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By

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Vegan diet and its effects on the dog's health

Veganiška dieta ir jos efektas šunų sveikatai

MASTER THESIS

of Integrated Studies of Veterinary Medicine

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CONFIRMATION OF THE INDEPENDENCE OF DONE WORK

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SUMMARY

This research was conducted in the Department of Animal Husbandry in the Veterinary Academy of the Lithuanian University of Health Sciences in 2019. In this investigation, dogs in Scheer, Germany, were fed two different diets: vegan and meat-based.

The nutritional adequacy of a vegan diet was determined by analysis of blood samples from 40 dogs, 20 of which were fed a 100% plant-based vegan diet for an average of 2.15 years, and a control group of 20 fed a meat-based diet. The results showed the same number of surpluses in both groups; however, the vegan group had only two nutritional deficiencies compared to 11 in the meat fed group. Statistically significant differences ($p < 0.01$) were found between the groups in iron, vitamin B12 and folic acid concentrations. Total protein, calcium and magnesium were not significantly different ($p > 0.05$).

To further evaluate the impact of a plant-based diet on dog health, eight dogs were put on a six-week feeding trial. The dogs were split into two groups of four dogs each; the control group was fed a meat-based diet, and the other group was fed a vegan diet. Blood analyses were performed prior to the start and at the end of the trial. The results showed that most of the values were not significantly changed. Some folic acid, B12 and iron deficiencies detected prior to the trial reached recommended healthy ranges during the trial on a vegan diet, although one dog experienced a folic acid surplus and another dog a folic acid deficiency.

All participants from all groups were determined to be in overall good health or in a condition that would not affect the blood chemistry parameters. These included total protein, vitamin B12, folic acid, calcium, magnesium, iron, taurine and L-carnitine. Laboratories analysing blood samples in Germany were Laboklin (seven samples), EasyLAB (two samples), IDEXX (37 samples), SYNLAB (one sample); in Australia, ASAP LABORATORY (two samples); and in England, AXIOM VETERINARY LABORATORIES (two samples). Veterinarians performed physical examinations during blood sample collection in various cities in Germany (including Stuttgart and Regensburg), England (Newton Abbot) and Australia (Melbourne).

To collect additional data from dog owners feeding a vegan or partially vegan diet, a questionnaire (initially presented to several thousand potential participants) was completed by 250 people.

Blood chemistry analysis and physical examinations of the vegan dogs in this study together clearly indicate that a vegan diet can be healthy and adequate for dogs, and in some cases, even improve overall health. The additional data collected from 250 dog owners feeding a plant-based diet strongly supported this conclusion.

Keywords: vegan, dog food, climate change, animal ethics, greenhouse gases, water usage

SANTRAUKA

Tyrimas buvo atliktas 2019 m. Lietuvos sveikatos mokslų universiteto Veterinarijos akademijos Gyvulininkystės mokslų fakultete ir Scheeryje, Vokietijoje. Bandymas buvo atliekamas su šunimis kurie buvo šeriami skirtingais pašarais, bandomoji grupė buvo šeriama 100% augalinės kilmės racionu, bandomoji grupė buvo šeriama pašarais su mėsa, Scheeryje, Vokietijoje.

Tyrime dalyvavo 40 šunų, po 20 kiekvienoje grupėje, veganų dietos tinkamumas šunims buvo nustatytas iš kraujo ir bendros šunų apžiūros. Rezultatai parodė, abiejų grupių šunų mityba buvo subalansuota ir visavertė, statistiškai reikšmingi skirtumai buvo stebėti tarp: geležies, vitamino B12 ir Folio rūgšties, P <0,01, tarp bendro baltymo, kalcio ir magnio statistiškai reikšmingo skirtumų nebuvo (P> 0,05).

Norint įvertinti ir įvertinti veganinės dietos pakeitimo poveikį šunų sveikatai, papildomai buvo įvykdytas bandymas, panaudojant 8 šunis. Šunys buvo padalyti į 2 vienodo dydžio grupes po 4, kontrolinė grupė buvo šeriama pašarais pagamintais mėsos pagrindu, bandomoji grupė buvo šeriama 100% augalinės kilmės racionu, prieš tyrimą ir jo pabaigoje buvo imtas kraujas. Visi metu dalyviai buvo geros sveikatos arba tokios būklės, kuri nepaveiktų kraujo parametrų, įskaitant bendrą baltymą, vitaminą B12, Folio rūgštį, kalcį, magnį, geležį, tauriną ir L-karnitinas. Kraujo mėginiai buvo analizuojami šiose Vokietijos laboratorijose: Laboklin (7 mėginiai), EasyLAB (2 mėginiai), IDEXX (37 mėginiai), SYNLAB (1 mėginys), ASAP LABORATORY (2 mėginiai), AXIOM VETERINARINĖ LABORATORIJA (2 mėginiai). Šunų fizinę būklė buvo įvertinta veterinarijos klinikoje Voketijoje (Stuttgart), Anglijoje (Newton Abbot) ir Australijoja (Melbourne).

Papildomus duomenys iš šunų savininkų, maitinančių veganų ar iš dalies veganų racioną buvo surinki įvygdžius apklausą. Keliems tūkstančiams potencialių dalyvių buvo pateiktas klausimynas, iš kurių 484 pradėjo apklausą, tačiau 250 užpildė apklausą. Apklausos rezultatai parodė, kad veganiška dieta gali būti sveika ir tinkama šunims.

Tyrimo tikslas buvo atlikti palyginamąją šunų sveikatos ir veganų dietos suderinamumo analizę. Papildomai atlikti apklausą siekiant surinkti duomenis apie šunis, šeriamus veganiška ar vegetariška dieta.

Raktažodžiai: veganas, šunų pašaras, klimato pokyčiai, gyvūnų etika, teisės, šiltnamio efektą sukeliančios dujos, vanduo;

INTRODUCTION

Our planet is changing at an unprecedented rate due to human intervention, and multiple anthropogenic influences have led to the current ongoing mass extinction, only the sixth in Earth's history. Today, up to one million animal and plant species are under threat of extinction (1); Atmospheric concentrations of carbon dioxide, methane and nitrous oxide are unusually high compared to the last 800,000 years, the rate of sea-level rise in the previous 70 years is higher than its mean rate of the last 2000 years and 1983-2012 was likely the warmest 30-year period in the last 1400 years (2).

Change is urgently needed as continued greenhouse gas emissions increase the likelihood of irreversible damage for all life on earth. Pollution and environmental destruction are the top concerns among young people in Germany (3), and all EU countries are predicted to fall short of the Paris Agreement goals by 2030 (4). Humans required more than 200.000 years to reach a world population of 1 billion, and in the last 200 years, the world population has increased to more than 7 billion people (5). Approximately 6.5% of all people ever born are currently alive (5).

With the opportunity to write a master thesis and the freedom of choosing a topic, the first choice might have been in the field of surgery but knowing the newest climate data it would not have made much sense focusing mainly on professional skills while facing the sixth mass extinction and running towards catastrophic future prediction on how climate change will soon affect our all lives. One aim of this study was to produce a thesis in the veterinary field that could be of potential importance in addressing climate change, loss of biodiversity, species extinction and pollution and, therefore the violation of animal and human rights. After many hours of research and studying the scientific consensus, I determined that the greatest impact may be in the field of nutrition. Livestock systems occupy 45% of global land surface area (6) and the conversion of feed to edible meat is largely inefficient: for 100kg feed, cattle produces only 4kg of edible meat, pork produces 11kg, chicken produces 22kg and fish produces 56kg (7). Livestock production contributes 18 (8)–51% (9) of all global CO_2 emissions and is, therefore, one of the largest contributors to climate change, even more than all transportation systems combined (including automobiles, aircraft and shipping) (7). Additionally, animal agriculture is a major source of water quality degradation and ocean dead zones.

There is limited awareness in the general public about the environmental impacts of a non-vegan diet. Students have almost no knowledge about the environmental impact of the food they consume, and while most are aware of the climate crisis, many are not strict practitioners of pro-environmental behaviour (10). In general, the impacts different sources of nutrition have on our planet are greatly underestimated.

Therefore, the aim of this study was to investigate the possibility of replacing the most resource-intensive ingredients of the canine diet (animal products) with those that can be more efficiently produced (plant products), while maintaining or potentially improving dog health. Dogs that had been fed a purely vegan diet for several months to years were recruited and blood samples collected to compare to official recommended healthy ranges and to a control group. Additional information was obtained through physical examinations. For further investigation, several dogs were put on a vegan dog food trial. For every vegan-fed dog in this study, a conventional, meat-based fed dog was used for comparison.

Hypothesis

Plant-based alimentation for canines could drastically reduce the demand for high impact products from animal agriculture, which is arguably the leading greenhouse gas emitter and primary driver of climate change.

Research objectives

The goal of this research is to determine the nutritional adequacy of a vegan diet for dogs.

Research tasks

1. Evaluation of the adequacy of a vegan diet for dogs by analysing blood from dogs being fed a vegan diet to compare values to officially recommended ranges and to a control group fed a meat-based diet.
2. Evaluation of the adequacy of a vegan diet for dogs by comparing before and after blood chemistry values of eight dogs subject to a vegan diet trial.
3. Evaluation of the adequacy of a vegan diet for dogs by directly analysing vegan food ratios of randomly selected vegan dog owners, with the aid of the official and licensed FutterMedicus veterinary feed calculator.
4. Collection and analysis of questionnaire data from 250 dog owners feeding a completely or partially vegan diet.

1. LITERATUR REVIEW

1.1 Proteins and amino acids

Proteins are defined as a complex organic compound containing hydrogen, carbon, oxygen, nitrogen and depending on type of protein also sulfur, the characterizing element is nitrogen. All proteins have a common characteristic, being build up from single units called amino acids (AA) (12). If we were to compare all macromolecules in the dog's organism, the protein would have the most diverse range of function of them all. Proteins serve as structural, regulatory, protective or even contractile component. Additionally, proteins can serve as enzymes and can be used as transport vehicle, integrated in membranes and used for storage or even posses' toxic properties (13). All proteins are made up of multiple amino acids called polymers.

Proteins are the building blocks for all cells in the dog's organism, needed to create hormones, antibodies, organs, the brain and every single hair making up the dog's fur. The main structural component of all body organs and tissues are proteins taking the form of: collagen and elastin which can be found in tendons; ligaments and cartilage; contractile proteins known as actin and myosin in muscle tissue; keratin that are found in nails, hair and skin; Proteins also are also of great importance when we examine the blood, hemoglobin, transferrin, albumin and globulin are all blood proteins; Hormones like insulin, enzymes and antibodies are functional proteins; breaking of amino groups by deamination or transamination resulting in amino acids are a source of energy (13).

Of especial importance are the 10 essential amino acids for dogs (11 for cats), meaning the body is not able to synthesize those particular amino acids by himself, to be exact, the carbon skeletons of these 10 AA are can´t be synthesized by the organism (13). Those amino acids are essential as if they are not present as building blocks for several biological active compounds, the synthesis of new proteins and enzymes can´t occur, leading to ultimately to severe illness (see deficiency and outcomes). The non-essential AA can be synthesized by the body itself from carbon and nitrogen building blocks, meaning those AA do not need to be present in the food in order to be formed, however they are of equal importance as the essential AA for metabolic processes (13). Those 10 essential AA are: Arginine, Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Tryptophan, Threonine, Valine (13). Non-essential AA include: Alanine, Asparagine, Aspartate, Cysteine, Glutamate, Glycine, Proline, Serine, Tyrosine, Taurine (13). The AAFCO established official minimal amount for the 10 essential amino acids for dogs *(see Table 1)*.

Table 1 *AAFCO Nutrient requirements for dogs 2014 (14)*

AAFCO Nutrient Requirements for Dogs (2014)			
Nutrient (% or per kg/diet	Growth and Reproduction Minimum	Adult Maintenance Minimum	Adult Maintenance Maximum
Protein (%)	22,0	18,0	N/A
Arginine (%)	0,62	0,51	N/A
Histidine (%)	0,22	0,18	N/A
Isoleucine (%)	0,45	0,37	N/A
Leucine (%)	0,72	0,59	N/A
Lysine (%)	0,77	0,63	N/A
Methionine + cystine (%)	0,53	0,43	N/A
Phenylalanine + tyrosine (%)	0,89	0,73	N/A
Tryptophan (%)	0,20	0,16	N/A
Threonine (%)	0,58	0,48	N/A
Valine (%)	0,48	0,39	N/A
Nutrient requirements indicated on a dry-matter basis per kg/diet. The AAFCO made this AAFCO nutrient profile for doog foods with a presume energy density of 3,5kcal ME/g dry matter.			

Table 2 *Representing the official recommended minimal nutrient requirements for dogs according to the FEDIAF (15) (The European Pet Food Industry Federation).*

FEDIAF Nutrient Requirements for Dogs (2019)				
Nutrient (Unit per 100g dry matter (DM))	Early Growth (<14 weeks) & Reproduction Minimum	Late Growth (≥14 weeks)	Adult Maintenance Minimum (95kcal/kg)	Adult Maintenance Maximum (110 kcal/kg)
Protein	25	20	21,0	18,0
Arginine	0,82	0,74	0,60	0,52
Histidine	0,39	0,25	0,27	0,23
Isoleucine	0,65	0,50	0,53	0,46
Leucine	1,29	0,80	0,95	0,82
Lysine	0,88	0,70	0,46	0,42
Methionine	0,35	0,26	0,46	0,40
Methionine + cystine	0,70	0,53	0,88	0,76
Phenylalanine	0,65	0,50	0,63	0,54
Phenylalanine + tyrosine	1,30	1,00	1,03	0,89
Threonine	0,81	0,64	0,60	0,52
Tryptophan	0,23	0,21	0,20	0,17
Valine	0,68	0,56	0,68	0,59
	Nutrient requirements indicated on a dry-matter basis per 100g/diet. And are recommended minimum values on an average daily energy intake of 95 kcal/kg or 110 kcal/kg. The FEDIAF calculated the values for adult dogs according the NRC (2006) recommendations, assuming a moderate-sized lean adult dog of 15kg bodyweight.			

1.1.1 Assessing Protein Quality:

There are two methods of assessing the protein quality for dogs, the “In vivo” and “In vitro” method. The In vivo method is expensive and time-consuming as the to be tested protein is being fed to animals and the response is being measured, such as: Nitrogen retention, weight gain, relative protein value, relative nutritive value, whole body nitrogen content (16). The in vitro technique is less expansive, it determines the amino acid profile which is then compared to a reference protein, normally being egg protein, the score is then a relative to the reference protein. The big issue with the in vitro method is that it can predicts the quality of the protein according to the amino acid profile but does not consider digestibility and effects of processing (16)

1.1.2 Protein levels in dog food

No matter if the source of the proteins are plants or animals, the minimal required protein amount need to be available in the dog food given, the AAFCO recommends a total protein content of 18% (14), it is unlikely to overfeed in proteins for dogs if the source is from animals or plants, but a amino acid toxicity can occur if fed synthetic sources, however high-protein diets should be avoided if renal or liver disease is suspected.

The research is clear, protein is an essential part for a healthy dog diet, if diet contains too few proteins, several clinical signs can be observed, such as: anemia, anorexia, reduced growth rate, loss of hair, infertility, decreased production of milk, poor appearing coat and unhealthy hair, lethargy, increased catabolism of muscle tissue and other proteins like blood proteins. Eventually leading to severe muscle atrophy, anemia and even fatty liver can occur (13).

1.1.3 Taurine

Taurine can be found as a free AA in several different tissues such as: retina, skeletal muscle, myocardium, liver, brain, milk, bile salts. Taurine assists in the absorption of ingested fat compounds (16). Another important function of taurine is in the nervous system, where it acts as a neurotransmitter and neuromodulator, being an important part of brain development, retinal function, heart function and regulation of the body temperature. Research suggests that taurine is also involved in cell volume regulation, osmolarity, stability of cell membranes and more (13). Unlike for dogs, for cat’s taurine is classified as an essential aa, due to several factors like taurine loss in feces and minimal ability of the cat’s organism to synthesize taurine (13). Taurine deficiencies: As previously discussed, taurine is an essential aa for cats, however for dogs, research does not proof it to be essential for dogs, but there are several scenarios in which it can become essential even for dogs, for example when feeding a high-fat containing feed at 24%DM, which caused Taurine levels to decrease in test subjects

and even reached slight deficiencies in some dogs (17). Low Taurine levels have been identified to be associated with dilated cardiomyopathy, and the dilated cardiomyopathy patients have shown low concentrations of taurine in the myocardial muscle tissue. (18).

1.1.4 Arginine

Arginine is of such importance that dogs consuming a meal lacking arginine develop rapid onset of clinical symptoms such as vomiting, increased salivation, hyperglycemia and tremors. Arginine is a crucial component in the urea cycle, therefore being a crucial component for neutralizing nitrogenous waste material like ammonia (19). Arginine is very abundant in most protein sources, this is the reason why the majority of pet food producer do not add arginine as a supplement. As described in the AAFCO nutrient requirements for dogs above the minimal required percentage of arginine in the food product should be 0,62% for growth (puppies) and reproduction, while 0,51% are found to be the minimal requirement for an adult dog during maintenance (14). These findings correspond to a study that found arginine levels of 0,4-0,56% of DMB supported the maximum weight gain (20).

1.1.5 Glutamine and Glutamate

Those two amino acids were classified as non-essential aa, research has proven that certain conditions can deplete those aa, however glutamate is still considered non-essential and glutamine is considered conditionally essential, implying that it is non-essential in healthy animals, but studies have shown that the organisms own synthesis and storage of glutamine might not be sufficient in certain conditions like severe infections, serious illness, chemotherapy, diarrhea and post cardiac surgery (21,22).

1.2 Carbohydrates

Carbohydrates, Proteins and fats are all part of the macronutrient category. Carbohydrates (composed of carbon, hydrogen and oxygen (23)) do count as a main source of energy and supply fiber that can be of benefit of health for the gastrointestinal system. The nutritional and functional capabilities are expressed in arrangements of the monomers, being alpha-type or beta-type. Therefore, we can group carbohydrates into mono, di, oligo and polysaccharides (23) (see fig. 1.)

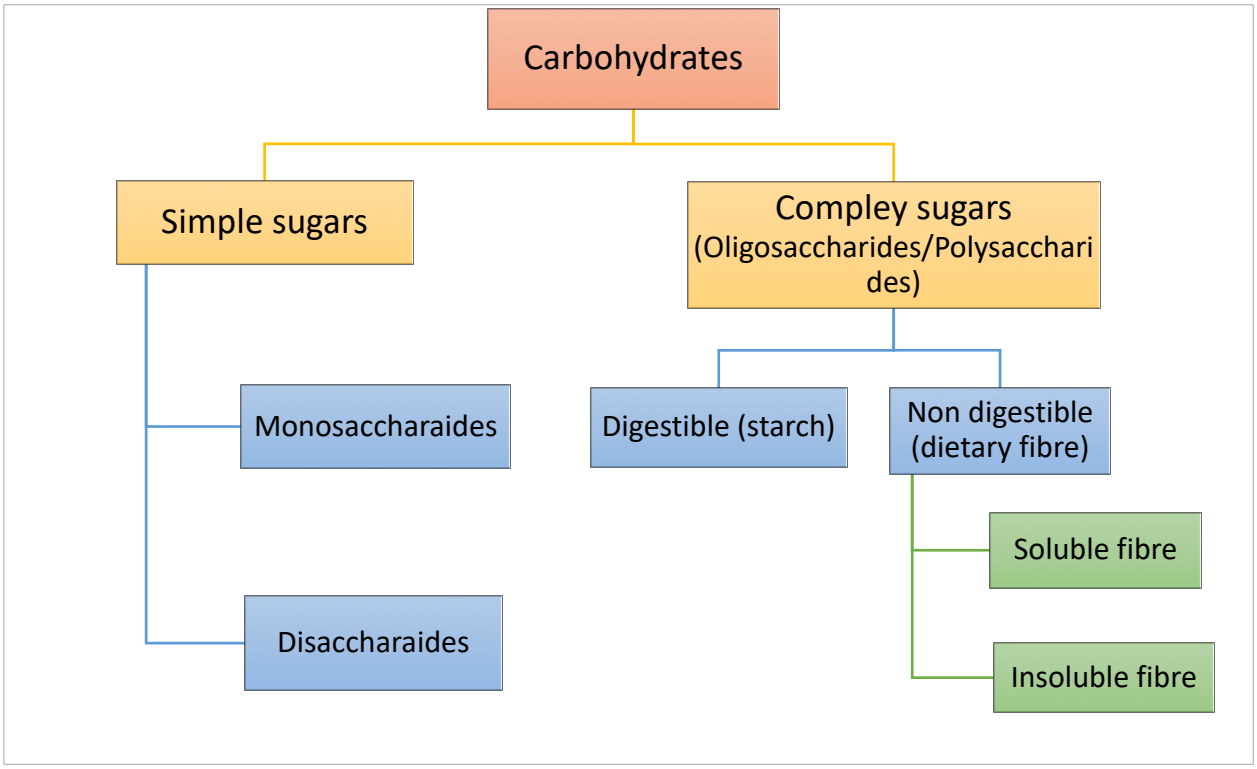

Fig. 1. *Classification of Carbohydrates*

1.2.1 Monosaccharides

Known as a simple sugar, representing carbohydrate in its simple form, examples are: Glucose, fructose and galactose (23).

1.2.2 Disaccharides

Representing the most available carbohydrates in nature. It´s structure is represented by two monosaccharides joined together to form sucrose, lactose and maltose. Sucrose is also known as table sugar, constituting of one glucose molecule joined to one fructose molecule. Lactose, also known as milk sugar, constitutes of one glucose molecule joined to one galactose molecule and maltose are two glucose molecules linked together (23).

1.2.3 Oligosaccharides

Also known oligomers, constituting of 3-9 monosaccharide molecules, mostly joined with beta-type bounds. Examples: Raffinose, stachyose (23).

1.2.4 Polysaccharides

Source of polysaccharides are plant materials and glycogen found in animal tissue, whereby the number of polysaccharides in animal tissue is by no means comparable to the prevalence of plant derived polysaccharides in nature. Plant source examples: Starches, Inulin, Gums, Mucilages, Plant

cell-wall polysaccharides (23). Starches are glucose molecules joined by alpha-type glyosidic bonds. Starch production generates an energy storage system for the plants. Inulin represents another form of energy storage in plants, mainly built from fructose molecules. Plant cell-wall polysaccharides, also known as non-starch polysaccharides are building blocks of the plant cell walls, examples: celluloses, hemicelluloses, beta-glucans, pectin's.

Animal source example: Glycogen, being the energy storage unit in animals, glucose monomers joined with alpha-type glyosidic bonds, mostly found in the liver and muscle tissue. (23; 24)

1.2.5 Digestion of carbohydrates in dogs

Digestion involves the mechanical breakdown of carbohydrate food source, enzymatic processes and microbial processes. Dogs do not produce alpha-amylase in their saliva, meaning digestion by enzymes do not start in the oral cavity of a dog, however new research has proven amylase production in dog's saliva (25). In the stomach little digestion of carbohydrates occur, therefore the real digestion and absorption of simple carbohydrates and starches happens in the small intestine. As several studies suggest, dogs do digest carbohydrates far better than wolves due to a drastic increase in copies of the gene that is coding for digestion of carbohydrates, produced in the pancreas, the AMY2B (26), which is the gene that made it possible for dogs to thrive and be healthy on a starch-rich diet (27,28). Dogs, when fed a diet containing 30-57% extruded barley, corn, oats and rice showed that to all starches from all sources were to almost 100% digested, meaning almost no starch passed from the small intestine into the colon (28). Other studies compared uncooked to cooked starch digestibility in dogs and showed than some starches like rice starch are digested in its raw and cooked form by almost the same degree, however other starches from potatoes were when given raw not digested at all. Therefore, strongly indicating the increase in digestibility of cooked foods over raw food sources (29), again showing that the dog is of omnivorous nature (30).

1.2.6 Absorption of carbohydrates

Absorption happens through active transport processes across the mucosa of the small intestine, if a carbohydrate malabsorption or intolerance is being observed, this can be due to a deficiency in needed enzymes or issues with the active transport processes, another reason for decreased absorption is when damage is being done to the mucosal lining of the intestine due to infections. Bacteria colonization can also cause destruction of amylase enzymes, therefore hindering the uptake of nutrients (23).

1.2.7 Sources of carbohydrates (23)

D-Glucose: Fruits, in most plant foods, maple sugar, honey; D-Fructose: Fruits, in most plant foods, maple sugar, honey; Pectins: Fruits; Sucrose: Beet sugar, fruits, cane sugar, maple sugar; Maltose: Sprouted grain, product of starch digestion; Amylose: Grains, starchy plants; Amylopectin: Grains, starchy plants, thickener in processing foods; Glycogen: Also known as the animal starch, found in muscle and liver; Lactose: Dairy products, milk; Cellulose: Cell walls of plants, wheat bran; Hemicellulose: Plant cell walls; Lignin: Plant cell walls; Carrageenan: Red seaweed, used for food processing; Raffinose, stachyose, verbacose: Plants protection, antifreeze substances; Dextrins, Corn syrup, high-fructose syrup: Used for food processing.

1.3 Fiber in the dog's diet

Fiber has been shown to decrease the time food needs to pass through the intestinal tract and to prolong the transition time in dogs with fast transition rates (31). Fiber has been shown to help in normal bowel function. Epithelial cells of the colon are shown to be in optimal function when fiber is administered to the diet, overall the whole gastrointestinal tract of dogs does perform at optimal levels on diets with fiber (32). Therapeutic management of some diseases require specific dietary fiber levels. Research in humans have shown that fiber can have positive effects on a variety of conditions such as: constipation, colorectal cancer, irritable bowel syndrome, Crohn´s disease and many more (23).

1.4 Important nutrients concerning a plant-based dog food

1.4.1 Folic acid (Water-soluble Vitamin)

Folic acids are a family of vitamers (having similar biological activity) (33). Folic acids are also known as folates or folacin. Important is the interplay between Vitamin B12 and folic acids on the production of methionine from homocysteine. Folic acids are involved in: Phospholipid synthesis, creatinine formation, metabolism of amino acids, production of neurotransmitter and nucleotide biosynthesis. Folic acid is metabolized by a hydrolysis process in the intestine, the enzyme gamma-glutamyl initiates this process, Folylmonoglutamate is being formed in the hydrolysis process, this form is then being absorbed into the organism through the epithelial cells of the intestine, therefore folylmonoglutamate is circling in the animal's system, after being absorbed by target cells folylmonoglutamate undergoes further enzymatic conversions. Folates are so important as no storage are available in the organism, IDEXX laboratories in Germany recommend folate levels of 9,3-23,8 ng/ml. Dificiencies can cause anorexia, megablastic anemia, leukopenia, poor weight gain, decreased immune function and glossitis, recommended tests to check suspected folate deficiencies is a blood test (34). Folate can be found in a variety of foods like green vegetables, egg yolks and liver. As

folates are sensitive to heating and processing, commercial pet foods supplement folates to counteract folate degradation of heating and processing.

1.4.2 Iron

Iron is a crucial micromineral that is a main player in a variety of metabolic functions and processes. The most known function of iron is the transport of oxygen in hemoglobin. Iron also serves as integral part of many enzymes like cytochromes which is needed for drug metabolism and the generation of energy. Most functional iron can be found in hemoglobin, myoglobin (oxygen transport) and cytochromes (electron transport) (35). There are two different forms of iron, heme iron and nonheme iron. Heme iron is the form of iron present in animal tissue in hemoglobin and myoglobin, while nonheme iron can be found in grains and plants. Dietary Iron is absorbed to a big part in the duodenum (36, 37). After iron enters across the enterocytes by ferroportin and being attached to transferrin in blood plasma to be transported (36, 37, 38). The dog's organism cannot efficiently excrete over excessive iron, homeostasis has mechanisms to assure steady iron levels by controlling iron uptake in the intestine (36, 37).

1.4.3 Vitamin B12

Vitamin B12, also known as cyanocobalamin is against the main believe in society, not produced by animals, but by a few bacteria and Archaea, therefore its production is caused by microbial fermentation (39), which is the method used for large scale industrial production. The main used bacteria to produce B12 are: Propionibacterium shermanii, Pseudomonas denitrificans and Sinorhizobium meliloti (40). Cyanocobalamin acts as a cofactor for metabolic processes such as the synthesis of nucleic acids and amino acid, citric acid cycle and functional conversation of epithelial cells (41). As animals do not produce Vitamin B12, it needs to be presented in sufficient quantities in the food sources. The absorption of cyanocobalamin is a complex process that can be disrupted due to a variety of gastrointestinal pathologies and therefore potentially cause hypocobalaminemia, such as exocrine pancreatic insufficiency, intestinal lymphoma or any other enteropathy leading to a chronic illness (42). Vitamin B12 deficiencies can lead to a variety of clinical signs like leukopenia, non-regenerative anemia, hyperammonaemia, hypoglycemia, neuropathies, anorexia, diarrhea, vomiting, failure to thrive (42). Most farmed animals are living in unnatural environments, exposed to insufficient Vitamin B12 producing bacteria's or archaea's, therefore most are given Vitamin B12 supplements, leading to usually sufficient B12 sources if fed with meat, which is the same reason why people consuming a vegan diet have to take Vitamin B12 supplements and non-vegans don't as they are indirectly supplementing through the supplemented animal product, however Vitamin B12 deficiencies are common in people and malabsorption is most commonly seen in elderly people (43).

A deficiency underlies 3 primary reasons: 1. Malabsorption through gastrointestinal pathologies, 2. Dietary insufficiencies, 3. Autoimmune disorders (44,45,46). In dogs especially, pancreatic pathologies and functional disorders of the Cubam-receptors are held accountable for B12 deficiencies (47). Another theoretical reason for B12 deficiencies is believed to be dysbiosis, caused by for example Clostridium spp or Bacteroides spp (48). Some dogs can present hereditary disorders of the cubam-receptors in the ileum (49, 50), if such a mutation is present in a dog, it is a autosomal recessive trait, which is shown as a severe deficiency of B12 already in young age of the dog, this condition is named after two scientists that discovered this mutation in humans, it is named the same in dogs "Imerslung-Gräsback-Syndrom (IGS) (51,52). In Chinese Shar Pei another hereditary B12 deficiency has been described, but it is associated with gastrointestinal disorders and it occurs in older age (53). Needed B12 for dogs is 1,27µg/kg bodyweight during growing and pregnancy; 0,47µg/kg bodyweight during maintenance (54). General official guidelines for parental and oral therapy for dogs do not exist yet, however recommendations are given.

2. METHODOLOGY

2.1 Study Design

This research was carried out over 15 months (June 2018–September 2019) in the Department of Animal Husbandry in the Veterinary Academy of the Lithuanian University of Health Sciences in Kaunas. Blood was collected from dogs that were fed a vegan diet (for least three months to a maximum of 10 years) to compare blood chemistry with recommended reference levels and with that of the meat-based food control group. The participants were required to be in good overall health or only have conditions that would not affect the following blood chemistry parameters: total protein, vitamin B12, folic acid, calcium, magnesium, iron, taurine and L-carnitine.

The vegan dogs were selected to represent a wide age range, from 10 months to 15+ years, to evaluate the adequacy of a vegan diet for almost all life stages. No puppies younger than 10 months were available during the recruitment period. Blood analyses were performed in six different official recognized laboratories that specialize in veterinary analytics (Laboklin, easyLAB, IDEXX, SYNLAB, ASAP Laboratory and AXIOM Veterinary laboratories) located in Germany, England and Australia. To evaluate the impact of a vegan diet under more controlled conditions, eight dogs were subject to a six-week feeding trial. Four dogs were fed a meat-based diet as a control and four dogs were fed a vegan diet. Blood was sampled prior to and at the end of the trial. The results were compared to official recognized healthy ranges for each blood chemistry parameter.

The final assessment was a questionnaire completed by 250 dog owners feeding a vegan or vegetarian diet and included a variety of topics including rationale for choosing a plant-based diet and changes observed during the diet.

Daily feeding ratios of some participants were analysed using the official licensed and registered veterinary feed calculator “FutterMedicus” under the supervision of Dr Uwe Romberger (an expert on vegan dog food and advisor on vegan dog food ratios).

2.2 Recruitment of participants for study

Finding dogs on a long-term vegan diet was a challenge itself. Participants were recruited via online platforms such as Facebook, word of mouth, a public survey and posts in several forums that were specifically designed for sharing information about plant-based dog nutrition (‘Vegan Dogs of Australia’; ‘Vegan Dog Nutrition-UK’; ‘Vegan Dog Nutrition’; ‘Vegan Hund!? Ja klar!’ and others.)

2.2.1 Requirement to qualify for the study

To be accepted as a participant, the dog had to be fed a 100% vegan diet for at least three months prior to the start of the study. Additionally, it was important to select individuals of different ages to better understand suitability of the diet for different stages of life (see table 3).

2.3 Laboratories used for analysis of blood samples

Blood samples were sent for analysis to one of six laboratories. In Germany, the laboratories included Laboklin *(*seven samples), easyLAB (two samples), IDEXX (37 samples) and SYNLAB (one sample); in Australia, ASAP LABORATORY (two samples) and in England, AXIOM VETERINARY LABORATORIES (two samples). The blood parameters included total protein, vitamin B12, folic acid, calcium, magnesium, iron, taurine and L-carnitine.

2.4 Average length of diet fed per group

The average length of feeding a vegan diet in category LT was 2.15 years. The average length of feeding was calculated by dividing the total time of vegan diet feeding in the long-term fed vegan diet (LT) category by the number of participants in that group (20). The result of 2.15 years (25.85 months) representing the average time dogs have been fed a vegan diet in category long-term. The control group was fed a conventional meat-based dog food for their entire life, meaning any healthy meat-based fed dog could have qualified as a participant of this group.

The vegan trial group was fed a diet for six weeks. The control group were conventional meat-based fed dogs that have received commercially available meat-based dog food for their entire life, meaning the diet of the control group participants did not change for the trial.

2.5 Collection of samples

Biochemical, serological and haematological tests were performed on the participants (n=48) in various cities around Germany (Rohrbach, Stuttgart, Gronau, Herne, Regensburg, Bedburg-Hau and others), England (Newton Abbot) and Australia (Melbourne) between the 04-02-2019 and 29-11-2019. Details on breed, sex, weight, date of analysis, laboratory and diet see Tables 3 and 4.

2.5.1 List of participants

Long-term vegan diet (LT)

Table 3 *Long-term vegan diet participant details*

Part. No.	Dog Owner	Dog Name	Breed	Sex	Dogs age	Dog´s weight (kg)	Date of blood analysis	Laboratory	On vegan diet for:
1.	I. Pfeilmeyer	Eli	Whippet	M.	3 yrs.	14	04.02.2019	IDEXX Laboratory	1 yr. 6 mos.
2.	I. Pfeilmeyer	Lewis	Whippet	M.	4 yrs.	15	04.02.2019	IDEXX Laboratory	1 yr. 6 mos.
3.	P. James	Archie	Golden Retriever	M.	10 mos.	25	10.10.2019	ASAP Laboratory	6 mos.
4.	Dr. U. Romberger	Rosine	Whippet	F.	4 yrs.	11,2	24.10.2019	IDEXX Laboratory Ludwigsburg	4 yrs. 8 mos.
5.	Dr. U. Romberger	Dori	Whippet	F.	1 yr.	13,8	24.10.2019	IDEXX Laboratory Ludwigsburg	11 mos.
6.	L. Scheffel	Sissi	Mixed	F.	9 yrs.	8,5	01.11.2019	IDEXX Laboratory Ludwigsburg	5 yrs.
7.	R. Kählert	Mei	Mixed	M.	10 yrs.	20	05.11.2019	SYNLAB Augsburg	6 yrs.
8.	V. Dickersbach	Emma	Mixed	F.	4 yrs.	20	06.11.2019	easyLAB	1 yr.
9.	V. Dickersbach	Summer	Australian Mini Shephard	F.	12 yrs.	11	06.112019	easyLAB	1 yr.
10.	L. May	Nyima	Collie	F.	2 yrs.	19	12.11.2019	IDEXX Laboratoy	1 yr.
11.	C. Burgdorf	Juri	Mixed	M.	13 yrs.	8,7	12.11.2019	IDEXX Laboratory	2 yrs.
12.	K. Sauer	Zombie	Cocker-Mixed	M.	11 yrs.	21	13.11.2019	IDEXX Laboratory Ludwigsburg	10 yrs.
13.	M. Brücker	Zolly	German Shephard Mix	M.	5 yrs.	16	15.11.2019	LABOKLIN	6 mos.
14.	M. Brücker	Benny	Dachshund Mix	M.	8,5 yrs.	10	15.11.2019	LABOKLIN	6 mos.
15.	M. Brücker	Jenny	German Shephard Mix	F.	5 yrs.	17	15.11.2019	LABOKLIN	6 mos.
16.	M. Brücker	Susi	Mixed	F.	7 yrs.	16	15.11.2019	LABOKLIN	6 mos.
17.	M. Knezevic	Brego	Husky	M.	6 yrs.	21	15.11.2019	LABOKLIN	3 mos.
18.	M. Knezevic	Mailo	Husky	M.	3 yrs.	21	15.11.2019	LABOKLIN	3 mos.
19.	M. Brücker	Amber	Shepherd-Mix	F.	4 yrs.	21	19.11.2019	LABOKLIN	6 mos.
20.	N. Stahlschmid	Bobby	Boston Terrier	M.	13 yrs.	12,5	19.07.2019	IDEXX Laboratory	5 yrs.

2.5.1.1 Control group for long-term fed vegan diet (LT)

The control group with equal numbers of dogs were collected from IDEXX laboratories in Germany that were subject to routine health checks and had no known pathologies. Participant selection was performed randomly and anonymously. The dogs were healthy and fed conventional diets (see Table 7).

2.5.2 List of participants in the vegan trial study category (VT)

Table 4 *Vegan trial test group participant details*

Part. No.	Dog Owner	Dog Name	Breed	Sex	Dogs age	Dog´s weight (kg)	Start of vegan diet	Laboratory	Vegan diet trial
1.	L.Kiemer	Sally	Labrador	F.	15 yrs.	34,5	15.10.2019	IDEXX Laboratory Ludwigsburg	6 Weeks
2.	M.Tannert	Alpha	American Bulldog	F.	3 yr.	41,6	22.10.2019	IDEXX Laboratory Ludwigsburg	6 weeks
3.	C. Tinkler	Maddie	Dachshund Miniature Wirehaired	F.	9 yrs.	4,4	16.10.2019	AXIOM Veterinary laboratories	6 weeks
4.	R. Mau	Mila	Mixed	F.	10 yrs.	18	05.10.2019	IDEXX Laboratory Ludwigsburg	6 Weeks

2.5.2.1 List of participants for vegan trial control group (VT):

Table 5 *Vegan trial control group participant details*

Part. No.	Dog Owner	Dog Name	Breed	Sex	Dogs age	Dog´s weight (kg)	Start of control group	Laboratory	Meat based diet
1.	C. Flemmer	Macy	Mixed	F.	8 yrs.	14,9	01.10.2019	IDEXX Laboratory	6 Weeks
2.	S. Braun	Sam	Cavalier King Charles Spaniel	M.	6 yr.	7,8	04.10.2019	IDEXX Laboratory Ludwigsburg	6 weeks
3.	A. Smith	Pearl	French Bulldog	F.	7 yrs.	11,3	14.10.2019	AXIOM Veterinary laboratories	6 weeks
4.	D. Lorenz	Jumper	Pembroke Welsh Corgi	M.	3 yrs.	12	13.10.2019	IDEXX Laboratory	6 weeks

2.6 Physical examination of participants

To confirm the health status of the participants and for better evaluation of the blood results, physical examinations were performed. The examination included assessment of body condition, general appearance, hydration status, lymph node condition, mucous membrane condition, skin and coat condition, cardiac and respiratory function and abdomen and oral cavity condition (see section 3.4).

2.7 Questionnaire data from 250 dog owners feeding a vegan or partially vegan diet

As information about vegan diets for dogs is scarce, additional information was collected by a questionnaire given to dog owners feeding a complete or partial vegan diet. The questionnaire was posted in forums and groups dedicated to pet owners feeding plant-based vegan diets. The questionnaire was presented to several thousand potential participants, of which 484 started and 250 completed the survey. Most of surveys were completed by computer or laptop, followed by mobile device and a small number were completed on a tablet.

2.8 Food ratio analysis

Several study participants were selected for direct evaluation of feeding ratios to determine if the diet contained sufficient amounts of all needed nutrients prior to feeding. Advice for this task was provided by an expert in plant-based dog nutrition, Dr. med. Vet. Uwe Romberger at the Tiergesundheitszentrum Regensburg. The analysis was performed with the licensed program FutterMedicus (for analysis of daily feeding ratios, see Annex 1).

2.9 Statistical Analyses

Data was analysed by the IBM SPSS Statistics program, v. 20, using the student-t and chi-square tests. The results were considered statistically significant at $p \leq 0.05$, and as not statistically significant when $p < 0.01$ (see table 9).

2.10 Research funding

For detailed information on research funding please see attached Annex 2. (55, 56)

3. RESULTS

3.1 Results of long-term vegan diet bloodwork analysis and comparison to official adequate reference levels

Table 6 shows the blood results of all 20 long-term vegan diet dogs. All values are compared to official recommended healthy ranges provided by each laboratory. A yellow arrow pointing upwards (↑) next to a value indicates that the result is higher than the recommended maximum, a red downward pointing arrow indicates the opposite, a result below the recommended minimum (↓).

The bloodwork results of long-term vegan diet dogs showed only two deficiencies, while the control (meat-fed) group experienced 11 deficiencies. The only two detected deficiencies in the 20 long-term vegan diet dogs were from two dogs belonging to the same owner, who reported they were diagnosed with giardia infection shortly after blood sampling, potentially explaining the low folic acid results. Two surpluses were detected in the long-term vegan fed group, both of which were the only samples analysed in SYNLAB. The same number of surpluses were detected in the control group.

Table 7 shows taurine and L-carnitine levels of the same category. Due to the high cost of taurine and L-carnitine testing, the study includes results of only three participants.

Table 6 *Bloodwork results of long-term vegan diet dogs*

Participant/Dog	**Total Protein (IDEXX)** 5,4-7,6 g/dl	**Iron (IDEXX)** 84-230 ug/dl ***Laboklin** 15-45 umol/l ****SYNLAB** 19,5-30,1 umol/l	**Vitamin B12 (IDEXX)** 234-812 pg/ml	**Folic Acid (IDEXX)** 9,3-23,8 ng/ml ***Laboklin** 3-10 ng/ml ****SYNLAB** 7,5-17,5 ng/ml ***Lab nmol/l (5,2-26,8)	**Calcium (IDEXX)** 2,1-2,9 mmol/l	**Magnesium (IDEXX)** 0,7-1,1 mmol/l
Dog 1 (C. Burgdorf/Juri)	6,7	230,8	566	11,2	2,3	1,0
Dog 2 (U. Romberger/dog 1)	5,9	177,9	474	10,6	2,7	0,9
Dog 3 (U. Romberger/dog2)	5,6	164,6	456	10,2	2,3	0,8
Dog 4 (K. Sauer/Zombie)	7,0	207,1	459	12,0	2,7	0,9
Dog 5 (L. Scheffel/Sissi)	6,8	182,1	364	16,8	2,4	1.0
Dog 6 (M. Brücker/Amber)	5,9	*31,3	607,3	*3,64	2,4	0,9
Dog 7 (M. Brücker/Benny)	5,9	*40,0	489,7	*5,43	2,5	1

Continuation of ***Table 6***

Dog 8 (M. Brücker/Brego)	6,2	*42,7	387,5	*4,79	2,6	0,9
Dog 9 (M. Brücker/Jenny)	6,2	*41,2	405,4	*5,02	2,6	0,9
Dog 10 (M. Brücker/Mailo)	5,9	*37,0	578,7	*3,22	2,7	0,9
Dog 11 (M. Brücker/Susi)	5,9	*24,2	477,2	*6,23	2,4	1,0
Dog 12 (M. Brücker/Zolly)	6,8	*34,1	679,2	*6,16	2,5	1,0
Dog 13 (R. Köhlert/Mei)	7,0	**56,2↑	503	**7,7	2,42	0,9
Dog 14 (L. May/Nyima)	6,2	**46,2↑	608	***19,2	2,68	0,8
Dog 15 (V. Dickersbach/Australian Mini)	6,0	123,1	427	6,4↓ (Giardia inf.)	2,4	0,9
Dog 16 (V. Dickersbach/Mischling)	6,4	202,5	420	5,8↓ (Giardi inf.)	2,2	0,9
Dog 17 (N. Sathlschmidt/Bobby)	6,2	-	-	-	2,82	-
Dog 18 (P. James/Archie)	6,0	-	-	-	2,68	-
Dog 19 (I. Pfeilmeyer/Eli)	5.8	-	-	-	2,6	0,7
Dog 20 (I. Pfeilmeyer/Lewis)	6.0	-	-	-	2,4	0,8

N/A*= Due to no financial support from the university or other organizations, each blood test was partially or fully paid for by the private owner of the dog, resulting in some parameters not measured to reduce costs.*

Table 7 *Taurine and L-carnitine results of long-term vegan diet dogs*

Participant/Dog	**Taurine** (44-224 umol/l)	**L-Carnitine** (16-42 umol/l)
Dog 4 (K. Sauer/Zombie)	111,86	-
Dog 5 (L. Scheffel/Sissi)	159,8	75,8↑
Dog 17 (N. Sathlschmidt/Bobby)	119,85	-

N/A*= Due to no financial support from the university or other organizations, each blood test was partially or fully paid for by the private owner of the dog, resulting in some parameters not measured to reduce costs.*

3.1.1 Results of control group for LT and comparison to official adequate reference levels

Table 8 shows the blood testing results of all 20 control group dogs. All values are compared to official recommended healthy ranges provided by the laboratories. The same indicator for surpluses and deficiencies are those used in Tables 6 and 7 (↑; ↓).

Table 8 *Bloodwork results of the LT control group*

Participant/Dog	**Total Protein (IDEXX)** 5,4-7,6 g/dl	**Iron (IDEXX)** 84-230 ug/dl	**Vitamin B12 (IDEXX)** 234-812 pg/ml	**Folic Acid (IDEXX)** 9,3-23,8 ng/ml	**Calcium (IDEXX)** 2,1-2,9 mmol/l	**Magnesium (IDEXX)** 0,7-1,1 mmol/l
Dog 1	5,8	116	440	8,9↓	2,6	1
Dog 2	6,6	78↓	301	8,8↓	2↓	0,9
Dog 3	7	201	270	9,4	2,7	1
Dog 4	5,9	198	188↓	12,5	2,8	0,9
Dog 5	5,6	153,6	310	14,1	2,4	0,8
Dog 6	6,6	177,4	198↓	14,3	2,2	1,1
Dog7	6,9	241,1↑	440	16,8	2,5	0,9
Dog 8	6,1	127,7	704	15,2	2,6	1
Dog 9	5,5	192,9	264	9,9	1,9↓	1
Dog 10	5,7	93	453	11,7	2,4	1,1
Dog 11	6,4	119,5	222↓	15,7	2,6	0,9i
Dog 12	6,8	231,7↑	341	13,1	2,5	0,8
Dog 13	5,9	155	353	12,6	2,3	0,8
Dog 14	7,1	148,6	312	16,4	2,7	1
Dog 15	5,2	188,2	707	10,2	2,4	1,1
Dog 16	6	202,1	266	11,1	2,5	0,7
Dog 17	5,9	116,4	206↓	7,9↓	2,7	0,7
Dog 18	6,1	99,3	506	9,1↓	2,8	1,1
Dog 19	6,9	198,7	389	12,1	2,1	1
Dog 20	6,9	210,4	654	14,6	2,4	0,8

3.1.2 Statistical analyses of LT category data

Statistical analyses of long-term vegan dogs and corresponding control group (see table 9) revealed statistically significant differences in mean concentrations of iron, vitamin B12 and folic acid ($p < 0.01$). No statistically significant differences were found for protein, calcium or magnesium ($p > 0.05$).

Table 9. *Statistical analyses of LT and corresponding control group*

Parameters		Sum of Squares	df	Mean Square	F	Sig.
Protein (5,4-7,6 g/dl)	Between Groups	,006	1	,006	,025	0,876
	Within Groups	9,561	38	,252	-	-
	Total	9,568	39	-	-	-
Iron (84-230 ug/dl)	Between Groups	15112,169	1	15112,169	6,646	**0,014**
	Within Groups	77315,180	34	2273,976	-	-
	Total	92427,349	35	-	-	-
Vitamin B12 (234-812 pg/ml)	Between Groups	123088,050	1	123088,050	6,761	**0,014**
	Within Groups	618970,910	34	18205,027	-	-
	Total	742058,960	35	-	-	-
Folic Acid Lab 1(9,3-23,8 ng/ml) Lab 2 (3-10ng/ml)	Between Groups	129,753	1	129,753	9,511	**0,004**
	Within Groups	463,856	34	13,643	-	-
	Total	593,609	35	-	-	-
Calcium (2,1-2,9 mmol(l)	Between Groups	,036	1	,036	,773	0,385
	Within Groups	1,769	38	,047	-	-
	Total	1,805	39	-	-	-
Magnesium (0,7-1,1 mmol/l)	Between Groups	,005	1	,005	,434	0,515
	Within Groups	,431	35	,012	-	-
	Total	,437	36	-	-	-

3.2 Results of VT bloodwork and comparison to official reference levels

Table 10 shows the blood testing results of the vegan diet trial dogs and comparison of those results to official recommended healthy ranges provided by the testing laboratories. The same indicators for surpluses and deficiencies are those used in Tables 6–8 (↑; ↓).

During the six-week vegan trial, most blood chemistry values were maintained, and several deficiencies detected before the trial were resolved. Deficiencies in folic acid, vitamin B12 and iron were detected in 2 out of 4 dogs (50%) prior to the start of the trial, when the dogs were still fed a commercial meat-based diet. Dog 1 was found to be deficient in vitamin B12 and iron before the trial with concentrations of 194 pg/ml and 69.1 ug/dl, respectively. At the end of the trial, dog 1 did not present any deficiencies and reached optimal levels of vitamin B12 and iron (350 pg/ml and 125.2 ug/dl, respectively). Dog 4 was found to be deficient in vitamin B12 and folic acid before the trial with concentrations of 186 pg/ml and 4.6 ng/dl, respectively. At the end of the trial, dog 4 did not present any deficiencies and reached optimal levels of vitamin B12 and folic acid (263 pg/ml and 10.4 mmol/l, respectively). Dog 3 began with folic acid levels in the optimal range of 24 mmol/l and ended the trial with a slight surplus of 36.9 mmol/l. Dog 2 maintained blood concentrations except for developing a deficiency in folic acid during the vegan trial. The owner of dog 2 explained that the

dog likely ingested something that led to diarrhoea 4 days before the end of the trial which resolved within 48 hours after onset.

Results of the physical examinations of the participants were normal.

Table 10 *Bloodwork results of dogs in the vegan diet trial*

	Before	After	Before	After	Before	After	Before	After	Before	After	Before	After
	Total Protein (5,4-7,6 g/dl)		**Vitamin B12 IDEXX** (234-812 pg/ml) ***AXIOM** (200-408 pmol/l)		**Folic Acid IDEXX** (9,3-23,8 ng/ml) ***AXIOM** (12-30nmol/l)		**Iron IDEXX** (84-230 ug/dl) ***AXIOM** (20-37 umol/l)		**L-Carnitine** (16-42 umol/l)		**Taurine** (44-224 umol/l) ***AXIOM** (5,1-12.1 mg/l)	
Dog 1 (L.Kiemer/ Sally)	7,4	6,9	194↓	350	15,1	9,8	69,1↓	125,2	-	51,2↑	-	215,7
Dog 2 (M. Tannert/ Alpha)	6,4	6,6	241	292	11,3	5,5↓	177,4	179,8	-	58,5↑	-	159,8
Dog 3 (C. Tinkler/ Maddie)	5,95	5,75	*525↑	*493↑	*24	*36,9↑	*20,7	*20,8	-	-	-	*24,8↑
Dog 4 (R. Mau/ Mila)	6,4	6,2	186↓	263	4,6↓	10,4	-	-	-	-	-	-

N/A*= Due to no financial support from the university or other organizations, each blood test was partially or fully paid for by the private owner of the dog, resulting in some parameters not being measured to reduce costs.*

3.2.1 Results of VT control group and comparison to official adequate reference levels

Table 11 shows the blood results of the VT control group, with all values compared to official recommended healthy ranges provided by the testing laboratories. The same indicator for surpluses and deficiencies are those used in Tables 6–8 and 10. (↑; ↓).

Table 11 *Bloodwork results of VT control group*

	Before	After	Before	After	Before	After	Before	After	Before	After	Before	After
	Total Protein (5,4-7,6 g/dl)		**Vitamin B12 IDEXX** (234-812 pg/ml) ***AXIOM** (200-408 pmol/l)		**Folic Acid IDEXX** (9,3-23,8 ng/ml) ***AXIOM** (12-30nmol/l)		**Iron IDEXX** (84-230 ug/dl) ***AXIOM** (20-37 umol/l)		**L-Carnitine** (16-42 umol/l)		**Taurine** (44-224 umol/l) ***AXIOM** (5,1-12.1 mg/l)	
Dog 1 (C. Flemmer/ Macy)	6,7	6,3	218	383	8,9↓	8,7↓	144,3	181,8	36,4	39,1	-	-
Dog 2 (S. Braun / Sam)	6,5	6,7	302	344	13,1	14,8	165,1	149,6	-	33	-	186,9
Dog 3 (A. Smith / Pearl)	6,0	5,9	245	212↓	15,2	13,5	155,5	137,3	-	-	-	-
Dog 4 (D. Lorenz / Jumper)	6,3	6,9	501	468	9,2↓	11,1	182	164,7	-	-	-	-

3.2.2 Statistical analyses of VT category and its control group

The statistical analyses of the vegan trial group and its corresponding control showed no statistically significant differences between the tested parameters ($p > 0.05$). Proofing that a plant-based diet did show no inferior or superior results over a conventional meat-based diet during the 6-week feeding trial. Further strengthening the plausibility of feeding a purely plant-based diet to dogs (see table 12-13).

Table 12 *Statistical analyses of vegan trial group and its corresponding control*

Paired Samples Test									
Vegan vs. meat fed trial group		Paired Differences					**t**	**df**	**Sig.** (2-tailed)
		Mean	**Std. Deviation**	**Std. Error Mean**	**95% Confidence Interval of the Difference**				
					Lower	Upper			
Pair 1	Before trial: **Total Protein**	,16250	,36827	,18414	-,42350	,74850	,882	3	,442
Pair 2	Before trial: **Iron**	-31,45000	61,87184	43,75000	-587,34646	524,44646	-,719	1	,603
Pair 3	Before trial: **Vitamin B12**	-133,33333	158,41191	91,45916	-526,85033	260,18367	-1,458	2	,282
Pair 4	Before trial: **Folic Acid**	-,06667	5,60476	3,23591	-13,98966	13,85633	-,021	2	,985
Pair 6	After trial: **Total Protein**	-,08750	,53288	,26644	-,93543	,76043	-,328	3	,764
Pair 7	After trial: **Iron**	-13,20000	61,37687	43,40000	-564,64929	538,24929	-,304	1	,812
Pair 8	After trial: **Vitamin B12**	-96,66667	94,29917	54,44365	-330,91879	137,58546	-1,776	2	,218
Pair 9	After trial: **Folic Acid**	-2,96667	5,55818	3,20902	-16,77394	10,84061	-,924	2	,453

Table 13 *Statistical analyses of only vegan group*

Paired Samples Test									
Vegan fed trial group (Before trial vs. After trial)		**Paired Differences**					**t**	**df**	**Sig.** (2-tailed)
		Mean	**Std. Deviation**	**Std. Error Mean**	**95% Confidence Interval of the Difference**				
					Lower	Upper			
Pair 1	**Total Protein**	,17500	,28723	,14361	-,28204	,63204	1,219	3	,310
Pair 2	**Iron**	-29,25000	37,97163	26,85000	-370,41160	311,91160	-1,089	1	,473
Pair 3	**Vitamin B12**	-94,66667	54,68394	31,57179	-230,50911	41,17577	-2,998	2	,096
Pair 4	**Folic Acid**	1,76667	6,55769	3,78609	-14,52355	18,05688	,467	2	,687

3.3 Physical examination of participants from long-term vegan diet and vegan trial groups

General observations prior to handling: One participant in the long-term vegan diet group did show a slight gait and postural abnormalities due to a cervical intervertebral disc protrusion. The remaining 23 dogs had symmetrical bodies and did show no abnormalities in gait or posture. **Body condition:** All participants were assessed according the Purina Small Animal Body Condition Scoring System. All participants of the long-term vegan diet group presented ideal body conditions (4–5). In the vegan trial group, one dog scored a "6", placing the dog in the slightly overweight category. **General appearance:** All dogs participating in the study showed normal behaviour and were responsive and alert, while two participants were described as fearful. There were no signs of depression or stupor. **Hydration status:** All study participants showed adequate hydration (0-5%), and skin immediately returned to normal position after tenting. **Lymph nodes:** No abnormalities for submandibular, prescapular, axillary, inguinal or popliteal lymph nodes. Lymph nodes were of normal size, shape, symmetry and firmness, and were freely movable. **Mucous membranes (MM):** All participants presented pink mucous membranes, indicating adequate perfusion and oxygenation of peripheral tissues. **Skin and coat:** 22 out of 24 dogs showed a healthy skin and coat without abnormalities. One participant in the "long-term vegan diet group" reported that her whippet had common breed-related alopecia (partial or complete hair loss) on her thighs which appeared during winter periods, and indicated that the alopecia was expressed long before switching to a vegan diet. One owner in the VT group reported that her dog had minor alopecia around her nose, which recently had regrown. **Cardiac System:** 23 out of 24 dogs showed no cardiac abnormalities. 1 participant of the VT group was diagnosed with congenital sub-aortic stenosis prior to the trial**. Respiratory System:** 23 out of 24 participants presented a clinically healthy respiratory system; no abnormalities were found. One participant from the long-term vegan diet group was diagnosed with arteriovenous fistulas with hypotension and was being treated for this condition prior to the start of the study and the illness was well under control. **Palpation of Abdomen / Evaluation of the Digestive tract:** The physical examination and palpation of the abdomen showed no pain or uncomfortable response in any participant. **Oral cavity:** 83.4% presented a healthy oral cavity and healthy teeth, while 16.7% had slight to moderate dental calculus. Not one participant was described as having foetor ex ore (bad breath).

Defecation frequency: 54.2% reported a defecation frequency of 2 times/day, 8.3% reported a frequency of 2–3 times/day, 20.8% reported 3 times/day and 16.7% reported >3 times/day (see Fig. 3).

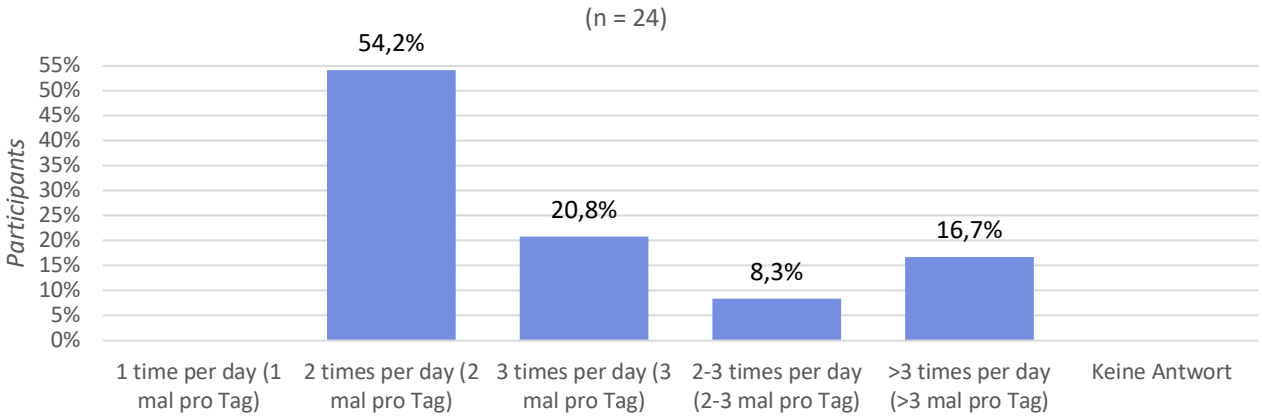

Fig. 3. *Defecation frequency of LT and VT dogs*

Stool consistency: All participants reported normal and healthy appearing stool consistencies; 70.8% described it as "sausage-shaped with cracks on the surface" and 45.8% as "smooth and soft sausage-like". Not one participant reported a mushy consistency or separate hard lumps or any other consistency abnormalities. **Stool colour:** Stool colour was reported to be primarily brown, from light/chocolate/red to dark brown, therefore representing normal colour variations. However, 12.5% reported a green stool colour, which normally serves as an indicator of increased ingestion of grass or a potential parasitic infestation. In the case of feeding a fully plant-based diet and the resulting increased amount of vegetables consumed, this can be easily explained as causal factor (see Fig. 4).

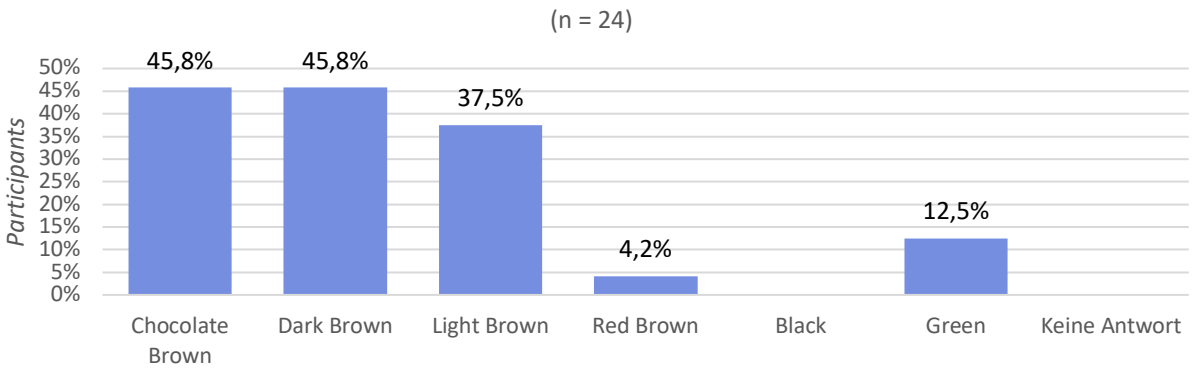

Fig. 4. *Color of stool of LT and VT dogs*

Volume of stool after switching to a vegan diet: 41.7% of all participants observed an increase in stool volume, 33.3% reported a maintained volume, 0% reported a decrease in stool volume and 25% did not know if the stool volume had changed (see Fig. 5).

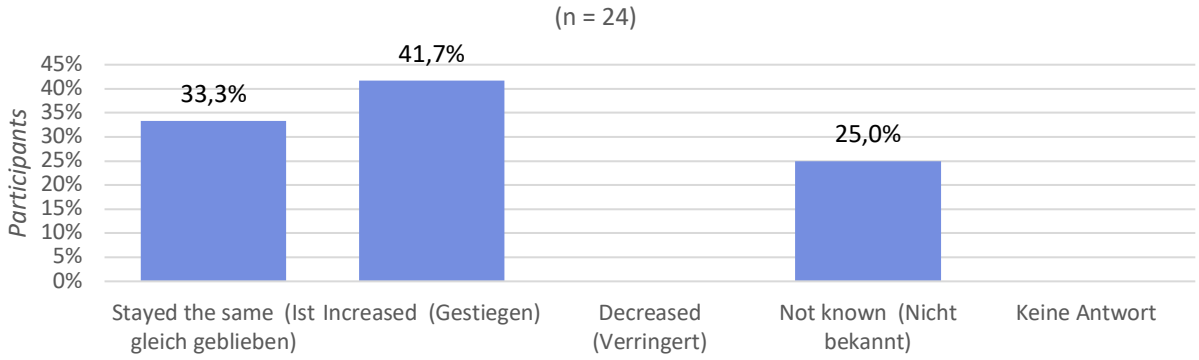

Fig. 5. *Stool volume changes after switching to vegan diet in LT and VT dogs*

Overall health observations after changing to a vegan diet: 66.7% reported maintained health status from prior to the start of feeding a vegan diet to being fed a vegan diet; 29.2% observed an increase in the dog's overall health, 4.2% did not know how to respond to the question, and 0% of the participants reported a decrease in health (see fig. 6).

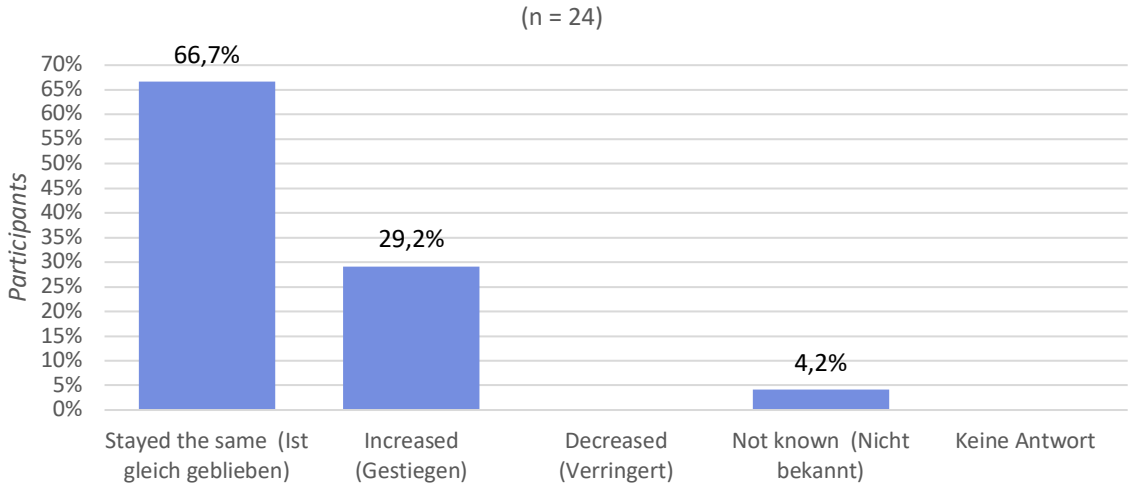

Fig. 6. *Overall health of LT and VT dogs*

3.3.1 Physical examination of participants from LT control group and VT control group

Both control groups were in overall good health with very similar findings as described for test groups in section 3.3.1; the exact physical examination data is available upon requested.

3.4. Results and analysis of collective data of 250 dog owners feeding a plant-based

Table 14 *Summery of questionnaire presented to dog owners feeding a plant-based diet to their dogs*

Survey title	Master Arbeit/Final Thesis in Veterinary Medicine (Questionnaire/Umfrage)
Date of report summary	Friday, 08. November 2019 19:24
Total surveys started	484
Unfinished surveys	234
Completed surveys	**250**
Completion percentage	**51.7%**

Information about participants

Most participants were between 31-40 years of age (26,8%), followed by the 26-30 year group (22,8%), the rest similarly distributed in the age groups 19-25; 41-50; 51 or older and only one participant was younger than 18 years. Surprisingly, the female fraction outnumbered all other genders by 90,4%, male participants represented by 7,8% and 4 participants (1,6%) responded with diverse *(see fig. 7.)*.

94,8% responded in consuming a vegan diet themselves, 3,2% a vegetarian diet and only 4 participants (1,6%) consuming an omnivorous diet *(see fig. 8.)*.

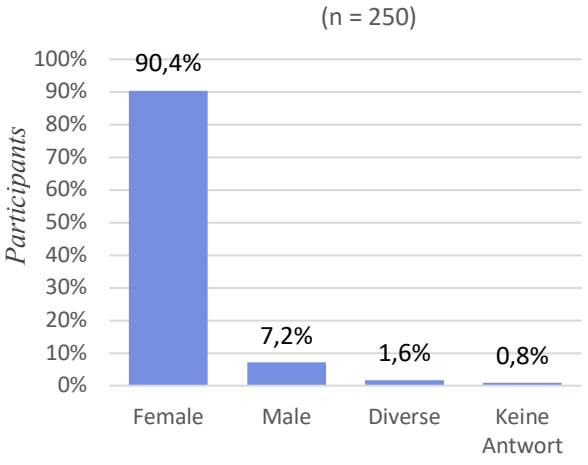

Fig. 7. *Sex of participants*

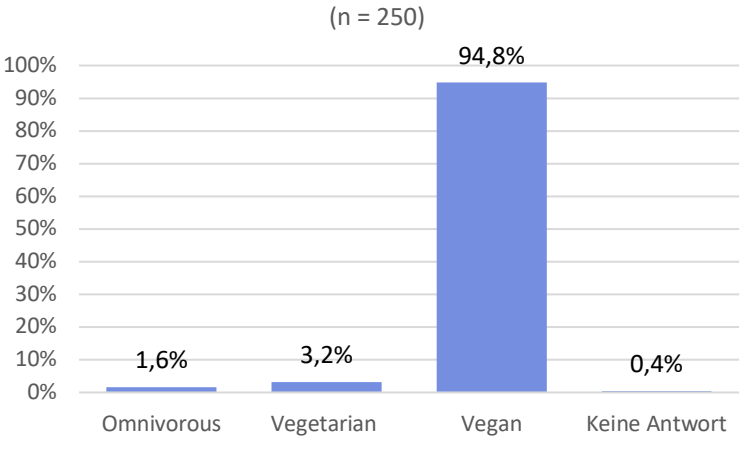

Fig. 8. *Diet of participants*

Reason for own diet choice

The reasoning behind the diet choice could be indicated from 0 (not applicable) to 100 (very accurate) according to pre-given statements *(see table 15)*. The result was very clear: 97,76-point average describing their diet choice as an ethical and moral obligation. Followed by a 96,41-point agreement on concerns towards animal welfare, treatment and living condition of farmed animals. 91,45-point average was given on the increased awareness on the environmental impact that animal products have compared to plant-based alternatives. 234 participants found it with an 83,38-point average healthier to not include animal products in their diets. A small number with a 28,38 points average reasoned their vegan/vegetarian diet choice as a result from a doctor's recommendation. The lowest numbers were represented with a point score average of 11,14 on following the current trend in becoming vegetarian/vegan and 6,49 by not having a real reason.

Additionally, 45 participants answered with a 46,67-point average to have "other reasons", whereby the most indicated were again regarding ethical issues, animal welfare and own health followed by the environmental issues. Some also indicated human rights issues due to the increased environmental burden and resource usage from animal products compared to plant-products and the human issues arising from it like "land grapping and climate justice".

The results clearly indicate that the majority of vegans/vegetarians have clear defined motifs behind their diet choices, mainly being driven by well-reasoned environmental and ethical issues.

Table 15 *Reason for own diet choice by questionnaire participants*

	Answers	No answer	Min.	Max.	Ø	M	Variance	Standard-deviation
I believe that we have the ethical and moral obligation to not harm other beings if there are cruelty free alternatives like a vegan diet compared to a diet that includes animal products.	245	5	0	100	97.76	100	117.48	10.84
I find it to be healthier not including animal products in my diet.	234	16	0	100	83.38	100	609.58	24.69
I have health issues and my doctor recommended me a vegan/vegetarian diet, which eased my symptoms.	105	145	0	100	28.38	10	1102.16	33.20
I do not agree with the treatment and living condition of farmed animals.	237	13	0	100	96.41	100	221.40	14.88
I am aware of the increased environmental impact that animal products have compared to plant-based alternatives.	235	15	0	100	91.45	100	324.39	18.01
I don´t have a real reason for my diet choice.	74	176	0	100	6.49	0	346.39	18.61
I wanted to give it a try as it is very hyped at the moment.	79	171	0	100	11.14	0	594.84	24.39
Other reason (please specify in textbox)	45	410	0	100	46.67	30	2077.27	45.58

M=Median Ø=Average

Information about participating dogs:

33,2% of the 250 dogs were in the age range of 2-5 years, followed by the 5-10 years group (27,6%); the 10-15 years group (19,6%) and the 1-2 year group (11,6%). The smallest two groups were also the oldest and youngest group, 0-1 year group (5,6%) and older than 15 years group (2,4%).

The dogs were put into weight categories *(see fig. 9.)*.

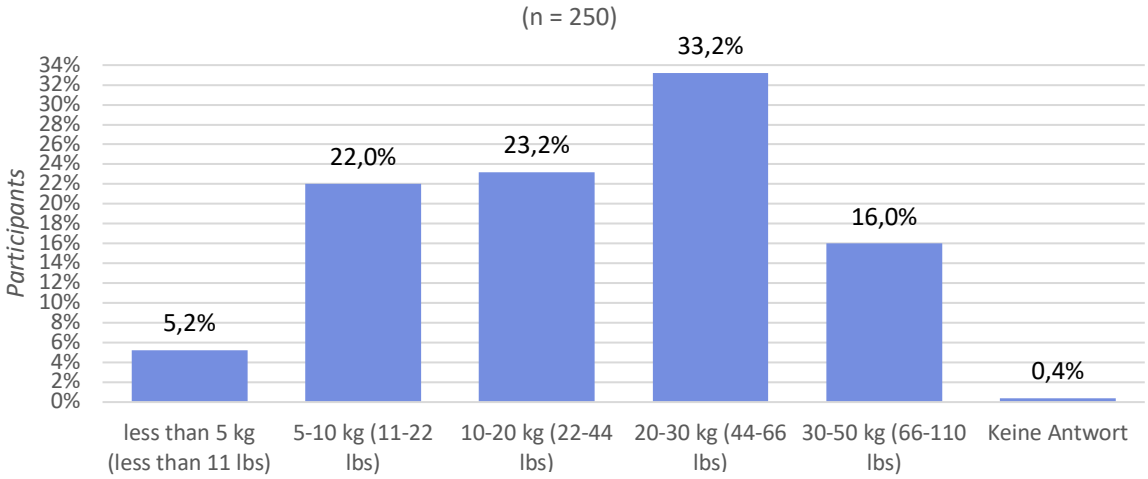

Fig. 9. *Weight of dogs*

53,6% of the participants listed their dog as "Single housed animal", meaning living with one dog only, while 46,4% specified "Group housed animal", living with more than one dog.

More than 70 different dog breeds contributed to the study. There was not really a significant portion for breed over the other, however mixed breeds were most prevalent *(see fig. 10.)*.

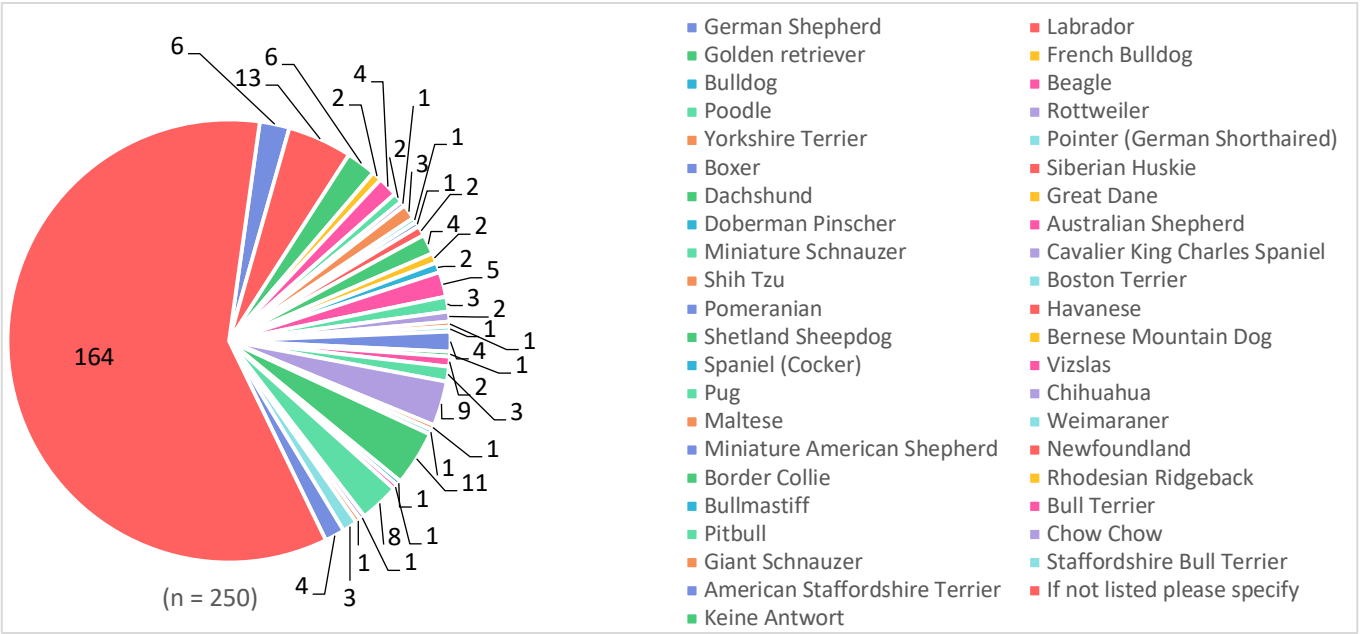

Fig. 10. *Dog breeds participating in survey*

Purpose of dogs

94,8% reported to own a dog for leisure (family dog) while 3,2% use their dogs for sport (include but are not limited to agility trails, hunting and racing). 1,6% use their dog for breeding purposes.

Information about health status of dogs

The majority of dogs (76,8%) report that the dog has no known disease while 23,2% do have a known disease. Most, if not all diseases clearly weren´t related to nutrition but to a pathogen, age-related changes or due to congenital diseases. Congenital diseases included: hip-dysplasia, blindness, deafness, patent ductus arteriosus, elbow dysplasia, stiffening of the spine since puppy age, sub aortic stenosis or brachiocephalic syndrome. Leishmania was listed in 3 participants, arthritis in 3 participants, Ehrlichiosis in 1 participant. Cancer was reported by 3 participants. Interestingly 3,6% reported "allergies" as disease, which most marked as improved or solved after switching to a vegan diet. The majority with 87,2% reported no use of permanent medication, while 11.2% reported to use permanent medication and 1,6% didn´t provide any answer to this question. Most frequent permanent medication included anti-inflammatory and pain relief medicaments like: Rymadil and Phenylbutazone. Other medicaments were used such as: Caniphedrin (treatment incontinence); Canitroid, Levothyroxine, Forthyron (treatment hypothyrpoidism); Gabapentin (pain relief due to old racing injury) Tramadol (opiod pain killer); Allopurinol, Allopurinol (prevention of kidney stones and lowering pH of urine); Prednisolone (Crohn´s disease); Clinadry, Optimune (eye drops); Some mentioned homeopathic treatments like "Zeel"; Digestive enzymes; Apoquel, piriton (allergy medicine); Proin (incontinence); Atenolol (beta blocker, cardiac arrhythmias, hypertension).

Alimentation of dogs

84,8% of all participants reported to feed their dogs an exclusively Vegan diet (100% plant based), while 9,6% reported to switch between a fully vegan and non-vegan diet as they reported to be "not convinced yet" that a purely vegan diet would fulfill all nutritional requirements. The rest were similarly distributed between an Ovo-lacto-vegetarian diet (plant-based diet including eggs and dairy) 2,4%, Ovo-Vegetarian diet (plant-based diet including eggs but not dairy) 1,6%, Lacto-Vegetarian diet (Plant-based diet including dairy but not eggs) 0,4% and commercial dog food (including meat, eggs and dairy) was fed by only 1,2% of surveyed dog owners. When asked how the participants became aware of vegan dog food, 76,4% reported that the internet was the main source for information about this topic. 7,2% reported their friends or colleagues as source of information. 3,6% reported their veterinarian as a source about vegan dog food and 2,8% reported professional journals as their source *(see fig. 11.)*.

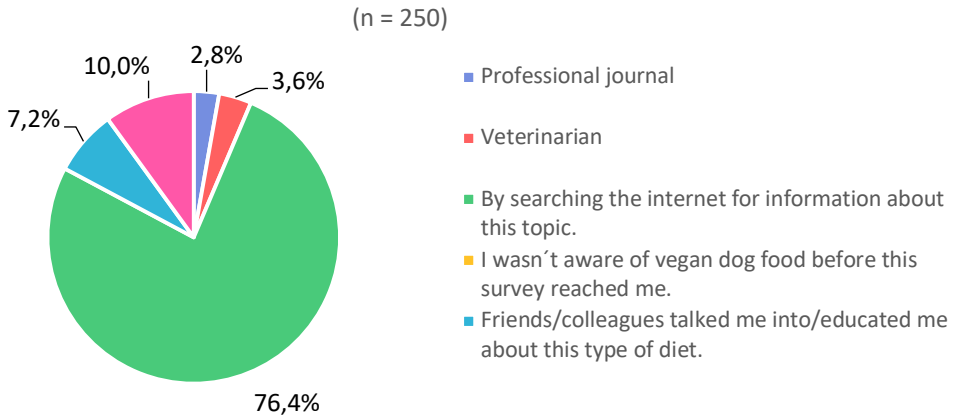

Fig. 11. *How participants became first aware about vegan dog food*

10% reported other sources that made them aware of vegan dog food, most of them also indicated sources from the internet and social media like Facebook groups and Instagram. One person reported to have seen the company VegDog on TV which made her aware of vegan dog food. Another person reported to have met a dog owner that made her aware of the possibility to aliment a dog vegan. Another person reported as he was vegan himself that there was no question to extend the ethical consideration to his dogs' bowl. Some reported that they have tried a vegan diet as a last possibility to resolve skin and digestive issues, most likely caused by allergic reactions, which many reported having resolved after switching to a fully plant-based diet. Another person reported to have gotten aware about vegan dog food through his wholefood shop supplier and another one through a vegan summer festival.

Interestingly, most dog owners (30%) started to feed their dogs a vegan diet at the age of 1 year. Overall the start of a vegan diet is distributed through all stages of life, while 96% started to feed their dogs a vegan diet before the dog reached an age of 15 years *(see fig. 12.)*.

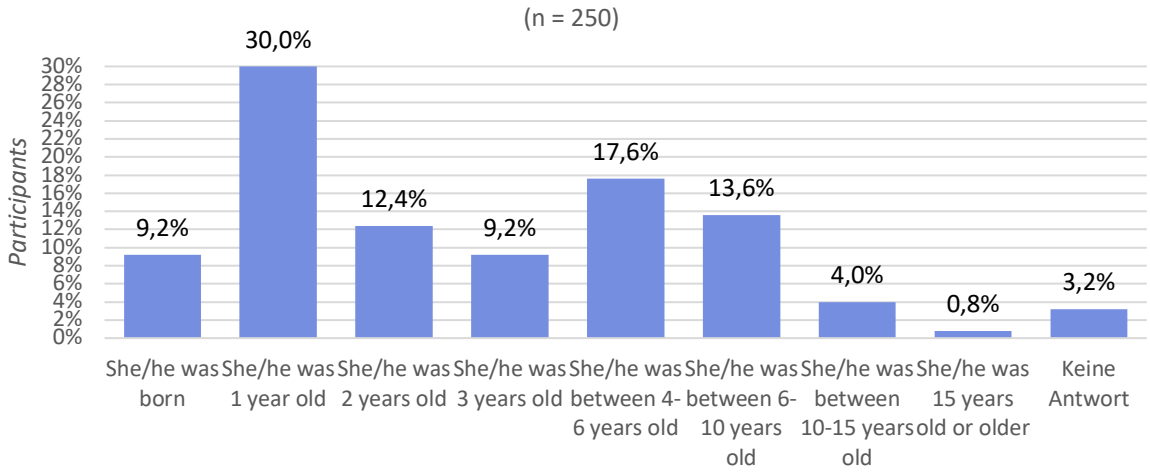

Fig. 12. *Age of dog, when vegan diet was first introduced*

Motifs for feeding a vegan/plant-based diet

Very clear responses by all participants, 91,2% reported to feel ethically and morally obliged to avoid being the cause of suffering for other beings such as farmed animals when there is a cruelty free alternative which is proven to be healthy for a pet will always be the correct moral choice to feed this type of diet. Followed by 66% that reported to know research that proofs that dogs can thrive and be healthy on a well-balanced vegan diet; 60% believe a vegan/plant-based diet is healthier for their dogs; 51.6% were not satisfied with the quality of commercial dog food and the use of animal by-products which are unfit for human consumption and that they don´t want to feed their dogs a product that contains the so-called "4D" livestock animals (dead, dying, diseased, disabled); Followed by 49,6% which believe many health issues such as increased cancer rates and chronic diseases arise due to poor quality feed and processing practices of commercial dog food industries. While agreeing that we should apply the same skeptical thinking that most have towards vegan dog food to commercial dog food as well, therefore giving the urge to rethink what we consider normal as feeding practices for our beloved companion animals; 42,4% reported to be against the practice of rendering (Industrial process that converts waste animal tissue into usable materials); 16,4% reported that the dog had allergic issues and a vegan diet resolved the issues, while 2,8% got a vegan diet recommended due to health issues and the equal amount of 2,8% were recommended a vegan diet form their veterinarian. 2% reported to have been talked into a vegan diet for dogs by friends/colleagues, another 2 % didn´t provide an answer and 0,8% are trying a vegan diet for their dogs out of curiosity.

Information about fed diet

Most participants (57,2%) were feeding a mixture of commercially available vegan dog food and homemade vegan dog food, followed by feeding solely commercial vegan dog food (38%). A surprisingly high number of 41 participants (16,4%) were feeding homemade vegan dog food. 4,4% choose non-vegetarian commercial dog food for their pet (including meat, dairy and other animal products). 3.6% reported feeding commercial vegetarian dog food and 2 participants didn´t provide an answer. 9 participants (3,6%) provided additional information about their feeding behavior, while 4 of them fit right into the homemade vegan dog food group (one of them specified to use cold-pressed vegan dog food); 2 of those 9 participants fit into the non-vegetarian group; 1 of those 9 into the vegetarian group and 1 of those 9 reported to mix commercial vegan and non-vegan dog food which would classify as feeding commercial dog food (including meat, dairy and other animal products) *(see fig. 13.)*.

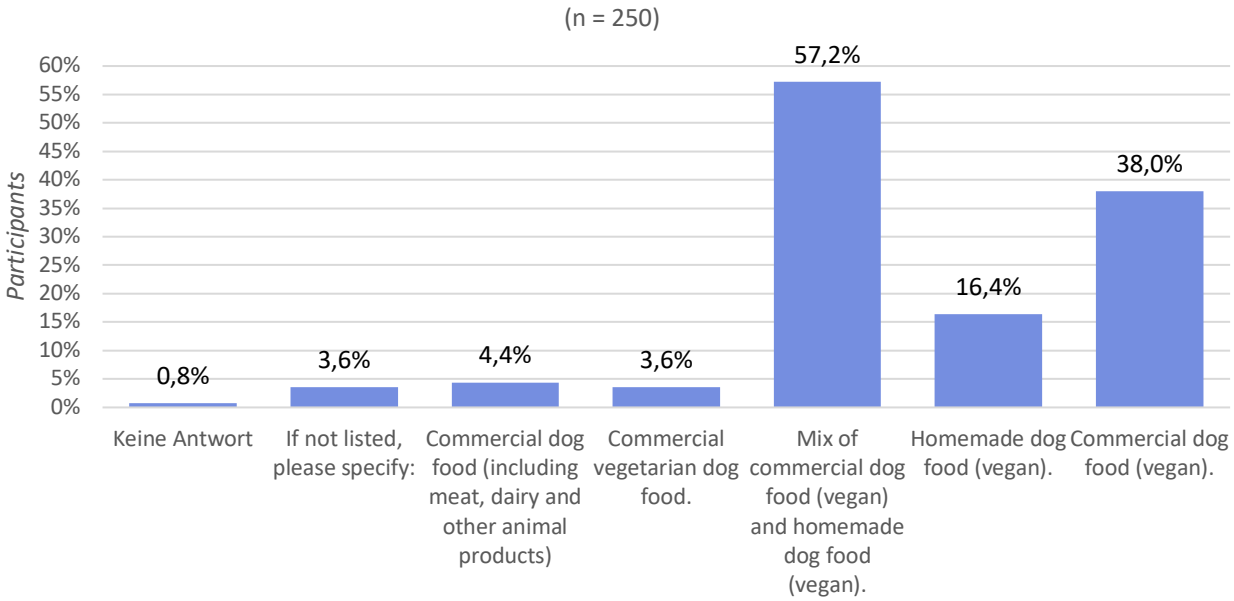

Fig. 13. *Type of diet fed by participants of survey*

Most used commercial brands by participants

The following brands were listed when the participants were asked to provide the brands they purchase their commercial vegan dog food from *(numbers representing quantities of responses):*

71: Vegdog (Munich, Germany)
50: Green Petfood (Veggiedog)
47: Benevo (U.K. based)
42: V-dog (USA)
51: Vegan4dogs/ Greta (Berlin, Germany)
24: Ami (Italian company)
21: No Answer given
19: Yarrah (organic bio)
13: Natural Balance (USA)
9: Halo (USA)
8: Biopet Vegan
7: Lukullus
6: Vegan Pet
4: Nature´s recipe (USA)
3: V-Dog (V-Planet)
2: Wild Earth
2: Prime100 (Pea and hemp roll)
2: Pitti Boris
2: Royal Canine (United States, France, South African, Brazil
1: Nature´s Recipe
1: Vegusto
1: Napani
1: veggieanimals
1: Chi Dog
1: Gather Endless Valley
1: Naftie

Most used ingredients for homemade food

The following column chart represents the percentage of participants using different types of ingredients for home preparation of vegan/vegetarian food portions *(see fig. 14.)*.

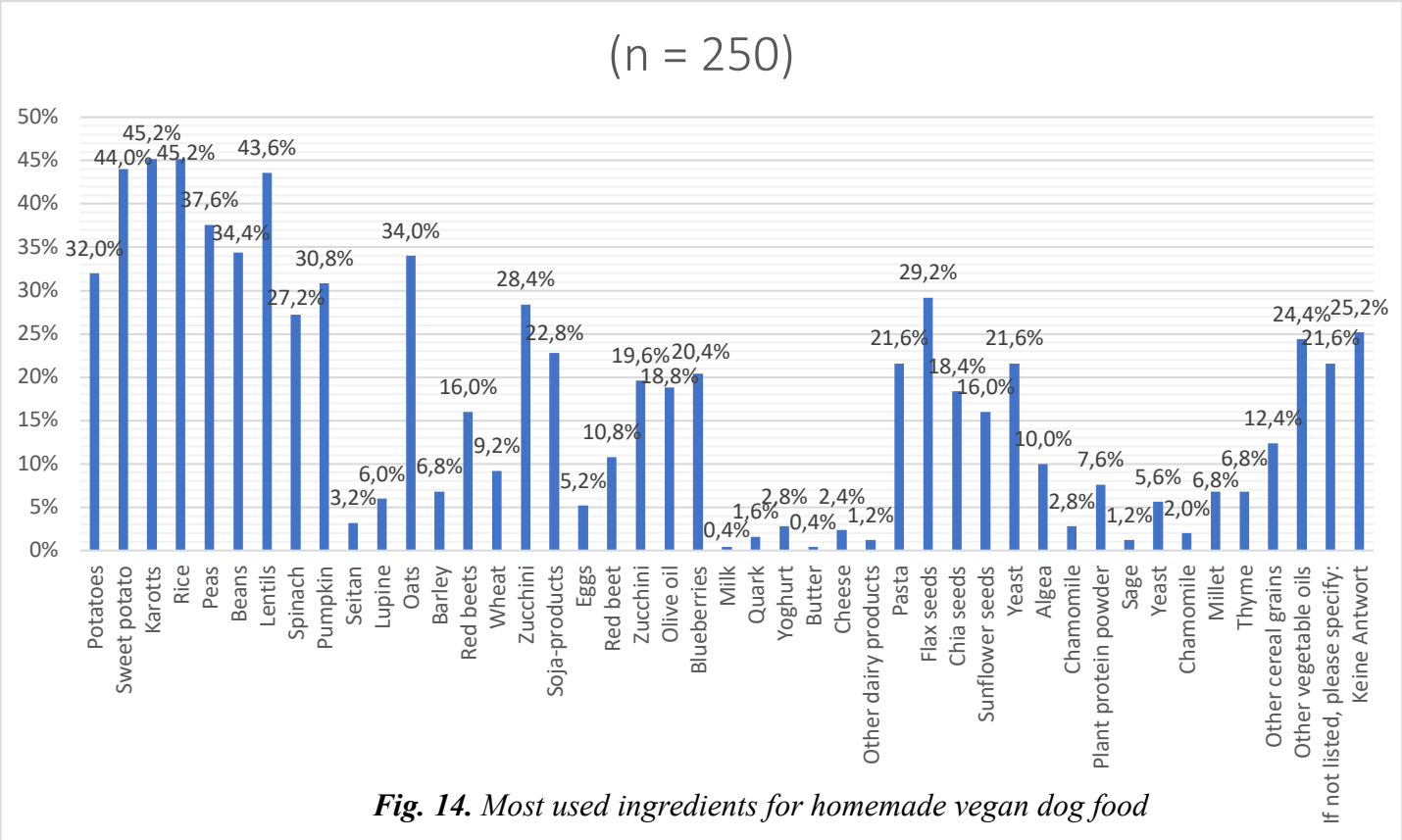

Fig. 14. *Most used ingredients for homemade vegan dog food*

21, 6% specified more ingredients which included: Broccoli, Coconut cream, peanut butter, chickpeas, quinoa, bananas, apples, buckwheat, cauliflower, tofu, Tempe, pasta, hemp seeds, hemp protein, nutritional yeast, tomato, coconut milk, eggplant, peppers, parsnip, herbs, cranberries, brussels sprouts, blueberries, soy milk, wholegrain pasta, cucumber, caraway seeds, savory, flaxseed oil, Coconut oil, coconut-hemp oil, hemp oil, seaweed powder, kale, stinging nettle, green cabbage, celeriac, strawberries, corn, flaxseed-, Cocos-flakes, yeast-flakes, oat-milk, wakame, shiitake mushrooms, celery, turmeric, polenta, green beans, split peas, pumpkin seeds, almonds, brazil nuts, walnuts, ginger salad, pear, seaweed, capsicum, tahini, kaki, artichoke, yeast extract (marmite).

Grain-free or not Grain-free

34,4% of the surveyed dog owners didn´t know of any difference between grain-free and grain containing diets. However, 40% reported to feed a diet containing grains and 22,4% feeding a grain-free diet while 3,2% of the participant did not provide any answer on this question.

When asked the grain-free feeders for reasons, upon the most common were:

1. Believes that a grain-free diet could be healthier (Most were unsure of the factuality of their claim)
2. Allergies, intolerances
3. The vegan dog food they chose happened to be grain-free
4. Two participants reported of less digestion issues without grains
5. Two participant reported of diarrhea due to a gluten intolerance in their dogs
6. One participant reported that she is feeding for 25 years grain-free diets and her personal experience is that her dogs are appearing to be healthier without grains

When asked the grain-included feeders for reasons, upon the most common were:

1. Grain is very well tolerated and digested
2. No evidence of grains being bad
3. The need of grains in the diet for a dog's overall health
4. Feeding grains not excessively
5. The believe that grain-free is unhealthy for dogs
6. Some mentioned that everything unbalanced can be unhealthy, the key is balance in whatever we give the dogs
7. One participant even mentioned a research conducted in Sweden that showed the adaptability of dogs towards digestion of starches and therefore his knowing that grains can be fed without any issues
8. One participant finds the grain topic overly hyped and it isn´t necessary to limit grains, except in a case of allergy

The discussion about grains in a dog's diet is very split between the surveyed participants and so are their feeding behaviors regarding this subject.

Protein content of dog food

77,2% of all participants claim to feed an average protein containing diet (20-30% protein content); 12,4% reported to feed a high in protein diet (>30% protein content), while 5,2% are feeding a diet low in protein (<20% protein content). 5,2% did not provide an answer to this question.
When asked if participants were open to try out new protein sources for their dog's alimentation the majority with 45,6% responded with "Yes definitely, I believe in using a variety of protein sources, my dog´s diet would be more complete". On the other hand, 39,6% showed also interest in new protein sources, but desire more feedback on newer protein sources. 10,4% of participants were more reluctant and prefer factual data about new protein sources and 1,2% do not want to include new protein sources and want to feed only the protein sources they know of. 3,2% did not provide an answer *(see fig. 15.)*.

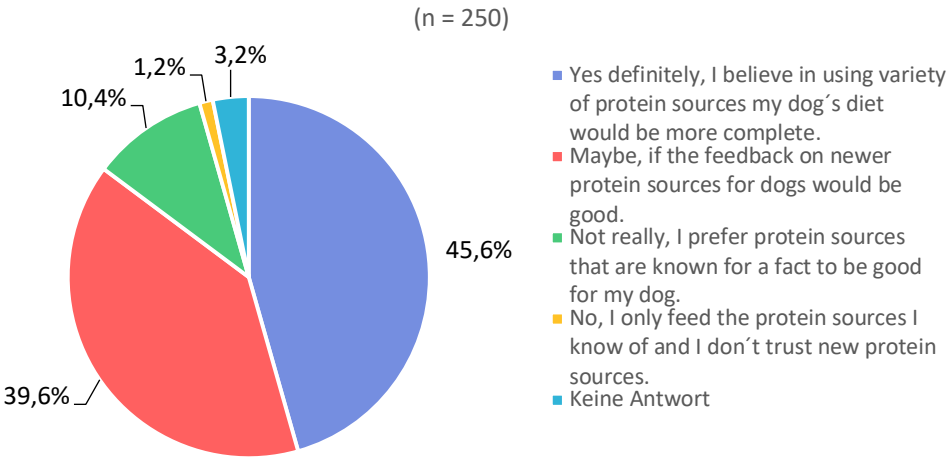

Fig. 15. *Openness of participants for trying new sources of proteins*

Assurance about the nutritional adequacy of the given plant-based food-ratio

The vast majority represented by 59,2% assure the nutritional adequacy of feeding a plant-based diet to their dogs by trusting the package claim of the vegan dog food producer which says "Complete-diet" *(Alleinfutter)*. The EU law defines a complete pet food as "Any food which, by reason of its composition, is sufficient for a daily ration" Regulation EU No. 767/2009 (57), therefore assuring the average total quantity of a specific pet food that is needed daily by a pet of a given species, age category and lifestyle or activity to satisfy all its energy and nutrient requirements. 24,8% reported to use the package claim and have had a blood test done for evaluation of adequacy. While 10,8% had a nutrition counseling with a dietitian (professional advice). 5,6% did not provide an answer to this question.

Frequency of feeding per day

66% report to feed their dogs twice per day, 15,2% three times per day, 10,4% once per day, 5,2% report to feed their dogs irregularly while 0,8% did not provide any answer *(see fig. 16.)*.

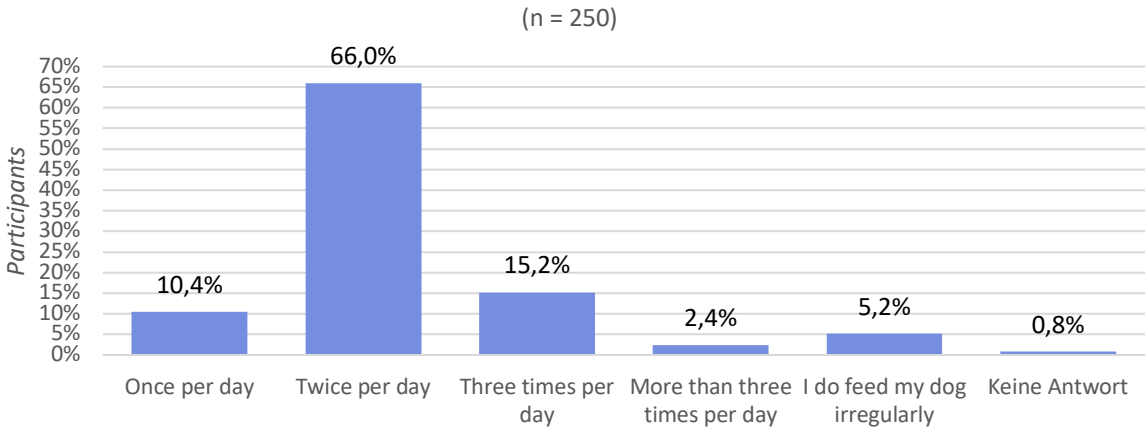

Fig. 16. *Number of feedings per day*

The amount of food given per day depends on a variety of factors like age, sex, breed, level of activity, reproduction state, etc. The results showed that 46% of all participants do feed portions between 200-400g per day. 100-200g (12,8%); 200-300g (22,8%); 300-400g (23,2%); 400-500g (12%); 500-600g (10,4%); >600g (10,4%); No answer given (8,4%) *(see fig. 17.)*.

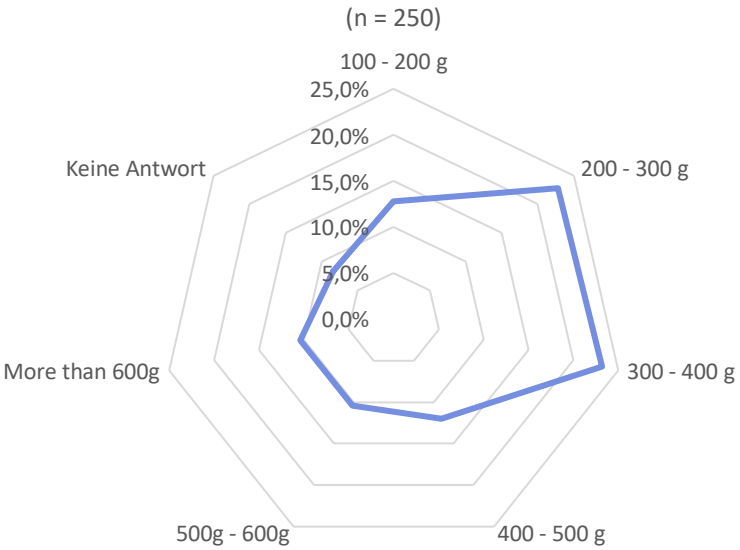

Fig. 17. *Amount of food given per day*

Determination of needed amount of food

54,8 % feed their dogs according their own experiences. 23,6% determine feeding amounts according to the manufacture guidelines given on the package or homepage of used dog food, 13,2% according to the appetite of their dogs and 7,2% feed ad libitum (meaning the food can be accessed as desired by the dog). Not one participant reported to arbitrarily determine feeding quantities. 1,2% did not provide an answer to the question.

Acceptance of different food types

Palatability of different dog food types are represented by a plot-diagram *(fig. 18.)*. The given options were: Commercial vegan; Homemade vegan; Mixture commercial and homemade vegan, Commercial meat-based, Homemade meat-based, Mixture of commercial and homemade meat-based, Mixture of Vegan and Meat-based. In general, all categories had a very high acceptance, this can be represented by the mean of the point-score of all categories, ranging from 74,44 to 93,70 (0 representing a very badly acceptance and 100 a very good acceptance) *(see table 16)*. The highest acceptance was represented by the vegan food category Mixture of vegan commercial and vegan homemade food with a mean point score of 93,70. In fact, the 3 highest groups were all representatives of the category “vegan” in the following sequence in decreasing order: Mixture of vegan commercial

and vegan homemade; Homemade vegan; Commercial vegan. The lowest acceptance was represented by category Commercial meat-based dog food with an average acceptance point score of 74,44.

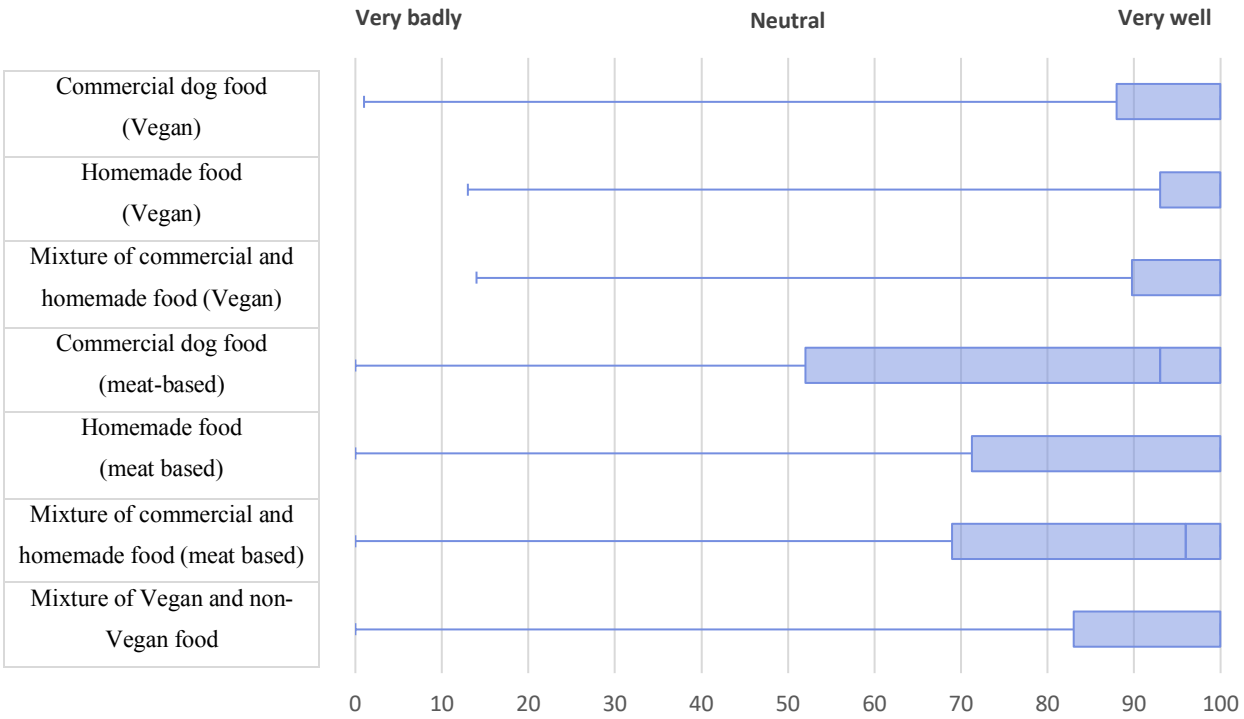

Fig. 18. *Acceptance of different food types*

Table 16 *Experience on acceptance of different food types by dog owners*

	N.	Not Applicable	Minimum	Maximum	Interval	Average	Median	Variance	Standard deviation
Commercial dog food (Vegan)	226	15	1.00	100.00	99	91.65	100.00	203.04	14.25
Homemade food (Vegan)	205	32	13.00	100.00	87	93.05	100.00	216.58	14.72
Mixture of commercial and homemade food (Vegan)	188	44	14.00	100.00	86	93.70	100.00	125.59	11.21
Commercial dog food (meat-based)	126	93	0.00	100.00	100	74.44	93.00	1048.46	32.38
Homemade food (meat based)	74	155	0.00	100.00	100	79.58	100.00	988.33	31.44
Mixture of commercial and homemade food (meat based)	61	158	0.00	100.00	100	77.39	96.00	1033.44	32.15
Mixture of Vegan and non-Vegan food	101	124	0.00	100.00	100	85.71	100.00	694.65	26.36

It is important to acknowledge that there are a multitude of factors influencing the acceptance of food and therefore the results, such as: The quality and quantity of food given; appetite; hunger and satiety; taste; palatability; sensory aspects; social setting; social context; meal patterns; psychological factors like stress, mood; eating disorders; health of individual; changing food behavior, personal bias and many more.

Tolerance of vegan dog food

87,6% report that their dogs tolerate vegan food without any issues and 10% report that the food is being tolerated. There hasn't been one dog owner reporting that the food is not well tolerated, and none reported that the food is not tolerated at all, however, 2,4% did not give an answer to this question.

Allergic reaction

97,6% of all participants responded to this question while 93,6% reported no allergic reaction while switching on a vegan diet. 3,6% reported an allergic reaction while switching to a vegan diet, with the following symptoms: ear itching; scratching; skin irritation; One participant reported about inflammation and swelling of the dog's lips after trying a certain commercially available vegan dog food. However, it is very important to acknowledge here that the allergen most likely causing "cutaneous adverse food reactions" (CAFRs) in Europe, Australia or North America are beef, dairy-products, chicken, wheat and lamb. 4 out of the top 5 CAFRs causing allergens are from animal products and only 1 from plants (58).

General knowledge of participants about nutritional requirements of dogs

The results showed that 46,4% of all participants would evaluate themselves as "well informed"; 27,6% as "knowing the basic requirements"; 20,4% as "very well informed"; 3,6% reported to be "not well informed" and 0,8% (representing 2 participants) reported to be "not informed at all". 1,2% did not provide an answer for this question.

General knowledge of participants about nutrient content of foodstuff

The results were similar to the self-evaluations in nutritional requirements of dogs. 49,6% described themselves as "well informed"; 23,6% as "knowing the basic requirements"; 20% as "very well informed"; 4% reported to be "not well informed" and 0,8% reported to be "not informed at all". 2% did not provide an answer for this question.

General knowledge of participants about potential deficiencies of a plant-based diet for dogs

The results again showed similar results to the two previous self-evaluations. 44% described themselves as "well informed"; 29,6% as "knowing the basic requirements"; 21,2% as "very well informed"; 3,2% reported to be "not well informed" and 0,4% reported to be "not informed at all". 1,6% did not provide an answer for this question.

Defecation frequency

54% report 2 defecation/day; 31,2% report 3 defecations/day; 10,4% report >3 defecations/day; 3,2% report 0-1 defecations/day; 1,2% did not provide an answer to the question.

Stool color

Reported stool color was described for the majority to be brown (96%), whereby different shades of brown were noted, described as light/chocolate/dark-brown. Only 2 participants reported a color "yellow" different than brown. 3,2% of the participants did not provide an answer to this question *(see fig. 19.)*.

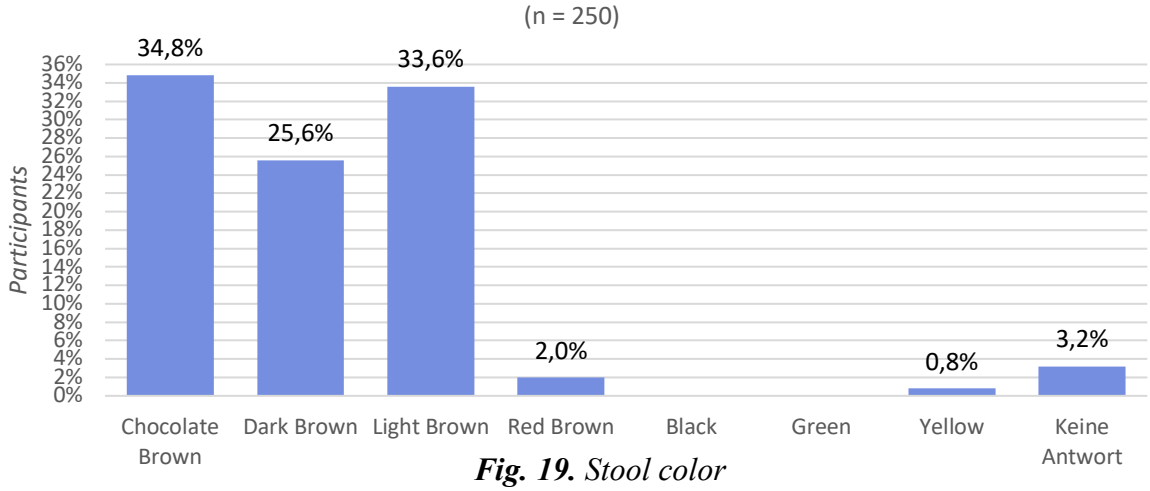

Fig. 19. *Stool color*

Stool consistency

97,2% reported a normal stool consistency, of which 51,2% were reported as "smooth and soft, sausage-like" and 46% as "sausage shaped with cracks on the surface". 0,8% reported a mushy consistency (mild diarrhea) and 1 participant (0,4%) indicated his dog to have separate hard lumps. 1,6% did not provide an answer to this question.

Supplementation of dog´s food

Participants were asked if they are supplementing their dog´s food and 50,4% reported to supplement and 47,6% did not supplement. 2% did not provide an answer to this question. 58 participants reported to use Vegdog; 26 are using Nutritional yeast; 18 to use V-complete, 8 to use single vitamins, 5 using brewers´s yeast and 3 supplementing with a Mixture of herbs.

Several more reported to supplement with the following: seaweed, taurine, spirulina, L-carnitine, lupine powder, digestive enzymes, green mush, MSM, glucosamine, CBD oil, Hokamix powder, Boswellia powder, Quercerin, Algae, chlorella, Omega 3 oils, seaweed powder, flaxseeds, sunflower seeds, pumpkin seeds, desiccated coconut, Goji berries, chia seeds, hemp seeds, Vitamin C, Calciumcitrat, Mineral nutrition mix, Augustine, Rose hip vitals, Cranium, Dorset greens, missing

link, Nepani, Vegepup, moringa powder, medical mushrooms, D-mannose tablets, curcuma powder, turmeric, berries, psyllium husk, cinnamon, ginger. 120 participants did not provide any answer.

Origin of treats

84,8% are reinforcing good behavior with treats, 13,2% do not reinforce good behavior with treats and 2 % did not provide any answer. 61,6% report to use vegan treats whereby 26,8% report to use vegan and non-vegan treats as sometimes other people are giving treats that can be derived from animal products. 3,6% report to use animal derived treats and 8% did not provide an answer to the question. The treats used are supplied by a variety of different brands, to mention a few: Benevo, Ami, Vegdog, V-dog, Halo, Vegan4dogs, Homemade treats, Whimzees, Yarrah, Greta, Wild earth, Variety, Lukkulus, Napani, Anibio, Rinti, Chewies, Nattura, Keksdieb, Hundsfutter, Veggiedog, Wainwright´s, Lily´s kitchen, Pawsome Organics, Dr. pogo, Joes vegan buiscuits, Premier (Fressnapf), Harrah, Forza Bio, Camon, Vetconcept, terra-pura bio, Fruitibles, Antos, pooch and mutt, healthy paws, soopa, Veggiedent, Zukes, Snooks, Canine carry outs, Vegepet, garden bites, Trixie, Dentagum.

Many of the participants also reported to use kibble and normal foodstuff as treats such as: Vegan cheese, carrots, vegan sausages, tofu, smoked tofu, chickpeas, cooked potato pieces, broccoli, dried bread, bananas, pumpkin, peanut butter, fruits, vegetables.

3 participants reported in giving non-vegan treats such as: Dried meat, dried fish, regional wild meat.

Recognized changes in dog after switching on a vegan diet

Interestingly, 54% reported to have observed changes in their dogs after switching on a vegan diet while 40,8% have not noticed any changes and 5,2% did not provide an answer on the question (*for detailed information on observed changes, please see fig. 30.).*

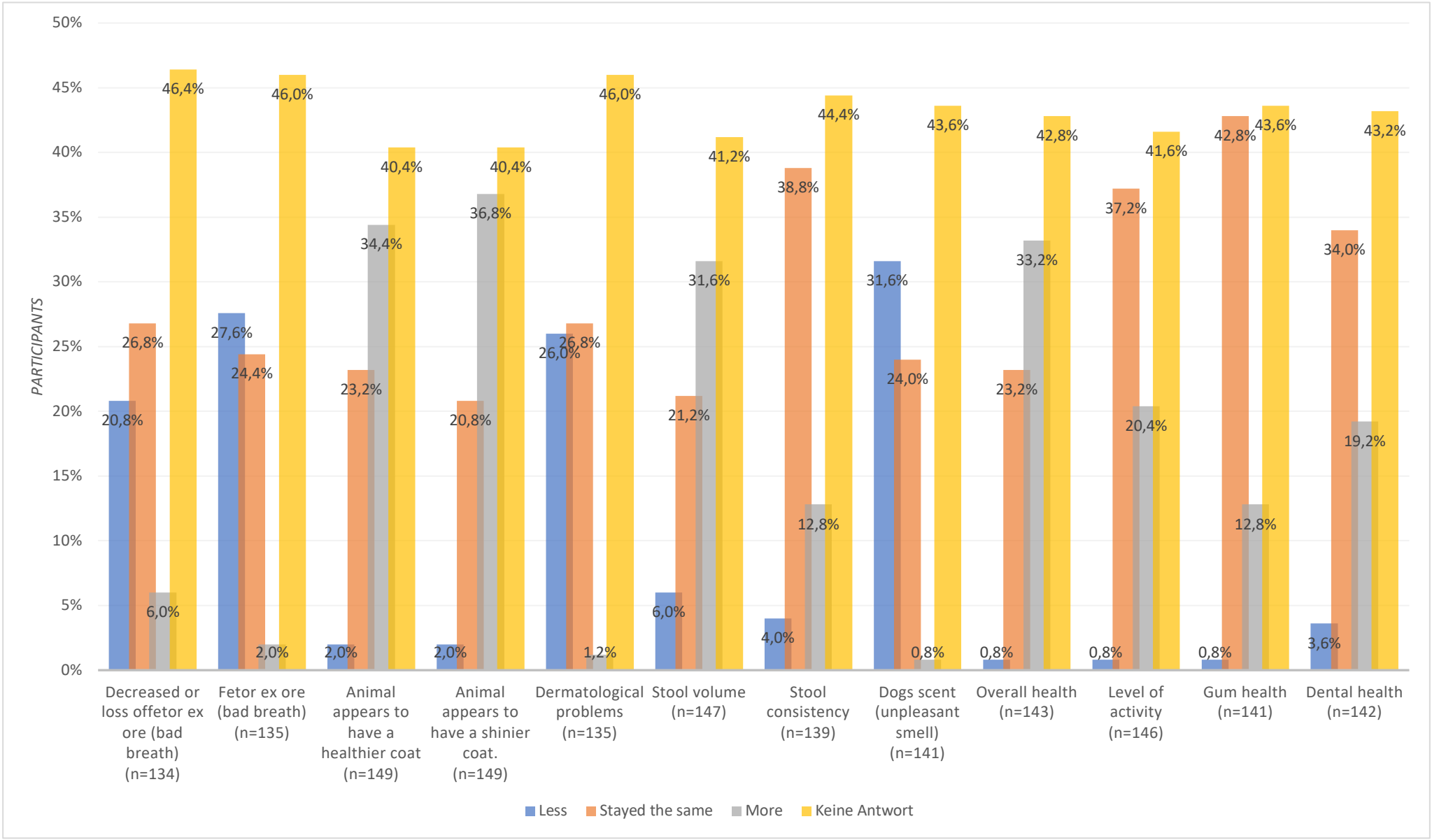

Fig. 30. *Observed changes after switching on a vegan diet*

Awareness of alkalization of urine (increased pH)

Meat and animal products when metabolized are considered acidic. Due to high amounts of purines present in RNA and DNA in animal products uric acid is formed, which is the acidifying agent behind decreased pH in urine. It is true that a plant-based diet, being less acidic than a meat-based diet, will increase the pH of the dog´s urine.

On the flip side, a too acidic diet would increase the risk for crystallization and building of Calcium Oxalate stones. It is about finding the optimal pH in order to prevent urinary tract issues.

The majority of participants were not aware of an increased pH of the dog's urine when feeding a vegan diet (38%). 31,2% reported to be aware of the urine alkalization and 28% were "More or less" aware of this potential issue, 2,8% did not provide an answer.

Awareness of Acidification of urine (decreasing pH) by food additives

When asked if participants were aware of the possibility to counteract the alkalization and the potential increased risk for Lower urinary tract inflammation, crystallization and building of struvite (stones in the urinary tract system) by natural food additives, the clear majority represented by 43,6% reported to be unaware of this. 27,2% reported to have heard about the possibility to acidify the dog's urine but still didn´t check their dogs' urine pH levels while 26% were aware of food additives as an acidifier to the dog's diet. 3,2% did not provide an answer.

Urine pH values of participants

Only 19 out of 250 participants were able to provide urinary pH levels, eight of which reported a urinary pH of between 6,0-6,5. Three participants had a urinary pH of 6,5-7,0; another three of 7,0-7,5; two had a pH-range of 7,5-8,0; two a pH of 5,5-6.0 and only showed a pH of 4,5-5,5 *(see fig. 31.)*. The recommended healthy pH range of a dog´s urine is 5-7, it is not abnormal for healthy dogs to have more acidic or alkaline urine, however, abnormal pH is known to promote crystals in the urinary system. Therefore, a constant pH value out of recommended urinary pH ranges indicate a higher risk for stone formation. Some studies have found no association between urinary pH and presence of Calcium oxalate uroliths (CaOx) (59), however the prevalence of CaOx-uroliths are increasing not only in dogs and cats but also in humans in the last decades. A strong evidence was shown in the database of Minnesota Urolith Center which showed that only 5% of canine uroliths in 1981 were made up from CaOx while in 2009/2010 it was 45% of all received samples (60), indicating the already existing issue in dogs, whether plant-based or not.

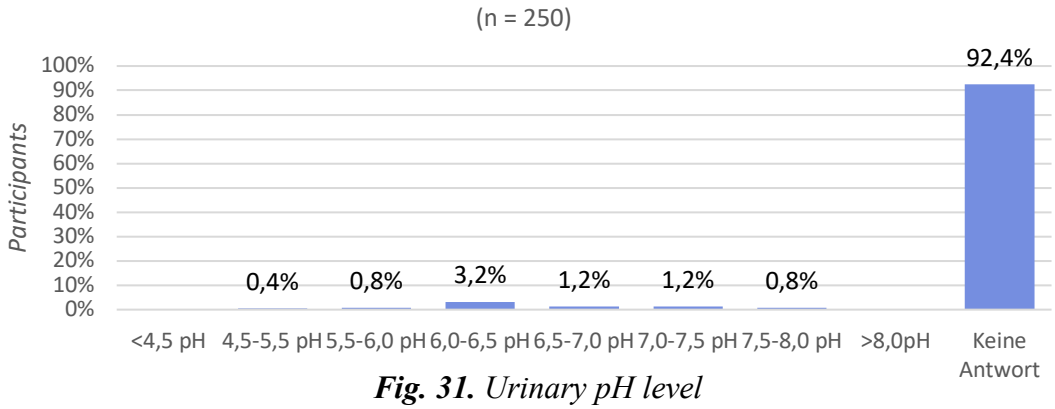

Fig. 31. *Urinary pH level*

Is there any association between a vegan diet and lower urinary tract infection (LUTI)?

The question was asked how many of the dogs participating in the study were diagnosed with a UTI while being on a vegan diet and to compare the prevalence to prevalence levels of meat-based fed dogs. 87,2% reported that no UTI have been diagnosed while feeding a vegan diet, whereby 13 participants (5,2%) reported a diagnosed UTI while being fed a vegan diet. 7,6% did not provide an answer.

Comparing the prevalence of UTI in the study participants to meat-based fed dogs it can be assumed, that even when mostly feeding a vegan diet without an added acidifying agent, there is a decreased prevalence of UTI disease in the study participants. As studies have shown the prevalence of LUTI to be around 26,6%, while the lifetime risk for LUTI is 14% (61; 62). The risk of a LUTI positive urine culture is 2,5 times higher in spayed female dogs over a neutered male and 1,5 times higher for an intact female over a neutered male (62). However, the highest risk group for LUTI are spayed females in the higher age group (63).

In this study the plant-based fed dogs had a LUTI prevalence of only 5,2% compared to 14% for meat-based fed dogs.

Experience on feeding a vegan diet

After having fed a vegan diet, the participants were asked if they would recommend a vegan diet for dogs to their friends and colleagues. 44,4% of all participants responded with “Yes, definitely” and that they do recommend it very often. 38% stated that they recommend a vegan diet for dogs but only if being asked from someone out of his/her own interest and 0,4% (1 participant) would not recommend a vegan diet for dogs.

Interestingly, 13,6% reported that they would like to recommend it but are afraid to be ridiculed and 8,8% stated to never mention feeding a vegan diet in order to avoid negative comments. This clearly shows that the broad society is approaching this topic with a strong prejudgment.

When asked if the participants find it easy talking to friends, colleagues or family members about a vegan diet for dogs, 46% answered with “sometimes” and 37,6% answered to not find it easy, while 14% do find it easy talking about this topic. 2,4% did not provide an answer.

The participants were asked why they find it hard having a conversation about this topic, the results are the following:

1. 41,2% say that most have an immediate prejudice about this topic.
2. 23,2% say that most believe that dogs are no different from wolves in terms of nutrition.
3. 11,6% say that most don´t even want to listen to scientific research, because their refusal towards this topic is hindering a constructive conversation.
4. 10,4% did not provide a specific answer.
5. 6,8% say that most believe they are experts on dog nutrition.
6. 3,6% say that most don´t realize that commercially available dog food already constitutes mostly of plants in order to produce a cheaper product and to increase profit.
7. 3,2% say that many are not keen talking about the topic “vegan” in whatever matter.

Is a vegan diet for dogs cruel or unhealthy?

72,4% answered with “No, it is the opposite of cruel as it is an act of compassion. Dogs can even be healthier on a vegan diet”. 22% responded with “No, I don´t find it to be cruel or unhealthy for dogs” and 3,2% responded with “I am not sure. It needs more research”. Not one single participant found feeding a vegan diet to dogs cruel nor unhealthy, however 2,4% of the participants didn´t provide an answer.

The biggest concerns of the participants feeding a vegan diet to dogs were:

34%: Not being taken serious from veterinarians; 30%: Do not have any concerns; 28,8%: Nutritional inadequacy; 19,2% Being labeled an animal abuser; 15,6%: Health issues; 8,4%: Rejection of the food; 3,2%: Did not provide an answer.

4. DISCUSSION OF RESULTS

The growing public awareness of the current climate emergency, the significant ongoing environmental destruction and the fact that humanity is in the midst of the sixth mass extinction have led the public to speak up and take action on personal and community levels while demanding more government initiatives. People are outraged and demand justice, which was demonstrated recently (Sept. 2019), when the largest climate strike in world history took place (64), with approximately six million participants in more than 150 countries (65). The consensus of 195 countries and hundreds of leading scientists have led to the Intergovernmental Panel on Climate Change (IPCC) and its comprehensive reports, which are reviewed by thousands of experts and undergo multiple drafting rounds (66, 2). The results are clear: anthropogenic activities are by far the main driver of climate change, loss of biodiversity, mass extinction and water degradation.

Animal agriculture is the main driver behind deforestation (67), water degradation (68), greenhouse gas emissions (8, 9) land grabbing (69) water pollution (68) and ocean death zones (70). Some studies even suggest a twofold to 20-fold increase in nutritionally similar food per cropland area if animal-based products were replaced with plant-based alternatives (71). This would allow tremendous amounts of land to recover, thereby creating carbon sinks and space for biodiversity to flourish. Some set the number even higher. Professor Peter Smith, a chair in Plant and Soil Science at the University of Aberdeen and convening lead author for the United Nations body, stated in an article for the BBC that the climate, land and water footprint can be up to 100 times greater for some animal products compared to plant-based replacements (72).

Several studies show that most people are unaware of the tremendous impact the choice of food source can have (10, 73) in terms of production resources and emissions. Unfortunately, many are reluctant to reduce or stop consumption of meat due to convenience and pleasure (73). Increased scientific consensus in human nutrition have led to the rise of one of the fastest-growing social movements in history, veganism.

Awareness of consuming less animal products for better health is rising, combined with the knowledge that animal products are one of the highest impact food sources (beef being the single food with the greatest impact on the environment) (74), have led to a tremendous acceleration in the vegan movement. The moral and ethical awareness of farmed animals, wildlife and fish and concerns regarding animal welfare are also increasing (64); however, many sharing those beliefs tend to extend their concerns, for obvious reasons, to their companion animals.

Many may consider it ethical to feed a vegan diet to dogs to spare many animals the unnecessity of a life filled with pain and suffering, destined for a crowded ride filled with fear to the kill floor. Is it unethical trying to avoid the breeding of animals into existence as sole purpose to

nourish animals society values more than others? Is it unethical feeding a diet to dogs that tries to abolish the idea of speciesism? Is it cruel to feed a diet to try to reduce the environmental impact and emissions needed for its production? For the majority of participants, it is the opposite; they describe it as an act of compassion, including not only their companion animals into their consideration but all living beings on earth.

The dog owners participating in this study show great awareness by well-defined and logical reasons for their own diet choices and their choices on the alimentation for their dogs and its direct and indirect effects on others, including humans.

As we can see from this research, the bloodwork results of the vegan participants were of no concerns to their owners. Even the longest vegan-fed dog (> 10 y) in this study showed healthy blood results. This is adequate motivation for this research, as there is a relatively small body of existing study on this topic.

The National Research Council, as well as the 5th Small Animal Clinical Nutrition book officially recognizes the dog as an omnivorous animal (75, 76). A study conducted in Sweden in 2013 showed the domesticated dog has adapted since descending from the wolf: metabolic and digestive adaptation has led to an increase in amylase expression, allowing dogs to more easily break down starches than wolves. Specifically, the amylase gene showed an approximately 28-fold increase in activity (28). The concern of protein in a vegan diet for dogs originates from the belief that only meat is rich in high-quality protein, but there is literature that shows that plants are excellent sources of proteins and amino acids, and are already used in almost every commercially available dog food. These plants include rice, wheat, corn, barley, gluten meal, soybeans, pea protein, lentils and many more (76), and therefore, a well-balanced vegan diet will not lack the needed amount of protein for a dog: all 20 long-term fed vegan dogs in this study had healthy recommended protein blood levels. The same observations were seen in other studies on vegan dog food (77). Dog physiology does not use or distinguish proteins and amino acids differently from plant or animal sources. Although the dog is not capable of synthesising needed proteins if the food ratios are lacking the needed amount of proteins, this can easily be avoided by adding sufficient quantities of protein to the diet (76).

As described in the results, more than 30% of owners feeding a vegan diet reported an increase in dog stool volume and 31% report a defecation frequency of 3 times per day, showing faster transition times for passing through the gut, explainable by the increased content of fibre in the diet. This is consistent with the observations of the National Research Council in 2006 (75). Several other studies have also shown that plant protein sources are feasible alternatives to poultry meal protein sources (78). Taurine does not need to be supplied as dogs can produce taurine themselves; however, it is recommended to supply some taurine to the food to assure sufficient levels as some breeds show low plasma taurine concentrations (79). Dogs do not require carbohydrates, but they require glucose

which, on a vegan, plant-based, diet are supplied in more than sufficient amounts. Vitamin B12 is a nutrient of concern in a vegan diet as this vitamin is produced by microorganism and are normally passively filtered through prey animals or consumed via water sources from rivers or lakes containing such microorganisms. Therefore, a vegan or vegetarian diet for dogs living in a modern environment over the long term will lead to deficiencies in Vitamin B12 (76). However, it is straightforward to add B12 to the dog's diet to avoid this issue, as suggested by the National Research Council (75), which also works well for humans following a vegan diet. In this study, none of the vegan dogs showed a vitamin B12 deficiency, not even in cases of a decade-long vegan diet, while four conventionally meat-based fed dogs showed a deficiency in Vitamin B12.

Currently, most farmed animals live in closed and confined spaces, without access to natural sources containing sufficient vitamin B12-supplying microorganisms; administered water has been processed and is free of microorganisms. Ultimately, all dogs need vitamin B12 supplementation, whether on a vegan or meat-based diet through passive supplementation. However, gastrointestinal pathologies can greatly decrease vitamin B12 absorption, especially in chronic gastrointestinal diseases (80), making the diet not the only concern.

A general concern of feeding a vegan diet is increased urinary pH. Even if this research proved the prevalence of LUTI to be lower than in the general dog population, it should be recommended to each dog owner feeding a vegan or vegetarian diet to frequently check the urine pH and add acidifying agents to the dog food if needed. Great natural acidifiers include vitamin C, oats, lentils, asparagus, peas and yeast (81).

There are some concerns when feeding a vegan diet, but there are also concerns when feeding a meat-based diet. In the six-week vegan trial, no significant differences were observed between the vegan or meat-fed group ($p > 0.05$). There are a multitude of factors influencing the quality and bioavailability in foodstuff, regardless of the source. However, as research suggests thus far, a vegan diet for dogs is not only possible but can maintain and in cases even increase health in dogs. As observed in the results of this study, a dog can be fed a vegan diet that is well-balanced and nutritionally adequate. Vegan dog food companies are continually improving their formulas, making it easier for consumers to feed a nutritional, well-balanced vegan diet.

CONCLUSION

This research has shown that the long-term vegan-fed dogs showed the same number of nutritional surpluses as the conventional meat-fed control group (all were detected for iron). The meat-based fed control group showed 11 deficiencies (four folic acid, four vitamin B12, two calcium and one iron), while the long-term vegan fed category presented only two deficiencies in total (lower than recommended folic acid values, explained by a giardia infection during the blood collection. When comparing the groups (plant- and meat-based), the mean differences in protein, calcium and magnesium showed no statistically significant difference ($p > 0.05$); the results showed statistically significant differences in iron, vitamin B12 and folic acid ($p < 0.01$). The physical examinations did not raise any suspicion of nutrimental-related issues.

The results of the six-week vegan trial showed that most of the blood chemistry values remained steady during the trial. Three deficiencies detected before the trial in folic acid, vitamin B12 and iron reached recommended healthy ranges during the vegan diet. However, no statistically significant differences were observed between the vegan and meat-based control groups during the trial ($p > 0.05$), further strengthening the plausibility of feeding a vegan diet to dogs. The physical examinations did not raise any suspicion on nutrimental caused issues.

The 250 surveyed dog owners responded with defined reasons (ethical, environmental and health) for feeding a vegetarian or vegan diet to their dogs and themselves. Of the 250 surveyed, 54% of dog owners feeding plant-based food observed positive health changes while feeding a plant-based diet, and the great majority reporting increased health and positive health changes. However, stool volume and frequency were reported by 31.6% to have increased. Out of 250 surveyed dog owners, only one would not recommend a vegan diet to others, which this shows the great satisfaction felt by dog owners being able to offer their dog a vegan diet.

The performed direct food ratio analyses with expert DMV Uwe Romberger also confirmed that a vegan diet, if well balanced, has an abundance of nutrients and proteins, supplying the dog with all needed elements (see Annex 1).

Feeding a vegan diet to dogs is shown to be possible, according to this research. The reasons behind such a practice are clear and well defined, as are the diet choices of the dog owners. Feeding a vegan diet to dogs is not a trend but a solution to diminish the consequences of climate change and simultaneously raise the standards of animal ethics in 2020.

I recommend consulting a veterinarian such as Uwe Romberger or Lisa Walther, who are specialized in vegan dog nutrition.

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I met another very special person along my research, **Randy Rau**, a person very concerned about animal welfare and the unnoticed pain and suffering in animal agriculture. She contemplated for a long time to feed her dogs a vegan diet but was unsure if it would be nutritionally adequate. Therefore, she joined the six-week vegan dog food trial and paid for the bloodwork costs on her own. Thank you, Randy Rau, for all your efforts in animal welfare.

Also, I want to thank **everybody** that contacted me for offering their dogs for a blood test. Thank you for understanding that my crowdfunding was not enough to cover the full costs of the bloodwork. Thank you all; very much. Without your dogs, this study would have never been possible.

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Be open to new ideas, even if they seem crazy at first glance, sometimes there is more truth to find than we might anticipate. If you might not agree with someone, inform yourself first before being judgmental. We can still turn the climate crisis around, let´s unite and if you have an idea yourself, go for it! Don´t wait for others to do it. I believe in you.

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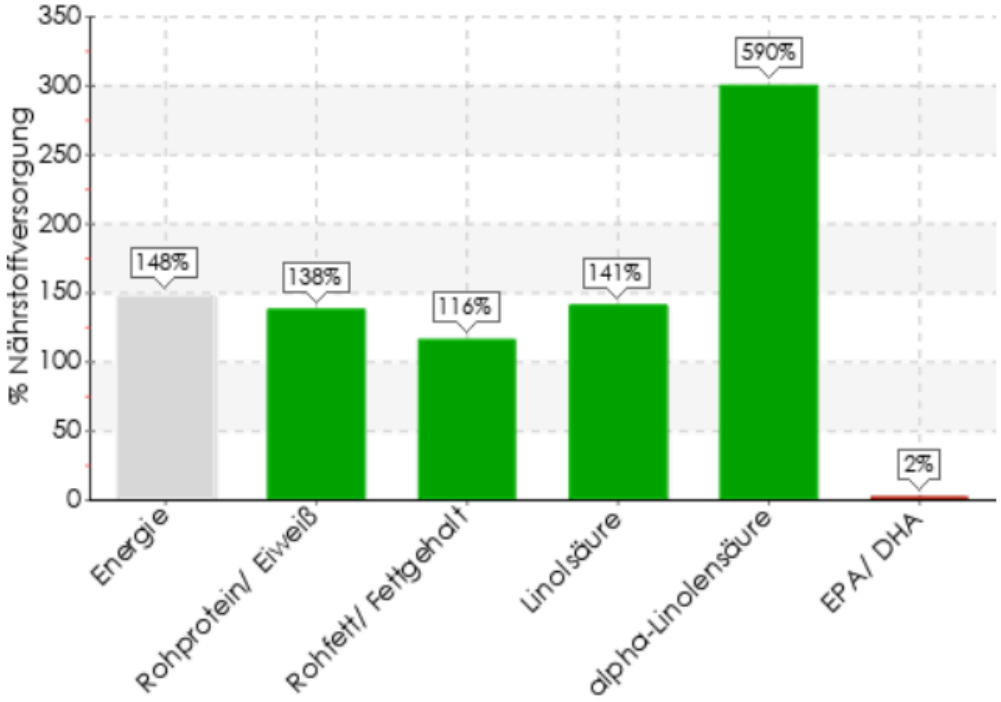
Annex 1

Futterplan vegan FÜR Rosine vom 07.11.2019

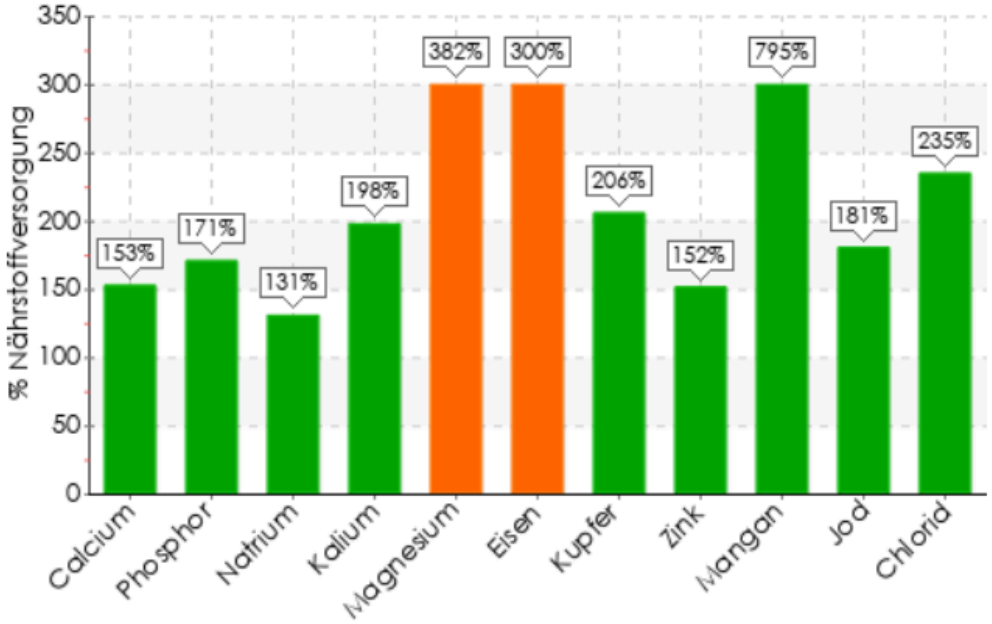
aktuelles Gewicht: 11.2 Kg, Alter: 4.9 Jahre, Endgewicht/ Idealgewicht: 11.8 Kg

Rassenvergleich 12 - 14 Kg, aktuelle Gewichts-Einschätzung: Untergewicht Besonderheit/ Erkrankung:

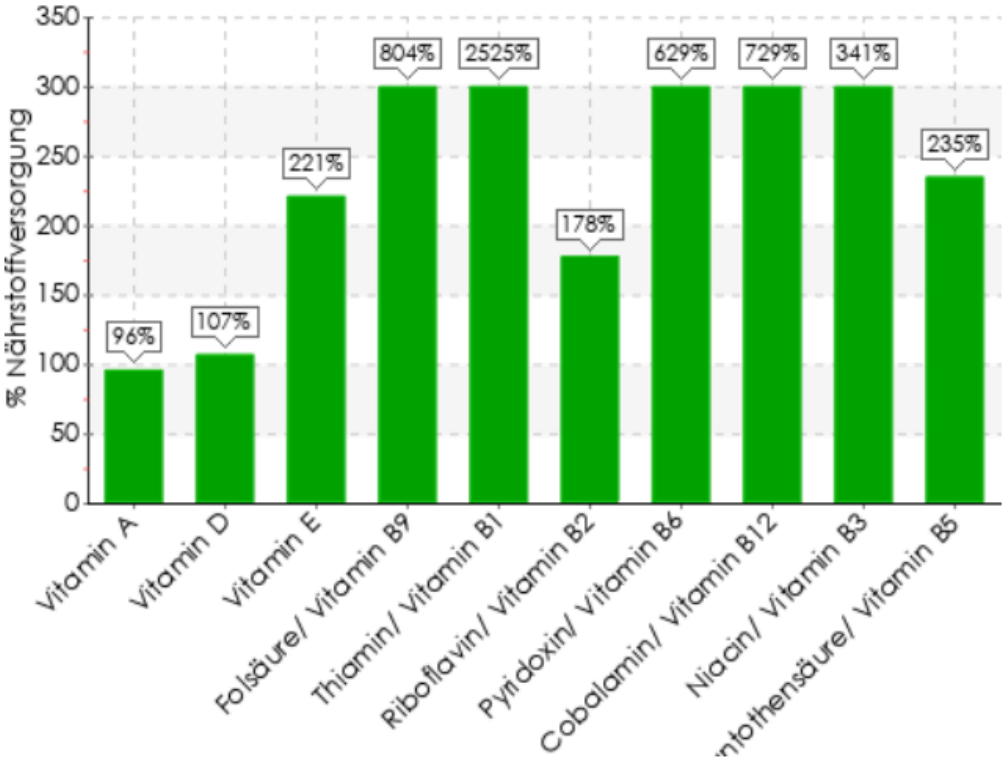
Kalorien-, Protein- und Fettversorgung



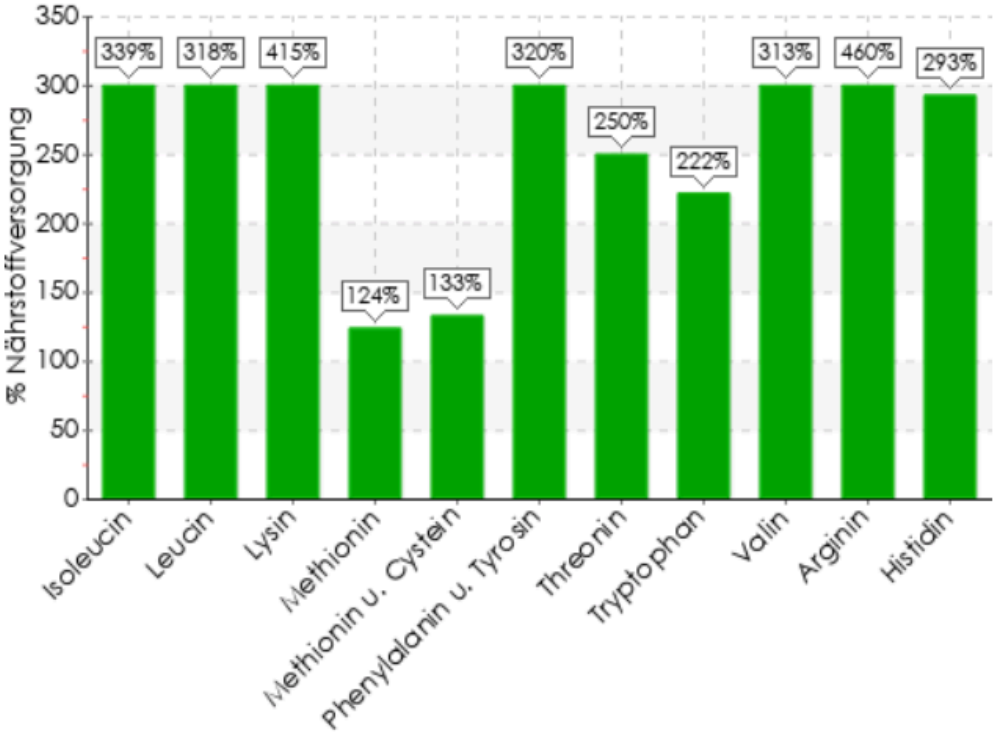
Mineralstoffversorgung



Vitaminversorgung



Aminosäurenversorgung



Nährstoff	täglicher Bedarf	GEFÜTTERTE Menge	%Deckung
Energie	604.8 (kcal)	895.1 (kcal)	148 %
Rohprotein/ Eiweiß	41.4 (g)	56.9 (g)	138 %
Rohfett/ Fettgehalt	11.4 (g)	13.3 (g)	116 %
Calcium	827.7 (mg)	1262.4 (mg)	153 %
Phosphor	620.7 (mg)	1062.6 (mg)	171 %
Natrium	165.5 (mg)	217.2 (mg)	131 %
Kalium	827.7 (mg)	1642.2 (mg)	198 %
Magnesium	124.1 (mg)	474 (mg)	382 %
Eisen	6.2 (mg)	18.6 (mg)	300 %
Kupfer	1.2 (mg)	2.6 (mg)	206 %
Zink	12.4 (mg)	18.9 (mg)	152 %
Mangan	1 (mg)	7.9 (mg)	795 %
Jod	182.1 (mcg)	328.7 (mcg)	181 %
Vitamin A	1045.6 (IE)	1000 (IE)	96 %
Vitamin D	112.6 (IE)	120 (IE)	107 %
Vitamin E	6.2 (mg)	13.7 (mg)	221 %
Folsäure/ Vitamin B9	55.9 (µg)	449.3 (µg)	804 %
Thiamin/ Vitamin B1	0.5 (mg)	11.7 (mg)	2525 %
Riboflavin/ Vitamin B2	1.1 (mg)	1.9 (mg)	178 %
Pyridoxin/ Vitamin B6	0.3 (mg)	2 (mg)	629 %
Cobalamin/ Vitamin B12	7.2 (mcg)	52.8 (mcg)	729 %
Niacin/ Vitamin B3	3.5 (mg)	12 (mg)	341 %
Biotin/ Vitamin H/ Vitamin B7	23.6 (mcg)	67.2 (mcg)	285 %
Pantothensäure/ Vitamin B5	3.1 (mg)	7.3 (mg)	235 %
Chlorid	248.3 (mg)	584.7 (mg)	235 %
Isoleucin	786.3 (mg)	2665.4 (mg)	339 %
Leucin	1407 (mg)	4480.5 (mg)	318 %
Lysin	728.3 (mg)	3023.1 (mg)	415 %
Methionin	687 (mg)	849 (mg)	124 %
Methionin u. Cystein	1349.1 (mg)	1795 (mg)	133 %
Phenylalanin	935.3 (mg)	2836.7 (mg)	303 %
Phenylalanin u. Tyrosin	1531.2 (mg)	4897 (mg)	320 %
Threonin	893.9 (mg)	2234.3 (mg)	250 %
Tryptophan	289.7 (mg)	642 (mg)	222 %
Valin	1018 (mg)	3191.1 (mg)	313 %
Arginin	728.3 (mg)	3348.9 (mg)	460 %
Histidin	397.3 (mg)	1162.1 (mg)	293 %
Linolsäure	2317.5 (mg)	3266.2 (mg)	141 %
alpha-Linolensäure	91 (mg)	536.8 (mg)	590 %
EPA/ DHA	91 (mg)	1.5 (mg)	2 %

Futtermittel in Gramm pro Tag

Gegarte Linsen	150 (g)
Eifreie gekochte Nudeln aus Hartweizengrieß	150 (g)
Frische Zucchini	100 (g)
Banane	50 (g)
Haferflocken	50 (g)
Kichererbsen aus der Dose	20 (g)
Reis Protein Raab Vitalfood	10 (g)
Lupinenpulver der Lupinus albus	10 (g)
Weizenkeimflocken	10 (g)
Bierhefepulver	5 (g)
Chlorellapulver	5 (g)
Spirulinapulver (Jod ca. 0.45mg/100g)	5 (g)
Futtermedicus Vitamin Optimix Cooking Pulver (1 ML = 3.5 g)	4 (g)
getrocknete Cranberry	3 (g)
Kokosnussraspeln	3 (g)
Kürbiskernpulver	3 (g)
Algenkalk (Calcium ca. 34%. Jod ca. 4mg/100g)	1 (g)

Hinweise

Die Beurteilung der Energieversorgung findet bei einer Rationsanpassung nicht statt (grauer Balken), da die Kalorienveränderungen erst nach einigen Wochen der Fütterung des neuen Planes sichtbar werden.

ANNEX 2

Research funding:

Drawing and analyzing blood from 48 dogs for the needed blood parameters (listed in conduct of study) exceeded 10 000€. The university did not provide any financial support for this research. Therefore, IDEXX laboratories in Germany was contacted, the planned research was presented, and it qualified for the IDEXX study section. However main costs would still need to be financed by myself.

Research objectives and methodology was presented to several organizations (55, 56), whereby the Pollination Project (TPP) responded with great interest and funded parts of this study. TPP was founded in 2013 as an international nonprofit organization, whose mission is to spark goodness and compassion in every person through a daily practice of generosity and grantmaking. "We know there are many ways to approach changing the world. It is our belief that uplifting and empowering individuals at the grassroots-level is a particularly potent way to achieve real and long-lasting change "(55).

The Pollination Project selected this study for funding on the 17. December 2019.

Crowdfunding campaign

A GoFundMe crowdfunding campaign titled Vegan dog food – An unconventional perspective, (with a goal to raise 1999€ to support this research) was conducted during a period of 5 months. A total of 535€ were raised to cover parts of the costs considering veterinary blood analysis.

Despite not being fully funded, many recruited vegan dog owners offered to partially or fully finance the bloodwork on their expenses.