コイの成長と体組成に及ぼす飼料の無機塩混合物添加量

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Effect of Different Dietary Levels of Salt-mixtures on Growth and Body Composition in Carp

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A 56-day feed trial was conducted to examine the effect of feeding graded levels of a salt-mixture (0–6%), as compared with an established mineral supplement (4%, McCOLLUM's salt-mixture no. 185 plus trace elements), on the growth and body composition of carp.

Fish fed the ration containing 4% McCOLLUM's salt-mixture plus trace elements displayed the highest overall growth response and feed efficiency over the experimental test period. For those fish fed graded levels of the salt-mixtures, maximum growth was obtained at the 2% inclusion level.

In contrast to fish fed mineral supplemented rations, fish fed a ration containing no mineral supplement displayed reduced growth, low feed efficiency, loss of muscle tone, decreased heamatocrit, decreased haemoglobin and serum glucose concentration, elevated carcass moisture, Ca, P and Na content, and reduced carcass protein, lipid, Mg, Fe, Zn and Cu content.

The data present indicates the necessity of a dietary mineral supplement for carp fed a purified ration based on casein, potato starch, dextrin and lipid.

In contrast to rainbow trout *Salmo gairdneri*, carp *Cyprinus carpio* appear relatively insensitive to the presence or absence of a complete mineral supplement in purified diets based on casein, starch and oil. PFEFFER and MESKE[1] found no differences in the growth rate, feed efficiency or mineral retention of carp (mean weight 100 g) given such diets containing between 0 and 8% ash from fish meal. Earlier OGINO and KAMIZONO[2] using carp of mean weight 2 g found little difference in growth over a 7-week period between fish given diets supplemented with McCOLLUM's salt-mixture no. 185 plus trace elements[3] (mean final weight 5.09 g) and those given diets lacking such a mineral supplement (mean final weight 4.59 g).

These results are surprising in that dietary requirements for the following minerals have been established in carp: Mg;[4,5] P;[6] Fe;[5,7] Zn;[5,8] Cu and Mn.[5,9]

Although the lack of overt deficiency symptoms in the studies of PFEFFER and MESKE[1] and OGINO and KAMIZONO[2] can probably be explained on the basis of the short duration of their feeding trials, the present experiment was carried out to examine the situation further by determining the effect of feeding graded levels of a new salt-mixture and comparing it with an established mineral supplement (McCOLLUM's salt-mixture no 185 plus trace elements; given at a level of 4% in the diet), on the growth, feed efficiency and body composition of juvenile carp contained within a water-recirculation system.

Materials and Methods

Diets

The composition of the diets is shown in Table 1. Five diets comprising mainly casein, starch, dextrin and oil were formulated: one contained no supplemental mineral premix; three others contained, at concentrations of 2, 4 and 6 g/100 g, diet, a salt-mixture containing all the major and trace elements for which a dietary requirement has been shown in higher vertebrate animals; a fifth diet contained, at a concentration of 4 g premix/100 g diet a mineral supplement (McCOLLUM's salt-mixture with added trace elements).[3] The mineral supplements in these diets replaced polyethylene in the basal diet. The diets were formulated to contain 31% crude protein and 9% lipid, and were prepared as described previously.[10]

Fish

Common carp were obtained from the Cotswold

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Table 1. Composition of the experimental diets (% by weight)

Ingredient	Diet No.				
	1	2	3	4	5
Casein	33	33	33	33	33
Dextrin	20	20	20	20	20
Potato starch	25	25	25	25	25
Corn oil	6	6	6	6	6
Cod liver oil	3	3	3	3	3
Binder*1	4	4	4	4	4
Vitamin premix*2	2	2	2	2	2
Polyethylene	7	5	3	1	3
Test salt-mixture*3	—	2	4	6	—
McCOLLUM's salt-mixture*4	—	—	—	—	4
Nutrient content (as analysed)					
Moisture (%)	10.76	9.56	11.45	9.97	10.21
Crude protein (%)	31.19	32.22	30.93	30.83	31.14
Lipid (%)	9.24	8.78	9.21	8.75	9.20
Ash (%)	1.22	2.54	3.95	5.68	2.98
Ca (mg/100 g)	<10	542	1024	1570	400
P (mg/100 g)	18	386	704	974	589
Mg (mg/100 g)	4	35	63	92	65
K (mg/100 g)	4	49	81	123	41
Na (mg/100 g)	10	51	99	172	162
Fe (mg/100 g)	<1	10	21	31	19
Mn (mg/100 g)	0.02	3.09	6.45	9.92	1.12
Zn (mg/100 g)	0.29	3.89	6.77	9.56	4.49
Cu (mg/100 g)	<0.01	0.13	0.64	0.88	0.29

*1 Hydroxypropylmethylcellulose (Celacol, British Celanese Ltd.)

*2 To supply/100 g diet: Thiamine HCl 5 mg; Riboflavin 5 mg; Calcium pantothenate 10 mg; Niacin 20 mg; Pyridoxine HCL 4 mg; Biotin 0.6 mg; Folic acid 1.5 mg; Cyanocobalamin 0.01 mg; Inositol 200 mg; Ascorbic acid 100 mg; Choline chloride 400 mg; Manadione 4 mg; Para amino benzoic acid 5 mg; Alpha tocopherol acetate 40 mg; Vitamin A acetate 200 IU; Vitamin D_3 1000 IU.

*3 To supply/100 g premix: $Ca(H_2PO_4)_2 \cdot H_2O$ 73.75 g; $MgSO_4 \cdot 7H_2O$ 12.75 g; KCL 5.00 g; NaCl 4.75 g; $FeSO_4 \cdot 7H_2O$ 2.50 g; $ZnSO_4 \cdot 7H_2O$ 0.55 g; $MnSO_4 \cdot 4H_2O$ 0.508 g; $CuSO_4 \cdot 5H_2O$ 98.7 mg; $CoSO_4 \cdot 7H_2O$ 47.7 mg; $CrCL_3 \cdot 6H_2O$ 25.5 mg; KI 10 mg; $Na_2MoO_4 \cdot 2H_2O$ 12.75 mg; NaF 2.5 mg; $NiCL_2 \cdot 6H_2O$ 5 mg; $NaBO_2 \cdot 4H_2O$ 3.25 mg; $NaVO_3$ 0.75 mg; $SuCl_2 \cdot 2H_2O$ 0.50 mg; $Na_2SeO_4 \cdot 10H_2O$ 0.25 mg.

*4 McCOLLUM's salt-mixture no. 185 plus trace elements.[3)] To supply/100 g premix: Calcium lactate 32.70 g; K_2HPO_4 23.98 g; $CaHPO_4 \cdot 2H_2O$ 13.58 g; $MgSO_4 \cdot 7H_2O$ 13.20 g; $Na_2HPO_4 \cdot 2H_2O$ 8.72 g; NaCl 4.35 g; Ferric citrate 2,97 g; $ZnSO_4 \cdot 7H_2O$ 0.30 g; $CoCl_2 \cdot 6H_2O$ 100 mg; $MnSO_4 \cdot H_2O$ 80 mg; KI 15 mg; $AlCl_3 \cdot 6H_2O$ 15 mg; CuCl 10 mg.

Carp Farm, Gloucestershire, U.K. Eleven fish (mean weight 55 g) were randomly allotted to each of the dietary treatments, they were reared in 50 *l* circular plastic tanks contained in a recirculation system. A 140 *l* header tank supplied freshwater by gravity, via a ring main, to the experimental tanks at a rate of 2 *l*/min/tank. A constant bleed-in of freshwater (0.5 *l*/min) was supplied to the header tank throughout the experiment. The water temperature was maintained at 28±0.5°C and a 14 h light cycle provided by a fluorescent lighting. The major and trace element content of the tank water during the experimental period is shown in Table 2.

Table 2. Major and trace element content of tank water over experimental test period

Element	Mean*
Ca (mg/*l*)	11.83
Mg (mg/*l*)	2.10
Na (mg/*l*)	6.17
K (mg/*l*)	0.80
Fe (μg/*l*)	76.57
Zn (μg/*l*)	254.43
Mn (μg/*l*)	14.53
Cu (μg/*l*)	13.60

* Based on three samples taken after 0, 4 and 8 weeks respectively.

At the start of the experiment six fish were killed (by means of a sharp blow on the head) and stored at −20°C for subsequent carcass analysis. Fish were fed twice daily, six days a week at the rate of

2.5% body weight per day, and were weighed individually at weekly intervals during the eight week test period as described previously.[10] On the last day of the experiment all eleven fish from each treatment were killed. Six fish were used for gross chemical and mineral analysis. Blood samples were taken from the caudal vein from the five remaining fish, small amounts were used for haematocrit and haemoglobin estimation. The remainder was allowed to clot at room temperature, centrifuged and the resultant serum samples stored at −20°C for subsequent calcium and glucose analysis.

Chemical Methods

Moisture, crude protein (N×6.25), lipid, ash and mineral analyses of the whole fish carcass and of diets, and blood haematocrit and haemoglobin measurements were performed as described previously.[10] Total serum Ca was assayed fluorimetrically using a Corning model 940 calcium analyser. Serum glucose was measured in a Beckman Glucose Analyser.

Statistical Methods

Differences between treatments in mean body weight, carcass composition and blood constituents were tested for significance (P 0.05) by DUNCAN's multiple range test.[11]

Results

Growth curves of carp given the five experimental diets are shown in Fig. 1. The curve for fish given diet 1 differs from that for fish given the other four diets, mean weight ceasing to increase during the last week although mean weights (86.84 g for diet 1 against a range of 98.00–104.37 for diets 2–5) at eight weeks were not significantly different. Carp given diet 1 also displayed the lowest specific growth rate and feed efficiency of all the treatments (Table 3).

Carp fed the ration without mineral premix (diet 1) became sluggish in their movements about five weeks after the experiment started and by the end of the experiment their bodies became flaccid with a lack of muscle tone.

Concentrations of certain constituents of the blood or serum were also significantly affected by the absence of a mineral supplement in the diet (Table 3). Carp fed diet 1 had significantly lower haematocrit, blood haemoglobin and mean

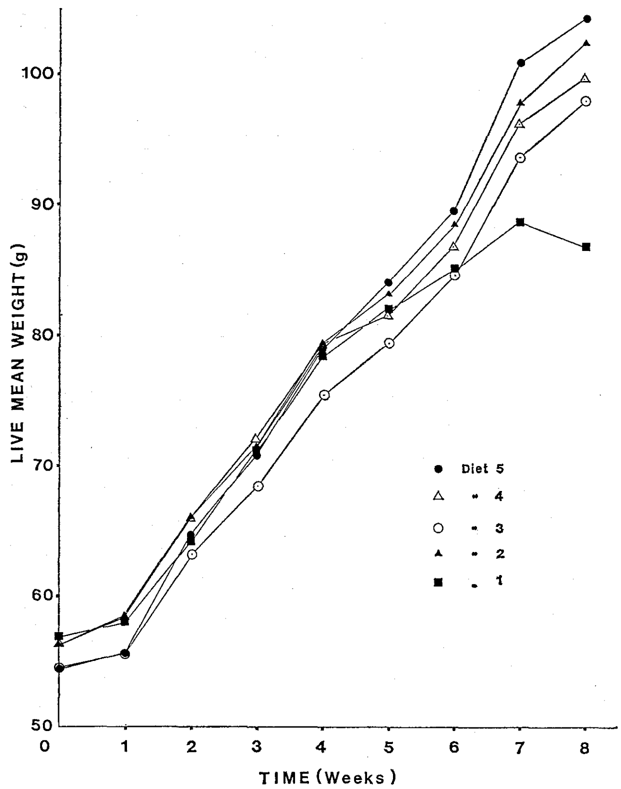

Fig. 1. Mean weight of fish on different diets at tuccessive weekly intervals.

Table 3. Growth, feed utilisation and blood analysis of common carp after 8 weeks

Diet No.	1	2	3	4	5	±SE*[1]
Mean initial weight (g)	56.88^{a}	56.13^{a}	54.56^{a}	56.20^{a}	54.52^{a}	5.183
Mean final weight (g)	86.84^{a}	102.26^{a}	98.00^{a}	99.66^{a}	104.37^{a}	9.095
Weight gain (%)	52.67	82.18	79.62	77.33	91.43	
Specific growth rate (%)*[2]	0.756	1.071	1.046	1.023	1.160	
Food intake (gpday)	1.372	1.377	1.337	1.363	1.369	
Weight gain (g/day)	0.535	0.824	0.776	0.776	0.890	
Feed efficiency*[3]	0.39	0.60	0.58	0.57	0.65	
Blood analysis						
Serum glucose (mg/100 m*l*)	53.5^{a}	75.8^{b}	81.8^{b}	81.7^{b}	109.0^{c}	6.817
Serum calcium (mg/100 m*l*)	9.61^{a}	9.30^{a}	9.73^{a}	10.14^{a}	9.30^{a}	0.408
Haematocrit (%)	27.75^{a}	38.83^{b}	34.19^{b}	35.00^{b}	35.12^{b}	1.550
Haemoglobin (g/100 m*l*)	7.30^{a}	10.79^{b}	9.24^{b}	9.55^{b}	10.03^{b}	0.417
MCHC (%)*[4]	26.31^{a}	27.79^{ab}	27.03^{ab}	27.29^{ab}	28.56^{b}	0.621

*1 Standard error, calculated from residual mean square in the analysis of variance
*2 Specific growth rate = $\log_e$ final body weight $-\log_e$ initial body weight,/time (days), $\times 100$
*3 Feed efficiency = weight gain, g/food fed, g
*4 Mean corpuscle haemoglobin concentration = haemoglobin concentration/haematocrit, $\times 100$
abc Mean values for components with the same superscripts are not significantly (p 0.05) different

Table 4. Proximate and mineral composition of whole fish carcass at the start and end of the 8 week feeding trial (values are expressed on a wet weight basis)

Constituent	Initial	After 8 weeks					
		1	2	3	4	5	±SE
Moisture (%)	80.08	78.45^{b}	74.40^{b}	74.43^{a}	73.43^{a}	75.19^{a}	0.393
Crude protein (%)	14.44	14.66^{a}	16.65^{a}	16.88^{b}	16.67^{b}	16.18^{b}	0.395
Lipid (%)	1.36	3.67^{a}	6.14^{c}	5.72^{bc}	6.13^{c}	4.77^{b}	0.365
Ash (%)	4.01	3.47^{c}	2.94$_{a}$	2.92^{a}	3.13^{ab}	3.29^{bc}	0.103
Ca (mg/g)	13.03	11.33	9.14	8.51	9.56	10.02	
P (mg/g)	6.42	5.47	4.61	4.53	5.35	5.24	
K (mg/g)	2.48	2.32	2.62	2.57	2.47	2.54	
Na (mg/g)	1.31	1.42	1.08	1.05	1.04	1.10	
Mg (mg/g)	0.42	0.27	0.36	0.35	0.39	0.40	
Fe (μg/g)	26.62	17.56	20.39	19.65	23.34	23.04	
Zn (μg/g)	47.81	30.17	43.52	46.03	50.48	47.14	
Cu (μg/g)	3.56	1.11	8.08	18.04	17.93	10.76	
Mn (μg/g)	4.28	2.15	2.19	2.38	2.48	1.51	

corpuscular haemoglobin content than fish given diets with mineral supplements. Serum glucose concentration was also significantly lower in these fish but, interestingly there was no significant difference in serum Ca concentration between treatments.

Table 4 shows that there were significant decreases in both carcass lipid and crude protein content and a significant increase in carcass moisture in fish fed diet 1 lacking a mineral supplement. These changes may explain in part the flaccidity of muscle observed after about 5 weeks in fish given this treatment. Carcass ash content of the diet 1 fish was significantly greater than in that of other treatments apart from diet 5, the trend in carp fed diets 2, 3 and 4 was for increasing carcass ash with increasing dietary intake of mineral premix.

Significantly higher concentrations of Ca and Na and significantly lower concentrations of Mg, K, Fe, Cu and Zn were present in the carcasses of carp fed diet 1 than in the carcasses of carp fed diets supplemented with minerals (Table 4). On the basis of deposition of minerals in the carcasses of carp over the experimental period (Table 5) fish fed diet 1 showed a net loss of Mg, Cu, Zn and Mn from the body and a net gain of Ca, P, Na and K. Within the other four dietary treatments all elements except Mn showed a positive

Table 5. Average dietary intake and carcass deposition of major and trace elements over the experimental test period

Mineral		Diet No.				
		1	2	3	4	5
Ca	Intake (g)	0.008	0.418	0.767	1.198	0.307
	Deposition (g)	0.243	0.203	0.123	0.220	0.335
P	Intake (g)	0.014	0.298	0.527	0.743	0.451
	Deposition (g)	0.110	0.111	0.094	0.172	0.197
Mg	Intake (mg)	3.1	27.0	47.2	70.2	49.8
	Deposition (mg)	−0.44	13.24	11.38	15.26	18.85
Na	Intake (mg)	7.7	39.3	74.1	131.3	124.2
	Deposition (mg)	48.80	36.91	31.43	30.02	43.39
K	Intake (mg)	3.8	37.8	60.6	93.9	31.4
	Deposition (mg)	60.41	128.7	116.5	106.8	129.9
Fe	Intake (mg)	<0.77	7.71	15.72	23.66	14.57
	Deposition (mg)	0.011	0.591	0.473	0.830	0.953
Cu	Intake (mg)	<0.007	0.097	0.475	0.672	0.224
	Deposition (mg)	−0.106	0.626	1.574	1.587	0.929
Zn	Intake (mg)	0.229	3.007	5.006	7.298	3.439
	Deposition (mg)	−0.099	1.767	1.902	2.344	2.313
Mn	Intake (mg)	0.013	2.387	4.832	7.571	0.859
	Deposition (mg)	−0.057	−0.016	−0.001	0.007	−0.076

balance over the experimental period. It is of interest that deposition of Ca, P, Na and K within fish given diet 1 and deposition of K and Cu within fish fed diets 2, 3, 4 and 5 was in excess of dietary intake.

Discussion

The trend toward reduced growth, low feed efficiency, loss of muscle tone and the evidence of anaemia in carp fed diet 1, containing no mineral supplement, contrast with the findings of OGINO and KAMIZONO[2] and of PFEFFER and MESKE.[1] The changes observed are, however, in line with pathologies associated with specific mineral deficiencies in carp. Anaemia in carp suffering from Fe deficiency has been described by SAKAMOTO and YONE.[7] Similarly, OGINO and CHIOU[4] described the gross symptoms of Mg deficiency in carp as sluggishness (together with loss of appetite). The significantly lower whole body levels of Mg, Fe, Cu and Zn further bear out the necessity for adequate dietary levels of these minerals.

The reduced carcass lipid content of carp fed diet 1 is surprising bearing in mind the increased carcass and viscera lipid content observed by OGINO and TAKEDA[6] with carp fed phosphorous deficient diets. Although a reduced carcass lipid content has also been observed in Chum salmon *Oncorhynchus keta*) fed a phosphorous deficient diet, the low carcass lipid content observed during the present investigation was probably consistent with the utilisation of body lipid reserves due to reduced feed intake. In addition, the high carcass phosphorous content of carp fed diet 1 suggests that body reserves of phosphorous had been sufficient to meet the animals requirement for this elements over the short 8-week feeding period.

In carp given diet 1 the levels of Ca, P, Na and K deposited in the carcass were in excess of the dietary intake over the 8-week feeding period. This can only be explained by absorption of these minerals from the surrounding rearing water. By the same argument it would appear that the water levels of Cu, Zn, Mg and Mn could not satisfy the requirements of the fish since there was a net loss of these minerals from the carcass of carp given diet 1.

The highest mean final body weight and feed efficiency was obtained in fish given the diet containing 4% MCCOLLUM's salt-mixture no. 185 plus trace elements. By contrast, in those carp given graded levels of the salt-mixture, maximum growth was obtained at the level of supplementation used in diet 2 (2%). However, this diet would almost certainly have been deficient in P, Mg, Fe and possibly Cu compared to the salt-mixture developed by OGINO *et al.*[12] (5% inclusion level) for carp on the basis of nutrient requirement data.

The data presented clearly indicates the necessity of a dietary mineral supplement for carp fed a purified ration based on casein, potato starch, dextrin and oil. The salt-mixture under examination was partially successful. However, the inclusion of additional P, Mg, Fe and Cu (so as to give a mean final dietary concentration of 600 mg/100 g available P, 50 mg/100 g Mg, 20 mg/100 g Fe and 0.3 mg/100 g Cu) may improve the performance of carp given this mineral salt-mixture. The absence of such trace elements as Cr, Se, V, Ni, B, Sn and Mo from the McCOLLUM's salt-mixture no. 185 plus trace elements was not obviously detrimental possibly because of the short duration of the experiment, possibly because these elements were present as contaminants of the dietary components and tank water. For example, the recent studies of SATOH *et al.*[5] have shown that more than 40 weeks were required to display overt trace element deficiency symptoms in carp fed a semi-purified ration in which white fish meal was used as the sole source of dietary protein.

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