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Title: The evaluation of protein and carbohydrate sources with an emphasis on their impact to taurine status in dogs.

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For a grain free diet to be a contributing factor on dilated cardiomyopathy in dogs several ingredient related factors must be demonstrated relative to lower circulating taurine. Previous research has implicated limited bioavailability of precursor amino acids CYS and MET and the impact of fermentable substrates on TAU destruction or elimination. Various animal proteins, legumes and tubers have been implicated recently. Bioavailability of amino acids differs from direct measurement and can be influenced by structural protein composition, thermal processing, and interference from protease inhibitors or structural carbohydrates. Direct evaluation of a single ingredient in the target species is difficult, so surrogate species such as poultry or rodents are often used. More recently *in vitro* methods have been explored such as the protein digestibility corrected amino acid score and others. These methods can aid in the comparison among protein sources for the availability of amino acids such as CYS and MET whether of animal or plant origin. The legume seeds are well established to possess protease inhibitors such as the trypsin inhibitor which can reduce protein utilization. These are generally considered to be inactivated by thermal processing. Fiber sources, such as beet pulp have been shown to reduce TAU in the circulation. Other non-structural carbohydrates such as the oligosaccharides may act in a similar fashion. Legume seeds contain appreciable levels of these non-structural carbohydrates. They are not quantified by fiber analysis so may not be accounted for during formulation. They are fermentable substrates which may affect colonic fermentation and bile acid metabolism; thus, possibly disrupting TAU availability. Examples from our laboratory evaluating implicated ingredients in the dog and model systems will be shared to provide context to the discussion about where they may, or may not, be involved in this canine health issue.