.7±0.88	27.5±0.76	33.6±0.87
	GIL	29.7±1.37	28.1±1.03	30.1±1.06	30.4±0.72	33.7±1.61	35.9±1.72
	SHE	44.8±0.27	46.6±0.17	49.8±1.56	44.2±1.64	48.1±0.77	53.1±0.87

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.25 Interaction means for effects of year, wheat variety and site on Iron (Fe) concentrations

		Fe mg kg^{-1}					
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	30.3±2.20	30.2±1.30	28.2±1.00	32.01±3.10	32.1±1.70	31.03±0.76
	GIL	41.5±1.35	41.3±0.80	43.06±2.50	40.47±0.70	44.5±2.00	38.04±1.80
	SHE	31.5±0.50	30.2±1.00	33.5±1.30	31.4±0.90	32.7±0.45	33.8±0.60
2007	**COU**	22.7±0.80	20.3±0.30	22.5±0.30	25.5±0.50	26.7±0.36	23.9±0.50
	GIL	33.4±2.08	30.1±1.50	32.8±2.00	33.7±1.36	38.5±1.68	38.2±0.40
	SHE	35.1±0.70	31.0±0.30	36.3±0.36	35±0.40	39.5±0.80	36.2±0.10

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Grain heavy metal concentration assessments

For grain **heavy metal concentrations** analysed (Table A6.26), the season/year only had a significant effect on cadmium and lead concentrations. Grain concentrations of Molybdenum were significantly lower at Gilchesters and for Cadmium and Nickel lower at Courtyard. Concentrations of Aluminium and Nickel were significantly higher at Gilchesters. Lead had no significant differences for year, site or variety. Otherwise, Zebra had the highest levels of all heavy metal concentrations, Monsun had significantly higher levels of cadmium and Paragon and Tybalt had significantly lower levels of molybdenum. Significant interactions were detected between season/year and site, year and variety, site and variety, and between year, site and variety (Table A6.26). Interaction means for year, site and variety (Table A6.27) identified that Mo concentrations were lower for all varieties in both years at Gilchesters but there was a significant increase in grain concentrations for all varieties at Gilchesters in 2007. For Cd, year, site and variety interaction means identified that grain concentrations at Gilchesters were higher in 2006 for all varieties and that concentrations there decreased in 2007. At both Courtyard and Sheepdrove grain concentrations of Cd were lower in 2006 with grain concentrations increasing for all varieties in 2007 (Table A6.28). Zebra and Monsun recorded the highest concentrations at Gilchesters and Sheepdrove.

Site and year interaction means for grain concentrations of nickel and lead, identified significant increases of nickel in 2007 at Courtyard and Gilchesters and for lead significant increases in 2007 at Gilchesters and Sheepdrove, otherwise there were no significant site or variety interactions.

Table A6.26 Main effect means ± SE and *p* values for the effects of harvest year, site and variety on wheat grain heavy metal content

	Mo µg kg^{-1}	**Al** µg kg^{-1}	**Cd** µg kg^{-1}	**Ni** µg kg^{-1}	**Pb** µg kg^{-1}
Year					
2006	0.8±0.08	2.0±0.09	36.5±2.18	145.7±10.85	56.6±5.46
2007	0.7±0.04	2.5±0.12	45.0±1.57	137.0±3.67	109.6±6.67
Site					
Courtyard	1.1±0.04**a**	2.1±0.12**b**	34.9±2.53**b**	94.7±5.59**c**	88.2±7.04
Gilchesters	0.1±0.02**b**	2.7±0.16**a**	44.1±2.35**a**	201.8±11.07**a**	90.5±9.47
Sheepdrove	1.0±0.06**a**	1.9±0.08**b**	43.2±2.15**a**	130.3±5.43**b**	69.7±8.34
Variety (VR)					
Amaretto	0.5±0.07**d**	2.1±0.13**b**	36.2±3.32**b**	136.7±12.9**b**	82.4±12.28
Fasan	0.8±0.11**b**	2.1±0.19**b**	36.7±2.89**b**	131.8±12.45**b**	88.7±12.1
Monsun	0.8±0.12**b**	2.1±0.14**b**	48.0±3.17**a**	130.9±13.25**b**	76.2±11.3
Paragon	0.7±0.09**c**	2.2±0.15**b**	35.8±2.69**b**	133.2±11.85**b**	78.8±10.09
Tybalt	0.7±0.09**c**	1.9±0.11**b**	38.1±3.61**b**	154.2±17.51**ab**	84.2±13.36
Zebra	0.9±0.14**a**	2.9±0.29**a**	49.7±3.72**a**	162.3±16.27**a**	86.0±12.54
ANOVA					
Main Effects					
YR	ns	0.053	**0.0087**	ns	**0.01**
ST	**<.0001**	**0.0011**	**0.0002**	**<.0001**	ns
VR	**<.0001**	**<.0001**	**<.0001**	**0.039**	ns
Interactions					
YR:ST	**0.0001**	**0.0333**	**<.0001**	**<.0001**	**0.0087**
YR:VR	**<.0001**	ns	Ns	ns	ns
ST:VR	**<.0001**	ns	**0.0008**	ns	ns
YR:ST:VR	**<.0001**[1]	**0.0007**	**0.0091**	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1] See Table 6.27 for interaction means SE.

Table A6.27 Interaction means for effects of year, wheat variety and site on grain Molybdenum (Mo) concentrations.

		Mo mg kg^{-1}					
Year	**Site**	**Amaretto**	**Fasan**	**Monsun**	**Paragon**	**Tybalt**	**Zebra**
2006	**COU**	0.7±0.1**b**	1.2±0.02**b**	1.2±0.016**b**	0.9±0.22**ab**	0.9±0.15**ab**	1.4±0.09**b**
	GIL	0.0±0.01**e**	0.0±0.01**e**	0.0±0.0**e**	0.0±0.02**d**	0.0±0.01**d**	0.0±0.0**e**
	SHE	0.9±0.05**a**	1.4±0.11**a**	1.6±0.1**a**	1.0±0.04**a**	1.0±0.08**a**	1.9±0.01**a**
2007	**COU**	0.8±0.03**a**	1.2±0.02**b**	1.1±0.09**b**	1.1±0.07**a**	1.1±0.03**a**	1.3±0.04**b**
	GIL	0.2±0.06**d**	0.3±0.07**d**	0.2±0.06**d**	0.3±0.10**c**	0.3±0.07**c**	0.3±0.07**d**
	SHE	0.5±0.03**c**	0.7±0.03**c**	0.6±0.02**c**	0.7±0.04**b**	0.7±0.01**b**	0.7±0.01**c**

Means labelled with the same letter within the same column for each year are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.28 Effect means ± SE and *p* values for the effects of harvest year and variety on wheat grain heavy metal content for each Farm

	Mo mg kg^{-1}			Al mg kg^{-1}			Cd μg kg^{-1}		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year									
2006	1.0±0.08	0.0±0.01	1.3±0.08	2.1±0.23	2.2±0.10	1.6±0.09	19.5±1.49	57.0±1.99	32.9±2.51
2007	1.1±0.04	0.2±0.03	0.6±0.02	2.1±0.1	3.3±0.27	2.1±0.10	50.2±1.85	30.1±1.39	53.6±1.84
Variety (VR)									
Amaretto	0.8±0.05	0.1±0.05**c**	0.7±0.08**d**	2.7±0.23	2.5±0.20	1.7±0.16	32.4±6.43	39.2±6.08**c**	37.1±5.21**b**
Fasan	1.2±0.1	0.1±0.06**a**	1.1±0.14**bc**	1.8±0.22	2.9±0.40	1.7±0.08	33.5±6.23	41.2±4.45**bc**	35.3±4.36**b**
Monsun	1.1±0.09	0.1±0.05**bc**	1.1±0.2**ab**	1.9±0.11	2.5±0.33	1.9±0.19	39.0±4.91	51.3±5.99**a**	53.7±4.62**a**
Paragon	1.0±0.11	0.2±0.08**a**	0.8±0.08**cd**	2.0±0.19	2.7±0.31	2.1±0.24	33.5±3.97	36.6±5.06**c**	37.3±5.42**b**
Tybalt	1.0±0.08	0.1±0.06**ab**	0.9±0.08**cd**	1.8±0.11	2.4±0.20	1.6±0.15	27.1±6.51	46.3±6.35**ab**	40.8±4.16**b**
Zebra	1.3±0.05	0.1±0.07**ab**	1.3±0.24**a**	3.2±0.49	3.5±0.70	2.2±0.21	43.8±8.47	50.0±6.09**a**	55.3±4.04**a**
ANOVA									
Main Effects									
YR	ns	**0.0437**	**0.002**	ns	**0.0223**	ns	**0.0002**	**0.0008**	**0.0016**
VR	**<.0001**	**0.0008**	**<.0001**	**0.0004**	0.0816	0.0688	**<.0001**	**0.0004**	**<.0001**
Interactions									
YR:VR	ns	**0.0143**	**<.0001**	0.0758	**0.0168**	ns	**0.0001**	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$ COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A6.28 (cont.) Effect means ± SE and *p* values for the effects of harvest year and variety on wheat grain heavy metal content for each Farm

	Ni $\mu g\ kg^{-1}$			Pb $\mu g\ kg^{-1}$		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year						
2006	65.3±4.85	244.2±16	127.7±9.66	85.5±10.30	50.1±6.93	34.2±7.86
2007	124.1±5.36	155.6±6.97	132.9±5.16	91.0±9.78	134.7±12.91	105.1±10.65
Variety (VR)						
Amaretto	92.0±11.87	201.8±19.5	116.2±13.01	70.0±14.39	87.0±26.58	90.1±23.25
Fasan	94.5±14.57	181.2±22.52	119.7±15.55	70.9±15.62	109.8±20.57	85.5±25.81
Monsun	78.2±16.73	191.3±21.53	123.3±7.77	88.3±17.09	93.0±22.06	47.3±17.3
Paragon	95.3±14.05	171.4±22.7	137.8±17.15	97.2±18.17	79.6118.94	59.6±14.81
Tybalt	104.6±15.25	229.9±37.03	128.0±12.28	98.4±23.07	89.5±27.30	64.7±19.88
Zebra	103.4±11.37	235.9±36.01	156.7±10.60	104.6±15.30	82.0±28.40	70.9±22.29
ANOVA						
Main Effects						
YR	**0.0045**	**0.0305**	ns	ns	**0.0217**	**0.0138**
VR	ns	ns	ns	ns	ns	ns
Interactions						
YR:VR	ns	ns	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$
COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

For the assessment of **Phytic Acid levels** in the grain along with the concentrations of phosphorous (P) and the molar ratios of zinc (Zn) and iron (Fe) there were no significant effects of year or site for the grain phytic acid levels or P concentrations (Table A6.29). The phytic acid Zn and Fe ratios were both highest in grains from Courtyard with the lowest Zn ratio in grains from Sheepdrove and the lowest Fe ratio in grains from Gilchesters (Table A6.29). Fasan was the variety with the highest levels of phytic acid, P concentrations in the phytic acid and with the highest phytic acid Zn and Fe ratios. Tybalt had the lowest levels (Table A6.29). For site and variety interactions (Table A6.30) phytic acid and P concentrations increased significantly at Courtyard in 2007, Fasan had the highest levels there but not significantly different to Paragon and Zebra. Significant differences existed between varieties at each site but Fasan remained the variety with the highest levels at each farm and Tybalt with the lowest (Table A6.30)

Table A6.29 Main effect means ± SE and *p* values for the effects of harvest year, site and variety on spring wheat grain phytic acid content.

	Phytic acid mg g^{-1}	**Phytic acid P** mg g^{-1}	**Phytic acid Zn** molar ratio	**Phytic acid Fe** molar ratio
Year				
2006	9.4±0.15	2.6±0.04	30.7±0.92	23.5±0.55
2007	9.9±0.15	2.8±0.04	29.2±1.01	28.8±1.08
Site				
Courtyard	9.7±0.2	2.7±0.06	38.2±0.76**a**	31.6±1.29**a**
Gilchesters	9.5±0.14	2.7±0.04	28.9±0.7**b**	21.8±0.65**c**
Sheepdrove	9.8±0.21	2.7±0.06	22.7±0.71**c**	24.7±0.70**b**
Variety (VR)				
Amaretto	9.3±0.19**c**	2.6±0.05**cd**	30.4±1.67**b**	25.4±1.46**b**
Fasan	10.9±0.23**a**	3.0±0.06**a**	35.3±1.73**a**	31.8±1.78**a**
Monsun	9.5±0.27**c**	2.7±0.08**bc**	28.3±1.37**c**	26.0±1.56**b**
Paragon	9.7±0.26**bc**	2.7±0.07**bc**	29.8±1.5**bc**	25.7±1.30**b**
Tybalt	8.8±0.21**d**	2.5±0.06**d**	27.8±1.65**c**	21.8±1.11**c**
Zebra	9.8±0.25**b**	2.7±0.07**b**	28.3±1.73**c**	26.0±1.47**b**
ANOVA				
Main Effects				
YR	ns	ns	ns	**0.0238**
ST	ns	ns	**<.0001**	**<.0001**
VR	**<.0001**	**<.0001**	**<.0001**	**<.0001**
Interactions				
YR:ST	**0.0001**	**0.0001**	**0.0006**	**<.0001**
YR:VR	ns	ns	**0.0015**	**0.0347**
ST:VR	**0.0008**	**0.0008**	**0.0092**	**0.0225**
YR:ST:VR	ns	ns	0.0549	**0.0261**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table A6.30 Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat grain phytic acid content for each Farm

	Phytic acid mg g^{-1}			**Phytic acid P** mg g^{-1}		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year		P45			P46	
2006	8.6±0.18	9.6±0.17	10.0±0.33	2.4±0.05	2.7±0.05	2.8±0.09
2007	10.8±0.19	9.5±0.22	9.5±0.27	3.0±0.05	2.6±0.06	2.6±0.08
Variety (VR)						
Amaretto	9.4±0.40**bc**	8.9±0.17**c**	9.4±0.38**bcd**	2.6±0.11**bc**	2.5±0.05**c**	2.6±0.11**bc**
Fasan	10.6±0.32**a**	10.3±0.28**a**	11.7±0.42**a**	3.0±0.09**a**	2.9±0.08**a**	3.3±0.12**a**
Monsun	9.2±0.62**c**	9.3±0.28**c**	10.1±0.42**b**	2.6±0.17**c**	2.6±0.08**c**	2.8±0.12**b**
Paragon	10.0±0.55**ab**	10.1±0.26**ab**	9.0±0.4**cd**	2.8±0.16**ab**	2.8±0.07**ab**	2.5±0.11**c**
Tybalt	8.9±0.46**c**	9.2±0.02**c**	8.4±0.37**d**	2.5±0.13**c**	2.6±0.06**c**	2.3±0.1**c**
Zebra	10.0±0.46**ab**	9.5±0.50**bc**	10.0±0.36**bc**	2.8±0.13**ab**	2.6±0.14**bc**	2.8±0.1**bc**
ANOVA						
Main Effects						
YR	**0.0037**	ns	ns	**0.0037**	ns	ns
VR	**0.0007**	**0.0008**	**<.0001**	**0.0007**	**0.0008**	**<.0001**
Interactions						
YR:VR	ns	ns	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

Table A6.30 (cont.) Effect means ± SE and *p* values for the effects of harvest year and variety on spring wheat grain phytic acid content for each Farm

	Phytic acid Zn molar ratio			**Phytic acid Fe** molar ratio		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year		P48			P49	
2006	39.2±1.20	27.4±0.91	25.6±0.96	24.2±0.74	19.8±0.58	26.5±0.95
2007	37.3±0.94	30.6±0.97	19.8±0.65	39.1±1.19	23.9±1.04	22.9±0.91
Variety (VR)						
Amaretto	40.2±1.60**b**	28.1±0.87**b**	22.9±1.27**b**	31.6±3.15**b**	20.6±1.03**bc**	24.0±1.01**b**
Fasan	44.4±1.43**a**	32.9±1.92**a**	28.6±2.18**a**	37.6±3.82**a**	25.4±2.05**a**	32.5±1.33**a**
Monsun	35.0±1.61**c**	27.9±1.47**b**	21.9±1.23**b**	31.9±3.4**b**	21.4±1.76**bc**	24.6±1.10**b**
Paragon	36.1±1.40**c**	31.8±1.02**a**	21.6±1.34**b**	30.6±2.76**b**	23.1±1.38**ab**	23.0±1.00**b**
Tybalt	37.3±1.08**bc**	25.8±1.52**b**	20.5±1.47**b**	26.3±2.26**c**	19.1±1.08**c**	19.9±1.15**c**
Zebra	36.4±2.11**c**	27.3±1.98**b**	20.9±1.49**b**	31.8±2.99**b**	21.3±1.42**bc**	24.2±1.02**b**
ANOVA						
Main Effects						
YR	Ns	ns	**0.0059**	**0.0018**	ns	**0.0218**
VR	**<.0001**	**0.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**
Interactions						
YR:VR	0.0949	**0.0064**	0.0589	ns	**0.0004**	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$)

APPENDIX 7: Chapter 5 Results tables for all three sites for both years

Effect of fertiliser input type and rate on disease severity

Table A7.1 Effect means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat disease severity

	MILD L1	MILD L2	SEPT L1	SEPT L2
	%	%	%	%
Year				
2006	0.6±0.11	0.8±0.13	0.3±0.06	5.1±0.64
2007	0.0±0.00	1.0±0.15	0.0±0.00	3.3±0.27
Site				
Courtyard	0.7±0.16 **a**	2.1±0.2 **a**	0.4±0.09 **a**	4.6±0.26 **b**
Gilchesters	0.0±0.00 **b**	0.1±0.04 **c**	0.0±0.00 **b**	0.5±0.09 **c**
Sheepdrove	0.2±0.04 **b**	0.5±0.12 **b**	0±0.01 **b**	7.4±0.82 **a**
Fertility rate				
125 kg N ha^{-1}	0.3±0.09	0.9±0.14	0.1±0.03	3.9±0.46
250 kg N ha^{-1}	0.3±0.07	0.9±0.14	0.2±0.06	4.4±0.53
Fertility Type				
CP	0.6±0.19**a**	1.3±0.24**a**	0.2±0.11**a**	4.4±0.59
FYM	0.1±0.04**b**	0.6±0.12**b**	0.1±0.03**b**	4.8±0.91
FYM+CP	0.3±0.13**ab**	1.1±0.23**a**	0.1±0.03**b**	3.9±0.51
GWC	0.1±0.04**b**	0.6±0.16**b**	0.1±0.04**ab**	3.7±0.76
ANOVA				
Main Effects				
Year (YR)	**0.0107**	ns	**0.0244**	**0.0324**
Site (ST)	**0.0002**	**<0.0001**	**0.0002**	**<0.0001**
Fertility rate (FR)	ns	ns	ns	ns
Fertility type (FT)	**0.0004**	**<0.0001**	**0.0267**	ns
Interactions				
YR x ST	**0.0002**	ns	**0.0002**	**<0.0001**
YR x FR	ns	ns	ns	ns
ST x FR	ns	ns	ns	ns
YR x FT	**0.0004**	ns	**0.0267**	ns
ST x FT	**0.0034**	**0.012**	**0.0055**	**0.0232**
FR x FT	ns	ns	ns	ns
YR x ST x FR	ns	ns	ns	ns
YR x ST x FT	**0.0033**	**0.0001**	**0.0055**	**0.0008**
YR x FR x FT	ns	**0.0236**	ns	ns
ST x FR x FT	ns	ns	ns	ns
YR x ST x FR x FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Table A7.2 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat disease severity for each site

	MILD L1			MILD L2			SEPT L1			SEPT L2		
	%			%			%			%		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year												
2006	1.5±0.26	0.0±0.00	0.3±0.07	2.1±0.26	0.0±0.00	0.1±0.05	0.8±0.15	0.0±0.00	0.0±0.00	3.6±0.28	0.0±0.00	11.7±1.18
2007	0.0±0.00	0.0±0.00	0.0±0.00	2.1±0.31	0.1±0.07	0.8±0.17	0.0±0.00	0.0±0.00	0.0±0.00	5.6±0.36	1.1±0.13	3.1±0.41
Fertility rate												
125 kg N ha^{-1}	0.8±0.26	0.0±0.00	0.1±0.05	2.3±0.28	0.0±0.06	0.4±0.12	0.3±0.06	0.0±0.00	0.0±0.00	4.4±0.36	0.6±0.17	6.8±1.09
250 kg N ha^{-1}	0.7±0.2	0.0±0.00	0.1±0.07	2.0±0.29	0.0±0.03	0.6±0.16	0.5±0.16	0.0±0.00	0.0±0.00	4.8±0.37	0.5±0.09	8±1.25
Fertility Type												
CP	1.4±0.49**a**	0.0±0.00	0.3±0.14**a**	2.8±0.43**a**	0.1±0.12	1.0±0.27**a**	0.7±0.31**a**	0.0±0.00	0.0±0.00	4.8±0.17**ab**	0.7±0.31	7.7±1.21**ab**
FYM	0.4±0.11**b**	0.0±0.00	0.0±0.03**b**	1.3±0.24**c**	0.0±0.06	0.2±0.10**bc**	0.3±0.08**b**	0.0±0.00	0.0±0.00	4.5±0.66**bc**	0.5±0.13	9.3±2.17**a**
FYM+CP	0.8±0.34**ab**	0.0±0.00	0.1±0.08**ab**	2.6±0.46**ab**	0.0±0.00	0.6±0.21**b**	0.2±0.08**b**	0.0±0.00	0.0±0.00	5.7±0.56**a**	0.5±0.13	5.5±0.98**b**
GWC	0.4±0.11**b**	0.0±0.00	0.0±0.01**b**	1.7±0.34**bc**	0.0±0.03	0.0±0.06**c**	0.3±0.1**ab**	0.0±0.00	0.0±0.00	3.5±0.42**c**	0.5±0.12	7.1±1.95**ab**
ANOVA												
Main Effects												
YR	**0.0147**	ns	**0.0679**	ns	ns	**0.0168**	**0.0224**	ns	ns	**0.0127**	**0.0053**	**0.0071**
FR	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
FT	**0.0083**	ns	**0.0203**	**0.0116**	ns	**0.0001**	**0.0321**	ns	ns	**0.0029**	ns	ns
Interactions												
YR:FR	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
YR:FT	**0.0083**	ns	**0.0203**	**0.0256**	ns	**0.0025**	**0.0321**	ns	ns	**0.0311**	ns	**0.0226**
FR:FT	ns	ns	**0.08**	ns	ns	ns	ns	ns	ns	ns	ns	ns
YR:FR:FT	ns	ns	**0.08**	ns	ns	ns	ns	ns	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)
COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Effect of fertiliser input type and rate on crop performance

Table A7.3 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain yield, plant height SPAD and TGW

	Grain Yield	Plant height	SPAD	TGW
	t ha^{-1}	cm	GS65	g
Year				
2006	4.5±0.09	93.7±0.69	49.8±0.42	3.7±0.03
2007	2.8±0.07	84.2±0.78	42.5±0.42	37.5±0.5
Site				
Courtyard	3.6±0.1 **b**	88.4±0.49	43.9±0.43 **b**	20.3±2.13
Gilchesters	4.2±0.22**a**	89.4±0.87	49.8±0.59 **a**	20.3±2.64
Sheepdrove	3.2±0.08**c**	89.2±1.59	44.8±0.76 **b**	18.7±1.93
Fertility Rate				
125 kg N ha^{-1}	3.7±0.12	89.2±0.91	45.7±0.55	20.1±1.8
250 kg N ha^{-1}	3.7±0.12	88.7±0.86	46.7±0.57	19.4±1.82
Fertility Type				
CP	3.5±0.2**b**	88.9±1.28	47.8±0.65**a**	19.1±2.39**c**
FYM	3.8±0.16**ab**	89.3±1.23	45±0.8**b**	20.3±2.72**a**
FYM+CP	3.6±0.17**ab**	88.5±1.31	47.1±0.74**a**	19.7±2.53**b**
GWC	3.8±0.15**a**	89.2±1.22	44.9±0.91**b**	20±2.66**a**
ANOVA				
Main Effects				
Year (YR)	**0.0009**	**0.0027**	**0.001**	**<.0001**
Site (ST)	**0.0005**	ns	**<.0001**	**<.0001**
Fertility rate (FR)	ns	ns	**0.0255**	ns
Fertility type (FT)	**0.0235**	ns	**<.0001**	**<.0001**
Interactions				
YR x ST	**<.0001**	**<.0001**	**0.0006**	**<.0001**
YR x FR	ns	ns	ns	ns
ST x FR	ns	ns	ns	ns
YR x FT	**0.0032**	ns	**0.0136**	**<.0001**
ST x FT	**0.0005**	ns	**0.0136**	ns
FR x FT	ns	ns	**0.073**[1]	ns
YR x ST x FR	**0.0139**	ns	ns	0.0701
YR x ST x FT	ns	ns	ns	0.0804
YR x FR x FT	ns	**<.0001**	ns	ns
ST x FR x FT	ns	ns	ns	ns
YR x ST x FR x FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$) [1] See Table 7.5 for interaction means SE.

Table A7.4 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain yield, plant height SPAD and TGW for each site

	Grain Yield			**Plant height**			**GS65 SPAD**			**TGW**		
	t ha^{-1}			cm			%			g		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year												
2006	4.2±0.11	5.5±0.1	3.7±0.05	86.5±0.42	93.4±0.6	101.2±0.6	45.6±0.55	53.8±0.33	50.2±0.31	3.5±0.03	3.9±0.03	3.5±0.02
2007	3±0.1	2.5±0.15	2.7±0.11	90.3±0.76	85.3±1.28	77.1±0.76	42.3±0.53	45.8±0.55	39.4±0.61	37.1±0.43	43.1±0.73	34±0.54
Fertility rate												
125 kg N ha^{-1}	3.6±0.12	4.3±0.3	3.2±0.12	88.7±0.58	90.3±1.13	88.6±2.44	43.1±0.58	49.3±0.82	44.6±1.06	20.7±3.09	21.3±3.66	18.5±2.7
250 kg N ha^{-1}	3.6±0.18	4.2±0.32	3.2±0.11	88.1±0.8	88.4±1.32	89.7±2.09	44.8±0.61	50.3±0.87	45±1.1	20±2.98	19.2±3.89	19±2.82
Fertility type												
CP	3.5±0.27	4.4±0.42**a**	2.8±0.22**b**	88.4±1.13	90.2±1.81	88±3.25	46.3±0.84**a**	50.2±1.04	46.9±1.24**a**	19.3±4.15**b**	21.1±4.96**a**	16.9±3.47
FYM	3.6±0.16	4.5±0.44**ab**	3.3±0.13**a**	88.1±0.83	90±1.68	89.9±3.26	42.5±0.58**b**	49.2±1.05	43.2±1.67**b**	21.2±4.58**a**	19.6±5.77**b**	20±4.28
FYM+CP	3.6±0.23	3.8±0.45**b**	3.3±0.15**a**	88.4±1.12	87.6±1.9	89.5±3.33	44.9±0.81**a**	50.5±1.29	45.8±1.25**a**	20.1±4.31**ab**	20.8±5.41**ab**	18.2±3.78
GWC	3.8±0.17	4.3±0.44**ab**	3.4±0.1**a**	88.7±0.9	89.6±1.67	89.3±3.21	42±0.8**b**	49.4±1.41	43.2±1.74**b**	20.6±4.4**a**	19.5±5.7**b**	19.8±4.21
ANOVA												
Main Effects												
YR	**0.0179**	**0.0005**	**0.0138**	**0.0266**	**0.0265**	**0.0004**	**0.044**	**0.0048**	**0.0012**	**<.0001**	**0.0001**	**<.0001**
FR	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
FT	ns	**0.0479**	**<.0001**	ns	ns	ns	**<.0001**	ns	**<.0001**	**0.0077**	**0.0033**	**<.0001**
Interactions								ns				
YR:FR	**0.0164**	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
YR:FT	**0.0583**	ns	**0.0004**	ns	ns	ns	ns	ns	**0.0003**	**0.0171**	**0.0007**	**<.0001**
FR:FT	ns	ns	ns	ns	ns	ns	ns	ns	**0.0245**	ns	**0.0004**	ns
YR:FR:FT	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	**<.0001**	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)
COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A7.5 Interaction means for effects of fertility type and fertility rate on wheat SPAD at Sheepdrove farm

	GS65 SPAD			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	45.75±0.27**Ba**	43.06±0.29**Ab**	45.92±0.3**Aa**	43.62±0.27**Ab**
250 kg N ha^{-1}	48.07±0.22**Aa**	43.36±0.27**Ac**	45.69±0.27**Ab**	42.7±0.28**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Table A7.6 Interaction means for effects of fertiliser rate and type on grain (kernel) weight at Gilchesters farm

	Thousand Grain Weight g			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	21.42±6.68**Ab**	17.86±8.79**Ac**	23.24±7.25**Aa**	21.99±8.46**Aac**
250 kg N ha^{-1}	20.65±7.95**Ba**	21.12±8.23**Aa**	17.66±8.71**Aa**	16.67±8.12**Ba**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Effect of fertiliser input type and rate on quality parameters

Table A7.7 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain protein and N content

	NIR Protein	**Grain N**
	%	%
Year		
2006	11.6±0.15	2.2±0.03
2007	12.5±0.11	2.4±0.02
Site		
Courtyard	10.8±0.17**c**	2.1±0.04**b**
Gilchesters	12.9±0.11**a**	2.5±0.02**a**
Sheepdrove	12.4±0.09**b**	2.4±0.02**b**
Fertility Rate		
125 kg N ha^{-1}	11.9±0.14	2.3±0.03
250 kg N ha^{-1}	12.1±0.14	2.3±0.03
Fertility Type		
CP	12.6±0.18**a**	2.5±0.04a
FYM	11.6±0.20**c**	2.2±0.04c
FYM+CP	12.2±0.20**b**	2.4±0.04b
GWC	11.5±0.19**c**	2.2±0.04c
ANOVA		
Main Effects		
Year (YR)	**0.0102**	**0.009**
Site (ST)	**<.0001**	**<.0001**
Fertility rate (FR)	**0.0004**	**0.0165**
Fertility type (FT)	**<.0001**	**<.0001**
Interactions		
YR x ST	**0.0013**	**0.0003**
YR x FR	ns	ns
ST x FR	**0.0033**	**0.0062**
YR x FT	**0.0171**	**0.0004**
ST x FT	**0.0011**	**0.0001**
FR x FT	**0.0001**[1]	**0.0317**[2]
YR x ST x FR	ns	ns
YR x ST x FT	**<.0001**	**<.0001**
YR x FR x FT	ns	ns
ST x FR x FT	**0.0009**	**<.0001**
YR x ST x FR x FT	**0.0025**	**0.004**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$) [1] see table 9.9 for interaction means SE.

Table A7.8 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain protein and N content for each site

	NIR Protein			**N**		
	%			%		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year						
2006	9.8±0.16	12.9±0.10	12.1±0.08	1.9±0.03	2.4±0.02	2.3±0.02
2007	11.9±0.14	13±0.22	12.8±0.15	2.3±0.03	2.5±0.05	2.5±0.03
Fertility rate						
125 kg N ha^{-1}	10.6±0.22	12.8±0.13	12.4±0.11	2.1±0.05	2.4±0.03	2.4±0.03
250 kg N ha^{-1}	11.1±0.26	1.03±0.18	12.4±0.14	2.2±0.05	2.5±0.03	2.4±0.03
Fertility type						
CP	11.6±0.35**a**	13.2±0.16**a**	13.1±0.18**a**	2.3±0.08**a**	2.5±0.03**a**	2.6±0.06**a**
FYM	10.5±0.35**c**	12.4±0.22**b**	12.0±0.11**c**	2.0±0.08**c**	2.3±0.04**b**	2.3±0.03**c**
FYM+CP	11±0.33**b**	13.3±0.21**a**	12.5±0.08**b**	2.1±0.07**b**	2.5±0.04**a**	2.5±0.03**b**
GWC	10.3±0.26**c**	12.6±0.21**b**	11.8±0.10**c**	2.0±0.05**c**	2.4±0.05a**b**	2.3±0.03**c**
ANOVA						
Main Effects						
YR	**0.0086**	ns	**0.0022**	**0.0063**	ns	**0.0018**
FR	**0.0007**	ns	ns	**0.0037**	ns	ns
FT	**<.0001**	**0.0008**	**<.0001**	**<.0001**	**0.0005**	**<.0001**
Interactions						
YR:FR	ns	0.4591	ns	ns	ns	ns
YR:FT	**0.0257**	**0.0036**	**0.0001**	**0.0071**	**0.0059**	**<.0001**
FR:FT	**<.0001**	ns	**0.0091**	**0.0001**	0.0664	ns
YR:FR:FT	ns	**0.0113**	ns	ns	**0.0197**	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove.

Table A7.9 Interaction means for effects of fertiliser rate and type on wheat grain protein content at courtyard farm

	Grain NIR Protein %			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	10.9±0.39**Ba**	10.56±0.56**Aa**	10.58±0.44**Ba**	10.18±0.36**Ab**
250 kg N ha^{-1}	12.32±0.48**Aa**	10.46±0.45**Ac**	11.37±0.46**Ab**	10.36±0.34**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$).

Table A7.10 Interaction means for effects of fertiliser rate and type on wheat grain protein content at Sheepdrove farm

	NIR Protein %			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	12.97±0.26**Aa**	12.01±0.16**Ac**	12.44±0.15A**b**	12.06±0.14**Ac**
250 kg N ha^{-1}	13.24±0.25**Aa**	11.98±0.16**Ac**	12.61±0.07**Ab**	11.63±0.11**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$).

Table A7.11 Interaction means for effects of fertiliser rate and type on wheat grain N content at courtyard farm

	Grain N %			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	2.13±0.08**Ba**	2.05±0.13**Aab**	2.06±0.1**Bab**	1.2±0.07**Ab**
250 kg N ha^{-1}	2.41±0.11**Aa**	2.03±0.1**Ac**	2.21±0.09**Ab**	2.01±0.08**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$).

Effect of fertiliser input type and rate on grain macro nutrient concentrations

Table A7.12 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain P, K and S content

	Grain P	**Grain K**	**Grain S**
	%	%	%
Year			
2006	0.36±0.003	0.45±0.002	0.14±0.001
2007	0.42±0.002	0.5±0.005	0.15±0.001
Site			
Courtyard	0.39±0.005**b**	0.45±0.002**c**	0.13±0.002**b**
Gilchesters	0.36±0.005**c**	0.49±0.007**a**	0.15±0.001**a**
Sheepdrove	0.41±0.003**a**	0.47±0.005**b**	0.15±0.001**a**
Fertility Rate			
125 kg N ha^{-1}	0.39±0.004	0.47±0.004	0.14±0.001
250 kg N ha^{-1}	0.38±0.004	0.47±0.004	0.14±0.001
Fertility Type			
CP	0.39±0.007	0.48±0.007**a**	0.15±0.002**a**
FYM	0.39±0.006	0.46±0.005**b**	0.14±0.002**c**
FYM+CP	0.38±0.006	0.47±0.007**ab**	0.14±0.002**b**
GWC	0.38±0.005	0.46±0.005**b**	0.14±0.002**c**
ANOVA			
Main Effects			
Year (YR)	**0.0004**	**0.0023**	**0.0059**
Site (ST)	**<.0001**	**0.0001**	**<.0001**
Fertility rate (FR)	**0.0431**	ns	**0.0242**
Fertility type (FT)	ns	**<.0001**	**<.0001**
Interactions			
YR x ST	**0.0097**	**<.0001**	**0.0001**
YR x FR	**0.0102**	ns	ns
ST x FR	**0.0219**	ns	**0.0187**
YR x FT	**0.0028**	**<.0001**	**0.0011**
ST x FT	**0.0289**	**0.0001**	**<.0001**
FR x FT	ns	ns	**0.024**
YR x ST x FR	ns	ns	ns
YR x ST x FT	**0.0898**	**0.0008**	**<.0001**
YR x FR x FT	ns	ns	ns
ST x FR x FT	ns	ns	**0.0027**
YR x ST x FR x FT	ns	ns	**0.0219**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$). See table 7.15 for interaction means SE.

Table A7.13 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain P, K and S content for each site

	P			K			S		
	%			%			%		
	COU	**GIL**	**COU**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**COU**
Year									
2006	0.35±0.003	0.33±0.003	0.12±0.002	0.12±0.002	0.12±0.002	0.44±0.002	0.12±0.002	0.15±0.001	0.14±0.001
2007	0.42±0.002	0.39±0.003	0.14±0.001	0.14±0.001	0.14±0.001	0.5±0.005	0.14±0.001	0.15±0.002	0.15±0.002
Fertility rate			0±0	0±0	0±0		0±0	0±0	0±0
125 kg N ha^{-1}	0.39±0.006	0.36±0.006	0.13±0.002	0.13±0.002	0.13±0.002	0.47±0.007	0.13±0.002	0.15±0.001	0.15±0.002
250 kg N ha^{-1}	0.39±0.007	0.36±0.007	0.13±0.002	0.13±0.002	0.13±0.002	0.47±0.007	0.13±0.002	0.15±0.002	0.15±0.002
Fertility type									
CP	0.39±0.011	0.35±0.008**b**	0.14±0.003**a**	0.14±0.003**a**	0.14±0.003**a**	0.49±0.013**a**	0.14±0.003**a**	0.15±0.001**a**	0.16±0.003**a**
FYM	0.39±0.009	0.36±0.01a**b**	0.12±0.003**c**	0.12±0.003**c**	0.12±0.003**c**	0.46±0.006**b**	0.12±0.003**c**	0.14±0.002**b**	0.14±0.002**c**
FYM+CP	0.39±0.01	0.36±0.01a**b**	0.13±0.003**b**	0.13±0.003**b**	0.13±0.003**b**	0.47±0.01**b**	0.13±0.003**b**	0.15±0.002**a**	0.15±0.001**b**
GWC	0.39±0.01	0.36±0.009**a**	0.13±0.003**c**	0.13±0.003**c**	0.13±0.003**c**	0.46±0.006**b**	0.13±0.003**c**	0.15±0.002**ab**	0.14±0.001**c**
ANOVA									
Main Effects									
YR	**0.0007**	**0.0008**	**0.0053**	**0.0053**	**0.0053**	**0.0011**	**0.0053**	ns	**0.0034**
FR	ns	ns	0.0129	0.0129	0.0129	ns	0.0129	ns	ns
FT	ns	**0.0871**	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**0.0009**	**<.0001**
Interactions									
YR:FR	**0.0598**	ns	ns	ns	ns	ns	ns	ns	ns
YR:FT	ns	ns	ns	ns	ns	**<.0001**	ns	**0.0001**	**0.0002**
FR:FT	0.0748	ns	**0.001**	**0.001**	**0.001**	ns	**0.001**	ns	ns
YR:FR:FT	ns	ns	ns	ns	ns	ns	ns	**0.0015**	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$) COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Effect of fertiliser input type and rate on grain micro nutrient concentrations

Table A7.14 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain Ca, Fe and Zn content

	Grain Ca	Fe	Zn
	%	mg kg^{-1}	mg kg^{-1}
Year			
2006	0.04±0.001	34.8±0.41	29.9±0.59
2007	0.05±0.001	31.6±0.46	34.9±0.53
Site			
Courtyard	0.05±0.001**a**	29.6±0.48**c**	27.7±0.63**c**
Gilchesters	0.04±0.001**b**	37±0.39**a**	30.6±0.31**b**
Sheepdrove	0.05±0.001**a**	33.6±0.37**b**	38.4±0.42**a**
Fertility Rate			
125 kg N ha^{-1}	0.05±0.001	33.3±0.48	32.1±0.61
250 kg N ha^{-1}	0.04±0.001	33.3±0.45	32.5±0.64
Fertility Type			
CP	0.05±0.002**a**	35.1±0.56**a**	33±0.96**a**
FM	0.04±0.001**b**	31.6±0.59**c**	32±0.82**bc**
FM+CP	0.05±0.002**a**	34±0.67**b**	32.6±0.89**ab**
GWC	0.04±0.001**b**	32.3±0.69**c**	31.5±0.84**c**
ANOVA			
Main Effects	p45	p51	p49
Year (YR)	**0.0014**	**0.0529**	**0.0026**
Site (ST)	**<.0001**	**<.0001**	**<.0001**
Fertility rate (FR)	ns	ns	ns
Fertility type (FT)	**<.0001**	**<.0001**	**0.0023**
Interactions			
YR x ST	**0.0004**	ns	**0.0002**
YR x FR	ns	ns	ns
ST x FR	ns	0.0708	ns
YR x FT	0.0559	0.041	0.0243
ST x FT	ns	**<.0001**	0.0306
FR x FT	ns	ns	ns
YR x ST x FR	**0.0792**	ns	0.0957
YR x ST x FT	**0.0872**	0.0015	0.0709
YR x FR x FT	ns	0.0843	ns
ST x FR x FT	ns	0.0056	ns
YR x ST x FR x FT	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A7.15 Interaction means for effects of fertiliser rate and type on wheat grain S content grown at courtyard farm

	Grain S %			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	0.13±0.004**Ba**	0.12±0.005**Ab**	0.13±0.004**Aab**	0.12±0.004**Ab**
250 kg N ha^{-1}	0.14±0.004**Aa**	0.12±0.004**Ad**	0.13±0.004**Ab**	0.13±0.003**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Table A7.16 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain Ca, Fe and Zn content for each site

	Ca			**Fe**			**Zn**		
	%			mg kg^{-1}			mg kg^{-1}		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year									
2006	0.04±0.001	0.03±0.002	0.04±0.001	31.6±0.73	37.8±0.35	35.1±0.5	23.3±0.45	30.3±0.45	36.1±0.39
2007	0.05±0.001	0.04±0.001	0.06±0.001	27.7±0.39	36.1±0.75	32.2±0.42	32.1±0.38	31±0.41	40.7±0.47
Fertility rate	0±0	0±0	0±0						
125 kg N ha^{-1}	0.05±0.001	0.04±0.002	0.05±0.002	29.1±0.67	37.1±0.52	34±0.59	27.4±0.8	30.7±0.5	38.2±0.63
250 kg N ha^{-1}	0.05±0.001	0.04±0.001	0.05±0.002	30.1±0.69	37±0.6	33.3±0.47	28±0.98	30.5±0.36	38.6±0.55
Fertility type									
CP	0.05±0.002**c**	0.04±0.001	0.06±0.004**a**	32±1.07**a**	36.7±0.52b	36.8±0.62**a**	28.3±1.41**a**	30.7±0.33	39.9±1.09**a**
FM	0.05±0.002b**c**	0.03±0.001	0.05±0.002**b**	27.9±0.65**c**	35.8±0.75b	31.9±0.39**c**	28.1±1.15**a**	29.4±0.36	38±0.61**b**
FM+CP	0.05±0.002b**c**	0.04±0.003	0.05±0.003**b**	30.2±0.87**b**	38.5±0.84a	33.6±0.66**b**	27.8±1.41**ab**	31.7±0.88	38.3±0.9**b**
GWC	0.05±0.002a**b**	0.03±0.002	0.05±0.002**b**	28.5±0.92**c**	37±0.9ab	32.3±0.64**c**	26.6±1.1**b**	30.4±0.61	37.4±0.55**b**
ANOVA									
Main Effects	p45								
YR	**0.0084**	**0.041**	**0.0008**	ns	ns	**0.0103**	**0.0059**	ns	**0.0046**
FR	ns	ns	**0.0971**	**0.0497**	ns	ns	ns	ns	ns
FT	**0.0127**	ns	**0.0001**	**<.0001**	**0.0041**	**<.0001**	**0.0137**	0.0823	**0.009**
Interactions									
YR:FR	**0.0853**	ns	ns	ns	ns	ns	0.0599	ns	ns
YR:FT	ns	ns	**0.0009**	**0.0075**	**0.0082**	ns	**0.0602**	ns	**0.0087**
FR:FT	ns	ns	ns	**0.0127**	**0.0234**	ns	ns	ns	ns
YR:FR:FT	ns	ns	ns	ns	**0.0261**	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$) [1] see table 7.17 for interaction means SE
COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A7.17 Interaction means for effects of fertiliser rate and type on wheat grain Fe content grown at courtyard farm

	Grain Fe mg kg^{-1}			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	30.5±1.73**Ba**	28.2±0.92**Aab**	29.3±1.34**Aab**	28.3±1.32**Ab**
250 kg N ha^{-1}	33.4±1.14**Aa**	27.4±0.97**Ac**	31.0±1.10**Ab**	28.5±1.37**Ac**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Table A7.18 Interaction means for effects of fertiliser type and fertiliser rate on wheat grain Fe content grown at Gilchesters farm

	Grain Fe mg kg^{-1}			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	36.8±0.71**Aab**	36.6±1.22**Ab**	38.8±0.93**Aab**	35.7±1.18**Aa**
250 kg N ha^{-1}	36.6±0.83**Aab**	35.0±0.9**Ab**	38.1±1.53**Aa**	38.3±1.25**Aa**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$)

Effect of fertiliser input type and rate on grain phytic acid

Table A7.19 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain phytic acid content

	Phytic acid	**Phytic acid P**	**Phytic acid Zn**	**Phytic acid Fe**
	mg g^{-1}	%	moler ratio	moler ratio
Year				
2006	9.9±0.07	78.8±0.78	34.4±0.82	24.5±0.32
2007	9.9±0.11	67.0±0.8	28.7±0.55	27.0±0.47
Site				
Courtyard	10.0±0.12	73.2±1.35**b**	37.1±1.06**a**	28.8±0.51**a**
Gilchesters	9.9±0.11	78.7±1.03**a**	32.2±0.46**b**	22.7±0.33**c**
Sheepdrove	9.9±0.1	68.4±0.89**c**	25.8±0.4**c**	25.1±0.32**b**
Fertility Rate				
125 kg N ha^{-1}	9.9±0.09	72.4±0.98	31.6±0.73	25.6±0.44
250 kg N ha^{-1}	10.0±0.08	74.0±1.02	31.8±0.81	25.8±0.40
Fertility Type				
CP	10.0±0.12	73.5±1.44	31.2±1.11	24.3±0.48**b**
FYM	9.8±0.10	71.8±1.17	31.4±0.93	26.7±0.56**a**
FYM+CP	9.9±0.16	73.4±1.67	31.4±1.20	25.0±0.6**b**
GWC	10.0±0.11	74.0±1.35	32.7±1.09	26.8±0.65**a**
ANOVA				
Main Effects				
Year (YR)	ns	**0.0024**	**0.0115**	**0.0482**
Site (ST)	ns	**0.0001**	**<.0001**	**<.0001**
Fertility rate (FR)	ns	**0.0956**	ns	ns
Fertility type (FT)	ns	ns	ns	**<.0001**
Interactions				
YR x ST	ns	**ns**	**0.0006**	ns
YR x FR	ns	ns	ns	ns
ST x FR	**0.0988**	ns	ns	ns
YR x FT	ns	ns	ns	ns
ST x FT	ns	ns	ns	ns
FR x FT	ns	ns	**0.0578**	**0.0736**
YR x ST x FR	ns	ns	**0.0857**	ns
YR x ST x FT	ns	ns	**0.0359**	**0.0163**
YR x FR x FT	**0.0443**	0.0403	**0.0265**	**0.0171**
ST x FR x FT	ns	ns	ns	ns
YR x ST x FR x FT	ns	ns	ns	ns

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, $p < 0.05$).

Table A7.20 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain phytic acid content for each site

COURTYARD	Phytic acid mg g^{-1}			Phytic acid P %			Phytic acid Zn molar ratio			Phytic acid Fe molar ratio		
	COU	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
Year												
2006	10.1±0.13	9.8±0.13	10.0±0.13	80.7±1.32	83.1±0.98	72.6±0.99	43.5±1.14	32.1±0.63	27.5±0.43	27.3±0.50	21.9±0.30	24.2±0.38
2007	9.9±0.20	10.1±0.18	9.8±0.16	65.6±1.41	72.7±1.21	64.2±1.04	30.7±0.78	32.4±0.70	24.0±0.52	30.4±0.81	23.9±0.60	25.9±0.48
Fertility rate												
125 kg N ha^{-1}	9.8±0.20	9.7±0.15	10.0±0.13	71.7±1.94	77.5±1.53	68.4±1.16	36.8±1.45	31.7±0.70	26.3±0.58	28.9±0.82	22.3±0.41	25.1±0.4
250 kg N ha^{-1}	10.1±0.12	10.1±0.16	9.8±0.16	74.6±1.88	79.8±1.35	68.4±1.36	37.4±1.56	32.8±0.58	25.3±0.55	28.8±0.62	23.2±0.53	25.0±0.51
Fertility type												
CP	10.1±0.21	9.7±0.2	10.1±0.22	74.5±3.00	77.4±1.78	68.8±2.10	36.9±2.33	31.3±0.72	25.5±1.00	27±0.81**b**	22.3±0.52	23.4±0.62**b**
FYM	9.8±0.16	9.9±0.19	9.8±0.18	71.2±2.02	78.0±1.65	67.4±1.38	35.6±1.66	33.3±0.78	25.6±0.62	30.0±0.73**a**	23.5±0.80	25.9±0.59**a**
FYM+CP	9.7±0.36	10±0.24	10.0±0.24	71.5±3.61	79.2±2.08	69.7±2.27	36.6±2.70	31.5±1.10	26.1±0.97	27.5±1.12**b**	22.1±0.79	25.3±0.70**a**
GWC	10.2±0.15	10.1±0.23	9.7±0.19	75.5±1.90	80.1±2.78	67.6±1.21	39.2±1.69	33.1±1.00	25.9±0.60	30.9±1.09**a**	23.2±0.47	25.6±0.50**a**
ANOVA												
Main Effects												
YR	ns	ns	ns	**0.008**	**0.0101**	**0.0101**	ns	ns	**0.0102**	**0.0957**	ns	**0.065**
FR	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
FT	ns	ns	ns	ns	ns	ns	ns	ns	ns	**0.0008**	ns	**0.0172**
Interactions												
YR:FR	ns	**0.0562**	ns	ns	ns	ns	ns	ns	ns	0.794	ns	ns
YR:FT	ns	**0.0635**	ns	ns	ns	ns	ns	ns	**0.0988**	**0.0208**	**0.0754**	ns
FR:FT	ns	ns	ns	**0.069**	ns	ns	ns	ns	ns	ns	ns	ns
YR:FR:FT	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	0.0677

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Effect of fertiliser input type and rate on grain heavy metal concentrations

Table A7.21 Effects means ± SE and *p* values of harvest year, site, fertiliser rate and type on wheat grain heavy metal content

	Mo	**Al**	**Cd**	**Ni**	**Pb**
	mg kg^{-1}	mg kg^{-1}	µg kg^{-1}	µg kg^{-1}	µg kg^{-1}
Year					
2006	0.6±0.04	2.2±0.13	29.9±1.27	88.9±3.37	41.0±3.23
2007	0.8±0.04	2.3±0.10	37.7±1.30	121.0±2.95	114.7±5.43
Site					
Courtyard	0.9±0.03**a**	2.2±0.10**b**	33.8±2.06**ab**	97.6±3.22**b**	77.1±7.37
Gilchesters	0.2±0.03**b**	2.7±0.22**a**	36.6±1.56**a**	121.6±5.31**a**	72.2±7.18
Sheepdrove	0.9±0.03**a**	1.9±0.06**b**	30.9±1.12**b**	95.9±4.00**b**	78.9±6.88
Fertility Rate					
125 kg N ha^{-1}	0.7±0.04	2.2±0.10	33.7±1.31	105.3±4.07	72.1±5.64
250 kg N ha^{-1}	0.7±0.04	2.2±0.13	33.6±1.39	103.3±3.04	80.5±5.99
Fertility Type					
CP	0.5±0.04**d**	2.5±0.20**a**	36.6±1.94**a**	109.5±5.13	76.8±7.76**b**
FM	0.9±0.06**a**	2.0±0.07**b**	31.8±1.85**b**	100.4±4.12	71.5±7.66**b**
FM+CP	0.6±0.05**c**	2.3±0.18**b**	33.7±1.94**b**	108.2±6.12	87.9±9.55**a**
GWC	0.8±0.06**b**	2.1±0.16**b**	32.2±1.86**b**	98.7±4.60	68.2±7.64**b**
ANOVA					
Main Effects					
Year (YR)	**0.015**	ns	**0.0134**	**0.0068**	**0.0012**
Site (ST)	**<.0001**	**0.0512**	**0.0224**	**0.0008**	ns
Fertility rate (FR)	ns	ns	ns	ns	ns
Fertility type (FT)	**<.0001**	ns	**0.0056**	ns	ns
Interactions					
YR x ST	**0.0013**	ns	**<.0001**	ns	ns
YR x FR	ns	ns	ns	ns	ns
ST x FR	**0.0245**	ns	ns	**0.0523**	ns
YR x FT	**0.0002**	ns	ns	ns	**0.003**
ST x FT	**<.0001**	ns	**0.0229**	ns	**0.0852**
FR x FT	**0.0008**[1]	ns	ns	ns	**0.0355**[2]
YR x ST x FR	ns	ns	ns	ns	ns
YR x ST x FT	**<.0001**	ns	ns	ns	ns
YR x FR x FT	ns	ns	ns	ns	ns
ST x FR x FT	**0.0002**	ns	ns	ns	ns
YR x ST x FR x FT	ns	ns	ns	ns	**0.032**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$) [1] See Table 7.23 for interaction means SE. [2] See Table 7.24 for interaction means SE.

Table A7.22 Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain heavy metal content for each site

	Mo			**Al**			**Cd**		
	mg kg^{-1}			mg kg^{-1}			μg kg^{-1}		
Year	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	0.8±0.04	0.1±0.01	0.9±0.04	2.5±0.17	2.4±0.32	1.7±0.06	19.3±1.07	44.5±1.16	25.8±1.18
2007	1.0±0.04	0.4±0.04	0.8±0.04	1.9±0.09	3.0±0.27	2.0±0.09	48.2±1.61	26.2±1.67	35.9±1.42
Fertility rate									
125 kg N ha^{-1}	1.0±0.04	0.2±0.03	0.8±0.04	2.2±0.17	2.7±0.21	1.8±0.08	33.0±2.81	36.7±2.18	31.6±1.58
250 kg N ha^{-1}	0.9±0.06	0.2±0.04	0.9±0.04	2.2±0.11	2.7±0.4	1.9±0.08	34.5±3.04	36.5±2.26	30.2±1.59
Fertility type									
CP	0.7±0.05	0.2±0.02**b**	0.6±0.03**b**	2.4±0.3	3.1±0.49	2.0±0.13	39.9±4.63	36.7±2.84**b**	33.3±1.96
FYM	1.2±0.05	0.3±0.09**a**	1.0±0.05**a**	2.0±0.14	2.2±0.11	1.8±0.11	31.7±3.97	32.8±3.74**c**	31.1±1.84
FYM+CP	0.9±0.05	0.2±0.03**b**	0.7±0.02**b**	2.3±0.18	2.9±0.5	1.8±0.1	33.9±4.22	37.1±2.90**b**	30.3±2.66
GWC	1.0±0.07	0.2±0.05**b**	1.0±0.03**a**	2.0±0.15	2.5±0.49	1.9±0.11	29.7±3.47	39.7±3.11**a**	28.7±2.43
ANOVA									
Main Effects									
YR	**0.0616**	**0.0082**	ns	ns	ns	**0.045**	**0.0005**	**0.0151**	**0.0144**
FR	**0.0215**	ns	ns	ns	ns	ns	ns	ns	ns
FT	**<.0001**	**<.0001**	**<.0001**	ns	ns	ns	**0.0008**	**0.0245**	ns
Interactions		ns			ns			ns	
YR:FR	ns	**0.0879**	ns	ns	ns	ns	ns	ns	ns
YR:FT	**0.0001**	**<.0001**	ns	ns	ns	ns	ns	ns	ns
FR:FT	**0.0001**	**0.0232**	ns	ns	ns	ns	ns	ns	ns
YR:FR:FT	ns	**0.0095**	ns	ns	ns	ns	**0.0716**	ns	ns

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, $p < 0.05$). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A7.22 cont. Effects means ± SE and *p* values of harvest year, fertiliser rate and type on wheat grain heavy metal content for each site

	Ni			Pb		
	μg kg^{-1}			μg kg^{-1}		
Year	**COU**	**GIL**	**SHE**	**COU**	**GIL**	**SHE**
2006	84.5±3.75	105.6±7.55	76.7±4.33	35.7±4.23	39.2±4.92	48±7.13
2007	110.7±4.12	142.8±4.54	115±4.77	118.6±9.59	116.1±9.87	109.7±8.94
Fertility rate						
125 kg N ha^{-1}	94±4.65	130.8±8.5	93.5±5.88	68.9±9.99	71.9±10.37	75.3±9.29
250 kg N ha^{-1}	101.2±4.44	111.6±5.74	98.3±5.50	85.3±10.79	72.4±10.1	82.4±10.25
Fertility type						
CP	101.7±8.27	127.5±7.24	100.3±9.61	70±11.99	81.7±14.38	78.9±14.61
FYM	91.5±6.05	114.4±7.35	98.1±7.22	70.5±12.13	72.3±14.45	71.8±14.2
FYM+CP	103.4±5.03	129.2±15.27	93.4±8.02	102.7±19.33	83.7±14.7	77.1±15.37
GWC	93.8±6.10	113.1±9.95	91.8±7.57	65.3±13.71	47.8±13.02	87.7±11.61
ANOVA						
Main Effects						
YR	**0.0195**	**0.0286**	**0.0318**	**0.0026**	**0.0035**	**0.0159**
FR	ns	ns	ns	ns	ns	ns
FT	ns	ns	ns	**0.0196**	ns	ns
Interactions		ns			ns	
YR:FR	ns	ns	ns	ns	ns	ns
YR:FT	ns	ns	ns	**0.0062**	ns	ns
FR:FT	ns	ns	ns	ns	**0.0157**[b]	ns
YR:FR:FT	ns	ns	ns	ns	ns	**0.0216**

Means labelled with the same letter within the same column are not significantly different (Tukey's honestly significant difference test, *p* < 0.05). COU: Courtyard; GIL: Gilchesters; SHE: Sheepdrove

Table A7.23 Interaction means for effects of fertility type and fertility rate on wheat grain Mo content

	Grain Mo μg kg^{-1}			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	0.8±0.06**Ac**	1.1±0.07**Aa**	0.9±0.05**Ab**	1.0±0.09**Aab**
250 kg N ha^{-1}	0.5±0.02**Bd**	1.2±0.06**Aa**	0.7±0.06**Bc**	0.9±0.09**Ab**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, p < 0.05)

Table A7.24 Interaction means for effects of fertiliser type and fertiliser rate on wheat grain Mo content

	Grain Mo μg kg^{-1}			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	0.5±0.06**Ab**	0.8±0.09**Aa**	0.6±0.07A**b**	0.7±0.08**Aa**
250 kg N ha^{-1}	0.4±0.04**Bd**	0.8±0.09**Aa**	0.5±0.06**Ac**	0.7±0.09**Ab**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, p < 0.05)

Table A7.25 Interaction means for effects of fertiliser type and fertiliser rate on wheat grain Pb content

	Grain Pb μg kg^{-1}			
	CP	**FM**	**FM+CP**	**GWC**
125 kg N ha^{-1}	75.4±9.86**Aab**	64.9±11.81**Ab**	94.9±12.23**Aa**	51.4±9.77**Ab**
250 kg N ha^{-1}	78.1±12.30**Aa**	77.6±9.94**Aa**	80.6±14.90**Aa**	85.7±10.82**Ba**

Means labelled with the same letter within the same column are not significant different (Tukey's honestly significant difference test, p < 0.05)

APPENDIX 8: Chapter 6 Wheat classification tables

Table 8.1 UK wheat classification

Grade	Description
Group 1 (hard)	Premium bread
Group 1 (hard)	Bread
Group 2 (hard)	Milling
Group 3 (soft)	Biscuit
Group 4 (hard/soft)	Feed

Table 8.2 Austrian wheat classification

Grade	Description
Class BQ 7-9	Top Baking Quality
Class BQ 4-6	Baking Quality blends
Class 3	Biscuit
Class 2	Hard feed
Class 1	Soft feed

Table 8.3 Austrian scoring system for wheat grades

Score (1-9)	maturity	plant height	septoria	fusarium	grains/ear	TGW	HFN	Protein	Zeleny Sediment	loaf volume
1. v. low						1	1	1	1	1
2. v. low - low						2	2	2	2	2
3. low						3	3	3	3	3
4. low to mid		BQ 4-6				BQ 4-6				BQ 4-6
5. mid							BQ 4-6		BQ 4-6	
6. mid to high								BQ 4-6		
7. high		BQ7-9					BQ7-9		BQ7-9	BQ7-9
8. high to v. high								BQ7-9		
9. v. high										

Table 8.4 German wheat classification

Grade	Description
E	Elite wheat
A	Quality Wheat
B	Bread wheat
C(k)	Biscuit wheat
C	Feed wheat

Table 8.5 German scoring system for wheat grades

Score	Bread volume	HFN	Zeleny sedimentation	Protein	Protein concentration	Flour yield
1. v. low						
2. v. low - low				C	C	
3. low			C		A	
4. low to mid	C	C		A	E	C
5. mid		A	A			A,E
6. mid to high	A	E		E		
7. high			E			
8. high to v. high	E					
9. v. high						