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Technical Symposium
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California Animal Nutrition Conference

Utilizing Engineered Systems & Advanced Microbiology Approaches to Improve Animal Performance and Mitigate Climate Change

Matthias Hess, Ph.D.
Associate Professor, Head, Systems Microbiology & Natural Products Laboratory
Department of Animal Science, University of California Davis

In order to remain profitable, the livestock industry will have to find economical solutions to enhance animal performance, while also reducing its environmental impact. The Hess Lab has developed an artificial (in vitro) rumen system that allows a rapid and highly reliable evaluation of how ruminant animals might respond to new feed additives, feed formulation, etc.

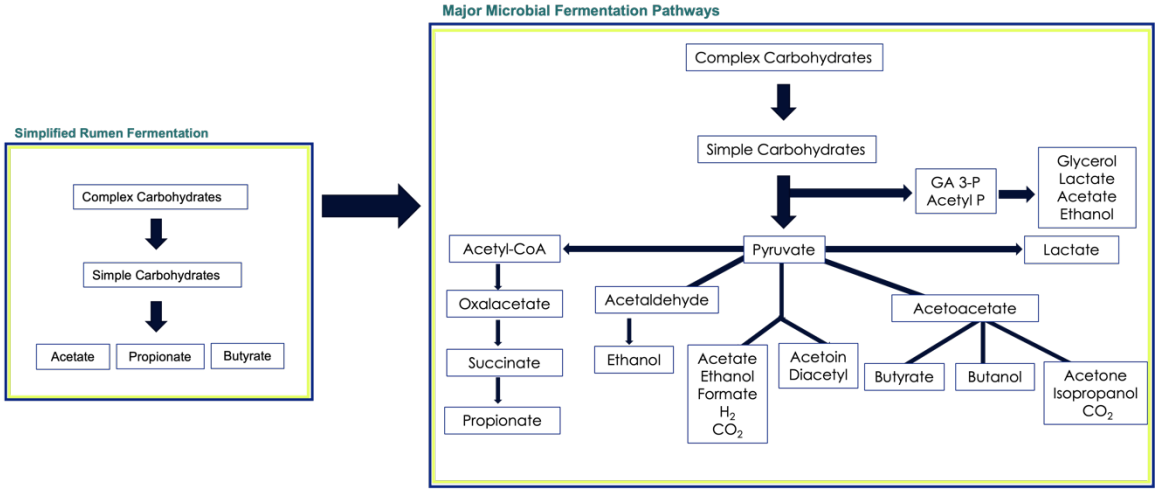
The in vitro system we developed has allowed us to pre-screen hundreds of treatments at a relatively low cost and resulted in the identification of promising candidates, such as the red seaweed Asparagopsis taxiformis which reduced methane production (by up to ~90%) when added to the animal diet without negatively affecting the animal performance.

Over the last months we have tested many more feed additives, especially agricultural waste products, and some of the results will be presented here.

An alternative model of microbiome-driven carbon flow in the rumen

Mallory Embree, Native Microbials, Inc.

The rumen microbiome is a highly complex community comprised of microorganisms originating from all three domains of life. In ruminants, this microbial community plays a vital role in providing nutrition for the host animal through fermentation, primarily through the conversion of feed ingredients into volatile fatty acids (VFAs) that the host ruminant then utilizes as energy. Although correct, this model ignores complexities arising from bacterial fermentative metabolism due to the variation in biochemical pathways, end-product inhibition, as well as influence from the physicochemical dynamics of the rumen environment. Here, we describe in-vivo experiments and existing literature that support an alternative flow of carbon in the rumen that is ultimately driven by the fermentability of the diet. Under ideal conditions, VFAs are the common fermentation by-product. As these compounds and other common fermentation by-products (including CO_2) accumulate, the microbial population will shift towards producing common overflow fermentative by-products such as ethanol, butanol, and acetone via alternative fermentative pathways such as mixed acid and abe fermentation. Chronic accumulation of high concentrations of solvent by-products forces measurable and permanent changes in microbiome composition that are detectable using next generation sequencing. Here, these alternative by-products are explored, as well as their influence on animal health, physiology, and ultimately productivity. Their potential use for rapid, in-field detection of acidosis is also addressed.



Which microbes drive nutritional functions?

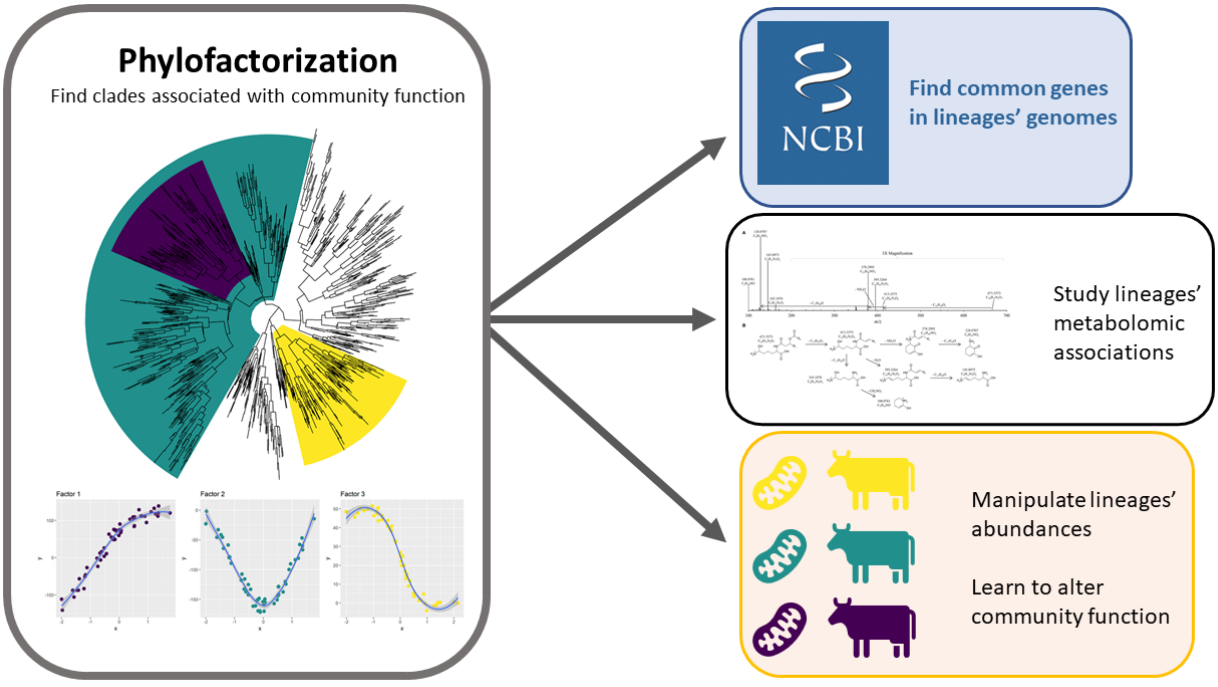
Alex Washburne, PhD

Phylofactorization can find out & advance our understanding of animal nutrition.

Ruminants rely on microbes to digest feed and the microbial determinants of ruminant feed efficiency may lead to products that enhance animal nutrition through modified animal microbiomes. Connecting research and discovery (R&D) with product development, however, requires researching microbial communities with tools capable of identifying specific sets of microbes whose metabolic functions impact animal nutrition. Identifying a lineage of interest allows researchers to study the lineages as possible levers for the ecological control of ruminant microbiomes and nutrition.

In this talk, I will present a relatively novel statistical method for generating insights about animal nutrition from microbiome studies. The method – phylofactorization – scans the evolutionary tree of microbes to find lineages of close relatives who share a common pattern of association with macroscopic variables such as feed efficiency.

Phylofactorization identifies clades associated with community function, connecting microbiome studies with downstream studies of microbial genomic & metabolomic traits, and experimental modifications of microbiomes to improve animal nutrition.



My talk will focus on conveying the intuition of the method by showing how the method has helped us learn new things in microbiomes, conservation, epidemiology, and more. While the mathematical machinery is sophisticated - it is a constrained neutral network, limited to neurons with a phylogenetic interpretation - the question answered by phylofactorization is simple: which lineages of microbes are associated with our meta-data, controlling for confounds? For example, phylofactorization can find lineages of microbes positively associated with milk yield, controlling for nonlinear yield curves, study, and more. Lineages of interest have high within-group similarities relative to between-group differences. The identified clades are hypothesized to have common ecological associations due to common traits inherited from their common ancestor. These clades may be the functional ecological building blocks linking community structure to macroscopic functions such as host nutrition.

After identifying a lineage of microbes associated with animal nutrition, researchers can then look closer at the genomic and metabolic traits of those close relatives to generate hypotheses about the enzymatic and metabolic mechanisms by which microbes modulate animal nutrition. The lineages can also be targeted for experimental enrichment or depletion, as levers of ecological control to modify microbiomes and improve animal nutrition.

Citations:

Washburne, Alex D., et al. "Phylogenetic factorization of compositional data yields lineage-level associations in microbiome datasets." *PeerJ* 5 (2017): e2969.

Washburne, Alex D., et al. "Phylofactorization: a graph partitioning algorithm to identify phylogenetic scales of ecological data." *Ecological Monographs* 89.2 (2019): e01353.

Milk yield and microbial community composition changes in response to a rumen-native direct fed microbial

C. Marotz *, A. Washburne *, F. Yang*, B. Anderson*, J. Lefler*, M Embree*

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Ruminant digestion is dictated by the complex ecosystem of microbial organisms inhabiting the rumen. Over the past two decades, biotechnological advances in DNA sequencing have enabled an unprecedented view of the tens of thousands of unique bacterial and fungal species that comprise the vast majority of ruminal biomass. We harnessed these new technologies to determine which microbial species are most positively associated with healthy, high performing dairy cows. Using specialized machine learning algorithms, we sought to identify organisms that have an outsized influence on their microbial community, the keystone rumen microorganisms. We hypothesized that supplementary feeding of keystone organisms that are highly associated with milk production metrics could alter the microbial ecosystem and lead to more efficient dairy cows.

We isolated these keystone organisms using anaerobic cultivation techniques and developed a preservation method that allows them to survive the harsh journey through TMR. The result, Galaxis™ Frontier (GF), is a daily feed additive containing four live, rumen-native microorganisms:

1. *Clostridium beijerinckii*; a bacterium specializing in the conversion of glucose to butyrate and acetate.
2. *Pichia kudriavzevii*; a fungus which produces catabolic enzymes evolved to digest cellulose and hemicellulose.
3. *Ruminococcus bovis*; a bacterium first isolated by Native Microbials [1] that produces volatile fatty acids, can degrade resistant starch, and is largely influential of the surrounding microbial community.
4. *Butyrivibrio fibrisolvens*; a bacterium capable of diverse catabolic potential with clearly defined biohydrogenation pathways leading to vaccenic acid [2], the precursor for 75-90% of the CLA found in milk [3].

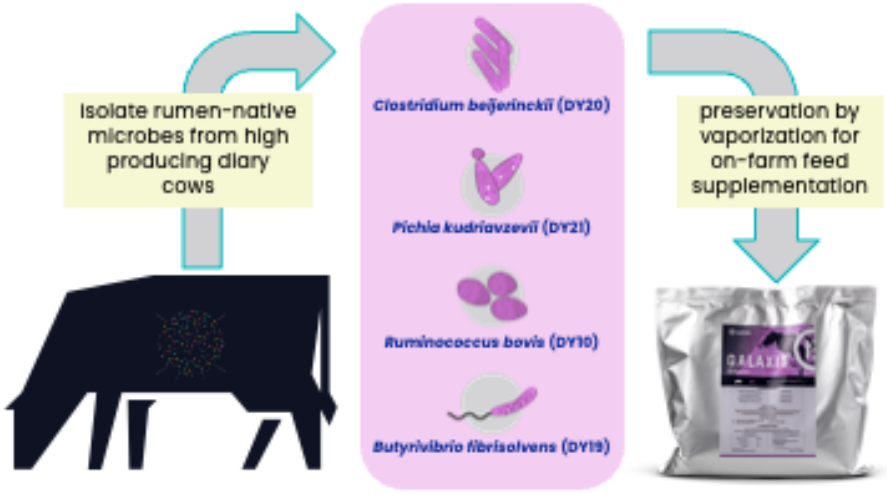

Figure 1. Galaxis Frontier is comprised of four live microorganisms isolated from healthy, high-producing dairy cows, designed to be fed daily through the total mixed ration.

We measured the effects of feeding Holstein cows Galaxis Frontier in six independent academic studies; University of Florida, University of Illinois, Michigan State University, South Dakota State University, Cornell University, and the DairyExperts CRO. A meta-analysis of these trial results shows improved milk yield and feed efficiency in lactating cows, but the mechanism of action and impact on the rumen microbiome remains unclear.

	# cows in trial					measurements			
Trial Site	Control	Galaxis Frontier	Length of trial (weeks)	Avg DIM @start (range)	application	Per-cow milk yield	Per-cow components	intake	Rumen fluid samples (weeks of trial)
DairyExperts	30	30	40	50 (40–60)	Mixing wagon	daily	daily	Individual daily	39
University of Illinois	23	23	20	80 (43–145)	Top-dressed	daily	weekly	Individual daily	0, 6, 11, 20
MSU	30	30	16	90 (70-160)	Top-dressed	daily	weekly	Individual daily	0, 16
University of Florida	39	39	20	61 (31 – 87)	Top-dressed	daily	weekly	Individual daily	0, 10, 18
South Dakota State	29	27	16	-25 (-39)-(-12)	Top-dressed	daily	weekly	Individual weekly	-5, -4, -3, -2, -1, 1, 2, 3, 4, 5, 8, 10, 11, 15
Cornell	74	76	20	105 (47–160)	Mixing wagon	daily	weekly	Pen-level daily	0, 11, 15, 19

Figure 2. Studies included in meta-analysis. In all experiments, animals were matched by parity, days in milk, and baseline milk yield then randomly assigned to either the control group (basal TMR) or treatment group (basal TMR supplemented with 5g/head/day Galaxis Frontier consisting of 4 x 10^7 CFU of *C. beijerinckii*, 1 x 10^9 CFU of *P. kudriavzevii*, 1 x 10^8 CFU of *B. fibrisolvens*, and 1 x 10^8 CFU of *R. bovis*).

To elucidate microbial changes underlying these results, we performed 16S rRNA gene amplicon sequencing on 703 rumen fluid samples collected from 150 cows across the 6 trial sites and compared the rumen microbial composition of cows supplemented daily with GF to control animals not fed GF (352 vs 351 samples, respectively). After quality filtering, our dataset contained approximately 24,239 unique amplicon sequence variants. While sharing similar microbial populations, the cows from each trial had distinct microbial compositions, likely due to differences in geography, diet and genetics.

We used phylofactorization [4] to identify the isometric log-ratio abundances of microbes with a false-discovery rate <0.05 that were consistently increased on GF supplementation across all studies, while controlling for DIM and study. Relevant taxa found to be associated with altered microbiomes in treatment cows included members of the genera *Prevotella* and *Selenomonas,* and a member of the family Lachnospiraceae. These results highlight the ability of native microbe supplementation to alter the rumen microbiome in a consistent way regardless of study variables, including diet.

The microorganisms in Galaxis Frontier are part of the core rumen microbiome and found in nearly all animals. We looked exclusively at control animals to see if microbes that changed abundance after feeding Frontier are associated with greater milk production. These microbes trended towards significance as bioindicators of milk yield controlling for study and days-in-milk. When including treatment cows, these microbes were significantly correlated with milk production ($p<0.05$), suggesting that supplementing TMR with Galaxis Frontier alters the rumen microbiome in a way that supports increased milk production. In particular, the lineage of Lachnospiraceae was positively associated with greater milk production.

This demonstrates that supplementing GF leads to reproducible changes in the rumen microbiome that underly the observed positive production effects. Together, these results illustrate the close association

of microbial composition with milk production, and at least partially explains a mechanism of action for the ability of rumen-native direct fed microbials to improve animal efficiency.

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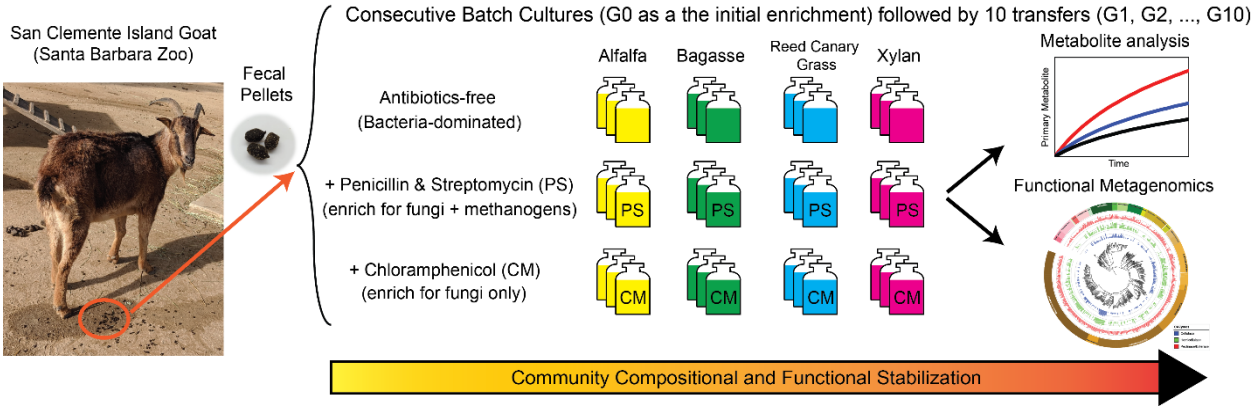
The Biotech Potential of Anaerobic Fungi from Ruminant Herbivores

Michelle A. O'Malley, Department of Chemical Engineering, University of California, Santa Barbara, Santa Barbara, CA, United States

Anaerobic fungi are the primary colonizers of biomass within the digestive tract of large herbivores, where they have evolved unique abilities to break down lignin-rich cellulosic biomass through invasive, filamentous growth and the secretion of powerful lignocellulolytic enzymes. Despite these attractive abilities, considerably less genomic and metabolic data exists for gut fungi compared to well-studied anaerobic bacteria and aerobic fungi that hydrolyze cellulose. We have addressed these knowledge gaps by isolating and characterizing a collection of fungi from large herbivores using a combination of 'omics' tools. Hundreds of novel carbohydrate active enzymes (CAZymes) and components of fungal cellulosomes (enzyme complexes) were identified from several strains of anaerobic fungi, which were discovered through a combination of homology modeling and catabolite repression. Many of these CAZymes share high homology with those found in anaerobic bacteria, and likely arose through horizontal gene transfer. Additionally, high-resolution genomic sequences have revealed a rich set of biosynthetic genes across the fungi that likely regulate diverse processes from fungal development and maturation to microbial defense in the rumen microbiome. A wealth of diverse membrane transporters (SWEET, MFS, etc.) were also identified across anaerobic fungal genomes, which were verified to enhance sugar transport activity in the yeast *S. cerevisiae*. To better characterize the role of anaerobic fungi in herbivores, collected fecal samples were challenged by different types of biomass substrates during cultivation to identify important microbial partnerships; 10 billion metagenomic reads spread across 402 enrichment samples tracked biological diversity as the cultures converged to a set of stable microorganisms. 724 genomes were assembled for previously uncultured gut microbes. Surprisingly, consortia dominated by anaerobic fungi generated more than twice the amount of methane compared to prokaryotic consortia, suggesting that fungi accelerate biomass breakdown and methane release in herbivores. Overall, our analysis points to natural compartmentalization between anaerobes as a means to degrade crude biomass in herbivores. Moreover, our work has unmasked a rich repertoire of novel biomass-degrading enzymes, transporters, biosynthetic gene clusters, and a wealth of horizontally transferred genes from anaerobic fungi that can be used for biotechnology and bioprocessing.



Almond Hulls: Chemical Composition and Feeding Value

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INTRODUCTION

Almond (*Prunus dulcis*) belongs to the family Rosaceae that is related to stone fruits including peaches and cherries. Field weight yields of almonds at harvest are 23% meats (nuts), 13% debris, 14% shells, and 50% hulls (EPA 1995). The Almond Board of California reported the fruit weight to be 31% kernels, 20% shells and 49% hulls on an As Is basis and 32% kernels, 20 shells, and 48% hulls on a DM basis (Huang 2018 unpublished). Almond hulls are a byproduct in the production of almond nuts. Almond hulls (AH) are anatomically similar to the fleshy portion of a peach that humans consume, and hulls contain the mesocarp and exocarp (Figure 1). Consequently, almond hulls are high in sugars and a byproduct feedstuff high in nutritive value that is fed to ruminants in various regions of the world.

The story of almond hulls as a byproduct feedstuff in California has an interesting history. In the 1940s, almond hulls were not used as a livestock feed. Instead, the majority were burned while the remainder were plowed under in fields (Cruess 1949). Velasco et al. (1965) stated “As recently as 1948, almond hulls were considered of little or no value, and most of them were burned or otherwise destroyed. Then as a result of work by University of California researchers (1948-1951), hulls were found to have an energy value 65 to 90% of barley”. The early research at the University of California clearly demonstrated the nutritive value of almond hulls for ruminants (Weir, 1951; Velasco et al. 1965). Subsequently, the research of Aguilar et al. (1984) was of paramount importance to expanding the use of almond hulls as a feedstuff for dairy cattle. Almond hulls are now a common, highly valued byproduct feedstuff that are

used in the diets of lactating dairy cows in California. A survey (Castillo et al. 2012) of 40 dairy farms in California found almond hulls to be an ingredient in 39 out of 104 TMR evaluated with an average feeding rate of 1.5 kg/cow/day and a range of feeding from 0.2 to 3.0 kg. A more recent survey (Heguy 2019, unpublished) found that the feeding amount had increased to approximately 2.3 kg (5 pounds) per lactating cow daily. Even though almond hulls are commonplace in diets of lactating cows, there is a paucity of data on the chemical composition and nutritive value of almond hulls. To achieve higher amounts of feeding to lactating dairy cows, more comprehensive information is required on the nutritive value and chemical composition of almond hulls.

California is the world's leading producer of almond nuts with a production of 1.16 billion kg in crop year 2019/2020 (Almond Almanac 2020). Associated with the yield of nuts (kernels) was 1.83 billion kg of hulls and 0.75 billion kg of shells. For the 2019/2020 crop year, almond tree fruit weight was 31% kernels (nuts), 49% hulls, and 20% shells. Almond nut production in the 2019/20 crop year increased by 63% from the 630 million kg produced in 2007. Orchard plantings of almond trees are increasing rapidly. In 2019, there were 1.18 million acres of bearing orchards and 350,000 acres of nonbearing orchards. Almond nut production can be expected to increase dramatically when the nonbearing orchards come into production in the next 5 to 10 years, which will create a large supply of almond hulls for feeding to livestock. The focus of the current research was/is to evaluate the feeding potential of almond hulls.

This paper is a brief review of the past and current work on assessing the nutritive value of almond hulls. The approach will be:

1. Nutrient Composition of Almond Hulls
2. Survey of California Nutritionists on Almond Hull Usage
3. *In Vitro* Assessment of Almond Hulls
4. Feeding Value of Almond Hulls
5. Variation in Composition and Regulatory Issues
6. Current and Future Research
7. Summary

1. Nutrient Composition of Almond Hulls (AH)

The aim was to begin an evaluation of the chemical composition and nutritive value of almond hulls and to investigate differences in the chemical composition of almond hulls as it relates to the contribution of debris that includes shells and sticks. A concern about almond hulls as a feedstuff is the variability in nutrient (chemical) composition. The variability in nutrient composition can be attributed to a number of factors, but of utmost importance are the variety of almond, the harvesting methods, and the age of the orchard (trees). Nutritionists know that Nonpareil almond hulls are superior in nutritional quality to Pollinator (varieties other than Nonpareil) almond hulls. Consequently, Nonpareil almond hulls are preferred for feeding high producing dairy cows. Most almond varieties are not self-pollinating so two or more varieties are planted in an orchard with consideration to when each variety blooms. Nonpareil produces a high quality nut for human consumption. There is a tendency to refer to the “other” varieties as pollinators because often their role is to pollinate the Nonpareil variety in an orchard. That is how the terms “other variety” and “pollinators” are referred to in this paper with the two descriptions being synonymous.

The harvesting of almonds involves the shaking of trees so the fruit falls to the orchard floor where the hulls dry. However, along with the almond fruit that falls in response to tree shaking, there are sticks and leaves that also fall from the trees. Sweepers with brushes put the fruit, along with sticks, into windrows on the orchard floor. Sweepers have blowers to remove some of the leaf material. Next, the harvester picks up the fruit from the orchard floor to be placed into a reservoir cart. The harvester does remove leaf

material, dirt/rocks, and the sticks of large size, but there are considerable amounts of sticks of short length that remain in the fruit. A shuttle cart follows the reservoir cart, and the fruit is transferred to the shuttle cart. This action all happens on the go, in swift motion and precise timing. Once loaded the shuttle cart races off to an elevator at the edge of the orchard where the fruit is transferred to semi-trailer containers that will transport the fruit to an almond huller. The harvester will remove large sticks.

Age of orchard has not been critically evaluated as far as we are aware.
Antidotal information indicates that older trees have more foliage so that the amount of sticks in the almond fruit on the orchard floor is greater than the amount of sticks for younger orchards. A contributing factor with older trees is that the current agronomic practices often do not involve the pruning of trees so the potential for more sticks (debris) contributing to the harvest of almonds may have increased. This debris contributes to the variation in chemical composition or what nutritionists refer to as "variation in quality".

Nutritionists were asked what influenced the decision to include almond hulls in diets. The #1 consideration was price, but consistency was a close 2nd (Heguy 2019; unpublished survey). Frequently overlooked is the publication by Aguilar et al. (1984) that reported the variation in composition of three varieties of almond hulls that were varieties of major production at the time the study was conducted (**Table 1**). Variability was high even for Nonpareil almond hulls, which are viewed as high quality almond hulls because of their hull size and chemical composition (nutritive value).

Table 1. Variation in chemical composition of hulls from three almond varieties (Aguilar et al. 1984).

Item	Merced[1]	Nonpareil[2]	Neplus[3]
Crude fiber, %	14.4	14.3	21.1
range[4]	14.0 – 14.8	12.1 – 16.6	17.4 – 24.9
ADF, %	21.5	27.3	29.9
range	20.6 – 22.5	19.9 – 34.8	24.6 – 35.2
Cellulose, %	13.3	15.5	18.3
range	12.8 – 13.8	12.9 – 18.1	15.9 – 20.7
Lignin, %	7.9	12.1	11.7
range	7.5 – 8.4	7.7 – 16.6	7.9 – 15.6
Soluble sugars, %	26.4	31.7	23.9
range	19.6 – 33.2	20.8 – 33.7	18.5 – 29.4
Crude protein, %	5.4	6.7	6.1
range	4.9 – 5.8	4.7 – 8.8	5.4 – 6.7
Ash	7.3	6.1	7.6
range	7.0 – 7.7	5.2 – 7.0	6.8 – 8.3

[1]Merced: mid to late season variety with papery shell.
[2]Nonpareil: early season variety with thin shell.
[3]Neplus: requires a pollinator, soft shell.
[4]Range refers to the chemical component listed above it.

Our research was an extension of the previous work (Aguilar et al. 1984) conducted at UC Davis. Commercial almond hulls that are available in the market are often a mix of varieties, Nonpareil variety mixed with Pollinator varieties. Commercial almond hulls are a “commodity” that contains debris composed predominately of sticks and shells and are often a mix of Nonpareil and Pollinator varieties. Chemical analysis of commercial almond hulls does not look specifically at the composition of the “hull” because of the debris component. Our approach was to hand-sort commercial almond hulls to separate the hulls from the debris (sticks and shells). This created what we referred to in our research as “Pure” Hulls.

Twelve different samples of almond hulls were obtained. Samples included 5 Nonpareil, 2 Butte/Padre pollinator mixes, 1 Butte/Mission pollinator mix, and 4 pollinators that had no variety designation. For our research, the “Other Variety” designation included the seven samples that were designated by the source supplier as not a Nonpareil variety. Each sample of almond hulls was thoroughly mixed and divided into two samples. One sample was retained for chemical analysis and represented Total almond hulls (TAH) while the other half was hand sorted to separate hulls from debris (sticks and shells) to create samples of Pure almond hulls (PAH) and Debris (wood sticks and shells). Thus, in our research study, TAH represented commercial almond hulls because TAH contained debris. The PAH represented the hull material that is the important fraction with respect to nutritional value of commercial almond hulls. The methods of chemical analysis were described previously (DePeters et al. 2020a).

The proportion of debris in the 5 Nonpareil almond hull samples was 4.7% As-Is basis (S.D. = 3.08) while for the Other Variety, the debris was 6.8% and more variable (S.D. = 4.07) As Is basis. Our results agree with those of Offeman et al. (2014) who sorted 36 samples of Nonpareil and found 4.5% debris. In our study there were two samples of the Monterey variety that averaged 5.1% debris while Offeman et al. (2014) found 7.4% debris in 21 samples. There were also two samples of Butte/Padre mixed varieties with 9.1% average debris. These two samples were numerically lower in debris than the 14.7% for Butte and 13.0% for Padre reported previously (Offeman et al. 2014). Offeman et al. (2014) noted that some varieties, including Butte and Padre, tended to

have a portion of the shell adhering to the hull so it might be assumed that there is likely more debris in the almond hull fraction from these varieties. In more recent work in our research program, in contrast to Offeman et al. (2014), we found that some samples of Nonpareil almond hulls had shell retained to the hull. We wish to acknowledge here that Mr. Dave Phippen (Travaille & Phippen, Inc. Manteca, CA) has and is a valuable resource for us with respect to providing information about all aspects related to the production of almonds. Shell that is closely associated with the hull is often referred to as “stick-tights”. In our hand-sorting process, it was extremely difficult to remove the shell “stick-tights” from the hull of the Nonpareil almond hulls. Why stick-tights occur is unknown, and it seems to vary with season of harvest and variety of almond. For our research, it was an interesting observation that probably deserves further study since “stick-tights” will impact the nutritional value of commercial almond hulls. Overall, our data agree with the literature that Nonpareil are large in size and contained the lowest amount of debris numerically.

Our study did not evaluate growing region in California. Information on the impact of growing region on the chemical composition of almond hulls is lacking and deserves attention. Offeman et al. (2015) evaluated one sample of Nonpareil almond hulls each from two counties in CA as a descriptive measure for their leaching study. The sample from Kern Co. contained 94.39% hulls, 3.94% shell, 1.43% twigs, and 0.24% other material (As Is basis). The sample from Colusa Co. contained 92.46% hulls, 2.52% shell, 2.43% twigs, and 2.59% other material (As Is basis). The almond hulls were from different harvest seasons, and other information on hulling methods and age of orchard

was not provided. The difference in total debris was 5.61 versus 7.54% for Nonpareil. We are unaware of any other data for California with respect to the impact of growing region, variety, harvest and hulling practices, and agronomic practices on debris contribution to almond hulls.

Dry matter content was highest for Debris and lowest for PAH. The sticks and shells were low in moisture content so when these were removed from the TAH, the DM content of PAH decreased for both the Nonpareil (**Table 2**) and the Other Variety (**Table 3**). This difference in moisture content could be important since the legal definition of almond hulls in California states not higher than 15% crude fiber (CF; As Is basis) and not higher than 13% moisture. Moisture impacts the estimate of CF on an As Is basis. The Nonpareil almond hulls were 14.64% CF (DM basis) and 12.7% CF (As Is basis). The proportion of CF As Is basis to CF DM basis was 86.8% for TAH. In contrast, the proportion of CF As Is basis to CF DM basis was 85.2% for PAH. A similar pattern occurred for the Other Variety where the proportion of CF As Is basis to CF DM basis was 88.1% for TAH and 87.4% for PAH.

Table 2. Chemical composition (DM basis) of total almond hulls (TAH), pure almond hulls (PAH), and debris for Nonpareil variety.

	TAH				PAH				Debris			
Chemical Composition, % DM	Avg	SD	Min	Max	Avg	SD	Min	Max	Avg	SD	Min	Max
DM	86.82	1.41	85.60	88.50	85.20	1.91	83.50	87.70	91.54	1.16	90.20	93.00
OM	92.97	0.51	92.29	93.56	92.54	1.21	91.17	93.90	95.68	0.44	95.03	96.18
CP	5.08	1.10	3.80	6.40	5.14	1.14	3.80	6.70	6.94	1.94	4.50	9.00
Soluble Protein	2.12	0.71	1.50	3.10	2.26	0.77	1.40	3.30	1.84	1.05	1.10	3.60
NDF	21.40	1.83	18.50	22.90	19.26	1.19	18.00	21.20	62.28	6.39	55.90	72.30
NDFom	20.98	1.71	18.20	22.40	18.84	1.15	17.50	20.60	60.72	6.49	54.20	71.00
ADF	15.36	1.33	13.50	16.70	13.38	0.76	12.60	14.60	46.44	4.96	41.20	53.70
ADFom	15.04	1.05	13.50	16.00	13.04	0.64	12.60	14.10	45.48	4.69	41.20	52.70
CF	14.64	0.89	13.20	15.40	12.96	1.05	12.10	14.60	44.36	4.83	39.10	52.20
CF (As Is)	12.71	0.77	11.46	13.37	11.04	0.89	10.31	12.44	40.61	4.42	35.79	47.78
Lignin	8.59	0.71	7.64	9.41	7.63	0.70	7.02	8.78	22.38	2.73	19.40	25.81
Ash	7.03	0.51	6.44	7.71	7.46	1.21	6.10	8.83	4.32	0.44	3.82	4.97
EtOH Soluble CHO	32.57	4.00	27.32	36.39	33.56	4.32	28.03	39.88	7.87	3.32	4.80	13.41
Starch	0.32	0.18	0.10	0.60	0.44	0.46	0.00	1.20	0.44	0.29	0.20	0.90
NFC	65.43	3.07	62.10	69.37	67.20	2.79	63.99	70.96	22.14	7.59	14.07	34.12
NSC	32.88	3.86	27.90	36.50	34.00	4.38	28.20	40.40	8.30	3.26	5.70	13.90
TDN	68.56	0.74	67.80	69.80	69.46	1.02	68.60	70.80	49.26	9.71	37.50	63.60
NEL (Mcal/lb)	0.71	0.03	0.68	0.74	0.74	0.01	0.73	0.75	0.47	0.14	0.31	0.68
Ca	0.21	0.03	0.16	0.25	0.19	0.02	0.17	0.21	0.55	0.23	0.26	0.80
P	0.12	0.03	0.09	0.16	0.12	0.03	0.10	0.16	3.09	6.66	0.06	15.00
Mg	0.09	0.02	0.07	0.11	0.09	0.02	0.07	0.11	2.89	6.21	0.07	14.00
K	2.81	0.35	2.42	3.27	2.88	0.34	2.53	3.34	1.19	0.15	0.98	1.36
Na	0.02	0.00	0.02	0.02	0.01	0.01	0.01	0.02	0.02	0.01	0.01	0.04
Fe (PPM)	196.0	21.5	168.0	217.0	209.8	68.1	136.0	282.0	378.8	202.2	252.0	734.0
Mn (PPM)	13.40	2.79	11.00	18.00	12.80	1.30	12.00	15.00	26.60	2.70	23.00	30.00
Zn (PPM)	11.60	1.95	9.00	14.00	10.60	3.78	7.00	16.00	42.40	10.55	28.00	55.00
Cu (PPM)	3.80	1.30	3.00	6.00	2.80	1.30	1.00	4.00	11.60	7.70	5.00	20.00

Abbreviations: SD = standard deviation; DM = Dry matter; OM = organic matter; CP = Crude protein; ADF = Acid detergent fiber; ADFom = Acid detergent fiber on an organic matter basis; NDF = Neutral detergent fiber; NDFom = Neutral detergent fiber on an organic matter basis; CF = crude fiber; EtOH Soluble CHO = ethanol soluble carbohydrates; NFC = non-fiber carbohydrates calculated by: NFC% = 100 – (CP% + Fat% + Ash% + NDF% + NDFICP%); NSC = non-structural carbohydrates calculated by: NSC% = EtOH CHO% + Starch% ; TDN = total digestible nutrients; NEL = net energy of lactation; Ca = calcium; P = phosphorus; Mg = magnesium; K = potassium; Na = sodium; Fe = iron; PPM = parts per million; Mn = manganese; Zn = zinc; Cu = copper.

Table 3. Chemical composition (DM basis) of total almond hulls (TAH), pure almond hulls (PAH), and debris for Other varieties.

	TAH				PAH				Debris			
Chemical Composition, % DM	Avg	SD	Min	Max	Avg	SD	Min	Max	Avg	SD	Min	Max
DM	88.09	1.70	85.60	89.90	87.26	2.10	84.20	90.20	92.47	1.09	91.00	94.00
OM	92.44	1.00	90.94	93.66	91.54	0.89	90.04	92.63	96.37	0.51	95.85	97.10
CP	5.04	1.36	4.00	8.00	4.87	1.44	3.80	8.00	5.39	2.02	3.30	9.60
Soluble Protein	2.06	1.15	0.90	4.40	1.97	1.18	1.00	4.50	1.77	1.71	0.70	5.40
NDF	25.54	3.76	20.40	31.70	22.07	1.33	19.60	23.60	69.23	6.44	59.20	78.10
NDFom	24.90	3.63	20.00	31.30	21.54	1.08	19.50	22.90	68.27	6.10	59.10	77.10
ADF	18.11	3.52	13.60	24.10	15.89	1.55	14.00	18.40	50.54	5.78	43.00	58.20
ADFom	17.84	3.57	13.60	24.10	15.57	1.41	13.60	17.50	50.29	5.60	43.00	57.60
CF	18.10	1.46	15.90	19.70	15.07	1.33	13.30	17.20	49.39	5.57	39.80	54.80
CF (As Is)	15.94	1.29	14.01	17.35	13.15	1.16	11.61	15.01	45.67	5.15	36.80	50.67
Lignin	9.74	2.44	6.94	12.45	8.69	1.88	7.30	12.84	22.70	2.61	17.92	26.22
Ash	7.56	1.00	6.34	9.06	8.46	0.89	7.37	9.96	3.63	0.51	2.90	4.15
EtOH Soluble CHO	27.98	3.43	21.38	31.19	29.49	3.66	23.31	35.01	5.39	2.06	3.70	9.61
Starch	0.26	0.11	0.10	0.40	1.66	3.68	0.20	10.00	0.20	0.14	0.10	0.40
NFC	60.69	3.36	55.55	64.43	63.31	1.94	60.08	65.81	18.33	6.81	6.00	28.36
NSC	28.23	3.47	21.60	31.60	29.71	3.64	23.50	35.20	5.59	2.17	3.80	10.00
TDN	65.80	2.86	62.50	69.60	67.00	1.77	63.60	68.60	44.61	10.73	37.30	66.90
NEL (Mcal/lb)	0.65	0.04	0.61	0.70	0.66	0.02	0.62	0.68	0.43	0.14	0.33	0.74
Ca	0.26	0.04	0.22	0.31	0.24	0.03	0.19	0.29	0.40	0.14	0.21	0.56
P	0.12	0.04	0.07	0.18	0.12	0.04	0.07	0.19	0.09	0.05	0.04	0.19
Mg	0.11	0.01	0.10	0.13	0.11	0.02	0.09	0.13	0.10	0.03	0.06	0.17
K	3.25	0.40	2.83	3.88	3.45	0.40	2.93	3.98	1.08	0.48	0.57	2.00
Na	0.02	0.01	0.01	0.03	0.07	0.15	0.01	0.40	0.01	0.00	0.01	0.02
Fe (PPM)	201.9	70.2	119.0	322.0	229.4	57.3	163.0	304.0	258.4	251.1	118.0	822.0
Mn (PPM)	17.86	5.61	11.00	25.00	17.43	6.11	10.00	26.00	20.86	8.86	10.00	38.00
Zn (PPM)	14.29	4.50	9.00	22.00	13.71	5.77	7.00	24.00	46.00	36.13	17.00	122.00
Cu (PPM)	4.57	1.27	2.00	6.00	3.71	1.11	2.00	5.00	13.71	9.11	4.00	26.00

Abbreviations: SD = standard deviation; DM = Dry matter; OM = organic matter; CP = Crude protein; ADF = Acid detergent fiber; ADFom = Acid detergent fiber on an organic matter basis; NDF = Neutral detergent fiber; NDFom = Neutral detergent fiber on an organic matter basis; CF = crude fiber; EtOH Soluble CHO = ethanol soluble carbohydrates; NFC = non-fiber carbohydrates calculated by: NFC% = 100 – (CP% + Fat% + Ash% + NDF% + NDFICP%); NSC = non-structural carbohydrates calculated by: NSC% = EtOH CHO% + Starch%; TDN = total digestible nutrients; NEL = net energy of lactation; Ca = calcium; P = phosphorus; Mg = magnesium; K = potassium; Na = sodium; Fe = iron; PPM = parts per million; Mn = manganese; Zn = zinc; Cu = copper.

The issue of using the chemical component of CF on an As Is basis and not on a DM basis for regulatory purposes should be reassessed. CF As Is may not adequately reflect changes in moisture content with due to weather or storage. For instance, the CF content of a sample of almond hulls obtained from the outside of a pile may differ from a sample obtained from deep within the pile. In addition, the CF method does not adequately reflect the cellulose, hemicellulose, and lignin composition of the cell wall fraction. A more appropriate chemical method might be neutral-detergent fiber.

The fiber (aNDF, aNDFom, ADF, ADFom, and CF) and lignin compositions followed a pattern similar to DM content with highest content of fiber fractions and lignin in Debris and lowest content of fiber fractions and lignin in PAH. The TAH was intermediate but only slightly higher in fiber and lignin content than PAH.

There was large variation in fiber composition for our study. A summary from the literature (**Table 4**) also showed that the fiber composition of almond hulls was quite variable.

Table 4. Chemical composition of almond hulls reported in the literature. All values are on a DM basis.

	1	2	3	4a	4b	4c	4d	5a	5b	6	7	8	9	10	11
NDF	28.0	-	62.0	27.5	31.3	27.1	26.7	36.2	34.7	-	37.1	33.9	33.7	21.1	16.0
ADF	28.8	-	30.4	25.7	21.7	17.9	18.6	24.0	23.8	29.2	24.3	28.7	26.2	13.7	22.3
CF	-	10.6	-	13.2	13.3	13.3	12.2	-	-	15.1	-	-	-	13.5	-
CP	2.7	4.1	10.3	6.0	6.6	6.7	5.7	2.7	2.2	62.0	2.9	6.0	5.7	7.0	2.9
Ash	6.1	6.1	9.9	-	-	-	-	7.7	5.3	7.4	6.5	7.1	5.0	12.0	9.0

Lignin	7.1	-	-	10.6	7.9	6.6	6.3	11.2	10.6	11.9	11.8	12.4	10.2	4.1	11.4
Sugars	26.6	26.6	14.1	-	-	-	-	-	-	30.2	-	-	-	-	56.9
Crude Fat	3.3	3.3	2.7	2.4	3.3	2.7	-	1.6	3.4	-	1.7	3.6	2.4	2.5	-
Pectins	4.0	-	-	-	-	-	-	-	-	-	-	-	-	-	-

1. Saura-Calixto et al. 1983
2. Saura-Calixto and Cañellas 1982
3. Elahi et al. 2017
4. Alibes et al. 1983
    a-d: 1976-1979 samples
5. Yalchi and Kargar 2010
    a. Stone shell variety (hard shell)
    b. Paper shell variety (soft shell)
6. Aguilar et al. 1984
7. Yalchi 2011
8. DePeters et al. 2000
9. Arosemena et al. 1995
10. Norollahi et al. 2006
11. Jafari et al. 2011

In California commercial almond hulls are often a mix of Nonpareil and Other Varieties (Pollinator varieties). Nonpareil TAH contained 15.04% ADFom (S.D. 1.05; Range 13.5 to 16.0%). The variation in ADFom was much larger numerically for Other Variety with a S.D. of 3.57 for an average content of 17.84%. Average CF content for Nonpareil TAH was 14.64% (S.D. = 0.89) while the CF for Other Variety was higher numerically (Avg. = 18.1%) and more variable (S.D. = 1.46). The higher fiber content of Other Variety was likely related to smaller hull size (lower weight contribution to the total sample) relative to the debris, but also it could be related to shell adhering to the almond hulls.

Aguilar et al. (1984) studied three varieties of almond hulls and found considerable variation in chemical composition within each variety. In their work, Nonpareil contained on average 27.3% ADF (Range: 19.9 to 34.8%) while Neplus averaged 29.9% ADF (Range: 24.6 to 35.2%). Variation in chemical composition was also noted for almond hulls of different varieties in Iran (Jafari et al. 2011; Jafari et al. 2015).

In a study of byproduct feedstuffs common to California (DePeters et al. 2000), almond hulls that were collected at various hullers contained on average 33.9% NDF (S.D. = 4.5) and 28.7% ADF (S.D. = 4.2). In that study, variety was not considered. In addition, harvesting practices and hulling methods have changed in recent years to remove more debris from almond hulls (Dave Phippen, personal communication). Indeed, the fiber content of almond hulls was lower in our current study compared with DePeters et al. (2000). Nonpareil contained 21.4% aNDF (S.D. = 1.82) and the Other Variety contained 25.5% aNDF (S.D. = 3.76) in the current study.

Nonpareil TAH contained 21.4% aNDF (S.D. = 1.83) and 21.0% aNDFom (S.D. = 1.71). Both average content and variation of aNDF were higher for the Other Variety with 25.5% aNDF (S.D. = 3.76) and 24.9% aNDFom (S.D. = 3.63) compared with Nonpareil. A similar pattern was observed for ADF. Nonpareil TAH contained 15.4% ADF (S.D. = 1.33) and 15.0% ADFom (S.D. = 1.05). The fiber content and variation were higher for the Other Variety with 18.1% ADF (S.D. = 3.52) and 17.8% aNDFom (S.D. = 3.57).

Almond hulls were low in CP content. The CP content of TAH was similar Nonpareil compared with Other Variety. Removing debris did little to change the CP of PAH although the difference was larger for the Other Variety where debris accounted for a larger proportion of the TAH weight. Average CP of almond hulls was previously reported to be 6.0% (DePeters et al. 2000) for California. However, Elahi et al. (2017) reported 10.3% CP while Saura-Calixto et al. (1983) reported 2.7% CP and Saura-

Calixto and Cañellas (1982) reported 4.11% CP for almond hulls in different growing regions of the world.

Ethanol-soluble carbohydrates (EtOHSC) were lower in Debris compare with both TAH and PAH. Sequeira and Lew (1970) analyzed two samples of AH of unknown variety, and reported that almond hulls contained 31.5% total carbohydrates. The predominant sugars found were glucose (10.4%), fructose (8.8%), and sucrose (5.25%). Holtman et al. (2015) reported that Nonpareil almond hulls (not sorted) contained 37.3% fermentable and 9.0% nonfermentable sugars in close agreement to Nonpareil almond hulls (sorted) in the study of Offeman et al. (2014) that contained 32.7% fermentable sugars and 9.3% nonfermentable sugars. However, Nonpareil almond hulls contained 6.1% glucose, 5.9% fructose, and 1.9% sucrose in Holtman et al. (2015) compared with 15.8% glucose, 13.0% fructose, and 3.9% sucrose in Offeman et al. (2014).

Pectins were not measured in our research. Offeman et al. (2014) reported that about 60% of the DM in almond hulls was extracted with water while total sugars across varieties ranged from a low of 30.6% for Fritz to a high of 42.0% for Nonpareil. The 60% water extracted material agrees with the 55 to 62% reported by Jafari et al. (2015) and the 58.8 to 63.9% reported by Offeman et al. (2015). Offeman et al. (2014) suggested that the difference between total water soluble content and total sugar content represented other water-soluble components including pectins, gums, tannins, and ash. Saura-Calixto et al. (1983) reported 3.98% total pectins, 0.09% gums, and 6.02% polyphenols measured as D-catechin while Jafari et al. (2015) reported 2.32 to 2.84%

tannins and 3.2 to 3.57% total phenolic compounds. Holtman et al. (2015) reported AH contained 3.5% ash and 2.1% soluble ash. Using an average value for tannins and phenolics from Jafari et al. (2015), the total would be 18.07% for other water-soluble components, which agrees with Nonpareil (60% – 42% = 18%) for Offeman et al. (2014).

Variety plays a major role in the chemical composition of commercial almond hulls. In an orchard, two or more varieties are planted in alternating rows for pollination (**Figure 1**) since most varieties are not self-pollinating.

Figure 1. Almond orchard with different varieties.

Each variety is harvested separately because both the quality of the nut and the harvest time of each variety differ. Nonpareil has a large size hull so as a proportion of the total sample weight, the debris (shells and sticks) can be a lower proportion of the almond hull weight compared with a pollinator variety that typically has a smaller hull size. Another complicating factor is that the shell type also differs with variety with soft shell and hard shell almond varieties. The proportion of sticks and shells in the hull product of pollinators can be high compared with Nonpareil almond hulls. Even though Nonpareil hulls are the highest quality based on fiber and sugar composition, many hullers blend the hulls from different varieties to create commercial almond hulls that meet the California legal definition of almond hulls of 15% CF or less on an As Is basis. This blending of almond hull varieties tends to minimize the importance of understanding what factors impact the chemical composition and nutritive value of commercially available almond hulls.

The most complete comparison of composition (**Table 5**) with respect to variety of almond that we are aware of was conducted by the Almond Board of California (Huang 2018 unpublished). Total sugar content varied from a low of 13.4% for Aldrich to a high of 32.2% for Independence. Nonpareil and Independence were both greater than 30% total sugar, but the total sugar content was more variable for Nonpareil than Independence. Crude protein content was low for all varieties with the exception of Wood Colony at 9.7%. Moisture content was low for all varieties except for Padre (15.9%) and Carmel (14.8%), which were above the CDFA definition of not more than 13% moisture. The variability in moisture content was also high for Carmel. For NDF

content only Independence averaged below 20% NDF. Acid detergent lignin ranged from a low of 2.8% for Price to a high of 5.3% for Fritz. Potassium varied with variety, but calcium, magnesium, and phosphorus varied little with variety.

Table 5. Composition of California Almond Hulls by Variety- G. Huang, Associate Director, Food Research and Technology, ABC unpublished data, 2018

Composition of Clean Almond Hulls by Variety on Dry Matter Basis (%)

Analyte	Aldrich	Butte	Butte/ Padre	Carmel	Fritz	Independence	Mission
Moisture	12.6±9.9	11.3±4.8	10.1±2.1	14.8±10.6	11.8±6.0	8.2±3.4	11.7±8.5
Protein	6.4±1.5	4.6±1.4	5.0±1.1	7.0±1.3	5.0±1.1	5.5±0.5	4.2±0.9
Fat	2.2±0.6	2.4±0.4	2.5±1.0	2.0±0.7	2.1±0.6	1.9±0.1	2.3±0.7
Ash	10.8±1.4	10.5±2.5	8.5±1.1	11.0±1.3	9.4±1.2	10.1±1.8	10.9±1.6
Fructose	3.8±1.0	6.4±1.8	7.8±0.8	3.7±0.7	7.2±2.0	6.3±1.0	4.8±0.8
Glucose	6.4±1.5	10.6±2.7	12.1±1.6	6.6±2.4	9.1±1.9	14.6±2.7	7.0±2.2
Sucrose	3.4±2.2	3.1±1.9	3.4±1.1	0.9±0.8	2.4±1.3	11.3±1.6	1.9±1.3
Total Sugar	13.4±4.3	20.1±4.7	23.3±1.7	10.8±3.9	18.6±3.4	32.2±1.4	13.5±4.2
NDF	28.1±1.8	26.1±2.7	23.9±2.1	28.9±3.1	25.7±2.3	19.2±1.4	26.3±2.3
ADF Seq	19.0±1.7	18.1±2.0	17.3±1.2	19.9±2.1	16.9±2.8	14.1±0.5	17.9±1.4
AD Lignin	4.2±0.8	4.1±0.9	3.6±0.4	4.7±0.7	5.3±1.1	3.0±0.5	4.4±0.5
Potassium	4.1±0.5	3.7±0.9	3.1±0.4	4.0±0.3	3.3±0.3	3.1±0.4	3.8±0.1
Calcium	0.3±0.0	0.2±0.0	0.2±0.0	0.3±0.0	0.2±0.0	0.2±0.0	0.3±0.1

Magnesium	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.2±0.0
Phosphorus	0.2±0.0	0.1±0.1	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.1	0.1±0.0

Composition of Clean Almond Hulls by Variety on Dry Matter Basis (%) continued

Analyte	Monterey	Nonpareil	Padre	Price	Sonora	Wood Colony	Average
Moisture	11.5±10.2	8.3±3.1	15.9±6.5	12.8±6.0	10.9±7.2	12.4±7.9	11.6±6.6
Protein	5.1±0.7	4.0±0.8	4.4±1.8	5.5±1.2	7.3±2.7	9.7±3.6	5.6±2.2
Fat	2.5±0.9	2.0±1.0	2.0±1.0	2.2±1.1	1.5±0.6	2.1±0.7	2.2±0.7
Ash	10.8±1.8	8.1±2.0	9.7±2.7	9.5±0.8	9.6±0.8	12.3±3.3	10.1±2.1
Fructose	5.8±1.3	8.5±1.0	8.5±1.4	8.6±1.5	7.1±0.8	5.4±2.0	6.5±2.0
Glucose	8.5±1.8	16.9±2.9	13.4±3.8	13.2±2.0	10.6±2.4	6.1±2.5	10.5±4.0
Sucrose	2.6±1.4	5.1±1.5	4.6±3.0	3.1±1.9	5.4±2.4	3.8±1.7	3.8±2.7
Total Sugar	16.9±3.8	30.5±4.3	26.3±6.8	24.8±2.3	23.1±3.6	15.3±5.2	20.7±7.3
NDF	26.6±2.6	21.2±3.3	22.0±2.6	20.9±1.2	23.6±1.6	26.2±3.5	24.6±3.6
ADF Seq	16.9±4.2	15.1±2.1	15.8±1.5	15.0±1.2	16.0±1.2	17.9±2.3	17.0±2.5
AD Lignin	4.2±0.7	3.1±0.6	3.4±0.8	2.8±0.1	3.1±0.4	3.6±1.1	3.8±1.0
Potassium	3.8±0.7	2.6±0.6	4.1±1.1	3.0±0.4	3.4±0.6	4.2±1.1	3.6±0.8
Calcium	0.2±0.0	0.2±0.0	0.3±0.1	0.2±0.0	0.2±0.0	0.3±0.1	0.2±0.1
Magnesium	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0
Phosphorus	0.1±0.1	0.1±0.0	0.1±0.1	0.1±0.0	0.1±0.1	0.2±0.1	0.1±0.1

The predominate sugar (**Table 6**) in almond hulls was glucose at approximately 50% of the total sugar content (Huang 2018), which agrees with Sequeira and Lew (1970). Fructose and sucrose were found in lower concentrations.

Table 6. Composition of Almond Hulls by Types- G. Huang, Associate Director, Food Research and Technology, ABC unpublished data, 2018

Composition of Almond Hulls (%, 2017)

Analyte	California	Hardshell	Nonpareil	Average
Moisture	13.0±3.3	10.3±1.8	10.1±4.0	10.9±3.2
Protein	6.0±2.2	3.7±0.8	4.9±0.9	4.7±1.5
Fat	2.6±0.8	2.0±0.5	2.6±1.0	2.4±0.8
Ash	8.7±2.3	8.6±2.3	7.1±1.6	8.1±2.1
Fructose	6.9±1.4	7.2±2.3	8.3±1.3	7.5±1.8
Glucose	11.3±2.0	11.5±4.6	15.8±2.8	13.1±3.9
Sucrose	4.1±1.3	3.5±1.7	5.6±2.5	4.4±2.1
Total Sugar	22.2±3.7	22.3±8.2	29.7±3.6	25.1±6.6
NDF	31.3±6.5	34.3±14.2	22.4±2.6	29.1±10.4
ADF Seq	21.1±5.1	24.6±10.0	15.6±2.3	20.3±7.5
AD Lignin	5.1±1.7	6.6±3.5	3.4±0.9	5.0±2.7
Potassium	3.0±0.8	2.5±0.7	2.3±0.4	2.6±0.7
Calcium	0.2±0.0	0.2±0.1	0.2±0.0	0.2±0.0
Magnesium	0.1±0.0	0.1±0.0	0.1±0.0	0.1±0.0
Phosphorus	0.0±0.1	0.1±0.0	0.1±0.1	0.1

Take Home Messages: The chemical composition/nutritional value of almond hulls was influenced by the Debris fraction and by the variety of almond. Reducing the proportion of Debris in almond hulls decreased the fiber and ash content. Nonpareil hulls were superior in quality as measured by higher sugar content and lower content of ash, lignin, and NDF in the hulls compared with Other Varieties. Almond

hulls are an excellent source of readily available carbohydrates (sugars) in the diet of ruminants.

2. Survey of California Nutritionists on Almond Hull Usage

Members of the California Chapter of the American Registry of Professional Animal Scientists (ARPAS) were surveyed on almond hull usage practices. In February 2019, an electronic survey was emailed to the entire California ARPAS membership list. Forty-two surveys were returned by 40 nutritionists and two feed suppliers.

In the previous five years (2014 – 2018), almond hull usage increased (41%) or remained the same (44%), while only 15% of respondents reported decreased usage. Average feeding rates for lactating cows across herds and almond hull feeding levels in nutritionists' highest almond hull fed herds are presented in **Table 7**. The reported average feeding rate depicts an increase from a previous California survey (Castillo et al. 2012) that reported an average feeding rate of 1.5 kg/lcow/day, with a range of 0.2 to 3.0 kg.

Table 7. Average and maximum almond hull feeding rates (kg/lactating cow/day) in California lactating rations.

	Average	Range
Average feeding rate	2.3 kg	0.5 – 4.5 kg
Maximum feeding rate	4.6 kg	0.9 – 8.2 kg

Table 8 describes almond hull utilization in lactating cow, dry cow, and heifer growing rations. Sixty-two percent of respondents said that changes in the price of almond hulls affected how the hulls were utilized in ration formulations, and was mostly dependent on the price of almond hulls compared with forage/silage prices. Price, consistency, mold,

and quality were variables that respondents felt they were “very responsive” to addressing when including almond hulls in rations (**Table 9**).

Table 8. Almond hull utilization in California dairy rations.

	Forage	Concentrate	Forage & Concentrate
Lactating Cow Ration	30%	0%	70%
Dry Cow Ration	31%	7%	62%
Heifer Growing Ration	29%	9%	62%

Table 9. Responsiveness of inclusion of almond hulls in diets related to different variables.

	Very	Somewhat	Not
Price (n=38)	32	6	0
Consistency (n=38)	30	7	1
Mold (n=35)	29	5	1
Quality (n=37)	27	9	1
Crude Fiber Levels (n=36)	15	16	5
ADF (n=35)	15	16	4
Ash (n=34)	14	16	4
Sugar (n=36)	13	19	3
NDF (n=36)	11	21	3

Other results of potential interest include 79% of respondents tested almond hulls for chemical composition. Frequency of lab testing varied between monthly and yearly, or when there was reason for concern. Most nutritionists reported concerns when including almond hulls in lactating cow rations (66%) and in dry cow/heifer growing rations (70%).

Quality issues were a top concern for feeding almond hulls, and most concerns related to the amount of stick and shell that impact the nutritional quality of the hulls.

Take Home Messages: Almond hull feeding is an important topic in California as rising almond orchard acreage increases hull availability for feeding to dairy cattle and other livestock. The topic of byproduct feeding will become increasingly important as decreased water availability impacts forage production in the State. Given the large range in reported feeding rates, results from this survey suggest there may be opportunity to increase almond hull inclusion rates in California dairy rations.

3. *In Vitro* Assessment of Almond Hulls

The aim of this study was to determine the *in vitro* digestibility and *in sacco* disappearance of dry matter (DM) and neutral detergent fiber (NDF) in total almond hulls (TAH), pure almond hulls (PAH), or Debris. The TAH were used because there are no data on the effect of debris (non-hull material) on the nutritional value of almond hulls. The hulling process yields commercial AH (< 15.0% crude fiber) that are predominately hulls, with the amount of debris (sticks and shells) varying, at least, for the variety of almond (DePeters et al., 2020a). We know from previous research (DePeters et al., 2020a) that the contribution of debris impacts the chemical composition of AH by increasing the fiber, lignin, and ash content.

The few in vitro studies with AH did not adequately describe the AH used (Arosmena et al., 1995; Jafari et al., 2011; Elahi et al., 2017). In the case of Arosemena et al. (1995), the AH were commercial AH and therefore contained debris. Based on the lignin and fiber content, it is unlikely that the AH used in other studies (Jafari et al., 2011; Jafari et al., 2015; Elahi et al., 2017) were pure AH. To the best of our knowledge, there are no reports in the literature for the in vitro fermentability of pure AH.

The aim of this study was to investigate the impact of debris on the in vitro and *in sacco* rumen fermentability of AH by evaluating 12 samples of commercial AH (Total almond hulls; TAH) of which a portion of each was hand sorted to create Pure almond hulls (PAH) and Debris (non-hull material).

Twelve different samples of AH were obtained from five hullers throughout California. Samples contained 5 Nonpareil, 2 Butte/Padre pollinator mixes, 1 Butte/Mission pollinator mix, and 4 pollinators that had no variety designation. Each huller supplied a sample of Nonpareil hulls as well as 1-2 samples of “other” varieties. Samples were designated either Nonpareil or Other Variety. Each sample of AH was thoroughly mixed and divided into two samples. One sample represented TAH while the other half was hand sorted to separate AH from debris to create samples of PAH and Debris (wood sticks and shells). Three samples from one of the hullers did not have enough Debris to be used for in vitro analysis, so only the PAH and TAH samples were used from that huller. This resulted in 12 PAH samples, 12 TAH samples, and 9 Debris samples.

In vitro gas production was measured by incubating 33 samples using the syringe method (Menke & Steingass, 1988). In addition, total gas produced at 24 h was used to calculate metabolizable energy (ME) values with the equation determined by Melesse et al. (2018). *In vitro* true digestibility on a dry matter (DM) basis (IVTD) and neutral detergent fiber digestibility (NDFD) determinations were carried out using multilayer polyethylene polyester cloth bags in the ANKOM Daisy incubator. Bags were incubated for 12, 24, 48, or 72 h and the ANKOM fiber analyzer was used to determine remaining NDF. More detailed methods and statistical analysis can be found in Swanson et al., 2021b.

In vitro total estimated gas production (**Table 10**) was overall significantly higher for PAH (270 ml/g) compared with both TAH and Debris (261 and 79 ml/g respectively).

The rate of gas production was significantly higher for PAH and TAH (0.10098 and 0.101 /h respectively) compared with Debris (0.074 /h), but there was no difference between PAH and TAH. Estimated gas production was significantly greater for PAH (283 ml/g) than TAH (267 ml/g) for Nonpareil but the difference was not significant for the Other Variety. As anticipated, total estimated gas production was significantly lower for Debris (94 ml/g Nonpareil and 69 ml/g Other) compared with both PAH and TAH for both varieties. Estimated rate of gas production was similar for PAH and TAH for both varieties but was significantly lower for Debris compared with TAH. There was a greater numerical difference between Nonpareil and Other Variety for PAH (283 ml/g and 261 ml/g, respectively) than for TAH (267 ml/g and 257 ml/g, respectively). A similar pattern was observed with the estimated rate of gas production, with the Nonpareil Debris having a numerically greater estimated rate (0.0989 /h) than Other Variety (0.061 /h).

Table 10. Estimated potential gas production (ml/g) of almond hulls (AH) for each Type (Total AH, Pure AH, Debris) and Variety (Nonpareil or Other). The effects of Type (Total AH, Pure AH, Debris) on parameters of the gas production function are shown. The estimate is the asymptote or total volumes (ml/g) of gas produced for each Type and Variety from the model. The corresponding rate constants for each Type and Variety are expressed as /h.

					P-value[1]		
	Total AH	Pure AH	Debris	S.E.M.	Total AH vs Pure AH	Total AH vs Debris	Pure AH vs Debris
Asymptote (ml/g)							

Nonpareil	267	283	94	3.3	0.006	<0.001	<0.001
Other	257	261	69		0.463	<0.001	<0.001
Type Avg	261	270	79	3.2	0.009	<0.001	<0.001
Rate Constant (/h)							
Nonpareil	0.11	0.10	0.09	0.003	0.627	0.007	0.067
Other	0.10	0.10	0.06		0.796	<0.001	<0.001
Type Avg	0.10	0.10	0.07	0.003	0.320	<0.001	<0.001

The contribution of debris was reflected in the significantly lower amount of estimated total gas produced for TAH compared with PAH even though the overall estimated rate was not significantly different. Jafari et al. (2011) evaluated the impact of AH variety on in vitro rumen fermentation. Total gas produced (ml/g DM), rate of gas production (ml/h), and organic matter coefficient of digestibility differed by variety as seen with Rabei (79.5, 0.13, 0.823), Mamaei (78.9, 0.13, 0.815), Shahroud15 (63.1, 0.11, 0.68), and Shokoufe (70.1, 0.12, 0.715) respectively. Similar to the results in this study, the Rabei variety that had the greatest gas production also had the highest non-fiber carbohydrates (NFC) and lowest acid detergent lignin (ADL) concentrations (Jafari et al., 2011; DePeters et al., 2020a). The Nonpareil variety in our study also had numerically the highest estimated amount and rate of gas production along with greater NFC and lower lignin content for all types when compared with the Other Variety (DePeters et al., 2020a). Offeman et al. (2014) also found that Nonpareil AH had the highest fermentable sugar content when compared with other varieties grown in California. Rumen microorganisms are able to easily break down and ferment NFC, while lignin is mostly undegradable, so greater NFC content could lead to an overall increase in fermentation and improved digestibility (Nocek and Russell, 1988).

The calculated metabolizable energy (ME) concentration was numerically greater for PAH (9.3 MJ/kg and 8.7 MJ/kg) than TAH (9.0 MJ/kg and 8.5 MJ/kg) while both were significantly greater than Debris at approximately half the energy content (4.7 MJ/kg and 3.8 MJ/kg) for both Nonpareil and Other Variety respectively. There were small numerical differences within Variety between TAH and PAH, but over all Types, Nonpareil had significantly higher calculated ME content compared with the Other Variety. The Other Variety had a greater numerical proportion of Debris (6.8%) compared with Nonpareil (4.7%) (DePeters et al., 2020a).

The lower ME concentration of Debris contributed to the numerically lower energy content of TAH compared with PAH. The larger difference in estimated ME content for Nonpareil compared with the Other Variety was likely due to the differences in aNDF, lignin, ash, and NFC content. As reported previously (DePeters et al., 2020a), Nonpareil TAH contained 21.4% aNDF, 8.6% lignin, 7.0% ash, and 65.4% NFC compared to the Other Variety TAH that were 25.5% aNDF, 9.7% lignin, 7.6% ash, and 60.7% NFC. Similar trends were observed for PAH and Debris for Nonpareil and Other Varieties (DePeters et al., 2020a). These differences in composition would account for the lower ME concentration of Other Variety compared with Nonpareil across all Types.

The IVTD and NDF digestibility were measured at 12, 24, 48, and 72 h (**Table 11**). The IVTD and NDFD were significantly greater for PAH than TAH at 48 and 72 h, and Debris

was significantly lower in digestibility than both TAH and PAH for IVTD at every time point and for NDFD at 24, 48, and 72h.

Table 11. Daisy *in vitro* true digestibility on a dry matter (DM) basis (IVTD) and neutral detergent fiber digestibility (NDFD) of almond hulls (AH). The effects of Type (Total AH, Pure AH, Debris) on IVTD and NDFD coefficients at each timepoint measured *in vitro* are shown.

					P-value[1]		
	Total AH	Pure AH	Debris	S.E.M.	Total AH vs Pure AH	Total AH vs Debris	Pure AH vs Debris
Daisy *in vitro* True Digestibility on DM basis coefficient							
12hr	0.79	0.81	0.36	0.010	0.088	<0.001	<0.001
24hr	0.84	0.87	0.37		0.003	<0.001	<0.001
48hr	0.87	0.91	0.40		<0.001	<0.001	<0.001
72hr	0.88	0.92	0.42		<0.001	<0.001	<0.001
Daisy NDFD coefficient							
12hr	0.14	0.11	0.06	0.021	0.244	<0.001	0.072
24hr	0.32	0.36	0.08		0.148	<0.001	<0.001
48hr	0.46	0.57	0.11		<0.001	<0.001	<0.001
72hr	0.51	0.61	0.13		<0.001	<0.001	<0.001

[1]Pure vs Total = contrast between Pure almond hulls and Total almond hulls; Debris vs Total = contrast between Debris and Total almond hulls; Pure vs Debris = contrast between Pure almond hulls and Debris samples.
Total AH = contains AH and Debris; Pure AH = sorted to contain only hulls; Debris = sticks and shells sorted from TAH.

The lower IVTD and NDFD for Debris contributed to the lower digestibility of TAH compared with PAH at 24, 48, and 72h for IVTD and 48 and 72h for NDFD. The digestibility of aNDF for PAH compared with TAH at 12 h of does not agree with

changes in IVTD, but could be linked to the greater amount of aNDF due to the presence of sticks and shells in TAH compared with PAH. Lignin, a fraction of aNDF, was measured as ADL in this study. The ADL does not include soluble lignin, unlike Klason lignin, which is usually measured at greater amounts than ADL (Hatfield et al., 1994). Queirós et al. (2020) found that almond shells had between 5-9% soluble lignin and 27.9-30.5% Klason lignin. Hall (2000) reported that AH have 16.9% soluble fiber, which would include soluble lignin. It is possible that some of the greater aNDF (24.3% TAH; 20.9% PAH) observed in TAH in this study was soluble lignin, which quickly solubilized within the first 12 hours of incubation. This would lead to a deceptively high aNDF digestibility amount at 12 hours for TAH compared with PAH. At this time more research still needs to be done on the type and amount of lignin in AH.

In addition to the *in vitro* digestibility measurements, two nonlactating, nonpregnant, rumen cannulated Holstein cows were used to measure *in sacco* dry matter and NDF digestibility. The TAH and PAH samples were weighed into monofilament nylon bags that were heat sealed before being placed in the rumen of the cannulated cows. Two series of *in sacco* incubations were conducted with bags of TAH or PAH incubated in the rumen for 0, 1, 2, 4, 8, 16, 32, and 64 h as described by Nocek (1988). Bags were then analyzed for DM and NDF disappearance. A non-linear mixed effects model was used to analyze the rate and extent of digestibility for both DM and NDF of the TAH and PAH.

The estimated asymptote for the coefficient of DM disappearance was significantly greater for PAH (0.3547) and TAH (0.3435; **Table 12**). The estimated fractional rate of *in sacco* DM disappearance was 0.064 /h for TAH and 0.0768 /h for PAH, which was not significantly different. The calculated coefficient for the proportion of DM disappearance (P) was numerically higher for PAH (0.9325) than TAH (0.8985). A similar response was observed for estimated potential disappearance of NDF (Table 12). The estimated asymptote for the proportion of NDF disappearance was 0.80796 for TAH and 0.892 for PAH, which was significantly different. The estimated rate of NDF disappearance was also significantly greater for PAH (0.060 /h) compared with TAH (0.052 /h). The calculated coefficient for the proportion of NDF disappearance (P) was numerically higher for PAH (0.7439) than TAH (0.6659).

Table 12. *In sacco* dry matter (DM) and neutral detergent fiber (NDF) disappearance for each Type (Pure almond hulls (AH) or Total AH). Effects of Type (Pure AH or Total AH) on coefficients of the *in sacco* disappearance function. The estimates were determined from the model. A are the estimated asymptote for the coefficient of disappearance for each Type. k are the corresponding rate constants (/h) for each Type. Int are the corresponding intercepts of time 0 with the coefficient of disappearance on the y-axis for each Type. P are the estimated Asymptote (A) + Intercept (Int) or total estimated proportion of disappearance for each Type.

			P-value[1]
Variable	Estimate	S.E.M.	Pure AH vs Total AH
DM Disappearance			
A- Pure AH	0.35	0.008	0.018

A- Total AH	0.34		
k- Pure AH	0.07	0.002	0.195
k- Total AH	0.06		
Int- Pure AH	0.58	0.008	<0.001
Int- Total AH	0.55		
P- Pure AH[2]	0.93		
P- Total AH[2]	0.89		
NDF Disappearance			
A- Pure AH	0.89	0.032	<0.001
A- Total AH	0.80		
k- Pure AH	0.06	0.003	0.007
k- Total AH	0.05		
Int- Pure AH	-0.15	0.023	0.071
Int- Total AH	-0.14		
P- Pure AH[2]	0.74		
P- Total AH[2]	0.66		

[1] Pure vs Total = contrast between Pure almond hulls and Total almond hulls.
[2]These are calculated values: A + Int = P (proportion of disappearance).
Total AH = contains AH and Debris; Pure AH = sorted to contain only hulls.

Yalchi and Kargar (2010) compared stone shell and paper shell (similar to soft shell of Nonpareil and hard shell of Other Variety) AH in the rumens of four sheep. Degradation rate of DM for stone shell (0.067 /h) and paper shell (0.063 /h) differed. Proportion of degradation of DM was also greater for stone AH (0.81) compared with paper AH (0.77). Degradation rate of NDF was 0.054 and 0.046 /h and degradation coefficients were 0.56 and 0.52 for stone shell and paper shell AH, respectively. Yalchi (2011) evaluated PAH in the rumen of three sheep at seven time points ranging from 2 to 96 h. Digestibility coefficients of DM for PAH were 0.47 at 2 h and 0.77 at 96 h compared with 0.24 at 2 h and 0.67 at 96 h for alfalfa hay. Interestingly, the digestibility of NDF in PAH was lower than in alfalfa hay at all times points except for the 96 h time point. The perception is that the fiber fraction of AH is highly digestible. However, the findings of Yalchi (2011) question this view. In fact, earlier work by DePeters et al. (1997) reported

that for three samples of AH, the proportion of NDF remaining after 72 h of in situ digestion averaged 0.14, for a digestibility coefficient of 0.86. In contrast, the proportion of NDF remaining for beet pulp was 0.036 and 0.042 for soy hulls. The NDF digestibility of AH deserves further study

Take Home Messages: Overall, Debris was not as digestible as PAH and TAH, and Debris contributed to TAH having significantly lower IVTD and NDF digestibility at 48 and 72h, along with numerically lower calculated ME and significantly lower gas production when compared with PAH. This is important for dairy producers in California who need high quality, digestible feeds to support milk production. Reducing the amount of Debris contamination in commercial AH is one important approach to improving the nutritive value of AH for ruminants and to improving the overall monetary value of the hulls for almond hullers.

4. Feeding Value of Almond Hulls

In vitro rumen fermentation data (Section 3) indicate that digestibility of fiber (NDF) in almond hulls is low (Swanson et al. 2021b), and the fiber in almond hulls may not be as high in digestibility as the fiber in alfalfa hay (Swanson, unpublished). Early research in our lab (DePeters et al. 1997) observed lower fiber digestibility in almond hulls compared with other by-product feedstuffs including soy hulls and citrus pulp measured using an *in sacco* (*in situ*) disappearance study (**Table 13**).

Table 13. Estimated and calculated *in sacco* digestion parameters for DM and NDF from DePeters et al., 1997.

Feed	DM Disappearance	DM kd	NDF Disappearance	NDF kd
Almond hulls	29.2	0.062	16.5	0.043
Soy hulls	87.2	0.047	59.7	0.038
Beet pulp	63.1	0.084	39.1	0.083
Wheat Mill Run	36.7	0.135	19.8	0.122

kd = rate constant for disappearance (h^{-1})

Jafari et al (2015) evaluated almond hulls from four different almond varieties as well as alfalfa and sugar beet pulp for in situ rumen degradability using steers. These researchers found high levels of rumen DM degradability. Extent of ruminal DM degradation of almond hulls ranged from 77.4% to 84.7%. The reason the difference between studies (Jafari et al. 2015 and DePeters et al., 1997) is not apparent.

Norollahi et al. (2006) measured *in vivo* digestibility of almond hulls in sheep. It appears that the diet was 100% almond hulls, but it was not stated as such in the paper. Apparent digestibility was 73.1%, 29.6%, 40.6%, and 84.4% for DM, crude protein, crude fiber, and nitrogen-free extract, respectively. Yalchi (2011) determined in sheep the apparent digestibility of two diets, 100% alfalfa hay (Basal diet) and 70% alfalfa hay/30% almond hulls (Mixed diet). Compared with the 100% alfalfa hay diet, when almond hulls were added to the basal diet (100% alfalfa hay) the apparent digestibility of NDF and CP decreased and there was a tendency for the apparent digestibility of ADF and hemicellulose to also decreased when almond hulls were added to the basal diet (100% alfalfa hay).

The aim of the following study was to evaluate the *in vivo* apparent digestibility of almond hulls. One lot of commercial almond hulls was obtained, which is a limitation of the study. The chemical composition is reported in **Table 14**. Almond hulls (unprocessed) were cubed with alfalfa hay in the following proportions, 0:100, 10:90, 20:80, and 40:60 almond hulls:alfalfa hay (wt:wt As Is basis), to create four experimental diets. The cubes were broken apart by hand. Eight wether sheep were used in a replicated 4 x 4 Latin square design with 4 wethers, 4 periods with each period 14 d in length, and 4 diets. Apparent digestibility was determined using a total feed and fecal collection approach. Each wether was fitted with a fecal harness. Sheep were fed twice daily, and feed intake recorded. Feces were collected and weighed twice daily. A regression approach was used to estimate the digestibility of almond hulls.

Table 14. Chemical composition of almond hulls used in sheep digestibility study. All values are on percentage of DM basis unless otherwise noted.

	Almond hulls
CP	4.4
ADF	21.1
ADFom	20
aNDF	27.3
aNDFom	26.7
Crude Fiber	19.6
Lignin	7.8
EtOH soluble CHO	31
Water soluble CHO	36
Starch	0.2
Ether Extract	1.55
Ash	6.33
TDN	65.9
NEL (Mcal/lb)	0.68
NFC	62
NSC	31.2

Abbreviations: CP = Crude protein; ADF = Acid detergent fiber; ADFom = Acid detergent fiber on an organic matter basis; NDF = Neutral detergent fiber; NDFom = Neutral detergent fiber on an organic matter basis; CF = crude fiber; EtOH Soluble CHO = ethanol soluble carbohydrates; NFC = non-fiber carbohydrates calculated by: NFC% = 100 – (CP% + Fat% + Ash% + NDF% + NDFICP%); NSC = non-structural carbohydrates calculated by: NSC% = EtOH CHO% + Starch% ; TDN = total digestible nutrients; NEL = net energy of lactation

The apparent DM digestibility of almond hulls was 60.9%, significantly lower than the 63.3% for alfalfa hay (**Table 15**). Apparent digestibility of NDFom was 23.5% for almond hulls and 44.4% for alfalfa hay. Apparent digestibility of crude protein was 32.6% for almond hulls and 73.7% for alfalfa hay. This study only evaluated one lot of commercial almond hulls. But, based on this study, the *in vivo* digestibility of fiber (NDFom) and crude protein were low compared with alfalfa a hay. The low fiber (NDFom) digestibility

should be considered when almond hulls are used to replace a portion of the forage, for example corn silage, in the diet of high producing dairy cows.

Table 15. Calculated apparent digestibility of nutrients from almond hulls and alfalfa in sheep.

Apparent Digestibility	Almond Hulls	Alfalfa Hay
DM	60.9	63.3
NDFom	23.5	44.4
ADFom	17.7	45.6
Crude Protein	32.6	73.7

Aguilar et al. (1984) used steers to determine apparent digestibility of three varieties of almond hulls including Nonpareil (NP), Neplus (NE), and a commercial mix (CM). Almond hulls replaced a portion of the control diet (**Table 16**).

Table 16. Diets for digestibility (Aguilar et al 1984)

Ingredient	Control	20% AH	40% AH
Alfalfa	25.0	19.8	14.7
Oat hay	35.0	27.75	20.55
Barley	40.0	31.7	23.50
Almond hulls		20.0	40.00
Urea		0.75	1.25

As the proportion of almond hulls in the diet increased, apparent digestibility of dry matter, ADF, cellulose, and energy decreased (**Table 17**).

Table 17. Apparent digestibility in sheep of various diets and almond hull varieties as reported in Aguilar et al. 1984.

Item	Control	20%CM	40%CM	20%NP	40%NP	20%NE	40%NE
DM	70.3	67.3	65.4	68.7	66.3	69.4	66.4
ADF	51.3	38.9	33.3	40.7	28.3	51.1	40.2
Cellulose	63.4	57.7	54.9	60.4	51.3	66.0	57.3
Energy	69.7	65.8	63.9	67.6	64.6	67.9	64.0

Calculated digestibility of almond hull varieties using regression analysis is reported in **Table 18**.

Table 18. Apparent digestibility and DE concentration of almond hull varieties from Aguilar et al. 1984.

Item	Nonpareil	Neplus	Commercial Mix
DM, %	61.2	62.1	59.6
ADF, %	19.4	23.3	14.8
Energy, %	57.0	56.3	54.5
DE, Mcal/kg	2.52	2.45	2.38

The DE content of the Control diet was 3.08 Mcal/kg. Replacing a portion of the Control diet with commercial almonds reduced the energy concentration to 2.91 Mcal/kg (5.5% reduction) and 2.82 Mcal/kg (8.4% reduction) with 20% and 40% inclusion of commercial almond hulls in the diet, respectively. The decrease in DE concentration with the inclusion of almonds hulls likely reflected the low digestibility of fiber in almond hulls since the soluble sugars should be highly digestible. Aguilar et al. (1984) found that the correlation between ADF (%) and DE (Mcal/kg) was -0.99 while the correlation between soluble sugars (%) and DE (Mcal/kg) was +0.49. Lignin and crude fiber contents were also negatively correlated to DE concentration.

The apparent digestibility of DM for commercial almond hulls (59.6%) observed with steers by Aguilar et al. (1984) agrees closely with our observation (60.9%) with sheep. Likewise, Aguilar et al. (1984) observed fiber (ADF) digestibility of 14.8% while we observed fiber digestibility of 17.7% (ADFom) and 23.5% (NDFom).

There are a few studies with lactating dairy cows. Aguilar et al. (1984) included almond hulls in a TRM for lactating cows as a forage ingredient, replacing alfalfa hay at 12.5 and 25%. The Control diet was 61% alfalfa hay so the TMR was high forage. There was no difference in animal performance. The inclusion of almond hulls with associated fiber content had no impact on any milk component although milk fat % was low across all diets in their study as seen in **Table 19**.

Table 19. Milk composition of cows from the Aguilar et al. 1984 study.

Item	Control	12.5% AH	25% AH
DM Intake, kg/d	19.4	20.1	19.8
Milk, kg/d	25.3	25.5	24.8
Fat, %	3.2	3.2	3.2
Protein, %	3.2	3.2	3.2
SNF, %	8.8	8.8	8.8
Solids, %	12.0	12.0	12.0

More recently Williams et al. (2018) replaced 27.5% of alfalfa cubes in the diet with almond hulls. The average intake of almond hulls was 3.9 kg DM (8.6 pounds) daily. Intake of DM did not differ with diet. However, yields of milk, energy-corrected milk, milk protein, and milk lactose decreased with the feeding of almond hulls (**Table 20**).

Table 20. Feed intake and milk production performance (Williams et al. 2018)[1].

Item	Control	Almond Hull
DM Intake, kg/d	22.3	22.6
Milk yield, kg/d	27.4[a]	24.6[b]
Energy-correct milk yield, kg/d	26.4[a]	24.6[b]
Fat, kg/d	1.04	1.00
Fat, %	3.81	4.14
Protein, kg/d	0.87[a]	0.78[b]
Protein, %	3.22	3.20
Lactose, kg/d	1.36[a]	1.19[b]
Lactose, %	4.99	4.88

[a–b] Means in the same row followed by different superscripts differ significantly ($P < 0.05$).
[1]Control cows consumed 14.2 kg alfalfa cube DM and Almond hull cows consumed 10.5 kg alfalfa cube DM + 3.9 kg almond hull DM.

In 2019, a dairy cattle feeding study was conducted at UC Davis where increasing amounts of almond hulls were added to the TMR to replace the concentrates (Swanson et al., 2021a). As previously stated, almond hulls are low in crude protein but high in fermentable carbohydrates. The highly fermentable sugars, such as sucrose and glucose, in AH could make them a better replacement for concentrates in a lactating cow diet instead of forages that offer more digestible fiber. The aim of this study was to determine if AH could be fed in varying amounts as a replacement for corn and soyhulls in a lactating cow diet to support production performance and digestibility and if there are changes in production with different AH levels substituted for concentrates.

The study used 12 lactating Holstein dairy cows averaging 96 ± 30 days-in-milk that were assigned to dietary treatments using a 4 x 4 Latin square experimental design.

Healthy cows were assigned to their respective Latin square based on parity with 4 primiparous cows to square 1, 4 multiparous second-lactation cows assigned to square 2, and 4 multiparous third-lactation cows assigned to square 3. There were 4 periods, with each period 21 days in length, with the last 7 days of each period used for data collection, with feed offered and milk production recorded twice a day during the study and feed refusals recorded every morning. There were four diets fed where the TMR composition was based on formulating a diet for 28 kg DM intake per cow that would provide cows with 0, 1.8, 3.6, or 5.5 kg/d of commercial AH. This created 4 TMR's with 0, 7, 13, or 20% AH (**Table 21**). As the amount of AH increased in the diet, corn and soyhull pellets decreased while soybean meal increased. Details for the experimental design were reported previously (Swanson et al. 2021a).

Table 21. Composition of total mixed ration for lactating cows.

	0% AH	7% AH	13% AH	20% AH
Ingredient and % of TMR on DM basis				
Alfalfa	38.7	38.7	38.7	38.5
Rolled Corn	32.5	30.1	28.3	23.3
Soy Hulls	11.5	8.0	2.0	0.0
Almond Hulls	0.0	7.0	13.2	20.0
Oat Hay	3.3	2.5	2.5	2.5
Soybean Meal	1.5	1.8	2.9	3.7
DDG	6.2	6.2	6.2	6.2
Cottonseed	3.8	3.8	3.8	3.8
Limestone / Oystershell	1.3	0.3	0.3	0.3
Sodium Bicarb	0.6	0.6	0.6	0.6
Mineral[1]	0.2	0.2	0.2	0.2
Mag ox	0.2	0.2	0.2	0.2
Salt	0.1	0.1	0.1	0.1

[1] Nutrius LLC, Kingsburg, CA Provides to the diet 0.56 % DM Crude Protein, 0.92 % DM ADF, 0.48 % DM NDF, 0.02 Mcal/ lb NE lactation, 1.7 % DM TDN, 12.37 % DM of Calcium, 5.33 % DM Phosphorus, 9.15 % DM Sodium, 0.08% DM Potassium, 4.28 % DM Magnesium, 2.16 % DM Sulphur, 25.06 ppm DM Cobalt, 668.80 ppm DM Copper, 58.54 ppm DM Iodine, 2664.5 ppm DM Manganese, 22.79 ppm DM Selenium, 4473.59 ppm DM Zinc, 1982.07 ppm DM Iron, 933.33 g/Ton of Monensin, 242.68 KIU/ lb of

DM Vitamin A, 84.0 KIU/ lb of DM Vitamin D-3, 1.9 KIU/ lb of DM Vitamin E, 26.67 mg/ lb of Biotin, 0.0103 % DM Lysine, and 0.246 % DM of Methionine, 0.24 % DM Methionine-3, 13.31 mg/ lb EDDI, 0.02 % DM Diflubenzuron, 13.30 lbs Live BCFU's, 1.55 % DM Almond shells, 0.23 % DM Rice Hulls.

The chemical composition of the almond hulls used is shown in **Table 22**. Sampling almond hulls was difficult because of the size of the hulls as well as the distribution and particle size of the debris, which included sticks and shells. The variation in crude protein was small while there was larger variation in the fiber fractions, water soluble carbohydrates, and lignin compositions. The almond hulls were high quality, sometimes referred to in the industry as "prime". The crude fiber on an As Is basis was 12.78%, and crude fiber ranged from a low of 11.88% to a high of 15.06% with sampling.

Table 22. Chemical composition of 4 grab samples of almond hulls added to total mixed-rations in this study.

	Avg	SD	Min	Max
Chemical Composition (% of DM basis unless otherwise noted)				
DM	86.1	2.03	83.6	88.3
CP	4.5	0.24	4.2	4.7
Soluble protein	1.5	0.05	1.5	1.6
aNDF	23.8	2.04	22.2	26.6
aNDFom	23.5	2.08	21.9	26.4
ADF	14.9	2.17	12.9	16.8
ADFom	14.0	2.35	11.5	16.1
CF	14.9	1.77	13.8	17.5
Lignin	7.2	0.78	6.3	8.1
Ash	5.9	0.33	5.6	6.3
OM	94.1	0.33	93.7	94.4
EtOH soluble CHO	32.0	2.16	29.7	34.1
Water soluble CHO	34.7	2.24	31.8	37.2
Ca	0.2	0.02	0.2	0.2
P	0.1	0.01	0.1	0.1
Mg	0.1	0.01	0.1	0.1
K	2.5	0.08	2.4	2.6
Na	0.02	0.01	0.02	0.03

Fe (mg/kg)	225	103	161	378
Mn (mg/kg)	17	2.4	15	20
Zn (mg/kg)	17	3.4	12	20
Cu (mg/kg)	4	0.5	4	5
NFC	64.0	3.04	60.0	66.7
NSC	32.0	2.16	29.7	34.1
NEL (Mcal/kg)[1]	1.6	0.02	1.5	1.6

[1] NEL was calculated based on the equation outlined in the Dairy NRC (2001).
Abbreviations: ADFom = Acid detergent fiber on an organic matter basis; aNDF = Neutral detergent fiber from alpha-amylase; aNDFom = Neutral detergent fiber on an organic matter basis; CF = crude fiber; EtOH Soluble CHO = ethanol soluble carbohydrates; Water soluble CHO = water soluble carbohydrates

The inclusion of AH in the diet resulted in lower intake of NDF for the 13% AH and 20% AH diets compared with 0% and 7% AH diets (**Table 23**). Intake of ADF was lower for the 13% AH compared with the other diets. There were no differences in DM, crude protein, calcium, phosphorus, or estimated net-energy intake due to diet. As anticipated, the intakes of DM, CP, ADF, NDF, and NEL were different for parity.

Table 23. Intake of dry matter and chemical components for cows consuming each almond hull (AH) diet.

						P-value[1]				
	0% AH	7% AH	13% AH	20% AH	SEM	Diet	Parity	L	Q	C
Intake in kg/d										
Dry Matter	26.7	27.6	26.4	26.6	0.72	0.16	0.01	0.35	0.50	0.05
CP	4.6	4.5	4.5	4.6	0.13	0.41	0.02	0.96	0.10	0.83
ADF	5.1	5.1	4.8	5.0	0.14	0.02	<0.01	0.15	0.07	0.03
aNDF	7.5	7.5	6.6	6.5	0.19	<0.01	0.01	<0.01	0.53	<0.01
Calcium	0.29	0.31	0.29	0.34	0.02	0.06	0.10	0.10	0.27	0.05
Phosphorus	0.08	0.08	0.08	0.08	0.003	0.44	0.04	0.66	0.62	0.14
NEL (Mcal)	45.8	45.6	43.9	43.9	0.57	0.11	0.02	0.03	0.90	0.23
DMI/BW %	4.1	4.2	4.0	4.1	0.11	0.30	0.61	0.69	0.64	0.07

[1] Diet = effect of different AH % diets on intake; Parity = effect of parity on intake; L, Q, C = linear, quadratic, and cubic contrasts of diets averaged over levels of parity and period; There were no significant Diet x Parity interactions for any measurements

Actual milk yield (**Table 24**) tended to decrease at the higher amount of AH feeding, with the 7% AH diet numerically the highest milk yield (39.3 kg). There was no effect of diet on yields of ECM, fat, lactose, and total solids. Protein yield was greater for the 7% almond hull diet resulting in the highest protein production (1.34 kg). For fat percentage, there was a significant effect of diet with the 13% and 20% almond hull diets higher than the 0% and 7% almond hull diets. Protein percentage was significantly different due to diet, with the 0% and 7% almond hull diets higher in protein content compared with the 13% and 20% almond hull diets. Milk urea nitrogen (MUN) concentration was lower for the 13% and 20% almond hulls diets compared with the 0% and 7% almond hull diets.

Table 24. Yield and composition of milk and components for cows consuming each almond hull (AH) diet.

						P-value[1]				
	0% AH	7% AH	13% AH	20% AH	SEM	Diet	Parity	L	Q	C
Yield (kg/d):										
Milk	38.8	39.3	36.9	37.7	1.37	0.05	0.01	0.09	0.99	0.03
ECM[2]	41.8	42.2	40.1	41	1.22	0.20	<0.01	0.36	0.96	0.05
Fat	1.46	1.47	1.44	1.48	0.05	0.65	0.02	0.47	0.99	0.29
Protein	1.33	1.34	1.23	1.25	0.04	<0.01	0.02	<0.01	0.85	<0.01
Lactose	1.95	1.99	1.86	1.9	0.07	0.07	0.02	0.16	0.84	0.03
Total Solids	4.85	4.91	4.63	4.73	0.15	0.09	0.01	0.17	0.89	0.03
Concentration (%):										
Fat	3.81	3.78	3.95	3.97	0.12	<0.01	0.70	<0.01	0.92	0.13
Protein	3.46	3.43	3.35	3.33	0.07	<0.01	0.35	<0.01	0.78	0.11
Lactose	5.02	5.06	5.04	5.04	0.05	0.48	0.87	0.44	0.26	0.39
Total solids	12.58	12.58	12.65	12.64	0.20	0.49	0.65	0.16	0.74	0.57
MUN (mg/dL)	10.65	10.13	8.62	8.08	0.58	<0.01	0.77	<0.01	0.98	0.29
SCC (1000's cells/mL)	47.82	31.42	47.33	49.25	1.25	0.47	0.18	0.76	0.16	0.54

[1] Diet = effect of different AH % diets on production; Parity = effect of parity on production; L, Q, C = linear, quadratic, and cubic contrasts of diets averaged over levels of parity and period; There were no significant Diet x Parity interactions for any measurements

[2] ECM = energy-corrected milk: [(0.327 x lbs of milk) + (12.95 x lbs of fat) + (7.65 x lbs of total protein)]

The decrease in aNDF intake seen in this study was likely due to the decrease in soyhull pellets (60.3% aNDF and 44.6% ADF compared with 23.8% aNDF and 14.9% ADF for AH) in the diet. Even though aNDF intake decreased as AH inclusion increased, ADF intake had a cubic response to diet, with the 13% AH diet resulting in the lowest intake. The smaller numeric differences in ADF intake compared with aNDF intake were likely due to the smaller differences in ADF content of AH and soyhull pellets compared to that of aNDF content.

Aguilar et al. (1984) found that feeding a TMR with up to 25% AH had no negative effects on DMI (21.8, 23, 22.7 kg/day for 0, 12.5, and 25% AH diets respectively), milk yield (24.9, 25.2, and 24.7 kg/day for 0, 12.5, and 25% AH diets respectively), and milk composition of fat and protein. These researchers added urea to the diets containing AH so that diets were isonitrogenous. Their approach to diet formulation was different than the current study since the forage proportion of the diet was 61% alfalfa hay for the control (no AH diet) and the forage decreased with each addition of AH to a high AH diet with 35% forage and 25% AH. More recently in an approach similar to Aguilar et al. (1984), AH were used to supplement alfalfa for dairy cows, with no urea added, which resulted in a decrease in CP intake, an increase in aNDF intake, and no effect on DMI (Williams et al., 2018). However, yields of milk (27.4 and 24.6 kg/day for control and AH diets respectively), ECM, milk protein, and milk lactose decreased in response to replacing alfalfa cubes with AH (Williams et al., 2018). In the current study AH were replacing concentrate ingredients and not the forage ingredients. Numerically, ECM and

milk yield were lowest for the 13% AH diet, but highest for the 7% AH diet showing a cubic effect of diet. There was a decrease in both milk protein percentage and yield as the amount of AH in the diet increased. When lactating goats were fed diets containing varying amounts of AH supplemented with urea, milk protein percentage was the highest for the 25% AH diet, but lowest for the 35% AH diet, with no change in milk protein yield (Reed and Brown, 1988). This was potentially due to the higher amount of non-protein nitrogen in the 25% AH diet, although all diets likely exceeded nitrogen requirements, resulting in higher blood and milk urea nitrogen concentrations as the AH and urea supplementation increased (Reed and Brown, 1988). Both ECM production and DMI responses followed a similar cubic pattern as AH inclusion in the diet increased, so it is possible that the changes in DMI could have been a primary driver in ECM production. Body condition score (BCS) was not analyzed in this study. Without BCS for the cows, it is difficult to say whether maintaining this level of ECM production would be sustainable for cows consuming 20% AH long term.

Dry matter, OM, and ADF, aNDFom, and crude protein apparent digestibilities were affected by diet (**Table 25**). For DM and OM digestibility, the AH diets were greater compared with the 0% AH diet, with the 20% AH diets having the highest digestibility for DM and OM. A similar pattern was seen with CP apparent digestibility with the 20% AH diet having a higher digestibility than the 0% AH diet. Digestibility of ADF was significantly higher for the 20% AH diet (46.9%) when compared with the 0% AH diet (41.6%). For ADFom digestibility, the 20% AH diet was numerically greater than that of the other diets. For aNDF digestibility, all of the AH diets were numerically higher than

the 0% AH diet. For aNDFom apparent digestibility, the 7% AH diet was higher than the 0% AH diet. There were no interaction effects of diet and parity for any digestibility parameters.

Table 25. Apparent total-tract digestibility for cows consuming each almond hull (AH) diet.

						P-value[1]				
	0% AH	7% AH	13% AH	20% AH	SEM	Diet	Parity	L	Q	C
Apparent Digestibility (%)										
DM	69.1	72.8	72.2	75.1	0.76	<0.01	0.40	<0.01	0.66	0.03
OM	70.8	74.0	73.5	76.2	0.74	<0.01	0.39	<0.01	0.72	0.04
ADF	41.6	43.5	43.4	46.9	1.24	0.03	0.16	0.01	0.41	0.31
ADFom	42.2	44.2	43.1	46.4	1.58	0.28	0.19	0.12	0.62	0.30
aNDF	47.5	51.4	49.0	50.1	1.19	0.13	0.27	0.34	0.26	0.06
aNDFom	47.9	52.6	50.5	51.6	1.24	0.07	0.28	0.13	0.16	0.07
CP	66.2	68.1	66.8	70.0	0.97	0.03	0.53	0.02	0.42	0.06

[1] Diet = effect of different AH % diets on digestibility; Parity = effect of parity on digestibility; L, Q, C = linear, quadratic, and cubic contrasts of diets averaged over levels of parity and period; There were no significant Diet x Parity interactions for any measurements
Abbreviations: ADFom = Acid Detergent Fiber on an Organic Matter Basis; aNDF = Neutral detergent fiber from alpha-amylase; aNDFom = Neutral detergent fiber on an organic matter basis

Previous studies reported that AH were highly digestible both *in situ* and *in vivo* (Alibés et al., 1983; Norollahi et al., 2006; Yalchi and Kargar, 2010). Commercial AH were reported to have 24 hour *in situ* DM digestibility of 70-71% with *in vivo* DM apparent digestibility of approximately 73% (Norollahi et al., 2006; Yalchi and Kargar, 2010; Yalchi, 2011). In diets where AH were added to account for up to 40% of the total ration, *in vivo* DM digestibility still ranged from 64-70% depending on the diet. In the present study, DM and OM apparent digestibilities ranged from 69 to 76%. Apparent digestibilities of DM. OM, ADF, and CP increased as the amount of AH in the diet increased. The results from this study contradict some of the previous work done on AH

digestibility. Digestibility studies conducted with sheep and goats found decreases in CP, aNDF, and ADF digestibilities when AH were added to the diet in place of alfalfa, but DM digestibility was decreased in the feeding study with goats (Reed and Brown, 1988; Yalchi, 2011). When steers were used to assess the digestibility of AH substituted for both grain and forage, researchers found no difference in DM digestibility but ADF digestibility was significantly decreased (Aguilar et al., 1984). In these previous studies, AH were mostly replacing forages (mainly alfalfa hay) in the diets, which could account for the differences in diet effect seen in the current study were AH are mainly replacing the concentrates in the diet.

The increase in DM, OM, ADF, and CP digestibilities seen in our study could be due to various factors. When goats were fed soybean hulls, a fibrous by-product, instead of corn grain, aNDF and ADF digestibilities increased although DM and CP digestibilities decreased (López et al., 2014). Dried citrus pulp, a fibrous but carbohydrate-rich by-product, when added to the diet of goats to replace corn grain, resulted in an increase in ADF digestibility, a decrease in CP digestibility, but no change in either DM or aNDF digestibilities (López et al., 2014). The authors also noted an increase in acetic acid production with a decrease in propionic acid when feeding citrus pulp (López et al., 2014). This increase in acetic acid production could be the result of increased microbial activity from fermentable carbohydrates such as pectin, which in turn could account for the higher digestion of fiber. Similarly, when beet pulp, a fibrous carbohydrate rich by-product, was added to replace barley in lactating Holsteins' diets, there was an increase in DM and aNDF digestibilities along with an increase in acetic acid production

(Poorkasegaran and Yansari, 2014). Like beet pulp and citrus pulp, AH are high in non-fiber carbohydrates (NFC) as well as soluble fiber that includes pectin (Saura-Calixto et al., 1983; Yalchi and Kargar, 2010). The NFC content is highly fermentable by rumen microorganisms and this increased substrate availability could contribute to microbial growth, increasing fermentation, which in turn could potentially increase fiber digestibility (Nocek and Russell, 1988). The easily fermentable carbohydrates, including pectin, in AH that can increase acetate and butyrate production could result in increased milk fat production (Poorkasegaran and Yansari, 2014; Urrutia and Harvatine, 2017), similar to what we saw with the cows consuming 20% AH in this study along with the increased amount of chewing.

Time spent in activities associated with resting, eating, and ruminating was affected by diet with the cows receiving the 20% AH diet resting less and eating and ruminating more when compared with the other three diets. As the amount of almond hulls in the diet increased, the number of minutes a cow spent ruminating increased. Cows on the 20% almond hull diet spent approximately 60 minutes more each day ruminating. This increase in chewing likely supported a rumen environment that supported high milk fat percent.

The fiber in AH is associated with lignin at a somewhat high percentage compared with typically fibrous feeds such as alfalfa (Yalchi and Kargar, 2010). This likely is reflected in the increasing amount of lignin in our diets as the percent of AH increased. While normally lignin would be associated with decreased digestibility, it could aid in fiber

digestibility by increasing the fiber mat in the rumen, which in turn would increase retention time of the AH, thereby decreasing passage rate (Poorkasegaran and Yansari, 2014). In addition, commercial AH have a relatively large particle size (about 35cm in diameter) compared to chopped forage or grain. This larger particle size could also lead to an increase in retention time (Poorkasegaran and Yansari, 2014). This is reflected in the increased percentage of time spent ruminating and eating for the cows consuming increasing amounts of AH. The increased time spent ruminating and chewing could have been a result of decreased passage rate, which would lead to increased fiber digestibility (Poorkasegaran and Yansari, 2014). Given the lack of a linear decrease in aNDF digestibility seen in this study, it is more likely that the size of the AH, not the lignin content, played a role in the increased rumination and chewing.

Take Home Messages: Almond hulls are an excellent, palatable feedstuff for lactating dairy cows. Almond hulls fed in our study were approximately 13% Crude Fiber As Is Basis so the hulls were high quality. Almond hulls of high quality replaced up to 20% of the concentrate ingredients in a TMR with no negative effects in production performance (feed intake, milk yield, milk composition, rumination time). Higher levels of feeding may be possible depending on the level of milk production. Higher amounts of feeding will be based on various factors identified in our survey of nutritionist, but cost of competitive ingredients and the consistency of the chemical composition of almond hulls are of utmost importance.

5. **Variation in Composition and Regulatory Issues**

The harvesting methods and current agronomic practices impact the contribution of debris to commercial AH. The almond huller removes a large portion of this debris although it is challenging to remove the sticks that are shorter than about 2 inches (Phippen, personal communication).

Commercial feed laws and regulations (CDFA validation 2773.5) define AH hulls as: "*Almond hulls are obtained by drying that portion of the almond fruit which surrounds the nut. They shall not contain more than 13.0 percent moisture, nor more than 15.0 percent crude fiber, and not more than 9 percent ash. If they contain more than 15.0 percent but less than 29.0 percent crude fiber, they shall be labeled "Almond Hulls and Shell" …"*.

We conducted a descriptive study to evaluate a 5-year period of information for commercial AH analysis using data from the CDFA to determine if there were differences associated with month and year and if any differences in the percent CF were related to moisture content of the AH.

Data for a 5-year period, 2014 to 2018, were obtained from the CDFA. The data included month and year of sampling, the percent CF As Is basis, and the percent moisture. The number of samples collected each year for analysis varied. The CDFA does not establish a priori the number of samples that will be collected during any given year. The CDFA Commercial Feed Program is not based on a statistical sampling approach with random sampling of AH. The CDFA Commercial Feed Program is focused on feed safety and label compliance, in the case of this study with AH, for percent CF As Is and percent moisture.

For the purpose of our research, a percent CF As Is basis greater than 15% CF was designated as a violation. A moisture content greater than 13% was designated as a violation. A description of the statistical analysis approaches is described (DePeters et al. 2020b)

There were 673 samples of AH analyzed during the 5-year period studied. The percentage of total AH samples analyzed that were found to be in violation were 62.1, 54.3, 39.3, 51.4, and 45.2%, for 2014, 2015, 2016, 2017, and 2018, respectively (**Table**

26). There was no obvious trend across years for the proportion of AH samples analyzed that were a violation for the percent CF As Is basis.

Table 26. Number of samples with no crude fiber (CF) violation (<15% CF), number of samples with crude fiber violation, total samples, and percent of samples that were a violation by year.

Year	Samples with No CF Violation	Samples with CF Violation	Total Samples	Percent that were Violation
2014	61	100	161	62.1
2015	85	101	186	54.3
2016	71	46	117	39.3
2017	51	54	105	51.4
2018	57	47	104	45.2

The percent CF (17 % CF As Is basis) in AH that were in violation was similar across years as was the percent CF (13 % CF As Is basis) for samples with no violations (**Table 27**).

Table 27. Count of violations by year including the average percent crude fiber (CF; As Is basis) for almond hull samples that were and were not a violation. A crude fiber greater than 15% is a violation.

Year	Number of violations	%CF in Violations	SD	%CF in No Violations	SD
2014	100	17.6	2.4	13.5	1.2
2015	101	17.4	2.3	13.7	1.0
2016	46	17.2	1.7	13.1	1.2
2017	54	17.4	1.8	13.0	1.4
2018	47	17.6	2.7	13.0	1.3

There was no obvious trend or difference found number of violations for percent CF As Is basis by month (**Table 28**). These violations were based on percent CF expressed on an As Is basis. A sample greater than 15% CF As Is basis was a violation.

Table 28. Count of violations by month and the average percent crude fiber (CF; As Is basis) for almond hulls samples that were a violation (> 15%CF) and were not a violation averaged over 5 years.

Month	Number of violations	Total samples	%CF in Violation	SD	%CF with No violations	SD
1	30	55	17.8	2.4	13.6	1.2
2	26	62	16.8	1.8	13.1	1.2
3	24	57	18.0	1.8	13.3	1.4
4	40	108	17.5	2.0	13.3	1.2
5	36	62	17.6	2.1	13.7	1.3
6	52	74	17.4	2.2	13.5	1.1
7	39	54	17.5	1.9	13.7	1.0
8	16	29	17.7	3.8	12.7	1.5
9	16	38	18.2	3.0	13.0	1.1
10	26	50	17.2	1.6	13.3	1.1
11	15	30	17.2	1.5	13.4	1.4
12	28	54	16.9	2.9	13.3	1.5

The percent moisture in AH samples that were a violation was similar to the percent moisture of samples that were not violations. Interestingly, the evaluation of violations adjusted for percent moisture provided the best statistical model fit to these data. From the statistical model, the coefficient for the percent moisture was -0.07 ($\pm$ 0.04), the negative slope indicating that as the percent moisture increased, the risk of violation declined.

Across all years, AH samples found in violation were 4.1 percentage units higher in CF As Is basis than those samples that were not in violation.

Analysis of percent CF As Is basis adjusted for percent moisture had a coefficient for percent moisture that was -0.14 (± 0.05), the negative slope indicating that as the percent moisture increased in the AH, the percent CF As Is basis decreased.

The number of AH samples hulls found in violation of percent moisture content (> 13% moisture) were too few counts for statistical analysis. Summer months had no violations while the moisture violation occurred during the winter months. Moisture content can be a concern for the growth of fungi since AH are high in sugar content.

The aim of the CDFA program of sampling and analyzing feeds is to ensure feed safety. Only a small portion of the AH marketed were actually sampled and tested for crude fiber and moisture during the 5-year period of review so it is difficult to know what quality of AH that were actually fed to lactating cows on dairy farms.

Take Home Messages: The rank from most to least violations (violation stated as > 15% CF As Is basis) as a proportion of total samples was 2014> 2015 > 2017 > 2018 > 2016.The percent crude fiber differed by year, but across all years crude fiber content was highest in summer compared with other seasons. When moisture content was included in the statistical model for predicting the number of violations, as the percent moisture increased, the risk of a percent crude fiber (As Is basis) violation decreased.

Including moisture in the model for predicting percent crude fiber by month and year indicated that as the percent moisture increased, the percent crude fiber decreased. From a legal definition perspective, setting a maximum percent moisture of 13% moisture prevents the intentional addition of moisture to almond hulls to reduce the risk of a violation. Almond samples in violations were 4.1 percentage units higher in percent crude fiber (17%CF) compared with samples found not to be in violation (13 %CF). From a practical perspective of feeding animals and purchasing almond hulls, it is likely wise to periodically obtain a respective sample from lots of almond hulls delivered to a dairy farm for chemical analysis. For commercial almond hulls: ***Test, Don't Guess*** when it comes to the chemical composition of AH.

6. Current and Future Research

We are currently looking at the chemical composition of almond hulls collected during the 2021 harvest season. This involves both wet chemistry analysis and NIR (near infrared) analysis. One aim is to assess the lignin content of almond hulls. The fiber in almond hulls is less digestible than might be expected. The question being addressed is does the type of lignin have an effect. We are measuring both the acid detergent lignin (ADL) and the Klason lignin (KL) content of commercial almond hulls and “pure” almond hulls (sorted to remove sticks and shells). Klason lignin content is great than ADL. The difference between ADL and KL is often times referred to soluble lignin. We are studying the difference in lignin type/methodology as it relates to the digestibility of hulls.

A lactation study with Holstein cows will evaluate the feeding of cubes that contain both alfalfa hay and almond hulls. The justification for this approach was to include almond hulls with lower quality alfalfa hay for the international export-market.

Almond hulls effectively replaced concentrate ingredients in the diets of lactating cows. However, how high can almond hulls go in a lactating cow diet as a forage ingredient is yet to be studied. Water is a scare resource in California, and that is not likely to change in the near future. In fact, with climate change and a growing human population, water will only become more restrictive to both plant and animal agriculture in California. How much of the diet silage can almond hulls replace?

Future research is yet to be determined. However, information on chemical composition of almond is need as agronomic practices evolve. The almond industry is exploring harvesting methods that do not involve the ground floor in the orchard. New, self-pollinating varieties will be developed and well as more water efficient almond varieties. The form of the almond hull product will also be explored. Pelleting, for example, is an approach to increasing the density for shipping almond hulls nationwide. However, pelleting, similar to cubing, changes the physical form of the hulls. In addition, pelleting, in particular, but also cubing reduce the ability of cows to sort the debris, sticks and shells, from the diet.

7. Summary

Almond hulls are a byproduct created in the production of almonds for human consumption. Almond hull composition is high in sugars and fiber, but low in protein. The chemical composition of almond hulls is quite variable as reflected by the high proportion of samples collected by CDFA Inspectors that were found in violation, greater than 15% CF As Is Basis. Almond hulls are high in energy content based on in vitro and in vivo determinations. The fiber content of almond hulls may not be as high as generally viewed. Almond hulls can successfully be used as either a concentrate and/or a forage ingredient in the diet of lactating dairy cows and for this reason almonds are a unique and important byproduct feedstuff for dairy cattle.

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Can increasing forage sorghum berry size improve berry processing and starch digestibility?

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TAKE HOME MESSAGES

- In the southern Ogallala Aquifer region dairy cattle inventory continue to rise while aquifer level is decreasing. With over 850,000 dairy cows and over 2 million cattle on feed in this region, increasing water efficiency use will be key to meet forage demand in the future.
- Current strategies used or under consideration to increase water efficiency use include growing water efficient crops, hydroponic systems, and use of buffer strips for forage production[1].
- Forage sorghum is a drought tolerant, water efficient crop that could be used to partially meet beef and dairy cattle forage demand. However, berry processing and starch digestibility remains an issue when using forage sorghum for silage production.
- Results from this study suggest that increasing sorghum berry size, at least for the size difference of the two forage sorghum hybrids compared, might not increase in situ starch digestibility.
- Future research should evaluate the effect of using hybrids with larger berry size and using a sorghum hybrid with a 45:55 panicle to vegetative parts ratio (i.e., a ratio similar to corn) on starch digestibility.

INTRODUCTION

As dairy cattle inventory in the southern Ogallala Aquifer region continues to increase farmers are seeking drought tolerant, quality forages to continue meeting forage demands (Fig. 1). With roughly 850,000 dairy cows and over 2 million cattle on feed, the beef and dairy cattle industries generate substantial economic benefits. Roughly 12% of the milk in the country is produced in the southern Ogallala Region[2], and dairy farmers and milk processing plants contribute over $4 billion in annual economic output and generate over 14,000 jobs[3].

The significant increase in dairy cattle inventory that occurred since 1990 (roughly 800,000 dairy cows more) increased forage demand. However, because forage production displaced cash crops, water use from the Ogallala Aquifer did not increase with the migration of dairies. Furthermore, this region has been grain deficit since 1970 and imports over one third of the feed grains used[3]. Therefore, a significant amount of the water use for feed production comes outside of this region as imports of corn grain and soybean meal. However, the rate of decline of the Ogallala Aquifer remains

Figure 1. Southern Ogallala Region (light blue area below dashed line). Adapted from Guerrero et al., 2012.

constant and dairy farmers are seeking water efficient crops to meet forage demands.

Corn silage has historically been the silage of choice, but forage sorghum adoption has increased in recent years[4] (Fig. 1). However, variable berry sizes results in poor berry processing, digestibility and starch availability. This article will provide a summary of previous and current research on forage sorghum berry processing and starch digestibility.

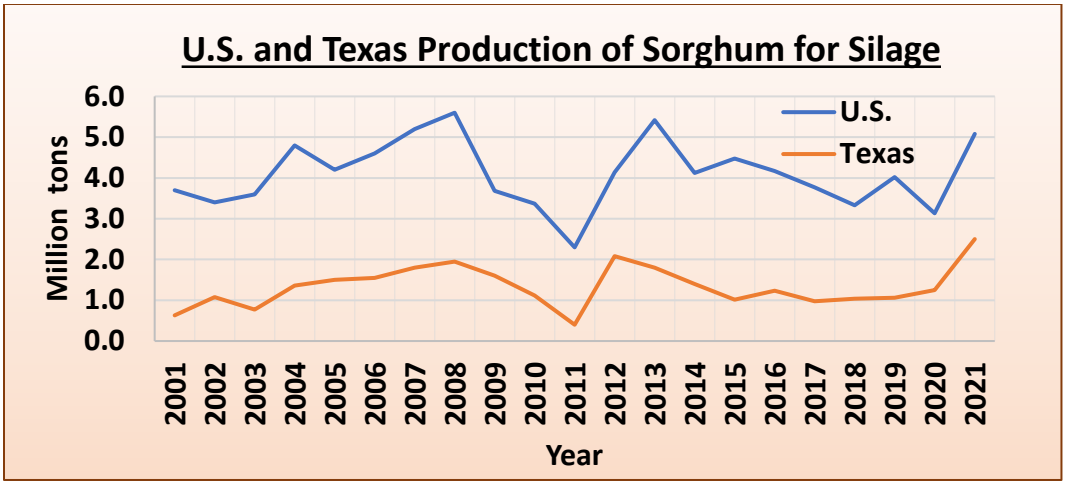

Figure 1. Annual production of sorghum for silage in the U.S. and Texas. From 2020 to 2021 production of sorghum for silage increased in the U.S. and Texas by 62 and 100%, respectively[4].

Previous research on forage sorghum berry processing and starch digestibility

The main disadvantages of Whole Plant Sorghum Silage (WPSS) compared to Whole Plant Corn Silage (WPCS) are the higher content of aNDF, ADF and lignin and lower starch concentration and starch digestibility of the former[5]. The higher content of lignin in conventional WPSS causes the decreased NDF digestibility (NDFD) compared to WPCS. However, BMR WPSS hybrids may potentially reach levels of NDFD similar to conventional WPCS[6].

Sorghum berry processing and starch digestibility could be improved with increase mechanical processing. Decreasing the theoretical length of cut from 0.86 to 0.59 inches and changing roll gap settings from 3 mm to 1 mm increased starch passing the 2.36 mm sieve roughly by 20%-points[7]. In turn, increased berry processing should increase starch digestibility. Compared to whole sorghum berries, berries that were cut in 2 and 4 parts showed improved ruminal in situ starch digestion kinetics (effective ruminal disappearance was 15.2%, 22.6%, 40%, respectively)[8]. Improvements in starch digestibility would be explained by disruption of the pericarp and starch-protein matrix as well as increased surface area for microbial digestion. However, more aggressive kernel processing may imply higher fuel costs, increased wear out of kernel processors and increased labor and logistics to change harvesting settings. Overall, this would lead to increase harvesting costs for the dairy farmer. An alternative strategy to increase sorghum berry processing without changing mechanical processing could be to use forage sorghum hybrids with increased berry size.

Current research on forage sorghum berry processing and starch digestibility

Recently, the starch digestibility of forage sorghum was evaluated on a commercial West Texas dairy under center pivot irrigation[9]. The objective was to assess the effect of forage sorghum berry size on berry processing score and in situ starch digestibility.

Corn and two forage sorghum hybrids were evaluated: 1) F24, larger berry size and 2) F10, smaller berry size (Fig. 2). Plots were blocked by irrigation section, sorghum hybrids were randomly allocated within blocks and replicated five times (Fig. 3). Forage sorghum hybrids were harvested at soft dough stage, 30% dry matter and using kernel processors set 2 mm apart. Whole plant and chopped (processed) samples were obtained the day before and at harvest, respectively. Whole and processed grain samples were screened to determine particle size distribution.

Figure 2. The berry processing score and in situ starch digestibility of forage sorghum hybrid F10 (smaller berry size, left) and F24 (larger berry size, right) were compared. Picture courtesy of Diego Druetto.

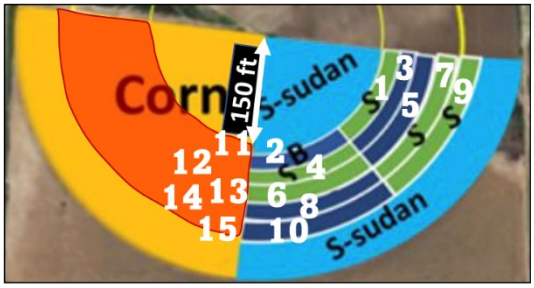

Figure 3. Corn and sorghum crops were randomly allocated to be planted on the west or east side. Then sorghum hybrids F10 (**light green**) and F24 (**dark blue**) plots were randomly allocated within blocks and replicated five times. Sorghum hybrids were seeded on 05/24/21 and harvested 09/03/21.

Three important questions were answered with this study:

1) Were berries from forage sorghum F24 bigger than F10? Yes, we did a particle size separation of the intact berries (obtained one day before harvest) with two consecutive sieves (4 mm and 3.3 mm) and a pan. Before harvest, F24 had less intact berries passing the 3.35 mm sieve compared to F10, validating the larger berry size on F24.

Intact sorghum berry particle size distribution			
Berry size	**F10**	**F24**	***P*-value**
>4mm, %	0^{a} (± 2.7)	41^{b} (± 2.7)	<0.01
>3.35, %	42 (± 3.9)	49 (± 3.9)	0.24
<3.35, %	58^{a} (± 4.5)	10^{b} (± 4.5)	<0.05

Table 1. Intact berry particle size distribution from panicles obtained the day prior to harvest for silage. Means within the same row with different superscripts are significantly different.

Interestingly, the reproductive to vegetative parts ratio (i.e., panicle : leaf + stems for sorghum hybrids, cobs : leaf + stems for corn) was significantly lower in forage sorghum hybrids (30:70) compared with corn (45:55).

2) Did berry processing score of F24 and F10 differ? There was very little difference in favor of F10. Two different sieves were used: 1) 2.36 mm sieve: more starch from processed berries from F10 passed the sieve compared to F24; 2) 1.7 mm sieve: no difference. Considering the initial size of the F24 berry was significantly bigger and starch from processed berries passing the 1.7 mm screen did not differ between hybrids, this could indicate potentially more F24 berries were broken compared to F10.

Processed berry particle size distribution	F10	F24	*P*-value
Starch above 2.36 mm screen, %	68^{a} (± 1.6)	75^{b} (± 1.6)	<0.001
Starch passing 2.36 mm screen, %	32^{a} (± 1.6)	25^{b} (± 1.6)	<0.001
Starch above 1.7 mm screen, %	84 (± 0.9)	83 (± 0.9)	0.34
Starch passing 1.7 mm screen, %	16 (± 0.9)	17 (± 0.9)	0.34

Table 2. Processed berry particle size distribution from samples obtained at harvest for silage. Means within the same row with different superscripts are significantly different.

3) Did F24 have better starch digestibility than F10, and how do they compare with corn? There was no difference in starch digestibility between F24 and F10. Corn had better starch digestibility compared to both forage sorghum hybrids.

Crop	Sorghum (F10)	Sorghum (F24)	Corn	*P*-value
In-situ starch digestibility, % starch	59.5^{a} (± 3.03)	59.3^{a} (± 3.03)	74.8^{b} (± 3.03)	0.001

Table 3. In-situ rumen starch digestibility (7 h) of forage sorghum hybrids F10 and F24 and corn silage. Means within the same row with different superscripts are significantly different.

As expected, sorghum hybrids had higher ADF, aNDF and lignin content and lower NDFD and starch content compared to corn. Sorghum hybrid F24 had lower lignin, lower crude protein and higher starch content compared to sorghum hybrid F10 (Table 4).

Crop	Sorghum (F10)	Sorghum (F24)	Corn	SEM
CP, % DM	9.9^{a}	8.9^{b}	9.0^{b}	0.12
ADF, % DM	29.8^{a}	29.2^{a}	21.3^{b}	0.54
aNDF, % DM	44^{a}	44^{a}	38^{b}	0.76
Lignin, % DM	4.4^{a}	4.1^{b}	3.7^{c}	0.09
Starch, % DM	23.9^{a}	26.6^{b}	31.4^{c}	1.00
WSC, % DM	0.38^{a}	0.42^{a}	3.21^{b}	0.14
NDFD30, % NDF	44.5^{a}	45.5^{a}	55.0^{b}	0.65
uNDF120, % DM	16.9^{a}	16.5^{a}	11.2^{b}	0.32

Table 4. Nutrient value of sorghum hybrids F10, F24 and corn. Means within the same row with different superscripts are significantly different.

Discussion and Future Directions

Results from this study suggest that increasing sorghum berry size, at least for the size difference of the two forage sorghum hybrids compared, might not increase in situ starch digestibility. Future research should evaluate the effect of using hybrids with larger berry size than F24. In addition, the ratio of the panicle to vegetative parts (leaves + stems) of sorghum vs. corn was very different (30:70 vs. 45:55), and this might have affected berry processing. Hence, future research should also assess the value of using a sorghum hybrid with a 45:55 panicle to vegetative parts ratio on starch digestibility.

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Enhancing milk component yields in lactating dairy cows and effects of monensin on the performance of lactating dairy cows fed contemporary diets

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INTRODUCTION

In many milk markets across the U.S., milk volume is being managed while milk components are in high demand. This puts more pressure on nutritionists to develop diets that optimize milk fat and protein yields which means several factors related to diet composition, and source of nutrients need to be evaluated. Also, this requires diets that optimize rumen function and efficiency and make sure all requirements are met. There are several dietary factors that positively impact milk components, and this paper will briefly touch on a few of them and then describe a study where the concept of a "modern diet" was evaluated in the context of different levels of monensin and the effects on milk components.

The primary factor impacting the ability to optimize rumen function and provide substrates for milk components and maintain rumen health is digestible aNDFom, especially in the form of forage fiber. When working to evaluate diets and models, the factor accounting for most of the variation in amino acid supply is digestible aNDFom (Higgs et al., 2015) and this is intuitive given the nature and function of the rumen. One of the most limiting factors in diet formulation can be digestible aNDFom or a combination of highly digestible fiber with a low inventory, so the opportunity for the high digestibility forage cannot be used to optimize the diets. Acetate from fiber digestion is one of the primary energy sources for the cow and can account for approximately 30% of the energy supply and is also the primary source for de novo fatty synthesis in the gland. There are many studies describing the role of acetate in milk fat synthesis and reducing equivalents necessary for elongation of milk fat (Bauman et al., 1970; Van Soest, 1994; Shephard and Combs, 1998; Palmquist, 2007; and Maxin et al., 2011) thus, providing highly digestible aNDFom is one of the primary dietary means to ensure high acetate production and also formulate diets that are first limiting on total aNDFom intake and not negatively impacted by uNDF levels. From the calculations of Mertens (2009) concerning intake capacity of aNDFom, the general expected levels of aNDFom intake for high cows just post-peak are approximately 1.2% to 1.30% of body weight (BW). Since those guidelines were published, and with the ability to directly measure and estimate the uNDF of the forages and byproducts, we have data suggesting that the upper limits to intake and rumen fill as suggested by Mertens are consistent with high digestibility forages and that uNDF in forages can be high enough to be first limiting dry matter intake (DMI) (Cotanch et al, 2014; Van Amburgh et al., 2018). From data generated at Miner Institute, University of Bologna and Cornell University using rumen emptying and intake experiments, total aNDFom intake can reach about 1.5% of BW with good digestibility and some legume containing diets and that translates into approximately 1.3% BW of rumen fill. Intake of uNDFom with high forage diets is about 0.4% BW and rumen mass is about 0.48% to 0.62%. Therefore, to optimize rumen function, and acetate production, when available, diets need to be formulated with the largest digestible pool of

aNFDom to ensure adequate DMI and fiber digestion for acetate yield keeping the relative limits to DMI in mind.

In high quality pastures, the typical NFC fractions formulated for in North America are reversed with large water and ethanol soluble carbohydrates and lower starch. This suggests that the ruminant is more adapted to high sugar than high starch diets and from a component basis this makes sense as sugars will ferment to butyrate, a fat precursor (Oba et al., 2015 and Penner et al., 2011). In addition, sugar fermentation improves rumen pH by enhancing bacterial and protozoal yield (Penner et al., 2009; Dineen et al., 2020) and enhancing rumen energy status and epithelial transport of acids and glucose (Penner et al., 2011). Increased butyrate production can enhance milk fat percentage and yield and enhance fiber digestibility (Broderick et al., 2008). Data from Hoover (1987) and Hoover and Miller-Webster (1998) would suggest that in TMR fed cattle, rumen function is optimized at sugar levels of 6% to 8% dry matter in the diet. This would provide more substrates for milk fat synthesis, greater microbial yield for milk protein synthesis and enhance fiber digestion which would improve DMI, substrates and microbial yield.

Further, to optimize milk components, amino acid (AA) supplies need to be optimized to meet the requirements for milk volume, fat, and protein (Yoder et al, 2020; LaPierre et al., 2019). With more data, it appears meeting requirements for amino acids, especially methionine and lysine with some data on histidine, that energetic efficiency, not protein efficiency is increased. This concept of horizontal integration was discussed by Lobley, (2007) where he demonstrates that AA will be used to fill any pathway necessary to improve the energetic efficiency of the animal and that protein and energy are integrated in metabolism and not separate entities as we normally formulate for. To that point, data have been published integrating the AA requirements on both a metabolizable energy basis (Higgs and Van Amburgh, 2016) or a digestible energy basis (Lapierre et al., 2019). This approach makes amino acid formulation much more precise and estimated requirements tend to be higher than previously described. Modeling and cow response data from Higgs and Van Amburgh (2016) demonstrated the approach of describing AA requirements on a gram per megacalorie of metabolizable energy (ME) was a useful approach for defining more precise requirements for lactating dairy cattle. Recent work by LaPierre et al, (2019) suggests that when applying this approach using Cornell Net Carbohydrate Protein System (CNCPS) v6.55 (Van Amburgh et al., 2015) the values for methionine are 1.19 g/Mcal ME and for lysine is 3.21 g/Mcal ME based on energy corrected milk responses from lactating dairy cattle. Thus, to improve milk and milk component yield, those AA should be formulated close to those values.

Some feed additives such as yeasts and ionophores are shown to impact milk components and the efficiency of microbial activity in the rumen. Although monensin is associated with improved feed efficiency, negative effects on milk fat production and synthesis have been previously reported. Monensin altered the content of saturated and unsaturated fatty acids (FA) in ruminal fermenters through inhibition of biohydrogenation (Fellner et al., 1997), thus it is hypothesized that the mode by which monensin decreases milk fat is through an accumulation of conjugated FA in the rumen that inhibit milk fat synthesis (Alzahal et al., 2008; Baumgard et al., 2000). More recently, the effect of monensin on milk fat production was greatest in studies that fed diets high in unsaturated FA (Alzahal et al., 2008; He et al., 2012), and a reduction in milk fat synthesis was predicted to be caused by an accumulation of long chain FA in the rumen that inhibit de novo FA synthesis (Dubuc et al., 2009). Further, monensin in high starch diets has been associated with a decrease in milk fat

production due to a reduction in biohydrogenation caused by monensin and high levels of rumen fermentable starch that decrease ruminal pH (Bradford and Allen, 2004; Van Amburgh et al., 2008). And more recently, Akins et al. (2014) reported a numerical decrease in milk fat content with monensin feeding in average starch (27%) diets, but not in reduced starch (21%) diets.

Using diet formulation systems nutritionists can monitor rumen unsaturated FA load (RUFAL), dietary fat, starch, and NDF content to help minimize diet induced milk fat depression, and therefore understand how to optimize the use of monensin in lactating dairy cows. Previous studies that reported a decrease in milk fat production with monensin feeding were performed decades ago when dietary nutrients in dairy diets were not as well understood as they are today, and more recent monensin studies have reported no effect on milk fat production (Akins et al., 2014; Hagen et al., 2015; Vasquez et al., 2021).

The FDA has approved the use of monensin in lactating dairy cattle diets at levels of 11 g/ton to 22 g/ton (DM basis), but recently, few studies have been conducted evaluating lactation performance at various monensin concentrations using more contemporary diets formulated with refined nutrient requirements and supplies. Therefore, the amount of monensin in the diet needed to effect milk production and composition, intake, and shifts in milk FA profile is of interest. The objective of this study was to evaluate increasing dietary monensin (Rumensin, Elanco Animal Health, Greenfield, IN) concentration on milk performance, milk FA profile, and production efficiencies (component-corrected milk/ DMI) in lactating dairy cows fed contemporary diets. We hypothesized milk performance and feed efficiency would improve with increasing levels of dietary monensin with no negative effects on milk component yield or shifts in FA profile.

MATERIALS AND METHODS

Experimental Design and Treatments

The experiment was conducted from September to December 2020 at the Cornell University Ruminant Center (Harford, NY), and all procedures were approved by Cornell University Animal Care and Use Committee. One-hundred ninety-two cows (120 ± 50 DIM; mean ± standard deviation) were stratified by parity, DIM, and pre-trial milk production, and assigned to 1 of 12 pens housing 16 cows per pen (12 multiparous and 4 primiparous) in a 91-day longitudinal study with a 29 day covariate and 62 day experimental period. All cows were fed 11 g/ton (DM basis) monensin for the adaptation and covariate period. Following the covariate period, pens were randomly assigned 1 of 4 treatment diets stratified by milk performance and BW data collected in the covariate period. Cattle were housed in freestall pens with 16 headlocks and sand-bedded stalls, and had free access to feed, water, and bedding. Cows were milked three times daily at 0700h, 1500h, and 2300h in a double-16 parallel parlor. Feed was delivered once daily as a TMR at 0600h ad libitum to allow for 5% refusals.

Diets were formulated to meet or exceed nutrient demands for high producing lactating dairy cows using CNCPS (v6.55; Van Amburgh et al., 2015). Methionine and lysine were balanced using the latest information on requirements and supply as generated in the studies of LaPierre et al. (2020) where amino acid requirements are described on a gram per unit of ME basis (Higgs and Van Amburgh, 2016). For diet formulation, the methionine requirement was set at 1.19 g methionine per Mcal ME and lysine was set at 3.21 g per Mcal ME (or 2.7 times the grams methionine). All diets consisted of (DM basis) 34.9 % corn silage, 19.4 % grass haylage, 18 % corn meal, 6.8 % soybean meal, and 21 % pre-

mix containing monensin (Purina Animal Nutrition, Caledonia, NY; Table 1). Treatments were 0 g/ton monensin (CON), 11 g/ton monensin (R11), 14.5 g/ton monensin (R14.5), and 18 g/ton monensin (R18) on a DM basis, and monensin intake was formulated to be 305 mg/d, 404 mg/d, and 515 mg/d for R11, R14.5, R18, respectively.

Forages and TMR were sampled twice weekly, composited, and sent to Cumberland Valley Analytical Services (Waynesboro, PA) once per week for nutrient analysis. Additionally, FA profile was determined on TMR samples. Grains were sampled once weekly, and a 4 wk composite was sent once monthly for chemical analysis. Grain mixes were sent for determination of monensin concentration upon delivery of a new batch (Eurofins Food Chemistry Testing US, Inc, Greenfield, IN). Feed DM was determined twice weekly for diet adjustment and calculation of DMI. Pen level intake was obtained daily using Feedwatch (Valley Agricultural Software, Tulare, CA), and determined using observations of feed offered and feed refused.

Milk production was recorded at every milking (Delpro, DeLaval Inc, Kanas City, MO) and milk samples were taken at 3 consecutive milk sessions once weekly during the last two weeks of the covariate period and every week of the experimental period. Samples were analyzed for fat, true protein, anhydrous lactose, and MUN using a FTIR spectrophotometer (Lactoscope model FTA, Delta Instruments, Drachten, the Netherlands) at the Department of Food Science at Cornell University (Ithaca, NY). De novo, mixed-origin, and preformed FA were analyzed by FTIR on all milk samples according to PLS prediction models described by Woolpert et al. (2016) and calibration was carried out using gas-liquid chromatography reference chemistry described by Wojciechowski and Barbano (2016). The same calibration set was used for milk components and FA analysis with concentrations ranging from 0.05 to 1.4 g/100g milk de novo FA, 0.08 to 2.2 g/100g milk mixed FA, and 0.06 to 1.9 g/100g milk preformed FA. In addition, FA chain length (mean carbon number per FA) and unsaturation (double bonds per FA) were measured as previously described by Wojciechowski and Barbano (2016). Body weight (BW) was obtained once weekly following the 1500h milk session as well as body condition score (BCS) using a 5-point scale according to Wildman et al. (1982). Blood samples were collected once weekly via the coccygeal vein into tubes containing sodium heparin. Samples were centrifuged (3,000 × *g* for 20 min at 4˚C), and plasma was harvested and frozen at -20 ˚C for urea nitrogen analysis (No. 640, Sigma-Aldrich, St. Louis, MO). Finally, rumination time (minutes per day) was obtained from cows with a pre-existing Smartbow ear tag (Zoetis, Parsippanny, NJ; CON: n = 34, R11: n = 38, R14.5: n = 42, and R18: n = 42).

Statistical Analysis

All data, excluding BCS, were analyzed through SAS version 9.4 (SAS Institute Inc., Cary, NC) using PROC MIXED and LSMEAN statements to compare treatment means. When individual cow variables with covariate structure and repeated weekly measurements (milk production, milk composition and FA profile, BW, rumination, and PUN) were analyzed, pen was the experimental unit and cow was the observational unit as previously described by Fessenden et al. (2020) and Bellow et al. (2016), and the following model was used:

$$Y_{ijklm} = \mu + T_i + W_j + TW_{ij} + P_{k:i} + B_{l:k:i} + BX_{lik} + \varepsilon_{ikklm},$$

where Y_{ijklm} = dependent variable, μ = overall mean, T_i = fixed effect of treatment i, W_j = fixed effect of week j, TW_{ij} = fixed interaction of treatment i and week

j, $P_{k:I}$ = random effect of pen k within treatment i, $B_{l:k:i}$ = random effect of cow within pen k within treatment i, BX_{lik} = the covariate adjustment for each cow, and ε_{ikklm} = residual error. An auto-regressive structure [AR(1)] was used to

Table 1. Ingredient composition of experimental diets

	Diet[1]			
Ingredient, % of DM	CON	R11	R14.5	R18
Corn silage	34.9	34.9	34.9	34.9
Grass haylage	19.4	19.4	19.4	19.4
Corn meal	18.0	18.0	18.0	18.0
Soybean meal	6.81	6.81	6.81	6.81
SoyPass[2]	5.83	5.83	5.83	5.83
Citrus pulp	4.49	4.49	4.49	4.49
Wheat middlings	4.49	4.49	4.49	4.49
Dextrose	1.60	1.60	1.60	1.60
Bloodmeal	1.00	1.00	1.00	1.00
Berga fat F100[3]	0.60	0.60	0.60	0.60
Energy Booster 100[4]	0.60	0.60	0.60	0.60
Ground limestone	0.54	0.54	0.54	0.54
Min AD[5]	0.45	0.45	0.45	0.45
Sodium bicarbonate	0.42	0.42	0.42	0.42
White salt	0.27	0.27	0.27	0.27
Vitamin and mineral mix[6]	0.22	0.22	0.22	0.22
Magnesium oxide	0.11	0.11	0.11	0.11
Smartamine M[7]	0.10	0.10	0.10	0.10
Smartamine ML[7]	0.10	0.10	0.10	0.10
Levucell SC[8]	0.05	0.05	0.05	0.05
Rumensin 90[9]	-	0.006	0.008	0.01

[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin.
[2]Lignotech USA, Rothschild, WI.
[3]Berg + Schmidt America LLC, Libertyville, IL.
[4]Milk specialties, Eden Prairie, MN.
[5]Calcium (22%) and magnesium (12%) supplement (Min-AD, Winnemucca, NV).
[6]Contained (DM basis) 27.4% Ca; 223 ppm Fe; 24,997 ppm Zn; 5,765 ppm Cu; 18,473 ppm Mn; 134.5 ppm Se; 568 ppm Co; 568 ppm I; 2021 KIU/kg vitamin A; 562 KIU/kg vitamin D; 9660 IU/kg vitamin E)
[7]Adisseo Inc, Alpharetta, GA.
[8]Lallemand Inc, Milwaukee, WI.
[9]Monensin, 90.7 g/lb. (Elanco Animal Health, Greenfield, IN).

analyze repeated measurements with cow in pen within treatment. For pen level variables (DMI and production efficiencies), a random effect of pen within treatment was used. Three cows did not complete the experiment due to health issues (1 and 2 cows from R14.5 and CON, respectively). The BW data from wk 6 to 9 of the experimental period were removed from statistical analysis due to scale malfunctions during extreme cold weather conditions, with wk 5 BW was used as final BW to determine BW change. Degrees of freedom were determined using Kenward-Roger option and least square means were adjusted by Tukey method for multiple comparison tests. Body condition score data was analyzed using a non-parametric analysis (PROC NPAR1WAY) with treatment as the classification variable. Statistical significance was reported as $P \leq 0.05$ and tendencies as $0.05 < P \leq 0.10$.

RESULTS AND DISCUSSION

Ingredient composition and chemical analysis of the diets are in Table 1 and 2, respectively, and chemical analysis of the forages and concentrate mixes are in Table 3. The analyzed monensin concentration for all treatment pre-mixes, on a DM basis, are as follows: CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, and R18 = 19.3 g/ton monensin. The actual monensin intake was 0, 384, 465, and 589 mg/d for CON, R11, R14.5, and R18, respectively. Lactation performance results are in Table 4. We observed a numerical increase in DMI in the R18 group compared to CON, R11, and R14.5 (27.7 vs. 26.9, 26.8, and 26.7 kg/d, respectively). Monensin treatment tended to have a quadratic effect on DMI ($P = 0.10$) where R11 and R14.5 had slightly decreased DMI compared to CON, but DMI increased in the R18 group. This finding is not consistent with previous studies as increasing dietary monensin has been associated with either no change or a slight decrease in DMI (Akins et al., 2014; Hagen et al., 2015), although Recktenwald et al. (2014) reported a trend for increased DMI in cows fed monensin compared to none in diets high and low in starch and protein content. Milk yield was not affected by monensin treatment in agreement with experiments of Alzahal et al. (2008) and Hagen et al. (2015) (Table 4). The lack of an adaptation period for the CON group following the covariate diet of 11 g/ton monensin was predicted to decrease the ability to detect treatment effects because we observed a decrease in milk yield in the CON group compared to all monensin treated groups from wk 4 to 9 (data not shown) indicating cows were still adjusting to the removal of monensin in the beginning 3 wk of the experimental period. This is consistent with lactose production data as we observed a decrease in lactose yield in the CON group compared to all monensin treated groups following wk 3 of the experimental period (data not shown). In agreement, Akins et al. (2014) reported an increase in milk yield in cows fed monensin from wk 4 to 12, but not from wk 1 to 3, suggesting cows were still adapting to monensin changes in the diet. Additionally, the experimental period for Akins et al. (2014) was 3 wk longer than the current study, allowing for greater detection of monensin effects on milk yield over time.

No significant treatment effects were observed for milk fat concentration or yield; however, milk fat percentage increased numerically with increasing monensin concentration (4.60, 4.67, 4.71, and 4.66 for CON, R11, R14.5, and R18 respectively; Table 4). The numerical increase in milk fat was most likely an effect of monensin on de novo FA synthesis as there was a linear increase ($P < 0.05$; Table 5) in de novo and mixed fat content with increasing levels of

monensin. Previous research has shown monensin decreases milk fat concentration with increasing monensin levels (Dubuc et al., 2009; Duffield et al., 2008b), while others (Martinez et al., 2009; McCarthy et al., 2018) have reported no effect on milk fat. More recently, monensin has been shown to interact with other dietary factors such as starch content and unsaturated oils to reduce milk fat, rather than causing milk fat depression independently (McCarthy et al., 2018). Van Amburgh et al. (2008) also reported monensin diets high in

Table 2. Analyzed nutrient composition (mean ± SD) of experimental diets

	Diet[1]			
Item	CON	R11	R14.5	R18
DM, % as-fed	43.4 ± 1.5	44.0 ± 1.2	43.5 ± 1.3	44.1 ± 1.4
CP, % of DM	15.3 ± 0.3	14.9 ± 0.6	15.0 ± 0.6	15.4 ± 0.6
ADF, % of DM	19.4 ± 1.6	20.4 ± 1.6	19.7 ± 1.0	18.8 ± 1.4
aNDF, % of DM	32.0 ± 1.4	32.8 ± 0.9	31.7 ± 1.1	31.3 ± 1.7
Sugars, % of DM	5.7 ± 0.3	5.7 ± 0.7	5.8 ± 0.2	5.9 ± 0.4
Starch, % of DM	25.6 ± 1.6	24.9 ± 1.0	25.3 ± 0.9	26.2 ± 1.2
Ether extract, % of DM	4.4 ± 0.2	4.2 ± 0.3	4.4 ± 0.2	4.2 ± 0.3
Ash, % of DM	7.2 ± 0.3	7.0 ± 0.3	7.1 ± 0.4	7.1 ± 0.3
NFC, % of DM	43.7 ± 1.2	43.7 ± 0.9	44.5 ± 1.6	44.6 ± 1.4
NSC, % of DM	31.3 ± 1.5	30.5 ± 1.1	31.1 ± 0.8	32.1 ± 1.1
ME, Mcal/kg[2]	2.7	2.7	2.7	2.7
FA, % of DM				
Total	3.56 ± 0.31	3.47 ± 0.11	3.73 ± 0.27	3.78 ± 0.28
16:0	1.12 ± 0.13	1.04 ± 0.03	1.14 ± 0.11	1.19 ± 0.10
18:0	0.33 ± 0.05	0.31 ± 0.03	0.33 ± 0.06	0.35 ± 0.05
18:1 *cis*-9	0.50 ± 0.07	0.49 ± 0.02	0.53 ± 0.05	0.54 ± 0.06
18:2 *cis*-9, *cis*-12	1.13 ± 0.08	1.11 ± 0.05	1.20 ± 0.07	1.20 ± 0.07
18:3 *cis*-9, *cis*-12, *cis*-15	0.31 ± 0.04	0.34 ± 0.02	0.33 ± 0.04	0.32 ± 0.03
RUFAL[3]	1.94	1.94	2.06	2.06

[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin.
[2]Predicted using the Cornell Net Carbohydrate and Protein System v6.5 (Van Amburgh et al., 2015).
[3]Rumen unsaturated fatty acid load = 18:1 + 18:2 + 18:3 from the chromatographic analysis of the diets.

Table 3. Nutrient analysis (mean ± SD) of diet ingredients

Item	Corn Silage	Grass Haylage	CON Mix	R11 Mix	R14.5 Mix	R18 Mix
DM, % as-fed	29.3 ± 0.7	39.5 ± 4.0	90.5 ± 0.3	90.7 ± 0.9	90.5 ± 0.4	90.4 ± 0.3
CP, % of DM	7.5 ± 0.4	15.7 ± 0.7	21.9 ± 0.5	23.9 ± 1.9	21.2 ± 1.3	22.4 ± 1.5
ADF, % of DM	24.1 ± 1.1	34.4 ± 1.4	14.7 ± 2.0	14.0 ± 2.6	14.8 ± 2.9	14.3 ± 2.4
aNDF, % of DM	39.7 ± 1.7	52.0 ± 1.9	22.7 ± 3.3	22.1 ± 3.6	23.4 ± 3.5	22.5 ± 2.2
Sugars, % of DM	0.4 ± 0.2	3.4 ± 0.6	17.5 ± 0.9	16.2 ± 2.0	18.3 ± 0.9	18.4 ± 1.0
Starch, % of DM	34.5 ± 1.6	1.4 ± 0.3	5.0 ± 0.7	5.3 ± 3.9	5.4 ± 1.7	5.8 ± 3.3
Ether extract, % of DM	3.2 ± 0.1	3.7 ± 0.3	6.7 ± 0.9	5.6 ± 1.3	6.8 ± 1.7	6.9 ± 2.2
Ash, % of DM	3.4 ± 0.3	8.5 ± 0.5	13.0 ± 1.9	12.1 ± 2.5	12.8 ± 0.3	13.2 ± 1.2
NFC, % of DM	46.8 ± 1.4	23.1 ± 1.4	38.8 ± 2.0	35.1 ± 2.5	39.6 ± 3.1	40.4 ± 1.5
NSC, % of DM	34.9 ± 1.6	4.8 ± 0.6	22.5 ± 0.7	21.5 ± 3.6	23.7 ± 1.4	24.2 ± 2.5

[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin.

starch content and unsaturated oil might have a stepwise negative effect on milk fat production, whereas rumen unsaturated FA increase, the risk of milk fat depression increases with monensin. In the current study, monensin concentration had no negative effect on milk fat production, rather milk fat content increased with monensin treatment due to the change in de novo and preformed fat synthesis. This finding is consistent with the expected increase in propionate production which would provide more energy for productive functions in the gland (Prange et al., 1978; Van Maanen, et al., 1978).

Milk FA profile results are in Table 5. The de novo and mixed FA concentration linearly increased in cattle fed monensin compared to CON but yields were not significantly different (P = 0.21) although there was a trend for a linear increase in both de novo (P < 0.06) and mixed FA (0.09). Both Duffield et al. (2008b) and Alzahal et al. (2008) reported a significant decrease in de novo FA concentration per total FA with monensin treatment, so the results of this experiment are not consistent with previous observations. The mixed FA yield and percent of total FA did not differ among treatment groups (P < 0.10), but mixed FA content linearly increased compared to CON (P = 0.02). The preformed FA concentration and yield were not different among treatment groups nor was preformed FA as a percentage of total FA. Alzahal et al. (2008) also found monensin treatment had no effect on preformed concentrations as a function of total FA. There was a trend for C16 concentration and yield tended to be greater (P = 0.09) with a significant linear effect of monensin consistent with the mixed FA results. The C18 and *cis*-9 C18:1 concentration and yield were not affected by monensin treatment. The biohydrogenation of oleic acid to stearic acid is achieved by gram-negative bacteria (Alzahal et al., 2008; Harfoot and Hazelwood, 1988) who, unlike gram-positive bacteria, are not inhibited by monensin treatment, therefore, this theory might explain the lack of treatment effects on stearic and oleic acid in the current study. The level of unsaturation of FA decreased with increasing monensin levels and was likely due to the level of de novo and mixed FA contents of the milk across treatments (P = 0.01; Table 5). All monensin treated groups approached a tendency for a reduction in FA chain length compared to CON (P = 0.11, 0.14, and 0.16 for R11, R14.5, R18, respectively) likely due to an increase in de novo synthesis in the monensin treated groups. Alzahal et al. (2008) and Fellner et al. (1997) suggest monensin has a role in inhibiting ruminal biohydrogenation which would reduce milk fat synthesis, but in the current study, the milk fat concentration levels, de novo FA levels, and FA unsaturation suggests that monensin treatment enhanced biohydrogenation in the rumen or had some effect on FA synthesis. An alternative observation is that monensin did not impact biohydrogenation and the increased concentration of saturated FA was related to the increase in de novo and mixed FAs which would dilute out the unsaturated FA given the level of milk fat yield. We did not measure other C18:1 or C18:2 isomers that would have given more insight into the effect of monensin on biohydrogenation, although the high levels of fat production and the reduction in FA unsaturation in monensin fed cows suggest monensin did not play a role in inhibiting biohydrogenation or milk fat synthesis in the current study.

Table 4. Effect of increasing dietary monensin concentration on lactation performance

	Diet[1]					*P*-value[2]			
Item	CON	R11	R14.5	R18	SEM	Linear	Quad	Trt	Trt x Wk
Days in milk[3]	190	168	193	184	7.2	-	-	-	-
Monensin, mg/d	0	384	465	589	-	-	-	-	-
DMI, kg/d	26.9	26.8	26.7	27.7	0.31	0.29	0.09	0.22	< 0.01
Milk, kg/d	39.3	39.9	39.7	39.6	0.34	0.48	0.38	0.69	< 0.01
Fat, %	4.60	4.67	4.71	4.66	0.04	0.16	0.40	0.38	0.16
Fat, kg/d	1.79	1.83	1.85	1.83	0.02	0.15	0.52	0.40	< 0.01
Protein, %	3.35	3.37	3.36	3.39	0.02	0.15	0.89	0.41	< 0.01
Protein, kg/d	1.30	1.33	1.33	1.33	0.01	0.13	0.46	0.41	< 0.01
Lactose, %	4.63	4.65	4.63	4.63	0.01	0.98	0.27	0.51	< 0.01
Lactose, kg/d	1.82	1.85	1.84	1.84	0.02	0.34	0.50	0.71	< 0.01
MUN, mg/dL	8.96[a]	10.24[b]	9.61[ab]	9.52[ab]	0.28	0.12	0.04	0.05	< 0.01
PUN, mg/dL	9.11	9.13	9.04	8.89	0.17	0.42	0.42	0.72	< 0.01
ECM[4], kg/d	46.0	46.9	47.1	46.8	0.50	0.17	0.47	0.46	< 0.01
3.5% FCM[5], kg/d	46.0	46.9	47.2	46.8	0.53	0.19	0.51	0.49	< 0.01
SCM[5], kg/d	42.5	43.3	43.5	43.2	0.46	0.17	0.41	0.42	< 0.01
BW, kg	692	691	694	693	2.1	0.74	0.67	0.83	0.26
BW change, kg/d	0.16	0.27	0.16	0.44	0.09	0.07	0.33	0.08	-
BCS[6]	2.93	2.93	3.04	2.93	0.40	-	-	-	< 0.01
Rumination, min/d	647	645	639	641	6.2	0.40	0.91	0.77	0.01

[a-b]Means within a row differ with different superscripts ($P < 0.05$).
[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin
[2]Week effect for all estimates ($P < 0.01$).
[3]Average of experimental period.
[4]Calculated according to Tyrell and Reid (1965).
[5]Calculated according to NRC (2001).
[6]Largest standard deviation of treatment means.

Table 5. Effect of increasing dietary monensin concentration on de novo, mixed, and preformed fatty acid production

Item	Diet[1]				SEM	*P*-value[2]			
	CON	R11	R14.5	R18		Linear	Quad	Trt	Trt x Wk
Total FA, g/100 g milk	4.33	4.39	4.43	4.37	0.04	0.22	0.34	0.41	0.31
De novo[3]									
g/100 g milk	1.13	1.16	1.17	1.16	0.01	0.05	0.32	0.17	0.35
g/d	438	452	458	454	6.3	0.06	0.46	0.21	0.06
g/100 g FA	26.1	26.4	26.2	26.3	0.11	0.24	0.54	0.41	< 0.01
Mixed[4]									
g/100 g milk	1.85	1.88	1.91	1.90	0.02	0.02	0.79	0.10	0.07
g/d	720	737	753	746	11.8	0.09	0.76	0.28	< 0.01
g/100 g FA	42.8	42.9	43.0	43.1	0.18	0.25	0.66	0.64	< 0.01
Preformed[5]									
g/100 g milk	1.34	1.35	1.36	1.33	0.02	0.95	0.27	0.61	< 0.01
g/d	520	527	533	521	7.1	0.61	0.28	0.54	< 0.01
g/100 g FA	31.0	30.7	30.8	30.6	0.21	0.15	0.98	0.46	< 0.01
Chain length	14.57	14.54	14.54	14.54	0.01	0.02	0.27	0.08	< 0.01
Level of unsaturation	0.235[a]	0.231[ab]	0.227[b]	0.227[b]	0.002	<0.01	0.94	0.01	< 0.01
Fatty acids									
16:0, g/100 g milk	1.79[y]	1.81[xy]	1.85[x]	1.83[xy]	0.02	0.02	0.74	0.09	0.07
16:0, g/d	695[y]	712[xy]	728[x]	720[xy]	9.6	0.02	0.67	0.09	< 0.01
18:0, g/100 g milk	0.36	0.36	0.37	0.36	0.01	0.80	0.33	0.60	< 0.01
18:0, g/d	140	142	145	141	2.3	0.35	0.26	0.32	< 0.01
18:1 *cis*-9, g/100 g milk	0.79	0.79	0.79	0.78	0.01	0.91	0.59	0.86	< 0.01
18:1 *cis*-9, g/d	305	308	311	306	4.0	0.57	0.42	0.66	< 0.01

[a-b]Means within a row differ with different superscripts ($P < 0.05$).
[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin.
[2]Week effect for all estimates ($P < 0.01$).
[3]C4 to C14 (Barbano and Melilli, 2016).
[4]C16, C16:1, and C17.
[5]Greater than or equal to C18.

The increase in de novo and mixed FA synthesis and yield in mid- to late lactation dairy cattle was an interesting and exciting observation and one that is not well documented. The increase in de novo and mixed FA through the feeding of monensin could be due to a couple different substrate supplies. Monensin is known to increase the supply of propionate and under certain conditions, propionate can be part of an initiation sequence where synthesis of acyl chains from carbon atoms could potentially lead to incorporation into chain elongation of FA (Palmquist, 2007). In addition, with increased propionate, there will be greater glucose and capacity for reducing equivalents which means increased NADPH +H supply which would allow for an increase in the FA synthase reaction allowing for production and elongation of FA. The protein sparing effect of monensin could increase the supply of certain amino acids, including the branched chain amino acids and their conversion to branched chain volatile FA and these could serve as precursors for chain elongation for chain lengths less than 16 carbons (Massart-Leen et al., 1981; Ha and Lindsay, 1990; Liu et al., 2018). Diets were not formulated to contain high quantities of fat, thus it is possible that with lower exogenous FA, there was less competition for certain enzymes related to glycerol production and utilization, but de novo FA synthesis could be increased. Finally, it is also possible, that some of the fat content and yield was related to the supply of methionine and lysine. In the current study, the methionine and lysine were supplied at what we believe are closer to the true requirements and, with the DMI observed, the metabolizable methionine level was approximately 85 g/d and the lysine levels were approximately ≥225 g/d, levels much higher than typically fed. This data would suggest that overcoming the limitation of at least two essential amino acids (EAA) allowed for greater milk fat synthesis in these cows. There is emerging data to suggest there is a link between mTOR signaling, EAA, and the regulation of milk fat synthesis (Li et al., 2016; Nichols et al., 2020).

There is a strong correlation between true protein yield and de novo FA content of milk (Barbano et al. 2019), demonstrating an integrated outcome of metabolism and the metabolic signaling related to nutrient supply (Lobley, 2007; Rius et al., 2010). Milk protein concentration and yield were unaffected by monensin treatment ($P = 0.41$; Table 4), however, milk protein content and yield were both high, and paralleled the de novo and mixed FA yields again likely due to some effects of the level of EAA fed in this study. Milk protein responses to monensin treatment have been inconsistent in many studies where some have reported a decrease (Akins et al., 2014; Martinez et al., 2009), no effect (Alzahal et al., 2008; McCarthy et al., 2015), or an increase in protein content with monensin feeding (Van Amburgh et al., 2008). A meta-analysis by Duffield et al. (2008b) found monensin reduced milk protein concentration but increased milk protein yield suggesting dilution effect might be a factor as monensin increases milk production (Alzahal et al., 2008; Ipharraguerre & Clark, 2003). Given the previously described protein sparing effect of monensin on ruminal feed digestion (Poos et al., 1979; Chen and Russell, 1991; Ruiz et al, 2001), under certain conditions it is possible when feeding monensin that more feed protein can escape fermentation and flow to the small intestine, which would provide more amino acids independent of any microbial yield effects. That outcome, combined with a shift in propionate production (Prange et al., 1978; Van Maanen, et al., 1978), could possibly result in an enhancement of milk protein yield. The milk lactose concentration and yield did not differ among treatment groups ($P = 0.51$ and $P = 0.71$, respectively; Table 4). In agreement with the current study, Akins et al. (2014) and Hagen et al. (2015) found monensin had no effect on milk lactose concentration.

Although non-significant, ECM, FCM, and SCM all increased with monensin treatment compared to CON likely from the increase in milk component production

in the monensin fed groups (Table 4). Previously, experiments by He et al. (2012) and Martinez et al. (2009) found monensin had no significant effect on component corrected milk yield. We observed an average 7 kg/d increase in ECM and FCM yield compared to actual milk yield across all treatment groups, and a 3.5 kg/d increase in SCM yield, again likely a result of the diet formulation of higher EAA levels, modest fat levels and strong rumen fermentation conditions. The CON group tended (P = 0.09) to have greater feed efficiency (actual milk/DMI) and R11 and R14.5 were significantly greater than R18 (P = 0.02 and P = 0.04, respectively) than R18 treatment due to the increased DMI of the cows on the R18 treatment (Table 6). However, there was a quadratic effect on ECM/DMI, FCM/DMI, and SCM/DMI by monensin treatment due to the level of DMI in the R18 treatment (Table 6). A couple of factors impacting the ability to identify differences in production efficiency are the numerical increase in DMI of the cows on the R18 treatment and the re-adjustment to the treatment diet following the covariate period as previously outlined. Although non-significant, the 0.8 kg difference in DMI of the cows on the R18 treatment obscured the typical outcome of enhanced feed efficiency at that level of monensin intake (Akins et al., 2014; Hagen et al., 2015), and likely more relevant, the re-adjustment to the CON diet from the covariate period appeared to impact treatment effects on milk yield. In the current study, monensin had no effect on estimated diet energy while Akins et al. (2014) and Hagen et al. (2015) reported an increase in estimated diet energy in cows fed 18 g/ton monensin compared to no monensin.

Milk urea nitrogen concentration was significantly greater in R11 compared to CON (P = 0.04), but not different in R14.5 or R18 (Table 4). Martinez et al. (2009) found monensin had no effect on MUN while Akins et al. (2014) reported an increase in MUN with monensin treatment. Additionally, McCarthy et al. (2015) reported significantly higher MUN values in early lactation cows who were fed diets top-dressed with monensin. Plasma urea nitrogen was unaffected by monensin treatment, although a meta-analysis(Duffield et al., 2008a) reported blood, plasma, and serum concentration increased with monensin treatment (Table 4). Recktenwald et al. (2014) suggests monensin plays a role in retaining urea N in the blood as they observed higher PUN values and larger plasma N pools with monensin treatment; however, that was not observed in the current study. The R11 and R18 treatment groups had a nonsignificant increase in BW compared to CON with R18 approaching a tendency to be greater (P = 0.11), although this observation warrants the recognition that wk 5 BW data is used to determine final BW due to an error with the scale (Table 4). In a previous study, Phipps et al. (2000) reported a significant increase in BW change with increasing levels of monensin. In the current study, BCS was not significantly different among treatment groups. This data suggests cows with few nutritional limitations will partition as much energy and nutrients towards milk production and away from BW and BCS gain even in later lactation as many of these cows were greater than 200 DIM while on treatment and not gaining appreciable amounts of weight or BCS. This observation requires further study and suggests BW accumulation in later lactation might be partially due to inadequate nutrient supply for milk and component yield, thus nutrients are retained in the tissue at a greater rate. Monensin treatment had no effect on rumination time and the values were quite high indicating good rumen health (Table 4).

CONCLUSION

Overall, the milk and component yield of these mid- to late lactation cattle was high and unprecedented suggesting the conditions of evaluating monensin feeding in cattle fed more contemporary diets was achieved. Increasing the supply of monensin had no significant effects on milk yield, DMI, or

production efficiencies; however, some of that lack of difference is likely due to shift from a covariate period with monensin feeding to a control diet where

Table 6. Effect of increasing dietary monensin concentration on milk production efficiency

	Diet[1]					*P*-value[2]			
Item	CON	R11	R14.5	R18	SEM	Linear	Quad	Trt	Trt x wk
Milk/DMI	1.47[a]	1.48[a]	1.48[a]	1.42[b]	0.01	0.11	< 0.01	0.02	< 0.01
ECM/DMI	1.71	1.74	1.76	1.69	0.02	0.63	0.04	0.13	0.13
3.5% FCM/DMI	1.71	1.74	1.76	1.70	0.02	0.66	0.04	0.13	0.12
SCM/DMI	1.58	1.61	1.62	1.56	0.02	0.71	0.03	0.10	0.09
Estimated diet energy[3]	1.64	1.65	1.65	1.68	0.02	0.34	0.49	0.62	-

[a-b]Means within a row differ with different superscripts ($P < 0.05$).
[1]CON = 0 g/ton monensin, R11 = 13 g/ton monensin, R14.5 = 15.8 g/ton monensin, R18 = 19.3 g/ton monensin.
[2]Week effect for all estimates ($P < 0.01$).
[3]Estimated diet energy content = [0.08 × BW, $kg^{0.75}$ + BW change, kg/d × 5.34 + milk, kg × (0.0929 × milk fat, % + 0.0563 × milk protein, % + 0.0395 × milk lactose, %)]/DMI, kg (NRC, 2001).

monensin was removed and an inadequate adjustment period. We observed a positive response to monensin treatment with linear increases in de novo and mixed FA concentration which resulted in enhanced milk fat yield. This indicates monensin can be fed at higher concentrations to achieve high milk component yields in lactating cows fed contemporary diets optimized for component yield, and more research is warranted to understand the relationship between monensin and ruminal FA synthesis, especially the de novo and mixed FA.

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Technical Symposium
Speaker Biographies

Matthias Hess, Ph.D. is an Associate Professor and the Head of the Systems Microbiology and Natural Product Discovery Laboratory in the College of Agriculture and Environmental Sciences at the University of California Davis. Prof Hess' research centers on the microbial processes and enzymes that shape complex anaerobic ecosystems, such as digestors and the cow's digestive system, and the development of sustainable approaches to optimize the function of these systems. Prof Hess was the first to successfully reconstruct genomes from the rumen microbes and he identified more than 27,000 novel enzymes that allow rumen microbes to break down recalcitrant plant biomass. He has also identified seaweed species that grow offshore the Californian Coast and that reduce enteric methane production. Prof Hess teaches Animal Biochemistry and Metabolisms, which is one of the largest classes offered at UC Davis, and he was awarded the Faculty Award for Outstanding Mentorship from the University Honors Program in 2021.

Mallory Embree, Ph.D. received her Ph.D. in Bioengineering from the University of California, San Diego. She is currently Chief Science Officer and the technical co-founder of Native Microbials. Her research has focused on developing new methodologies to study both natural and synthetic complex microbial communities. Her work has centered around the integration of bioinformatics with multiple-omics datasets, physiological measurements, and metabolic modeling (flux balance analysis) to examine microbial communities from a species-centric perspective. She also has extensive microbial cultivation experience, particularly in anaerobic microbiology, and has isolated and characterized a vast number of "unculturable" species from environmental samples. In addition to livestock and companion animal microbiomes, she has studied microbial communities from a diverse range of environments including liver-disease mouse models, human skin microbiomes, brewery wastewater digesters, slow-growing methanogenic alkane-degrading enrichments, and low-biomass deep subsurface sediments from the ocean gyres. Currently, her research has culminated in 27 peer-reviewed manuscripts and 94 patent filings (21 issued).

Alex Washburne, Ph.D. received his PhD in Quantitative and Computational Biology from Princeton University in 2015 studying the mathematics of competition in ecological, epidemiological, and economic systems. He went on to a post-doc at Duke University, developing novel statistical tools for microbiome data analysis. The mathematical tools developed during his research have been applied to a broad range of problems across fields ranging from ecology to finance. Most recently, his tools provided insights into which pathogens spilling over from which wildlife to people, and he has developed novel tools to evaluate the

competitiveness of variants of concern and forecast outbreak trajectories during the COVID-19 pandemic. He's now running a consulting company and, among other things, developing new methods to identify the microbial determinants of feed efficiency in livestock.

Lisa Marotz, Ph.D. obtained her Ph.D. in Biomedical Sciences from UC San Diego in 2020. During her graduate research, she developed novel protocols and computational tools to elucidate complex microbial communities through next-generations sequencing which led to more than 20 peer-reviewed publications. After graduating she spent a year as a postdoctoral fellow where she focused on scaling up rapid qPCR testing at the start of the COVID-19 pandemic and identify the microbial communities co-associated with the SARS-CoV-2 virus. Lisa joined Native Microbials in early 2021 where she designs and analyses both commercial and academic studies to validate novel dairy cow endomicrobials.

Michelle O'Malley, Ph.D. is a Professor in the Department of Chemical Engineering at the University of California, Santa Barbara and the Associate Director of UCSB's Bioengineering Program. She earned a B.S. in Chemical Engineering and Biomedical Engineering from Carnegie Mellon University in 2004 and a PhD in Chemical Engineering from the University of Delaware in 2009, where she worked with Prof. Anne Robinson to engineer overproduction of membrane proteins in yeast. O'Malley was a USDA-NIFA postdoctoral fellow in the Department of Biology at MIT. At UCSB, her research group engineers protein synthesis within anaerobes and consortia for sustainable chemical production, bioremediation, and natural product discovery. O'Malley's research has been featured on NPR's *Science Friday*, the *BBC Newshour*, the *LA Times*, and several other media outlets. She was named one of the 35 Top Innovators Under 35 in the world by *MIT Technology Review* in 2015, one of the 10 "Scientists to Watch" by *Science News* in 2019 and is the recipient of the Presidential Early Career Award for Scientists and Engineers (PECASE) – the highest honor bestowed on early career scientists by the US government. She is also the recipient of the Allan P. Colburn Award from the AIChE, the ASM Award for Early Career Applied and Biotechnological Research, the AIChE Division 15 Early Career Award, a DOE Early Career Award, an NSF CAREER award, the Camille Dreyfus Teacher-Scholar Award, the ACS BIOT Division Young Investigator Award, an ACS PMSE Division Young Investigator Award, an ACS WCC "Rising Star" Award, and a Hellman Faculty Fellowship. She was elected to the American Institute of Medical and Biological Engineers in 2020 as is the Chair-Elect of the ACS Division of Biochemical Technology (BIOT).

CANC Speaker Biographies

Andy Stumpf is a retired Navy SEAL, professional skydiver and base jumper, public speaker, and host of the podcast "Cleared Hot."

Michael Ballou, Ph.D. is the Professor and Chair of the Department of Veterinary Sciences in the Davis College of Agricultural Sciences and Natural Resources at Texas Tech University. His research program is focused on understanding how nutrition and management influence the health of calves, heifers, and transition cows. Michael received his Ph.D. in Nutritional Immunology from UC Davis in 2007. He has authored or co-authored over 80 peer-reviewed publications and given over 125 invited presentations.

Ed DePeters, Ph.D. is a ruminant nutritionist and Distinguished Professor of Animal Science at UC Davis. He teaches Animal Nutrition (NUT 115) and Dairy Cattle Production (ANS 146). His research areas involve dairy cattle nutrition including evaluation of by-product feedstuffs and the impact of nutrition on milk production and composition. He is also a Master's Advisor for the Animal Science major.

Albert De Vries, Ph.D. is a professor in the Department of Animal Sciences at the University of Florida. He grew up on a dairy and swine farm in the Netherlands. He went to Wageningen University where he received a BS and MS in Animal Science with a minor in agricultural economics in 1991. In 1995, he came to the US to pursue a Ph.D. in Animal Sciences at the University of Minnesota in St. Paul with a focus on dairy science, applied economics, operations research, and statistics. After graduation in 2001, Albert accepted a faculty position at the University of Florida in Gainesville. He currently teaches two undergraduate dairy courses and advises undergraduate dairy students and graduate students. His research interests are in optimization of culling and replacement strategies, statistical process control, economics of reproduction and genetics, and precision dairy farming. In his extension role, he works with the allied dairy industry and dairy farmers on farm financial management and to apply the results of dairy systems management research. Albert is married to Kim who is a small animal veterinarian. Together they have twin daughters Grace and Karen and four cats. They live near Newberry, Florida.

Juan M. Piñeiro, Ph.D. grew up in Argentina, worked with beef cattle at his family cow calf operation and obtained his D.V.M. degree in 2012 at the University of La Plata. He moved to the U.S. in 2013 and worked in dairies located in Texas and Colorado for a year. In 2016 and 2018 he obtained his M.S. and Ph.D. degrees at The Ohio State University. Dr. Piñeiro is currently an Assistant Professor and Texas A&M AgriLife Extension Service Dairy Specialist in the Animal Science Department, Texas A&M University. During his professional career, his research efforts focused on the impact, prevention and treatment of transition period diseases of dairy cows. More recently he has been involved in research trying to increase the starch digestibility of forage sorghum hybrids. He has taught as a guest lecturer on five Animal Science and Veterinary Medicine courses at three different universities, is a faculty member for the U.S. Dairy and Education Training Consortium and has expertise training dairy farm personnel. As the co-chair of the High Plains Dairy Conference and Southwest Dairy Day and a committee member of five

associations, he is involved in organizing several conferences and field days for dairy farmers, allied industry members, academia and local communities. Piñeiro received over $250,000 from five funded grants and gifts, published eleven peer-reviewed journal articles, 30 extension articles and has been involved in over 50 Extension and educational activities.

Mike Van Amburgh, Ph.D., is a Professor in the Department of Animal Science and a Stephen H. Weiss Presidential Fellow at Cornell University where he has a dual appointment in teaching and research. His undergraduate degree is from The Ohio State University and his Ph.D. is from Cornell University. He teaches multiple courses and leads the Cornell Dairy Fellows Program, advises approximately 50 undergraduate students and is the advisor for the Cornell University Dairy Science Club. Mike currently leads the development of the Cornell Net Carbohydrate and Protein System (CNCPS/CPM Dairy), a nutrition evaluation and formulation model used worldwide. Through the modeling effort, he focuses on enhancing the efficiency of nutrient use by ruminants to improve the environmental impact of animal food production. A significant component of his current work is to understand whole animal and ruminal nitrogen metabolism and amino acid supply and requirements to enhance the development of the Cornell Net Carbohydrate and Protein System. Further, his group is active in developing methods to better describe the interaction between forage and feed chemistry, rumen function and nutrient supply to compliment the model. He has authored and co-authored over 100 journal articles and many conference proceedings and is the recipient of several awards including the American Dairy Science Foundation Scholar Award, the Land O'Lakes Teaching and Mentoring Award from ADSA, the American Feed Ingredient Association Award for Research, Journal of Dairy Science Most Cited Award, the CALS Professor of Merit Award and the CALS Distinguished Advisor Award. In 2016, he was named a Stephen H. Weiss Presidential Fellow, the highest teaching award given by Cornell University.

Alexander N. Hristov, Ph.D., is a Distinguished Professor of Dairy Nutrition in the Department of Animal Science at The Pennsylvania State University and is a member of several professional societies and of the Feed Composition Committee of the U.S. National Animal Nutrition Program. He has a Ph.D. in Animal Nutrition from the Bulgarian Academy of Agricultural Sciences and has worked as a research scientist in his native Bulgaria, USDA-ARS Dairy Forage Research Center in Madison, WI, and the Ag Canada Research Center in Lethbridge, AB. He was on the faculty at the Department of Animal and Veterinary Science, University of Idaho from 1999 to 2008 and is at Penn State since 2008. Hristov's main research interests are in the areas of protein/amino acid nutrition of dairy cattle and mitigation of nutrient losses and gaseous emissions from dairy operations. He is currently the co-Chair of the Network on Feed and Nutrition in Relation to Greenhouse gas Emissions, which is an activity of the Livestock Research Group within the Global Research Alliance on Agricultural Greenhouse Gases and is on the Scientific Advisory Board of the European Joint Programming Initiative on Agriculture, Food Security and Climate Change (FACCE-JPI). Dr. Hristov initiated and led the development of the world's first Dairy MOOC (Massive Open Online Course) with a current enrollment of over 40,000 (https://www.coursera.org/learn/dairy-production). He has given over 90 invited presentations and has published over 220 books, book chapters, and peer-reviewed journal articles.

California Animal Nutrition Conference
2022 Steering Committee

Chairperson: Zachery Meyer
Zachery Meyer was raised in Ixonia, Wisconsin. He grew up immersed in his family's business, Rock River Laboratory. Meyer spent many hours helping in various jobs around the laboratory, seeing first-hand the dedication and commitment his father and the late Twilah Kulow had to the business and their customers. Meyer gathered business experience at Clear Channel and GE Medical while working toward his degree from the University of Wisconsin-Milwaukee. In 2007, Meyer resumed his involvement in Rock River Laboratory, starting as a soil sampler, moving to outside sales, and eventually taking on his current role of director of operations. Meyer still gathers inspiration from the Rock River Laboratory employees and mentors who cultivated his drive for customer satisfaction and service, while continuing to learn and deepen his understanding of animal nutrition and agronomy. When he isn't building relationships with customers or overseeing laboratory operations, Meyer spends his time playing or watching sports and sharing family time with his wife and two young daughters.

Vice Chairperson: Ruben Almada, B.A.Sc., Turlock Dairy & Refrigeration
Ruben Almada was born and raised in the Hilmar, California area. Growing up on a dairy steered him toward a life in the dairy world. He graduated from California Polytechnic State University-San Luis Obispo in 2006 with a Dairy Science Degree. Upon completing his degree Ruben joined Cargill Animal Nutrition in the fall of 2006 where he was a Dairy Management Consultant for 3 years. He then joined Kemin Animal Health and Nutrition in July of 2010 as the Key Account Manager covering California and Arizona for the Dairy Segment. In April of 2021, Ruben joined Turlock Dairy & Refrigeration as a Farm Management Support Specialist helping dairymen merge cows with automation. He is married to his wonderful wife, Jennifer, and they have two children, Kinley (8) and Jaxson (6).

Ex-Officio: Jennifer Heguy, M.S., P.A.S. – UCCE Farm Advisor
Jennifer Heguy is a native of California's San Joaquin Valley. She received her B.S. in Animal Science, with an emphasis in Livestock and Dairy, at the University of California, Davis. In 2006, she received her M.S. degree at UC Davis, focusing on dairy cattle nutrition. Jennifer currently serves as the University of California Dairy Farm Advisor in Merced, Stanislaus, and San Joaquin Counties where milk is a major agricultural commodity. Jennifer's research and extension program focuses on feeding management practices.

Committee Members:

Brian Rainey, M.S., MBA, P.A.S., Pine Creek Nutrition Service, Inc.
Upon graduating from Kansas State University, Brian made a gradual progression west seeking career fulfillment in working hands-on with livestock producers. Brian joined Pine Creek Nutrition Service, Inc. in May 2010 and brings a science, business, and industry portfolio to the consulting staff. Brian received a Bachelor of Science degree in Animal Science in 2001 from Kanas State University, Manhattan, KS, a Master of Science in Ruminant Nutrition in 2004 from Montana State University, Bozeman, and a Master of Business Administration, with Distinction, Phi Kappa Phi, May 2010 California State University, Fresno.

Kyle Thompson, Ph.D.
Kyle Thompson received his B.S. degree in animal science from Fresno State (2006) and his master's and Ph.D. degrees in animal science from Oklahoma State (2011/2015). He joined the Fresno State staff in the fall of 2016 after taking classes and teaching at Oklahoma State from January 2007-June 2016 and serving as the graduate student assistant manager of the campus dairy cattle center. His research included dairy nutrition research trials and lactating cow probiotics. He also assisted in research for bovine respiratory disease, rumen temperature bolus, milk production by weigh-suckle-weigh, and swine antimicrobial replacements. He also assisted in 4-H and FFA Field Day dairy judging competitions. While in Stillwater, OK, he owned and operated Wild Acre Farms and Exotics, which raised ewes, game birds, free-range hens, and other fowl/animals, and produced grasses and winter wheat for grazing and hay production. As a Fresno State student, he worked in the sheep unit for three years, served as a campus farm tour guide, and dairy unit herdsman and feed/hospital technician. He also worked as an exotic animal nutrition intern (2009) and a global nutrition fellow at the San Diego Zoo (2013).

Beau Williamson, B.S., Adisseo USA Inc.
Beau Williamson was born and raised in Clovis, California. Being the son of an animal science professor – Scott Williamson, Ph.D – he grew up around animal agriculture. His passion for animal agriculture grew through his involvement with 4-H and FFA. This led him to attend school at Purdue University, where he graduated with a degree in agricultural economics. Beau began his career in the dairy industry at Elanco Animal Health and he currently works for Adisseo, a global leader in animal nutrition. Beau works with nutritionists and dairymen to help them meet key nutritional needs of their animals. When he is not working, he loves spending time with his family, being involved in his church, and enjoying the great outdoors.

Joanne Verstuyft, B. A. Sc., Zinpro Corporation
Joanne Verstuyft was born and raised on her grandparent’s ranch in the East Bay near El Sobrante, CA. Joanne started early in horses and cattle with her grandfather and uncle’s influence. She competed in 4-H and jackpots with her horses and purebred Angus cattle at a young age. Joanne graduated from California Polytechnic University-San Luis Obispo in 2003 with an Agriculture Business degree concentration in marketing and beef cattle. After two successful college internships with Elanco, she joined Elanco as a Beef Cattle Sales Associate covering Western Nebraska, Northeastern Colorado, and Wyoming calling on feedlots and cow-calf operations. She returned to California as a Pharmaceutical Sales Representative for Lilly,

and in late 2009, she joined Elanco's Dairy Team promoting rBST in the Central Valley. After fifteen years with Elanco, Joanne left to work for Pinnacle Premix in sales covering California and Arizona. In January 2019, Joanne joined Zinpro Corporation as an account manager covering dairy and equine in California. She promotes Zinpro performance minerals in sharing data while also providing farm support, lameness evaluations, and hoof trimming. Joanne enjoys working in the animal agriculture industry and matching her passion and career. Joanne lives in the Bay Area, where she enjoys riding her horse and spending time with family and friends.

Levi Schwieterman, Native Microbials – No biography was provided.

California Animal Nutrition Conference History

YEAR	CHAIRPERSON	COMPANY AFFILIATION
2022	Mr. Zachery Meyer	Rock River Laboratory, Inc.
2021	Jennifer Heguy, M.S., P.A.S.	University of California, Coop. Ext.
2020	NO CANC CONFERENCE	
2019	David Ledgerwood, M.S., P.A.S.	Chr-Hansen
2018	Jason Brixey, M.S., P.A.S.	Pine Creek Nutrition Service
2017	Dr. Phillip Jardon, DVM, MPVM	Elanco Animal Health
2016	Dr. Phillip Jardon, DVM, MPVM	Elanco Animal Health
2015	Mr. Ben Tarr	Adisseo USA Inc.
2014	Dr. Jeffrey M. DeFrain	Zinpro Performance Minerals
2013	Mr. Doug DeGroff	Diversified Dairy Solutions, LLC
2012	Mr. Eduardo Galo	Novus International, Inc.
2011	Dr. Michael A. DeGroot	DeGroot Dairy Consulting
2010	Dr. Jim Tully	Pine Creek Nutrition Service, Inc.
2009	Mr. Michael Braun	Phibro Animal Health
2008	Dr. Luis Rodriguez	Zinpro Corporation
2007	Dr. Marit Arana	A.L. Gilbert Company
2006	Mr. Dennis Ervin P.A.S.	Prince Agri Products, Inc.
2005	Dr. Lawson Spicer	Nutri Management Inc.
2004	Dr. Luis Solorzano	Purina Mills, Inc.
2003	Dr. Alfonso Mireles, Jr.	Foster Farms
2002	Mr. Edmund Vieira	Pine Creek Nutrition Service, Inc.
2001	Dr. Melinda Burrill	California State Polytechnic University - Pomona
2000	Mr. Dave Fischer	Foster Farms
1999	Dr. M. Steven Daugherty	California State Polytechnic University - SLO
1998	Dr. Doug Dildey	Alltech, Inc.
1997	Ms. Carla Price	Nutritionist
1996	Dr. H.John Kuhl, Jr.	Nest Egg Nutrition
1995	Mr. Dennis Ralston	M. Rinus Boer Co., Inc.
1994	Dr. Doug Dildey	Alltech, Inc.
1993	Dr. Mark Aseltine	ConsultingAnimal Nutritionist
1992	Dr. Carl Old	MacGowan-Smith Ltd.
1991	Mr. Nick Ohanesian	Ohanesian & Associates
1990	Mr. Rod Johnson	M. Rinus Boer Co., Inc.
1989	Mr. Timothy Riordan	Nutri-Systems, Inc.
1988	Dr. Russ W. Van Hellen	Great West Analytical
1987	Dr. John E. Trei	California State Polytechnic University, Pomona
1986	Dr. A.A. Jimenez	Ancon, Inc.
1985	Dr. Wm. A. Dudley-Cash	Foster Farms
1984	Dr. Joel Kemper	Penny-Newman Co.
1983	Dr. Alex J. Kutches	O.H. Kruse Grain & Milling Co.
1982	Dr. Howard Waterhouse	Bell Grain & Milling
1981	Mr. Don Ulrich	Diamond Shamrock Chemical Co.
1980	Mr. Tom Geary	PMS-West, Inc.
1979	Dr. Frank Parks	Kemlin Industries
1978	Mr. Fred Pfaff	Zacky Farms
1977	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.
1976	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.

California Animal Nutrition Conference History - Continued

YEAR	CHAIRPERSON	COMPANY AFFILIATION
1975	Dr. R.D. Hendershott	Nulaid Foods
1974	Dr. R.D. Hendershott	Nulaid Foods
1973	Dr. Leland Larsen	Nutri-Systems, Inc.
1972	Dr. Leland Larsen	Nutri-Systems, Inc.
1971	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.
1970	Mr. Fred Pfaff	Balfour Guthrie
1969	Mr. Fred Pfaff	Balfour Guthrie
1968	Mr. Fred Pfaff	Balfour Guthrie
1967*	Mr. Gary L. Frame	J.G. Boswell Co.
1966*	Mr. Gary L. Frame	J.G. Boswell Co.
1965*	Mr. Arne Jalonen	Topper Feed Mills
1964*	Mr. Arne Jalonen	Topper Feed Mills
1963*	Dr. W.P. Lehrer	Albers Milling Co.
1962*	Dr. H.J. Almquist	The Grange Co.
1961*	Dr. H.S. Wilgus	The Ray Ewing Co.
1960*	Mr. Bert Maxwell	Nulaid Foods
1959*	Mr. Bert Maxwell	Nulaid Foods
1958*	Mr. Robert Caldwell	Anderson Smith Milling Co.
1957*	Mr. Emery Johnson	P.C.A., Los Angeles
1956*	Mr. Emery Johnson	P.C.A., Los Angeles
1955*	Dr. H.J. Almquist	The Grange Co.
1954*	Dr. H.J. Almquist	The Grange Co.
1953*	Mr. Clifford Capps	California Milling Co.
1951*	Mr. Dolph Hill	Golden Eagle Milling Co.
1950*	Dr. H.J. Almquist	The Grange Co.
1949*	Dr. H.J. Almquist	The Grange Co.
1948*	Dr. H.J. Almquist	The Grange Co.

* California Animal Industry Conference

History of the California Animal Nutrition Conference

The California Animal Nutrition Conference (CANC) originated in the 1940s as the California Animal Industry Conference, sponsored by the California Grain & Feed Association (CGFA). CGFA wanted to expand the continuing education program into a forum encompassing animal health, nutrition, and management. The expectations were that communications between (nutritionists) industry, educational institutions, and regulatory agencies would be improved. In 1972, CGFA discontinued sponsoring the Animal Industry Conference.

After the conference was discontinued, a small group of nutritionists began meeting annually in Fresno. Two or three invited speakers from industry or the universities presented information on nutrition, especially poultry.

In 1975 a set of organizational bylaws were developed by the steering committee. CANC was established and was provided support by CGFA. The CGFA Board of Directors appointed a chairperson annually and approved the steering committee. In 1978, Dr. Frank Parks, the Chairperson, requested that CANC be granted independent status and be established as a self-governing committee of CGFA. This request was granted.

For a few years, meetings were held in Fresno and Corona, California. For a couple of years starting in 1978, CANC published “Nutri-Facts,” a “newsletter” consisting of articles on animal production.

In 1979, donations were requested from industry companies to help keep registration fees low. During the 1980s and through the 1990s the attendance at CANC continued to grow as the quality of the conference improved and the conference became known nationwide. In the 1990s a pre-symposium was added. The pre-symposium is sponsored by a company selected by the CANC Steering Committee and this process allows the selected company to showcase its research and products. In the year 2000, posters on research by students were included.

Attendance at the conference has grown from 50 in the 1970s to over 300 attendees. To encourage attendance, different activities have been tried such as keynote speakers, skiing expeditions, and a very successful barbeque dinner put on by the Animal Science Department at California State University, Fresno.

The California Grain & Feed Association has supported and allowed CANC to work and grow. The premise of the CGFA and CANC relationship is to work together to educate the feed industry with information for problem-solving and to disseminate valuable research information. CANC is not an industry, university, or government entity, but a committee collectively working together for the good of agriculture in California.