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ANNALES AGRICULTURAE FENNIAE, VOL. 29: 91—105 (1990)

Seria ANIMALIA DOMESTICA N. 88 — Sarja KOTIELÄIMET n:o 88

COMPARISON OF ENERGY FEEDING STANDARDS FOR GROWING CATTLE

2. SILAGE-BARLEY RATIONS

MARTTI LAMPILA and ANGEL MICORDIA

LAMPILA, M. & MICORDIA, A. 1990. **Comparison of energy feeding standards for growing cattle. 2. Silage-barley rations.** Ann. Agric. Fenn. 29: 91—105. (Agric. Res. Centre, Inst. Anim. Prod., SF-31600 Jokioinen, Finland.)

The present paper is a continuation of the series in which energy requirement and intake of growing Ayrshire bulls are compared according to five standards. The standards applied (ARC, DDR, DUTCH, FU, MAFF) have been presented in the first part of the series.

The results presented are based on an experiment in which animals with an average age of about 151 d at the start were fed rations based on grass silage offered *ad libitum*. The experiment comprised 4 groups of 11 animals the feeding of which was complemented either with constant amounts of 1.5 (G1), 2.0 (G2) or 3.0 (G3) kg barley meal, or according to the metabolic weight (50 g per kg $W^{.75}$) (G4). The experimental time was 278 days divided into 20 periods the length of which — excluding the last one — was 14 days.

The intake of the digestible organic matter (DOM) increased along with the concentrate supply by 13.7 % from G1 to G4. Corresponding increments of the estimated net energy (NE) intakes varied according to different systems from 12.1 to 17.3 %. The average daily liveweight gains, however, were very similar (1.10—1.14 kg) in all groups. Because of this discrepancy, the accuracy of any one of the systems could not be very good in all groups.

On the lowest level of concentrate supply (G1) the difference between the NE requirement and intake was within the limits of error only according to the DDR system. Correction of the growth to a 50 % carcass weight did not change the situation. In the case of MAFF and FU the deficits, 15.7 and 11.7 %, respectively, were the largest and increased further when the growth was corrected.

According to MAFF, the difference between the NE requirement and intake was within the limits of error only at the highest level of concentrate supply (G4). When corrected growths were applied with the DDR system, the differences were statistically insignificant in all groups. With ARC, DUTCH and FU, the differences were within the limits of error in three groups independently of the correction. The use of the Gompertz function for the mathematical description of the growth did not change the significancies of the differences.

The NE value of DOM of the rations decreased with increasing share of barley in ARC and DDR, rose in DUTCH and FU and remained constant in MAFF. The changes were small, however, as also were those of the ME values of DOM.

Index words: forages, growing bull, energy standards, feeding standards.

INTRODUCTION

In the first paper of this series (LAMPILA et al. 1988) the accuracy of five feeding standards for growing cattle was compared by feeding growing Ayrshire bulls rations based on different roughages. In the present paper, the same standards (ARC, DDR, DUTCH, FU and MAFF) have been compared with grass silage rations supplemented with different amounts of barley meal. A preliminary report on the results of the feeding experiment has been published by VARVIKKO and LAMPILA (1984).

Owing to the relatively short experimental time and its position relative to the animals' growth phase, some of the equations employed for the mathematical description of the events in our first paper — most commonly the second degree polynomial — were also suitable in the present work. None of them, however, are well applicable for describing longer periods of growth and/or of other parameters. Therefore, comparative calculations were performed in the present work using the Gompertz function and its derivatives according to LEHMANN (1975, 1980 a, b).

EXPERIMENTAL METHODS

Animals and feeding

At the beginning of the preliminary phase of the experiment, 48 Ayrshire bull calves at the mean age of 81 days were divided into two groups of 24 animals each. With the aim of reducing the variation in age, the animals were taken into the study in six lots of eight animals at 2-week intervals. Both groups of 24 calves were fed grass silage *ad libitum* as the only forage and either 1.5 or 2.0 kg of concentrate composed of barley meal supplemented with a small amount of soybean meal. Each lot of animals was kept in this preliminary phase for 70 days. Thus their average age at the beginning of the experiment was 151 days. After this period, each lot of 8 animals was allotted (at 2-week intervals) in pairs to four feeding groups, such that one animal in each pair originated from the lower (1.5 kg) and one from the higher (2.0 kg) level of concentrate supply during the preliminary phase.

The experiment lasted 278 days, divided into 20 periods of 14 days, except for the last period which lasted 12 days. Mainly due to health problems, the original number of 12 animals per group was reduced to 11 during the experiment.

The animals were fed twice daily. Grass silage was offered *ad libitum* and individual intakes were measured. The daily supply of barley meal, as the only concentrate, was constant in three groups: 1.5 (G1), 2.0 (G2), or 3.0 (G3) (air-dry weight) per head per day. In the fourth group (G4), the supply was 50 g per kg $W^{.75}$, the amounts being adjusted according to the liveweights, which were determined fortnightly. The aim was to reach the same total supply in this group, during the course of the experiment, as with the constant daily supply of 3.0 kg, however, the objective was exceeded by about 15 %. Mineral, trace element and vitamin supplements were given to satisfy the estimated requirements. Water was freely available.

Other experimental methods

The animals were weighed at the beginning of the experiment, and thereafter at the end of

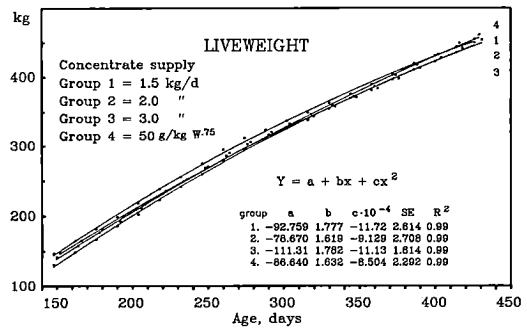


Fig. 1. Mean liveweights as a function of age.

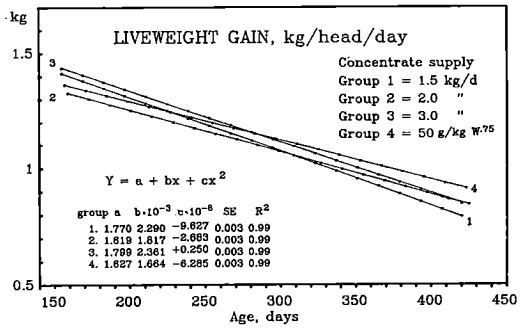


Fig. 2. Mean liveweight gains as a function of age.

each experimental period. For reasons mentioned in our previous paper (LAMPILA et al. 1988) the results of weighings were individually regressed using the same four types of equation. When periodic daily liveweight gains (DLWGs) and their group means were calculated (Fig. 2), individual weights were taken from the equations fitting best to the results of weighings in each case. Weights and weight gains, thus obtained, were also applied when energy requirements and intakes were calculated. Instead, in the description of the liveweights presented in Figs. 1 and 3, group means are based directly on the results of weighings.

When using the Gompertz function, the calculation method was on an individual basis as described above. Parameters of the functions describing the intake of the digestible organic matter (DOM) and of the net energy (NE) according to the DDR system, were calculated for comparative purposes, using equations 20 and 24 presented by LEHMANN (1980 b).

The group means of the total NE requirement and intake were calculated from the results of the integration of the corresponding best-fit equations determined individually, and the significance of the difference between the means was tested by the F-test. Comparative calculations were carried out also on the basis of the individually determined cumulative figures, but the results differed from those of integration only very little. Tukey's test was used when the significance of the differences regarding energy requirement or intake was compared between the different systems.

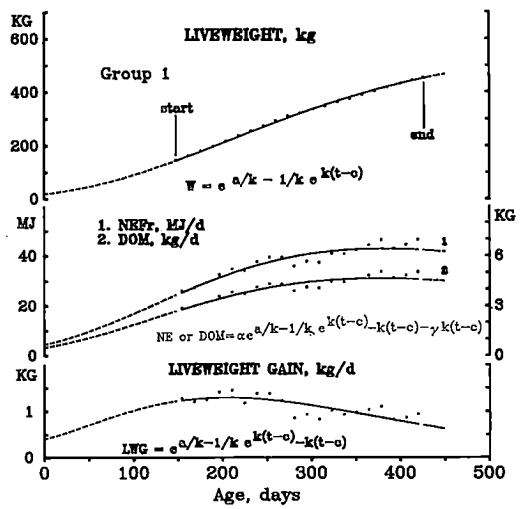


Fig. 3. Means of LW, DLWG and expenditure of NEFr and DOM as calculated according to Gompertz function. Parameter values of the functions are presented in Table 1.

RESULTS AND DISCUSSION

Weights and weight gains

The animals' weights and periodic DLWGs are shown in Figs. 1 and 2, respectively, as the means (dots) of the groups regressed as functions of age. In both cases, the quadratic function was found to fit best to the data.

When corrections of the weights and DLWGs to 50 % carcass weight were made, as described in the first paper, the regression equations for the mean liveweights were:

Group 1:
$y_1 = -92.182 + 1.765\,x - 1.117\,10^{-3}\,x^2$
Group 2:
$y_2 = -78.050 + 1.610\,x - 8.824\,10^{-4}\,x^2$
Group 3:
$y_3 = -108.121 + 1.736\,x - 9.310\,10^{-4}\,x^2$
Group 4:
$y_4 = -85.178 + 1.611\,x - 7.652\,10^{-4}\,x^2$

and those for DLWGs:

Group 1:
$y_1 = 1.883 - 3.005\,10^{-3}\,x - 1.210\,10^{-6}\,x^2$
Group 2:
$y_2 = 1.598 - 1.671\,10^{-3}\,x - 1.670\,10^{-7}\,x^2$
Group 3:
$y_3 = 1.735 - 1.855\,10^{-3}\,x - 1.300\,10^{-8}\,x^2$
Group 4:
$y_4 = 1.611 - 1.524\,10^{-3}\,x - 2.201\,10^{-8}\,x^2$

The regression methods used, to mathematically describe the animals' weight development and to simplify the calculations concerning energy requirement, had a modifying effect on the results. As appears from Fig. 2, the maximum DLWGs occur at the beginning of the experimental time, while, according to Gompertz, the maxima are somewhat later (Fig. 3). The difference is relatively small, however, and did not essentially affect the comparison of the systems, as will be seen later.

Fig. 3 gives the weight and DLWG curves for Group 1, calculated according to the Gompertz function, as well as the periodic variation of the means of the DLWGs (dots) based directly on weighings and used in the calculation of the curves. The figure also illustrates the intakes of DOM and that of NE, calculated according to the DDR system, both regressed according to LEHMANN (1980 b, Eq. 24). Table 1 presents the parameter values for the equations, calculated for all groups and both for original and corrected weights. Values of the constant (c) are means used in the calculation.

When the dates of the maximum DLWGs were calculated, from the equations based on the uncorrected weights, the results varied between 203 and 225 days of age. The maxima

Table 1. Parameter values defining LW and DLWG (a, k, c) according to Gompertz function and the expenditure of total NEFr in MJ/kg gain, and DOM in kg/kg gain, (α, γ), as calculated with "original" and "corrected" liveweights.

Group	Growth parameters				NEFr expenditure			DOM expenditure		
	a	k	c	R^2	α	γ	R^2	α	γ	R^2
					ORIGINAL					
1	.0385	.00604	—643	.94	.8864	—.652	.98	.1035	—.640	.98
2	.0350	.00545	—737	.95	1.1250	—.619	.98	.1309	—.608	.98
3	.0392	.00616	—612	.96	1.2179	—.611	.97	.1455	—.597	.97
4	.0355	.00550	—722	.95	.9565	—.658	.99	.1048	—.660	.99
					CORRECTED					
1	.0377	.00588	—664	.93	.9424	—.639	.97	.1108	—.626	.97
2	.0346	.00537	—751	.94	1.1705	—.611	.97	.1364	—.600	.97
3	.0371	.00572	—669	.91	1.2730	—.596	.91	.1522	—.582	.91
4	.0344	.00529	—756	.93	1.0720	—.634	.98	.1179	—.635	.98

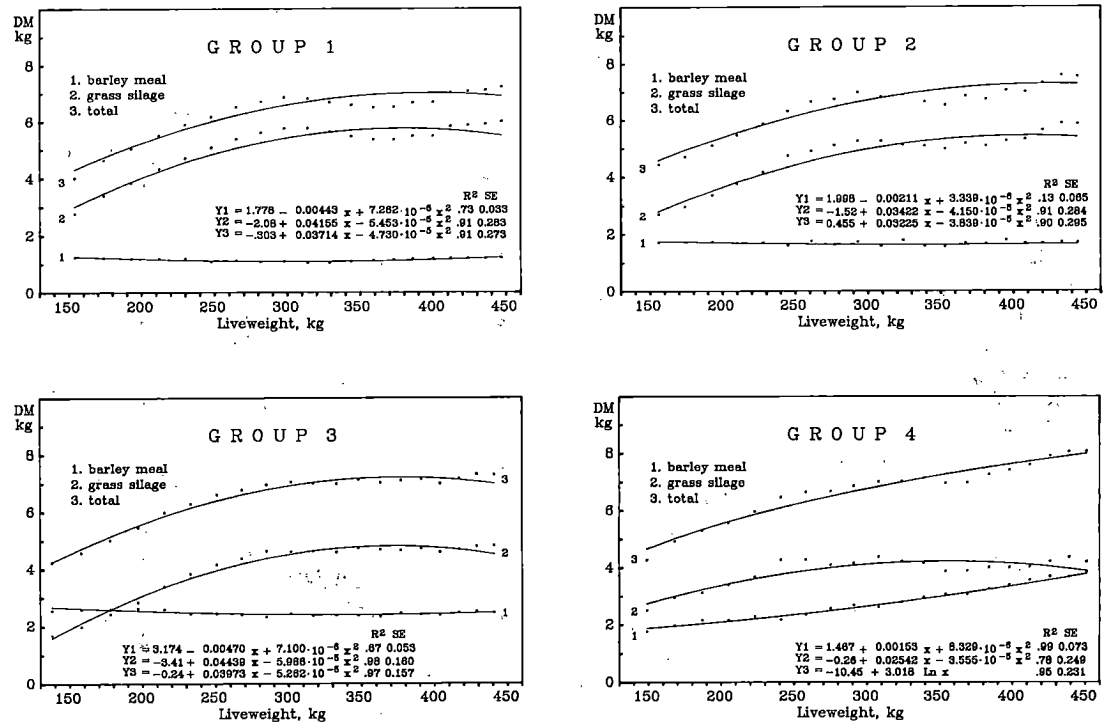


Fig. 4. Dry matter intakes as a function of liveweight.

were thus about 50 to 75 days later than those shown by the quadratic equations. The earliest date occurred in Group 1 and the latest one in Group 4. The notable periodic variation, even of the means of the DLWGs (Fig. 3), gives an expression, however, to the problem concerning exact determination of the "real" growth during short intervals of time, a problem referred to already in our previous paper.

Feed intake

Fig. 4 presents the periodic means of feed intake (dots) regressed as functions of liveweight. The variation occurring in silage intake is worth noticing; because of the restricted supply of concentrate, it is reflected in the total intake of DM. Deviations from the regression lines occurred in different groups, generally to the same direction at the same time, which points to a variation in the palatibility of silage. An explanation for the latter may be found from the fact that the silage fed originated from different lots of preparation for which different silage additives were applied.

Energy requirement and intake

Comparison of the NE intakes with corresponding requirements is presented in Table 2 and Fig. 5. Equations for the curves in Fig. 5 are given in Appendices 1 and 2.

Comparison on the basis of original weights shows that the difference at the lowest level of concentrate supply in Group 1 is within the limits of experimental error only in case of the DDR system. It is true that the difference is small also with ARC and DUTCH while with FU and especially with MAFF the requirement exceeds the intake quite clearly. In the other

Table 2. Means of total NE intake in MJs during the experiment compared with requirement as calculated on the basis of "original" and "corrected" liveweights.

SYSTEM	ARC	DDR	DUTCH	FU	MAFF
			ORIGINAL		
Group 1					
Intake	11231^{b}	10560^{ab}	10035^{a}	10754^{ab}	11074^{b}
Requirement	12000^{b}	10573^{a}	10610^{a}	12181^{b}	13129^{c}
Difference	-769^{s}	27^{NS}	-574^{s}	-1428^{s}	-2055^{s}
Group 2					
Intake	11667^{b}	11034^{ab}	10632^{a}	11355^{b}	11564^{b}
Requirement	11848^{b}	10465^{a}	10443^{a}	12036^{b}	12863^{b}
Difference	-182^{NS}	569^{NS}	190^{NS}	-682^{NS}	-1299^{s}
Group 3					
Intake	12104^{a}	11425^{a}	11204^{a}	12012^{a}	12102^{a}
Requirement	11835^{b}	10442^{a}	10545^{a}	12109^{b}	13047^{b}
Difference	270^{NS}	983^{s}	659^{NS}	-97^{NS}	-945^{s}
Group 4					
Intake	12616^{a}	11880^{a}	11776^{a}	12609^{a}	12668^{a}
Requirement	12077^{abc}	10727^{a}	10837^{ab}	12416^{bc}	13291^{c}
Difference	539^{NS}	1153^{s}	939^{NS}	192^{NS}	-624^{NS}
			CORRECTED		
Group 1					
Intake	11242^{b}	10600^{ab}	9998^{a}	10760^{ab}	11088^{b}
Requirement	12161^{b}	10750^{a}	10822^{a}	12412^{bc}	13391^{c}
Difference	-919^{s}	-150^{NS}	-824^{s}	-1652^{s}	-2303^{s}
Group 2					
Intake	11671^{b}	11034^{ab}	10611^{a}	11357^{b}	11570^{b}
Requirement	11937^{ab}	10567^{a}	10580^{a}	12165^{ab}	13020^{b}
Difference	-266^{NS}	467^{NS}	31^{NS}	-808^{NS}	-1450^{s}
Group 3					
Intake	12132^{b}	11425^{ab}	11094^{a}	12030^{b}	12138^{b}
Requirement	12281^{ab}	10948^{a}	11116^{a}	12759^{b}	13722^{b}
Difference	-148^{NS}	477^{NS}	-22^{NS}	-730^{NS}	-1584^{s}
Group 4					
Intake	12628^{a}	11880^{a}	11723^{a}	12617^{a}	12684^{a}
Requirement	12304^{ab}	10978^{a}	11156^{a}	12745^{ab}	13661^{b}
Difference	324^{NS}	902^{NS}	568^{NS}	-128^{NS}	-977^{NS}

Figures for intake and requirement in the same row without a common superscript letter differ significantly ($P<0.05$). In the case of differences between intake and requirement, S = significant and NS = not significant.

groups, the difference remains statistically insignificant according to three systems. In the case of DDR, the intake exceeds the requirement significantly in Groups 3 and 4, while, according to MAFF, the difference is insignificant only at the highest level of concentrate supply (Group 4).

When calculations are based on corrected weights, statistical significance of the differences changes only in case of the DDR system for Groups 3 and 4, according to which all differences are now within the limits of error. However, in these two groups the intakes still exceed the requirement by 4.4 % and 8.2 %, respectively.

Table 3 presents the requirements and intakes of NE when they are calculated on the basis of weights and corresponding DLWGs derived by regressing original weights using Gompertz function. Compared to the corresponding figures in Table 2, the changes are small and the statistical significancies of the differences

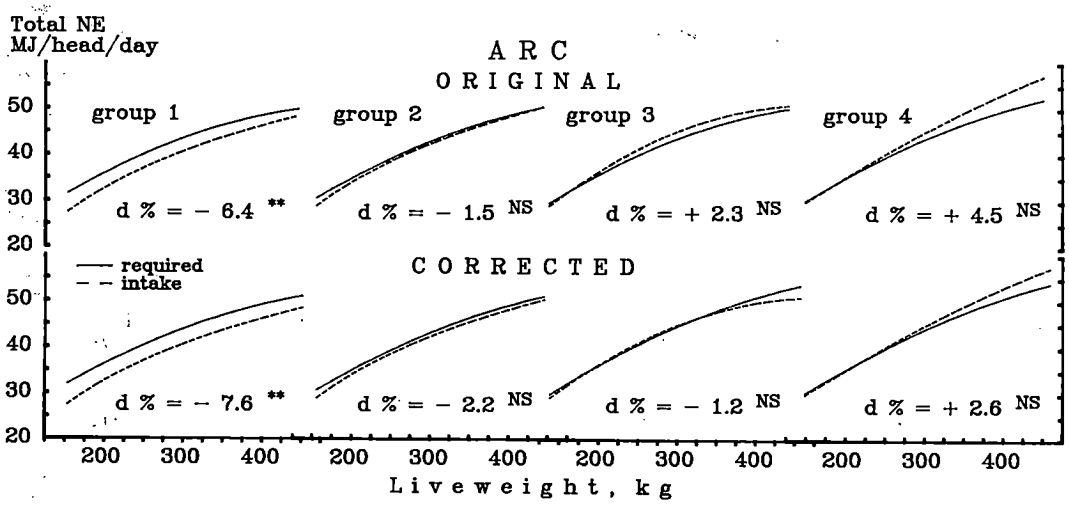
Table 3. Means of total NE intake in MJs during the experiment compared with requirement as calculated on the basis of "original" and "corrected" liveweights, derived from Gompertz function.

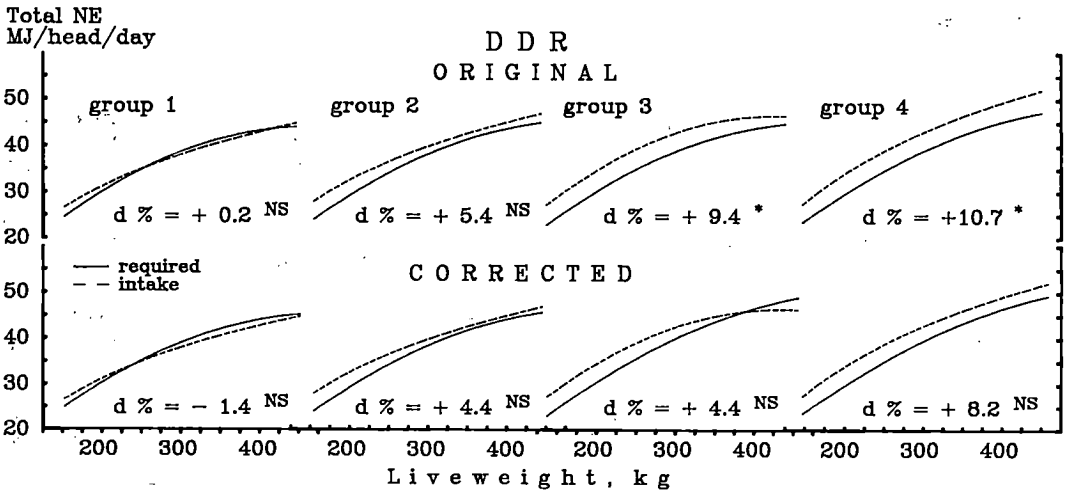
SYSTEM	ARC	DDR	DUTCH	FU	MAFF
			ORIGINAL		
Group 1					
Intake	11227^{b}	10600^{ab}	10020^{a}	10752^{ab}	11071^{b}
Requirement	12004^{b}	10592^{a}	10611^{a}	12212^{b}	13118^{c}
Difference	-777^{s}	8^{NS}	-591^{s}	-1460^{s}	-2047^{s}
Group 2					
Intake	11664^{b}	11034^{ab}	10607^{a}	11353^{b}	11561^{b}
Requirement	11873^{bc}	10500^{ab}	10489^{a}	12087^{c}	12912^{c}
Difference	-209^{NS}	535^{NS}	118^{NS}	-734^{NS}	-1351^{s}
Group 3					
Intake	12102^{a}	11425^{a}	11198^{a}	12010^{a}	12100^{a}
Requirement	11841^{bc}	10449^{a}	10575^{ab}	12125^{c}	13067^{c}
Difference	261^{NS}	976^{s}	623^{NS}	-115^{NS}	-967^{s}
Group 4					
Intake	12613^{a}	11880^{a}	11769^{a}	12607^{a}	12665^{a}
Requirement	12076^{abc}	10731^{a}	10847^{ab}	12422^{bc}	13296^{c}
Difference	537^{NS}	1149^{s}	922^{NS}	185^{NS}	-631^{NS}
			CORRECTED		
Group 1					
Intake	11237^{b}	10600^{ab}	9973^{a}	10758^{b}	11085^{b}
Requirement	12202^{bc}	10818^{a}	10882^{ab}	12506^{c}	13439^{c}
Difference	-965^{s}	-218^{NS}	-909^{s}	-1748^{s}	-2354^{s}
Group 2					
Intake	11668^{b}	11034^{ab}	10584^{a}	11356^{b}	11568^{b}
Requirement	11995^{ab}	10636^{a}	10687^{a}	12262^{ab}	13138^{b}
Difference	-326^{NS}	398^{NS}	-103^{NS}	-906^{NS}	-1570^{s}
Group 3					
Intake	12128^{b}	11425^{ab}	11046^{a}	12027^{b}	12135^{b}
Requirement	12408^{ab}	11096^{a}	11341^{a}	12971^{ab}	13979^{b}
Difference	-280^{NS}	330^{NS}	-295^{NS}	-943^{NS}	-1844^{s}
Group 4					
Intake	12624^{a}	11880^{a}	11697^{a}	12616^{a}	12681^{a}
Requirement	12363^{ab}	11050^{a}	11275^{a}	12848^{ab}	13796^{b}
Difference	261^{NS}	830^{NS}	422^{NS}	-232^{NS}	-1115^{NS}

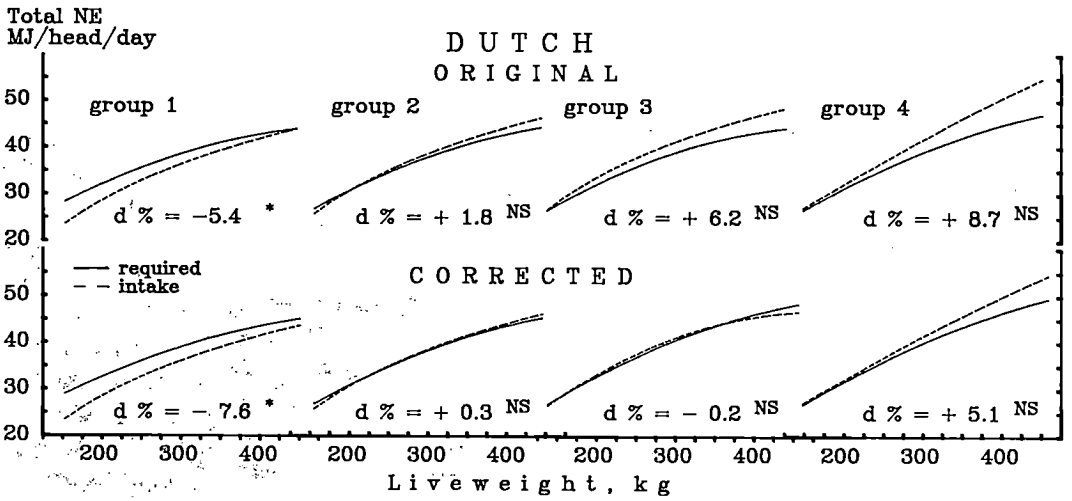
For explanation regarding significancies, see footnote in Table 2.

remain unchanged. As regards the comparison of the systems, both methods applied in the calculation of the progress of weight thus appear to have been of about equal value. The comparison of the requirement and intake curves, illustrated in Fig. 6, to those presented in Fig. 5 shows, however, that the requirement curves based on the Gompertz function are more bent. As a result, the progression of the curves in pair comparison is generally less uniform than that presented in Fig. 5.

The effect of increased concentrate supply on the animals' growth was unexpectedly small. Because of this, the energy requirement calculated, even on the basis of corrected weights, rose from Group 1 to Group 4 by only 1.2—3.1 %, as can be calculated from the figures in Table 2. During the same comparison interval, calculated energy intake rose considerably more, 12.1—17.3 %. When increments in the requirement are subtracted from those of the intake, some 10—14 percent units


Total NE
MJ/head/day
A R C
ORIGINAL
group 1
group 2
group 3
group 4
d % = − 6.4 **
d % = − 1.5 NS
d % = + 2.3 NS
d % = + 4.5 NS
required
intake
C O R R E C T E D
d % = − 7.6 **
d % = − 2.2 NS
d % = − 1.2 NS
d % = + 2.6 NS
50
40
30
20
200 300 400
L i v e w e i g h t , k g


Total NE
MJ/head/day
D D R
ORIGINAL
group 1
group 2
group 3
group 4
d % = + 0.2 NS
d % = + 5.4 NS
d % = + 9.4 *
d % = +10.7 *
required
intake
C O R R E C T E D
d % = − 1.4 NS
d % = + 4.4 NS
d % = + 4.4 NS
d % = + 8.2 NS
50
40
30
20
200 300 400
L i v e w e i g h t , k g


Total NE
MJ/head/day
D U T C H
ORIGINAL
group 1
group 2
group 3
group 4
d % = −5.4 *
d % = + 1.8 NS
d % = + 6.2 NS
d % = + 8.7 NS
required
intake
C O R R E C T E D
d % = − 7.6 *
d % = + 0.3 NS
d % = − 0.2 NS
d % = + 5.1 NS
50
40
30
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200 300 400
L i v e w e i g h t , k g

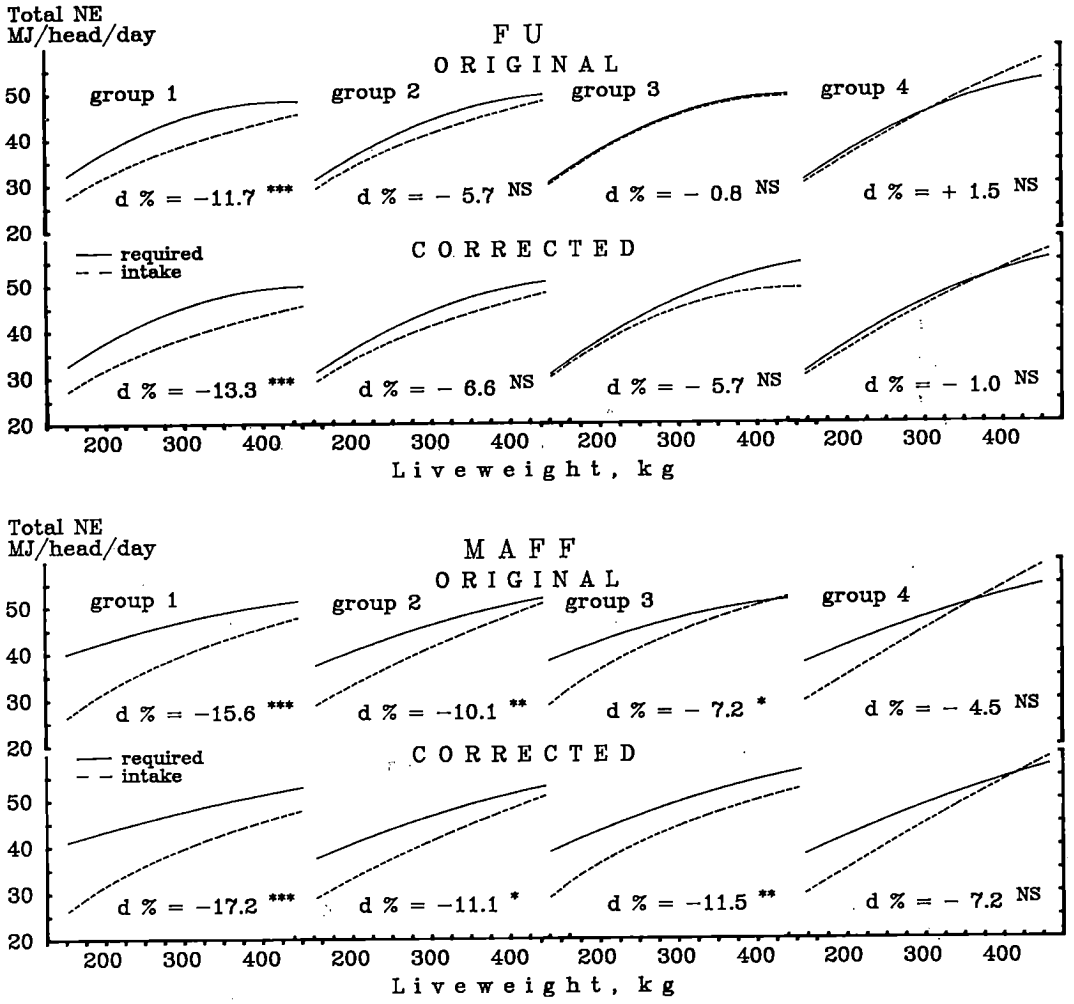
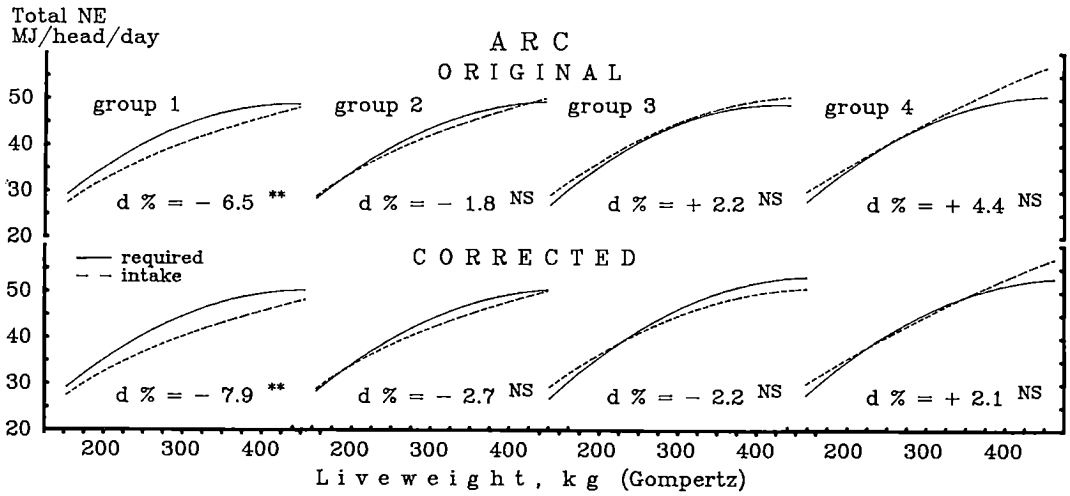


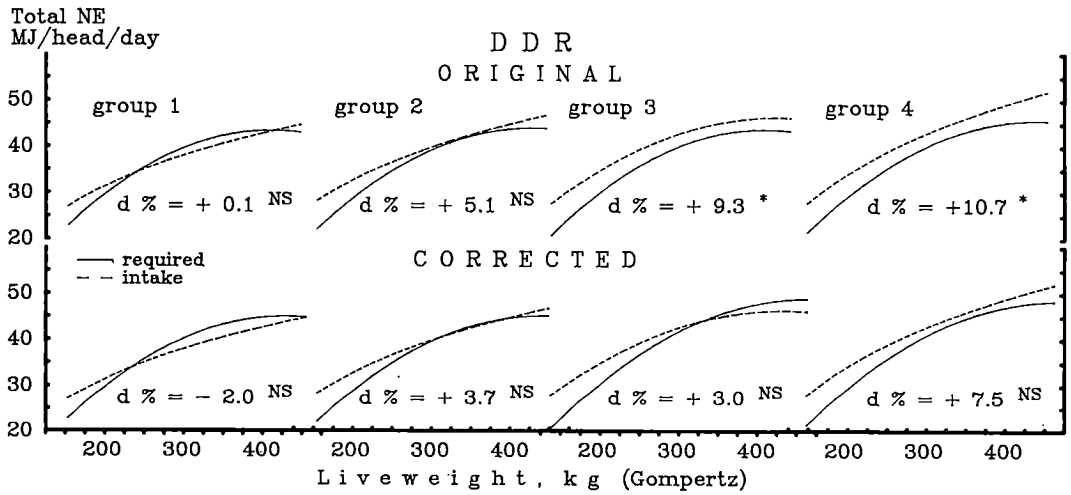
Fig. 5. Comparison of net energy requirement and intake in different experimental groups as calculated according to different systems. Equations of the curves are presented in Appendices 1 and 2.

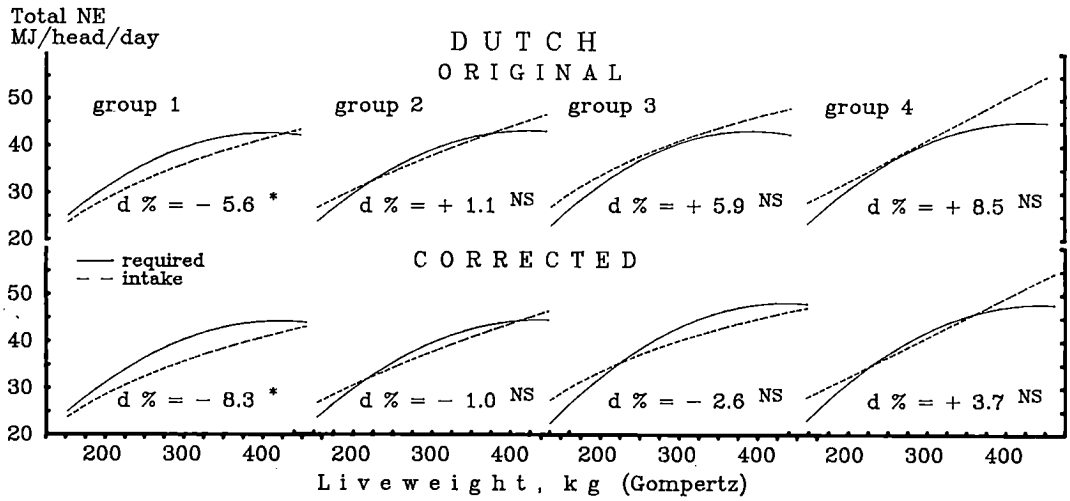
of the latter still remain "unexplained". On account of this, no system could be very exact at all levels of concentrate supply.

The situation might have appeared better if possible changes in energy storage and digestibility of feeds could have been determined. For example, a rise in body energy concentration is indicated by the fact that the mean totals of fats separated from the surface of internal organs and from the abdominal cavity increased, from about 17.5 to 27.8 kg from Group 1 to Group 4, respectively.

Some similarities in the above mentioned elevations of the NE intake can be found among systems but also essential differences, especially when considering the fact that concentrate intake varied within relatively narrow limits. The increases from Group 1 to Group 4 were the lowest in the DDR and ARC systems (12.1 and 12.3 %, respectively) and the highest in the


Total NE
MJ/head/day
A R C
ORIGINAL
group 1
group 2
group 3
group 4
d % = − 6.5 **
d % = − 1.8 NS
d % = + 2.2 NS
d % = + 4.4 NS
required
intake
C O R R E C T E D
d % = − 7.9 **
d % = − 2.7 NS
d % = − 2.2 NS
d % = + 2.1 NS
50
40
30
20
200 300 400
L i v e w e i g h t , k g (Gompertz)


Total NE
MJ/head/day
D D R
ORIGINAL
group 1
group 2
group 3
group 4
d % = + 0.1 NS
d % = + 5.1 NS
d % = + 9.3 *
d % = +10.7 *
required
intake
C O R R E C T E D
d % = − 2.0 NS
d % = + 3.7 NS
d % = + 3.0 NS
d % = + 7.5 NS
50
40
30
20
200 300 400
L i v e w e i g h t , k g (Gompertz)


Total NE
MJ/head/day
D U T C H
ORIGINAL
group 1
group 2
group 3
group 4
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d % = + 1.1 NS
d % = + 5.9 NS
d % = + 8.5 NS
required
intake
C O R R E C T E D
d % = − 8.3 *
d % = − 1.0 NS
d % = − 2.6 NS
d % = + 3.7 NS
50
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200 300 400
L i v e w e i g h t , k g (Gompertz)

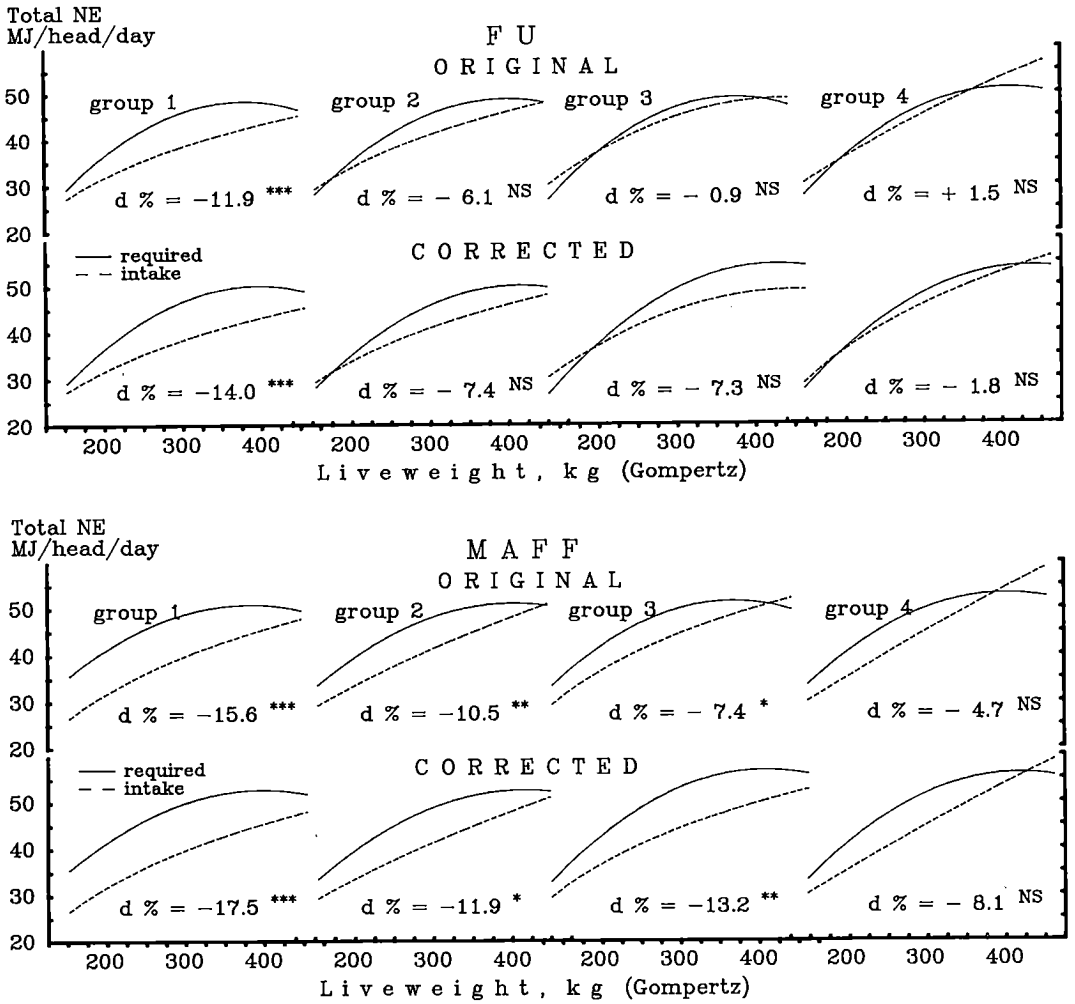

Fig. 6. Comparison of net energy requirement and intake in different experimental groups as calculated according to different systems on the basis of Gompertz function.

DUTCH and FU systems (17.3 and 17.2 %, respectively). An explanation for the difference of the changes may be sought mainly from the NE values calculated for DOM by different systems.

NE and ME values of DOM

Table 4 presents at first the mean daily intakes of DM, organic matter (OM) and DOM. In addition, NE values of DOM are given for DDR and FU systems which allow the calculation separately for both feeds. This is completely true, however, with the FU system only and even then the same share of the energy should be taken for maintenance from both feeds. In case of the DDR system, the correction factor (v = DE/GE) is applied for the whole ration which often results in an elevation of the value of roughage on the cost of concentrate. Due to

Table 4. Mean daily intakes of DM, OM and DOM of barley and grass silage, and the estimated NE values of DOM according to DDR and FU systems.

Group	Feed	DM		OM		DOM		NE, MJ/kg DOM	
		kg/d	%	kg/d	%	kg/d	%	DDR	FU
1	Barley	1.162	18.4	1.128	19.8	0.946	22.7	8.55[a]	10.78[b]
	Silage	5.157	81.6	4.567	80.2	3.216	77.3	9.34[b]	8.86[a]
	Total	6.319		5.695		4.162		9.16[a]	9.29[b]
2	Barley	1.690	26.1	1.641	27.9	1.374	31.5	8.58[a]	10.69[b]
	Silage	4.783	73.9	4.235	72.1	2.983	68.5	9.38[b]	8.79[a]
	Total	6.473		5.876		4.357		9.13[a]	9.38[b]
3	Barley	2.462	37.8	2.391	40.0	2.000	44.1	8.62[a]	10.58[b]
	Silage	4.052	62.2	3.588	60.0	2.530	55.9	9.42[b]	8.71[a]
	Total	6.515		5.979		4.530		9.07[a]	9.53[b]
4	Barley	2.815	41.9	2.736	44.2	2.302	48.6	8.62[a]	10.55[b]
	Silage	3.902	58.1	3.455	55.8	2.431	51.4	9.42[b]	8.67[a]
	Total	6.717		6.190		4.733		9.03[a]	9.59[b]

Means for NE with different superscript letter in the same row differ significantly ($P<0.05$).

a high digestibility of the silage, such an effect was in the present case quite small.

As appears from the figures, the relationship of the NE values is essentially different in these systems. In case of FU, the value of the barley DOM is about 22 % higher than that of silage while, on the contrary, the value of the silage DOM according to DDR exceeds that of barley by about 8—9 %. The main reason for the difference with the former can be found from the value numbers, which favour barley (95 against 80 in this work). The higher value of the silage DOM with the latter is mainly due to the high NE value given to the digestible ether extract (DEE) the share of which in the DOM of silage is relatively high compared to that of barley.

Table 5 presents the NE and ME values of DOM as means for groups and whole rations. Standard deviations (S.D.) are calculated from the periodic group means. In case of the FU system, no equation is available for the calculation of the ME values.

It may be noticed at first that the mean intake of DOM increases from Group 1 to Group 4 along with the increasing supply of concentrate by 13.7 %. That is slightly more than corresponding increments in the intake of NE (Table 2) according to DDR and ARC (about 12.3 %). It was thus to be expected that the NE value of DOM decreases with increasing share of concentrate in the ration. Contrary to these systems, the NE intake according to DUTCH and FU increases along with the share of concentrate definitely more (about 17.3 %) than the intake of DOM. The main reason for that with FU appears to be the high rating of concentrate compared to silage, as discussed above. With DUTCH the factors resulting in practically the same end appear, however, to be more complex in nature.

Generally the mean NE values of DOM are relatively stable within each system, as is especially the case with MAFF. Standard deviations of the means indicate, however, as also in DUTCH and ARC, that the variation was relatively wide during the experimental time. This appears to be mainly due to the changes in the ratio of the energy requirements for maintenance and growth. The NE value of ME and — accordingly — of DOM are rated in these three systems so much different in maintenance and growth that relatively small changes in the said ratio may result in noticeable changes in the NE value of DOM.

In case of the DDR system the said effect is

Table 5. Mean daily intake of DOM and the estimated NE and ME values of DOM according to different systems.

GROUP	DOM	NE, MJ per kg DOM					ME, MJ per kg DOM			
		ARC	DDR	DUTCH	FU	MAFF	ARC	DDR	DUTCH	MAFF
1 MEAN	4.162	9.71^{e}	9.16^{b}	8.68^{a}	9.29^{c}	9.57^{d}	16.05^{c}	16.52^{d}	15.39^{a}	15.95^{b}
S.D.	.590	.25	.13	.33	.08	.33	.09	.11	.07	.03
2 MEAN	4.357	9.64^{d}	9.13^{b}	8.80^{a}	9.38^{c}	9.56^{d}	16.00^{b}	16.44^{c}	15.37^{a}	15.96^{b}
S.D.	.624	.21	.12	.25	.06	.28	.09	.10	.06	.04
3 MEAN	4.530	9.61^{c}	9.07^{b}	8.90^{a}	9.53^{c}	9.61^{c}	15.90^{b}	16.32^{d}	15.33^{a}	15.98^{c}
S.D.	.653	.21	.09	.24	.10	.28	.08	.10	.06	.03
4 MEAN	4.733	9.59^{b}	9.03^{a}	8.95^{a}	9.59^{b}	9.63^{b}	15.86^{b}	16.26^{d}	15.30^{a}	15.98^{c}
S.D.	.835	.17	.07	.30	.11	.28	.09	.08	.05	.02

Means with different superscript letter in the same row differ significantly ($P<0.05$).

excluded because the same energy unit (NEFr) is applied for both requirements. This is an apparent reason for the low S.D. values of the means. Together with the small differences between the means they suggest that DOM could have been used approximately as well as energy units for the expression of the animals' requirements. The same opinion was reached when comparison of the DOM and NEFr intakes was carried out using Gompertz function, as illustrated in Fig. 3, and parameter values calculated for original weights (Table 1).

High stability of the ME values of DOM within each system (Table 5) is a further indication towards the same direction in the case of systems expressing the needs as metabolizable energy or starting calculations from it. This is not to say, however, that DOM would be the best possible basis for the simplification or unification of the various calculation systems which often end up at approximately similar results.

REFERENCES

LAMPILA, M., MICORDIA, A. & VÄÄTÄINEN, H. 1988. Comparison of energy feeding standards for growing cattle. 1. Rations based on different forages. Ann. Agric. Fenn. 27: 247—258.

LEHMANN, R. 1975. Matematische Grundlagen zur Analyse des Wachstums von landwirtschaftlichen Nutztieren. 1. Aufgabenstellung und Problemanalyse. Arch. Tierzucht 18: 163—174.

— 1980 a. Anwendung eines Wachstumsmodells in der Tierernährung. 1. Matematische Grundlagen. Arch. Tierernährung 30: 427—435.

— 1980 b. Anwendung einer Wachstumsmodells in der Tierernährung. 2. Energiebedarf und Energieaufwand. Arch. Tierernährung 30: 437—446.

VARVIKKO, T. & LAMPILA, M. 1984. Väkirehutason vaikutus lihamullien kasvu- ja teurastulokseen säilörehuruokinnalla. Koetoim. ja Käyt. 3.7.1984.

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SELOSTUS

Energianormistojen vertailu kasvavien ayrshire-sonnien ruokintakokein

2. Eri väkirehutasot säilörehudieeteissä

MARTTI LAMPILA ja ANGEL MICORDIA

Maatalouden tutkimuskeskus

Kirjoitus on jatkoa sarjassa, jossa kasvavien ay-sonnien energian saantia ja normin mukaista tarvetta verrataan keskenään laskien ne viiden järjestelmän mukaan. Järjestelmät on esitelty sarjan ensimmäisessä osassa. Tässä esitetyt tulokset perustuvat kokeeseen, johon noin 151 päivän keski-iässä otettuja ay-sonnivasikoita ruokittiin vapaaseen säilörehun tarjontaan perustuvilla dieeteillä.

Koe käsitti neljä 11 eläimen ryhmää, joissa energian saantia täydennettiin 1.5 (G1), 2.0 (G2) tai 3.0 (G3) ohrakilon vakinaisilla päiväannoksilla tai metabolisen painon perusteella nousevan annostuksen (50 g per kg $W^{0.75}$) mukaisesti (G4). Koeaika oli 278 päivää jaettuna kahteenkymmeneen jaksoon, jotka viimeistä lukuunottamatta olivat 14 päivän pituisia.

Keskimääräinen päiväkasvu oli eri ryhmissä likimäärin samansuuruinen (1.10—1.14 kg ryhmästä G1 ryhmään G4). Kun väkirehun annostuksen myötä noin 13.7 % kohonnut (G1→G4) sulavan orgaanisen aineen (DOM) saanti ei juuri näkynyt kasvussa, ei minkään normiston tarkkuus voinut olla erityisen hyvä kaikissa ryhmissä.

Alimmalla väkirehun annostustasolla (G1) energian saannin poikkeama lasketusta tarpeesta sopi virherajojen puitteisiin vain DDR:n järjestelmässä eikä tilanne muuttunut, vaikka kasvu korjattiin 50 prosentin teuraspainon mukaiseksi. Vajaus normitarpeeseen verraten oli erityisen suuri MAFF- ja FU-järjestelmissä, 15.7 ja 11.7 % vastaavasti, ja suureni painokorjauksen vaikutuksesta.

MAFF-järjestelmässä tarpeen ja saannin välinen ero sopi virherajojen puitteisiin vain eniten väkirehua saaneessa ryhmässä (G4). Korjatuilla painoilla DDR:n järjestelmän mukaan laskien olivat erot virherajojen sisällä kaikissa ryhmissä, muilla kolmessa ryhmässä (ARC, DUTCH, FU), kuten myös ilman sanottua korjausta. Kun vertailut tehtiin käyttäen Gompertz-funktion mukaan laskettuja eläinten painoja ja kasvuja, erojen merkitsevyydet säilyivät ennallaan. Tarve- ja saantikäyrien kulku oli tällöin kuitenkin vähemmän yhdenmukainen kuin käytettäessä laskentaperustana toisen vaihtoehdon mukaista painonkehitystä.

Rehuannosten sulavan orgaanisen aineen (DOM) keskimääräinen nettoenergia-arvo aleni ohran osuuden kohotessa ARC- ja DDR-järjestelmässä, kohosi DUTCH- ja FU-järjestelmässä ja pysyi vakinaisena MAFF:ssa. Muutokset, jotka johtuivat säilörehun ja ohran DOM:n keskinäisen arvostuksen eroista, olivat kuitenkin pieniä. Samoin olivat myös muutokset ilmaistaessa DOM:n arvo muuntokelpoisena energiana.

Appendix 1. Net energy intake and requirement as best-fit second degree polynomial ($NE = a + b \cdot W + c \cdot W^2$), logarithmic ($NE = a + b \cdot LnW$) or power ($NE = a \cdot W^b$) functions of "original" liveweights calculated according to different systems.

Group/ System	REQUIREMENT				INTAKE				
	a	$b \cdot 10^{-2}$	$c \cdot 10^{-4}$	SE	a	$b \cdot 10^{-2}$	$c \cdot 10^{-4}$	R^2	SE
Group 1									
ARC	12.874	13.941	—1.268	0.028	—71.690	1965.1	log.	.988	0.717
DDR	0.512	18.802	—2.021	0.372	—59.232	1706.8	log.	.976	0.873
DUTCH	12.181	12.081	—1.124	0.080	—72.786	1909.3	log.	.981	0.882
FU	8.904	18.367	—2.140	0.063	—59.360	1718.7	log.	.974	0.934
MAFF	29.707	7.698	—0.628	0.011	—75.358	2019.7	log.	.986	0.779
Group 2									
ARC	11.905	13.704	—1.137	0.020	—75.030	2055.8	log.	.979	0.991
DDR	0.572	17.885	—1.773	0.333	—63.125	1807.1	log.	.967	1.093
DUTCH	9.483	12.797	—1.119	0.074	—73.912	1971.3	log.	.965	1.223
FU	9.080	16.850	—1.740	0.046	—62.566	1816.8	log.	.962	1.169
MAFF	24.909	9.236	—0.704	0.009	1.946	53.567	pow.	.977	1.062
Group 3									
ARC	12.226	14.347	—1.305	0.019	7.076	18.546	—1.962	.987	0.815
DDR	1.727	17.953	—1.846	0.384	4.542	19.727	—2.312	.986	0.757
DUTCH	10.260	13.538	—1.328	0.075	—66.309	1884.4	log.	.981	0.947
FU	9.273	17.976	—2.010	0.085	8.425	18.392	—2.085	.985	0.754
MAFF	27.177	9.394	—0.869	0.022	—69.202	1992.6	log.	.987	0.821
Group 4									
ARC	12.224	13.572	—1.039	0.037	11.831	13.069	—0.665	.991	0.828
DDR	1.626	17.110	—1.538	0.336	—83.884	2226.7	log.	.988	0.845
DUTCH	9.933	12.659	—0.976	0.062	10.849	11.414	—0.369	.988	0.964
FU	9.763	16.380	—1.520	0.019	11.907	13.202	—0.714	.987	0.977
MAFF	25.860	8.980	—0.565	0.006	13.396	11.345	—0.285	.992	0.820

Appendix 2. Net energy intake and requirement as best-fit second degree polynomial ($NE = a + b \cdot W + c \cdot W^2$), logarithmic ($NE = a + b \cdot LnW$) or power ($NE = a \cdot W^b$) functions of "corrected" liveweights calculated according to different systems.

Group/ System	REQUIREMENT				INTAKE				
	a	$b \cdot 10^{-2}$	$c \cdot 10^{-4}$	SE	a	$b \cdot 10^{-2}$	$c \cdot 10^{-4}$	R^2	SE
Group 1									
ARC	14.260	13.105	—1.106	0.030	—70.864	1949.3	log.	.988	0.707
DDR	1.883	17.845	—1.816	0.370	—58.162	1686.4	log.	.977	0.865
DUTCH	13.979	11.087	—0.936	0.075	—71.049	1874.5	log.	.981	0.861
FU	10.982	16.986	—1.855	0.041	—58.554	1703.2	log.	.974	0.923
MAFF	32.569	6.158	—0.361	0.042	—74.639	2005.9	log.	.987	0.772
Group 2									
ARC	11.967	13.604	—1.087	0.024	—74.675	2048.8	log.	.979	0.991
DDR	0.581	17.783	—1.712	0.313	—62.647	1797.9	log.	.966	1.093
DUTCH	9.443	12.753	—1.059	0.074	—72.656	1947.2	log.	.965	1.212
FU	9.346	16.528	—1.628	0.036	—62.185	1809.5	log.	.962	1.169
MAFF	24.776	9.286	—0.656	0.007	1.962	53.406	pow.	.977	1.061
Group 3									
ARC	12.475	14.008	—1.102	0.035	7.766	17.920	—1.856	.987	0.819
DDR	2.492	16.995	—1.486	0.360	5.363	18.973	—2.182	.985	0.768
DUTCH	10.271	13.395	—1.111	0.077	6.898	16.492	—1.708	.978	0.965
FU	10.169	16.892	—1.568	0.039	9.266	17.633	—1.951	.985	0.779
MAFF	26.868	9.628	—0.692	0.006	—67.769	1963.6	log.	.987	0.826
Group 4									
ARC	12.213	13.516	—0.960	0.042	11.900	13.052	—0.684	.991	0.825
DDR	1.755	16.854	—1.405	0.332	—82.349	2196.5	log.	.988	0.829
DUTCH	9.661	12.821	—0.904	0.063	10.830	11.510	—0.437	.988	0.959
FU	10.058	15.968	—1.329	0.009	12.006	13.160	—0.730	.987	0.978
MAFF	25.418	9.348	—0.520	0.008	13.353	11.426	—0.325	.992	0.819

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THE EFFECT OF UREA TREATMENT AND AMMONIATION ON THE NUTRITIVE VALUE OF OAT STRAW

ILMO ARONEN

ARONEN, I. 1990. **The effect of urea treatment and ammoniation on the nutritive value of oat straw.** Ann. Agric. Fenn. 29: 107—112. (Agric. Res. Centre, Inst. Anim. Prod., SF-31600 Jokioinen, Finland.)

A 4 × 4 Latin square digestion trial was conducted with four growing full-sib Ayrshire cattle, produced by embryo-transfer technique, to determine the effects of ammonia treatment (A) and urea treatment with two levels of urea, either low (LU) or high (HU), on digestion of oat straw in comparison to field dried control straw (C). The effects on voluntary feed intake were determined in four wethers.

Except for crude protein (CP), urea treatment had no statistically significant effect on the apparent digestibility of the dietary constituents of oat straw. Ammonia treatment increased the apparent digestibility of organic matter, CP and also crude fibre.

Due to lack of response in the digestibility of nutrients, the calculated feed value of LU and HU was improved only slightly and nonsignificantly compared to C. Instead, the feed value of A was 14 % higher than that of C.

Ammonia treatment had a positive effect on straw dry matter intake, while urea treatment only tended to improve intake. Both A, HU and LU efficiently preserved wet oat straw.

The factors influencing the effects of urea and ammonia treatment on digestion and voluntary intake are discussed.

Index words: ammonia treatment, urea treatment, straw.

INTRODUCTION

In practical farming ammoniatreated straw has been widely used as a roughage for ruminants. However, increasing interest has been focused on urea treatment because of its low costs and convenience. Recently, extensive research was carried out at the Agricultural Research Centre to evaluate the treatment of straw with ammonia, urea or a urea—ureaphosphate mixture (ALASPÄÄ 1986). The feed values in that experiment, however, were based on digestibility coefficients determined in wethers. The purpose of the present experiment was to study the effect of ammonia treatment and urea treatment with two levels of urea application on the digestion of oat straw in the diets of growing cattle and on the voluntary intake of wethers. Unlike in many previous experiments, round bales were used.

MATERIAL AND METHODS

Treatment of straw

Untreated oat straw (C) was dried (dry matter (DM) content 846 g/kg) on the field and baled with a round-baler. Urea treatment with fertilizer grade urea in liquid form was carried out in connection with baling. Urea solution was prepared according to ALASPÄÄ (1986). Two levels of urea were used. The high level of urea (HU) was intended to match the recommendation for urea treatment of straw in Finland (3.5—4.4 % urea of wet straw (DM 500—750 g/kg) corresponding 56—70 g urea/kg straw DM). The inclusion rate actually achieved averaged 46 g urea/kg DM. Concentration of urea at the low level of urea treatment (LU) was in an average 25 g urea/kg DM.

Baling and treatment of straw was carried out on the 1st of September 1988, the mean ambient temperature and humidity being 12.2 °C and 95 %, respectively. After baling, the urea treated bales were placed into plastic bags and the air in the bags partly evacuated.

Ammonia treatment was conducted by injecting anhydrous ammonia at the rate of 42.1 g/kg DM into the round bales in the plastic bags.

All the bales were stored in an unheated barn until feeding. The first bales were opened at approximately 150 days post-treatment. Mean ambient temperatures in September, October, November and December were 10.8, 4.2, —3.8 and —6.9 °C, respectively.

Digestibility and voluntary intake measurements

Four growing full-sib cattle of the Finnish Ayrshire breed, produced by embryo transfer, were assigned to treatments in a 4 × 4 Latin square arrangement. The treatments were the diets containing C, LU, HU or A. Each period of the trial consisted of a 14-day adjustment phase and a 7-day collection phase during which digestibility was estimated from total collection of feaces.

The animals were of a mean initial live weight of 326 kg reaching a mean final live weight of 370 kg. They were housed in individual metabolism crates with free access to water. During the collection phase the bulls were offered 35 g of straw DM and 35 g of barley DM per kg of metabolic live weight (kg $W^{0.75}$). In order to achieve isonitrogenous diets, C- and LU-diets were supplemented with 100 g and 50 g of urea, respectively.

Feed value, based on the digestibility coefficients determined in the present experiment, was calculated according to SALO et al. (1982) using a factor of 0.42 to correct the calculated energy content of digestible nutrients.

Ad libitum intake of the four different straw was determined in wethers in another trial of 4 × 4 Latin square design. In addition to straw, 300 g of barley meal was included in each diet and sufficient amounts of urea were included in the diets C and LU to achieve isonitrogenous conditions.

Chemical analyses

Proximate feed analyses were performed conventionally. Dry matter content was determined according to ALASPÄÄ (1986). Total and water soluble nitrogen were determined from fresh samples using the Kjeldahl method and ammonium nitrogen was determined colorimetrically (Mc CULLOUGH 1967).

Statistical analyses

The GLM procedure of the Statistical Analysis System (SAS 1987) was used for statistical analyses of digestibility and intake data for a Latin square design. The model included diet, animal and period. Differences among treatment means were tested using Tukey's test.

RESULTS

Chemical composition

Chemical composition of oat straw treated with different methods is summarized in Table 1. Both of the urea treatments and the ammonia treatment elevated the crude protein (CP) content of straw, the increase in CP content being most prominent with treatment A and lowest with treatment LU and intermediate with treatment HU. Also the proportion of NH_3-N and soluble N of the total N was highest in A and lowest in C.

The ash content in untreated straw was slightly higher than in the treated ones merely reflecting the difficulty of obtaining similar batches of straw in field conditions. However, there were no significant differences in contents of crude fiber (CF) and ether extract (EE) between the treatments.

Digestibility, feed value and voluntary feed intake

Digestibility of nutrients is reported in Table 2. Except for CP, urea treatment had no statistically significant effect on apparent digestibility of dietary constituents of oat straw. CF digestibility tended to increase only slightly. Instead, ammonia treatment increased the appar-

Table 1. Chemical composition of untreated oat straw (C), oat straw treated with ammonia (A) and with a low level (LU) or high level of urea (HU).

	C	LU	HU	A
Dry matter (DM), g/kg	845^{b}	673^{a}	674^{a}	658^{a}
In DM, g/kg				
Ash	68^{b}	62^{a}	60^{a}	58^{a}
Crude protein (CP)	74^{a}	105^{b}	136^{c}	158^{d}
Ether extract (EE)	16	16	17	16
Crude fiber (CF)	429	430	414	435
Nitrogen free extract (NFE)	413^{c}	386^{b}	372^{b}	332^{a}
pH	7.1^{a}	8.1^{ab}	8.5^{b}	8.7^{b}
Nitrogen, %				
Total	1.18^{a}	1.67^{ab}	2.28^{bc}	2.50^{c}
Soluble	0.42^{a}	0.81^{ab}	1.39^{b}	1.50^{b}
Ammonia	0.04^{a}	0.26^{b}	0.55^{c}	0.62^{c}

Means in the same row without the same superscript are significantly different ($P<0.05$).

Table 2. Digestion coefficients (%) of nutrients and calculated feed value of untreated and treated oat straw.

	C	LU	HU	A	SEM
OM	52.2^{a}	53.3^{a}	55.4^{ab}	60.1^{b}	1.28
CP	26.8^{a}	31.6^{ab}	45.1^{b}	47.4^{b}	3.86
EE	45.4	46.6	53.9	41.0	3.98
CF	66.8^{a}	68.6^{a}	69.3^{a}	80.0^{b}	0.95
NFE	41.6	42.6	43.3	40.5	2.40
Feed value,					
kg DM/FFU[1]	3.42^{b}	3.30^{ab}	3.18^{ab}	2.94^{a}	0.08
g DCP/FFU	69.1^{a}	108.9^{ab}	198.0^{bc}	223.6^{c}	2.44

Means in the same row without the same superscript are significantly different ($P<0.05$).
[1] FFU = fattening feed unit (= 0.7 kg Starch Equivalents).

ent digestibility of organic matter (OM), CP and also crude fiber (CF) ($P < 0.05$).

Due to lack of response in digestibility of nutrients, the feed value of urea-treated straw was improved only slightly and non-significantly, whereas the increase in feed value of ammonia treated straw was 14 %. HU treatment increased the digestible crude protein content in the feed unit (DCP/FFU) almost three-fold.

Voluntary feed intake of straw is presented in Table 3. Ammonia treatment increased significantly ($P < 0.05$) straw DM intake, but urea treatment only tended to improve it ($P > 0.05$).

Table 3. Effect of ammonia and urea treatment of straw on voluntary feed intake determined in wethers.

	C	LU	HU	A	SEM
Intake, g/d	646[a]	700[ab]	714[ab]	879[b]	45.9

Means in the same row without the same superscript are significantly different ($P < 0.05$).

DISCUSSION

In urea treatment the urea is hydrolysed to ammonia and the liberated ammonia prevents not only the growth of fungi but also that of undesirable bacteria, such as *Clostridia* (BLOCK et al. 1988). Indeed, in this experiment the overall quality of all of the straws was good and the smell of ammonia, also in LU-treated straw, was strong. However, it must be acknowledged that the cold weather itself may have prohibited mold growth, which was also the case in the experiment by ALASPÄÄ (1986). In her experiment straw quality was good during the winter and early spring but mold growth became apparent during the summer months.

If the urea is hydrolysed rapidly and a high concentration of ammonia is reached, an increase in the digestibility of nutrients can be expected (BLOCK et al. 1988). However, it has been shown, that the concentration of ammonia that can be generated from urea is related to several factors, the moisture content of the straw, level of urea, storage time and mean ambient temperature the last one being the most important (WILLIAMS et al. 1984, BLOCK et al. 1988, HADJIPANAYIOTOU 1988).

In this experiment, the DM content of the ammonia or urea treated straw during baling varied between 62 and 73 %. In regard to urea treatment, this is in the range of the practical recommendation (50—75 %) but is somewhat lower than that found to be optimal in laboratory conditions. BLOCK et al. (1988) found maximal digestibility improvement at a water content of 50—60 % and WILLIAMS et al. (1984) recorded 100 % hydrolysis of urea when the straw DM was 45 %. Increased water content has enhanced also ammoniation at low temperatures (MANDELL et al. 1988).

At HU treatment the level of urea inclusion averaged 4.6 % on a dry matter basis and 3.0 % on a wet basis. These values are lower than the practical recommendation in Finland (5.5—7.0 % on a dry matter basis), but well in the range of recommendations for practice in the German Democratic Republic (3.0 to 3.5 % of wet straw, BLOCK et al. 1988). The optimum urea dosage on a DM basis was reported to be 8 % at a water content of 50—60 % (BLOCK et al. 1988). On the other hand, an inclusion level higher than 4 % (on a wet basis) has been questionned (HADJIPANAYIOTOU 1988).

Another reason for the only modest changes seen in the digestibility of nutrients in response to urea treatment in this experiment may have been the rather low mean ambient temperature. The effect of the ambient temperature on urea hydrolysis and further that on nutrient digestibility has been well demonstrated by BLOCK et

al., 1988. In tropical (SAADULLAH et al. 1981) and subtropical conditions (HADJIPANAYIOTOU 1982) urea treatment has increased the digestibility of straw to the same extent as that usually found in experiments with ammonia.

In general, urea treatment, also at the higher level of application, failed to show any significant improvement in digestibility and feed value. These results are in agreement with those of WANAPAT et al. (1985) and ALASPÄÄ (1986). Contrary to the present study, a significant response in digestibility to urea treatment has been reported by HADJIPANAYIOTOU (1982) and BLOCK et al. (1988). Contradictions can, at least partly, be explained by differences in the level of urea dosage, DM content of the straw and especially the ambient temperature as stated above.

In agreement with WANAPAT et al. (1985), ALASPÄÄ (1986) and MANDELL et al. (1988) ammonia treatment in this experiment significantly increased crude fiber digestion and an improvement in feed value was also recorded. Especially the hemicellulose component of the straw cell wall is solubilized by ammonia treatment (DRYDEN and LENG 1988, GIVENS et al. 1988).

Enhanced digestibility of straw A increased voluntary intake. This is in agreement with the observations of ORSKOV et al. (1983) and MANDELL et al. (1988) but in disagreement with those of ALASPÄÄ (1986). Compared to C the intake of LU or HU was not significantly higher. It has to be recognised, that in diet C a small amount of urea was included in order to obtain isonitrogenous diets. The urea inclusion in it may have enhanced straw digestion, as was the case in the experiment of WANAPAT et al. (1985), and thereby also DM intake.

The utilization of treated straw depends on the level of feeding and the proportion of straw in the diet (SUNDSTOL and COXWORTH 1984). Indeed, urea or ammonia treated straw has been used for dry beef cows also in Finland. In order to further improve the utilization of cereal straw as a ruminant feed by urea treatment, the effects on digestion and DM intake of straw and level of urea dosage in relation to ambient temperature should be further studied in Finnish conditions.

REFERENCES

ALASPÄÄ, M. 1986. Treatment of straw with ammonia, urea or a urea + ureaphosphate mixture: effect on dietary intake and growth of young ayrshire bulls raised for beef. Ann. Agric. Fenn. 25: 91—97.

BLOCK, H.-J., WEISSBACH, F. & SCHMIDT, L. 1989. Improving the feeding value of straw by urea treatment. Symposium on developments in the use of new and non-conventional feeds in ruminant nutrition (Geneva, 16—20 January 1989). Economic Commission for Europe.

DRYDEN, G. McL. & LENG, R.A. 1988. Effects of ammonia and sulphur dioxide gases on the composition and digestion of barley straw. Anim. Feed Sci. Technol. 19: 121—133.

GIVENS, D.I., ADAMSON, A.H. & COBBY, J.M. 1988. The effect of ammoniation on the nutritive value of wheat, barley and oat straws. II. Digestibility and energy value measurements *in vivo* and their prediction from laboratory measurements. Anim. Feed Sci. Technol. 19: 173—184.

HADJIPANAYIOTOU, M. 1982. The effect of ammoniation using urea on the intake and nutritive value of chopped barley straw. Grass Forage Sci. 37: 89—93.

— 1989. Nutritional and economic aspects of the use of urea-treated barley straw in diets of lactating friesian cows and growing heifers. Symposium on developments in the use of new and non-conventional feeds in ruminant nutrition (Geneva, 16—20 January 1989). Economic Commission for Europe.

MANDELL, I.B., CHRISTISON, G.I., NICHOLSON, H.H. & COXWORTH, E.C. 1988. The effect of variation in the water content of wheat straw before ammoniation on its nutritive value for beef cattle. Anim. Feed Sci. Technol. 20: 111—124.

MCCULLOUGH, H. 1967. The determination of ammonia in whole blood by a direct colorimetric method. Clin. Chim. Acta 17: 297—304.

ORSKOV, E.R., REID, G.W., HOLLAND, S.M., TAIT, C.A.G. & LEE, N.H. 1983. The feeding value for ruminants of straw and whole-crop barley and oats treated with

anhydrous or aqueous ammonia or urea. Anim. Feed Sci. Technol. 8: 247—257.

SAADULLAH, M., HAQUE, M. & DOLBERG, F. 1981. Effectiveness of ammonification through urea in improving the feeding value of rice straw in ruminants. Trop. Anim. Prod. 6: 30—36.

Statistical Analysis System (SAS). 1987. SAS/STAT User's Guide for Personal Computers. SAS Institute Inc., Carry, N.C., U.S.A.

SUNDSTOL, F. & OWEN, E. 1984. Ammonia treatment. Straw and other fibrous by-products as feed. Chapter 7: 196—247. Elsevier.

WANAPAT, M., SUNDSTOL, F. & GARMO, T.H. 1985. A comparison of alkali treatment methods to improve the nutritive value of straw. I. Digestibility and metabolizability. Anim. Feed Sci. Technol. 12: 295—309.

WILLIAMS, P.E.V., INNES, G.M. & BREWER, A. 1984. Ammonia treatment of straw via the hydrolysis of urea. I. Effects of dry matter and urea concentrations on the rate of hydrolysis of urea. Anim. Feed Sci. Technol. 11: 103—113.

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SELOSTUS

Ammoniakki- ja ureakäsittelyn vaikutus kauran oljen ravintoarvoon

ILMO ARONEN

Maatalouden tutkimuskeskus

Kauran olki käsiteltiin ammoniakilla (A), ja kahdella urean käsittelytasolla, joista korkeammalla (HU) pyrittiin suositusten mukaiseen tasoon ja alemmalla (LU) puoleen edellisestä. Kontrolliolkena käytettiin pellolla kuivattua käsittelemätöntä kauran olkea (C). Olkien ureakäsittely tehtiin pyöröpaalauksen yhteydessä ja ammonointi välittömästi paalauksen jälkeen. Kaikki käsitellyt oljet suljettiin muovipusseihin.

Olkien sulavuus määritettiin kokonaiskeruumenetelmällä neljällä alkionsiirtotekniikalla tuotetulla ay-sonnilla. Maittavuus määritettiin neljällä pässillä.

Kaikki oljet säilyivät hyvin koko kokeen ajan. Kumpikaan ureakäsittelyistä ei vaikuttanut oljen ravinteiden sulavuuteen raakavalkuaista lukuunottamatta, mutta ammonointi lisäsi oljen orgaanisen aineen, raakavalkuaisen ja raakakuidun sulavuutta ja tätä kautta myös energia-arvoa 14 %. Kumpikaan ureakäsittelyistä ei lisännyt energia-arvoa.

Toisin kuin ammonointi ureakäsittely ei lisännyt oljen syöntiä. Kirjoituksessa arvioidaan lisäksi kirjallisuudessa esitettyjä tekijöitä, jotka vaikuttavat oljen ammonoinnin ja ureakäsittelyn tehoon.

ANNALES AGRICULTURAE FENNIAE, VOL. 29: 113—126 (1990)

Seria ANIMALIA DOMESTICA N. 90 — Sarja KOTIELÄIMET n:o 90

EFFECTS OF TWO FEEDING REGIMENS ON PRODUCTION AND EGG QUALITY OF SIX LAYING HEN HYBRIDS

TUOMO KIISKINEN and LEA HUIDA

KIISKINEN, T. and HUIDA, L. 1990. **Effects of two feeding regimens on production and egg quality of six laying hen hybrids.** Ann. Agric. Fenn. 29: 113—126. (Agric. Res. Centre, Inst. Anim. Prod. SF-31600 Jokioinen, Finland.)

An experiment involving six laying hybrids and two dietary treatments was conducted to study the performance and egg quality of hens. Three hybrids were Finnish (MÄ-16, LSK-61, SK-51) and three were foreign (LSL, HISEX, SVE). Diet A was a control diet and diet B based on phase feeding including rapeseed meal.

The results showed significant differences between hybrids and feeding regimens in egg production, egg weight, feed intake and feed conversion ratio. Diet A produced more eggs with better feed efficiency than diet B, but efficiency of protein conversion was better ($P<0.05$) on diet B and that of ME equal on each diet. Hybrid LSL laid more (51.4 g/hen/day) and SK-51 less eggs (41.8 g/hen/day) than the other hybrids ($P<0.05$). Excluding the MÄ-16 hens, LSL consumed significantly less ($P<0.05$) feed, protein and ME per kilogram eggs than the other genotypes. Feed conversion ratios for LSL, MÄ-16 and the other hybrids were 2.72, 2.82 and 2.96—3.15, respectively. Diet × hybrid interaction in daily feed, protein and ME intake could be ascertained. Hens fed diet B had lower body weight ($P<0.001$), less abdominal fat ($P<0.05$), larger thyroid gland and gizzard ($P<0.001$) and better plumage condition ($P<0.001$) at the end of the laying period than those fed diet A. Also between hybrids significant differences could be found in the above mentioned traits. No significant differences in mortality were detected between diets and hybrids. Incidence of liver lesions was higher on diet B, which contained rapeseed meal.

Both in the physical composition (percentage of shell, yolk and albumen) and quality (Haugh, yolk colour, spec.weight, shell strength) of egg significant differences between hybrids and diets could be found. Diet A produced eggs with slightly more albumen and less yolk than diet B ($P<0.05$). In the chemical composition of egg, differences between hybrids and diets could be detected only in the dry matter content of albumen. Aging of the hens caused changes in the composition of egg: albumen and its dry matter content decreased ($P<0.001$), yolk and its dry matter increased ($P<0.001$), fat content of yolk decreased ($P<0.001$) and most amino acids in albumen decreased. No differences between hybrids and diets in amino acid content of egg could be found.

Index words: laying hen, hybrid, phase feeding, rapeseed meal, egg production, egg quality.

INTRODUCTION

Egg production in Finland has been poorly profitable during the last years. As a result, producers have become increasingly interested in the genetic quality of their birds. In Finland,

only domestic laying hybrids were used earlier and applications for the import of foreign hybrids have increased. In order to obtain more information on the competence of the Finnish hen on an international level, some foreign hybrids (LSL, HISEX, SVE) were included for several years in the production control of the Finnish Poultry Breeders Association (KIISKINEN et al. 1981, EISKONEN and HONGISTO 1982, HONGISTO 1984, ALASPÄÄ 1985, ALASPÄÄ and KUKKOLA 1987, KUKKOLA and ALASPÄÄ 1988). A 1983 decision by the Ministry of Agriculture and Forestry ordered that a large experiment be organized to compare the most important Finnish hybrids with various foreign ones. The present study was conducted to support and to supplement the results obtained in the main experiment which was performed on a private farm. In addition to six hybrids the trial included two dietary regimens, a complete diet and a phased diet with different protein sources. An objective of this experiment was also to study the physical and chemical quality of eggs.

MATERIAL AND METHODS

Birds and housing

Six commercial hybrids of the White Leghorn breed LSL, HISEX, SVE, MÄ-16, LSK-61 and SK-51 were housed under the same practical conditions during the egg-laying period (20—84 weeks). The three last mentioned hybrids were bred in Finland. Around 1300 pullets were placed in three-tier batteries with three hens per cage (700 cm^2/bird). The duration of lighting was increased by 20 minutes weekly to 16 hours, with at intensity of 10 lux. A room temperature of 17 °C was maintained.

Experimental design and diets

The experiment had a 2×6 factorial design. Hens from each hybrid were equally distributed between the two dietary treatments A and B. Hens on treatment A (control) received the same diet (Table 1) throughout the laying period, whereas hens on treatment B were subjected to phase-feeding consisting of two phases between 20—36 and 37—84 weeks. The main difference between diet A and diet B during the first phase was in their supplementary protein. In diet B, soybean meal (SBM) was to a great extent replaced by some domestic protein sources; rapeseed meal (RSM expeller) from the low-glucosinolate cultivar Vankka *(Brassica campestris),* meat and bone meal and Pekilo single cell protein. During the last phase of treatment B, fish meal and SBM were totally replaced by the domestic protein sources and the calculated dietary protein level was reduced from 16.3 to 13 % and that of methionine and lysine from 0.34 to 0.30 % and from 0.86 to 0.60 %, respectively (Table 1). Metabolizable energy (ME) of the diet was lowered from 10.4 to 10.1 MJ/kg by using more oats. Because the RSM contained 15 % oil, supplementary fat (rapeseed oil) was omitted in diet B. During the second phase the calcium content of diet B was raised by increasing limestone. The birds had free access to feed and water.

Six replicates of 30 hens were assigned to each hybrid and eighteen replicates to each dietary treatment.

Experimental procedures

Production of each replicate was measured daily and feed consumption at four-week intervals. All birds were weighed individually at the ages of 20 and 83 weeks. Mortality was recorded and as far as possible all dead birds were sent

Table 1. Composition of the diets.

Diet	A	B	
Age (weeks)	20—84	20—36	37—84
Ingredient (%):			
Fish meal	3	3	—
Meat and bone meal	—	2	3
Soybean meal	16	6	—
Rapeseed meal (expeller)[1]	—	8	8
Pekilo[2]	—	1	1
Barley	45.5	47	29
Oats	25	25	50
Rapeseed oil	1	—	—
Limestone	7	6.5	7.92
Dicalcium phosphate	1.5	0.5	—
Salt	0.35	0.35	0.35
Micronutrients[3]	0.60	0.60	0.60
DL-methionine	0.05	0.05	0.08
L-lysine	—	—	0.05
Composition (calculated):			
ME MJ/kg	10.5	10.4	10.1
Crude protein %	16.3	16.3	13.0
Methionine »	0.34	0.34	0.30
Lysine »	0.84	0.86	0.60
Calcium »	3.27	3.12	3.63
Phosphorus »	0.75	0.80	0.78
Composition (analyzed):			
Crude protein %	16.3 ± 0.58	16.2 ± 0.12	12.9 ± 0.60
Methionine »	0.31	0.30	0.27
Lysine »	0.83	0.81	0.59
Calcium »	3.07	3.10	3.43
Phosphorus »	0.72	0.78	0.64

[1] Low-glucosinolate cultivar Vankka *(B. campestris)* containing 93 % dry matter, 31 % crude protein, 15 % ether extract, 11.21 MJ ME/kg d.m. and 44.8 µmol/g total glucosinolates.
[2] Single cell protein.
[3] Supplies per kilogram of diet: 15000 IU vitamin A, 1700 IU vitamin D, 20 mg vitamin E, 1 mg vitamin K, 3 mg riboflavin, 1.5 mg pyridoxine, 18 mg niacin, 10 µg B_{12}, 0.3 mg folic acid 500 mg choline chloride, 5 mg Carophyll Orange, 20 mg Fe, 45 mg Zn, 50 mg Mn, 4 mg Cu, 0.5 mg I, 0.1 mg Se.

to the National Veterinary Institute for pathological investigations.

Eggshell and albumen quality and yolk colour were determined twice (35 and 68 weeks) using conventional methods; specific gravity by the flotation method, shell strength with a compression force meter, Haugh unit (HU) with a micrometer and yolk colour using the Hoffman — La Roche colour fan. Thirty eggs per group were collected from the same replicate rows for each determination.

At the end of the study, ten birds per group were killed, and their thyroid gland, abdominal fat, gizzard and liver more loosened and weighed. The condition of plumage was evaluated for each replicate row when the birds had laid eggs around at five months and the individual survey was made at the end of the trial for 10 birds from each replicate row. The condition of plumage in the neck, back, breast, wings and tail was evaluated on a scale from 1 to 4 (1 = bare or nearly bare, 4 = good or complete plumage).

Analyses and statistics

Proximate feed analysis was done for each lot of the diets according to the standard methods used by the Institute of Animal Production

of the Agricultural Research Centre. Amino acids were determined with a gas chromatograph (Hewlet Packard 4570). This was performed for a common sample of each diet. The physical and chemical composition of eggs were determined at the same age as egg shell and albumen quality. The physical analyses (percentage of shell, albumen and yolk) and the chemical analyses (dry matter, protein, fat, amino acids, cholesterol) were performed for each replicate the sample consisting of the ten first eggs of each row. Before analyses, albumen and yolk of the eggs were separated and first mixed with a Pamix mixer and then homogenized with an Ultra Turrex mixer. Yolk fat content was analyzed by a modified Bligh—Dryer's method (HOLOPAINEN 1972) and egg yolk cholesterol by an HPLC method described by DUNCAN et al. (1979).

Fat content of the liver was determined by eluting the fat with dichloromethanemetanol according to the method of MAXWELL et al. (1980). The serum value of cholesterol was determined twice (33 and 72 weeks) and serum protein and urea once (72 weeks) using a compact Clinical Analyser manufactured by Kone Oy. Cholesterol and urea analyses were based on enzymatic colorimetric methods and total protein on the biuret method. Data were statistically analysed by a generalized multivariate analysis of variance and covariance (MANOVA). The comparisons between treatments were performed using Tukey's test (STEEL and TORRIE 1960).

RESULTS

The domestic hybrids reached 50 % production at the age of 147 (±1.3) days and the foreign ones at the age of 156 (±1.9) days ($P<0.001$). There were significant ($P<0.001$) differences among the hybrids for all production and feed consumption traits measured (Tables 2, 3, 4). In hen-day production, LSL was superior ($P<0.05$) to the other hybrids producing 51.4 g per day the rate of lay being 81.3 % (Table 2). On the contrary, SK-51 had significantly ($P<0.05$) the poorest egg production at 41.8 g and 70.2 %. The largest egg weight was ascertained for HISEX and LSL at 63.8 and 63.4 g ($P<0.05$), and SK-51 and LSK-61 laid the smallest eggs at 59.7 and 60.0 g, respectively. As regards hen-housed production, the foreign hybrids were significantly ($P<0.05$) superior to the Finnish hybrids, excluding MÄ-16. Daily feed consumption was higher ($P<0.001$) among the foreign hybrids, than among the Finnish hybrids (Table 3). LSL and MÄ-16 were superior to the other genotypes in feed conversion and efficiency of protein and ME utilization. Feed conversion ratio (kg/kg egg) for LSL, MÄ-16 and the other hybrids was 2.72, 2.82 and 2.96—3.15, respectively.

Between the dietary treatments, differences were significant in most production traits. Hen-day and hen-housed production were for diet A 47.4 g and 20.2 kg and for diet B 45.6 g ($P<0.001$) and 19.14 kg ($P<0.01$), respectively. There was also a significant ($P<0.001$) difference in egg weight due to the effect of diet (Table 4 and Fig. 1). In laying intensity no significant difference was detected and the laying curves of the dietary treatments were rather equal. (Table 4, Fig. 2). Significant diet × hybrid interaction ($P<0.05$) was found only for the daily feed, protein and ME intakes (Table 4).

Differences occurred among hybrids ($P<0.001$) and between the diets for final body weight, weight gain (%) and relative weight of abdominal fat, thyroid gland, liver and gizzard (Table 5). By the end of the 68th week of age the birds had gained weight in the range 16—21 % of their 20-week body weight, the highest

Table 2. Egg production data for six laying hybrids fed two different diets.

Diet	Hybrid	Egg output		Rate of lay %	Egg weight g
		kg/housed hen	g/hen/day		
A	LSL	22.50	52.0	81.1	64.4
(control)	MÄ-16	20.36	46.4	74.8	62.2
	HISEX	20.55	49.8	77.6	64.5
	LSK-61	18.35	45.8	76.1	60.1
	SK-51	18.71	42.6	70.8	60.4
	SVE	20.75	47.9	77.8	62.0
B	LSL	20.37	50.7	81.5	62.4
(phased,	MÄ-16	19.35	46.3	75.6	61.3
RSM)	HISEX	19.51	46.5	74.1	63.2
	LSK-61	18.60	43.0	71.9	60.0
	SK-51	17.48	40.9	69.6	59.0
	SVE	19.52	46.2	76.5	60.7
Average					
A		20.20	47.4	76.4	52.3
B		19.14	45.6	74.9	61.1
	LSL	21.43^{a}	51.4^{a}	81.3^{a}	63.4^{a}
	MÄ-16	19.85^{ab}	46.3^{bc}	75.2^{b}	61.8^{b}
	HISEX	20.03^{a}	48.1^{b}	75.9^{b}	63.8^{a}
	LSK-61	18.48^{b}	44.0^{c}	74.0^{bc}	60.0^{cd}
	SK-51	18.10^{b}	41.8^{d}	70.2^{c}	59.7^{d}
	SVE	20.14^{a}	47.0^{bc}	77.1^{b}	61.3^{bc}
SE		0.17	0.54	0.65	0.30

$^{a-d}$ Hybrid means without a common superscript letter are significantly different ($P < 0.05$). Means without superscript letters have no significant differences.
SE = standard error of mean.

Table 3. Feed, protein and energy (ME) intake of hens.

Diet	Hybrid	Feed intake		Protein intake		ME intake	
		g/hen/day	kg/kg eggs	g/hen/day	g/kg eggs	MJ/day	MJ/kg eggs
A	LSL	139	2.72	22.7	444	1.46	28.6
	MÄ-16	126	2.80	20.7	457	1.33	29.5
	HISEX	144	2.94	23.7	480	1.52	30.9
	LSK-61	130	2.92	21.3	477	1.37	30.8
	SK-51	126	3.05	20.7	497	1.33	32.1
	SVE	141	3.00	23.1	490	1.49	31.6
B	LSL	136	2.72	18.5	370	1.39	27.8
	MÄ-16	127	2.83	17.3	388	1.29	28.9
	HISEX	136	2.99	18.5	405	1.39	30.5
	LSK-61	131	3.15	17.9	429	1.34	32.1
	SK-51	127	3.24	17.3	442	1.29	33.1
	SVE	137	3.00	18.7	409	1.39	30.6
Average							
A		135	2.90	22.0	474	1.42	30.6
B		132	2.99	18.0	407	1.35	30.5
	LSL	137^{a}	2.72^{a}	20.6^{b}	407^{a}	1.42^{b}	28.2^{a}
	MÄ-16	127^{b}	2.82^{ab}	19.0^{a}	423^{ab}	1.31^{a}	29.2^{ab}
	HISEX	140^{a}	2.96^{bc}	21.1^{b}	442^{bc}	1.45^{b}	30.7^{bc}
	LSK-61	131^{b}	3.04^{cd}	19.6^{a}	453^{cd}	1.36^{a}	31.5^{cd}
	SK-51	126^{b}	3.15^{d}	19.0^{a}	470^{d}	1.31^{a}	32.6^{d}
	SVE	139^{a}	3.00^{cd}	20.9^{b}	450^{cd}	1.44^{b}	31.1^{cd}
SE		0.2	0.005	0.40	7.5	0.013	0.28

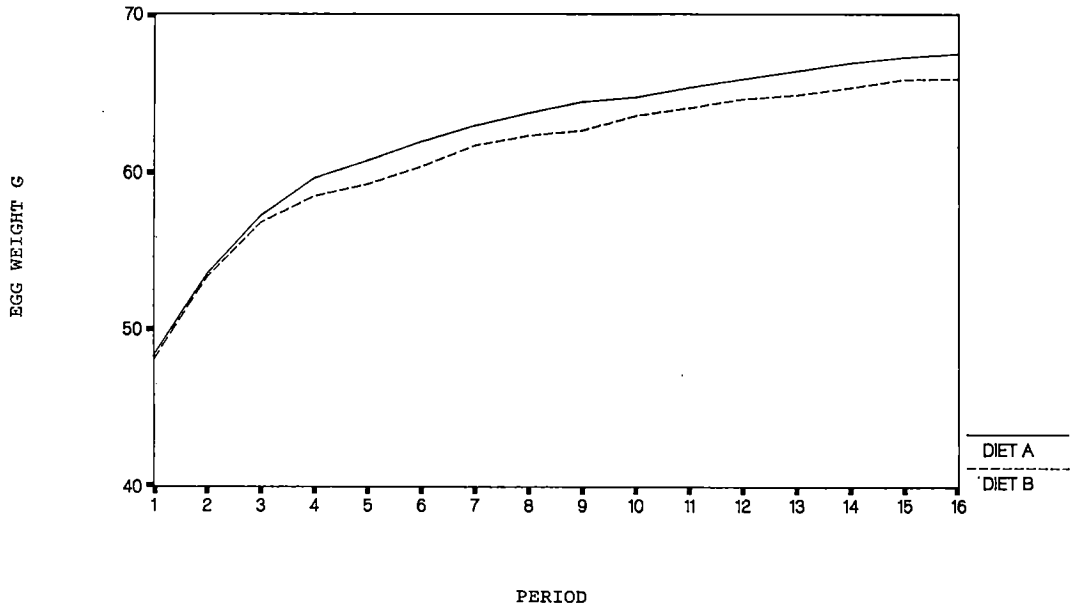

Fig. 1. Egg weight of the dietary treatments.

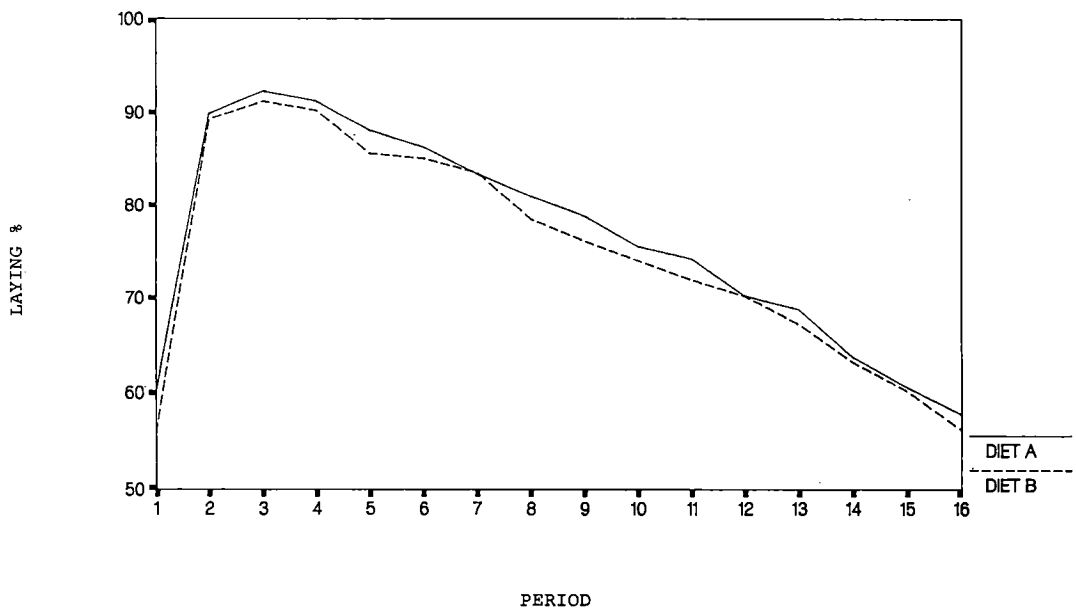

Fig. 2. Percentage egg production of the dietary treatments.

Table 4. Significance of F-values of the analysis of variance for egg production and feed intake.

	Source of variation		D × H
	Diet (D)	Hybrid (H)	
Egg output			
kg/housed hen	**	***	NS
g/hen/day	***	***	NS
Rate of lay	NS	***	NS
Egg weight	***	***	NS
Feed intake			
g/hen/day	*	***	*
kg/kg eggs	*	***	NS
Protein intake			
g/hen/day	***	***	**
g/kg eggs	***	***	NS
ME intake			
MJ/hen/day	***	***	*
MJ/kg eggs	NS	***	NS

* P<0.05
** P<0.01
*** P<0.001
NS = non-significant

gain being for the SVE hybrid (P<0.05) and the lowest for LSL, HISEX and LSK-61. Diet B produced less weight gain (P<0.001), less abdominal fat (P<0.05) and larger thyroid gland (P<0.001) and gizzard (P<0.001) than diet A. No significant diet × hybrid interactions were detected.

Mortality ranged from 10.2 to 19.0 % among six hybrids (Table 6). However, no significant differences were found. Liver lesions (fatty, brittle or broken) occurred more in dead hens fed diet B and it caused slightly but not significantly higher mortality than diet A. In general, no significant dietary or hybrid differences or diet × hybrid interactions were ascertained in the content of liver fat or serum values. Diet B seemed to produce higher serum cholesterol values and the difference compared with diet A was significant (P<0.05) in the second determination. SK-51 had the lowest serum protein value differing significantly (P<0.05) from that of MÄ-16.

At the end of the experiment, the SK-51 hens had better plumage than the others and excluding the SVE hens the differences were significant (P<0.05) (Table 7). Diet B produced slightly better plumage than diet A (P<0.001).

Among the six hybrids, significant differences (P<0.001) could be ascertained for Haugh unit (Table 8). Eggs from LSL, MÄ-16 and HISEX had in each determination higher albumen quality (HU) than the other hybrids. Albumen quality was slightly but significantly

Table 5. Mean final body weight (b.w.), weight gain and relative weight of abdominal fat, thyroid gland, liver and gizzard of hens.

	Final body weight kg	Weight gain %	Abdominal fat % b.w.	Thyroid gland mg/100 g b.w.	Liver g/100 g b.w.	Gizzard g/kg b.w.
Diet	(n = 60)					
A	2.03	21.8	5.04	8.4	2.86	11.6
B	1.92	13.9	4.39	14.3	2.86	15.9
Hybrid	(n = 20)					
LSL	1.81^{a}	16.1^{ab}	4.20^{ab}	11.3^{ab}	3.03^{b}	12.7^{ab}
MÄ-16	2.01^{b}	21.3^{c}	5.73^{b}	8.9^{a}	2.96^{b}	14.9^{ab}
HISEX	1.83^{a}	18.2^{bc}	3.27^{a}	10.6^{ab}	3.02^{b}	16.9^{a}
LSK-61	1.81^{a}	14.1^{a}	4.24^{ab}	10.8^{ab}	2.71^{ab}	16.2^{a}
SK-51	2.01^{b}	19.2^{bc}	5.33^{b}	13.3^{b}	2.48^{a}	11.3^{b}
SVE	2.39^{c}	18.3^{bc}	5.52^{b}	13.1^{b}	2.94^{ab}	10.6^{b}
SE	0.010	0.37	0.177	0.42	0.045	0.69
Source of variation			Significance of F-value			
Diet (D)	***	***	*	***	NS	***
Hybrid (H)	***	***	***	***	***	***
D × H	NS	NS	NS	NS	NS	NS

Table 6. Mortality, incidence of liver lesions, fat content of liver and serum values.

Age weeks	Mortality %	Liver[1] lesions	Liver fat %	Serum values mmol/l			
				cholesterol		protein	urea
				33	72	72	72
Diet	(n = 18)	—	60	54	48	48	48
A	12.2	5/24	11.0	3.85	3.36	55.9	1.90
B	14.6	9/29	11.8	4.29	3.94	54.3	1.88
Hybrid	(n = 6)	—	20	18	16	16	16
LSL	13.4	1/9	10.9	4.72	3.66	55.5^{ab}	1.88
MÄ-16	10.2	4/9	13.7	4.05	3.56	57.0^{a}	1.97
HISEX	19.0	2/11	8.4	3.78	3.15	55.1^{ab}	1.71
LSK-61	13.9	2/8	10.2	4.21	4.39	55.0^{ab}	2.23
SK-51	10.2	1/8	13.0	3.32	3.03	52.6^{b}	1.25
SVE	13.9	4/8	11.8	4.33	3.92	54.7^{ab}	2.24
SE	1.13	—	0.49	0.16	0.14	0.46	0.10
Source of variation			Significance of F-value				
Diet (D)	NS	—	NS	NS	*	NS	NS
Hybrid (H)	NS	—	NS	NS	NS	*	NS
D × H	NS	—	NS	NS	NS	NS	NS

[1] Number of liver lesions (fatty, fragile, haemorrhagic or broken) per dead and obducted hens.

Table 7. Points for plumage condition of hens (score 1—4).

	Total points in the 1st survey[1]	Points for different parts in the 2nd survey					
		Neck	Back	Breast	Wings	Tail	Total
Diet	(n = 18)	(n = 180)					
A	10.34	2.96	2.63	2.01	2.54	1.72	11.86
B	10.58	3.09	2.94	2.28	2.78	2.01	13.10
Hybrid	(n = 6)	(n = 60)					
LSL	10.37^{ab}	3.03^{bc}	3.08^{b}	2.05^{a}	2.67^{ab}	1.63^{a}	12.47^{b}
MÄ-16	13.28^{b}	2.97^{b}	2.75^{ab}	2.05^{a}	2.85^{b}	1.72^{a}	12.33^{ab}
HISEX	7.29^{a}	2.58^{a}	2.32^{a}	1.88^{a}	2.45^{a}	1.65^{a}	10.88^{a}
LSK-61	8.31^{a}	2.90^{ab}	2.57^{a}	2.02^{a}	2.62^{ab}	1.62^{a}	11.72^{ab}
SK-51	11.00^{b}	3.28^{c}	3.17^{b}	2.62^{b}	2.90^{b}	2.32^{b}	14.28^{c}
SVE	12.53^{b}	3.37^{c}	2.83^{ab}	2.23^{ab}	2.50^{a}	2.25^{b}	13.18^{bc}
SE	0.42	0.039	0.048	0.043	0.036	0.051	0.17
Source of variation			Significance of F-value				
Diet (D)	NS	NS	***	***	***	**	***
Hybrid (H)	***	***	***	***	***	***	***
D × H	NS	NS	NS	NS	*	NS	NS

[1] Neck, back, breast and tail included.

($P < 0.001$) affected by the dietary treatment in the second determination (68 weeks). In the first determination (35 weeks) no differences were found in yolk colour, but in the second one yolks of eggs from the LSL hens were significantly paler ($P < 0.05$) than eggs from the other genotypes. Diet B produced slightly paler ($P < 0.001$) yolks than diet A. A significant diet × hybrid interaction ($P < 0.001$) occurred for yolk colour at the later age. Shell quality as measured by specific weight was not influenced by diet and hybrid in the first determination. On the contrary, shell strength expressed as a compression force was significantly ($P < 0.05$)

Table 8. Eggshell and albumen quality and yolk colour.

	1st determination (35 weeks)				2nd determination (68 weeks)			
	Haugh unit	Yolk colour	Specific weight of egg	Shell strength	Haugh unit	Yolk colour	Specific weight of egg	Shell strength
Diet (n = 180)								
A	92.4	12.0	1.0937	3.50	83.0	13.0	1.0844	2.87
B	91.9	12.0	1.0939	3.51	81.0	12.6	1.0830	2.76
Hybrid (n = 60)								
LSL	94.6^{b}	12.0	1.0934	3.50^{ab}	86.8^{c}	11.8^{a}	1.0820	2.53^{a}
MÄ-16	94.5^{b}	12.1	1.0946	3.52^{ab}	85.2^{c}	13.0^{b}	1.0843	3.10^{c}
HISEX	92.9^{ab}	12.0	1.0938	3.34^{a}	83.2^{c}	13.1^{b}	1.0839	2.65^{ab}
LSK-61	91.1^{a}	12.0	1.0949	3.35^{a}	79.2^{ab}	12.9^{b}	1.0837	2.80^{abc}
SK-51	88.7^{a}	12.1	1.0936	3.55^{ab}	77.2^{a}	13.0^{b}	1.0843	2.92^{bc}
SVE	91.2^{a}	12.0	1.0926	3.75^{b}	80.2^{b}	13.0^{b}	1.0839	2.91^{bc}
SE	0.28	0.02	0.00033	0.032	0.35	0.05	0.00027	0.036
Source of variation	Significance of F-value							
Diet (D)	NS	NS	NS	NS	***	***	**	NS
Hybrid (H)	***	NS	NS	**	***	***	NS	***
D × H	NS	NS	NS	NS	NS	***	NS	NS

Table 9. Physical composition of eggs.

	% from egg weight								
	Albumen			Yolk			Shell		
Age of hen (weeks)	35	68	Average	35	68	Average	35	68	Average
Diet (n = 180)									
A	57.7	55.7	56.7	29.6	31.4	30.5	12.2	12.5	12.4
B	57.3	55.2	56.3	30.0	31.8	30.9	12.3	12.8	12.5
Hybrid (n = 60)									
LSL	58.2^{a}	57.0^{a}	67.6^{a}	28.7^{a}	30.1^{a}	29.4^{a}	12.5^{a}	12.7^{ab}	12.6^{bc}
MÄ-16	57.9^{ab}	54.6^{c}	56.2^{bc}	29.4^{ab}	31.7^{ab}	30.5^{b}	12.3^{ab}	13.2^{a}	12.7^{c}
HISEX	58.4^{a}	56.5^{ab}	57.5^{a}	28.8^{a}	30.4^{a}	29.6^{a}	12.3^{ab}	12.7^{ab}	12.5^{bc}
LSK-61	56.6^{b}	54.5^{c}	55.5^{c}	30.8^{bc}	32.7^{bc}	31.7^{c}	12.4^{ab}	12.7^{ab}	12.6^{bc}
SK-51	57.4^{ab}	55.7^{bc}	56.5^{b}	30.2^{b}	32.2^{b}	31.2^{bc}	12.0^{b}	11.9^{c}	12.0^{a}
SVE	56.7^{b}	54.7^{c}	55.7^{c}	31.0^{c}	32.4^{b}	31.7^{c}	12.1^{b}	12.6^{b}	12.3^{b}
SE	0.12	0.14	0.10	0.11	0.12	0.09	0.04	0.06	0.04
Source of variation	Significance of F-value								
Age (A)	—	—	***	—	—	***	—	—	***
Diet (D)	NS	NS	*	NS	NS	*	NS	*	**
Hybrid (H)	***	***	***	***	***	***	***	***	***
A × H[1]	—	—	*	—	—	NS	—	—	***

[1] Other significant interactions were not ascertained.

lower in eggs produced by HISEX and LSK-61 than in eggs from the SVE hybrid. Later, diet B produced eggs with a lower ($P < 0.01$) specific weight than diet A and eggs from LSL had lower ($P < 0.01$) shell strength values than those from MÄ-16, SK-51 and SVE.

There were dietary and hybrid differences in the physical composition of egg (Table 9). LSL and HISEX produced eggs with higher albumen and lower yolk content ($P < 0.05$) than the other genotypes. Also in percentage of shell significant ($P < 0.001$) differences were observed. Hens fed diet B laid eggs with slightly lower albumen ($P < 0.05$) and higher yolk ($P < 0.05$) and

Table 10. Chemical composition of albumen and yolk of eggs.

Age of hen (weeks)	Albumen				Yolk							
	Dry matter %		Protein % dm.		Dry matter %		Protein % dm.		Fat % dm.		Cholesterol %	
	35	68	35	68	35	68	35	68	35	68	35	68
Diet (n = 18)												
A	12.4	11.3	89.0	88.0	49.9	50.3	32.7	31.2	59.1	56.6	0.90	0.90
B	12.3	11.2	89.8	88.1	49.7	50.6	32.8	32.8	59.2	56.0	0.96	0.90
Hybrid (n = 6)												
LSL	12.3^{b}	11.0^{a}	89.9	88.3	49.6	50.1	33.2	31.0	58.2	55.7	0.91	0.90
MÄ-16	12.8^{a}	11.4^{b}	88.7	88.3	50.0	50.7	33.1	32.9	59.4	55.4	0.87	0.92
HISEX	12.4^{b}	11.4^{b}	89.7	88.2	49.8	50.4	32.8	33.0	58.6	56.6	0.95	0.88
LSK-61	12.5^{b}	11.4^{b}	89.3	88.2	49.8	50.8	32.6	30.4	59.2	56.3	0.92	0.89
SK-51	12.1^{c}	11.2^{ab}	89.6	87.7	49.9	50.4	32.5	32.5	59.7	57.7	0.90	0.90
SVE	12.1^{c}	11.2^{ab}	89.3	88.2	49.6	50.6	32.2	32.3	59.7	55.9	1.00	0.90
SE	0.05		0.19	0.10	0.08	0.13	0.10	0.52	0.22	0.30	0.014	0.010
Source of variation	Significance of F-value'											
Age (A)	***		NS		***		NS		***		—	—
Diet (D)	*		NS		NS		NS		NS		*	NS
Hybrid (H)	***		NS		NS		NS		NS		NS	NS
A × H[1]	*		NS		NS		NS		NS		—	—

[1] Among other interactions no significance was ascertained.

shell content ($P<0.01$) than birds fed diet A. When the hens aged, the percentage of albumen decreased ($P<0.001$) and that of yolk and shell content increased ($P<0.001$). The only interactions found in the physical composition of egg were between age and hybrid for percentage of albumen ($P<0.05$) and shell ($P<0.001$).

Significant differences among hybrids and the diets for the chemical composition of the egg components were found only for dry matter content (DM) of the albumen (Table 10). In the first determination, eggs produced by diet B contained more cholesterol in the yolk ($P<0.05$) than eggs produced with diet A. No difference appeared in the second determination.

In the amino acid composition of the albumen or the yolk, no dietary or hybrid differences were discovered (Table 11). Most of the amino acids of the albumen were significantly lower in the later determination. On the contrary, in yolk protein the changes were significant only in the case of methionine and tyrosine where opposite changes appeared. Glutamic acid increased both in albumen and yolk protein when the hens were older.

DISCUSSION

Differences in the performance of hens were considerable among the hybrids. The results are mainly in agreement with the results reported from the simultaneous experiment conducted on the private farm (ANON. 1986). The results of the present trial and those of the control station of the Finnish Poultry Breeders Association (KIISKINEN et al. 1981, ALASPÄÄ 1985, ALASPÄÄ and KUKKOLA 1987, KUKKOLA and ALASPÄÄ 1988) suggest that the average Finnish hen material is not quite competitive with the best foreign hybrids.

Table 11. Amino acid composition of albumen and yolk of eggs.[1]

Age of hen (weeks)	Albumen		Yolk	
	35	68	35	68
Amino acid g/16 g N	n = 36	n = 36	n = 36	n = 36
Methionine	3.74(3.43—4.57)	2.39(2.07—2.63)***	2.09(1.95—2.23)	1.70(1.60—1.87)***
Lysine	6.96(6.88—7.15)	6.54(6.27—6.83)*	7.41(7.27—7.55)	7.29(7.02—7.63)
Arginine	5.58(5.38—5.88)	5.39(5.20—5.53)	6.70(6.58—6.88)	6.56(6.38—6.82)
Isoleucine	5.46(5.30—5.78)	5.02(4.65—5.32)**	5.10(4.92—5.30)	5.08(4.65—5.37)
Leucine	8.49(8.35—8.62)	7.98(7.78—8.45)*	8.33(8.20—8.43)	8.13(7.75—8.28)
Phenylalanine	6.30(6.17—6.45)	5.62(5.08—6.10)***	4.24(4.13—4.32)	4.39(4.13—4.86)
Tyrosine	4.51(4.38—4.62)	4.06(3.90—4.20)***	4.75(4.50—4.92)	4.50(4.27—4.63)**
Threonine	4.60(4.53—4.68)	4.44(4.27—4.55)	4.78(4.65—4.92)	4.88(4.47—5.17)
Valine	6.79(6.62—7.07)	6.08(5.60—6.43)***	5.41(5.15—5.67)	5.47(5.13—5.73)
Alanine	6.05(5.95—6.15)	5.46(5.02—5.87)***	4.98(4.83—5.08)	4.91(4.68—5.05)
Aspartic acid	10.23(10.13—10.37)	10.01(9.62—10.60)	9.10(8.73—9.30)	9.39(8.83—9.93)
Glutamic acid	12.41(12.03—12.65)	13.13(12.52—14.03)*	11.17(10.70—11.40)	12.25(11.80—12.68)***
Glycine	3.71(3.63—3.82)	3.39(3.20—3.53)**	2.96(2.85—3.05)	2.99(2.88—3.10)
Proline	3.57(3.47—3.65)	3.56(3.35—3.83)	3.84(3.73—4.00)	3.88(3.72—4.03)
Serine	6.81(6.70—6.90)	6.71(6.37—6.85)	7.64(7.48—7.73)	7.76(7.41—8.23)

* Significant difference between ages. No significant differences between the diets or the
** hybrids and interactions were ascertained.
***

[1] In parentheses, the variation of the hybrid means.

Diet B, based on phase-feeding and the use of RSM, was inferior to diet A in egg production. This was mainly a result of decreased egg weight. The reduction in egg size often found with RSM may have been caused by reduced feed and energy intake as SUMMERS et al. (1987) suggest. Although feed efficiency on diet B was inferior to that on diet A, utilization of ME was equal in each regimen and phase-feeding (diet B) saved protein almost 70 g per kilo eggs. The lower body weight, weight gain and abdominal fat of hens fed diet B revealed that these birds used less feed for weight gain and fat deposits.

Mortality of birds was higher in this experiment than in the simultaneous comparison on the private farm (ANON. 1986). However, it was more equal among the hybrids in the present trial. In each experiment, mortality of the HI-SEX hybrid was highest, but the results of the control station do not support this observation (KIISKINEN et al. 1981, ALASPÄÄ 1985, ALASPÄÄ and KUKKOLA 1987).

Due to RSM, diet B caused a higher incidence of liver lesions and larger thyroid glands than diet A. Fatty liver haemorrhagic syndrome in laying hens is a typical phenomenon associated with the use of RSM, and genetic differences in sensitivity to RSM have been reported (MARCH et al. 1975, CAMPBELL and SMITH 1979, PROUDFOOT et al. 1983). In the present study, differences in the incidence of liver lesions between the hybrids could be found but they could not be ascertained statistically. A possible explanation for the slightly better plumage condition on diet B could be the lower production level of hens fed that diet.

Differences in albumen quality (HU) were remarkably great between the hybrids especially at the end of the laying period. This was also found in the farm trial (ANON. 1986). In each experiment the lowest HU values were obtained for SK-51, LSK-61 and SVE. The shell quality parameters of the LSL hybrid were lower in the second determination than those of the other genotypes. The corresponding values of the farm trial (ANON. 1986) were not similar in all respects, but they support a possible con-

clusion that the shell quality of LSL hens is a weakness of this hybrid. Incidence of cracked eggs of LSL and HISEX was highest in the quality control of the egg packer (ANON. 1986). These results seem to support the conclusion of BOUGON et al. (1981), that the increase in the laying hen's productivity leads to a reduction in egg shell quality and an improvement in the albumen.

It is difficult to find any explanation for the slightly but significantly lower albumen quality of eggs produced by diet B at the end of the laying period. The lower specific weight of eggs on that diet could be caused by RSM. In several studies more cracked eggs or decreased shell thickness has been reported to a result of inclusion of RSM in the diet. (VOGT and TORGES 1976, HULAN and PROUDFOOT 1980 a, b, PROUDFOOT et al. 1983, THOMKE et al. 1983, KIISKINEN 1983). Rapeseed meal was also a possible factor for slightly reduced yolk colour on diet B. Eggs from hens given rapeseed have shown a higher degree of yolk mottling (KARUNAJEEWA 1978).

It is generally known that the portion of the yolk increases with the age of the hen and that the large eggs produced by hens of the same age have smaller yolks than those of smaller eggs. Eggs produced by LSL and HISEX were largest and they also contained more albumen and less yolk than the eggs of the other hybrids. Differences in egg weight account also for differences in the physical composition of eggs between the diets.

Significant hybrid differences in the chemical composition of eggs were ascertained only in the DM of albumen. SØRENSEN and AMBROSEN (1978) presented that the heritability of albumen solids ranged from 0.5 to 0.6 if the hens were of a similar age. Slightly, but significantly lower albumen solids of eggs produced by diet B could be the result of amino acid deficiencies. AL BUSTANY and ELWINGER (1987) have found that increased dietary lysine levels raised the DM of albumen.

The hen's age seems to be the most important factor in the chemical composition of the egg. The dry matter content of the albumen decreases and that of the yolk remains unchanged or increases slightly with age of the hen. According to some Scandinavian reports the DM of the albumen decreases from 13 % (26—28 weeks) to 12 % (51—56 weeks) and further to 11.5 % at the age of 78—82 weeks (ANDERSSON et al. 1978, AMBROSEN and ROTENBERG 1981, AL BUSTANY and ELWINGER 1987, AIMONEN 1988). The decrease of total lipids in DM of the yolk was approximately three percentage units, and this was also reached in a study by ANDERSSON et al. (1978). As regards amino acid composition of egg protein, AMBROSEN and ROTENBERG (1981) could not show significant differences between hen-lines, but most amino acid in the albumen decreased slightly towards the end of the laying period which is in agreement with the present study. The great difference in the methionine content of the albumen between hen ages cannot be accurate, but is obviously due to errors in the analytical method used. Apparently, the effect of feeding on the amino acid composition of egg protein is of minor importance as no significant differences between the dietary treatments could be shown. SCHOLTYSSEK (1977) found that the dietary protein level had only a slight effect on the contents of some amino acids in the egg.

REFERENCES

AIMONEN, E. 1988. Not published.

ALASPÄÄ, M. 1985. Kanatalouskoeaseman tiedonanto koekaudelta 1984—1985. Siipikarja 67: 299—301.

— & KUKKOLA, I. 1987. Kanatalouskoeaseman tiedonanto koekaudelta 1985—1987. Siipikarja 69: 89—91.

AL BUSTANY, Z. & ELWINGER, K. 1987. Shell and interior qual-

ity and composition of eggs from hens of different strains and ages fed different dietary lysine levels. Acta Agric. Scand. 37: 175—187.

AMBROSEN, T. & ROTENBERG, S. 1981. External and internal quality and chemical composition of hen eggs as related to hen age and selection for production traits. Acta Agric. Scand 31: 139—152.

ANDERSSON, K., ELWINGER, K. & PAMLENYI, I. 1978. Restricted feeding and different protein levels to two strains of SCWL hybrids. 2. Effects on egg composition. Swed. J. Agric. Res. 8: 241—247.

ANON. 1986. Kanahybridikokeen tulokset ja koetyöryhmän suositukset. Siipikarja 68: 169—176.

BOUGON, M., LAHELLEC, C. & PROTAIS, J. 1981. Variations in egg quality in connection with yields in production. Quality of eggs. Proc. First Eur. Symp., Abeldoorn. p. 226—235.

CAMPBELL, L.D. & SMITH, T.K. 1979. Responses of growing chickens to high dietary contents of rapeseed meal. Br. Poult. Sci. 20: 231—237.

DUNCAN, I.W., CULBRETH, P.H. & BURTIS, C.A. 1979. Determination of free, total and esterified cholesterol by high-performance liquid chromatography. J. Crom. 162: 281—292.

EISKONEN, A. & HONGISTO, M. 1982. Kanatalouskoeaseman tiedonanto koekaudelta 1981—1982. Siipikarja 64: 311—313.

HOLOPAINEN, M. 1972. Kananmunan lipidikoostumuksesta. Acta Agr. Fenn. 125: 1—48.

HONGISTO, M. 1984. Kanatalouskoeaseman tiedonanto koekaudelta 1983—1984. Siipikarja 66: 134—137.

HULAN, H.W. & PROUDFOOT, F.G. 1980 a. The nutritional value of rapeseed meal for caged layers. Can. J. Anim. Sci. 60: 139—147.

— & PROUDFOOT, F.G. 1980 b. The nutritional value of rapeseed meal for layer genotypes housed in pens. Poult. Sci. 59: 585—593.

KARUNAJEEWA, H. 1978. Effect of rapeseed and dried citrus pulp meals on egg yolk colour and performance of crossbred hens. J. Austr. Inst. Agric. Sci. 44: 208—209.

KIISKINEN, T. 1983. Effects of Regent rapeseed meal fed during the rearing and laying period on the performance of chickens. Ann. Agric. Fenn. 22: 221—231.

—, TUISKULA, M., EISKONEN, A. & EKLUND, K. 1981. Kanatalouskoeaseman tiedonanto koekaudelta 1980—1981. Siipikarja 63: 307—313.

KUKKOLA, I. & ALASPÄÄ, M. 1988. Kanatalouskoeaseman tiedonanto koekaudelta 1986—1988. Siipikarja. 70: 325—328.

MARCH, B.E., BIELY, S. & SOONG, R. 1975. The effects of rapeseed meal fed during the growing and/or laying periods on mortality and egg production in chickens. Poult. Sci. 54: 1875—1882.

MAXWELL, R.J., MARMER, W.N., ZUBILLAGA, M.P. & DALICKAS, G.A. 1980. Determination of total fat in meat and meat products by a rapid column method. J. Assoc. Off. Anal. Chem. 60: 600—603.

PROUDFOOT, F.G., HULAN, H.W. & MCRAE, K.B. 1983. Effect of feeding poultry diets supplemented with rapeseed meal as a primary protein source to juvenile and adult meat breeder genotypes. Can. J. Anim. Sci. 63: 957—965.

SCHOLTYSSEK, S. 1977. Einfluss unterschiedlicher Proteingaben im Futter auf die Qualität von Hühnereiern. Arch. Geflügelk. 41: 10—13.

STEEL, R.D. & TORRIE, J.H. 1960. Principles and Procedures of Statistics. 481 p. New York.

SUMMERS, J.D., LEESON, S. & SPRATT, D. 1987. Canola meal and egg size. Can. J. Anim. Sci. 68: 907—913.

SØRENSEN, P. & AMBROSEN, T. 1978. Heritability of the proportion of solids in egg albumen and its relation to egg weight and albumen height. Acta Agric. Scand. 28: 3—9.

THOMKE, J., ELWINGER, K., RUNDGREN, M. & AHLSTRÖM, B. 1983. Rapeseed meal of Swedish low-glucosinolate type fed to broiler chickens, laying hens and growing-finishing pigs. Acta Agric. Scand. XXXIII: 75—96.

VOGT, H. & TORGES, H-G. 1976. Rapsextractionsschrot aus einer erucasäure- und glucosinolatarmen Sommerrapssorte im Legehennenfutter. Arch. Geflügelk. 40: 225—231.

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SELOSTUS

Kuuden kanahybridin tuotanto ja munanlaatu kahdella ruokinnalla

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Maatalouden tutkimuskeskus

Kokeessa verrattiin kolmen kotimaisen (MÄ-16, LSK-61, SK-51) ja kolmen ulkomaisen (LSL, HISEX, SVE) kanahybridin tuotantoa ja munanlaatua käyttäen kahta ruokintaa. Toisessa ruokinnassa (A) annettiin kanoille koko kokeen ajan (20.—84. ikäviikot) koostumukseltaan samaa täysrehua, joka sisälsi 16 % valkuaista ja 10.5 MJ ME/kg. Ruokinnassa B käytettiin vaihe- eli faasiruokintaa, jossa rehun valkuaispitoisuus alennettiin 36 viikon iässä 16 %:sta 13 %:iin ja ME-pitoisuus 10.4 MJ:sta 10.1 MJ:een rehukilossa. Lisäksi B-ruokinnan seoksissa kalajauho ja soijajauho korvattiin suurimmaksi osaksi kotimaisilla valkuaisrehuilla kuten rypsipuristeella ja viimeisen vaiheen (36.—84. ikäviikot) aikana kauramäärä lähes kaksinkertaistettiin.

Tulokset osoittivat merkitseviä eroja hybridien ja ruokintamenetelmien välillä munantuotannossa, munanpainossa, rehunkulutuksessa ja rehuhyötysuhteessa. A-ruokinnalla kanat munivat paremmin ja käyttivät rehua tehokkaammin kuin B-ruokinnalla, mutta proteiinin hyväksikäyttö oli viimeksimainitulla parempi ja energiahyötysuhde sama kummallakin ruokinnalla. LSL-hybridin munantuotanto (51,4 g/pv) oli paras ja SK-51:n huonoin (41.8 g/pv). Kotimaista MÄ-16-hybridiä lukuunottamatta LSL kulutti muita hybridejä merkitsevästi vähemmän rehua, valkuaista ja ME:a munakiloa kohden. Rehuhyötysuhteet olivat 2.72 (LSL), 2.82 (MÄ-16) ja 2.96—3.15 (muut).

B-ruokinnalla olleet kanat olivat kevyempiä, niissä oli vähemmän sisälmysrasvaa, niiden kilpirauhanen ja lihasmaha olivat suurempia ja höyhenpeite parempi kuin A-ruokinnalla. Merkitseviä eroja kuolleisuudessa ei voitu osoittaa hybridien ja ruokintojen välillä. Maksavikoja oli kuolleissa eläimissä enemmän B-ruokinnalla, jonka rehu sisälsi rypsipuristetta. Munan fysikaalisessa koostumuksessa (valkuaisen, keltuaisen ja kuoren osuus) ja laadussa (Haugh, keltuaisen väri, ominaispaino, kuoren lujuus) todettiin merkitseviä eroja hybridien ja ruokintojen välillä. Kemiallisen koostumuksen osalta tilastollisesti merkitseviä eroja oli vain valkuaisen kuiva-ainepitoisuudessa. Sen sijaan kanojen ikääntyminen aiheutti seuraavia muutoksia munan koostumuksessa: valkuaisen osuus ja sen kuiva-ainepitoisuus laski, keltuaisen osuus ja kuiva-ainepitoisuus nousi, keltuaisen rasvapitoisuus samoin kuin valkuaisosan useimpien aminohappojen pitoisuus aleni. Munan aminohappopitoisuuksissa ei todettu ruokintojen ja hybridien välillä merkitseviä eroja.

ANNALES AGRICULTURAE FENNIAE, VOL. 29: 127—129 (1990)
Seria ANIMALIA DOMESTICA N. 91 — Sarja KOTIELÄIMET n:o 91

Research note

EFFECT OF BETAINE SUPPLEMENTATION OF LOW METHIONINE DIET FOR GROWING PIGS

TIMO ALAVIUHKOLA and KAIJA SUOMI

ALAVIUHKOLA, T. & SUOMI, K. 1990. **Effect of betaine supplementation of low methionine diet for growing pigs.** Ann. Agric. Fenn. 29: 127—129. (Agric. Res. Centre, Swine Res. Sta., 05840 Hyvinkää, Finland.)

In an experiment with 100 growing pigs betaineanhydride was used to replace synthetic DL-methionine in a methionine deficient diet. Daily gain and feed: gain ratio were significantly different before methionine supplementation. Betaine seemed to have no methionine-saving effect as a methyl group donor.

Index words: betaine, growing pigs.

Betaine is produced by extracting sugar beet molasses wich containes 3.1—5.4 % of betaine in dry matter (STEINLE and FISHER 1978). Betaine has been shown to improve the growth rate of chickens in a methionine deficient diet (MISHLER et al. 1949). PESTI et al. (1979) found that betaine supplementation increased the growth rate of broilers fed a diet low in sulphur containing amino acids.

Methionine acts in the metabolism as a raw material in protein synthesis and as methyl group donor. Other possible donors are choline and betaine. Additional betaine improves gain only when methionine and choline are limiting factors in the feed.

In Finland, methionine supplements are used quite routinely in feed manufacture. As the price of betaine is lower than that of methionine, an attempt was made to find out if betaine would have any methionine-saving effect in the feeding of growing pigs.

One hundred piglets at around 25 kg live weight were allotted to five experimental groups on the basis of their litter origin, sex and live weight.

Each pig group was fed one of five diets (Table 1) to 100 kg live weight. Pair feeding was used and water was offered *ad libitum*. In the experimental diets, fish meal was replaced with pea meal and SCP to achieve as low a methionine level as possible. Calculated and analyzed methionine, methionine + cystine as well as choline content of the feeds are shown in Table 2.

Significant differences were found between treatments in daily gain and feed: gain ratio. Betaine seemed to have no beneficial effect on the performance of growing pigs fed a low methionine diet.

Table 1. Composition of the experimental diets and calculated content of energy, protein and amino acids.

Diet	1	2	3	4	5
Barley, %	62.28	67.14	64.36	67.80	67.72
Oats, %	15.00	—	—	—	—
SBM, %	10.50	10.00	4.50	7.20	7.20
Pea meal, %	—	5.50	9.30	5.00	5.00
Fish meal, %	5.00	—	—	—	—
Meat and bone meal, %	2.00	2.00	3.80	3.80	3.80
Skimmed milk powder, %	1.00	1.00	1.00	1.00	1.00
Molasses, %	2.00	2.00	2.00	2.00	2.00
Dried sugar beet pulp, %	—	8.00	9.00	9.00	9.00
Yeast, %	—	1.00	3.70	2.00	2.00
Fat, %	—	0.50	0.50	0.40	0.40
Vitam. + min. mixt., %	2.22	2.62	1.42	1.42	1.42
L-lysine, %	—	0.12	0.10	0.14	0.14
DL-methionine, %	—	0.12	—	0.04	0.04
Betaineanhydride, %	—	—	0.12	—	0.08
FU/kg (air dry)	0.97	0.96	0.96	0.96	0.96
Crude protein, %	17.1	15.0	15.0	15.0	15.0
Lysine, %	0.91	0.85	0.85	0.85	0.85
Treonine, %	0.65	0.52	0.52	0.51	0.51

Table 2. Calculated and analyzed methionine (+ cystine) and choline content of the diets.

Diet no:	Methionine %		Met. + cystine		Choline anal. mg/kg
	calc.	anal.	calc.	anal.	
1	0.32	0.32	0.58	0.61	1250
2	0.34	0.32	0.55	0.57	1100
3	0.22	0.22	0.40	0.45	1050
4	0.26	0.26	0.45	0.51	1050
5	0.26	0.28	0.45	0.50	1000

Table 3. Daily gain, feed : gain ratio and lean meat content of the carcass of the pigs fed experimental diets.

Group (diet)	1	2	3	4	5
Daily gain g/d (25—98 kg)	852^{ac}	823^{abcd}	788^{bd}	804^{abcd}	800^{abd}
Feed : gain (FU/kg)	2.48^{ac}	2.55^{abcd}	2.68^{bd}	2.63^{abd}	2.65^{bd}
Lean meat, %	51.7	51.2	51.2	50.4	50.4

REFERENCES

MISHLER, D.H., CARRICK, C.W. & HAUGE, S.M. 1949. Methionine, choline, betaine and fish products in a simplified ration. Poultry Sci. 28: 24—30.

PESTI, G.M., HARPER, A.E. & SUNDE, M.L. 1979. Sulphur amino acid and methyl donor status of corn-soy diets fed to starting broiler chicks and turkey poults. Poultry Sci. 58: 1541—1547.

STEWLE, G. & FISHER, E. 1978. Hochdruckflussigkeitschromatographische Methode zur Bestimmung von Betain in Zuckerfabriksprodukten. Zuckerind. 103 (2): 129—131.

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SELOSTUS

Betaiini metioniinin korvaajana lihasikojen ruokinnassa

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Maatalouden tutkimuskeskus

Sioille valmistettiin rehuseoksia käyttäen raaka-aineita, joiden metioniinipitoisuus on alhainen. Rehua täydennettiin synteettisellä DL-metioniinilla tai betaiinianhydridillä. Ruokintakokeen tarkoituksena oli selvittää, voidaanko betaiinilla korvata metioniinia metyyliryhmien luovuttajana sikojen aineenvaihdunnassa samoin kuin siipikarjan ruokinnassa.

Sikojen kasvu ja rehun hyväksikäyttö olivat betaiinilla täydennetyllä rehulla merkitsevästi heikommat kuin metioniinilla täydennettyä rehua saaneilla sioilla. Käytetyllä metioniini-kystiini-koliinitasolla sioilla ei siten liene puutetta metyyliryhmän luovuttajista vaan pikemminkin metioniinista kudosproteiinin rakennusaineena.

ANNALES AGRICULTURAE FENNIAE, VOL. 29: 131—139 (1990)

Seria AGROGEOLOGIA ET -CHIMICA N. 155 — Sarja MAA JA LANNOITUS n:o 155

THE SELENIUM CONTENT OF SOME AGRICULTURAL CROPS AND SOILS BEFORE AND AFTER THE ADDITION OF SELENIUM TO FERTILIZERS IN FINLAND

TOIVO YLÄRANTA

YLÄRANTA, T. 1990. **The selenium content of some agricultural crops before and after the addition of selenium to fertilizers in Finland.** Ann. Agric. Fenn. 29: 131—139. (Inst. Crop and Soil Sci., SF-31600 Jokioinen, Finland.)

The addition of selenate to the most common multinutrient fertilizers used in agriculture and horticulture was started in Finland in 1984.

The mean selenium content of barley, oats and grasses at present is 0.1—0.2 mg/kg dry matter, which is up to 20 times higher than that before the beginning of selenium fertilization.

Ensiling loss of selenium in effluents in fresh grass silage was rather slight, varying in four silos from 6 to 15 %.

No statistically significant increase in plant available selenium contents in soils has been detected.

Index words: selenium content, agricultural crops, selenium fertilization, ensiling losses.

INTRODUCTION

The addition of selenium, as sodium selenate, to the most common multinutrient fertilizers used in agriculture and horticulture was started in Finland on July 1, 1984. Selenium has been added to the fertilizers used mainly for cereal crops at the rate of 16 mg/kg, and to those used mainly for grassland cultivation at the rate of 6 mg/kg. As the average fertilizer application is about 500 kg/ha, each cereal crop receives about 8 g/ha of selenium and each grass crop 3 g/ha. The target selenium content of plants has been 0.1 mg/kg dry matter (YLÄRANTA 1985).

In Finland, the effects of selenium fertilizers on the selenium contents of crops, animal fodder and foodstuffs of both animal and plant origin have been monitored carefully and systematically. In order to evaluate the effect of selenium application on the environment, the selenium contents of soils and of lake and river waters have also been measured.

This paper describes the selenium content of some agricultural crops before and after the addition of selenium to fertilizers. The ensiling loss of selenium in grass silage, and the plant available fraction of selenium in the soils give valuable information for evaluation of the fate of selenium in the environment.

MATERIAL AND METHODS

Selenium content of crops before the beginning of selenium fertilization

The selenium concentrations in the dry matter of 12 crops grown side by side are presented. The field experiments were carried out at nine sites in various parts of Finland. The experimental fields, collection of samples and their preliminary treatment have been described in an earlier paper (YLÄRANTA and SILLANPÄÄ 1984).

Selenium content of barley, oats and grasses after the beginning of selenium fertilization

The grain samples of barley and oats have been collected by the Finnish State Granary from different farms throughout the country. The grain samples are a random sample of the most common varieties cultivated in Finland. Thus, they represent very well the actual selenium contents of the Finnish barley and oats.

The grass samples have been collected from farms in different parts of Finland. They have been analyzed by the Soil Fertility Service of Finland (Viljavuuspalvelu Oy).

Ensiling loss of selenium in fresh grass silage

Ensiling loss of selenium in effluents was measured from the samples collected from the experimental silos of the Institute of Animal Production of the Agricultural Research Centre of Finland in 1986. The grass intended for silage was cocksfoot *(Dactylis glomerata)* and timothy *(Phleum pratense)* ley. The ley contained 46.2 % cocksfoot, 41.4 % timothy and 0.4 % red clover, the rest being weeds. The fertilizer applied contained 110 kg of nitrogen and 3.5 g of selenium.

The silage employed was direct cut silage made in silo 1 and 2 with the additive AIV-2, containing 80 % formic acid and 2 % orthophosphoric acid; in silo 3 a mixture of enzymes and bacteria was used, and in silo 4 enzymes were used.

The grass was prepared in four covered bunker silos of concrete. The size of silos was 5 m by 10 m. The height of the silage stack was 2.5 m. The total content of grass varied in the silos from 13 400 to 14 700 kg dry matter. The grasses were cut on 9—10 June (silo 1) and on 16—17 June, 1986 (silos 2—4). The dry matter content of the grass used in the ensiling process was 17.9 % in silo 1 and 23.1 %, 22.8 % and 22.8 % in silos 2—4, respectively. The silage samples were taken at the end of 1986 and in the beginning of 1987.

The selenium content of grass in the silos was analyzed from ten seprate samples each consisting of 15—30 subsamples taken during filling of the silos. The effluent measurement consisted of 12 separate samples. From silo 1, the first effluent sample was collected on 9—12 June, 1986 and the last between 15 September and November, 1986. The respective time period for silos 2—4 was the same as for silo 1, except that the first sampling period was 17—20 June, 1986, which was the third sampling time for silo 1.

The silage samples were taken during the removal of silage from the silos for feeding. The silage samples represented silage used in the feeding during two weeks period. Thus, each silage samples consisted of 14 subsamples collected on each feeding day. Four samples from each silo were used for determination of the selenium concentration of the silage.

The selenium concentrations used as control values have been analyzed from grass and silage effluent of silage making in the previous year, in 1985. The grass used as the control received no selenium fertilization.

Soluble selenium in agricultural soil

The hot water soluble selenium in soils was ana-

lyzed by the Soil Fertility Service of Finland. The soil samples represent all soils found in Finland, but the weighting of collection varied in different sampling years in different parts of Finland.

The solution for selenium determination is obtained by extracting the soil with hot water (HESSE 1971). On average, about 4 % of the total selenium present in Finnish soil is extractable in hot water (YLÄRANTA 1983 b). Soil samples for selenium determinations were taken from the spring till the autumn in each sampling year.

Selenium analyses

The selenium determinations were carried out with the hydride method using an atomic absorption spectrophotometer (YLÄRANTA 1983 a).

RESULTS AND DISCUSSION

The natural selenium content of different agricultural crops varied in 1977 and 1978 from 0.001 to 0.034 mg/kg of dry matter (Table 1). The greatest means of selenium concentrations were measured in the fresh growth of timothy, rye grass and in swede tops.

Before the beginning of selenium fertilization, the selenium concentration of barley and

Table 1. Two-year averages of selenium content (µg/kg dry matter) of 12 crops grown side by side at nine sites (some of the crops were grown at 4 or 5 sites).

Crop	No. of sites	Se µg/kg dry matter		
		Mean	S.D.	Range
Spring wheat, grain	9	4.7	0.79	3.9—6.3
Spring wheat, straw	9	6.1	2.3	3.8—10.6
Oats, grain	9	5.7	1.3	4.3—8.4
Barley, grain	9	4.8	0.94	4.0—6.8
Timothy, silage, I cut	8	6.5	2.6	3.4—11.8
Timothy, silage, II cut	8	8.5	3.5	4.7—14.0
Timothy, dry hay	8	6.2	3.2	1.2—10.9
Timothy, fresh growth	8	11.0	4.9	4.9—21.0
Rye grass, silage, I cut	6	9.3	3.6	6.7—16.6
Rye grass, silage, II cut	6	12.4	3.4	7.8—17.3
Red clover, dry hay	5	6.0	3.1	3.4—12.6
Red clover, fresh growth	5	9.3	4.3	4.0—17.6
Pea, seed	8	6.4	3.3	4.1—14.1
Turnip rape, seed	5	8.1	2.8	4.8—11.3
Turnip rape, stalk	5	7.9	3.7	5.0—12.8
Swede, root	5	7.4	2.1	5.9—11.1
Swede, tops	5	20.1	9.2	13.8—33.8
Red beet, root	4	5.0	0.76	4.4—6.1
Carrot, root	5	7.8	2.1	5.2—8.3
Onion, bulb	7	6.9	1.4	5.4—9.3

oat grain in Finland was 0.004—0.016 mg/kg of dry matter (Table 2). After using selenium fertilizers for the first time, in 1985, the mean selenium content of barley grain, 0.16 mg/kg dry matter, was 20 times higher than before the use of selenated fertilizers. The results were similar in the case of oats, too.

The mean selenium content of barley and oat grain were similar in 1985 and 1987. The selenium content of barley and oat grain in 1986 seems to be, on average, slightly higher than in 1985 and 1987. The reason for this difference is very difficult to define. The growing seasons and the yields were different.

The average selenium contents of barley and oats were found to be higher than those estimated before selenium fertilization. That is only a positive finding. It shows that selenium fertilization works in practice as well as in the experimental fields. Further, a selenium level of 0.2 mg/kg of dry matter contributes better to the selenium need of man and animals than does 0.1 mg/kg.

The effect of selenium fertilization on the selenium content of spring cereals was very clear. Increasing the selenium content of winter wheat and rye with selenated fertilizers, using common cultivation measures, is a much more complicated task. It is recommended in Finland to fertilize winter cereals in the autumn so that the amount of nitrogen applied is only about 50 kg/ha. It means that the selenium addition comes to only about half of that applied to spring cereals.

The average selenium content in the grain of spring wheat in 1985 and 1986 was 0.23 mg/kg dry matter. The selenium concentration of winter wheat and rye has varied from 0.02 to 0.05 mg/dry matter (VARO et al. 1988).

In spring, farmers commonly use nitrogen fertilizers without added selenium for winter wheat and rye. Therefore, the selenium content of winter cereals in Finland is still low. Another reason for the small selenium concentration may be selenate reduction and fixing or selenate leaching in the soil during many autumn and spring months. These questions await intensive research work. In point of fact, a high selenium content of winter wheat and rye grain is not very important with respect to man's selenium intake in Finland.

Table 2. The mean selenium content of barley and oat grain in Finland from 1984 to 1987.

Crop and year		Number of samples	Se mg/kg dry matter	
			Mean	Range
Barley	1984	50	0.008	0.004—0.016
	1985	240	0.16	0.007—0.56
	1986	211	0.21	0.013—0.58
	1987	177	0.15	0.013—0.40
Oats	1984	49	0.009	0.004—0.015
	1985	200	0.15	0.008—0.41
	1986	216	0.20	0.005—0.52
	1987	171	0.13	0.020—0.45

Selenium content of grasses

Selenate fertilizer has a greater effect on the selenium content of grasses than on that of cereal crops. Therefore, more selenium was added to the fertilizers used mainly for cereal crops than to those used mainly for grassland cultivation. Because farmers fertilize grasses to some extent with fertilizers containing Se 16 mg/kg, the selenium content of grasses can increase up to 1 mg/kg of dry matter (Table 3).

Another reason for the high selenium content of grasses may be the exceptionally high fertilization rate, particularly for the first cut. The fertilization rate and also the amount of selenium added is, in general, lower for the second and third cuts. In addition, the residual effect of selenium application seems to be rather slight. Such being the case, it is easy to understand why the grass selenium content in the second and third cuts was lower than in the first cuts. In Finland, selenium was added only to the multinutrient fertilizers. Hence, the use of some nitrogen fertilizers not containing added selenium (ammonium nitrate and urea) may

Table 3. The mean selenium content of timothy grass dominated leys intended for both silage and hay from 1984 to 1988 in Finland.

Crop, cut and year		Number of samples	Se mg/kg dry matter	
			Mean	Range
Grass				
I cut	1984	86	0.024	0.010—0.053
	1985	57	0.33	0.044—1.38
	1986	104	0.17	0.027—0.46
	1987	101	0.25	0.015—0.91
Silage				
I cut	1984	100	0.028	0.012—0.081
	1985	100	0.24	0.036—0.90
	1986	103	0.14	0.038—0.41
	1987	104	0.21	0.032—0.89
	1988	116	0.16	0.043—0.39
II cut	1984	101	0.036	0.013—0.16
	1985	101	0.21	0.041—0.96
	1986	89	0.14	0.032—0.48
	1987	98	0.20	0.056—0.64
	1988	87	0.14	0.045—0.33
III cut	1984	98	0.030	0.011—0.12
	1985	101	0.17	0.042—0.58
	1986	93	0.12	0.025—0.43
	1987	102	0.17	0.028—0.77
	1988	104	0.11	0.018—0.13
Hay				
	1984	99	0.022	0.009—0.059
	1985	100	0.19	0.035—0.70
	1986	100	0.13	0.040—0.41
	1987	83	0.15	0.025—0.56
	1988	100	0.12	0.028—0.57

lead to a grass selenium content close to the natural level.

The selenium content of grasses of the first cut in 1984 was sometimes surprisingly high. The natural selenium level in grasses should be 0.01—0.02 mg/kg dry matter, as reported by OKSANEN and SANDHOLM (1970), ETTALA and KOSSILA (1979) and SIPPOLA (1979). One explanation is that, when cutting the grass with the mower chopper, some soil containing selenium was mixed into the grass samples. The farmers had the opportunity to purchase and to use selenated fertilizers at least for the third cut in 1984.

The mean selenium content of grass in 1985, 0.33 mg/kg of dry matter, was very much higher than that of silage in the same year, varying from 0.17 to 0.24 mg/kg of dry matter. This finding does not mean that much selenium has been lost into the silage effluents. A better explanation is that the samples were not very representative.

Because the fertilization varies on different farms, the limited amount of samples may be overly selected. We have to remember, too, that a small yield may cause a high selenium concentration in plants. The overall picture is that the selenium content of grasses and hay was lower in 1986 than in 1985.

The mean selenium content of grass samples in 1987 was 0.15—0.25 mg/kg dry matter. The highest selenium content was analyzed in the raw material of silage. That was also the case in 1985 and in 1986. The mean selenium contents of grass and silage in 1987 were higher than in 1986, but less than in 1985. The selenium content of silage in 1988 was of the same magnitude as in 1986. The grass selenium content was not analyzed in 1988.

The ensiling loss of selenium

The ensiling loss of selenium in different silos was 6—15 % (Table 4). Because the selenium content of silage was also measured in addition to the selenium content of grass, it was possible to compare the selenium losses by subtracting the selenium content of silage from the selenium content of the raw material. This is a very uncertain method, because a slight variation in the selenium contents has a powerful effect on the final result of selenium loss. However, obviously for the reason of even selenium content of silage raw material and silage itself, both methods gave selenium losses quite near to each other; for silo 1 the figures were (selenium loss measured in effluents mentioned first) 14.7 % and 12.7 %, being 6.2 % and 4.3 %, 7.5 % and 12.8 %, 5.9 % and 9.4 % for silos 2—4, respectively.

The greatest loss of selenium was measured

Table 4. The mean selenium content of grass and the mean ensiling loss of selenium in fresh grass silage (DM = dry matter).

Silo No.	Grass, DM		Silage, DM Se mg/kg	Effluent		Se loss %
	kg/silo	Se mg/kg		l/silo	Se µg/l	
Control*	—	0.020	—	—	0.89	—
Silo 1	13 730	0.203	0.217	18 430	29.8	14.7
Silo 2	14 670	0.172	0.181	6 240	26.2	6.2
Silo 3	13 370	0.181	0.190	6 540	27.4	7.5
Silo 4	13 740	0.183	0.193	5 420	26.9	5.9

* Values for the grass without selenium application.

in the silage prepared from young fresh grass (silo 1). The grasses cut later, 16—17 June, 1986, gave smaller selenium losses, e.g. 5.9—7.5 % (silos 2—4, Table 4). The dry matter loss varied between 10 and 19 %. It has been reported elsewhere that the average loss of dry matter during the period from ensiling the raw material until its removal from the silo for feeding is 20—30 % (ETTALA and KOSSILA 1980, RINNE 1989).

The average selenium loss during ensiling was slight compared with the losses of most mineral constituents reported in the literature (ETTALA and KOSSILA 1980, RINNE 1989). Because the dry matter losses are commonly greater than the selenium loss during ensiling, the selenium content of silage should be higher than that of raw materials. This seems not to be the case in the results of silages presented in Table 3. However, in the ensiling experiment, the selenium content of silage was slightly higher than in the raw material grass (Table 4).

Selenium fertilization and the environment

According to the results of field experiments, cereal crops above the ground accumulate 5—20 % of the selenium applied in the form of selenate fertilizer (YLÄRANTA 1985). The selenium uptake of grasses is about 20 %. The selenium fertilization in practice in Finland indicates that apparently the selenium intake of plants is clearly higher than the experimental figures. However, if only a small proportion of selenium applied to the soil is removed each year along with the crop, the selenium content of the soil will rise.

In acidic soils, added selenate is obviously converted fairly rapidly into a form that is poorly available to plants. In Finnish experiments on coarse mineral soils with high humus content, treatment with selenate had, however, a remarkable residual effect on the selenium content of barley the following growing season (YLÄRANTA 1985).

The selenium content of Finnish agricultural soils usually varies between 0.1 mg/kg and 0.6 mg/kg. On average, about 4 % of the total selenium present in the Finnish soil is extractable in hot water (YLÄRANTA 1983 b).

The annual selenium application is only about 10 g/ha or less. It will take many years before it is possible to analyze the increase of the total content of soil selenium. Therefore, it is more useful to control the plant available selenium fraction in the soil.

Farmers have annually ordered about 700—800 selenium analyses from the Soil Fertility Service of Finland during recent years. Since the addition of selenium to fertilizers, the annual amount of selenium analyses has decreased clearly (Table 5).

The selenated fertilizers were in use throughout the farming area for the first time during 1985. It was possible to use some selenium fertilizers already in 1984. Thus, the selenated soil samples are from 1984 till 1989. It is impossible to find any statistically significant increase

Table 5. Soil sample means for hot water extractable selenium content (Se mg/l of soil) in Finland in 1983—1989.

Soil group	Sampling year	Number of samples	Se mg/l soil
Coarse mineral soils	1982	646	0.007
	1983	514	0.007
	1984	603	0.007
	1985	286	0.009
	1986	155	0.008
	1987	178	0.006
	1988	111	0.010
	1989	273	0.006
Clay soils	1982	71	0.007
	1983	78	0.008
	1984	86	0.011
	1985	27	0.009
	1986	14	0.008
	1987	5	0.005
	1988	7	0.011
	1989	55	0.006
Organogenic soils	1982	94	0.007
	1983	102	0.006
	1984	128	0.010
	1985	69	0.008
	1986	38	0.007
	1987	30	0.009
	1988	40	0.008
	1989	44	0.006
Totally	1982	811	0.007
	1983	694	0.007
	1984	817	0.008
	1985	382	0.009
	1986	207	0.008
	1987	213	0.007
	1988	158	0.009
	1989	372	0.006

in selenium contents caused by selenium fertilization.

Addition of selenium to the environment may also result in organisms experiencing a high initial intake of selenium. The importance of this fact depends very much upon how fast the selenium is eliminated after application. GISSEL-NIELSEN and GISSEL-NIELSEN (1973) have studied this elimination rate, which can be expressed by the biological half-life of selenium.

Samples representing mammals, birds, fishes, insects and annelids were included in the experiment. The biological half-life of selenium for these animals varied between 10 and 64 days, which is a short period when compared with the annual addition of selenium to the field.

Another problem that arises when the ecological effect of selenium addition is considered is whether an increase in the total selenium content of an ecosystem would alter the relative distribution of selenium between the inhabitants and cause accumulations in certain organisms. This was studied in a field experiment and a fresh water system (GISSEL-NIELSEN and GISSEL-NIELSEN 1973).

In the field, 75 g of selenium was added per hectare every year over a period of five years, which resulted in doubling of the selenium content in the soil. Soil arthropods (woodlouse, centipede, and crane fly larvae) were collected from the test field and from control fields. The animals were analyzed for selenium. The increase in the concentration for the three soil arthropods was roughly the same as the increase in the soil selenium, which indicates that an equilibrium was retained.

The results of the experiment of WATKINSON and DIXON (1979) are consistent with the findings of GISSEL-NIELSEN and GISSEL-NIELSEN (1973).

An unexpected effect of selenium fertilization could be an increased concentration of selenium in lake and river waters. It is very likely that long-term elevation of waterborne selenium to 8—10 µg/l can be harmful for the environment (LEMLY 1985). In addition to this, enrichments with selenium seem to improve the growth of freshwater green algae (LINDSTRÖM 1984).

It has been stressed by LEMLY (1985): "With the potential for significant biological magnification in food chains, a reasonable speculation would put the Maximum Acceptable Toxicant Concentration (MATC) — no observable long-term effects from dietary or waterborne exposure — for waterborne selenium within the range of 2—5 µg Se/liter (as total recoverable selenium in filtered water samples), or about 5—10 times background levels."

The National Board of Waters and the Environment has followed the selenium content in different natural waters since the beginning of selenium fertilization, without finding any selenium value above the determination level of 1 µg Se/l of water. The method of selenium determination used has now been improved.

The Public Health Institute has analyzed selenium concentrations below 1 µg Se/l, often only 0.1—0.2 µg Se/l, in the Finnish natural waters (ARO 1990). Commonly, most natural waters tend to have low concentrations of Se, less than 0.01 mg/l. Notable exceptions may occur if waters are alkaline or if they leach and drain seleniferous rocks and soils (NCNEAL and BALISTRIERI 1989).

It seems not to be easy to find a sound scientific basis for the fear that selenium fertilization would pose a serious environmental risk in Finland. It is wrong to compare the Finnish acid and reducing soil conditions to conditions where selenium can remain or even oxidize to the valence state six, which is a very soluble form of selenium. This kind of situation occurs primarily under arid or semi-arid conditions.

Thus, from the ecological point of view, field treatment with physiological levels of selenium has no negative impact on the environment (GISSEL-NIELSEN et al. 1984).

On the contrary, increasing the low concentration of selenium can have beneficial effects on various ecosystems. The role of selenium as a detoxifying microelement, e.g. in natural waters, by forming insoluble metal selenides, cannot be excluded (RUDD et al. 1980).

The serious examples of selenium toxicity, for instance, at the Kesterson Reservoir in the San Joaquin Valley of California (OHLENDORF 1989) serve to show us that we have to follow the selenium situation in our environment very carefully.

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REFERENCES

ARO, A. Personal communication with A. Aro, Public Health Institute, Mannerheimintie 166, SF-00300 Helsinki, Finland, January 1990.

ETTALA, E. & KOSSILA, V. 1979. Mineral content in heavily nitrogen fertilized grass and its silage. Ann. Agric. Fenn. 18: 252—262.

— & KOSSILA, V. 1980. Organic matter, mineral and nitrogen losses from fresh grass silage during ensiling. Ann. Agric. Fenn. 19: 9—20.

GISSEL-NIELSEN, G. & GISSEL-NIELSEN, M. 1973. Ecological effects of selenium application to field crops. Ambio 2: 114—117.

—, GUPTA, U.C., LAMAND, M. & WESTERMARCK, T. 1984. Selenium in soils and plants and its importance in livestock and human nutrition. Adv. Agron. 37: 397—460.

HESSE, P.R. 1971. A textbook of soil chemical analysis. 520 p. London.

LEMLY, A.D. 1985. Toxicology of selenium in a freshwater reservoir: Implications for environmental hazard evaluation and safety. Ecotoxicol. Environ. Safety 10: 314—338.

LINDSTRÖM, K. 1984. Selenium and algal growth. Proc. 3rd Int. Symp. Industrial uses of selenium and tellurium. Stockholm, October 15—17, 1984. p. 441—469.

MCNEAL, J.M. & BALISTRIERI, L.S. 1989. Geochemistry and occurrence of selenium: An overview. Proc. Symp. Selenium in agriculture and the environment. New Orleans, LA, 2 Dec. 1986. Ed. Jacobs, L. W. Soil Sci. Soc. Amer. Spec. Publ. 23: 1—13.

OHLENDORF, H.M. 1989. Bioaccumulation and effects of selenium in Wildlife. Proc. Symp. Selenium in agriculture and the environment. New Orleans, LA, 2 Dec. 1986. Ed. Jacobs, L. W. Soil Sci. Soc. Amer. Spec. Publ. 23: 133—177.

OKSANEN, H.E. & SANDHOLM, M. 1970. The selenium content of Finnish forage crops. J. Scient. Agric. Soc. Finl. 42: 250—253.

RINNE, K. 1989. Ensiling losses in fresh grass silage. Proc. XVI Intern. Grassland Congr., Nice, France, p. 981—982.

RUDD, J.W.M., TURNER, M.A., TOWNSEND, B.E., SWICK, A. & FURUTANI, A. 1980. Dynamics of selenium in mercury-contaminated experimental freshwater ecosystems. Can. J. Fish. Aquat. Sci. 37: 848—857.

SIPPOLA, J. 1979. Selenium content of soils and timothy (*Phleum pratense* L.) in Finland. Ann. Agric. Fenn. 18: 182—187.

VARO, P., ALFTHAN, G., EKHOLM, P., ARO, A. & KOIVISTOINEN,

P. 1988. Selenium intake and serum selenium in Finland: effects of soil fertilization with selenium. Am. J. Clin. Nutr. 48: 324—329.
WATKINSON, J.H. & DIXON, G.M. 1979. Effect of applied selenate on ryegrass and on larvae of solider fly, *Inopus rubriceps*. Macquart. N.Z.J. Exp. Agric. 7: 321—325.
YLÄRANTA, T. 1983 a. The hydride method for measuring the selenium content of plants. Ann. Agric. Fenn. 22: 18—28.
— 1983 b. Selenium in Finnish agricultural soils. Ann. Agric. Fenn. 22: 122—136.
— 1985. Increasing the selenium content of cereal and grass crops in Finland. Diss. 72 p.
— & SILLANPÄÄ, M. 1984. Micronutrient contents of different plant species grown side by side. Ann. Agric. Fenn. 23: 158—170.

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SELOSTUS

Seleenilannoituksen vaikutus eräiden viljelykasvien seleenipitoisuuteen ja ympäristöön

TOIVO YLÄRANTA

Maatalouden tutkimuskeskus

Suomessa alettiin lisätä selenaattimuotoista seleeniä peltoja puutarhaviljelyssä käytettäviin moniravinnelannoitteisiin vuonna 1984.

Viljelykasvien seleenipitoisuus oli ennen seleenilannoitusta keskimäärin alle 20 µg/kg kuiva-ainetta. Seleenilannoitus on kohottanut ohran ja kauran jyvien sekä nurmirehun seleenipitoisuuden keskimäärin 0.1—0.2 mg:aan/kg kuiva-ainetta.

Säilörehun valmistuksessa menetetään puristenesteen mukana kymmeniä prosentteja nurmirehun sisältämistä kivennäisaineista. Näihin lukuihin verrattuna oli seleenitappio pieni, neljässä eri siilossa vain 6—15 %, valmistettaessa säilörehua koiranheinä-timoteinurmesta.

Seleenin lisäämistä lannoitteisiin on pidetty jopa ympäristölle haitallisena toimenpiteenä. Seleenilannoitus on ollut käytössä täyspainoisesti viiden vuoden ajan. Tänä aikana ei ole ilmennyt mitään sellaista seikkaa, joka olisi esteenä seleenilannoituksen jatkamiselle.

Maasta kuumaan veteen uuttuvan seleenin pitoisuuden, joka kuvaa kasveille käyttökelpoista osaa maan sisältämästä seleenistä, ei ole havaittu kohonneen seleenilannoituksen vaikutuksesta. Luonnon vesienkään seleenipitoisuuden, joka on alle 1µg/l, usein 0.1—0.2 µ/l, ei ole osoitettu kohonneen seleenilannoituksen aikana.

ANNALES AGRICULTURAE FENNIAE, VOL. 29: 141—149 (1990)

Seria AGROGEOLOGIA ET CHIMICA N. 156 — Sarja MAA JA LANNOITUS n:o 156

EFFECTS OF LIMING AND THE ADDITION OF SULPHATE AND PHOSPHATE ON THE SELENIUM CONTENT OF ITALIAN RYE GRASS

TOIVO YLÄRANTA

YLÄRANTA, T. 1990. **Effects of liming and the addition of sulphate and phosphate on the selenium content of Italian rye grass.** Ann. Agric. Fenn. 29: 141—149. (Inst. Crop and Soil Sci., SF-31600 Jokioinen, Finland.)

The effects of liming and the application of sulphate and phosphate on the selenium content of Italian rye grass *(Lolium multiflorum* Lam.), grown on clay, sandy loam and Carex peat with naturally low selenium contents, was studied in a pot experiment. Selenium was added to the first crop in the form of either sodium selenite or selenate, sulphur in the form of calcium sulphate and phosphorus in the form of calcium dihydrogen phosphate.

In the pots to which no selenium was added, the mean selenium contents of rye grass varied between 8 and 10 µg/kg dry matter. In selenate treatments without liming or sulphate and phosphate additions, the Se concentrations varied from 1.45 mg/kg dry matter to 2.90 mg/kg dry matter. The selenium concentrations in the selenite treatment were only 7—18 % of those measured in the selenate treatment. There was a clear decrease in the Se concentrations from the first to the second cut.

The addition of sulphate decreased the selenium concentrations in selenate treatment on all soils in both cuts. The addition of phosphate decreased the Se concentrations in the selenate treatment only in the second cut on clay and sandy loam.

In selenite treatments, liming increased the selenium concentration in all soils in both cuts, but the increase was statistically significant only on clay and peat soil in the second cut. In the selenate treatment, liming decreased the selenium concentration in the first cut on all soils and in the second cut on sandy loam soil.

Index words: selenite and selenate application, liming, sulphate and phosphate addition, selenium content, Italian rye grass.

INTRODUCTION

In soil, selenite and selenate are the most readily available sources of selenium for plants (CARY and ALLAWAY 1969, GISSEL-NIELSEN and BISBJERG 1970). Selenite binds strongly with the hydrous sesquioxides (CARY et al. 1967, GEERING et al. 1968), with organic matter (HAMDY and GISSEL-NIELSEN 1976, JOHN et al. 1976), and with clay minerals in soil (HAMDY and GISSEL-NIELSEN 1977). Selenate is as much as 10—20 times more soluble than selenite (CARY and GISSEL-NIELSEN 1973).

The chemical form of selenium in soil depends largely on soil pH and on the redox potential. The predominant mobile inorganic forms of selenium are selenate in aerated alkaline soils, and selenite in acid, reducing soils such as those generally occurring in humid areas (GEERING et al. 1968).

The Finnish agricultural soils are mainly acidic in nature, so that liming can be assumed to increase the uptake of selenium by plants. In pot experiments in wich liming with $Ca(OH)_2$ was used to raise the pH($CaCl_2$) of clay, fine sand and Carex peat from 4.4—4.6 to 6.1—6.3, liming raised the selenium content of rye grass only in the selenite treatment of Carex peat (YLÄRANTA 1983 c). In the field experiments carried out by KORKMAN (1980), liming had a minor effect on the uptake of selenium from selenite by spring wheat and barley.

Although the use of fertilizers containing phosphate and sulphate should not be expected to bring about major changes in the solubility of soil selenium (CARY and GISSEL-NIELSEN 1973), they may cause some interactions in soil.

It is rather evident that selenate has analogies with sulphate. The appropriate analogy for selenite is phosphate (BARROW and WHELAN 1989). It has been found that sulphur, in the form of sulphate, effectively reduces the uptake of selenium by plants from selenate, but reduces only slightly that from selenite (e.g. GISSEL-NIELSEN 1973). Correspondingly, selenate can be assumed to interfere with the sulphur economy of plants (MILCHUNAS et al. 1983). This has raised further doubts about the suitability of applying selenate to soil as a source of selenium for plants.

The selenium content of cultivated crops can be raised most effectively by adding selenites or selenates into fertilizers, by spraying these salts onto the crops or by treating the seeds with aqueous solutions of these selenium compounds (BISBJERG and GISSEL-NIELSEN 1969, GISSEL-NIELSEN and BISBJERG 1970, GISSEL-NIELSEN 1977). In Finland, the addition of selenate to the most common multinutrient fertilizers used in agriculture and horticulture was started in 1984 (YLÄRANTA 1985).

Most of the selenium added to the acid Finnish agricultural soils in the form of selenite fixes quite rapidly into a form that is poorly available to plants. Sorption of selenite by soil seems to be weaker, however, than that of phosphate. Selenium added to soil in the form of selenate remains in a readily soluble form for several months. The results of some field experiments even suggest that some selenate is still available to other plants the following growing season (YLÄRANTA 1985). It would be very important to study in greater detail the reduction and fixation speed of added selenate under different conditions.

In a pot experiment in peat, clay and sandy loam soil, an effort was made to get a clearer picture about the effects of liming, sulphate and phosphate on the selenium content of plants. It was hoped that new information would be obtained by using much more powerful liming and sulphate and phosphate additions than are used in practical cultivation measures.

MATERIAL AND METHODS

The soils used in the pot experiment were Carex peat, sandy loam and clay. The particle size distributions of the mineral soils were determined using the pipette method of ELONEN (1971):

Particle size (ø)	Clay	Sandy loam
<0.002 mm	49 %	16 %
0.002—0.02	17	9
0.02 —0.06	16	10
0.06 —0.2	9	43
0.2 —2	9	22

The Carex peat had a degree of humification of H_{9-10} on the von POST scale, and contained 27.2 % inorganic matter as determined by ignition at 500 °C overnight. The clay contained 3.5 % organic carbon and the sandy loam 3.8 %. The pH($CaCl_2$) of the clay and sandy loam was 4.8 and of the Carex peat 4.0.

The following amounts of calcium, potassium, magnesium and phosphorus were extracted from the experimental soils into an acidic ammonium acetate solution (0.5 M CH_3COONH_4, 0.5 M CH_3COOH, pH 4.65; VUORINEN and MÄKITIE 1955):

	Extracted nutrients, mg/l of soil			
	Ca	K	Mg	P
Clay	1570	340	340	7.2
Sandy loam	1610	300	190	33.6
Carex peat	1450	110	410	7.6

The natural selenium content of the clay was 0.59 mg/kg, of the sandy loam 0.22 mg/kg and of the Carex peat 0.29 mg/kg.

Two-litre polythene pots were used in the experiment. There were four replicates, the plan being as follows:

Control, no addition of selenium
Selenite addition
Selenate addition
Selenite + liming
Selenate + liming
Selenite + phosphate
Selenate + phosphate
Selenite + sulphate
Selenate + sulphate

In the experiment, 2 200 g of a lightly moist clay soil (1 667 g of dry soil), 2 375 g of sandy loam soil (2 232 g) and 1 400 g of Carex peat (562 g) were weighed out per pot. The depth of soil in the pot was 12 cm. There were one selenite and one selenate addition for each soil, making a total of 108 pots.

The liming (1), sulphate addition (2), phosphate addition (3) and selenite (4) and selenate (5) additions were as follows:

	Carex peat	Clay	Sandy loam
1. $Ca(OH)_2$, g/pot	13.5	8.5	7.5
2. $CaSO_4 \cdot 2H_2O$, S mg/pot	800	800	800
3. $Ca(H_2PO_4)_2 \cdot 2H_2O$, P mg/pot	800	800	800
4. $Na_2SeO_3 \cdot 5H_2O$, Se mg/pot	0.200	0.200	0.200
5. $Na_2SeO_4 \cdot 10H_2O$, Se mg/pot	0.200	0.200	0.200

The aim of liming was to increase the pH($CaCl_2$) of the soils by around two pH units. Liming was carried out using analytical grade $Ca(OH)_2$ powder. The amount of lime required was determined by the $Ca(OH)_2$ titration method (MÄNTYLAHTI and YLÄRANTA 1980). However, the amount of $Ca(OH)_2$ powder actually added was 40 % higher than this figure in the light of experience gained in the laboratory with incubation experiments.

Sulphur and phosphorus were added in the form of calcium compounds, as calcium is the most common exchangeable basic cation in soil, and thus exerts the minimum effect on the experimental conditions.

The calcium hydroxide, calcium sulphate and calcium dihydrogen phosphate were mixed carefully through the whole amount of soil in the pot. About 250 ml of soil was taken from every pot for covering the seeds after sowing. Fertilizers were mixed into the remaining soil in each pot as follows:

Nutrient	mg/pot	Compound
N	470	NH_4NO_3, $(NH_4)_2HPO_4$
K	350	KCl, K_2SO_4
Mg	90	$MgCl_2 \cdot 6H_2O$
Ca	60	$CaCl_2 \cdot 2H_2O$
Fe	2	FeNa-EDTA
Mn	3.3	$MnSO_4 \cdot H_2O$
Cu	2.6	$CuSO_4 \cdot 5H_2O$
Zn	2.3	$ZnSO_4 \cdot 7H_2O$
B	0.35	H_3BO_3
Mo	0.4	$Na_2MoO_4 \cdot 2H_2O$
P	80	$(NH_4)_2HPO_4$
S	80	K_2SO_4, $MnSO_4 \cdot H_2O$, $CuSO_4 \cdot 5H_2O$, $ZnSO_4 \cdot 7H_2O$

The pots were treated with 5 ml aqueous sodium selenite solution (Merck, product number 6607) or 5 ml aqueous sodium selenate solution (BDH 10262). Selenium solutions were mixed into the soils separately after nutrient application.

Fifty-five seeds (200 mg) of Italian rye grass (*Lolium multiflorum* Lam., cv. Leda daehnfeldt) were planted in each pot. The seeds were covered with a thin layer of the experimental soil. The pots were watered to a moisture content corresponding to about pF 2. This was done daily throughout the entire growing period.

The pots were placed randomly on the tables in the greenhouse. The temperature of the greenhouse was adjusted to 20 °C in the daytime and to about 16 °C at night. The lighting, which was improved using daily high pressure sodium lamps and multielement lamps (OY AIRAM AB, SNaT-400 W, HgA-400 W), varied between 7 and 17 klux.

The seeds were sown on 21 August, 1987. The first cutting took place on 6 October and the second on 13 November.

The following nutrients were added to each pot two days after cutting the first crop: N 290 mg (NH_4NO_3), P 40 mg ($(NH_4)_2HPO_4$) and K 200 mg (KCl). No abnormalities were observed in the plants during growing.

The crops were cut at the silage stage. The shoots were cut with scissors leaving 2 cm of stubble, and the crops were dried in paper bags, in ovens provided with air circulation, for two days at 60 °C.

At the end of the experiment the contents of the pots were removed, and the pH($CaCl_2$) values were determined for each pot.

The selenium content of soil and plant samples was determined with the hydride method (YLÄRANTA 1983 a). For the determination of total selenium the soil samples were decomposed using a modification (YLÄRANTA 1983 b) of the method described by AGEMIAN and BEDEK (1980).

Statistical analysis of the results was carried out using the "ANOVA-1" software of the MSTAT-C Microcomputer Statistical Programme (Michigan State University, MI, U.S.A.).

For comparison of means of the results obtained in the experiment, DUNCAN's (1955) test was applied at the 5 % level of significance.

RESULTS

The average dry matter yields per pot obtained varied in the first cut from 6.4 g to 11.5 g and in the second cut from 7.4 to 13.0 g (Table 1). In the first cut the smallest yield was measured for clay soil and the highest for sandy loam soil. In the second cut the biggest yield was grown on sandy loam soil and the smallest on peat soil.

There were few statistically significant differences in both yields of rye grass grown on clay soil and in the first yield of grass grown on sandy loam and peat soil. In the first crop on clay soil, the reduction in yield could have been due to a phosphorus deficiency. Many of the other yield reductions are difficult to explain, because sometimes the yield was higher in the selenite and sometimes in the selenate treatments. The cause may possibly be mere coincidence.

The pH($CaCl_2$) of the unlimed clay and sandy loam soils at the end of the experiment averaged 4.7 and 4.8, and that of limed soils 6.4 and 6.5 (Table 2). The corresponding pH values for peat soil were 3.9 and 5.8. There were no statistically significant differences in pH values between the unlimed treatments for any type of soil. Sulphate addition clearly increased the content of soluble salts in all soils (Table 2).

In the pots to which no selenium was added,

Table 1. Mean yields of rye grass crops, g of dry matter. Figures given for both cuts for each soil not marked with a common letter differ from each other at the 5 % level of significance (DUNCAN 1955).

Treatments	I cut			II cut		
	Clay	Sandy loam	Carex peat	Clay	Sandy loam	Carex peat
Control, no Se addition	6.5^{c}	10.8^{ab}	8.1^{bcd}	10.8^{ab}	11.8^{a}	7.8^{ab}
Selenite addition	6.5^{c}	9.0^{c}	7.4^{d}	9.3^{b}	11.2^{a}	7.8^{ab}
Selenate addition	7.0^{bc}	11.0^{ab}	8.4^{bc}	10.5^{ab}	10.3^{a}	7.4^{b}
Selenite + liming	7.4^{b}	9.5^{bc}	9.5^{a}	10.5^{ab}	10.8^{a}	9.4^{a}
Selenate + liming	6.4^{c}	8.8^{c}	8.1^{bcd}	9.7^{ab}	11.1^{a}	9.6^{a}
Selenite + phosphate	8.6^{a}	9.9^{abc}	9.0^{ab}	11.0^{ab}	11.4^{a}	8.6^{ab}
Selenate + phosphate	8.6^{a}	10.2^{abc}	8.7^{abc}	11.7^{a}	11.7^{a}	8.9^{ab}
Selenite + sulphate	6.4^{c}	11.5^{a}	7.8^{cd}	10.1^{ab}	13.0^{a}	7.9^{ab}
Selenate + sulphate	7.1^{bc}	11.0^{ab}	8.2^{bcd}	10.6^{ab}	12.3^{a}	8.3^{ab}

Table 2. Mean pH($CaCl_2$) and electrical conductivity (Ec, 10^{-4} S/cm) of soils in different treatments after the second cut. The pH and Ec values given for each soil and not marked with a common letter differ from each other at the 5 % level of significance (DUNCAN 1955).

Treatments	Clay		Sandy loam		Carex peat	
	pH	Ec	pH	Ec	pH	Ec
Control, no Se addition	4.7^{b}	0.41^{c}	4.7^{b}	0.35^{c}	3.9^{b}	1.27^{b}
Selenite addition	4.8^{b}	0.36^{c}	4.7^{b}	0.42^{c}	3.9^{b}	1.21^{b}
Selenate addition	4.7^{b}	0.31^{c}	4.7^{b}	0.37^{c}	3.9^{b}	1.40^{b}
Selenite + liming	6.4^{a}	0.87^{b}	6.5^{a}	0.83^{b}	5.8^{a}	1.51^{b}
Selenate + liming	6.4^{a}	0.79^{b}	6.5^{a}	0.88^{b}	5.8^{a}	1.58^{b}
Selenite + phosphate	4.8^{b}	0.38^{c}	4.7^{b}	0.60^{bc}	3.9^{b}	1.34^{b}
Selenate + phosphate	4.8^{b}	0.37^{c}	4.7^{b}	0.48^{bc}	3.9^{b}	1.15^{b}
Selenite + sulphate	4.8^{b}	4.22^{a}	4.7^{b}	5.90^{a}	3.9^{b}	5.79^{a}
Selenate + sulphate	4.7^{b}	4.03^{a}	4.7^{b}	5.95^{a}	3.9^{b}	5.89^{a}

Table 3. Mean selenium contents of rye grass crops, mg/kg dry matter. Figures given for both cuts for each soil applied with selenium and not marked with a common letter differ from each other at the 5 % level of significance (DUNCAN 1955).

Treatments	I cut			II cut		
	Clay	Sandy loam	Carex peat	Clay	Sandy loam	Carex peat
Control, no Se addition*	0.009	0.010	0.009	0.008	0.009	0.010
Selenite addition	0.159^{de}	0.359^{d}	0.523^{cd}	0.199^{d}	0.359^{d}	0.217^{c}
Selenate addition	2.19^{a}	2.56^{a}	2.90^{a}	1.45^{a}	1.75^{a}	1.62^{a}
Selenite + liming	0.278^{cd}	0.396^{d}	0.725^{c}	0.303^{c}	0.367^{d}	0.596^{b}
Selenate + liming	1.89^{b}	2.03^{b}	1.94^{b}	1.39^{ab}	1.28^{b}	1.46^{a}
Selenite + phosphate	0.144^{de}	0.334^{d}	0.238^{d}	0.160^{d}	0.418^{d}	0.155^{c}
Selenate + phosphate	2.10^{a}	2.55^{a}	2.63^{a}	1.30^{b}	1.43^{b}	1.47^{a}
Selenite + sulphate	0.120^{e}	0.335^{d}	0.438^{cd}	0.182^{d}	0.362^{d}	0.273^{c}
Selenate + sulphate	0.391^{c}	1.11^{c}	0.680^{c}	0.352^{c}	0.726^{c}	0.634^{b}

* The control treatment is not included in the statistical comparison.

the mean selenium concentrations of rye grass varied between 8 and 10 µg/kg dry matter (Table 3).

In the selenite treatment without liming and sulphate and phosphate additions, the Se concentrations varied from 0.16 mg/kg to 0.52 mg/kg dry matter. The highest selenium concentration was measured in the first cut on peat soil. The Se concentrations were lower in the second cut compared to the first cut, except for that from sandy loam soil, where the mean selenium concentration was 0.36 mg/kg dry matter in both cuts.

In selenate treatments without liming and sulphate and phosphate additions, the Se concentrations varied from 1.45 mg/kg dry matter to 2.90 mg/kg dry matter. There was a clear decrease in the Se concentrations from the first to the second cut. The Se concentrations in the selenite treatment were only 7—18 % of those measured in the selenate treatment. In the treatments where liming material, sulphate and phosphate were added, a common feature was that the Se concentrations were higher or equal in the first cut compared to the second cut.

The addition of sulphate clearly decreased the selenium concentrations in the selenate treatment, on all soil types in both cuts. The selenium concentration of rye grass in the selenate treatment with the application of sulphate was only 18—43 % of the figures obtained without the addition of sulphate. The addition of phosphate decreased the Se concentrations only in the second cut in the selenate treatment on clay and sandy loam soil.

Liming increased the selenium concentration of the selenite treatments in all soils in both cuts, but the increase was statistically significant only on clay and peat soil in the second cut. In the selenate treatments, liming decreased the selenium concentration in the first cut on all soils, and in the second cut on sandy loam soil.

DISCUSSION

SMITH and WATKINSON (1984) reported that 48 and 320 µg Se/g dry matter of shoots — selenium applied as selenite and selenate, respectively — cause a 10 % reduction in dry matter yield for perennial rye grass *(Lolium perenne* L.). In this study, the selenium concentrations

were much lower than those, and they did not cause any measureable yield reduction in annual rye grass.

The adsorption capacity of clay minerals and sesquioxides decreases with increasing pH, thereby leaving more selenium available in soil to the plants. Thus liming increases the solubility of selenium in soil (CARY et al. 1967, GEERING et al. 1968, HINGSTON et al. 1971, FROST and GRIFFIN 1977). In a port trial by MORÉ and COPPENET (1980), where soil pH(H_2O) was increased from 5.2 to 6.5, the Se content of Italian rye grass at the grazing stage was significantly increased.

The effect of liming on the selenium content of rye grass was very slight. In this respect, the results are consistent with the results of the earlier pot and field experiments conducted in Finland by KORKMAN (1980) and YLÄRANTA (1983 c).

In the pot experiments carried out by YLÄRANTA (1983 c), the uptake of selenium by rye grass from selenate diminshed on Carex peat but not on either clay or fine sandy soil following the addition to the soil of 400 mg of sulphur, as calcium sulphate, per litre of soil.

In this pot experiment, the addition of sulphate on the clay, sandy loam and peat soil clearly decreased the selenium uptake by rye grass in the selenate treatment. These results confirm the results reported in the literature (e.g. GISSEL-NIELSEN 1973).

We should still be very cautious when drawing final conclusions concerning the effect of sulphate on the selenium uptake by plants. In practical cultivation, the fertilizers and the soils usually do not contain sulphate concentrations as high as in this pot experiment. Besides, the fixation of sulphate anion in soil is weak. Thus, sulphate is susceptible to leaching, which thus decreases the soluble sulphate concentrations in the soils.

For selenate, the appropriate analogy is sulphate. The binding constant for the SeO_4^{2-} ion is small, and so the net effect is weak adsorption with soil. Sorption also decreases appreciably with increasing pH because of the decreasing electric potential of the reacting surfaces. However, agronomic experience with sulphate would not be directly applicable to predict the behaviour of selenate as a fertilizer because the levels of application of selenate would be 1000-fold smaller (BARROW and WHELAN 1989).

The limited effect of the addition of sulphate on the selenium uptake by rye grass in the experiment conducted by YLÄRANTA (1983 c) may result from the differences in the soils and soil properties and from the way selenium was added into the pots. YLÄRANTA (1983 c) did not mix the selenium compounds into the whole soil in the pot. Therefore, the plants could obviously take the selenium from a higher concentration than in this pot experiment.

The exact mechanisms for the interaction between P and Se in soil are not known (CARTER et al. 1972). The application of phosphate has given contradictory results with regard to its effect on Se uptake by plants (ELRASHIDI et al. 1989). Sometimes the application of phosphate has increased the availability of Se on the treated soils, and sometimes Se uptake by plants has been reduced by the application of phosphate.

Many researchers have found that additions of phosphorus to soil increase the accumulation of selenium in plants. Phosphate is sorbed by soil more strongly than selenite (RĀJAN and WATKINSON 1976). When phosphorus is added to the soil, it evidently replaces some selenium on certain sorption sites, making selenium more available to plants.

In spite of plentiful addition of phosphorus to soils, the effect of phosphorus on the selenium uptake by rye grass was small in all soils. Hence, it is difficult to contend that phosphorus could play an important role in the uptake of selenium by plants under Finnish conditions.

REFERENCES

AGEMIAN, H. & BEDEK, E. 1980. A semi-automated method for the determination of total arsenic and selenium in soils and sediments. Anal. Chim. Acta 119: 323—330.

BARROW, N.J. & WHELAN, B.R. 1989. Testing a mechanistic model. VII. The effects of pH and of electrolyte on the reaction of selenite and selenate with a soil. J. Soil Sci. 40: 17—28.

BISBJERG, B. & GISSEL-NIELSEN, G. 1969. The uptake of applied selenium by agricultural plants. I. The influence of soil type and plant species. Plant and Soil 31: 287—298.

CARTER, D.L., ROBBINS, C.W. & BROWN, M.J. 1972. Effect of phosphorus fertilization on the selenium concentration in alfalfa *(Medicago sativa)*. Soil Sci. Soc. Amer. Proc. 36: 624—628.

CARY, E.E. & ALLAWAY, W.H. 1969. The stability of different forms of selenium applied to low-selenium soils. Soil Sci. Soc. Amer. Proc. 33: 571—574.

— & GISSEL-NIELSEN, G. 1973. Effect of fertilizer anions on the solubility of native and applied selenium in soil. Soil Sci. Soc. Amer. Proc. 37: 590—593.

—, WIECZOREK, G.A. & ALLAWAY, W.H. 1967. Reactions of selenite-selenium added to soils that produce low selenium forages. Soil Sci. Soc. Amer. Proc. 31: 21—26.

DUNCAN, D.B. 1955. Multiple range and multiple F tests. Biometrics 11: 1—42.

ELONEN, P. 1971. Particle-size analysis of soil. Acta Agr. Fenn. 122: 1—122.

ELRASHIDI, M.A., ADRIANO, D.C. & LINDSAY, W.L. 1989. Solubility, speciation, and transformations of selenium in soils. Proc. Symp. Selenium in agriculture and the environment. New Orleans, LA, 2 Dec. 1986. Ed. Jacobs, L. W. Soil Sci. Soc. Amer. Spec. Publ. 23: 51—63.

FROST, R.R. & GRIFFIN, R.A. 1977. Effect of pH on adsorption of arsenic and selenium from landfill leachate by clay minerals. Soil Sci. Soc. Amer. J. 41: 53—57.

GEERING, H.R., CARY, E.E., JONES, L.H.P. & ALLAWAY, W.H. 1968. Solubility and redox criteria for the possible forms of selenium in soils. Soil Sci. Soc. Amer. Proc. 32: 35—40.

GISSEL-NIELSEN, G. 1973. Uptake and distribution of added selenite and selenate by barley and red clover as influenced by sulphur. J. Sci. Food Agric. 24: 649—655.

— 1977. Control of selenium in plants. Risø Report 370. 42 p., 13 app.

— & BISBJERG, B. 1970. The uptake of applied selenium by agricultural plants. 2. The utilization of various selenium compounds. Plant and Soil 32: 382—396.

HAMDY, A.A. & GISSEL-NIELSEN, G. 1976. Relationships between soil factors and selenium content of Danish soils and plants. Risø Report 349. 13 p.

— & GISSEL-NIELSEN, G. 1977. Fixation of selenium by clay minerals and iron oxides. Z. Pfl.ernähr. Bodenkunde 140: 63—70.

HINGSTON, F.J., POSNER, A.M. & QUIRK, J.P. 1971. Competitive adsorption of negatively charged ligands on oxide surfaces. Disc. Faraday Soc. 52: 334—342.

JOHN, M.K., SAUNDERS, W.M.H. & WATKINSON, J.H. 1976. Selenium adsorption by New Zealand soils. I. Relative adsorption of selenite by representative soils and the relationship to soil properties. N. Z. J. Agric. Res. 19: 143—151.

KORKMAN, J. 1980. The effect of selenium fertilizers on the selenium content of barley, spring wheat and potatoes. J. Sci. Agric. Soc. Finl. 52: 495—504.

MILCHUNAS, D.G., LAUENROTH, W.K. & DODD, J.L. 1983. The interaction of atmospheric and soil sulfur on the sulfur and selenium concentration of range plants. Plant and Soil 72: 117—125.

MORÉ, É. & COPPENET, M. 1980. Teneurs en sélénium des plantes fourragères. Influence de la fertilisation et des apports de sélénite. Ann. Agron. 31: 297—317.

MÄNTYLAHTI, V. & YLÄRANTA, T. 1980. The estimation of soil lime requirement in soil testing. Ann. Agric. Fenn. 19: 92—99.

RĀJAN, S.S.S. & WATKINSON, J.H. 1976. Adsorption of selenite and phosphate on an allophane clay. Soil Sci. Soc. Amer. J. 40: 51—54.

SMITH, G.S. & WATKINSON, J.H. 1984. Selenium toxicity in perennial ryegrass and white clover. New Phytol. 97: 557—564.

VUORINEN, J. & MÄKITIE, O. 1955. The method of soil testing in use in Finland. Agrogeol. Publ. 63: 1—44.

YLÄRANTA, T. 1983 a. The hydride method for measuring the selenium content of plants. Ann. Agric. Fenn. 22: 18—28.

— 1983 b. Selenium in Finnish agricultural soils. Ann. Agric. Fenn. 22: 122—136.

— 1983 c. Effect of liming and sulphate on the selenium content of Italian rye grass *(Lolium multiflorum)*. Ann. Agric. Fenn. 22: 152—163.

— 1985. Increasing the selenium content of cereal and grass crops in Finland. Diss. 72 p.

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SELOSTUS

Kalkituksen sekä sulfaatti- ja fosfaattilisäyksen vaikutus italianraiheinän seleenipitoisuuteen

TOIVO YLÄRANTA

Maatalouden tutkimuskeskus

Kasveille käyttökelpoisimmat seleenin epäorgaaniset muodot maassa ovat seleniitti ja selenaatti. Seleniitti-ionin käyttäytyminen eri reaktioissa muistuttaa fosfaatti-ionin ja selenaatti-ionin käyttäytyminen sulfaatti-ionin käyttäytymistä.

Fosfaatti ja sulfaatti ovat yleisiä ioneja sekä lannoitteissa että maassa. On esitetty epäilyjä, että fosfaatti ja sulfaatti voivat häiritä kasvin seleenin ottoa maasta.

Seleenin liukoisuutta maassa voidaan ainakin teoriassa lisätä kohottamalla maan pH:ta kalkituksella. Suomalaiset maat ovat luonnostaan happamia. Siten myös maiden kalkitseminen saattaa muuttaa kasvien seleenin ottoa.

Astiakokeessa tutkittiin runsaan kalkituksen sekä suurien sulfaatti- ja fosfaattilisäysten vaikutusta italianraiheinän seleenin ottoon savimaassa, hietamaassa ja turvemaassa. Seleeni lisättiin maahan sekä seleniitti- että selenaattimuodossa. Kokeessa korjattiin kaksi satoa.

Kalkitus, jolla kohotettiin maiden pH-lukua lähes kaksi pH-yksikköä, vähensi kasvin seleenipitoisuutta silloin, kun seleeni lisättiin koeastioihin selenaattimuodossa. Vastaavasti kalkitus lisäsi raiheinän seleenipitoisuutta silloin, kun seleeni lisättiin maahan seleniittinä. Muutokset eivät kuitenkaan olleet aina tilastollisesti merkitseviä.

Koeastiat sisälsivät noin kaksi litraa maata. Lisätyt fosfori- ja rikkimäärät astiaa kohti olivat peräti 800 mg. Fosfaattilisäyksen vaikutus kasvien seleenipitoisuuteen oli pieni. Fosfaatti pienensi savi- ja hietamaassa italianraiheinän toisen sadon seleenipitoisuutta.

Sulfaattilisäys ei vaikuttanut oikeastaan ollenkaan raiheinän seleenipitoisuuteen seleniittikoejäsenissä. Kun seleeni lisättiin koeastioihin selenaattimuodossa, pienensi sulfaattilisäys raiheinän seleenipitoisuutta kaikissa maissa kummassakin sadossa. Seleenipitoisuus oli tällöin vain 18—43 % siitä, mitä se oli ilman sulfaattilisäystä.

Sulfaatin vaikutus kasvien selenaattiseleenin ottoon ei liene käytännön viljelyssä yhtä dramaattinen kuin tässä kokeessa, koska lannoitteiden sulfaattirikkipitoisuus ja maan sulfaattipitoisuus eivät yleensä ole näin suuria suhteessa seleenipitoisuuteen. Lisäksi sulfaatti-ioni pidättyy myös huonosti maahan, jolloin se on alttiina huuhtoutumiselle. Sulfaatti voi siis käytännössä toisin kuin astiakokeessa joutua pois kasvien juurten ulottuvilta häiritsemästä kasvien seleenin ottoa. Kasvuolot eivät ylipäätänsä ole astiakokeessa suoraan verrattavissa pelto-oloihin, minkä vuoksi tulokset ovat käytäntöön vain suuntaa antavia.

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Seria AGROGEOLOGIA ET -CHIMICA N. 157 — Sarja MAA JA LANNOITUS n:o 157

SELENIUM IN SOIL EXTRACTS AND PLANTS DETERMINED BY FLUOROMETRY

DACHENG WANG and JOUKO SIPPOLA

WANG D. & SIPPOLA J. 1990. **Selenium in soil extracts and plants determined by fluorometry.** Ann. Agric. Fenn. 29: 151—156. (Agric. Res. Centre, Inst. Crop and Soil Sci. SF-31600 Jokioinen, Finland.)

Methods for the fluorometric determination of selenium in soils and plants are described. The detection limit of the fluorometric determination was 0.3 ng selenium in 5 ml cyclohexane. The recovery of Se added to the soil samples was 99.3 % on average and that added to plant samples 100.8 %. The precision as coefficient of variation of duplicate determinations of plant selenium was 6.1 % and that for HNO_3-$HClO_4$ digestible soil selenium 2.3 %. For soil total selenium determination, digesting samples with aqua regia released almost twice as much selenium as the HNO_3-$HClO_4$ digestion. Water soluble selenium in soils constituted 2 % of aqua regia digestible selenium on average and about 5 % of the acid ammonium acetate-EDTA extractable selenium. Plant selenium correlated with soil selenium as follows: Water extr. $r = 0.33^{***}$, AAAc-EDTA extr. $r = 0.33^{***}$, HNO_3-$HClO_4$ digest. $r = 0.27^{**}$, aqua regia digest. $r = 0.23^{**}$. The results of this investigation showed that the multielement extractant AAAc-EDTA is also useful for the determination of plant available selenium in soils. Water extraction may be a good choice when selenium is under particular investigation.

Index words: selenium, soil, plant, extraction, fluorometry.

INTRODUCTION

Selenium is an essential element for animal health. Many methods for determining selenium in soils and plants have been described. The most popular ones include the fluorometric method using 2,3-diaminonaphthalene (DAN) and atomic absorption spectrometry employing either hydride generation or electrothermal atomization. The fluorometric method is both fast and sensitive for a variety of samples with low Se contents. In this method, Se is determined after sample digestion and the formation of Se-complex with 2,3-diaminonaphthalene which is then extracted in cyclohexane. Wet digestion methods for determining selenium in plant materials and soils have been further improved by many researchers (CARY and ALLAWAY 1969, CHAN 1976, HOU SHAOFAN et al. 1979). However, by these methods selenium is first oxidized to Se(+6) and then reduced with some reagent to Se(+4). This makes the digestion more complicated, and increases the possibilities of selenium losses during the digestion process.

Many laboratories in Europe use aqua regia to digest soil samples for the determination of total elemental composition. Because of the existence of HCl in this digest, selenium is maintained in the Se(+4) form. CUMMINS et al. (1965) used Na_2MoO_4-H_2SO_4-$HClO_4$ to digest animal tissue samples for selenium deter-

mination. Na_2MoO_4 acts as an indicator here. When the temperature of the digest is raised to 180 °C, its colour turns to light yellow-light green. This temperature is high enough for complete digestion but low enough to avoid selenium losses.

Some studies have shown that soil total selenium concentration does not well reflect Se uptake by plants (GISSEL-NIELSEN and HAMDY 1978, SIPPOLA 1979). Also many weaker extractants, such as water, diluted acetic acid, hydrochloric acid, ammonium acetate, ammonium sulphate, calcium chloride, sodium hydroxide, EDTA etc. have been tested. However, there is still a need to develope a soil extractant which would reliably predict the availability of soil selenium to plants. Acid ammonium acetate-EDTA is used in Finland as the extractant for plant available trace elements in soils (LAKANEN and ERVIÖ 1971). If it were possible to use the same extract as that prepared for routine testing also in the estimation of plant available selenium, the determination would then be most economical.

The aim of this work was to compare by using fluorometry some of the methods used to determine soil total and extractable selenium content and evaluate them in predicting plant available selenium.

MATERIAL AND METHODS

The sample material of the study was comprised of soil and winter wheat collected at the heading stage from 13 European countries. Samples were obtained as part of the activities of the European Cooperative Research Network on Trace Elements in 1986—87. Methodological tests were also performed using certified reference material.

Extraction of soil selenium

Digestion with HNO_3-$HClO_4$: An air-dried soil sample of 0.4 g was placed in a 100 ml flask and 10 ml of acid mixture (Conc.HNO_3-conc.$HClO_4$ = 2:1) was added. The flask was gently warmed on a hot plate and shaken at intervals. After boiling for 10 minutes, the flask was removed. When cool, 2 ml H_2O_2(30 %) was added and reheated until the brown fumes disappeared and then removed from the hot plate. 2 ml HCl was added, reheated until brown fumes disappeared and this process was repeated. After cooling to room temperature, the sample was diluted with 20 ml of de-ionized water (HOU SHAOFAN et al. 1979).

Digestion with aqua regia: 5 g of dry soil was weighed into an Erlenmeyer flask, 50 ml of aqua regia ($HCl:HNO_3$ = 3:1) was added and allowed to stand overnight. Then the suspension was boiled slowly for 2 h under a reflux condenser. Finally the extract was filtered into a 200 ml volumetric flask and filled to volume with water (ANON. 1986).

Extraction with water: 20 g soil was placed in a 200 ml plastic bottle and 100 ml de-ionized water was added. The bottle was shaken for 30 minutes (200 c.p.m.) and then centrifuged for 10 minutes (5000 r.p.m.). 50 ml of extract was transferred into a 100 ml flask, pH was adjusted to 8 with 0.1 N NaOH. Then the solution was evaporated on a hot plate until 5—10 ml was left and digested by the same procedure as that for the soil samples using HNO_3-$HClO_4$.

Extraction with AAAc-EDTA: 25 g soil and 250 ml extracting solution was shaken for one hour (end over end, 27 r.p.m.). The suspension was then filtered using Whatman No. 42 filter paper. A suitable aliquot of filtrate was taken

and digested as in the HNO_3-$HClO_4$ method. The acid ammonium acetate-EDTA (AAAc-EDTA) extracting solution was prepared by diluting 571 ml of glacial CH_3COOH, 373 ml of NH_4OH and 74.4 g Na_2EDTA to 10 liters with water. pH was adjusted to 4.65 with acetic acid or ammonium hydroxide (ANON. 1986).

Digestion of plant samples for selenium determination

A 1 g plant sample was weighed into a 100 ml flask, 3 ml Na_2MoO_4 solution (15 g/100 ml water) and 10 ml acid mixture (H_2SO_4:$HClO_4$ = 3:4) were added. The flask was gently warmed on a hot plate. After a vigorous reaction and clearing of the solution, the temperature was raised. As soon as the colour of the solution had changed to light yellow-light green, the flask was removed from the hot plate and cooled to room temperature and 20 ml de-ionized water was added (CUMMINS et al. 1965).

Fluorometric determination of selenium in digests

In the case of soil digests 10 ml and for plants 4 ml 0.2 M EDTA-1 % hydroxylammonium chloride solution was added to the samples. With 50 % NH_4OH the pH was adjusted to 1.5—2.0 and 2 ml 0.1 % DAN solution was added in a dark room. The DAN solution was prepared by dissolving 0.1 g 2,3-diaminonaphthalene in 100 ml 0.1 M HCl, and impurities were extracted in 10 ml cyclohexane. The DAN solution was kept in a dark bottle in a refrigerator. The flask containing the sample was heated in boiling water for 5 minutes and cooled to room temperature under flowing tap water. Then 5 ml cyclohexane was added, shaken for 5 minutes and cyclohexane was separated for the determination of selenium using on a Perkin — Elmer 3000 Fluorescence Spectrometer. The excitation wavelength was 377 nm and fluorescence was measured at 522 nm. A Se standard containing 0.1 µg Se and a blank were run throughout all steps of sample preparation.

RESULTS AND DISCUSSION

Determination of soil total selenium

Digestion with HNO_3-$HClO_4$ is used in China as a measure of soil total selenium (HOU SHAO-FAN et al. 1979). There were no apparent interferences in determining selenium using this method as the recovery of added selenium ranged from 95 to 104 % (Table 1). The mean of the coefficient of variation when performig double determinations of 128 soil samples was 2.3 %.

Aqua regia digestion for soil total selenium determination is a rather simple digestion method in which soil samples are boiled under a reflux condenser. Due to the existence of HCl, all forms of selenium are changed into the Se(+4) oxidation state. Aqua regia is strong enough to destroy any organic matter, so an H_2O_2 addition is not needed as in the HNO_3-$HClO_4$ digestion. The soil total selenium content by the aqua regia digestion was 75 % higher on average than that by the HNO_3-$HClO_4$ digestion (Table 2). This shows that the aqua regia digestion was more complete, most likely because some mineral crystal lattices

Table 1. Recovery of Se added to soil sample (mean of 3 replicates).

Se added (µg)	—	0.020	0.040	0.080	0.160
Se found (µg)	0.106	0.123	0.147	0.193	0.251
Recovery (%)	—	97.6	101	104	94.4

Table 2. Soil and plant Se concentration, µg/kg (n = 128).

	Mean	SD	Medium	Minimum	Maximum
Plant Se	42	28	32	3.8	127
Soil HNO_3-$HClO_4$ dig. Se	180	102	156	39	523
Soil aqua regia dig. Se	316	160	278	62	819
Soil AAAc-EDTA extr. Se	17	8.8	16	3.2	51.6
Soil water extr. Se	6.0	3.2	5.4	1.0	24

were destroyed thus releasing selenium. Still the digestion is not complete, however for most elements an extraction rate of over 90 % is common (BAGHDADY and SIPPOLA 1983). The correlation between soil total selenium determined with HNO_3-$HClO_4$ digestion and aqua regia digestion was $r = 0.87$ ($n = 128$, $P < 0.001$), showing that both methods were reliable (Table 3).

Estimation of plant available selenium in soil

Plant available selenium in soils was extracted by acid ammonium acetate-EDTA and water. The AAAc-EDTA extractable selenium was about 5 % and the water extractable selenium about 2 % of the soil total selenium determined by the aqua digestion, on an average. Similarly, PAASIKALLIO (1981) observed that, with the exception of a Sphagnum peat, in 5 Finnish soils the AAAc-EDTA extractable selenium ranged from 5 to 20 % of total selenium. HAMDY and GISSEL—NIELSEN (1976) found that water-soluble selenium constituted on average 2.4 % of the total soil selenium in Danish soils.

Of these two extraction methods, water extraction was simple and economical. Since the extractant contained little salts, the digestion was also easier. For these determinations, the detection limit was 0.3 ng Se/5 ml cyclohexane. Using 1 g samples this is sufficient for most low selenium samples.

Table 3. Recovery of Se added to reference material (mean of 3 determinations).

Se added (µg)	—	0.020	0.040	0.080	0.160
Se found (µg)	0.057	0.077	0.103	0.134	0.216
Recovery (%)	—	100	106	97.8	99.5

Determination of selenium in plants

The mean value of 10 determinations of the wheat flour reference material (no. 27, Agricultural Research Centre/Central Laboratory, Finland) was 57.2 ± 5.2 µg/kg. Comparison of this result with the certified value of 58 ± 4 µg/kg indicates that the fluorometric method employed is accurate. This is further shown by the good recovery of Se the reference material (Table 3). The mean coefficient of variation of duplicate determinations of selenium in 128 plant samples was 6.1 % suggesting that the H_2SO_4-$HClO_4$ digestion is an acceptable method for oxidizing plant material for selenium determination. Both accuracy and precision are satisfactory. When using Na_2MoO_4 as a temperature indicator, the end of digestion is easy to control and there is no need to add HCl during the digestion as in the HNO_3-$HClO_4$ method. This makes the digestion process both simple and fast.

Dependence of plant selenium content on soil selenium

Plant selenium correlated significantly with estimates of plant available ($P < 0.001$) and total selenium ($P < 0.01$) in soils, although not very closely (Table 4). NYE and PETERSON (1975)

Table 4. Correlation between soil Se fractions and plant Se.

	Soil Se (HNO_3-$HClO_4$)	Soil Se (aqua regia)	AAAc-EDTA extr. Se	Water extr. Se
Plant Se	0.27**	0.23**	0.33***	0.33***
Soil Se (HNO_3-$HClO_4$)		0.87***	0.66***	0.63***
Soil Se (aqua regia)			0.65***	0.63***
AAAc-EDTA extr. Se				0.70***

, * significant at 0.01, 0.001 levels, respectively (n = 128).

found a good correlation between water-soluble selenite selenium content in soils and plant selenium uptake. OLSON and MOXON (1939), CARY and ALLAWAY (1969) have shown that the plant uptake of selenium is closely correlated with the water-soluble selenium fraction in the case of soils rich in selenium. However, the work of GISSEL-NIELSEN and HAMDY (1978) showed a lack of correlation between the selenium content of ryegrass and total or water-soluble selenium in Se-deficient soils. SIPPOLA (1979) found that the selenium content of timothy correlated with both total and AAAc-EDTA extractable soil selenium, but according to multiple regression analysis total selenium appeared to be more important.

The low correlation of plant selenium with various fractions of soil selenium indicates that plant selenium uptake is affected by many soil factors eg. texture, pH, Eh, free iron and organic matter. Most of these factors affect plant selenium uptake by changing the strength by which selenium compounds are retained by soil colloids. The selenium level in soil solution is a result of all these effects. Although the total content is likely to be in some relation to the extractable content, it is expected that the extractable content correlates more closely, as indicated by the results obtained. There was no difference found in the closeness of correlation between water and AAAc-EDTA extractable contents thus allowing the possibility to use both of these methods. AAAc-EDTA has its advantages as a multielement extractant but water may be more useful when selenium is under particular investigation.

REFERENCES

ANON. 1986. Methods of soil and plant analysis. Dept. of Soil Sci., Agr. Res. Centre, Finland. 30 p.

BAGHDADY, N.H. & SIPPOLA, J. 1983. Total heavy metal recovery by aqua regia in soils of different origin. Ann. Agric. Fenn. 22: 175—185.

CARY, E.E. & ALLAWAY, W.H. 1969. The stability of different forms of selenium applied to low-selenium soils. Soil Sci. Soc. Amer. Proc. 33: 571—574.

CHAN, C.C.Y. 1976. Improvement in the fluorometric determination of selenium in plant materials with naphthalene-2,3-diamine. Anal. Chem. Acta 82: 213—215.

CUMMINS, L.M., MARTIN, J.L. & MAGG, D.D. 1965. An improved method for determination of selenium in biological material. Anal. Chem. 37: 430—431.

GISSEL-NIELSEN, G. & HAMDY, A.A. 1978. Plant uptake of selenium and selenium values in different soils. Z. Pfl. ernähr. Bodenkunde 141: 67—75.

HAMDY, A.A. & GISSEL-NIELSEN, G. 1976. Relationships between soil factors and selenium content of Danish soils and plants. Risø report No. 349.

HOU SHAOFAN, THU, T., WANG, W. & LI, D. 1979. Fluorometric determination of selenium in soils. Anal. Chem. (in Chinese) 9,1: 121.

LAKANEN, E. & ERVIÖ, R., 1971. A comparison of eight extractants for the determination of plant available micronutrients in soils. Acta Agr. Fenn. 128: 223—232.

NYE, S.M. & PETERSON, P.J. 1975. The content and distribution of selenium in soils and plants from seleniferous

areas in Eire and England. Trace Substances in Environmental Health 9: 113—121.

OLSON, O.E. & MOXON, A.L. 1939. The availability, to crop plants, of different forms of selenium in the soil. Soil Sci. 47: 305—311.

PAASIKALLIO, A. 1981. The effect of soil pH and Fe on the availability of 75 Se in Sphagnum peat. Ann. Agric. Fenn. 20: 15—24.

SIPPOLA, J. 1979. Selenium content of soils and timothy (Phleum pratense L.) in Finland. Ann. Agric. Fenn. 18: 182—187.

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SELOSTUS

Seleeni maauutteissa ja kasveissa fluorometrisesti määritettynä

DACHENG WANG ja JOUKO SIPPOLA

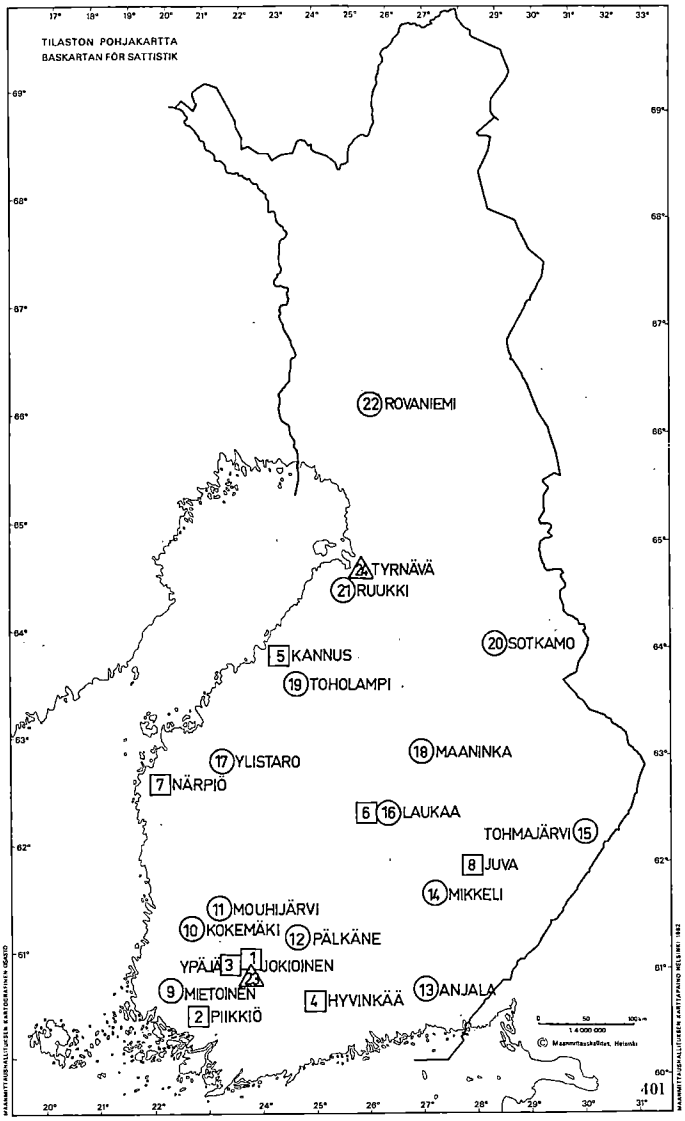
Maatalouden tutkimuskeskus

Lannoitteiden seleenilisäyksen myötä ei tarve määrittää seleeniä ole vähentynyt sillä muun muassa mahdollisten ympäristövaikutusten takia seleenipitoisuuden kehittymistä maaperässä tulee seurata. Tämän takia selvitettiin Suomessa vähän käytetyn fluorometrisen menetelmän käyttökelpoisuutta määritettäessä maan seleenipitoisuutta erilaisista maauutteista sekä kasveista. Samalla kokeiltiin eräitä Kiinassa maan seleenipitoisuuden määrittämiseen käytettyjä menetelmiä.

Seleenin fluorometrinen määritys osoittautui herkäksi ja toistettavaksi. Menetelmään mahdollisesti liittyviä häiriöitä selvitettiin lisäämällä analysoituun näytteeseen seleeniä vakiomääriä. Lisätyt määrät voitiin analysoida luotettavasti.

Käytetyistä uuttomenetelmistä kuningasvesi vapautti maasta eniten seleeniä. Hivenravinteiden uuttoon käytetty hapan ammoniumasetaatti-EDTA uutti 5 % kuningasveden uuttamista määristä. Ko. menetelmä kuvasti kasvien seleenipitoisuutta yhtä hyvin kuin vesiuutto, mitä menetelmää käytetään yleisesti liukoisen seleenin määrittämiseen. Tämän takia voidaan uuttamista hapan ammoniumasetaatti-EDTA menetelmällä suositella hivenravinteiden lisäksi myös maan kasveille käyttökelpoisen seleenin määrittämiseen.

CONTENTS



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