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Research Centre for Agriculture
State Laboratory for Soil and Crop Research
Department of Plant Biochemistry
Lottenborgvej 24
DK-2800 Lyngby

Chemical composition of barley varieties with different nutrient supplies

II. Concentration of amide and amino acids

Bygsorters kemiske sammensætning ved varierende næringsstoftilførsel
II. Koncentration af amid og aminosyrer

Ebbe Truelsen and Chresten Sørensen

Summary

21 commercial barley varieties, and breeders material KVL 468, and Risø 1508 were grown in pots and in the field with increasing nitrogen nutrition.

In relation to the protein contents, increasing nitrogen supply caused increases in proline, glutamic acid and amide, and decreases in the essential, limiting amino acids: lysine, threonine and cystine + methionine.

Risø 1508 had, in relation to the requirements of monogastric animals, the most advantageous amino acid composition, followed by KVL 468. The varieties Alva and Claudia were also better than average, but this was partly caused by a rather low protein content. Among the varieties with a less balanced amino acid composition the most prominent ones were Salka, Rupal and Duks, while the impaired composition in Mona, Aramir and Diva were caused by higher nitrogen concentrations.

Different growth conditions and soil types had no measurable influence on the varietal differences in the amino acid compositions.

Key words: Barley varieties, amino acids, amide, nutrition.

Resumé

I kerne fra 21 bygsorter samt Risø 1508 og KVL 468 tilført stigende kvælstofmængder blev der bestemt aminosyresammensætning i materiale fra karforsøg og amidindhold i materiale fra kar- og markforsøg.

Stigende kvælstofgødskning medførte foruden et stigende proteinindhold i kernen også en forskydning i proteinets aminosyresammensætning. Således fandtes en stigning i koncentrationen af prolin, glutaminsyre (glutaminsyre + glutamin excl. amidgruppen) og amid og et fald i de begrænsende, essentielle aminosyrer lysin, threonin og de svovlholdige aminosyrer cystin og methionin. Det relative indhold af prolin, glutaminsyre og amid var generelt højere i sorter med højt proteinindhold, og omvendt fandtes der højere indhold af begrænsende, essentielle aminosyrer i lavproteinsorter.

Risø 1508 skilte sig klart ud fra de øvrige ved at have et væsentligt højere indhold af lysin og threonin i proteinet og samtidigt lavere indhold af prolin, glutaminsyre og amid. Den efterfulgtes i karforsøgene af KVL 468, som overensstemmende med sit høje proteinindhold havde relativt høj amidkoncentra-

tion i markforsøgene. Alva og Claudia havde ligeledes relativt højt indhold af lysin og lavt indhold af prolin, glutaminsyre og amid, men det skyldtes delvis et lavere proteinindhold.

Blandt sorterne med højere indhold af prolin, glutaminsyre og amid i proteinet og lavere indhold af lysin havde Mona, Aramir og Diva et relativt højt proteinindhold, mens Salka, Rupal og Duks havde proteinindhold nær gennemsnittet, hvorfor årsagerne til disse sorters afvigende aminosyresammensætning må tilskrives andre forhold.

De forskellige vækstbetingelser ved kar- og markforsøg og jordtyper i Borris og Tystofte syntes ikke at medføre ændringer i de variationer, der blev fundet som følge af sort eller gødskning.

Nøgleord: Bygsorter, aminosyrer, amid, ernæring.

Introduction

The proteins in the barley grain have been the subject of extensive investigations due to their importance as monogastric feeding stuff (3, 8, 9). As a whole the barley proteins are characterized by a relative low content of the essential amino acids lysine and threonine, and a rather high level of proline and glutamine (5, 10). For this reason a great part of nitrogen will be lost in the urine from non-ruminant animals, if barley grain is their only diet, hence the barley protein has a rather low biological value (2, 3, 11).

Much effort has been made to find varieties and isolate mutants which have a more balanced amino acid composition. However, those which have been found, e.g. Hiproly (12), KVL 468 (19) and Risø 1508 (7) have all given smaller yields (9), and therefore have not been used for agricultural purposes.

The amino acid composition of the barley grain is strongly affected by nitrogen nutrition, so that increasing nitrogen supply causes a decrease in the contents of lysine and threonine in relation to the protein content despite an increase in the concentrations in dry matter being seen (15, 17, 19). The amide, which in the barley grain mainly derives from glutamine (8) increases with increasing nitrogen supply (15). A negative correlation between the relative concentrations of lysine and amide has been found (8, 16, 17). The relative contents of the sulphur-containing amino acids are little affected by nitrogen nutrition, but a slight decrease has been found with increasing N-supplies (4).

Previous investigations have shown differences in the amino acid composition among different barley varieties (15, 17, 19). The present work was carried out to discover any differences among newer common grown varieties including KVL 468 and Risø 1508.

Materials and methods

The barley varieties were supplied with increasing amounts of nitrogen and grown at different localities in Denmark. The grains used for the analyses originate from the same experiments as those from which results concerning concentrations of total nitrogen, tannins, β-glucans, phytate and minerals have been published elsewhere (18).

The amino acid analysis was performed according to *Spackman et al.* (14), following the modification by *Pedersen et al.* (13). The amide was determined as described by *Sørensen* (16).

Results

From the results in Table 1 it can be seen that the lysine concentrations in per cent of the protein contents decreased with increasing nitrogen supplies, and hence with increasing protein concentrations. Some varietal differences were found with the same nitrogen supply, the mutant Risø 1508 had a relatively high content of lysine, however the varieties Claudia, Alva, KVL 468, Zita, Canova and Varunda also had higher contents than average. The varieties with the lowest lysine contents were Mona, Salka, Aramir, Diva and Rupal. The rate of decrease with increasing

Table 1. Lysine-N and amide-N as per cent of total nitrogen in barley seeds (pot experiments).
Indhold af lysin-N og amid-N i procent af totalkvælstof i bygkerner (karforsøg).

g N per pot *g N pr. kar*	0.5	2.0	5.0	9.5	0.5	2.0	5.0	9.5
Variety *Sort*	Lysine-N *Lysin-N*				Amide-N *Amid-N*			
1 Zita	5.1	4.6	4.2	3.7	11.3	12.4	14.1	15.1
2 Alf	4.9	4.3	3.9	3.7	11.1	13.1	13.6	14.4
3 Alva	5.3	4.4	4.3	3.8	10.5	12.3	13.4	13.9
4 Aramir	4.9	4.2	3.8	3.3	11.3	13.5	15.0	15.7
5 Canova	5.2	4.5	4.1	3.8	10.6	12.0	14.6	15.7
6 Claudia	5.5	4.6	4.1	3.8	9.8	12.2	13.9	15.3
7 Diva	5.0	4.2	3.8	3.3	11.0	13.2	14.6	15.6
8 Duks	4.9	4.4	4.0	3.6	11.4	13.1	14.7	15.3
9 Georgie	5.2	4.2	4.1	3.6	11.1	12.9	15.1	14.9
10 Harry	5.1	4.3	4.0	3.6	10.8	12.7	14.0	15.2
11 KVL 468	5.1	4.6	4.2	3.9	11.0	12.1	13.3	13.8
12 Lami	4.8	4.3	3.9	3.6	11.1	12.5	13.8	14.7
13 Lofa	5.2	4.4	3.8	3.5	10.3	12.8	14.0	15.2
14 Mona	4.7	4.2	3.7	3.5	12.0	13.7	14.4	15.3
15 Nordal	4.9	4.6	4.2	3.6	11.1	12.2	13.6	15.1
16 Prisca	5.0	4.2	3.9	3.7	11.3	13.2	14.2	13.8
17 Proctor	5.0	4.6	4.1	3.8	10.8	12.1	13.8	14.6
18 Risø 1508	6.5	6.3	6.0	5.7	7.5	8.0	8.5	9.5
19 Rupal	4.7	4.1	3.9	3.6	11.5	13.6	14.3	15.7
20 Salka	4.8	4.3	3.7	3.3	11.8	13.1	14.5	15.1
21 Tron	5.0	4.3	3.9	3.8	10.8	12.6	13.7	14.4
22 Varunda	5.1	4.5	4.1	3.9	11.5	12.4	14.2	14.6
23 Vega	5.2	4.4	4.1	3.7	10.4	12.3	14.1	15.2

nitrogen supplies was about the same for the different varieties, except Risø 1508 which had a considerably smaller decreasing rate than the other varieties.

The results from the amide determinations are presented in the same Table. Opposite to lysine the amide contents increased with increasing nitrogen levels. The mutant Risø 1508 had a lower increasing rate than the other varieties. The highest amide contents were found in the varieties Aramir, Mona, Rupal, Duks, Salka and Diva, and the lowest in Risø 1508, Alva, KVL 468, Claudia and Proctor. In accordance with the results mentioned the varieties with the highest amide contents corresponded to those with the lowest lysine contents and vice versa. The relationship between lysine and amide contents from the pot experiments is shown in Fig. 1.

The results from the determinations of the amino acids threonine and glutamic acid (glutamic acid + glutamine excl. the amide group) are shown in Table 2. The variation in the concentrations of threonine is relatively small.

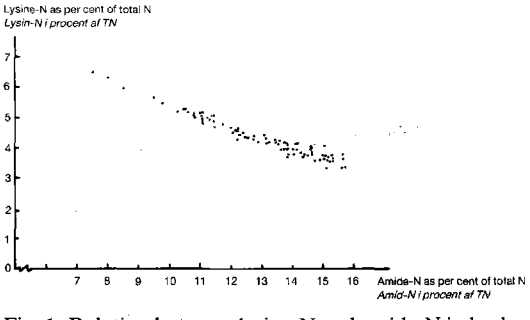

Fig. 1. Relation between lysine-N and amide-N in barley seeds (pot experiments).
Sammenhængen mellem lysin-N og amid-N i bygkerner (karforsøg).

Table 2. Threonine-N and glutamic acid-N as per cent of total nitrogen in barley seeds (pot experiments).
Indhold af threonin-N og glutaminsyre-N i procent af totalkvælstof i bygkerner (karforsøg).

g N per pot *g N pr. kar*	0.5	2.0	5.0	9.5	0.5	2.0	5.0	9.5
Variety *Sort*	Threonine-N *Threonin-N*				Glutamic acid-N *Glutaminsyre-N*			
1 Zita	2.6	2.5	2.4	2.3	12.7	14.2	15.2	15.8
2 Alf	2.6	2.5	2.3	2.3	11.8	14.9	14.6	15.6
3 Alva	2.7	2.4	2.3	2.2	11.5	13.4	14.2	15.1
4 Aramir	2.7	2.5	2.4	2.3	12.8	14.3	15.6	16.4
5 Canova	2.7	2.6	2.4	2.4	11.8	13.4	14.6	16.9
6 Claudia	2.9	2.5	2.4	2.4	11.6	13.1	14.6	16.3
7 Diva	2.7	2.5	2.5	2.2	12.9	14.1	15.3	16.1
8 Duks	2.7	2.6	2.5	2.4	12.3	14.3	15.9	16.4
9 Georgie	2.7	2.4	2.3	2.2	12.9	13.8	15.7	15.9
10 Harry	2.7	2.3	2.4	2.2	12.2	13.4	14.9	15.9
11 KVL 468	2.8	2.7	2.5	2.4	12.0	13.0	13.8	14.6
12 Lami	2.6	2.5	2.4	2.3	11.8	13.6	14.9	15.8
13 Lofa	2.7	2.4	2.3	2.2	11.8	13.9	14.7	15.9
14 Mona	2.5	2.4	2.3	2.1	13.0	15.1	15.5	15.9
15 Nordal	2.7	2.5	2.4	2.3	11.9	13.6	14.3	15.8
16 Prisca	2.7	2.4	2.3	2.2	13.0	14.9	15.4	15.8
17 Proctor	2.6	2.5	2.4	2.3	11.7	13.0	14.5	15.7
18 Risø 1508	3.1	3.0	2.9	2.7	9.2	9.1	9.6	9.9
19 Rupal	2.6	2.5	2.5	2.4	12.6	14.7	15.5	16.8
20 Salka	2.7	2.6	2.5	2.3	12.4	14.8	15.1	16.0
21 Tron	2.7	2.5	2.4	2.4	12.1	13.5	14.7	16.0
22 Varunda	2.6	2.5	2.3	2.4	12.1	13.6	14.7	15.8
23 Vega	2.7	2.5	2.3	2.3	11.7	13.8	14.7	16.0

Risø 1508, KVL 468, Duks and Salka have the highest concentrations and Mona and Harry the lowest. The glutamic acid contents were highest in Rupal, Mona, Aramir, Prisca and Duks, and lowest in Risø 1508, KVL 468, Alva, Proctor, Claudia and Nordal. In all varieties the threonine contents decreased with increasing nitrogen supplies, whereas the glutamic acid contents increased.

The results for the amino acids cyst(e)ine (cysteine + cystine), methionine and proline are presented in Table 3. The contents of the sulphur-containing amino acids cyst(e)ine and methionine decreased with increasing nitrogen concentrations, whereas proline increased. The highest concentrations of cyst(e)ine were found in the varieties Lofa and Proctor, and the lowest in Mona, Diva and Risø 1508. The differences in the contents of methionine were small.

The highest concentrations of proline were found in the varieties Mona, Duks, Prisca and Rupal, whereas the lowest were in Risø 1508, Alva, KVL 468 and Claudia. The increases in proline contents with increasing nitrogen concentrations were similar in all varieties except Risø 1508, which had a considerably smaller increase.

From the field experiments the results of the amide determinations are shown in Table 4. As in the pot experiments, the amide content increased with increasing nitrogen supplies, except at Borris, where a decrease from 0 to 60 kg N per ha was found. This is in agreement with the total nitrogen pattern, which has been discussed previously (18). At Tystofte the highest amide contents were found in the varieties Diva, Rupal, Aramir, Prisca and Duks, while Prisca, Duks, Salka, Mona and Rupal had the highest contents at Bor-

Table 3. Cyst(e)ine-N, methionine-N and proline-N as per cent of total nitrogen in barley seeds (pot experiments). *Indhold af cyst(e)in-N, methionin-N og prolin-N i procent af totalkvælstof i bygkerner (karforsøg).*

g N per pot *g N pr. kar*	0.5	2.0	5.0	9.5	0.5	2.0	5.0	9.5	0.5	2.0	5.0	9.5
Variety *Sort*	Cyst(e)ine-N *Cyst(e)in-N*				Methionine-N *Methionin-N*				Proline-N *Prolin-N*			
1 Zita	2.1	1.9	1.7	1.6	1.3	1.1	1.0	1.0	7.1	7.8	8.8	9.2
2 Alf	2.1	1.8	1.8	1.4	1.2	1.1	1.1	1.0	6.7	8.3	8.3	9.0
3 Alva	2.1	1.9	1.8	1.6	1.2	1.1	1.1	1.0	6.3	7.4	8.2	8.7
4 Aramir	2.1	1.9	1.6	1.5	1.2	1.1	1.1	1.0	6.8	8.1	9.1	9.7
5 Canova	2.1	1.9	1.8	1.6	1.3	1.3	1.2	1.1	6.7	7.4	8.6	9.9
6 Claudia	2.3	2.0	1.7	1.5	1.3	1.2	1.1	1.0	6.4	7.2	8.3	9.7
7 Diva	2.1	1.8	1.6	1.3	1.3	1.1	1.0	0.9	7.0	7.9	8.9	9.7
8 Duks	2.1	1.9	1.7	1.5	1.2	1.2	1.1	1.0	7.2	8.1	9.3	9.8
9 Georgie	2.2	1.8	1.7	1.5	1.2	1.1	1.1	1.0	7.2	7.7	9.1	9.2
10 Harry	2.5	1.8	1.7	1.6	1.3	1.1	1.0	1.0	6.9	7.9	8.9	9.8
11 KVL 468	2.1	1.9	1.7	1.6	1.2	1.1	1.2	1.0	6.8	7.4	7.9	8.8
12 Lami	2.1	2.0	1.7	1.5	1.2	1.2	1.1	1.0	6.5	7.8	8.5	9.3
13 Lofa	2.4	2.0	1.9	1.8	1.3	1.1	1.0	1.1	6.8	7.8	8.4	9.3
14 Mona	2.0	1.7	1.5	1.5	1.2	1.1	1.0	0.9	7.4	9.0	9.2	9.8
15 Nordal	2.1	2.0	1.7	1.5	1.2	1.2	1.1	0.9	7.1	8.1	8.6	9.7
16 Prisca	2.4	1.8	1.5	1.6	1.2	1.1	1.0	1.0	7.3	8.8	8.9	9.3
17 Proctor	2.3	2.1	1.9	1.7	1.2	1.2	1.1	1.0	6.8	7.6	8.7	9.6
18 Risø 1508	2.0	1.8	1.7	1.4	1.2	1.2	1.2	1.1	4.6	4.6	4.9	4.9
19 Rupal	2.1	1.9	1.8	1.6	1.4	1.2	1.2	1.1	7.1	8.4	9.0	9.7
20 Salka	2.1	1.9	1.9	1.5	1.2	1.2	1.1	1.0	7.1	8.3	8.7	9.3
21 Tron	2.1	1.9	1.7	1.5	1.3	1.2	1.1	1.0	6.9	7.6	8.6	9.6
22 Varunda	2.1	1.9	1.7	1.7	1.2	1.1	1.0	1.0	7.2	8.1	8.9	9.6
23 Vega	2.2	2.0	1.9	1.6	1.2	1.1	1.1	1.0	6.5	7.7	8.4	9.3

ris. The lowest concentrations were found at both research stations in Risø 1508, Alva and Claudia. The amide content in the protein was lower in the field experiments than in the pot experiments, and the concentrations at Tystofte were higher than those at Borris. For the three experiments the amide content was positively related to the nitrogen concentration presented previously (18).

Discussion

From the results (see also (18)) it can be calculated that the levels of proline, glutamic acid and amide increase with increasing protein contents in all experiments. On the other hand, in the protein, the contents of the essential amino acids lysine, threonine and the sulphur-containing amino acids decrease under these conditions. This is in agreement with other investigations where the prolamins increase and the albumins and globulins decrease with higher nitrogen contents (1, 8). However, it also appears that varieties with higher concentrations of lysine than average also have relatively lower concentrations of proline, glutamic acid and amide. Other varieties behave in an opposite manner. In the field experiments only amide determinations were made. According to Fig. 1 and (8, 16, 17) there is a close relationship between lysine and amide concentrations. Therefore it is highly probable that varieties with low amide contents have relatively high concentrations of lysine and vice versa.

In all experiments the lowest amide contents were found in Risø 1508, which is known to be low in prolamin (8), and which also had the highest lysine contents in the pot experiments. In the

Table 4. Amide-N as per cent of total nitrogen in barley seeds (field experiments).
Indhold af amid-N i procent af totalkvælstof i bygkerner (markforsøg).

Experiment *Forsøg*	Borris				Tystofte			
kg N per ha *kg N pr. ha*	0	60	120	180	0	60	120	180
Variety *Sort*	Amide-N *Amid-N*				Amide-N *Amid-N*			
1 Zita	11.3	11.1	12.8	13.7	11.9	12.2	13.0	13.5
2 Alf	11.1	10.8	11.7	13.0	12.4	12.1	12.8	13.0
3 Alva	10.8	10.4	10.8	12.5	11.8	11.6	12.0	12.9
4 Aramir	11.5	11.1	12.2	13.8	13.0	13.2	13.7	14.4
5 Canova	10.0	10.9	11.7	13.7	12.2	13.4	13.3	14.0
6 Claudia	10.2	10.0	10.8	12.7	11.2	11.9	12.3	13.2
7 Diva	11.0	11.2	12.6	14.1	12.8	13.7	14.8	14.4
8 Duks	12.0	11.4	12.8	14.5	12.9	13.0	13.8	14.2
9 Georgie	10.8	10.7	11.6	12.9	12.4	12.7	13.4	13.7
10 Harry	10.7	10.9	11.3	13.0	11.8	12.5	13.0	13.2
11 KVL 468	11.1	11.6	12.0	13.3	12.7	13.0	13.3	13.4
12 Lami	11.3	10.3	11.7	13.3	11.4	12.5	13.4	13.7
13 Lofa	11.5	10.6	12.5	13.2	12.1	12.9	13.2	14.0
14 Mona	11.7	11.2	12.7	14.0	12.4	13.4	13.8	14.0
15 Nordal	11.3	10.7	11.7	13.4	12.6	12.1	13.4	14.1
16 Prisca	11.9	11.7	13.0	14.3	12.6	12.9	14.1	14.7
17 Proctor	11.4	10.6	11.1	12.6	11.6	12.6	13.1	13.0
18 Risø 1508	8.6	7.9	8.3	8.5	8.0	8.4	8.8	8.7
19 Rupal	11.2	11.5	12.7	14.0	12.9	13.1	13.9	14.6
20 Salka	12.1	11.5	12.5	13.9	12.5	13.2	13.2	14.0
21 Tron	11.2	10.8	12.0	13.4	12.1	13.4	13.8	14.2
22 Varunda	11.8	10.4	12.1	13.1	12.4	11.6	12.7	13.0
23 Vega	10.6	10.1	11.3	13.1	11.7	12.5	13.1	13.4

field experiments, the varieties Alva and Claudia had, apart from Risø 1508, the lowest amide contents. In the pot experiments they were also among the lowest and at the same time among the highest in lysine contents. However, the varieties Alva and Claudia were also low in nitrogen contents, except Claudia in the pot experiments (18). From Tables 1 and 4 it can be seen that low N-supplies lead to high lysine and low amide concentrations in the protein, and the high lysine and low amide contents in Alva and Claudia may therefore partly be caused by their low nitrogen concentrations. KVL 468 had a high content of lysine and threonine and a low content of glutamic acid, proline and amide in the pot experiments, but was not especially low in amide in the field experiments (Table 4) because of its relative high nitrogen content (18).

Among the varieties with higher contents of glutamic acid, proline and amide and lower lysine contents than average, Mona, Aramir and Diva had relative high protein contents, while the protein contents in Salka, Rupal and Duks were near average.

When barley is fed to monogastric animals as a part of their diet the sulphur-containing amino acids can be the limiting amino acids in the mixture (6). According to *Eggum* (3) the best way to express the animal requirements for these amino acids is by using the sum of cyst(e)ine and methionine rather than methionine alone, because it looks like cyst(e)ine partly can replace

methionine. From the results it appears that the varieties Lofa and Proctor have a higher content than average of sulphur-bearing amino acids, and Mona and Diva are lower, but the differences are small.

Conclusion

The amino acid composition in the barley grain is highly influenced by its protein concentration. However, at equal protein contents varietal differences in the pattern of essential as well as non-essential amino acids among common grown varieties could be found. These varietal differences were maintained regardless of growth conditions or soil types.

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