Grain quality and N uptake of spring cereals as affected by nitrogen fertilization under Nordic conditions: a meta-analysis

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We reviewed quantitatively 40 Finnish field experiments related to the effect of nitrogen (N) fertilizer on the main parameters of grain quality and N uptake of spring cereals. The experiments were conducted on a wide range of mineral soils under varying growth conditions from the 1950s to the 1990s. Overall there was no statistically significant effect on 1000 grain weight and a slightly negative effect on grain test weight. Nitrogen fertilizer increased N uptake much more steeply in slightly acidic soils (SA, pH 5.8–6.9), located mostly in South Finland, than in moderately acidic soils (MA, pH 5.0–5.7), located in Central Finland. With increasing N rates, protein content increased to a larger extent in spring barley and oats than in spring wheat. In the light of the current trend to reduce N fertilizer application, the obtained regressions between N rates and the parameters of grain quality may be used to maintain yield quality at a desirable level, while optimizing N management.

Key words: nitrogen fertilization, protein content, 1000 grain weight, test weight, meta-analysis

Introduction

Nitrogen (N) fertilization has a decisive influence on the yield of arable crops. Moreover, protein content, 1000 grain weight and grain test weight (i.e. hectoliter weight) are important production parameters that affect the profitability of cereal cultivation. Thousand grain weight is used by breeders and flour millers to indicate kernel composition and potential flour extraction. Protein content is important for bread-making quality since it is related to many processing properties, such as water absorption and gluten strength.

Nitrogen input in soil has been decreasing due to increasing fertilizer prices, but also due to legislation and goals set in many countries to reduce nutrient emissions from agriculture (OECD 2001). The latter is closely linked to the need for agriculture to comply with national standards for nitrate emissions into aquatic environments. A number of international conventions and agreements, such as The European Union's Nitrate Directive (EEC 1991), The European Water Framework Directive (EEC 2000) and Framework Convention on Climate Change (UN 1992) also have the objective of limiting and reducing nutrient emissions from agriculture into surface and ground water, marine waters and the atmosphere.

In Finland, the national average N input to cultivated fields decreased from 160 kg ha⁻¹ at the beginning of the 1990s to 120 kg ha⁻¹ in 2005, due to a decline in the use of commercial N fertilizers from 114 to 74 kg ha⁻¹ (Salo et al. 2007a). According to current Finnish Agri-Environmental Programme (FAEP, 2007–2013) the maximum allowed N rate (N_{max}) for spring barley cultivated on clay and course-textured mineral soils in South and Central Finland is set to 90–100 kg ha⁻¹ for a yield expectation of 4000 kg ha⁻¹, while for spring wheat, the respective N_{max} is 110–120 kg ha⁻¹ (Ministry of Agriculture and Forestry 2011). The nitrate directive restricts N application to 170 kg ha⁻¹ for spring cereals cultivated throughout Finland (FINLEX 2000).

There have been concerns as to whether the current N_{max} is sufficient to produce high yields with good quality and the best economic result for growers. Reduced use of N fertilizer is likely to decrease both production costs and N leaching, while it may also result in reduced crop yields and quality if crops experience temporary N deficiency. Peltonen (1992) and Jensen and Schjoerring (2011) summarized the effects of N fertilizer on N uptake and crop quality in narrative reviews. In the present study, using meta-analytical techniques, we reviewed quantitatively 40 Finnish field experiments related to the effects of N fertilizer on the main parameters of yield quality (1000 grain weight, test weight, protein content) and grain N uptake, and we examined the sources of variation in the responses. We also evaluated whether N_{max} or economically optimal N rates (N_{opt}), which were justified in our previous study (Valkama et al. 2013), are sufficient to provide high quality yields.

Materials and methods The database

The database consisted of published and unpublished reports of experiments conducted at MTT Agrifood Research Finland (Jokioinen, Finland), at its Research Stations, at other experimental farms and on growers' fields in Finland (Appendix). The reports were retrieved from the library of MTT. Seven relevant published journal articles were found by searching the reference lists of previously published articles in relevant Finnish journals (Acta Agralia Fennica, Agricultural and Food Science, Agricultural and Food Science in Finland, Annales Ariculturae Fenniae, Journal of the Scientific Agricultural Society Finland, Kehittyvä Maatalous, and Maatalous ja Koetoiminta). Articles were also searched for by using key-words in the Web of Science Database ("nitrogen fertilizer or fertilization" AND cereal* or quality, or 1000 grain weight, or test weight, or protein content, or N uptake AND Finland).

In order to be included in the database, a study had to meet the following criteria:

- 1. The study had been carried out in Finland after 1950.
- 2. The N fertilizer source was ammonium nitrate (34% N), calcium ammonium nitrate (26% N) or calcium nitrate (16% N).
- 3. The experiments were conducted in the field.
- 4. The study had an appropriate control, i.e. fertilization with phosphorus (P) and potassium (K), but no N.
- 5. The effects of N fertilization on N uptake, protein content, 1000 grain weight or test weight of spring wheat (*Triticum* spp. L.), spring barley (*Hordeum vulgare* L.) and oats (*Avena sativa* L.) were assessed.
- 6. Responses to N fertilization were given in terms of either original data or means of the treatments (i.e. fertilized with PKN, \overline{X}_{PKN}) and controls (fertilized with PK, \overline{X}_{PK}), with standard deviations (S_{PKN} , S_{PK}) and sample sizes (n_{PKN} , n_{PK}).

An "experiment" was defined as a continuous sequence of consecutive years in a given field with fixed levels of annual N applications; the duration of the experiments that fulfilled these criteria varied between one and nine years, with a mean of three years. The spring-sown cereals included are the most important cereal crops in Finn-ish agriculture and are also widely cultivated in other northern countries such as Sweden, Norway, Denmark and Canada. The final database consisted of 24 experiments with barley, 12 experiments with wheat and 4 experiments with oats (Appendix). All 40 experiments were conducted between 1953 and 1999 at 17 sites. The soils were clay and coarse-textured mineral soils with a pH range of 5.0–6.9 and with soil organic matter (SOM) range of 2.9–8.5%. The annual N application rates ranged from 16 to 216 kg ha⁻¹ with a mean of 100 kg ha⁻¹. Since 1960s, N fertilizers were applied by placement technique. Phosphorus and K were supplied according to existing recommendations, with P rates ranging from 14 to 60 kg ha⁻¹ (mean 38 kg ha⁻¹), and K rates from 35 to 104 kg ha⁻¹ (mean 74 kg ha⁻¹).

Response and explanatory variables

Response variables included thousand grain weight (g), test weight (kg hl⁻¹), protein content of grain (%) and N uptake (kg ha⁻¹).

N uptake was calculated as uptake in harvested grain:

$$N \, uptake \, (kg \, ha^{-1}) = Protein \, (\%)/a \times Yield \, (kg \, ha^{-1}) \times 0.85/100 \, (\%)$$
 [1]

where a is coefficient equal to 6.25 for barley and oats, and 5.7 for wheat. The 0.85 coefficient was used to convert the grain yields from 15% moisture content to 100% dry matter content.

To explain the variation in the responses due to N fertilization, the categorical and continuous explanatory variables listed in Table 1 were included. The average yield without N fertilizer was for the whole database 2400 ± 800 kg ha⁻¹ The experiments were divided according to the yield level without added N into three groups, as in the previous study (Valkama et al. 2013). Soils were divided on the basis of their clay content. Cultivation zones were derived from the length of the average growth period.

(a) Categorical explanatory variables	Groups
Yield level without added N (kg ha-1)	low (1000–2000), medium (2000–3000), high (3000–4000)
Soil texture	clay (>30% clay), coarse-textured, i.e. loam, silt and sand (<30% clay
Soil pH (H ₂ O)	moderately acidic (5.0–5.7), slightly acidic (5.8–6.9)
Cultivation zones (average growth period, days)	I (175), II (165), III (155), IV (145)
Species of spring cereals	barley, wheat, oats
Decades	1950s, 1960s, 1970s, 1980s, 1990s
(b) Continuous explanatory variables	Range
Duration of experiment (years)	1–9
Soil organic matter (%)	2.9–8.5
N rates (kg ha ⁻¹)	16–216
P rates (kg ha ⁻¹)	14-60
K rates (kg ha-1)	35–104

Table 1. Categorical (a) and continuous (b) explanatory variables included in the meta-analysis.

Each observation in a meta-analysis is required to be independent. In the majority of the present experiments, several varieties were cultivated either simultaneously or in rotation during the experiments (Appendix). Thus, to avoid the problem of lack of independence, data for different varieties in the same study were pooled. Never-theless, the variation between the old and modern varieties in terms of response to N fertilizer was studied indirectly by means of grouping the dataset into the decades when experiments were conducted and by comparison of the responses between the decades. Table 2 shows the list of varieties of spring cereal species used for N fertilizer experiments in different decades.

Table 2. Varieties of spring cereal species used for N fertilizer experiments in different decades and the number of experiments pe	r
decade.	

Species	Decade	Variety (Year of release)	Number of experiments
Barley	50s	Balder (1945)	1
	60s	Pirkka (1952), Ingrid (1956), Otra (1959), Mari (1960), Arvo (1966), Karri (1967)	3
	70s	Olli (1927), Pirkka (1952), Ingrid (1956), Otra (1959), Vigdis (1964), Karri (1967), Pomo (1968), Etu (1970), Eero (1975)	8
	80s	Pirkka (1952), Ingrid (1956), Etu (1970), Aramir (1972), <i>Welam</i> (1976), <i>Harry</i> (1978), Ida (1979), Patty (1980), <i>Hankkijan Pokko</i> (1980), <i>Kustaa</i> (1981), <i>Kilta</i> (1982), <i>Arra</i> (1982), <i>Kymppi</i> (1985)	11
	90s	Loviisa (1989)	1
Wheat	50s	Timantti (1928), Timantti II (1937), Tammi (1939), Kärni (1948)	2
	60s	Apu (1949), Touko (1950), Norröna (1952), Svenno (1953)	5
	70s	Ruso (1967), Veka (1971)	1
	80s	Kadett (1981), <i>Heta</i> (1988)	2
	90s	Runar (1972), Reno (1975), Kadett (1981), Luja (1981) <i>, Heta</i> (1988), <i>Polkka</i> (1992), Laari (1990), <i>Satu</i> (1989)	2
Oats	50s	Sisu (1948)	1
	60s	Sisu (1948)	1
	90s	Puhti (1978), Veli (1981), Virma (1988), Yty (1989), Salo (1989), Leila (1991), Aarre (1994), Kolbu (1994), Katri (1995), Roope (1996)	2

Varieties which are currently cultivated in Finland are indicated in italics.

Linear regressions

The effects of SOM content on the parameters of grain quality and N uptake in control treatments (PK) were tested by linear regressions. The data were pooled for all cereal species and tested for normality and homoscedasticity (equal variance).

Meta-analysis

The effects of N fertilization on the grain quality and N uptake were analyzed using meta-analysis, which is the statistical analysis of a large collection of independent studies for the purpose of integrating their findings. The meta-analysis was carried out using the Meta Win 2.0 statistical program (see Rosenberg et al. 2000). Quantita-tive meta-analysis involves the calculation of an effect size (i.e., the magnitude of the treatment effect) that can be averaged across independent studies, giving an overall mean effect size.

For response variables, a separate estimate of response ratio (*r*) was calculated as an index of effect size for each site, cereal species and N rate as follows:

$$r = \overline{X}_{PKN} / \overline{X}_{PK}$$
^[2]

where \overline{X}_{PKN} and \overline{X}_{PK} represent the mean value for PKN-fertilized treatments and PK-fertilized treatment (i.e. control), respectively, averaged over the duration of an experiment.

However, it is desirable to perform statistical analyses in the metric of the natural logarithm of *r* since it has preferable statistical properties (Hedges et al. 1999):

$$\ln r = \ln \left(\overline{X_{PKN}} / \overline{X_{PK}} \right)$$
^[3]

To study the effect of increasing N rates, averaged 1000 grain weight, test weight, protein content and N uptake over the duration of experiment for each N rate in a study served as the value of the dependent variable. However, when studying the effects of other explanatory variables on the responses of 1000 grain weight and test weight, the data were averaged for all N rates used in each experiment to avoid lack of statistical independence. When studying the effects of explanatory variables on the responses of protein and N uptake, the experiments including 100–150 kg N ha⁻¹ were chosen to eliminate the effect of N rates, since the responses are known to be highly affected by increasing N rates (Gauer et al. 1992, Baker et al. 2004, Brennan and Bolland 2009, Mooleki et al. 2010, Kienzler et al. 2011). Thus, the number of experiments was reduced from 27 to 22 for the protein responses, and from 22 to 18 for the N uptake responses.

Details on meta-analysis of response ratio and calculations of the weighted means of $\ln r$ were described in our previous study (Valkama et al. 2013). To test whether $\ln r$ differed among the groups of categorical explanatory variables, the between-group heterogeneity ($Q_{\rm B}$) was examined using χ^2 test. Possible inter-correlation between the variables was also tested by using χ^2 test. To study the effect of continuous explanatory variables, weighted meta-regressions were run with $\ln r$ as the dependent variable and the continuous variables as independent ones. Model heterogeneity ($Q_{\rm M}$), which describes the amount of heterogeneity explained by the regression models, was examined using χ^2 test. To detect selection bias, the rank correlation (Kendall's tau) was calculated between standardized $\ln r$ and sample size (n) across the studies (Rosenberg et al. 2000).

Except for meta-regressions, log response ratios were back-transformed, and reported in the text and figures as the percentage changes from the control:

Response (%) =
$$[EXP(\ln r) -1] \times 100\%$$
 [4]

Responses to N fertilizer were considered to be significantly different from the control if their 95% confidence intervals (CI) did not overlap with zero.

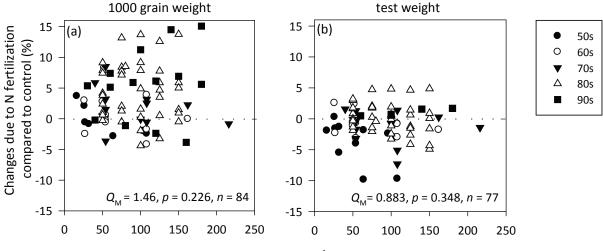
Results

Thousand grain and test weights

In the control treatments (PK), the average 1000 grain weight was 37.5 ± 5.3 g (Mean \pm SD) for barley (n = 18), 31.8 ± 4.1 g for wheat (n = 8), and 35.4 ± 5.4 g for oats (n = 3), whereas test weight was 60.5 ± 6.3 kg hl⁻¹ for barley (n = 18) and 77.7 ± 4.5 kg hl⁻¹ for wheat (n = 8). The test weight of oats was only measured in one experiment (Exp 2). Neither parameter had any relationship to SOM content when the cereal species were pooled together (1000 grain weight: $R^2 = 0.04$, p = 0.449, n = 17; test weight: $R^2 = 0.02$, p = 0.635, n = 14).

The responses of 1000 grain weight to N fertilizer were negative in 8 studies, positive in 17 studies and neutral in 4 studies. The weighted mean of the responses for all 29 experiments was 1%, but the effect was not statistically significant, since CI overlapped zero (-1% to 5%). The responses of test weight were negative in 13 studies, no changes were found in 7 studies and the effects were positive in 7 studies. The weighted mean of the test weight responses for all 27 experiments was slightly negative (-1.1%, CI -2.2% to -0.2%). The rank correlation test for this dataset showed no relationship between standardized ln *r* and sample size for either 1000 grain weight (Z = 1.331, p = 0.183, n = 29) or test weight (Z = 1.59, p = 0.112, n = 27). This indicates no selection bias in which larger effect sizes would be more likely to be present than smaller effect sizes. The results can therefore be considered to be reliable estimates of the true effects.

Neither the responses of 1000 grain weight (p = 0.413) nor that of test weight (p = 0.429) differed between decades, indicating that the old and modern varieties of spring cereals responded in a similar manner. Also the responses had no relationship to increasing N rates across the decades (Fig. 1).



N rates (kg ha⁻¹)

Fig. 1. Scatter plot between N rates and response of 1000 grain weight (a) and test weight (b) of spring cereals cultivated on clay and coarse-textured mineral soils in different decades, 1950s–1990s. Each symbol represents the average response for the duration of an experiment. $Q_{_{M'}}$ model heterogeneity; *n*, the number of observations.

The meta-regression showed that the response of 1000 grain weight to N fertilizer decreased with increasing SOM content (Fig. 2a). Thus, N fertilizer may increase 1000 grain weight by up to 10% in mineral soils with very low SOM content, but it may reduce the grain weights slightly in soils with high SOM content. In contrast, the meta-regression showed no associations between the response of test weight and SOM content (Fig. 2b).

However, the responses of 1000 grain weight did not differ significantly between cereal species (p = 0.883), soil textures (p = 0.208) or soil pH groups (p = 0.710), nor was the response modified by the amounts of PK fertilizers in the control treatments (p > 0.07). There was a tendency toward larger responses in studies with longer duration (p = 0.07). The responses varied between both cultivation zones (p = 0.03) and yield level groups (p = 0.003). However, these explanatory variables were inter-correlated ($\chi^2 = 22.4$, df = 6, p = 0.001). Thousand grain weight increase over the control due to N fertilizer was 6% in the "medium" yield groups, located mostly in zone I, whereas it was negligible in the "low" and "high" yield groups, located mostly in zones III and IV.

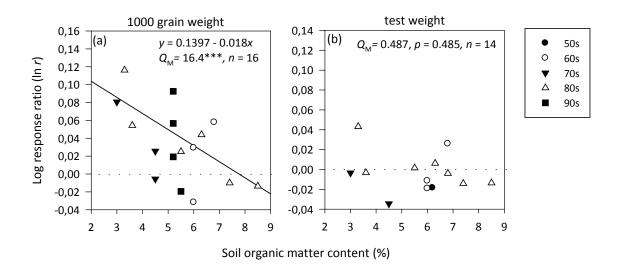


Fig. 2. The effect of SOM content on the log response ratio of 1000 grain weight (a) and test weight (b) to N fertilization of spring cereals cultivated on clay and coarse-textured mineral soils from the 1950s to the 1990s. Each symbol represents the average ln r for the duration of an experiment. For back-transformation of ln r see Eq. 4. $Q_{_{\rm M}}$, model heterogeneity; n, the number of observations. *** p < 0.001.

Likewise, the responses of test weight to N fertilizer did not differ between cereal species (p = 0.687), soil textures (p = 0.210), soil pH groups (p = 0.565) or yield level groups (p = 0.246), nor was the response of test weight modified by the amounts of PK fertilizers in the control treatments (p > 0.2). The responses varied between cultivation zones (p = 0.028), but when one experiment with extremely low response was excluded (-5.3%, spring barley, Exp 21), no difference was found (p = 0.855). The largest responses were found in studies with longest duration (p = 0.049).

N uptake

The effects of N fertilizer on N uptake were studied in 22 experiments (Appendix). In the controls, the average N uptake was 35.9 ± 8.1 kg ha⁻¹ for barley (n = 11), 35.8 ± 9.6 kg ha⁻¹ for wheat (n = 9) and 43.3 ± 14.3 kg ha⁻¹ for oats (n = 2). It had no relationship with SOM content for the cereal species pooled together ($R^2 = 0.08$, p = 0.361, n = 13). The increase in N uptake over the control due to N fertilizer (100-150 kg ha⁻¹) was 123 % (Cl 101-142%, 38 observations from 18 experiments). The effects of categorical explanatory variables on N uptake response are shown in Figure 3a.

The responses varied significantly between soil pH groups, cultivation zones and decades. On SA soils the response was more than twice that on MA soils. The response was also highly variable across the cultivation zones: that in zone I was three-fold that in zone III. However, soil pH and cultivation zones were highly inter-correlated ($\chi^2 = 21.8$, df = 2, p < 0.001): 28 out of 31 observations on SA soils were located in zone I, whereas most observations on MA soils were located in zones II and III. Therefore, it is not possible to separate the effect of pH from that of cultivation zones.

The response of N uptake increased over time: it was three-fold in the 1990s compared to that in the 1960s (Fig. 3a), indicating better N utilization by the modern varieties. However, the responses did not vary between the yield level groups (p = 0.176), soil textures (p = 0.257) or cereal species (p = 0.468). Nor was the response modified by the amounts of PK fertilizer in the control treatments (p > 0.05) or by SOM content (p = 0.401). There was a tendency toward larger N uptake responses in studies with longer duration (p = 0.09).

The magnitude of N uptake responses depended strongly on N rates as indicated by the weighted meta-regressions; it increased more steeply in SA than in MA (Fig. 4). However, we did not extrapolate the regression lines beyond the observed range and, thus, the regressions are valid for N rates of 30-216 kg ha⁻¹ (SA soils) and 50-150 kg ha⁻¹ (MA soils).

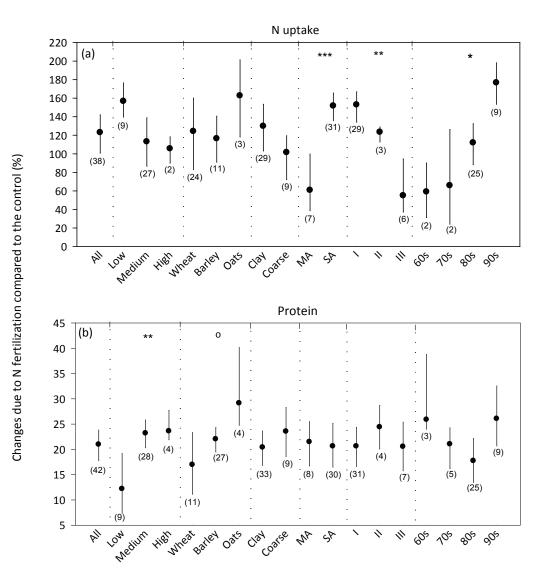
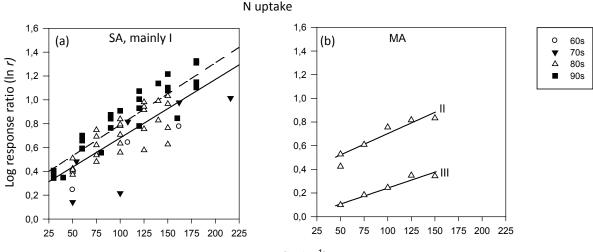


Fig. 3. Response of N uptake (a) and protein content (b) to N fertilizer (100–150 kg N ha⁻¹). Studies were further subdivided according to yield level without added N ("Low", "Medium", "High"), cereal species, soil texture (Clay, clay soils; Coarse, coarse-textured mineral soils), soil pH (MA, moderately acidic; SA, slightly acidic), cultivation zones (I–III), and decade. Each dot represents the weighted average N uptake or protein increase and the bars show 95% confidence intervals. The numbers in parentheses indicate the number of observations. ° p < 0.1; * p < 0.05; ** p < 0.01; *** p < 0.001.



N rates (kg ha⁻¹)

Fig. 4. The log response ratio of N uptake in relation to N rates in SA soils (a) and MA (b) soils, in cultivation zones I, and II and III, respectively. Each symbol represents the average ln r for the duration of an experiment. Solid lines represent the meta-regressions for the early studies (1960s–1980s). The broken line represents the meta-regression for the recent studies (1990s). Abbreviations are the same as in Fig. 3.

The parameters and fittings of the meta-regressions appear in Table 3. Within SA soils, an increase of 10 kg N ha⁻¹ was associated with 4.9% and 5.2% increases in the N uptakes over the controls in the early and recent studies, respective-ly. For MA, data were only available from three experiments on barley conducted in 1980s (Exp. 11, 14 and 22). Within MA, there was statistically significant difference between the cultivation zones (p = 0.004), and an increase of 10 kg N ha⁻¹ was associated with 3.8.% and 2.7% increases in N uptakes over the controls for zones II and III, respectively.

Table 3. Parameters and fittings of the weighted meta-regressions between N rates (N, kg ha⁻¹) and log response ratios of N uptake (ln r) for spring cereal species cultivated on SA and MA soils from the 1960s to the 1990s.

Soil pH	Cultivation	Decades	$\ln r = aN + y_0$		Model fitting	s
groups	zones	Decades	a (±SE)	$y_0(\pm SE)$	Q _M	n
C 4	mainly	1960s-1980s	0.0049 (0.0010)	0.1908 (0.1014)	26.3***	36
SA	mainly I	1990s	0.0052 (0.0004)	0.2705 (0.0476)	177.5***	26
MA	II	1980s	0.0038 (0.0015)	0.2642 (0.1420)	6.3*	8
MA	111	1980s	0.0027 (0.0006)	- 0.0294 (0.0600)	17.6***	5

SE, standard error; Q_{M} , model heterogeneity; n, the number of observations.For back-transformation of ln r see Eq. 4. * p < 0.05; *** p < 0.001. The equations are valid for N rates of 30–216 kg ha⁻¹ (SA soils) and 50–150 kg ha⁻¹ (MA soils).

Protein content

The entire database on the effect of N fertilizer on protein content consisted of 27 experiments (Appendix). In the control treatments, average protein content was $10.7 \pm 0.9\%$ for barley (n = 14), $12.7 \pm 2\%$ for wheat (n = 10) and $9.7 \pm 1.2\%$ for oats (n = 3). It had no relationship with SOM content for the cereal species pooled together ($R^2 = 0.02$, p = 0.682, n = 12).

The protein content increase over the control due to N fertilizer use (100–150 kg ha⁻¹) was 21% (Cl 18–24%, 42 observations from 22 experiments). The protein response varied significantly between the yield level groups (p = 0.007) and it varied marginally between cereal species (p = 0.074). The "medium" and "high" yield groups responded to a larger extent than did the "low" group, and barley and oats responded somewhat more than wheat (Fig. 3b). However, it was not possible to separate the effect of these explanatory variables from each other due to their interaction ($\chi^2 = 11.6$, df = 4, p = 0.02): 6 out of 11 observations with wheat belonged to the "low" yield group, while 24 out of 27 observations with barley belonged to the "medium" and "high" groups.

The protein responses to N fertilizer did not significantly vary between soil pH groups (p = 0.835), cultivation zones (p = 0.774), soil textures (p = 0.463) or decades (p = 0.127; Fig. 3b). Soil organic matter content did not modify the protein response (p = 0.923, n = 20). Finally, neither P nor K rates in the controls affected the protein response (p > 0.05). There was, however, a tendency for larger protein responses in studies with longer duration (p = 0.064, n = 42).

For the entire database the magnitude of protein response strongly depended upon N rates, as indicated by the weighted meta-regressions (Fig. 5). The parameters and fittings of the meta-regressions appear in Table 4, and the equations are valid for N rates \geq 30 kg ha⁻¹. An increase of 10 kg N ha⁻¹ was associated with a 2.2–2.4% increase in protein content over the control for barley and oats, and with a 1.7% increase for wheat.

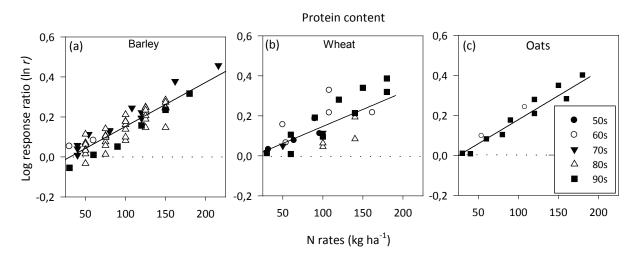


Fig. 5. Log response ratio of protein content in relation to N rates for barley (a), wheat (b) and oats (c) cultivated on clay and coarsetextured mineral soils in different decades, 1950s–1990s. Each symbol represents the average ln *r* for the duration of an experiment.

Chring corool chooses	$\ln r = aN + y_0$		Model fittings	5
Spring cereal species	a (±SE)	y ₀ (±SE)	Q _M	n
Barley	0.0022 (0.0002)	-0.0674 (0.0207)	115.7***	57
Wheat	0.0017 (0.0004)	-0.0230 (0.0378)	22.2***	25
Oats	0.0024 (0.0003)	-0.0614 (0.0272)	77.5***	12

Table 4. Parameters and fittings of the weighted meta-regressions between N rates (N, kg ha⁻¹) and log response ratios of protein (ln r) for spring cereal species cultivated on clay and coarse-textured mineral soils from the 1950s to the 1990s.

Abbreviations are the same as in Table 3. For back-transformation of ln *r* see Eq. 4.

The equations are valid for N rates \geq 30 kg ha⁻¹.

Estimates of protein increase at $\rm N_{_{opt}}$ and $\rm N_{_{max}}$

By using the linear regressions between N rates and the response of protein content (Table 4), we estimated protein increases with the application of N_{opt} for the different yield level groups. For the "low" and "medium" yield groups, N_{opt} would increase protein content over the control by 30–46%, resulting in absolute values of about 14% for barley and oats, and 16.5% for wheat (Table 5a). In contrast, for the "high" yield groups, N_{opt} would increase protein contents negligibly for barley and wheat, whereas for oats it would provide an adequate response, although the protein content in harvested yield would be only 11.6%.

The estimates of protein content and its response at N_{max} are shown in Table 5b. In FAEP, the yields without added N are not defined and therefore only one value of N_{max} is set in this comparison for each cereal species. N_{max} would increase protein content over the control by 16–20% for barley and wheat, and by 25% for oats. Thus, using the experimental data, the absolute values of protein content would be about 12% in barley and oats, and 15% in wheat.

Parameter	Barley			Wheat		Oats	
(a) N _{opt} :	145	145	57	170	45	184	101
Yield level without N (kg ha ⁻¹)	1500 (low)	2500 (medium)	3500 (high)	1500 (low)	3500 (high)	2500 (medium)	3500 (high)
Yield with N _{opt} (kg ha ⁻¹)	3500	4000	4000	3500	4000	5000	5500
Protein without N (%)	10.7	10.7	10.7	12.7	12.7	9.7	9.7
Protein with N _{opt} (%)	13.8	13.8	11.3	16.5	13.3	14.2	11.6
Response over control (%)	29	29	6	30	5	46	20
(b) N _{max} :	100			120		120	
Yield level without N (kg ha-1)	Not defined	I		Not defined		Not define	d
Yield with N_{max} (kg ha ⁻¹)	4000			4000		5000	
Protein without N (%)	10.7			12.7		9.7	
Protein with N _{max} (%)	12.4			15.2		12.1	
Response over control (%)	16			20		25	

Table 5. Estimates of protein content at application of N rates (a) justified as cost-effective (N _{ort}) ^a and (b) maximally permitte	d by
FAEP (N _{max}) ^b .	

^a Valkama et al. (2013). N_{opt} for the prices of N fertilizer 0.92 € kg ⁻¹, barley yield 0.157 € kg ⁻¹, wheat yield 0.181 € kg ⁻¹ and oats yield

0.164 € kg⁻¹. These fertilizer N-to-cereal price ratios are 5.86, 5.08 and 5.61 for barley, wheat and oats, respectively.

^b Ministry of Agriculture and Forestry (2011)

Discussion Thousand grain and test weights

The response of 1000 grain weight demonstrated some variation across the studies: it decreased with increasing SOM content, as did the yield response (Valkama et al. 2013). The summarized response of 1000 grain weight to N fertilizer was only 1% and not statistically significant. Similarly, it remained unaffected by N fertilizer in a Swedish study with different varieties of oats (Oscarsson et al. 1998). Previous long-term Finnish experiments conducted on clay soils demonstrated an increase of 1000 grain weight of wheat at increasing N rates (50–200 kg N ha⁻¹), but in barley, no increase was found at N application rates above 100 kg ha⁻¹ (Esala and Larpes 1986).

The summarized response of test weight was slightly negative, with no relationship with increasing N rates, supporting the findings of previous studies with barley, and both spring and winter wheat (Esala and Larpes 1986, Oscarsson et al. 1998, López-Bellido et al. 2001, Kienzler et al. 2011). Otherwise, the responses of 1000 grain and test weights were similar for the different soil textures, soil pH and cereal species.

Lack of variation between the decades suggests that the modern varieties of spring cereals did not differ from the old ones in terms of the responses of 1000 grain weight or test weight. Likewise, Rekunen (1988) demonstrated no relationship between the year of release (1921–1982) and 1000 grain weight or test weight of oats, based on long-term field experiments, in which the varieties of different periods were simultaneously compared under current fertilizer recommendation (100–120 kg N ha⁻¹). Also, he claimed that the impacts of environmental factors on test weight and other quality parameters such as protein content, as well as on cereal yield, were at least five times greater than the effect of genotypes.

Data based on Finnish farm surveys showed that, in 1990–2005, reduced N application rates were associated with lower test weight and 1000 grain weight (Salo et al. 2007b). However, the magnitude of grain quality reduction was not solely explicable by N application rates, since many other factors and management practices changed in Finnish cereal production during that period. For example, the decrease in cereal prices resulted in low investments in soil drainage and liming, which may be associated with reduced grain quality. The results based on the present meta-analysis of controlled field experiments indicate that lower N rates do not seem to affect 1000 grain or test weights within the diversity of Finnish conditions.

N uptake

Our results demonstrate that the main factors determining N uptake variability under northern conditions were soil acidity and the average length of the growth period, however, due to their inter-correlation, it was not possible to separate these effects. As a result, with increasing N rates, N balance increased more steeply in MA soils than it did in SA soils (Valkama et al. 2013). As the pH decreases, the solubility of aluminum (AI) and manganese (Mn) increases, and excess Al³⁺ and Mn²⁺ in the soil solution interferes with root and shoot growth and function, as well as restricting crop uptake of nutrients and water (e.g., Chesworth 2008). Therefore, to reduce N balances, liming could prove beneficial, since it is known to increase the N uptake and N concentration in grain (Lyngstad 1992, Soon and Arshad 2005).

The response of N uptake increased over the decades, indicating better N utilization by modern varieties of spring cereals. This is in accordance with the previous Finnish study by Muurinen et al. (2007), who demonstrated higher N uptake efficiency by modern varieties of barley, wheat and oats than by older ones. Similarly in the UK, the N uptake response of spring barley increased by 0.26% per year during the breeding period from 1930s to 2000s (Bingham et al. 2012).

Protein content

Regarding the variability of protein response to N fertilizer between the studies, wheat with "low" yields in the controls responded only half as well as barley or oats with "medium" or "high" control yields. This may indicate lower protein response in wheat compared to other cereal species. However, it was not possible to separate the factor of "cereal species" from that of "yield level group" due to their inter-correlation. A previous Finnish study showed that spring cereals differed in N accumulation and translocation according to their N use efficiency (NUE) values, and, in particular, the NUE of wheat was lower than that of barley and oats (Muurinen et al. 2007).

Another possible explanation of the smaller protein response in the "low" yield group is a dilution effect, i.e. protein synthesis increased due to N fertilizer to a lesser extent than did the synthesis of yield biomass. For example, the protein content increased over the control due to N fertilizer (100–150 kg ha⁻¹) by 13% (Fig. 3a), while the grain yield increased by 100–125% (Valkama et al. 2013, Fig. 3). By contrast, in the "high" yield group, both protein content and yield increased, due to N fertilizer, to the same extent (about 25%). Similarly, Clarke et al. (1990) demonstrated a decrease in protein content of wheat due to dilution of N by larger biomass. Cultivars with higher yield potential tended to have lower protein contents than did cultivars with low yield potential, at any given level of available N (Clarke et al. 1990). Peltonen-Sainio et al. (2012) showed the same tendency based on large Finnish datasets for spring cereals, except for some superior lines combining high grain protein concentration with relatively high grain yield.

The results indicated that soil parameters such as soil texture, pH and SOM, within the range of the dataset, did not modify the protein response to N fertilizer. This is in accordance with the study by Pettersson and Eckersten (2007), who reported that under northern conditions grain protein content was not correlated with soil pH, SOM or soil mineral N. Also our results suggest that protein response to N fertilizer did not differ among the varieties, since there was no obvious decade effect. Likewise, no significant variety × N rate interaction was recently found for the protein content of wheat (Swanston et al. 2012).

The current meta-analysis supports the results of previous studies showing a linear relationship between N rates and protein content of cereals (Baker et al. 2004, Brennan and Bolland 2009, Mooleki et al. 2010, Kienzler et al. 2011). However, increasing protein content by applying higher rates of fertilizer is relatively inefficient, as NUE decreases with increasing N level, especially under dry soil conditions (Gauer et al. 1992). Decreasing NUE reduces economic benefits of fertilizer application at higher rates. Therefore, beyond some point, addition of N fertilizer would not be a useful practice (Gauer et al. 1992).

We estimated whether N_{opt} or N_{max} provide high quality yields. For malting barley, low grain protein content is desirable, since higher protein levels result in lower starch content, less alcohol production and risks of cloudy beer, although yeast activity may be limited by N shortage at lower grain protein levels (Pettersson and Eckersten 2007). In our previous study, we demonstrated that N_{opt} (Valkama et al. 2013). Based on this, it seems that an N_{opt} of 145 kg ha⁻¹ is too high for malting barley within the "low" and "medium" yield groups, where protein content in harvested yield would be 14%. The ideal grain protein concentration for production of European lager beer is 10.7% of dry matter, with a permitted range of 9.5–11.5% (Jensen and Schjoerring 2011). In Finland the upper limit for protein content of malting barley is 12% (Kivi and Hovinen 1972). Therefore, for these yield level groups, the reduced limits set by FAEP may be preferable in order to achieve the desirable low protein content of the grains. In contrast, for the "high" yield group of barley, an N_{opt} of 57 kg ha⁻¹ may be sufficient to provide both high harvested yield and desirable low grain protein content.

For bread-making from wheat and oats, the minimum protein content is typically taken as 13%. Therefore, N_{opt} justified for the "low" and "medium" yield groups would give a favorable outcome, since protein increases can reach 30 - 46% over the control. In contrast, N_{opt} for the "high" yield groups may be insufficient, particularly for wheat to obtain desirable level of protein content. The estimates suggest that, despite the low protein response in wheat, an N_{max} of 120 kg ha⁻¹ would be enough to overcome the limit of 13%. However, initial protein content without N should not be below 10.8%. In contrast, for oats, N_{max} of 120 kg ha⁻¹ may not be enough to reach the limit, despite the good protein response, as its initial content was less than 10% in the experimental data of our dataset.

Conclusions

Knowledge given by reliable equations between N rates and grain quality is important in the light of current trends to reduce N fertilizer application, in order to maintain yield quality at a desirable level. Using a large number of experiments conducted on the different varieties of spring cereals on a range of mineral soils, under varying growth conditions and over several decades, the present study attempted to reveal the sources of variation in responses of grain quality parameters to N fertilizer use. The N uptake responses to N fertilizer showed variation among the soil acidity groups, cultivation zones and also among the decades (i.e. among the old and more modern varieties). In contrast, the protein response to N fertilizer varied between the yield level groups without added N, and to some extent between spring cereal species. The lack of response of 1000 grain weight and the negligibly small negative response of test weight clearly suggest that, in general, no reduction of these yield quality indicators is likely at reduced N fertilization rates.

The models of protein responses to N fertilizer gained in the present study may be combined with the yield response models obtained from our previous study (Valkama et al. 2013), to build a tool for adjusting N applications, in order to produce high yields of good quality as well as giving the best economic return to growers in Nordic conditions.

Acknowledgements

This study was funded by MTT Agrifood Research Finland and by the EU (project #040 Baltic COMPASS – "Comprehensive Policy Actions and Investments in Sustainable Solutions in Agriculture in the Baltic Sea Region, 2010– 2012"). We thank an anonymous reviewer and Prof. Hugh Riley (Bioforsk, Norwegian Institute for Agricultural and Environmental Research) for their comments on the manuscript and for English language editing.

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Appen	dix. Description of th	ne database used f	or the me	ta-analysis of	the effects o	f N fertilizer on 10	000 grain	Appendix. Description of the database used for the meta-analysis of the effects of N fertilizer on 1000 grain weight (GW), test weight (TW), protein content (Pr) and N uptake (Nup).	N), protein content (Pr) a	ind N uptake (Nup).	
z	Site and Cultivation zone	Soil type	рН (О ₂ Н)	Soil acidic group ^a	SOM (%) ^b	Experimental years	N of years	Variety	N rates	Response variables	Reference
							Spring	Spring barley			
1	Anjalankoski l	Silt clay	6.5	SA	I	1985–1987	ŝ	Ingrid, Kustaa, Patty, Kymppi, Hankkijan Pokko, Kilta	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of Kymenlaakso experimental station (ES)
7	Helsinki I, Tammisto experimental farm	Clay	I	I	I	1967–1969	ŝ	Arvo, Ingrid, Karri, Pirkka	30, 60	Pr, Nup	Kivi and Hovinen (1972)
m	Jokioinen I	Heavy clay	6.5	SA	6.3	1982–1987	9	Ingrid, Kustaa, Aramir, Ida, Harry, Patty,Welam, Hankkijan Pokko, Kilta, Pirkka	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of Plant Production Research
4	Jokioinen I	Loam clay	6.3	SA	5.2	1993–1996	4	Loviisa	30, 60, 90, 120, 150, 180	Pr, Nup, GW	Pietola et al. (1999)
ß	Kokemäki I	Fine sand clay	5.9	SA	3.7	1985–1986	2	Kustaa, Kilta, Kymppi	50, 75, 100, 125, 150	Pr, Nup	Report of Satakunta ES
9	Maaninka III	Fine sand	6.0	SA	I	1971–1975	ß	Olli, Otra, Vigdis	54, 108, 162, 216	Pr, Nup, GW, TW	Report of North Savo ES
7	Mietoinen I	Неаvy сlау	6.1	SA	3.3	1985-1987	c	Kustaa, Kilta, Kymppi	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of South–West ES
00	Mietoinen I	Fine sand	6.2	SA	I	1985–1988	ŝ	Kustaa, Kilta, Kymppi	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of South–West ES
6	Mikkeli II	Finer fine sand	I	I	6.8	1963–1965	ŝ	Otra	26, 52	GW, TW	Report of South Savo ES
10	Mikkeli II	Fine sand	I	I	6.8	1987–1989	ŝ	Arra	80	ТW	Report of South Savo ES
11	Mouhijärvi II	Silt	5.2	MA	I	1983–1984	2	I	50	Pr, Nup, GW, TW	Report of Sata–Häme ES
12	Kokemäki I	Clay	I	I	I	1974–1978	ŝ	Ingrid, Karri, Pirkka, Pomo	40, 80, 120	Pr	Lallukka et al. (1980)
13	Pihtipudas IV, Grower's fields ^c	Fine sand moraine soil	5.4	MA	3.0	1971–1972	2	Otra, Pirkka	54	GW, TW	The field book of fertilization experiments, MTT
14	Pälkäne II	Fine sand	5.6	MA	3.6	1985–1987	ŝ	Kustaa, Kilta, Kymppi	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of Häme ES
15	Pälkäne II	Finer fine sand	9	SA	I	1959–1960	2	Balder	26, 52, 103	GW, TW	Report of Häme ES
16	Vantaa I	Clay	I	I	I	1974–1978	4	Ingrid, Karri, Pirkka, Pomo	40, 80, 120	Pr	Lallukka et al. (1980)
17	Tohmajärvi III	Fine sand	5.7	MA	7.4	1983–1984	2	Etu	50, 100	GW, TW	Report of Karelia ES
18	Toholampi IV	Finer fine sand	5.7	MA	I	1975–1978	4	Eero	40	GW, TW	Report of Middle Ostrobothnia ES
19	Toholampi IV	Finer fine sand	5.0	MA	I	1963–1964	2	Otra, Mari	27, 54	GW, TW	Report of Middle Ostrobothnia ES
20	Toholampi IV	Finer fine sand	5.5	MA	4.5	1970–1973	œ	Otra	54, 108	GW, TW	Report of Middle Ostrobothnia ES
21	Toholampi IV	Finer fine sand	I	I	4.5	1970–1971	2	Eero, Etu	54, 108	GW	Report of Middle Ostrobothnia ES

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22	Ylistaro III	Silt clay	5.6	MA	5.5	1985–1986	7	Kustaa, Kymppi, Kilta, Hankkijan Pokko	50, 75, 100, 125, 150	Pr, Nup, GW, TW	Report of South Ostrobothnia ES
23	Ylistaro III	Finer fine sand	6.1	SA	8.5	1985–1987	2	Kustaa, Kymppi, Kilta, Hankkijan Pokko	50, 75, 100, 125	GW, TW	Report of South Ostrobothnia ES
24	Ylistaro III	Clay	I	I	I	1974–1978	4	Ingrid, Karri, Pirkka, Pomo	40, 80, 120	Pr	Lallukka et al. (1980)
							<u>Sprin</u>	Spring wheat			
1	Jokioinen I	Clay loam	6.5	SA	4.6	1988–1989	2	Heta, Kadett	100, 140	Pr, Nup	Esala (1991)
2	Jokioinen I	Clay loam	6.3	SA	5.2	1993–1996	4	Satu	30, 60, 90, 120, 150, 180	Pr, Nup, GW	Pietola et al. (1999)
ŝ	Jokioinen I	Fine sand clay	5.7	MA	6.0	1953	сı	Timantti, Timantti II, Kärni, Tammi	32, 64, 96	Pr, TW	Report of Plant Production Research
4	Luumäki I, Rikkihappo Oy's experimental farm	Fine sand clay	I	I	I	1968–1969	7	Apu, Svenno	06	Pr, Nup	Pessi et al. (1971)
5	Maaninka III	Fine sand	6.0	SA	I	1968–1970	ŝ	Apu	108, 162	Pr, Nup, GW, TW	Report of North Savo ES
9	Mietoinen I	Fine sand clay	6.2	SA	2.9	1988–1989	2	Heta, Kadett	100, 140	Pr, Nup	Esala (1991)
7	Mietoinen I	Fine sand clay	6.9	SA	I	1991–1996	9	Satu, Heta, Reno, Runar, Luja, Kadett, Polkka, Laari	60, 100, 140, 180	Pr, Nup, GW, TW	Report of South–West ES
80	Mietoinen I	Неаvy сlау	5.8	SA	6.0	1960–1963	ŝ	Norröna, Svenno	54, 108	GW, TW	Report of South–West ES
6	Nakkila I	Fine sand	5.9	SA	I	1970–1973	4	Ruso, Veka	50, 100	Pr, Nup, GW, TW	Report of Satakunta ES
10	Nakkila I	Fine sand clay	5.4	MA	I	1960–1964	Ŋ	Apu, Norröna	54, 108	Pr, Nup, GW, TW	Report of Satakunta ES
11	Pälkäne II	Fine sand	6.0	SA	I	1956–1957	2	Timantti II	16, 32, 64	GW, TW	Report of Häme ES
12	Helsinki I, Viikki experimental farm	Muddy clay	5.8	SA	9	1962–1963	2	Touko	50	Pr, Nup, GW, TW	Raininko (1966)
							O	<u>Oats</u>			
Ч	Jokioinen I	Clay loam	6.3	SA	5.2	1993–1996	4	Yty	30, 60, 90, 120, 150, 180	Pr, Nup, GW	Pietola et al. (1999)
2	Laukaa III	Silt	I	I	I	1957–1961	4	Sisu	27, 54, 108	GW, TW	Report of Middle Finnish ES
ŝ	Mietoinen I	Muddy clay	5.3	MA	I	1960–1963	4	Sisu	54, 108	Pr	Hakkola (1965)
4	Ylistaro III	Silt clay	5.9	SA	5.5	1991–1999	6	Aarre, Yty, Virma, Veli, Salo, Roope, Puhti, Leila, Kolbu, Katri	40, 80, 120, 160	Pr, Nup, GW	Report of South Ostrobothnia ES
^a MA, m ^b SOM, : ^c Statior	* MA, moderately acidic (pH 5.0–5.7); SA, slightly acidic (pH 5.8–6.9) b SOM, soil organic matter c Stationary experiments conducted on grower's fields.	.0–5.7); SA, slightly ac ucted on grower's fiel	idic (pH 5.8- ds.	-6.9)							

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