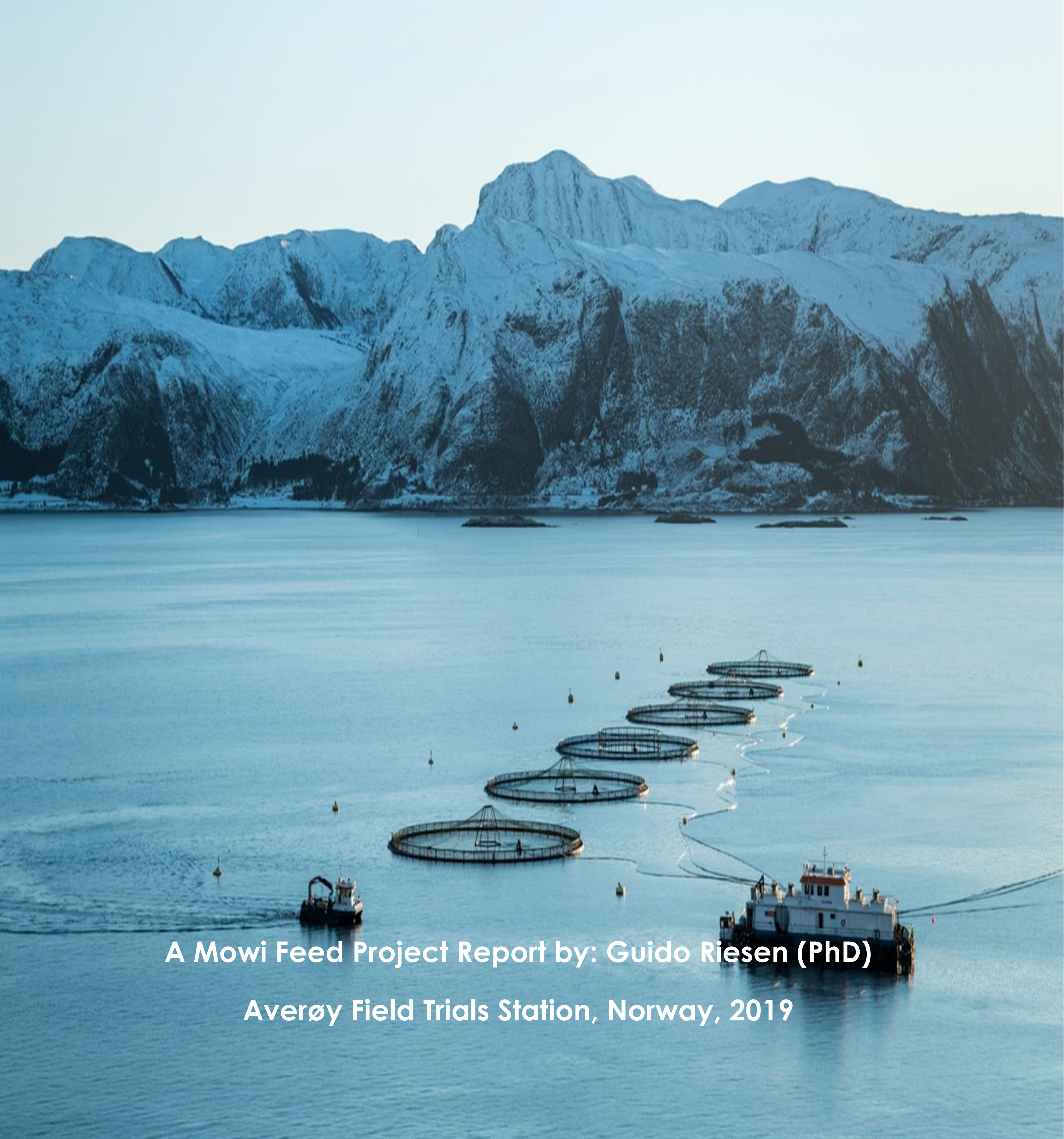
Impact of dietary digestible protein levels and pellet size on growth performance of large Atlantic Salmon, *Salmo salar*



A Mowi Feed Project Report by: Guido Riesen (PhD)

Averøy Field Trials Station, Norway, 2019

INTRODUCTION

Background

Achieving high growth performance with the lowest feed cost is a key component for successful salmon farming. It is known that dietary digestible protein (DP) levels are a major driver for growth performance in Atlantic salmon. It is recognised that dietary DP levels below the fish's requirement will reduce growth. On the other hand, DP levels above the requirement for growth can significantly increase feed cost and therefore reduce profitability in conjunction with poorer outcomes for the sustainability of salmon farming operations.

Optimal dietary DP content changes in accordance with fish size and environmental conditions such as water temperature, day light and disease pressure. Another influencing factor is the constant genetic selection for growth performance. Genetically improved growth performance is usually a result of the increased protein deposition capacity of an animal and might require diets with increased protein levels. The impact of feeding dietary digestible protein requirements needs regular reappraisal because optimal levels may change due to genetic selection and varying environmental conditions.

Recent reports from Mowi suggest that bigger pellet sizes seem to increase the feed intake and thus, the growth performance of salmon. This not because the pellets are intrinsically superior in terms of composition but, the effectiveness with which they are distributed and consumed increases. As the fish size of this project was appropriate to investigate this question more deeply, the hypothesis that pellet size influences growth was added as a secondary project objective.

Objectives

- To investigate whether current dietary digestible protein levels (DP) in MOWI feeds are adequate to support optimal performance of Atlantic salmon
- To investigate whether feeding larger pellet sizes improves the performance of Atlantic salmon.

MATERIAL & METHODS

Experimental feeds

Experimental feeds were formulated by Mowi Feed and produced at the Nofima Feed Technology Centre in Bergen (Norway) or at the Mowi factory at Valsneset, Norway. Seven experimental diets were produced. All diets were formulated to secure equal outcomes for digestible energy (DE) and to cover essential nutrient requirements with regards to the profile of digestible amino acids, essential fatty acids and key micronutrients as described by National Research Council NRC (2011). Six of the experimental feeds were formulated to contain different digestible protein levels (Diets A to F). Diet EE was produced with the same raw material mix as Diet E but with increased pellet size parameters. The composition and analysed proximate composition of each diet is shown in Table 1. The analysed nutritional values for each feed met expectations. However, pellet parameters were not only different between Diet E and Diet EE as expected but also between some of the other diets.

Fish and husbandry

The feeding trial was conducted at Mowi's Averøy Field Trials Station, Norway between 23rd October and 12th December 2019. Atlantic salmon were randomly distributed amongst 28 marine net pens (5x5x5m; 125 m^3) in the E- and F sections of the trial farm with an abundance of 80 fish per pen. The average mean weight was 3.386 kg, with ± 0.02 kg standard deviation between treatment averages. The fish were given a period of 4 days acclimation to environmental conditions whilst fed a commercial feed (Mowi RI 2500) before the study started. Upon commencement of the 49-day feeding period, the fish were fed one of 7 feeds (Table 1) with 4 pens allocated to each feed. The salmon were fed in excess using a combination of automatic feeders and a waste feed collection system. A daily over-feed of 10-15% was targeted and the excess feed amount was confirmed gravimetrically on dry weight corrected, recovered waste feed before feed intake and feed conversion factor were calculated. Water temperature during the trial period was 8.3 °C on average and spanned a range between 10.2 °C (October) and 6.8 °C (December), a natural photoperiod was observed. Fish health and welfare were monitored daily following Mowi's standard procedures.

The study was planned to be conducted until end of January 2020. However, by the beginning of December, increasing predation pressure on the fish by otters and mink was recorded. Due to the size of the fish, this did not cause direct mortality but significantly influenced feeding behaviour in the study pens. The decision was therefore made to terminate the trial so as to preserve results collected for feed intake and potential growth indices.

Fish sampling

At the end of the 49-day trial period, all fish were individually weighed and counted with further post-mortem assessments being carried out on 12 salmon / pen (48 fish / treatment). Fish were euthanised and each fish was subjected to the following: measurement of individual weight and fork length; gutting and weighment of the gutted carcass and liver; visual assessment of fat score of internal organs and collection of faeces. Based on these activities, indices such as condition factor (round and gutted weight basis) and viscera- and hepatosomatic index were calculated. Faeces were collected to determine the apparent digestibility of key nutrients in the feed including protein, amino acids and starch using an inert marker technique.

Table 1: Feed formulation, analysed proximate values and pellet size distribution

Formulations, % / Diet	A	B	C	D	E	F	EE
Fishmeals	10.00	10.00	10.39	10.00	10.00	10.00	10.00
Fish oils	10.63	10.60	10.39	15.58	15.04	13.91	15.04
Soy protein concentrate	11.37	13.30	14.95	19.57	7.82	6.26	7.82
Corn gluten	5.00	5.00	5.00	0.00	5.00	5.00	5.00
Wheat gluten	14.06	14.09	15.00	16.01	15.54	20.00	15.54
Pea Protein Concentrate	0.00	0.00	9.02	0.00	0.00	0.00	0.00
Guar meal	5.00	5.00	2.50	0.00	5.00	5.00	5.00
Vegetable oils	24.79	24.75	24.26	18.40	18.32	17.25	18.32
Wheat, whole	13.04	11.20	3.11	14.93	5.42	4.84	5.42
Field beans, dehulled	0.27	0.27	0.27	0.00	12.50	12.50	12.50
Vitamins and carotenoids	0.70	0.70	0.71	0.70	0.71	0.70	0.71
Minerals	1.80	1.79	1.69	1.81	1.82	1.83	1.82
Amino acids	1.60	1.47	1.18	0.98	1.19	1.11	1.19
Other	1.74	1.83	1.53	2.01	1.64	1.59	1.64
Post-production analysis							
Moisture, %	7.1	6.7	6.6	7.0	7.2	6.6	6.9
Crude protein, %	33.1	35.9	36.3	37.0	37.9	40.9	38.3
Crude fat, %	38.4	38.7	36.7	34.9	34.6	32.9	33.9
Total starch, %	10.8	9.0	9.5	11.3	11.1	11.1	11.3
Free astaxanthin, mg/kg	36	57	52	50	52	53	52
Mean pellet length (mm)	8.8	9.0	8.6	11.0	11.2	10.7	13.4
SEM	0.9	0.7	0.7	1.0	0.8	1.2	0.5
Mean pellet diameter (mm)	8.8	8.4	8.1	8.8	9.0	8.9	11.1
SEM	0.5	0.4	0.4	0.4	0.4	0.5	0.4
Mean no. pellets / kg	1712	1789	1979	1502	1396	1487	818
SEM	12	8	6	9	9	16	2

SEM: Standard error of the mean; Analysis of feed was carried out by Nofima BioLab, Bergen, Norway
To determine avr. pellet length, avr. pellet diameter 100 pellet each diet were measured. 3 x 100 pellets each diet were weighted to calculate the number of pellets / kg

From 6 of these fish, instrumental measurement of flesh colour (l*, a* and b*) using a Minolta CR-200 Chroma Meter (CR-410, C illuminate) was conducted. Colour outcomes were further derived in terms of chroma and hue.

Statistical analysis

All statistical analyses were performed using the statistical package of GraphPad Prism 9.1.2. If not otherwise stated in the report data were analysed to normality data and statistically evaluated with a one-way ANOVA and Tukey post-hoc test to find differences between dietary treatments. Differences were regarded as significant when $p \leq 0.05$. Unless otherwise stated, data are presented as mean value, standard error of the mean (SEM) and with an n-number of 4 replicates per dietary treatment.

RESULTS

Digestible protein (DP) and digestible energy (DE) content of experimental feeds

Digestible protein (DP), digestible energy (DE) content and the DP/DE ratio of the experimental feeds are shown in Table 2. Values are calculated using the measured apparent digestibility coefficients for protein, fat and starch. Digestible energy (DE) was calculated by adding calculated digestible energy contribution for protein, fat and starch, using standard energy values for each of the nutrients (23.6, 39.5, 17.1 MJ/kg respectively).

Table 2: Mean apparent digestibility coefficient (ADC) for protein, fat, starch and calculated digestible protein (DP), calculated digestible energy (DE) for each experimental diet on as is basis.

Parameter / Diet	A	B	C	D	E	F	EE
Mean ADC CP	92.8	92.8	92.4	93.8	93.4	94.4	93.7
SEM	0.3	0.2	0.2	0.3	0.4	0.2	0.2
Mean ADC fat	94.9	94.2	94.3	97.0	96.3	97.7	96.8
SEM	0.3	0.6	0.5	0.4	0.6	0.5	0.2
Mean ADC starch	75.2	78.7	73.6	80.9	70.1	74.4	71.5
SEM	1.5	0.3	0.3	0.8	1.0	0.7	0.8
Digestible protein (DP), (g/kg)	307	333	335	347	354	386	359
Digestible energy (DE), MJ/kg	23.0	23.5	22.8	23.1	22.8	23.2	22.8
DP/DE ratio[1)]	13.3	14.2	14.7	15.0	15.5	16.6	15.7

SEM: Standard error of the mean; ADC: apparent digestible coefficient; Energy values used for protein, fat, starch were 23.6, 39.5, 17.1 MJ/kg, respectively

Growth performance overview by treatment

Overall growth performance in this study was satisfactory with an average relative growth index (RGI) of 97%, with ± 17% standard variation (RGI calculated using Mowi's season and geography-adjusted anticipated growth rate model) until the trial was stopped after 49 days due to the adverse effect of predator activity on feeding behavior. Fish performance parameters are presented in Table 3. Average final weight, weight gain, specific growth rate (SGR) and biological feed conversion rate ($FCR_{biological}$) across all treatments were 4.373 kg, 0.988 kg, 0.52 (%/day), 0.92, respectively. The best performing feed was Diet EE which was characterised by larger pellet size parameters. The performance of salmon fed Diet EE was also significantly different from those fed Diet B and C but not from the other feeds. Other performance parameters were not significantly different from each other.

Table 3: Mean of initial weight, final weight, weight gain, specific growth performance, feed intake and feed conversion ratio by treatment and within the 49 day growth period. (Treatments differences were tested by a 1-way ANOVA model with Diet as variable)

Parameter / Diet	A	B	C	D	E	F	EE	ANOVA p-value
Initial weight, kg	3.359	3.408	3.400	3.382	3.417	3.366	3.371	0.975
SEM	0.039	0.031	0.032	0.061	0.066	0.052	0.06	
Final weight, kg	4.286[ab]	4.253[a]	4.256[a]	4.411[ab]	4.411[ab]	4.440[ab]	4.548[b]	0.011
SEM	0.061	0.054	0.057	0.083	0.023	0.044	0.06	
Weight gain, kg	0.927	0.857	0.856	1.029	0.994	1.074	1.177	0.166
SEM	0.085	0.040	0.044	0.143	0.082	0.083	0.11	
SGR, %/day	0.50	0.46	0.46	0.54	0.53	0.57	0.61	0.206
SEM	0.04	0.02	0.02	0.07	0.04	0.04	0.05	
FI , g/kg/day	19.0	19.4	19.1	17.8	18.8	18.5	18.8	0.254
SEM	0.4	0.2	0.4	0.1	0.3	0.6	0.7	
FCRb	0.93	0.95	0.93	0.87	0.92	0.91	0.92	0.252
SEM	0.02	0.01	0.02	0.01	0.01	0.03	0.03	

SEM: standard error of mean; SGR: specific daily growth rate; FI: feed intake as g/kg biomass gain/day; FCRb: biological feed conversion rate; Means sharing same superscipts are not significantly different from each other (Tukey's test, $P<0.05$)

Influence of pen section on growth performance

During visualisation of the data a variable performance trend was observed between fish allocated to the E-section (14 pens) and F-section (14 pens) of the trial farm. It appeared that salmon allocated to E-section had better growth performance compared to salmon allocated to F-section (Figure 1).

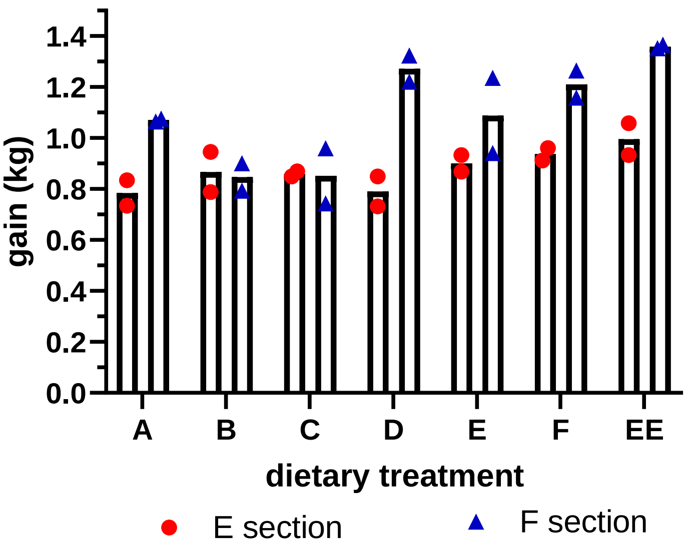


Figure 1: Weight gain differences by treatment and pen section. (individual pen performance visualised by symbol (n=2); each bar visualises average weight gain/ section)

A statistical comparison between the two sections showed significant differences between feed intake and weight gain but not feed conversion rate (FCRb) (Figure 2). This finding demonstrates that fish in both E and F sections did not differ in terms of feed efficiency but the feed intake of fish in section E was reduced. Higher predator pressure / interaction is proposed as a driver for the different feed intake for the two site sections.

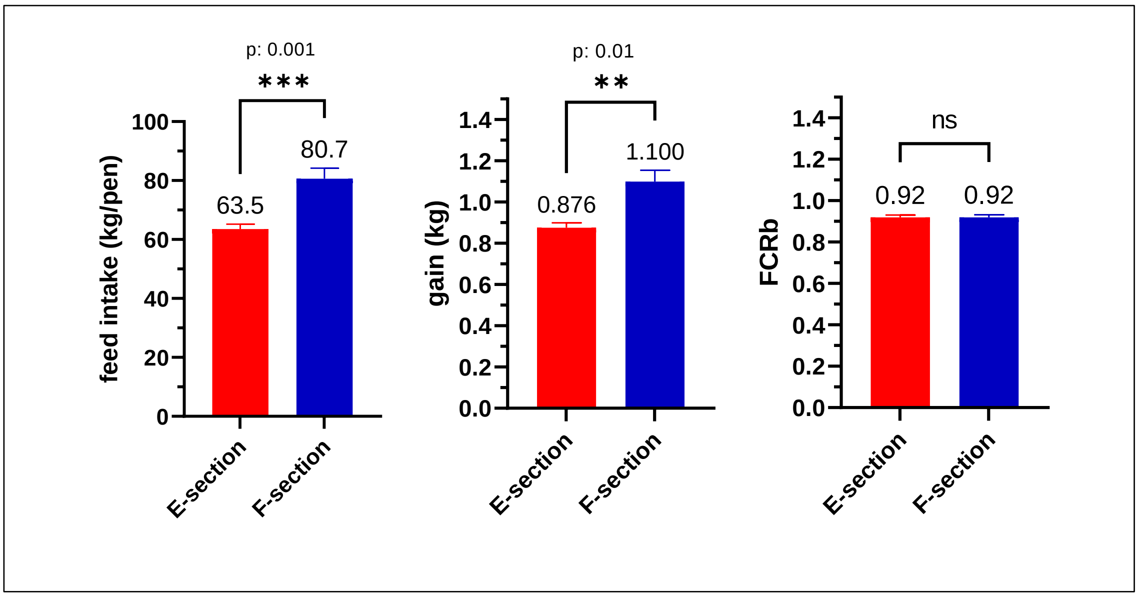


Figure 2: Mean feed intake, mean weight gain and mean biological feed conversion (FCRb) differences by pen section. (paired T-test, n =14 / section)

Influence of digestible protein (DP), digestible energy (DE), DP/DE ratio and pellet size on weight gain and feed conversion ratio (FCRb)

Different regression analysis (dose response models) were investigated with weight gain and FCR as the key performance parameters. The outcome of the digestible nutrient calculations showed that the experimental feeds were slightly different in digestible energy (DE) (Table 2) and that the best growth performance was achieved by salmon fed Diet EE (Table 3). Diet EE was designed with the biggest pellet size of all trial feeds. As there was marked variation in pellet sizes of the other experimental diets; regression analysis was therefore, not only modelled with DP and DP/DE ratio but also with DE and pellet number / kg as fixed effects. As 'pen section' was found to have a significant effect on performance, all regression analyses were performed for both E- and F-sections individually.

Figure 3 gives an overview of both linear and non-linear models applied to weight gain. It was apparent that higher dietary DP contributed to increased growth performance in both sections E and F (Figure 3a). A broken line model was the best fit for the relationship between dietary digestible protein (DP) and weight gain. The calculated break point of 36% DP for optimal weight gain should be received with caution due to the relatively low regression coefficient found. Digestible energy (Figure 3b) and the DP/DE ratio (Figure 3c) showed no significant linear or non-linear relationship to weight gain. As shown in Figure 3d, the number of pellets / kg affected weight gain in this study. For both pen sections significant linear regression models were found. Growth performance increased with increasing pellet size; which was expressed as number of pellets / kg.

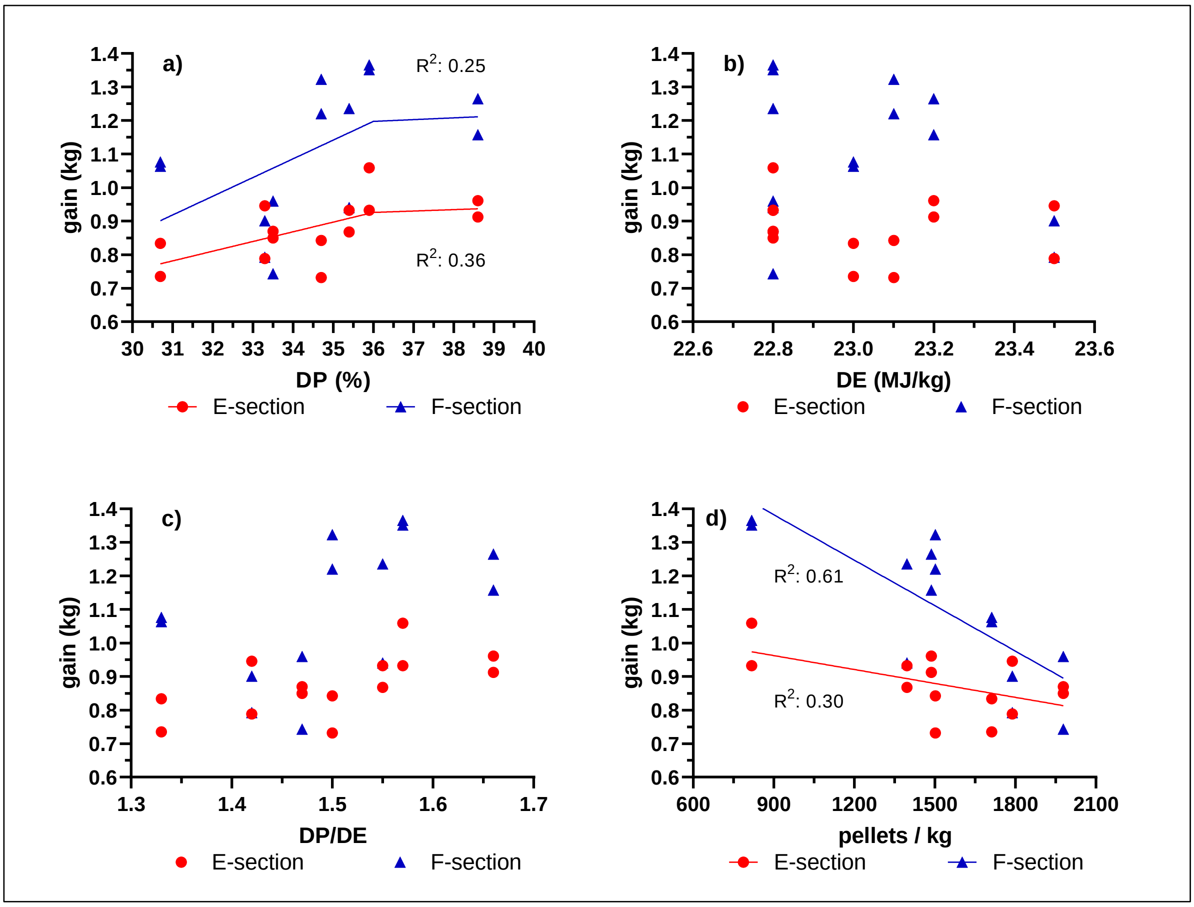


Figure 3: Relationships between digestible protein (DP), digestible energy (DE), DP/DE ratio, number of pellets / kg and weight gain (kg)

Figure 4 provides an overview of the influence of different dietary parameters on the biological feed conversion ratio (FCRb). Neither for dietary digestible protein (DP), dietary energy (DE), DP/DE ratio nor pellets / kg significant linear or non-linear relationship could be calculated (Figure 4 a-d).

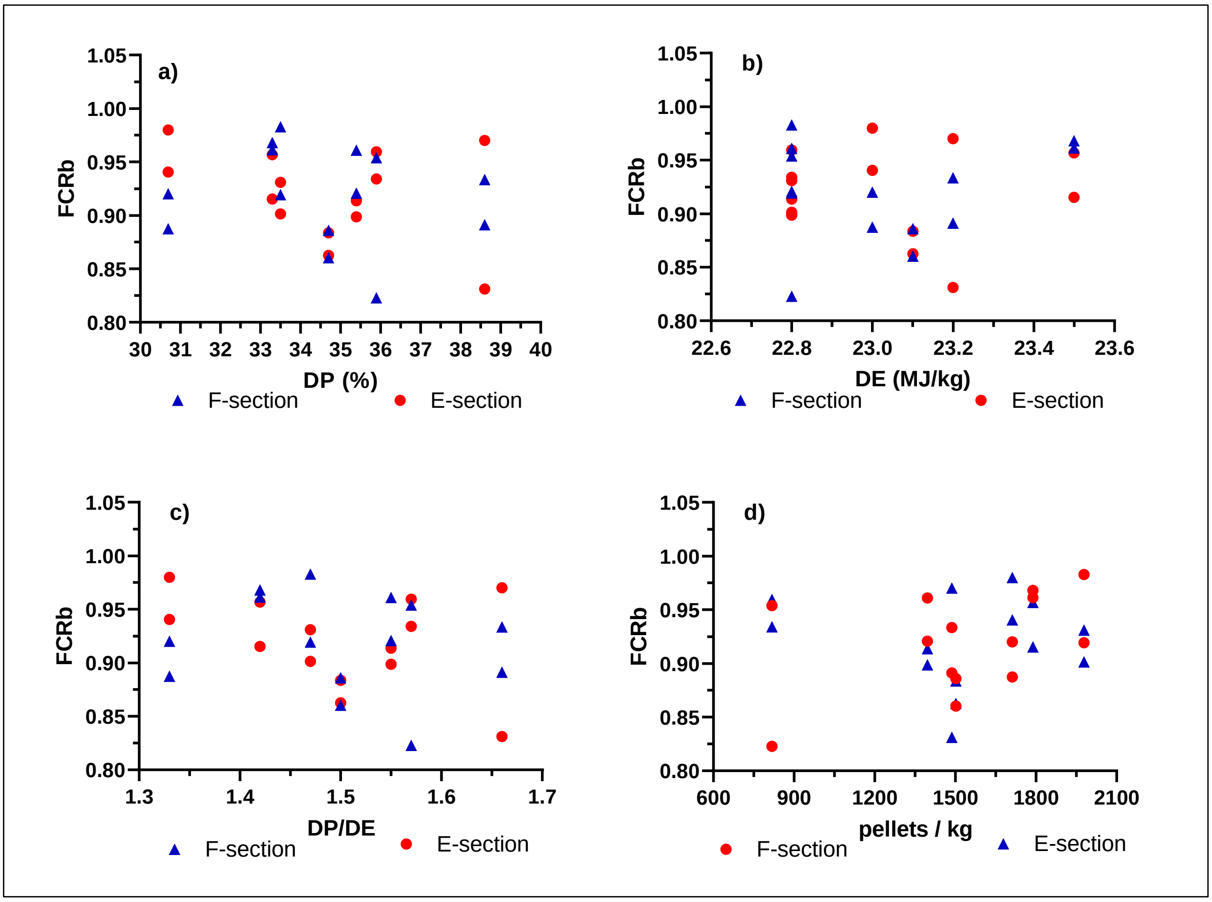


Figure 4: Relationships between digestible protein (DP), digestible energy (DE), DP/DE ratio, number of pellets / kg and biological feed conversion ratio (FCRb)

Possible interaction between response parameters

As shown in Figure 3a and 3b digestible protein (DP) and "number of pellets / kg" influenced the weight gain in this study. A multiple linear regression model (Gain ~ DP * number of pellet/kg) and simple linear regression models were applied to investigate whether DP and "number of pellets / kg" were correlated. Neither the multiple nor the simple linear model (Figure 5) detected significant interactions between the response parameters dietary DP and number of pellets / kg.

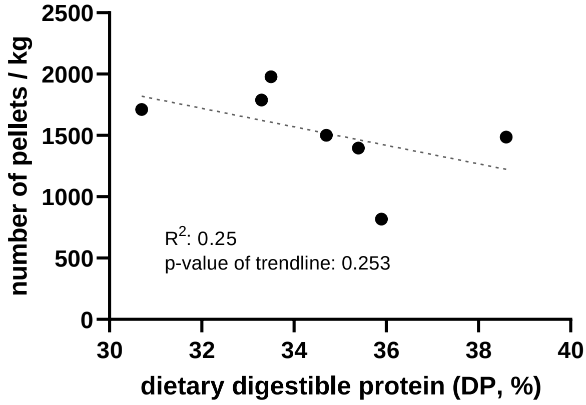


Figure 5: Relationships between digestible protein (DP) and number of pellets / kg. (R^2: 0.25; p: 0.253. Dotted trendline not significant

Yields and morphometric overview by treatment

Analysis of yields and morphometric results by treatment is presented in Table 4. As the fish for these analysis were selected within a certain final weight segment an influence of the two pen sections on the results was not expected. Assumption was tested with a 2-way ANOVA model. As assumed no influence of the pen section or interaction between Diet and Pen section were was found. The further evaluation was therefore not divided into separate pen sections as it was done for the performance parameters.

Mean condition factor for round fish, average carcass yield and average fat score for the analysed fish were, 1.40, 2.92 and 86.7%, respectively. Highest condition factors were found for Diet EE, the diet with the biggest pellet size, for both, round and gutted weight (1.45 and 1.25, respectively). Fish from treatment group EE had significant higher average condition factors than fish fed Diet C. Mean hepatosomatic index (HSI) values over all treatments was 1.13 and the highest for selected fish fed Diet EE. The calculated HSI values showed some differences between the treatment groups, with the general picture that Diets A, B and C, the treatments groups receiving the lowest dietary digestible protein diets, were significantly lower than Diets F, E, and EE.

Table 4: Means of condition factors, gutted weight, fat score, carcass yield and hepatosomatic index of analysed fish by treatment after a 49 day growth period. (Treatments differences were tested by a 2-way ANOVA model with Diet and Pen Section as variables)

Parameter / Diet	A	B	C	D	E	F	EE	2-way ANOVA p-values		
								Diet[1)]	Sec[2)]	Diet x Sec[3)]
Whole body weight, kg	4.082	4.155	4.224	4.342	4.277	4.422	4.446	0.117	0.538	0.244
SEM	0.091	0.126	0.064	0.084	0.134	0.055	0.074			
Condition factor, round	1.37^{ab}	1.40^{ab}	1.35^{a}	1.41^{ab}	1.40^{ab}	1.43^{ab}	1.45^{b}	0.042	0.366	0.335
SEM	0.01	0.03	0.02	0.02	0.01	0.02	0.02			
Weight after gutting, g	3.539	3.595	3.686	3.773	3.703	3.838	3.832	0.129	0.310	0.166
SEM	0.062	0.105	0.070	0.070	0.128	0.065	0.069			
Fat score	2.88	2.85	2.94	3.10	2.90	2.83	2.92	0.330	0.188	0.333
SEM	0.10	0.14	0.10	0.09	0.11	0.09	0.03			
Carcass yield, %	86.8	86.6	86.9	87.0	86.3	87.0	86.5	0.465	0.844	0.884
SEM	0.1	0.2	0.4	0.2	0.2	0.2	0.1			
Condition factor, gutted	1.19^{a}	1.20^{ab}	1.18^{a}	1.23^{ab}	1.20^{ab}	1.23^{ab}	1.25^{b}	0.010	0.449	0.108
SEM	0.01	0.02	0.02	0.02	0.01	0.01	0.01			
Hepato somatic index (HSI), %	1.07^{a}	1.09^{abc}	1.07^{ab}	1.13^{bc}	1.17^{c}	1.17^{c}	1.19^{c}	0.000	0.091	0.191
SEM	0.03	0.01	0.02	0.01	0.02	0.02	0.00			

SEM: standard error of mean; Means sharing same superscipts are not significantly different from each other (Tukey's test, P<0.05)

Influence of digestible protein (DP), digestible energy (DE), DP/DE ratio on yield and morphometric parameters

Condition factor for whole fish (CF_{round}), fat score, carcass yield and hepatosomatic index (HSI) were investigated for linear or non-linear relationships to dietary digestible protein (DP), digestible energy (DE) and DP/DE ration. No relationship was found for fat score and carcass yield to any of these response parameters. HSI and CF_{round} was influenced by DP, DP/DE ratio and number of pellets / kg but not by DE. Segmental line models were the best fitting models to describe the development of the condition factor and HSI (Figure 6). HSI but also CF_{round} increased with higher dietary DP levels until reaching a plateau (Figures 6a; 6b). With the response variable of weight gain, a plateau was reached at 36% DP. The same development for HSI and CF_{round} was analysed when modelling the DP/DE ratio as the linear response parameter with a breakpoint of 1.57(Figures 6c, 6d).

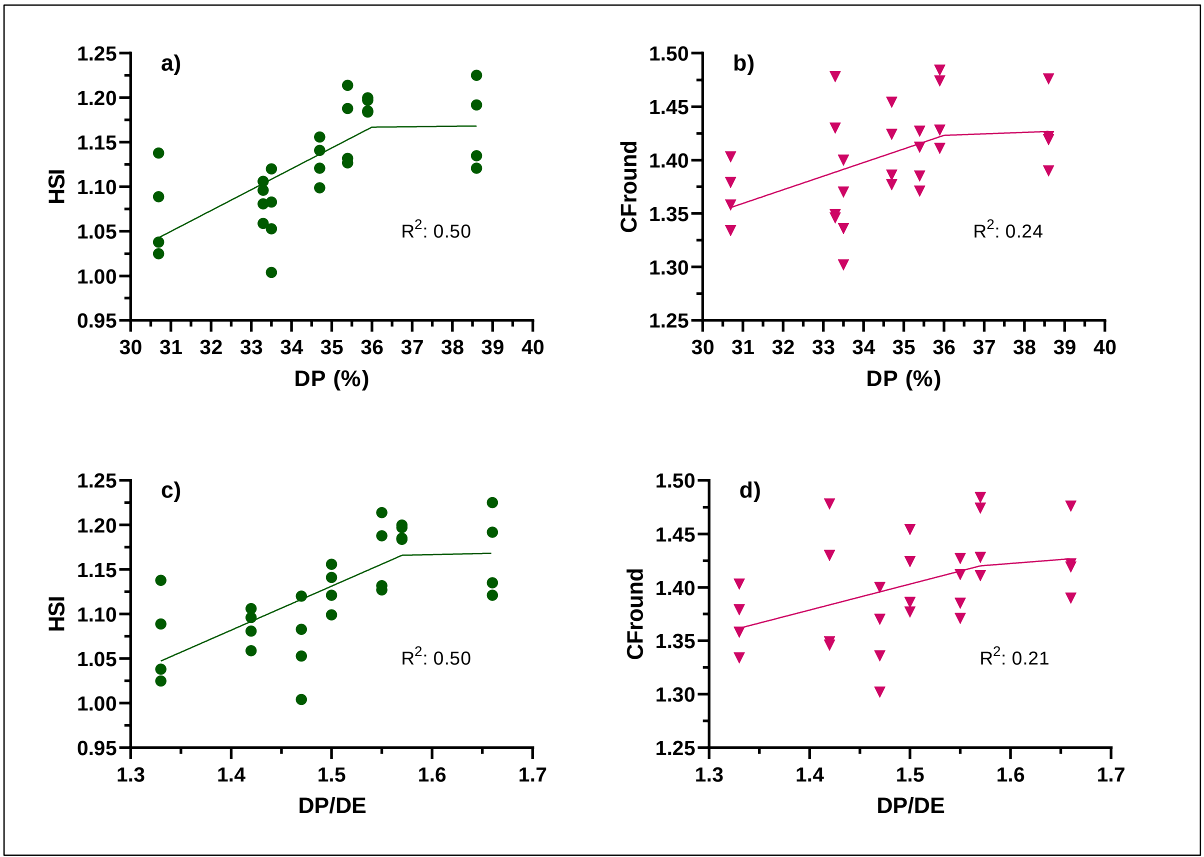


Figure 6: Relationships between digestible protein (DP) and DP/DE ratio for whole fish (CF_{round}) and hepatosomatic index (HSI).

Flesh colour by treatment and its relationship to digestible protein (DP), digestible energy (DE) and DP/DE ratio

To assess differences in the colour development of the salmon, Norwegian quality cuts (NQC) were taken and analysed using a Minolta Chroma Meter (Table 5). Colour development between the different treatments was not statistically different for any of the measured parameters. Data was also analysed with a linear or non-linear relationship between dietary digestible protein (DP), digestible energy (DE) and DP/DE ratio and colour development. Neither DP, DE nor DP/DE ratio of the diets had an influence on the colour development as measured using the Minolta reader.

Table 5: Mean Minolta Chroma meter colour parameters of analysed fish by treatment after a 49 day growth period. (Treatments differences were tested by a 2-way ANOVA model with Diet and Pen Section as variables)

Parameter / Diet	A	B	C	D	E	F	EE	2-way ANOVA p-values		
								Diet[1)]	**Sec[2)]**	**Diet x Sec[3)]**
Minolta L value	53.2	51.5	52.2	51.6	51.4	51.7	51.7	0.911	0.534	0.852
SEM	0.8	0.9	0.6	1.2	0.8	1.1	1.1			
Minolta a value	25.8	25.5	24.9	24.8	25.0	25.7	25.1	0.719	0.833	0.840
SEM	0.2	0.3	0.9	0.3	0.4	0.3	0.4			
Minolta b value	31.3	30.5	30.5	30.1	30.2	31.2	30.2	0.868	0.670	0.509
SEM	0.4	0.4	0.7	0.6	0.5	0.5	0.6			
Minolta Chroma	40.6	39.8	39.3	39.1	39.2	40.4	39.3	0.710	0.761	0.655
SEM	0.5	0.5	1.1	0.7	0.6	0.6	0.7			
Minolta Hue	50.5	50.1	50.8	50.5	50.3	50.5	50.3	0.303	0.865	0.529
SEM	0.2	0.2	0.4	0.3	0.1	0.2	0.2			

SEM: standard error of mean; 1) dietary treatment effect; 2) pen section effect 3) interaction effect of treatment x section; Means sharing same superscipts are not significantly different from each other (Tukey's test, $P<0.05$); L value: lightness; a value: redness; b value: yellowness; Chroma: saturation; Hue: yellow / red balance

DISCUSSION

This study was designed as a dose-response experiment with seven diets. Average fish weight at the start and end of the 49-day study period was 3.386 and 4.373 kg, respectively. Main objectives were to investigate the influence of dietary digestible protein levels (DP) and pellet size on the growth performance, yield, morphometric and colour parameters of Atlantic salmon under semi-commercial environmental conditions. To investigate furthermore the effect of pellet size on growth performance; Diets E and EE were designed to be different in pellet diameters. Diet analytics; however, revealed that there was pellet size variation not only between Diets E and EE but also between other experimental diets. Different pellet sizes and diameters will result in varying pellet weights and subsequently, different numbers of pellets / kg. Experimental diets were also found to vary in dietary digestible energy (DE). Therefore DE, DP/DE ratio and pellets/kg were considered as additional response parameters beside DP when analysing for linear or non-linear relationships. Calculations were conducted separately by splitting achieved data by the two different pen section or investigating differences by 2-way ANOVA models with Diet and Pen section as variables. This was necessary as predator interactions influenced the performance in the two section differently (Figure 2).

Weight gain was influenced by both digestible protein and pellet size in this study. Weight gain increased with increasing dietary DP in both pen sections until reaching a plateau at 36% DP (as-fed) (Figure 3a). Also Hepatosomatic index (HSI) and condition factor (CF_{round}) were increasing with increasing dietary DP and increasing DP/DE ratio until reaching plateau (Figures 6a-d). As for weight gain the model breakpoints for HSI and condition factor were found to be 36% DP (as-fed). Condition factor and HSI are measures for the quality of protein and energy metabolism. The finding that HSI, condition factor and weight gain had same model breakpoints confirmed that Atlantic salmon around 4500 g reached their optimum growth at 36% dietary DP in this study.

It is widely known that the optimal dietary DP content change with fish size. Further studies need to investigate whether found model breakpoints for the HSI, condition factor and DP/DE ration would also be valid in studies with other fish sizes than in the current study. If different fish sizes showing a similar reaction to the found DP/DE relationship as in this study, dietary DP inclusion models could be developed and safeguard optimal protein nutrition of Atlantic salmon.

Although significant, the associations described should be received with caution due to relatively low regression coefficients. A reason for the low explanatory power of the model may be related to the increasing predation pressure observed towards the end of the study. Different predator interactions are likely to be the reason for variations in feed intake and weight gain between the two cage sections of the study (Figure 2). Therefore, it should be taken into consideration that observed regression coefficients but also model break points could be different if the trial period had been extended as planned.

Pellet size and weight gain showed a positive linear relationship and higher average weight gains were achieved with bigger pellet sizes (less number of pellets / kg values) (Figure 3d). The underlying relationship between bigger pellet sizes and better performance could be easier feed accessibility in combination with lower metabolic effort to fill stomach capacity. Therefore, it cannot fully excluded that detected model break points were influenced by different feed intake behaviours in the different treatments, although no significant interactions between dietary DP levels and pellet size on weight gain or feed conversion detected (Figures 4, 5). Future studies need to focus on excluding any potential interactions between dietary digestible protein and pellet size parameters by ensuring the homogeneity of pellet size in production

Feed conversion ratio or colour development were not influenced by any of the analysed response parameters in this study.

CONCLUSION and IMPLEMENTATION

A reduced study runtime, predator interactions and physical feed parameter interactions made implementation of the findings difficult. The determined models and corresponding breakpoints were therefore considered as indications rather than absolute values. Depending on feed type and adopting a cautious approach, the following implementations were proposed:

- Dietary digestible protein requirement values were changed by 4 – 12% DP relatively, in diets for Atlantic salmon greater than 2500 g.
- Pellets for diets for Atlantic salmon greater than 2500 g should be produced with a minimum pellet size of 9 mm and a pellet diameter of 8.5 mm.

REFERENCES

National Research Council. 2011. *Nutrient Requirements of Fish and Shrimp*. Washington, DC: The National Academies