Dietary intervention for a growing Australian Kelpie with Intestinal Lipid Malabsorption

Introduction

Recently, an inherited form of intestinal lipid malabsorption (ILM) in Australian Kelpies, was described (O'Brien et al., 2020).

Intestinal long chain fatty acid absorption mainly occurs in the jejunum where fatty acids are transported into the enterocytes in which long chain fatty acids are conjugated with coenzyme A and these conjugates are subsequently used for triglyceride synthesis (Iqbal et al., 2009; Meller et al., 2013; von Engelhardt et al., 2015). This reaction is triggered by acetyl-CoA synthetases (Watkins, 1997). ILM is an autosomal recessively inherited disorder in Australian Kelpies (O'Brien et al., 2020). The condition is caused by a deletion of ACSL5 (Acyl-CoA synthetase long chain family member 5) gene (O'Brien et al., 2020). ACSL5 plays an important role for lipid metabolism and fat deposition in carnivores (Zhao et al., 2019). Australian Kelpies with ILM fail to thrive and show persistent diarrhoea and steatorrhea from early age (before 6 weeks) onwards which distinct ILM from differential diagnoses such as hereditary selective ileal cobalamin malabsorption and exocrine pancreatic insufficiency. Affected dogs seem to outgrow the condition from 6 months onwards, however remain smaller than their littermates, have higher faecal output and remain intolerant to high-fat diets throughout their life (O'Brien et al., 2020). Therapies to overcome the deficit in ACSL5 are currently unknown, however addition of pancreatic enzymes to the food was described to improve the clinical presentation (O'Brien et al., 2020). Therefore, dietary intervention sets an important cornerstone in the management of these patients.

<u>Clinical report</u>

A 12-weeks-old male, intact Australian Kelpie puppy was referred to the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zürich after being diagnosed with ILM. A positive DNA test performed in Australia revealed that the dog is a double carrier for ILM and therefore should not be used for breeding. The dog had soft faeces which were sometimes whitish or covered with slime. Supplementing the dog with pancreas enzymes (Lypex®, each capsule contains Lipase: 30'000 Ph. Eur. E., Amylase: 18'750 Ph. Eur. E., Protease: 1'200 Ph. Eur. E) improved the faeces quality. Additionally, the dog was prescribed Vitamin B12 supplementation by the referring veterinarian. Body and muscle condition scores

were not available. At the timepoint of referral, the dog had a body weight (BW) of 4.0 kg which was 1.3 kg below the ideal growth curve for the expected adult weight. The BW of the father was 15.5 kg, the BW of the mother was 12.5 kg. However, after consultation with the dog's breeders, a lower adult weight is expected for this puppy, contrary to the usual practice of judging adult weight by the same-sex parent and an adult weight of 12.5 kg was estimated.

Diet check

The ingredients of the patient's subsequent diets are specified in Table 1. The nutritional contents of the patient's subsequent diets (Table 2 and 3) and the ideal growth curve (Figure 1) were calculated with DietCheck Munich ©2005 Version 3.0 (RV Software; based on (NRC, 2006), modified by Dobenecker and Kienzle). The recommended nutrient levels by DietCheck correspond to the recommended allowance (RA) of the respective nutrient as specified by the NRC 2006. Deviations of the recommendation by DietCheck Munich ©2005 from the RA for dietetic reasons are specified in Table 2 and 3.

The diet fed by the owner at the timepoint of referral at the age of 12 weeks (Diet A_{12w}, table 1) was based on a commercial complete dry food for adult dogs which was marketed as "grainfree". Unfortunately, the manufacturer did not provide us with complete analytical values, therefore a diet check regarding trace elements, various minerals and vitamins was not possible. The recommendations for protein, calcium and phosphorous were met for a growing dog of the patients weight and age at time of referral. The fat content of diet A was 20% and therefore did not meet the criteria of a fat-reduced diet. Except for zinc, no nutritional additives were declared on the food label of the dry food, therefore it is highly questionable whether trace elements were sufficiently covered by the dry food. Diet A_{12w} contained 88% of the recommended metabolic energy content for growing dogs (or approximately 80% of the recommended metabolic energy supply calculated according to NRC 2006). At the timepoint of referral, the dog's body weight was 25 % below the growth curve. The goal of the nutritional consultation was to provide the dog with a nutritionally balanced diet, taking into account his impaired fat absorption. Regarding the body weight, the calculated growth curve illustrates the maximal weight for each timepoint. The patient's BW should slowly approach the growth curve, avoiding excessive weight gain in a short time period.

Diet recommendation

Diet B was prescribed to support the nutritional requirements of a growing dog with impaired fat absorption. The diet was adapted according to the different growth phases (diet $B_{14w} - B_{44w}$,

numbers are indicating the timepoint in weeks of diet adaptation to the next level). For each growth phase the nutritional requirements were calculated based on the ideal weight according to the growth curve and the adult weight.

The main dietary ingredients in the order of their contribution towards the daily energy recommendation were: horse meat (3% fat), polished rice, dried deer meat (treat) and corn germ oil, as well as carrots, fennel and an omega 3 supplement. Vitamins and minerals were supplemented with additional bone meal, a mineral vitamin supplement and salt (Table 1).

As described above, the initial energy content of the prescribed diet matched the daily ME content of diet A_{12w} (88% of ME, as calculated with DietCheck Munich) for the next growth phase. The amount of all diet ingredients were calculated with the recommendations valid for the expected weight at the end of each growth phase (for the changes in recommendations please refer to Table 2). The initial ration recommendation included 9.5% fat on a dry matter basis, while the recommendations for essential fatty acids were met.

As the dog did not gain as much weight as expected, especially in week 13 and 14 (Figure 1), the energy content of the diet was increased in week 16 (98% ME, as calculated with DietCheck Munich) by increasing the amount of rice from 70 to 100 g (see arrow in Figure 1). The owner was instructed to increase the amount of rice stepwise over several days to allow the gastrointestinal tract to adapt to the diet change. After the increase in energy uptake, the dog approached the growth curve. In week 20 the bodyweight was slightly below the growth curve and the owner was reminded to decrease the energy content of the diet back to the original 88% ME by decreasing the amount of rice to 75 g to avoid that the dog's weight exceeds the growth curve (Figure 1, dotted arrow). However, the owner had only decreased the energy intake of the dog in week 26, resulting in an increased bodyweight exceeding the growth curve by 9% at the end of week 27.

Further feedback was not received until the age of 44 weeks, when the dog's body weight was clearly exceeding the growth curve by 4%. Hematology and blood biochemistry including markers for pancreas health (TLI and cPLI) were performed at 35 weeks of age. As the pancreas markers were within the range for healthy dogs, the owners decided in accordance with the private veterinarian to discontinue the Lypex®-treatment. At 39 weeks of age, faeces were microscopically examined for fatty acids and neutral fat, and other nutrients. Except for muscle fibers (slightly positive result), the result was negative. However, the dog was found to be *Giardia spp.*-positive and was treated with Panacur® (fenbendazole). The owners reported increased faecal output with a slimy surface during that time. The Lypex® treatment was initiated again (1 tablet per day, divided equally to be fed together with 3 meals). The owner

moreover reported that the dog was refusing to eat rice and fennel since week 42 and therefore they had experimented with the feeding ratio themselves, including the temporary use of horse meet with a higher fat content (15% fat) without prior consultation. However, the increased fat content in the feeding ration resulted in refusal by the dog. At 45 weeks of age, the dog was fed an owner-modified diet as described in detail in Table 1 (diet C_{45w}). Even though the fat content was decreased againby the owers by feeding the previously prescribed hprse meat with 3% fat, the dog had unstable stool quality at the timepoint of follow up (45 weeks of age). The owner was offered 2 newly calculated diet options for 45 weeks up to 1 year (52 weeks) of age (diet D_{52wa} and D_{52wb}). These were formulated with different carbohydrate sources (sweet potatoes and oats) and had an increased fiber content to support gut health. Moreover, previously offered treats which were rich in connective tissue, were excluded from the diet again.

At the age of 21 months, the owner was contacted for further feedback. The owner reported to have stopped feeding the prescribed homemade diet at the age of 13 months and had changed to a commercial dry food without further consultation. The dog currently weighs 12.5 kg which corresponds to the estimated adult weight. Lypex®-treatment was continued since the last feedback. The owner reported to have changed the dry food several times in the past, when the dog stops eating it. At the age of 21 months the dog was fed diet E_{21mo} . The diet E_{21mo} does not safely cover the recommendations for magnesium, iodine, iron, copper and zinc and the contents of B-vitamins and essential fatty acids were not provided by the manufacturer. For this reasons we recommended an alternative dry food (diet F_{adult}), based on an intestinal low-fat dry food, with the options to mix it with cottage cheese, rice or buckwheat, as this was requested by the owner. Diet E_{21mo} and F_{adult} were calculated according to the recommendations for adult dogs (DietCheck Munich, version 3).

Discussion

This case report highlights the dietary intervention for an Australian Kelpie with ILM. The dog was prescribed a homemade diet with a low fat content ($\leq 10\%$ DM with exception of a diet containing oats, Diet D_{52wb} (12% fat in DM)) while covering the recommendations for essential fatty acids. The temporary trial of the owner to feed horse meat with a higher fat content resulted in refusal of the meals by the dog and suggests that feeding a low-fat diet was crucial for healthy development and wellbeing in this ILM-patient. It is suspected that similar dietary management may also be helpful in other Australian Kelpies with ILM, however further case reports and

studies are needed to confirm and generalize the presented dietary approach for Australian Kelpies with ILM.

Besides feeding a low-fat diet, the dog was supplemented with pancreatic enzymes throughout the dietary intervention. When these were discontinued at the age of 8 months, the dog showed higher faecal output of varying consistency. As pancreas markers were within the reference range before the pancreatic enzymes were discontinued, it seems unlikely that the dog is suffering a parallel exocrine pancreas insufficiency, which would cause similar clinical symptoms such as steatorrhea (Westermarck et al., 2012). As enzyme supplementation does not interfere with the measurement of TLI, it is not expected that the laboratory diagnostic result was influenced thereof (Roth, 2011).

The initial rationale for pancreatic enzyme supplementation was to facilitate lipid digestion in order to make essential fatty acids readily available for absorption. Pancreatic enzymes break down dietary triglycerides to fatty acids in the intestinal lumen and absorption of fatty acids into the enterocyte is mediated both by active transporters which are unlikely to be influenced by pancreatic enzyme activity and via diffusion into the cell (Mansbach et al., 2007; Iqbal et al., 2009; Wang et al., 2013; Karpińska et al., 2022). The passive diffusion step of dietary fatty acid absorption into the enterocyte may be indirectly influenced by the pancreatic enzymes, as they likely increase the amount of readily absorbable fatty acids, especially as it is likely that pancreas secretion will otherwise be decreased due to the low fat diet of this patient (Spannagel et al., 1996). Moreover, the pancreas enzyme supplementation and transition to a solid diet was reported to improve the clinical condition of Kelpies with ILM (O'Brien et al., 2020). Unfortunately, the dietary compositions were not presented in the previous study. However the solid diets might have contained less fat than the milk the puppies consumed (60% fat on a ME-basis (Kamphues et al., 2014) which is more than average standard kibbles).

In the present case faecal output increased when the Lypex®-treatment was discontinued. Shortly afterwards the dog was tested positive for *Giardia spp*. which may have altered intestinal morphology such as villus length and crypt depth and therefore decreased the absorption capacity of the intestine (Buret et al., 1990). It is possible that the clinical consequences of Giardiasis only became evident when the Lypex® was discontinued as additional pancreatic enzymes might have improved absorption of the readily available nutrients to a certain extent as small intestinal damage may have interfered with brush border enzyme activity which potentially was compensated in part by the addition of pancreatic enzymes (Hooton et al., 2015). The net effect of pancreatic enzyme supplementation in this case is questionable in the present case and supplementation in future cases should be based on

clinical evaluation of the individual patient (e.g. faecal scoring) until further research elucidating this topic has been performed. As side effects of pancreatic enzyme supplementation are rare (oral bleeding in high doses of non-coated enzymatic products were reported) it seems justifiable to evaluate the effect for each individual patient with a coated enzyme product (Rutz et al., 2002; Westermarck et al., 2012).

This case report shows that growing ILM-patients can reach their full growth potential when fed a low-fat diet, covering large parts of the energy requirements by carbohydrates and protein. However, in this case the dog exceeded the growth curve at 27 weeks of age which was due to an increased energy supply that was not reduced in time by the owner as previously suggested. Although the owners were very clearly informed about the consequences of excessive weight gain such as impaired bone health (Giordanella et al., 2021) they were too excited about the puppy's now rapid growth and did not carry out the agreed energy adjustment in time.

Previously, it was discussed that fat digestion is improved in Australian Kelpies with ILM after the age of 6 months (O'Brien et al., 2020). The present case however, showed adverse symptoms after being fed a higher fat-content at the age of 8 months. Until adulthood the present patient is very sensitive to changes in the dietary composition (higher content of connective tissue, changes in fat content, etc). It is therefore questionable if feeding a nonadapted diet to the patient for the first 12 weeks of age potentially leads to persistent changes in intestinal architecture which make the patient more prone to indigestions. Both intestinal microbiome and intestinal architecture develop strongly in the first months of life (Modina et al., 2021; Garrigues et al., 2022). Therefore, it might be a key factor to adapt the diet for these patients as early as possible. As ILM already can become evident in young puppies, strategies should be developed to help these puppies from early onset of clinical symptoms which may potentially include the period of milk consumption.

Conclusion

The present case report highlights the nutritional management of an Australian Kelpie with ILM who initially was 25% below the calculated growth curve. It shows that a patient with ILM can reach his full growth potential, if fed a low-fat diet covering the nutritional recommendations for growing dogs. In this case, the addition of pancreatic enzymes showed beneficial effects on faecal quality. However, it remains to be determined if pancreatic enzyme supplementation is generally necessary or if feeding a low-fat diet from early onset of clinical symptoms in affected dogs is sufficient.

Tables and figures

Table 1: Quantity of ingredients in the diets per day. All values were converted to daily amounts based on the owner's information and represent the raw weight.

Ingredient	Unit	Diet A _{12w}	Diet B _{14w}	Diet B _{18w}	Diet B _{44w}	Diet C _{45w}	Diet D _{52wa}	Diet D _{52wb}	Diet E _{21mo}	Diet F _{adul}
Commercial dry food ^a	g	160	/	/	/	/	/	/	/	/
Commercial semi-moist food ^b	g	/	/	/	/	/	/	/	150	/
Commercial dry food ^c	g	/	/	/	/	/	/	/	/	155
Dried venison meat ^d	g	10	10	10	10	/	/	/	/	/
Horse meat (3% fat)	g	/	250	250	250	340	340	340	/	/
Cottage cheese	g	/	/	/	/	/	/	/	6	/
Rice, polished	g	/	65	100	75-100	40	40	/	14	/
Sweet potatoes	g	/	/	/	/	40	150	/	/	/
Oats	g	/	/	/	/	/	/	80	/	/
Buckwheat	g	/	/	/	/	/	/	/	14	/
Carrots	g	/	50	50	50	50	35	35	/	/
Fennel	g	/	50	50	50	/	/	/	/	/
Zucchini	g	/	/	/	/	/	80	80	/	/
Corn germ oil	g	/	4	4	4	4	/	/	/	/
Hemp seed oil	g	/	/	/	/	/	5	5	/	/
Coconut oil	g	/	/	/	/	/	/	/	2	/
Omega 3 capsule ^e	piece	/	1	1	1	1	1	1	/	/
Mineral vitamin supplement ^f	g	/	5	5	5	5	5	5	/	/

Bone meal	g	/	2.5	3.5	3.5	3.5	3.5	3.5	/	/
Salt, non-iodized	g	/	0.9	0.9	0.9	0.9	0.9	0.9	/	/
Lungs (treat)	g	/	/	/	/	80	10	10	/	/
Beef chewing article	g	/	/	/	/	10	/	/	/	/

Diet A_{12w} was fed by the animal owner before consultation at the age of 12 weeks.

Diets $B_{14w-44w}$ were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich. The numbers refer to the week up to which the diet should be fed (e.g Diet B_{14} should be fed up to the age of 14 weeks).

Diet C_{45w} was fed by the animal owner after consultation at the age of 45 weeks and included changes made by the owner.

Diet D_{52wa and b} were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich for the age of 45 to 52 weeks.

Diet E_{21mo} was fed by the animal owner at the age of 21 months without prior consultation with the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich.

Diet F_{adult} was recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich at the age of 21 month for adulthood. Alternatives were calculated with addition of a small amount of rice or buckwheat and cottage cheese. Moreover, the dry food can be replaced by feeding 550 g of the corresponding low-fat wet food of the same brand.

^a The commercial dry food is a complete food for adult dogs. The food producer did not provide us with complete analytical values, therefore it was impossible to check the nutritional adequacy to detail.

^b The commercial semi-moist food is a complete food for adult dogs. The food producer did not provide us with complete analytical values for some nutrients and fatty acids (marked as NA in table 2 and 3). The product does not contain additional fat/oil.

^c The commercial dry food is marketed with the particular utritional purpose to compensate for maldigestion.

^d 10 g of dried venison meat correspond to 35 g of fresh product.

^e Each capsule contains 234 mg EPA and 156 mg DHA.

^f Mineral & vitamin supplement composition per 100g product: 19.8 g calcium, 6 g phosphorous, 1.2 g magnesium, 1.6 g potassium, 301 mg iron, 60 mg copper, 300 mg zinc, 37 mg manganese, 1.4 g chloride, 6.6 mg iodine, 35000 IU vitamin A, 3000 IU vitamin D, 300 mg vitamin E, 8.3 mg vitamin B1, 40 mg vitamin B2, 10 mg vitamin B6, 0.12 mg vitamin B12, 0.66 mg biotine, 56 mg niacin, 22.5 mg panthothenic acid

Table 2: Comparison of calculated proximate analysis and mineral contents in subsequent diets to the daily recommended allowance (RA) for a dog	
with 12.5 kg adult weight.	

Item	Unit	RA _{12w} ^a	Diet	RA _{14w} ^a	Diet	RA _{18w} ^a	Diet	RA _{44w} ^a	Diet	RA _{52w} ^a	Diet	Diet	Diet	RA _{adult} ^a	Diet	Diet
			A _{12w}		B _{14w}		B _{18w}		B44w		C _{45w}	D _{52wa}	D52wb		E21mo	Fadult
Energy; ME	MJ	2.9	2.6	3.3	2.9	3.5	3.4	3.5	3.2	3.5	3.4	3.4	3.4	2.7	2.2	2.2
		[3.2]		[3.7]		[3.8]		[3.9]		[3.8]				[2.6]		
Dry matter	g		150		158		189		168		173	196	186		132	141
Crude	g	43	68	50	68	51	69	52	69	53	92	81	87	36	45	38
protein						[40]		[40]		[40]				[22]		
Crude fat	g	[16]	30	[19]	15	[20]	15	[20]	15	[19]	18	18	23	[12]	13	9
NfE	g		29		62		90		70		48	75	57		62	79
Crude ash	g		14		11		12		11		14	18	14		8	12
Crude fiber	g		8		2.7		2.7		2.7		1.3	4	5.5		5	2.9
Methionine +	g	1.35	NA	1.55	4.41	1.60	4.49	1.61	4.43	1.65	5.49	5.47	5.71	1.41	NA	1.69
cysteine						[1.22]		[1.22]		[1.20]						
Calcium	mg	1159	3204	1324	1412	1372	1561	1380	1535	1409	1505	1368	1539	866	1060	1938
		[2319]		[2649]		[2748]		[2763]		[2708]						
Phosphorous	mg	966	2309	1103	1103	1143	1351	1150	1174	1174	1353	1217	1503	649	962	1395
		[1933]		[2207]		[2290]		[2302]		[2256]						
Magnesium	mg	77	NA	88	166	91	171	92	168	94	175	203	271	130	104	140
Potassium	mg	850	NA	971	1496	1006	1533	1012	1507	1033	1708	2144	1760	866	965	1163
														[933]		
Sodium	mg	425	NA	485	561	503	563	506	561	516	651	547	545	173	465	698
Chloride	mg	556	NA	635	691	659	691	663	691	676	683	728	730	260	NA	1256
Iron	mg	17	NA	19.4	29.8	20.1	30.0	20.2	29.9	20.7	36.9	34.1	36.3	6.5	NA	86.8

Copper	mg	2.1	NA	2.4	3.6	2.5	3.7	2.5	3.6	2.5	3.8	3.7	3.9	1.3	0.9	3.1
Zinc	mg	19.3	16	22.1	24.4	22.9	24.9	23.0	24.6	23.5	27.2	27.3	28.6	13.0	12.6	21.1
Manganese	mg	1.1	NA	1.2	2.2	1.3	2.2	1.3	2.2	1.3	2.3	2.8	5.8	1.0	3.0	5.3
Iodine	μg	170	NA	194	338	201	339	202	339	207	336	340	339	190	121	341

Abbreviations[:] NA, value not available

Diet A_{12w} was fed by the animal owner before consultation at the age of 12 weeks.

Diets $B_{14w-44w}$ were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich. The numbers refer to the week up to which the diet should be fed (e.g Diet B_{14} should be fed up to the age of 14 weeks).

Diet C_{45w} was fed by the animal owner after consultation at the age of 45 weeks and included changes made by the owner.

Diet D_{52wa and b} were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich for the age of 45 to 52 weeks.

Diet E_{21mo} was fed by the animal owner at the age of 21 months without prior consultation with the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich.

Diet F_{adult} was recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich at the age of 21 month for adulthood. Alternatives were calculated with addition of a small amount of rice or buckwheat and cottage cheese. Moreover, the dry food can be replaced by feeding 550 g of the corresponding low-fat wet food of the same brand.

^a Recommended allowance (RA) as calculated with DietCheck Munich Version 3.0 for growing dogs or adult dogs. Values in brackets [] show the respective RA as detailed by the NRC (2006) for growing dogs at 4-14 weeks of age or > 14 weeks of age or inactive adult dogs if older than 52 weeks depending on the actual weight of the patient in case of deviation of > 5% from the values given by DietCheck.

RA_{12w}: RA calculated with an actual body weight of 4 kg. Energy supply in Diet A corresponds to 88% ME as calculated with DietCheck Munich.

RA_{14w:} RA for week 13 - 14, calculated with body weight of 5.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B₁₄ corresponds to 88% ME as calculated with DietCheck Munich.

 RA_{18w} : RA for week 15 – 18, calculated with a body weight of 6.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B_{18} corresponds to 98% ME as calculated with DietCheck Munich.

 RA_{44w} : RA for week 19 – 44, calculated with a body weight of 10.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B_{44} corresponds to 88% ME as calculated with DietCheck Munich.

 RA_{52w} : RA for week 45 – 52, calculated with a body weight of 11.7 kg and expected adult weight of 12.5 kg. Energy supply in Diet C₄₅ and D₅₂ correspond to 98% ME as calculated with DietCheck Munich.

RA_{adult:} RA calculated with ideal and actual body weight of 12.5 kg. Energy supply in Diet F_{adult} corresponds to 83% ME as calculated with DietCheck Munich.

Table 3: Comparison of calculated vitamin and fatty acid contents in subsequent diets to the daily recommended allowance (RA) for a dog with
12.5 kg adult weight.

Item	Unit	RA _{12w} ^a	Diet	RA _{14w} ^a	Diet	RA _{18w} ^a	Diet	RA _{44w} ^a	Diet B _{44w}	RA _{52w} ^a	Diet	Diet	Diet	RA adult ^a	Diet	Diet
			A _{12w}		B _{14w}		B _{18w}				C _{45w}	D _{52wa}	D52wb		E21mo	Fadult
Vitamin A	IU	976	NA	1115	1932	1155	1932	1162	1932	1186	2114	2083	2083	1094	1205	2902
Vitamin D	IU	105	NA	120	180	124	180	125	180	128	191	191	191	118	135	287
Vitamin E	mg	6	NA	7	28	7	28	7	28	7	27	33	27	6	15	22
Vitamin B1	mg	0.26	NA	0.30	1.04	0.31	1.06	0.31	1.04	0.32	1.04	1.18	1.53	0.48	0.56	2.14
Vitamin B2	mg	1.02	NA	1.16	2.45	1.21	2.46	1.21	2.45	1.24	2.64	2.53	2.57	1.13	0.56	3.86
Vitamin B6	mg	0.29	NA	0.33	1.85	0.34	1.90	0.35	1.86	0.35	2.13	2.45	2.11	0.32	0.49	1.69
Vitamin B12	μg	7	NA	8	14	8	14	8	14	8	19	16	16	8	12	24
Biotin	mg	3	NA	4	43	4	43	4	43	4	48	43	59	2	45	158
Niacin	mg	3	NA	4	17	4	17	4	17	4	24	21	19	4	NA	18
Pantothenic	mg	2.9	NA	3.31	3.59	3.43	3.81	3.45	3.65	3.52	4.93	4.89	3.48	3.25	NA	4.53
acid																
Linoleic acid ^c	g	[2.6]	NA	[2.9]	2.9	[3.0]	3.0	[3.0]	2.9	[3.0]	3.16	3.66	5.54	[2.39]	NA	3.10
α-Linolenic acid ^c	g	[0.15]	NA	[0.18]	0.73	[0.18]	0.73	[0.18]	0.73	[0.18]	1.24	1.96	2.03	[0.09]	NA	0.34
Arachidonic Acid ^c	g	[0.06]	NA	[0.07]	0.30	[0.07]	0.30	[0.07]	0.30	[0.07]	0.51	0.42	0.42		NA	NA
EPA & DHA	g	[0.10]	NA	[0.11]	0.39	[0.12]	0.39	[0.12]	0.39	[0.12]	0.39	0.39	0.39	[0.20]	NA	0.279

Abbreviations: EPA, eicosapentanoic acid; DHA, docosahexaenoic acid; NA, value not available

Diet A_{12w} was fed by the animal owner before consultation at the age of 12 weeks.

Diets $B_{14w-44w}$ were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich. The numbers refer to the week up to which the diet should be fed (e.g Diet B_{14} should be fed up to the age of 14 weeks).

Diet C_{45w} was fed by the animal owner after consultation at the age of 45 weeks and included changes made by the owner.

Diet D_{52wa and b} were recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich for the age of 45 to 52 weeks.

Diet E_{21mo} was fed by the animal owner at the age of 21 months without prior consultation with the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich.

Diet F_{adult} was recommended by the Institute of Animal Nutrition and Dietetics of the Vetsuisse Faculty, University of Zurich at the age of 21 month for adulthood. Alternatives were calculated with addition of a small amount of rice or buckwheat and cottage cheese. Moreover, the dry food can be replaced by feeding 550 g of the corresponding low-fat wet food of the same brand.

^a Recommended allowance (RA) as calculated with DietCheck Munich Version 3.0 for growing dogs or adult dogs. Values in brackets [] show the respective RA as detailed by the NRC (2006) for growing dogs at 4-14 weeks of age or > 14 weeks of age or adult dogs if older than 52 weeks depending on the actual weight of the patient in case of deviation of > 5% from the values given by DietCheck.

RA_{12w}: calculated with an actual body weight of 4 kg. Energy supply in Diet A corresponds to 88% ME as calculated with DietCheck Munich.

 $RA_{14w:}$ recommendations for week 13 - 14, calculated with body weight of 5.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B₁₄ corresponds to 88% ME as calculated with DietCheck Munich.

 RA_{18w} : recommendations for week 15 – 18, calculated with a body weight of 6.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B_{18} corresponds to 98% ME as calculated with DietCheck Munich.

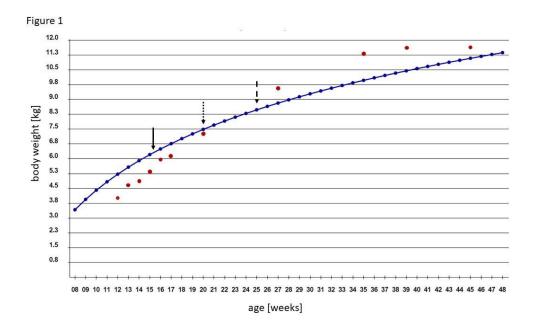
 RA_{44w} : recommendations for week 19 – 44, calculated with a body weight of 10.8 kg and expected adult weight of 12.5 kg. Energy supply in Diet B₄₄ corresponds to 90% ME as calculated with DietCheck Munich.

 RA_{52w} : recommendations for week 45 – 52, calculated with a body weight of 11.7 kg and expected adult weight of 12.5 kg. Energy supply in Diet C₄₅ and D₅₂ correspond to 98% ME as calculated with DietCheck Munich.

RA_{adult:} RA calculated with ideal and actual body weight of 12.5 kg. Energy supply in Diet F_{adult} corresponds to 83% ME as calculated with DietCheck Munich.

^c Fatty acid content of the feeding rations was calculated based on values published in SOUCI or USDA (deer).

Figure 1: Growth curve (blue dots/ line, calculated with DietCheck Munich Version 3.0) and actual body weight development (red dots).



Black arrow (solid line) shows increase of metabolic energy intake (from 88% to 98% ME as calculated by DietCheck Munich, version 3). The dotted black arrow shows the timepoint of suggested adaptation of ME-intake (88% ME) and the dashed black arrow shows the timepoint when the owner adapted ME-intake to 88% ME (as calculated with DietCheck Munich version 3).

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