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HONEY BEE NUTRITION A REVIEW AND GUIDE TO SUPPLEMENTAL FEEDING



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Photo Credit: Dr. Jennifer Tsuruda, University of Tennessee Knoxville.

ABOUT THE HONEY BEE HEALTH COALITION

The Honey Bee Health Coalition was formed in 2014 as a cross-sector effort to promote collaborative solutions to honey bee health challenges. The diverse Coalition brings together diverse stakeholders including beekeepers, growers, researchers, government agencies, agribusinesses, conservation groups, manufacturers and brands, and other key partners dedicated to improving the health of honey bees and other pollinators. The Coalition's mission is to collaboratively implement solutions that will help to restore and enhance the health of honey bees while also supporting the health of native and managed pollinators in the context of productive agricultural systems and thriving ecosystems.

A major tenet and founding principle of the Coalition is the recognition that the current decline in overall honey bee health is a multi-factorial problem, and all stakeholders have a role to play in managing bee health issues. The Coalition is focusing on accelerating improvement of honey bee health in four key areas: forage and nutrition, hive management, crop pest management, and outreach, education and communications. As part of the hive management focus area, the Coalition has developed this "Tools for Varroa Management" Guide that beekeepers can use to help focus on more effectively controlling the varroa mite in managed hives.

For more information on the Coalition and its key focus areas/products, please visit: <u>http://honeybeehealthcoalition.org/</u>

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1. INTRODUCTION

Honey bee nutrition is complex, varies seasonally and is based on the colony needs and beekeeping practices. When considering the nutritional requirements of the colony, the demands are diverse due to the complex social structure of the colony (reproductive division of labor and a distinct caste system). The overall nutritional needs of the colony vary from the nutritional demands of the larvae to those of the adults. The requirements will also vary depending on the caste (queens, workers and drones) and the age of the adults (for example young nurses versus the foragers). The beekeeper's needs can also drive the nutritional demands of the colony, for example, ramping up brood production in late winter for almond pollination requires increased feeding of the colony.

Honey bee colonies are called superorganisms because the large number of individual members (the honey bees) function as a single entity (the colony).

Foragers scout the landscape for floral resources looking for pollen and nectar. Pollen and nectar provide a wide array of vital macro and micronutrients that honey bees need. In the absence of pollen and nectar, beekeepers need to provide supplemental feeding to the colonies depending on the colony brood status, the nutritional needs of the colony in preparation for the season (for example, spring feeding versus fall feeding) and the food reserves in the hives. Typically, supplemental feeding involves either feeding proteins (pollen substitutes/pollen supplements) or sugars (syrup most commonly) or a combination of both proteins and sugars.

Protein patties can be either pollen-based (pollen supplements) or non-pollen based (pollen substitutes). Traditionally, it is believed that pollen-based supplemental protein patties can be given to stimulate brood production, especially

Pollen substitute patties contain no pollen and pollen supplement patties contain some pollen. While gamma irradiation is shown to remove pathogens on pollen, it can also potentially remove beneficial microbes. More information is needed to understand the holistic effect of radiation on the pollen nutritional quality.

when queens may not lay eggs typically in the season. While both pollen supplements and pollen substitutes provide the proteins (and lipids) required by bees, pollen has additional nutrients that may be scant or absent in the pollen substitute diets. Pollen can also act as a phagostimulant and thus adding it to the supplemental diets can improve the diet consumption in addition to enhancing the nutritional profile of the supplement. However it must be considered that natural pollen is difficult to procure in bulk at a reasonable cost and it may also carry the risk of pathogen transmission and pesticide exposure.

The history of supplementing colonies with diets other than pollen goes back to 1655 when Samuel Hartlib suggested a supplemental diet using dry meal or bean flour, bread and ale. Later in 1875, Amos Ives Root tested various supplements with corn meal, buckwheat, rye meal, wheat flour and syrups. By then it was evident that colonies stopped rearing brood after the pollen store was exhausted and no amount of winter supplemental "meal" resulted in spring brood until pollen was reintroduced. In fact, it was also recorded that colonies craved supplemental feeding ("meals") only if they lacked pollen stores inside the hives. Later, Mykola H. Haydak and Elton W. Herbert Jr. were the forerunners of pollen substitute research in the United States. This early pollen substitute research laid the foundation for the supplemental feeding practices that are currently used. However, it must be noted that a pollen diet has a higher nutritional value owing to the various macro and micronutrients present in pollen, and this currently cannot be replaced with any artificial supplements. However, when pollen is scarce in the landscape, supplemental feeding can be important to sustain colonies. Similar to pollen, nectar also has diverse micronutrients, in addition to carbohydrates, and syrup feeding alone will not be sufficient. Nevertheless, in times of nectar dearth, feeding supplemental sugar is also important.

2. OVERVIEW OF MACRO AND MICRONUTRIENTS

Macronutrients are important for bees and are required in larger amounts when compared with micronutrients. Micronutrients are equally important, even though they are required in much smaller quantities. Examples of macronutrients are proteins, carbohydrates and lipids. Examples of micronutrients are phytosterols, vitamins, minerals, phytochemicals etc. All vital macro- and micronutrients are needed in varying quantities to provide optimal nutrition to honey bees.

Proteins:

Generally, the protein content of pollen varies from 2.5% to 61%. A colony with 50,000 individuals collects about 312.5 lb (~142 kg) of pollen annually. Maximum protein consumption is by young nurse bees (65 mg per bee over ten days) to meet the physiological demands of brood food production. Most protein consumed is at day 5 post eclosion. For rearing just one larva, approximately 25 mg – 37.5 mg protein is required.

If the essential amino acids are not in the right proportions, even if the total protein content is 20% or more, the utility of the artificial diet will be limited to that of the limiting essential amino acids. However it can still be helpful even if it is less ideal.

Approximately 23%-30% crude protein content in the diets is optimal for feeding a honey bee colony. When purchasing or preparing any protein supplement for the colony, it is important to note that over 20% protein content is recommended when rearing brood. Proteins are composed of amino acids, the building blocks required for colony growth and development, and ten essential amino acids in specific ratios are important to sustain brood rearing (Table 1). With age, workers in

The ten essential amino acids are: arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine. a colony transition from high essential-amino acid diets to predominantly relying on carbohydrate diets to meet the demands of foraging.

Table 1: The proportions of the ten essential amino acids (as a proportion of tryptophan) required for optimal honey bee nutrition is shown in this table as reported by deGroot in 1953. For example, for every tryptophan, we need 3 arginine, 1.5 histidine, 4 isoleucine, etc. The table

is adapted from Hoover et al. 2022 (Appendix C).

Essential amino acids	Required proportions (compared with tryptophan)
Arginine	3
Histidine	1.5
Isoleucine	4.0
Leucine	4.5
Lysine	3.0
Methionine	1.5
Phenylalanine	2.5
Threonine	3.0
Tryptophan	1.0
Valine	4.0

Lipids:

Pollen lipid content varies from 1% - 20%. Important lipid groups such as fatty acids (oleic acid, omega 3 fatty acids, omega 6 fatty acids) can improve honey bee learning and form a crucial component of the total pollen lipids. When given a choice of artificial dietary supplements in a laboratory cage setting, nursing age honey bees consumed foods to attain a ratio between 1:2 and 1:3 for essential amino acids to lipids (approximately 1.25:1 protein to fat ratio). Research has also documented the importance of pollen protein to lipid ratios which may determine bee foraging in a landscape. The total lipid content for supplemental protein feed is recommended to be between 5% and 8%.

Micronutrients:

Some of the important micronutrients required in supplemental diets are phytosterols, salts, vitamins etc. Phytosterols are important micronutrients shown to improve honey bee longevity, enhance brood rearing and improve overall bee physiology. Pollen is the only natural source of phytosterols for bee pollinators. For honey bees, 24-methylenecholesterol is the most vital phytosterol that must be present in the diet (pollen or supplements). A minimum dry diet weight of 0.5% 24-methylenecholesterol is ideal. Other micronutrients, such as phytochemicals (phenolic acids) and flavonols (kaempferol, p-coumaric acid and quercetin) present in nectar are also important for improving honey bee longevity, reducing pathogenic infection, counteracting pesticide stress and increasing nurse honey bee hypopharyngeal gland size. Salts are equally important for the honey bee diet and pollen typically contains higher potassium concentrations than sodium. Honey bees source necessary salts from pollen. Studies have shown that honey bee foragers collect brackish water to compensate for dietary salts during times of pollen scarcity. However, excessive levels of sodium chloride, sodium and calcium are toxic for honey bees. Nurse honey bees require essential vitamins for rearing brood. These include ascorbic acid, pyridoxine, nicotinamide, thiamine, riboflavin, pantothenic acid, biotin and folic acid. Pantothenic acid is also important for caste differentiation (queen and workers). Pollen provides the necessary minerals and vitamins for honey bees. When considering artificial diet formulation, it is crucial to consider adding micronutrients for creating a holistic diet.

3. CARBOHYDRATES

Nectar provides the necessary carbohydrates that honey bees require to meet their energy needs. A worker honey bee needs approximately 11 mg of dry sugar daily which translates to approximately 2 lb (~0.91 kg) of 50% sugar syrup or 0.66 L of honey per day for a large colony of 50,000 individuals. When we consider the additional energy required for brood rearing and other chores, a large colony can easily require over 700 lb (~317.5 kg) of nectar/syrup each year (assuming 50% sugar concentration for nectar or syrup). Since floral nectar typically contains less than 50% sugar, this only emphasizes the large volume of nectar that worker bees need to forage for in a year. Adult honey bees can safely consume

50% sugar syrup is 1:1 ratio of sugar and water by weight.

and utilize glucose, fructose, sucrose, trehalose and maltose. However they are not able to digest starch and dextrins. Carbohydrates such as lactose, galactose and raffinose are toxic to honey bees and should be avoided in supplemental feeding.

4. SUPPLEMENTAL FEEDING Commercial Diets:

Pollen is mixed with glandular secretions and this mixture is then carried on the corbiculae of foraging worker honey bees and stored in the hive as bee bread (Figure 1). Some research has shown that unlike pollen, commercial diet patties are not stored in the colonies. In addition, commercial diets are eaten by a proportion of the adult honey bees in a hive and these patties are not directly fed to the larvae. Commercial diets will not be as attractive as multifloral pollen and these diets can never truly replace pollen for optimal nutrition. When supplementing or substituting pollen, beekeepers often rely on purchasing commercial diets or make protein patties at home with commonly available recipes containing soy flour, brewer's yeast, soybean flour, egg powder, pea protein, potato protein, corn meal, milk products etc. When providing colonies with protein supplements, seasonal colony needs vary. Traditional patties have high protein contents and help boost colony growth during spring. Winter patties, on the other hand, have lower protein contents and help provide enough nutrition for overwintering workers. Providing pollen supplements in fall and winter may boost queen egg-laying and is recommended only in the warmer winter regions such as the southwestern or the southeastern United States.

Some advantages of winter patties lacking pollen:

- 1. Queen gets much needed rest from constant egg laying.
- 2. Less brood production leads to fewer workers produced in fall/winter.
- 3. Smaller cluster of workers can stay warm at the center.
- 4. Break in brood also means break in mite cycles.
- 5. Smaller cluster also conserves the winter food stores.

Figure 1: Bee bread stored inside a hive. Image courtesy Dr. Jennifer Tsuruda, University of Tennessee-Knoxville.



A good pollen substitute for honey bees should ideally contain all the nutrients that pollen has and that honey bees need. While commercial diets in the past have focused primarily on proteins and sugars, we now recognize the importance of considering vital macronutrients (such as lipids) and micronutrients (such as vitamins and minerals) as a part of the wholesome diet (Table 2). Micronutrients (such as phytosterols) are as important as macronutrients, and commercial diet formulations are frequently micronutrient-limited. Commercial protein diets can either be in a ready-to-use patty form or they can be sold as dry formulations with additional instructions on patty formulation. The patties are usually placed on the top of the brood chamber (ideally right over the brood nest or cluster). Often, sugar syrup and vegetable oils are added to the dry powders when making a moist patty. Some vegetable oils, such as borage, can be a good source of 24-methylenecholesterol and may be added to boost the phytosterol and lipid profiles of the supplemental feeds. While some beekeepers also add essential oils to the patties, there is a lack of peer-reviewed scientific research on the nutritional benefits of these essential oil products. If the supplemental proteins are provided in a dry powder form, it is not recommended to place the powder inside the hive. Rather, a large dry powder feeder can be placed outside the colony and the foragers will collect this as they would for pollen naturally. However this must be protected from rain/moisture (Figure 2). Dry powder form of protein feeding works when natural

Figure 2. Feeder for supplementing honey bee colonies with dry protein powder formulation. Image courtesy Dr. Shelley Hoover, University of Lethbridge.



pollen is absent in the landscape. This feeding method also discourages robbing as the honey bees are more interested in sourcing the dry feed from the outside feeders enabling the beekeepers to work with the colonies.

The size of the pollen substitute or pollen supplement patty is also important. Giving a small quantity of the patty to a rapidly growing colony, especially when natural forage is limited, will hinder colony growth. This is because if the colony needs sufficient proteins to grow, limited pollen in the landscape and limited supplemental feeding will not be beneficial. In addition, if the colony rears a lot of brood and then encounters a food shortage, the workers may cannibalize the young larvae and cap older larvae early. On the other hand, a leftover unconsumed patty can attract pests and mold. In southeastern regions of the United States for example, it is important to provide pollen supplements or substitutes (in patty form) in smaller portions as large patties, if uneaten for several days, will encourage small hive beetle reproduction inside the hives (Figure 3). Checking the rate of consumption (change in diet mass over time) of the patties may be a good indication of the palatability of the supplemented patties. However, consumption, nutritional quality and digestibility are not the same and this is discussed more in section 7. Also, consumption will drop if pollen sources become available, indicating a beekeeper no longer needs to provide supplements while the honey bees are bringing in pollen.

Figure 3: (A) White arrow indicates small hive beetle eggs that were laid in uneaten protein patties. Image courtesy Dr. Jennifer Tsuruda, University of Tennessee-Knoxville. (B) In such instances, providing a small patty is more reasonable to ensure complete consumption. Image courtesy Angus Catchot III, Pollinator Health and Apiculture Lab, Mississippi State University.

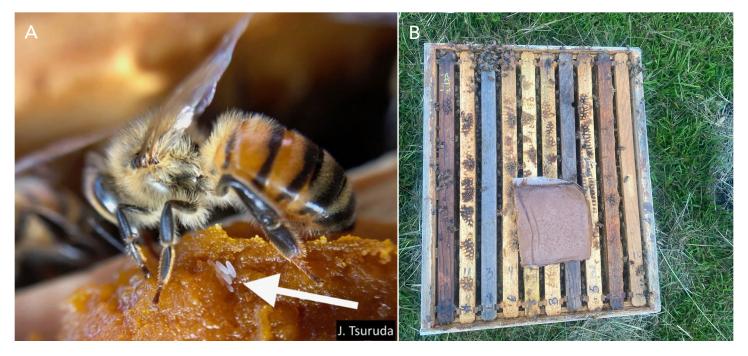


Table 2: This table lists some commonly found commercial diets (protein supplements and protein substitutes) sold in North America. Additional product nutrition information is available online for <u>Global Patties</u>, <u>Bee Pollen-Ate</u>, <u>Healthy Bees</u>, <u>Dadant products</u>, <u>Mann Lake products</u>, <u>Harvest Lane Honey products</u> and <u>MegaBee products</u>. In addition, other products are also available in the market such as the <u>Heritage Honeybee feeding supplements</u>. However not much information is available about the nutritional quality or the efficacy of these products. The author does not endorse, support or oppose any particular product, manufacturer or product website for the sale of the above-mentioned products. The product should be chosen depending on the individual needs. A Google search was conducted for product nutrition information and the most informative websites are linked. Additional information for liquid supplemental products containing amino acids and essential oils is also available at the manufacturers' websites.

Product	Manufacturer	Crude protein composition as on label (%)	Crude lipid composition as on label (%)	Notes from the manufacturer on the label
Global 15%	Global Patties	15.5	0.9	15% bee-collected pollen by weight
Bee Pollen-Ate	Alltech Inc.	35.0	-	Yeast based with zinc proteinate and selenium yeast
Global 4%	Global Patties	15.6	0.8	4% bee collected pollen by weight
Healthy Bees	Healthy Bees	15.9	4.18	Spirulina-algae based protein
AP23® Pollen Substitute	Dadant	47.0	3.5	Does not contain pollen
AP23 [®] with Honey- B-Healthy	Dadant	15.0	7.0	Contains high fructose corn syrup and essential oils
AP23 [®] Winter Patties with Honey-B-Healthy	Dadant	2.5	2.0	Contains high fructose corn syrup
Brood Builder™ Dry	Dadant	26.35	2.28	Contains soy beans and yeast
Brood Builder™ Patties with Honey- B-Healthy	Dadant	10.0	2.5	Contains high fructose corn syrup and essential oils

Product	Manufacturer	Crude protein composition as on label (%)	Crude lipid composition as on label (%)	Notes from the manufacturer on the label.
Ultra Bee Plus Patties with Natural Pollen	Mann Lake	15	-	5% natural pollen content
Bee-Pro Patties Pollen Substitute	Mann Lake	10	-	Contains additionally Pro Health
Bee-Pro dry feed	Mann Lake	40	2	High protein pollen substitute
HiveAlive Fondant Patties	Mann Lake	<0.5	<0.5	Contains a blend of vitamins and amino acids
Ultra Bee	Mann Lake	18	-	Includes amino acids, vitamins, lipids, minerals and high fructose corn syrup
Ultra Bee High Protein Pollen Substitute Patties	Mann Lake	15	-	Includes amino acids, vitamins, lipids, minerals and high fructose corn syrup
Ultra Bee High Protein Pollen Substitute Dry	Mann Lake	58	-	Contains no pollen
Pollen Patties	Harvest Lane Honey	15.6	0.8	Contains 4% pure pollen in addition to soy flour and brewer's yeast
MegaBee powder	MegaBee	38.3	2	Contains the essential amino acids and is plant protein based

Sugar Supplementation:

There are many ways to supplement honey bee colonies with sugar and there are also various modifications to the form of sugar that is commonly fed to the colonies. Beekeepers often provide sugar supplements as sugar syrup (made from cane sugar or beet sugar), granules or fondants depending on the season and colony needs. Some beekeepers may either directly provide high fructose corn syrup (HFCS) or make invert sugar syrup (by adding cream of tartar or citric acid). HFCS (produced from corn) is often a cheaper option to use instead of pure sugar syrup. The most common HFCS used for feeding colonies is HFCS 55 (contains 55% fructose). Compared to regular sugar (50% glucose and 50% fructose), both HFCS and invert sugar syrup have higher fructose

Hydroxymethylfurfural (HMF) is an organic byproduct that forms in sugar-containing foods due to overheating or prolonged heating and process is hastened with adding acidifying substances. 30 ppm of HMF in honey can be produced if honey is stored at 30°C for 250 days, heated to 50°C for 10 days or heated to 70°C for 10 hours.

contents. A byproduct of heating HFCS or invert sugar syrup is hydroxymethylfurfural (HMF) as the fructose component of sugar is more susceptible to forming HMF. HMF can also form when honey or regular syrup is heated or stored at warmer temperatures for a prolonged period of time.

HMF above 30 parts per million (ppm) is toxic for honey bees. Honey bees break nectar sugars down naturally using invertase enzymes. When chemically inverting sugar or overheating

Nectar/honey and syrup are not the same. Nectar and honey have additional nutrients that sugar syrup lacks. How much honey to leave in an overwintering hive will depend on the region and size of the colony. sugars, HMF is a dangerous byproduct of which beekeepers must be cognizant. Care must be taken when transporting or storing sugar syrup, honey and HFCS, as HMF can easily form at high temperatures. HFCS is especially more susceptible to forming HMF due to its high fructose content.

There are many ways to supplement a colony with carbohydrates, especially liquid syrup. While honey is considered adulterated if the colony is sugar supplemented in general, sugar supplementation is necessary when the colony critically relies on this. The amount to feed depends on the colony, seasonal and beekeeper needs. The method of feeding is also dependent on these factors in addition to the beekeeping scale. Migratory beekeepers will be overburdened if carrying extra equipment to supplement sugar syrup to the colonies. Some types of sugar feeders may also encourage robbing. Thus when choosing a sugar syrup feeder, the choice relies on the type and scale of beekeeping operation and the feeding season. Some of the more popular methods of sugar supplementation are listed as follows (Figure 4):

HIVE-TOP FEEDERS: These are very practical when feeding in any season. The colony does not need to leave the hive for feeding and this is especially helpful during unfavorable weather conditions. The feeders can be replaced or refilled without encouraging robbing or opening the hive. This is not a preferred method of feeding for commercial migratory beekeepers due to the inconvenience of hauling additional equipment. This style of feeder can be used to stimulate brood production in the colonies.

IN-HIVE FRAME (OR DIVISION BOARD)

FEEDERS: These in-hive feeders are also practical for spring feeding as the bees do not need to leave the colony to access feed. However, the hive has to be opened to refill these feeders. Not a recommended method to feed in winter.

PLASTIC RESEALABLE BAGS (SLIDERS OR

ZIPPERS): Plastic bags are filled with sugar syrup and holes are cut on the top of the bag to allow the honey bees to drink. However, if holes are cut at the bottom of the bag, the syrup will drip. Even though economical, these have to be replaced/refilled often and placed over the frames inside the hive. In addition, ekes or spacer rims have to be used to make room for the bags. Due to the nature of the plastic bags lying flat, small pockets of sugar syrup may form separated by empty stretches of the bag as the plastic bag will collapse. Even though this may seem to be a good option for a few colonies in a close apiary or backyard, the beekeeper must also be careful about the bags leaking and the syrup spilling over the combs.

BOARDMAN FEEDERS: While an entrance feeder is easy to use, particularly when colony stimulation is necessary, there are various disadvantages of using these. Not only do these feeders encourage robbing (as they are at the hive entrance), they also need to be refilled/replaced almost daily as they empty out quickly.

BARREL FEEDERS: Barrel feeding is a popular method of feeding large volumes of sugar syrup to honey bee colonies to prevent starvation, especially if apiary access is difficult. While barrel feeding is a common practice in some regions across North America (for example Ontario), it is either not recommended (for example Virginia, Alberta, Manitoba) or is illegal (for example British Columbia) in other regions. Hence beekeepers must be aware of the local laws of barrel feeding and recommendations may vary depending on the region. While barrel feeding can help put weight on a colony fast, the weaker colonies in the yard are likely to benefit the least. Barrel feeding can encourage robbing as it attracts honey bees from multiple nearby yards and as a result there is also a risk of disease transmission between colonies.

GRANULATED SUGAR, FONDANTS AND

SUGAR CANDIES: Dry sugar or fondants can be used in cold winter months to sustain a colony. However, honey bees will need water to moisten the dry sugar and, thus, fondant may be a preferred method for emergency winter feeding, especially for weaker colonies unable to forage for water. Fondant is also placed inside the brood chamber and is more easily accessible by the worker honey bees. However the primary goal must always be to ensure the colony has enough food reserves to overwinter successfully, thus minimizing the need to provide dry sugar or fondants as supplements. Figure 4: Examples of sugar syrup feeding. (A) Hive-top feeders; (B) In-hive frame feeders; (C) Boardman feeders and (D) Barrel feeders. Image courtesy A and C: Dr. Dewey Caron; B: Angus Catchot III, Pollinator Health and Apiculture Lab, Mississippi State University; D: Dr. Shelley Hoover, University of Lethbridge.



Another practice lacking scientifically proven efficacy is to add a small volume of bleach to prevent mold formation in syrup or an additive (pure essential oils and commercial additives such as "Feed for Bees", "Complete", "Hive Alive", "Honey-B-Healthy" etc.). Even if the products are marketed to make the feed more attractive, this is not recommended during seasons (late summer/early fall) when robbing occurs due to scarcity of natural forage. Individual essential oils (for example thyme oil) have been tested in research laboratories for their efficacy against parasites and diseases and the results are mixed.

5. PROBIOTICS AND PREBIOTICS

Probiotics are a mixture of living microorganisms (bacteria and yeast) and prebiotics are food/ substrates that promote the growth of these microorganisms. The gut microbiome in honey bees is relatively simple with five core members.

The 5 core members of the honey bee gut are: Gilliamella apicola, Lactobacillus Firm-5, Lactobacillus Firm-4, Snodgrassella alvi and Bifidobacterium asteroides.

Research shows that when probiotic formulations in the laboratory are developed using one or more of these core members, some beneficial effects are observed. These effects include counteracting American foulbrood infection, fungal pathogens and improving brood production and colony growth. In one study, the method of delivery determined the benefits observed: spray formulation to counteract infectious pathogens and patty formulation to boost colony growth and overall health.

There are also various commercial probiotic supplementations available in the market. These include either single strains or a combination of multiple strains of various bacteria. For example, in the United States, SuperDFM and ProDFM are sold commercially. Protexin Concentrate single-strain and Protexin Concentrate multi-strain probiotics are found in the United Kingdom and the European Union. Vetafarm Probotic is found in Australia, New Zealand and the UAE. However, most bacterial species included in these commercial formulations are not native to the honey bee guts and most of the strains are either relevant for cattle or for birds.

Despite not being members of honey bees' natural microflora, in one study, Vetafarm Probotic, Protexin Concentrate single-strain and Protexin Concentrate

Probiotics for honey bees are a group of microbes which have the potential to benefit overall bee health. Prebiotics are non-digestible ingredients in the diet that can promote the growth of beneficial microbes in the honey bee guts.

multi-strain commercial probiotics were tested to check their efficacy in reducing Nosema ceranae spore load counts and these products seemed promising. There is still limited understanding and a lack of peer-reviewed scientific evidence regarding the beneficial effects of the commercial probiotics sold in the United States and other countries. Research also shows that commercially available probiotic bacteria fail to establish in bee guts whereas bacterial strains cultured from the bee guts themselves establish successfully. A more beneficial probiotic supplementation may be eventually developed using bacteria, such as Lactobacillus sp., that are more commonly found in the honey bee guts. A few studies with prebiotics and honey bees have shown that feeding yeastderived 1,3–1,6 β -glucan significantly reduced deformed wing virus infections and feeding shellfish-derived chitosan enhanced immunity and resistance to Nosema infections. More research is needed before feeding the honey bee colonies with any prebiotic formulations.

6. MANAGEMENT NEEDS

In the absence of floral resources, honey bee colonies will slow down brood production and the queen will eventually cease egg laying. In such instances, brood cannibalism can also occur. Colonies that do not have sufficient honey stores inside the hive or adequate nectar in the landscape will starve. To maximize longevity and productivity, it is important that commercial colonies have access to optimal nutrition. In temperate climates, long winters, short flowering seasons and short colony growing seasons are common. Coupled with inclement foraging weather, insufficient pollen and nectar in the landscape and high colony stocking densities in landscapes with minimal forage, this can pose a problem for maintaining healthy colonies. Supplemental feeding is thus

Water is necessary, especially during the hot and dry summer months. Water is a valuable resource for the colonies. Water is used not only for hydration but also for liquefying crystallized honey or moistening dry sugar supplements. necessary in these instances and often is regulated depending on the beekeepers' needs – early spring pollination, honey production, sale of packages, splitting colonies or producing queens.

Regionally, nutritional management of colonies varies across the United States and Canada. Winters in the North are typically much colder than in the South, and the colonies may cease brood rearing completely by October or November. However, these colonies need sufficient food reserves to last the winter months and the diutinus bees need enough nutritional resources stored to overwinter successfully. In contrast, in the southern United States (especially in the Southwest and Southeast), where it is much warmer, the colonies may continue to produce brood and fly in search of natural forage and thus need a higher quantity of winter food reserves over the winter months than the northern colonies. Weather conditions, colony status, and forage availability determine the need for supplemental feeding. In spring, the colonies may need additional feeding to boost brood production and colony growth. Pacific Northwest US commercial beekeepers reported

Diutinus honey bees are the overwintering workers that are typically produced in fall and are the longest surviving worker cohorts in the colony. They need to store enough nutritional resources within their tissues to survive the winter. They are also responsible for rearing the new generation of brood the following spring.

feeding approximately 1.5 to 8 US gallons (~5.7 L to ~30.3 L) of sugar syrup in spring and 3 to 6.5 US gallons (~11.4 L to 24.6 L) of syrup in the fall. These beekeepers also reported feeding up to 6 lb (~2.7 kg) of protein supplements in the spring months and approximately 4 lb to 8 lb (1.8 kg to 3.6 kg) in the fall. In the Southwest region of the United States, a summer dearth and availability of rainfall determines the need to provide supplemental feeding to the colonies. In southern Alberta, Canada, beekeepers feed approximately 5 US gallons (~19 L) of syrup in fall, up to 3 US gallons (11.4 L) of syrup in spring and 3-5 lbs (1.4 - 2.3 kg) of protein supplements in spring.

Table 3: Table summarizes the protein supplementation needs for honey bee colonies depending on the season and the colony needs. Note that colonies being prepped for pollination will need supplemental feeding to strengthen the colonies and increase brood production in the hives. Also, ideally we must match protein and carbohydrate supplementations based on colony status and keep feeding if we start unless there is a steady flow of nectar and pollen in the landscape. Some beekeepers will provide dry pollen feeders to the colonies in spring to gauge nutritional stress vis-à-vis protein requirements for the colony. If the hive requires proteins and there is a dearth in natural pollen, the worker honey bees will visit the protein feeders. If there is ample pollen forage available, the foragers will prefer natural forage over the dry pollen feed.

Hive condition or season	Suggested protein feeding
Starting a package or hive with no foundation	First feed sugar supplementation (see Table 4) before feeding any proteins to the colonies.
Coving build up	Feeding proteins stimulate brood production, strengthens colonies for pollination services and ensures the colony has access to proteins in case of inclement weather. A pollen supplement patty is favored over a pollen substitute patty.
Spring build-up	Colony stimulation is only desirable when natural forage is available. The colony can be stressed if artificial protein supplementation stimulates the colony, population rapidly expands and there is a scarcity of natural forage availability.
Summer colony maintenance	Not needed unless the colony is expanding rapidly and unusually, or there is a lack of pollen in the landscape or prolonged inclement weather. Consider how much patty is needed. Be careful to not stimulate small hive beetle infestations with leftover patties inside the hives.
Late summer / Early fall colony supplementation and preparation for winter	Pollen substitute patties can be fed to the colonies to bolster diutinus honey bees ready to overwinter and thus strengthen the colonies. Pollen supplement patties (though not needed) may be fed in warmer regions without a brood break. If sufficient stored bee bread is present in the colony, there is no need to provide supplemental feeding.
Winter colony management	Not recommended and not needed.

Sugars (carbohydrates) are typically supplemented as regular liquid syrup during early spring (1:1 ratio of sugar and water) and as thicker sugar syrup (2:1 ratio of sugar and water) in fall. The regular sugar syrup can be prepared using white, refined cane or beet sugar. Feeding sugar is based on individual judgment (Table 4) and care must be taken to evaluate the colony needs, the seasonal fluctuations and the colony strength before deciding to start or stop feeding carbohydrates.

The cost of supplementing protein and carbohydrates is substantial. Especially for a commercial operation, this can run into thousands of dollars to maintain the hives seasonally. The National Agricultural Statistics Service reported feed to be the largest expenditure for beekeepers with costs for total feed (including syrup, sugar water, honey, pollen patties, and other types of feeds) to be \$42,582,000 and \$44,517,000 in 2022 and 2023 respectively. For large commercial operations, beekeepers need to prepare, store and dispense sugar syrups in large volumes. Thus this type of feeding system requires a feeding tank, a pump and a hose system and the total cost may be averaging > \$600-\$1500. The cost of raw granulated sugar is also significantly higher than the cost of refined granulated sugar. Even though HFCS is a cheaper alternative to regular syrup, current research shows that colonies fed HFCS have weaker immune systems and are more susceptible to pesticide exposures and other pathogens. Benefits of pollen over non-pollen supplements are also well described. Thus over the long-term, the benefits of providing wellbalanced and optimal supplemental nutrition to the honey bee colonies outweighs the costs of such maintenance.

General recommendations for amount of honey that should be in a standard hive for overwintering outdoors are as follows. The local experienced beekeepers can provide more information for your specific region depending on colony conditions and climatic variations.

- Warmer southern regions need at least 40 pounds and may need continual feeding depending on colony activity and forage availability.
- 2. Middle states need at least 60 pounds.
- Colder northern regions need at least 80 – 90 pounds. Further up north may require additional winter stores depending on weather conditions.

Feeding considerations:

- 1. Honey bees need water (from outside or condensation from inside the hive) to moisten dry sugar.
- 2. Do not feed weak colonies dry sugar.
- Honey bees will prefer nectar, honey or liquid syrup over dry sugar unless during fall/winter if temperature is too cold (syrup can freeze) or humidity is high (provides condensation). Honey bees will not store dry sugar and will only consume this when absolutely necessary.

Key points for sugar feeding:

- 1. If syrup is not completely consumed and left unattended in the feeder, mold or toxic yeast may grow.
- 2. Keep and mix syrup in clean containers.
- 3. Store syrup at a cool place to prevent formation of HMF.
- 4. Any syrup that may have discolored or fermented should not be fed to the honey bees.
- 5. Thoroughly clean feeders when convenient.

Table 4: Table summarizes the sugar supplementation needs for honey bee colonies depending on the season and the colony needs. Note that colonies being prepped for pollination will need supplemental syrup feeding to boost the hives. Nutritional stress can be caused when a large number of colonies are held in one location without adequate natural forage. If a colony has no capped honey, the colony can be assumed to be nearing starvation and requires immediate sugar supplementation. Thus it is important to recognize nutritional stress before overfeeding the colonies. For commercial beekeepers, a good measure of colony sugar supplementation needs can be the average weight of colonies in the apiary, with lighter colonies needing more supplementation than heavier colonies.

Hive condition or season	Suggested carbohydrate feeding
Starting a package or hive with no foundation	Honey bees will need to build comb and providing carbohydrates offers the energy needed. Continue feeding 1:1 syrup until at least there are five or six (preferably all nine or ten) fully drawn frames in the lower brood chamber (bottom box). Some beekeepers also feed until they have brood boxes with fully drawn combs. However, how long to feed depends on the beekeeper's resources and needs and the nectar flow.
Spring build-up	1:1 sugar syrup supplementation strengthens a colony coming out of winter, preventing colony starvation. However, beekeepers must stop feeding before the honey flow to avoid honey adulteration. Feeding during the flow when supers are on can result in contaminated honey and rejected loads. If you start supplementing syrup, do not stop until the honey flow starts. Colonies will rapidly start to expand with supplemental syrup and can crash without a sufficient nectar flow in the landscape to support brood production. Spring feeding boosts the colony, stimulates brood production and encourages worker honey bees to forage for pollen. Spring is also a great time to feed splits. If queen rearing, feed a small quantity ($0.5 - 1$ US gallon / $1.9 - 3.8$ L) of 1:1 sugar syrup every few days to the cell builders and queen banks.
Summer colony maintenance	During nectar dearth, 1:1 syrup supplementation maintains critical colony functioning and may prevent robbing of weaker colonies. Honey supers should be removed from hives when the nectar flow ends, depending on the region. Additional sugar supplementation can occur after the honey supers are off.
Fall colony supplementation and preparation for winter	Feeding is done in fall to build colonies up for winter and prevent starvation. This is particularly important if the hive feels light and adequate stored honey is lacking. Sugar syrup fed to bees in fall should be a thicker 2:1 ratio of sugar to water. This thicker syrup is easier to ripen (concentrate) by the honey bees for storage. Ideally, 4 US gallons (~15 L) of the thicker 2:1 syrup is needed per full-sized colony, but this depends on the region. In addition, transfer honey frames for overwintering preparations.
Winter colony management	Once the first frosts have appeared, it may be difficult to get bees to store liquid syrup. If additional sugar is necessary, consider granulated sugar, fondant, etc. If warmer winter conditions return, fall formula for liquid sugar syrup may be an option again. If we must feed the colonies during winter, then dry formulations are preferred. Stronger colonies can forage for water to reliquefy dry sugars, however weaker colonies will not be strong enough to forage for water.

In addition to outdoor wintering, indoor storage facilities are used for overwintering honey bee colonies. These indoor storages maintain a consistent airflow and temperatures between $37^{\circ}F - 42^{\circ}F$ (~3-6°C). Colony preparation for overwintering in indoor storage facilities is similar to that of colonies overwintering outdoors. The colonies need to be treated for mites and provided enough supplemental feed before they are moved indoors. While overwintering indoors, a general

recommendation for sugar supplementation is that assuming on an average colonies consume 2 oz (~57 g) of feed daily, a colony must have 150 oz or 9.5 lb (~4.3 kg) of supplemental carbohydrate feed available for a storage duration of 75 days. Many beekeepers are cautious, and to avoid colony starvation (especially for larger colonies), provide ample supplemental carbohydrate feed up to even 19 lb (~8.7 kg) per hive (4 oz / ~113 g per day) while the colonies are overwintering indoors. After leaving the indoor storage, beekeepers also feed immediately if the colony needs to be readied for almond pollination.

Reiterating the importance of natural pollen and nectar over any form of supplemental feeding, it is important to note that beekeepers can also provide colonies with adequate nutrition via accessible supplemental forage. A diverse and staggered bloom is important to provide honey bee colonies with adequate and diverse macro and micronutrients. Efforts are currently underway to partner beekeepers and growers in planting supplemental forage before/after commercial pollination in the large monocropping agricultural landscapes (for example Project Apis m. Seeds for Bees). There are numerous planting guidelines available through USDA, NRCS and Xerces Society. Many research laboratories have also joined efforts in evaluating the nutritional quality of bee forage to help select a nutritionally optimal habitat. A quick check of the stored bee bread inside the colonies during pollen flow can help one ascertain if honey bees are bringing back diverse types of pollen or not (Figure 5).

For smaller beekeeping operations (hobbyists and sideliners), it is possible to place pollen traps at the hive entrancer underneath the colony during times of pollen availability to trap the corbicular pollen (pollen collected by honey bee foragers and brought back to the hive) (Figure 6). The key is to not starve any one particular colony, but rather target the strong disease-free colonies that have plenty of brood (as they will forage for more pollen). The pollen traps can be placed for a period of 24 hours – 48 hours (depending on how much pollen is being brought back to the hives) on bright sunny days with ideal foraging conditions. The collected pollen can then be frozen in freezers at home for short-term and used during times of supplemental feeding. Frames of bee bread can also be reused similarly.

Purchasing natural pollen on a large scale can be expensive in addition to risking pesticide exposure and pathogen transmission from that pollen. Figure 5: An example of diverse forage habitat as evidenced by the diverse colors of stored pollen (bee bread) in the hive. Image courtesy Dr. Jennifer Tsuruda, University of Tennessee-Knoxville.



Figure 6: Pollen traps placed at the nest entrance can be an easy method to trap incoming natural pollen brought back by the foragers. Image courtesy: Audrey Sheridan, Pollinator Health and Apiculture Lab, Mississippi State University



7. GAPS IN OUR UNDERSTANDING OF HONEY BEE NUTRITION

Further research is needed to create a nutritionally optimal diet for honey bees, one that contains all the necessary macro- and micronutrients. Honey bee nutrition is extremely complex due to the variations in colony, seasonal and individual needs. The sociobiology of the colony and age polyethism of the workers (change in worker honey bee behavior with age) mandate different nutritional supplementations. Measures of "good nutrition" in the context of a colony response to supplemental feeding usually are evident by a strong populous colony, larvae with ample brood food (wet brood), a healthy brood pattern and thriving brood population (queen is healthy and lays eggs), enough workers to continue foraging during favorable conditions and better immunity of the individuals in a colony (no pests or parasites). However, the utilization and conversion of the supplemental diet to such colony health parameters are not clearly understood. Unlike cattle, where diet supplementation and return on investment is well understood, we are still unclear about the benefits of the supplements as concrete, identifiable markers of efficacy. For example, what quantity of protein supplemental feeding will increase brood area by a desired percentage or what volume of sugar syrup will increase colony strength by a desired number is not clear.

Especially when considering the acceptance of supplemental protein diets by honey bee colonies and the efficacy of these diets, palatability, digestibility and assimilation are all key factors. Most importantly, it is crucial to remember that palatability and digestibility are not the same. Palatability of the supplemental feed indicates that honey bees will readily consume the diet. Digestibility of the diet ensures that honey bees can easily digest the supplemental feed. Diet assimilation indicates that all the required valuable nutrients from the consumed and digested diet are absorbed by the honey bees. A palatable diet may not be nutritionally optimal and may not show beneficial effects such as increased brood rearing. Lipids, fatty acids and different protein sources are known to contribute to the attractiveness of natural pollen. Thus when considering palatability of supplemental diets, the lipids (fats, oils etc.)

An ideal supplemental diet must have the following characteristics:

- 1. Palatability.
- 2. Digestibility.
- 3. Optimal balance of the required macro and micronutrients, even though a pollen substitute can never truly replace natural pollen.
- Will provide the desired "good nutrition" effects (increase in brood, colony stimulation, increasing colony strength, supporting colonies during pollen and nectar dearth in the habitat).

and the protein sources in a diet can play a key role. Particle size of the meal is often considered an important aspect of an artificial diet. A recent study showed that when offered supplemental diets containing flours of different particle size, all experimental honey bee colonies preferred flours with small grain size and high lipid-to-protein ratios. However, studies have also shown that addition of milled pollen to artificial diets does not affect consumption, rather an increase in consumption was noted with an increasing addition of pollen to the artificial diet.

Another study showed that coconut oil, almond oil and linseed oil were highly attractive to honey bees in a laboratory cage study. The same study showed that artificial diets containing soybean proteins, powdered cellulose or milled oat hulls were favored. However, addition of 5% oil to the patties increased intake of the artificial diets. When considering all the proteins, lipids and other nutrients together, the digestibility and assimilation of the supplemental diet become as important as the palatability. Another study also indicated that brewer's yeast and soybean flour are palatable as protein sources. Researchers have attempted to increase diet palatability and consumption by adding synthetic brood pheromones to the brood nest, fermenting the diet, adding bee-collected pollen to the diet, adding phagostimulants, increasing the surface area of the patties etc. Nevertheless palatability is not an indication of

the nutritional quality of the diet. Even though a supplemental diet may be readily consumed, it may not be digested or assimilated easily and it may not offer optimal nutrition to the honey bee colonies. Ideally, the supplemental diet is affordable and also easy to feed to the honey bees. An abridged review of the colony impacts of various commonly available supplemental diets (commercial and homemade) is provided in Appendix A (adapted from Noordyke and Ellis, 2021).

Many questions remain unanswered and future research on supplemental feeding should address these:

- How long does it take for the conversion of pollen or the supplemented protein diet to royal jelly or brood food? As a recent study has shown that adult honey bee workers likely do not feed the supplemental protein patties directly to the larvae and they also do not store these as with beebread, it becomes critical to understand the conversion of the supplemental proteins to brood food.
- 2. Reiterating that studies have shown workers do not store supplemental protein diets as they do for pollen, do these artificial diets only provide short-term benefits to the adults in the colony?
- 3. Do these supplemental diets provide any transgenerational benefits?
- **4.** Even if the supplement increases brood area, is it economical, especially for a large-scale commercial operation?
- 5. When buying irradiated pollen commercially, does it have any effect on the nutritional quality of the pollen purchased? One study reported that when three different methods (gamma irradiation, ozone fumigation and ethylene oxide fumigation) of pollen sterilization were compared, ethylene oxide fumigation retained pollen quality as a food source for bumble bees while being able to suppress pathogens. We have limited understanding of the nutritional quality of pollen that has been gamma irradiated for honey bees.
- As bee bread is essentially stored pollen, can selling bee bread commercially be another avenue to explore? Importantly, the same concerns are valid about bee bread (pathogen

transmission and pesticide toxicity) as when purchasing commercial pollen.

The most economical solution is not always beneficial. With changing seasonal physiology, colony needs and individual demands, supplemental feeding should be holistic and adaptive. While we need to address these gaps in our understanding of supplemental feeding of the honey bee colonies, there are also opportunities for improvement when formulating a supplemental diet by considering all aspects of bee nutritional needs overall.

8. INTERACTION BETWEEN POOR NUTRITION AND OTHER STRESSORS

Optimal nutrition is the honey bee colony's first line of defense. Documented research has shown colonies that do not receive sufficient nutrition overwinter less successfully, are more susceptible to Nosema ceranae infection, have higher pathogen loads, produce less brood and have poor immunity. Emerging research also shows that pesticides and poor nutrition have synergistic impacts on honey bee health such as a reduction in longevity, loss of navigational abilities, impaired learning and olfactory memory and poor nutritional quality of the royal jelly produced by nurse honey bees in colonies exposed to pesticides. Thus it is crucial that honey bee colonies have access to optimal nutrition and beekeepers provide supplemental feeding when needed.

9. APPENDIX A: TABLE SUMMARIES

In this section, two review tables summarize the effects of commercial diets and some common homemade protein supplements on honey bees (individuals and overall colony). Several parameters have been measured such as adult honey bee population size, queen health, brood production etc. A detailed version of these reviews is available in supplementary tables 1 – 3 found in Noordyke and Ellis (2021) (Appendix C).

Table 5: A brief summary of the impacts of commercial diets on honey bees (adapted from Noordyke and Ellis, 2021).

Commercial diet name	Health impacts as shown in different peer- reviewed research articles when compared with sugar syrup or pollen limitation (# indicates number of relevant results)	Major phenotype response of the colonies
Bee-Pro® (Mann Lake)	1 positive, 6 negative and 11 neutral	No change in adult bee population, honey production or brood production
Brood Builder (Dadant)	3 neutral	No change in adult bee population and brood production
Feedbee®	8 positive and 1 neutral	Increase in adult bee population and honey production. No change in queen quality. Mixed results on brood production.
Global Patties	2 negative, 1 positive and 2 neutral	No change in brood production
MegaBee®	3 positive, 3 negative and 6 neutral	No change in adult bee population, brood production or queen quality
MegaBee Winter Patties (Dadant)	1 neutral	No change in <i>Nosema</i> load
UltraBee (Mann Lake)	1 positive, 1 negative and 7 neutral	No change in adult bee population, brood production and <i>Nosema</i> load

Table 6: A brief summary of the impacts of commonly formulated homemade patties on honey bees
(adapted from Noordyke and Ellis, 2021).

Homemade pollen substitute diet name/ ingredients	Health impacts as shown in different peer- reviewed research articles when compared with sugar syrup or pollen limitation (# indicates number of relevant results)	Major phenotype response of the colonies
Black gram, sucrose, skim milk powder, yeast, water	2 positive	Increase in adult bee population and honey production
Soybean meal, sucrose, skim milk powder, yeast, water	2 positive	Increase in adult bee population and honey production
Roasted soy flour, pollen, sucrose, honey (small (100 x 100 x 23 mm) surface area)	3 neutral	No change in brood production, honey production or pollen storage
Defatted soy flour, parched gram, brewer's yeast, sugar, glucose	2 positive	Increase in adult bee population and brood production
Soy milk powder, albumin, and sucrose solution	4 positive	Increase in brood production and honey production
Mungbean flour, honey, yeast extract	4 positive	Increase in adult bee population, brood production, honey production and pollen storage
Chickpea flour, honey, yeast extract	4 positive	Increase in adult bee population, brood production, honey production and pollen storage

10. APPENDIX B: BEEKEEPER INTERVIEWS

This section briefly summarizes common supplemental feeding practices of six commercial beekeepers across the United States. The beekeepers vary in their management and style of beekeeping operations. A series of interviews were conducted to understand what works for them depending on their location and their beekeeping practices for providing supplemental feeding to the honey bee colonies throughout the year.

BEEKEEPER 1

Beekeeper 1 is a migratory beekeeper based in Oregon and commercially pollinates multiple crops across the Pacific Northwest, including California almonds. Feeding during the first week of January (when the weather cooperates) is very helpful for preparing honey bee colonies for almonds. To take advantage of early brood, feeding is important as this will ensure presence of brood when the beekeeper moves colonies to the almond orchards. Prior experience with feeding colonies during Thanksgiving week was an inconvenience without any added benefit after which the beekeeper follows this feeding routine as mentioned.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	Soon after January 1, the beekeeper supplements the colonies with two cups of Baker's Drivert (CNH) placed over the brood cluster (to break the cluster) and 1 lb (~ 0.5 kg) of protein supplements (Global Patties containing 4% natural pollen). Before transporting the colonies to California, the colonies are fed 1:1 sugar syrup and 1 lb (~0.5 kg) of Global Patties containing 4% natural pollen. Depending on weather and colony health, another pound of the protein supplement may be fed in California. Before leaving California for Oregon, 1 gallon (~3.8 L) per week of 1:1 sugar syrup is supplemented mostly using in-hive feeders. The beekeeper also builds nucleus hives for sale after the almonds, so feeding the colonies to stimulate and restore strength is crucial. Back in Oregon, the colonies are moved for commercial pollination of the tree fruits (mostly pears) and depending on the colony status and environmental conditions (unfavorable weather and insufficient nectar flow), the beekeeper continues feeding unless there is a long stretch of nice weather and the maples start to bloom. May can be a harsh month in Oregon for honey bees with resource scarcity in the landscape after spring bloom. The beekeeper thus feeds the colonies as needed depending on weather conditions and colony status. Honey bee colonies in holding yards have dry pollen feeders outside so that honey bees can forage if they want.
Summer	Supplemental feeding for proteins is not necessary and can be dependent on colony status. Carbohydrate supplementation is not needed due to the honey flow which lasts for about two months (June-July).
Fall	About mid-August, the beekeeper removes all the honey supers and treats the colonies for mites. The beekeeper starts feeding the colonies to build the winter clusters and supplements about 4 feedings of thicker sugar syrup (between 1:1 and 2:1 as convenient) and a minimum of 3 lbs (~1.4 kg) of protein supplements (Global Patties with 4% pollen) per two-box colony. The beekeeper aims to supplement both sugar syrup and protein patties at the same time and tries to avoid any break in protein supplementation by replacing the protein patties just before the colonies fully consume the previous patty. The beekeeper aims to finish all feeding around the end of October.
Overwintering	The beekeeper overwinters the colonies outdoors. The beekeeper removes any unconsumed pollen substitutes on top of the hives (for avoiding mold). Last inspection and spot feeding as needed are done by the end of October/early November and the beekeeper will check the colonies next in January.

Beekeeper 2 is a migratory beekeeper based in Wyoming and commercially pollinates multiple crops across Wyoming, North Dakota and Nevada. In addition, the beekeeper also sells queens, packages and some honey. The beekeeper's main advice is to keep the mite numbers low and to supplement colonies with protein patties as early in the season as possible.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	The beekeeper starts feeding protein supplements (NutraBee) right before moving to the California almonds. In spring, the beekeeper feeds more sugar syrup than protein supplements. Usually 4-6 feedings of sucrose syrup (2:1 ratio; 6-8 gallons which is approximately 23-30 L) from January - May is fed to each colony. This can vary depending on the year.
Summer	No feeding is necessary for both proteins and carbohydrates.
Fall	Fall feeding vastly varies depending on how the summer season was and where the colonies were located. The honey bee colonies that spent summer in North Dakota are supplemented with NutraBee. The honey bee colonies that stay in Wyoming and Nevada in the summer months have enough beebread so no pollen supplement is usually needed. If the beekeeper needs to stimulate any colonies, then additional NutraBee feeding is required. For carbohydrate supplementation in Fall, the beekeeper prefers a syrup blend from Sweetener Products Inc. Fall carbohydrate feeding includes 4-5 gallons (~15-19 L) in about six weeks if the colonies are light in weight. If the colonies feel heavy, they are fed 1-2 gallons (~3.8-7.6 L) every six weeks. A lot of the feeding is also dependent on the queen egg laying.
Overwintering	The beekeeper overwinters colonies both indoors and outdoors and manages the colonies similarly. Only the feeding timeline changes slightly depending on whether the colonies are overwintering indoors or outdoors. If the colonies are overwintering outdoors, the beekeeper prefers to have all colonies ready by the middle of November with at least 4 lbs (~1.8 kg) of pollen per two-box colony and feeds them 2:1 sugar syrup to prepare them for overwintering. If the colonies are overwintering indoors, the supplemental feeding routine is just the same as the colonies that are overwintering outdoors. Only the timeline changes and these colonies overwintering indoors must be ready by the end of October.

Beekeeper 3 is a commercial beekeeper based in California and primarily provides pollination service. The beekeeper strongly feels that poor nutrition is the main stressor for honey bee colonies and that all beekeepers need to supplement as much and as often as is convenient. The beekeeper also suggested that honey bees can overcome other stressors if they are healthy, well fed and nutritionally supported.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	The usual protein supplementation is 2 lbs (~1 kg) of Ultra Bee every two weeks per colony. Right before the almond pollination, the beekeeper doubles the protein supplementation per colony. Natural forage in the landscape does not necessitate feeding after the almonds. In April, the beekeeper makes packages and feeds all parent colonies 2 lbs (~1 kg) of Ultra Bee to give them an advantage to grow and additional natural pollen in the landscape is also beneficial. The beekeeper follows the same schedule for feeding sugar supplements as for proteins. After almond pollination, if the colony weighs less than ideal, the beekeeper continues to feed them corn syrup (HFCS 55) every two weeks until nectar flow starts in the landscape and the colonies start to put on weight. After making packages, the beekeeper keeps feeding both parent colonies and packages. The packages are fed more in spring than any other time in the year.
Summer	The beekeeper feeds the colonies Ultra Bee before the colonies are moved for pollination. Depending on the crop the colonies are pollinating they may be well sustained naturally. The beekeeper does not feed any sugar syrup.
Fall	Starting in September, the beekeeper feeds the colonies 2 lbs (~ 1 kg) of Ultra Bee and corn syrup every two weeks.
Overwintering	The beekeeper overwinters the honey bee colonies outdoors and does not stop feeding the colonies in the winter months. Feeding continues all the way until the colonies are ready to be moved into almonds.

Beekeeper 4 is a commercial beekeeper based in Mississippi with colonies located primarily in South Mississippi and in one county in Central-East Mississippi (Noxubee county). The primary beekeeping operations include honey production, sale of nucleus colonies and packages and some occasional commercial pollination service. The beekeeper strongly feels that during times of pollen scarcity, supplementing the colonies with proteins is important. If budget permits, the beekeeper also suggested that feeding colonies sugar syrup is beneficial instead of high fructose corn syrup.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	The beekeeper supplements the colonies with NutraBee and Global Patties. The colonies can receive up to 2 lb (~1 kg) of protein supplements in spring and feeding is stopped in February when pollen is naturally abundant in the landscape. The beekeeper usually feeds the colonies 1:1 sugar syrup or mixes equal parts of 1:1 sugar syrup with high fructose corn syrup-55 (HFCS-55). The colonies receive 1 gallon (~3.8 L) of syrup every two weeks until honey flow starts and feeding is also dependent on the status of the colonies. Feeding typically starts in January and stops in February. Colonies are fed additional syrup if they feel light. The beekeeper also feeds the colonies syrup before splitting them and feeds additional syrup to all splits.
Summer	The beekeeper does not feed any protein supplements to the colonies in summer as they usually have stored beebread. For the last four years, the beekeeper has provided Ultra Bee dry pollen substitute only to the colonies located in Noxubee county. Year 2023 was unusual and all colonies in South Mississippi received additional NutraBee patties in summer due to drought and lack of forage. The honey bee colonies in Noxubee county do not need additional sugar supplementation in summer due to adequate nectar flow in the landscape. The beekeeper usually feeds 2.5 gallons (~9.5 L) of 1:1 sugar syrup per colony located in South Mississippi over the entire summer.
Fall	All colonies are supplemented with 1 lb (~ 0.5 kg) of Global Patties in fall. Sugar syrup supplementation varies depending on the colony location. Colonies located in Noxubee county receive 2 gallons of 1:1 sugar syrup per colony twice in fall. In south Mississippi, colonies receive 1 gallon (~3.8 L) of 1:1 sugar syrup at the beginning of fall and after one week receive an additional 1 gallon (~3.8 L) of HFCS-55 per colony.
Overwintering	The beekeeper overwinters the colonies outdoors. There is only a short brood break cycle. The colonies are not fed any protein supplements in winter and are spot fed sugar syrup if the colonies feel light.

Beekeeper 5 is a commercial migratory beekeeper with colonies in North Dakota, Washington and California. The beekeeper primarily is a honey producer and also commercially pollinates crops. The beekeeper strongly suggests providing adequate protein supplementation to colonies, especially for overwintering, and to keep supplementing colonies as needed. While the beekeeper has not found any long-term positive benefits from using probiotic supplements, the beekeeper is excited to try out newer supplemental feeds containing thymol and other essential oils.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	All colonies are in California at the beginning of the year. All colonies are fed once both sugar and protein supplements which includes 1 lb (~0.5 kg) of Global Patties and 1 gallon (~3.8 L) of Mann Lake syrup blend. Once natural forage is plenty in the landscape the colonies are well sustained without additional supplementation. When moving the colonies, the honey bees in California receive one 1 lb (~0.5 kg) Global Patties in California, the nucleus colonies receive one 1-2 lb (~0.5 - 1.0 kg) Global Patties and the colonies in Washington receive one 1 lb (~0.5 kg) Global Patties supplemental protein feed. Usually the colonies in Washington also receive 3-4 gallons (11.3 - 15.2 L) of syrup while the colonies in California receive 1-2 gallons (~3.8 - 7.6 L) of syrup blend per hive. The colonies are also spot fed syrup blend as needed.
Summer	All colonies are moved to North Dakota around May/June. Colonies receive one feeding of syrup before the honey flow. No additional pollen supplementation is given to the colonies. The colonies do not need feeding all summer.
Fall	All colonies are moved back to California for fall and overwintering. The colonies receive 1 lb (~0.5 kg) each of Global Patties three to four times from September to the end of the year. They also receive one high fructose corn syrup-55 feeding of about 1 gallon (~3.78 L) per colony between September to the end of the year.
Overwintering	The colonies overwinter outdoors and feeding is as indicated in Fall (September through the end of the year).

Beekeeper 6 is a commercial beekeeper in Northern California and the beekeeping operation focuses largely on commercial pollination (mainly almonds) and sale of queens and package bees. The beekeeper also sometimes makes a modest amount of honey for sale which varies across years. The beekeeper values natural pollen above any commercial protein supplement and suggests that pollen supplements may help stimulate colonies but cannot be a replacement for natural pollen. The beekeeper also actively plants supplemental forage, for example participating in the Project Apis m. "Seeds for Bees" program and working with growers to ensure there is sufficient supplemental forage for the colonies when pollinating almonds. The beekeeper strongly feels that instead of relying on the unpredictability of natural forage in the landscape due to varying climatic patterns or waiting for growers to plant additional forage around their crops, beekeepers can take a more active role and lead these efforts to help sustain their colonies.

Season	Usual protein and carbohydrate supplementations
Beginning of the year - Spring	Spring starts early in the year for the beekeeper and the colonies are fed beginning of January onwards. Depending on the colony status, they can receive 1-2 lb (~0.5-1 kg) of NutraBee per colony every ten days. Feeding will continue about 3-4 times until the almonds bloom. The beekeeper feeds 1 gallon (~3.8 L) syrup per colony at the same time when the protein supplements are provided. The beekeeper uses a premixed commercial sucrose syrup blend containing 66% solids and uses a hive-top feeder. This is done to stimulate the queen to lay eggs. The beekeeper continues to feed syrup to the colonies as needed. Spring generally has a lack of nectar plants in the landscape where the beekeeper is located so the colonies consume all the nectar that they forage for. Thus to stimulate colony growth for package production, the beekeeper continues to feed syrup as often as needed.
Summer	All colonies are moved to the mountains for summer. With abundance of pollen in the landscape, the colonies are not supplemented with any proteins. Depending on nectar flow and colony status, spot feeding 1 gallon (~3.8 L) syrup per colony is continued in summer.
Fall	All colonies are moved out of the mountains in preparation for overwintering. In fall, 1-2 lb (~0.5-1 kg) NutraBee per colony is fed to stimulate one or two rounds of brood production before the colony shuts down. Colonies can be supplemented with proteins three-four times depending on the weather and colony status. All protein supplementation is stopped before the end of November. The beekeeper switches to a commercial syrup blend containing 77% solids (50:50 blend of high fructose corn syrup-55 and sucrose syrup from Sweetener Products). This is done to allow the colonies to put on weight and not for stimulation. The colonies are fed 1 gallon (~3.8 L) syrup blend as often as needed.
Overwintering	Beekeeper overwinters colonies both outdoors and indoors. When overwintering outdoors: The colonies are supplemented with proteins as described before in fall. The colonies are spot fed sugar syrup blend (as in fall) as needed throughout the rest of the year depending on the colony status (if the colonies feel light). When overwintering indoors: The beekeeper chooses the best colonies when moving them from the mountains for overwintering indoors. These colonies are the heaviest and the strongest. Before moving the colonies to the indoor storage units, protein and sugar supplementation routines are the same as described before in fall. The colonies are not fed any supplements (proteins or carbohydrates) during the entire duration of time that they are held at the indoor cold storage unit.

11. APPENDIX C: FURTHER READING REFERENCES:

ESSENTIAL OIL AND ADDING SUPPLEMENTS TO PATTIES

Albo GN, Henning C, Ringuelet J, Reynaldi FJ, De Giusti MR et al. (2003) Evaluation of some essential oils for the control and prevention of American Foulbrood disease in honey bees. Apidologie 34(5): 417-427.

Bravo J, Carbonell V, Sepúlveda B., Delporte C, Valdovinos CE et al. (2017) Antifungal activity of the essential oil obtained from Cryptocarya alba against infection in honey bees by *Nosema ceranae*. Journal of Invertebrate Pathology 149: 141-147.

Sammataro D, Finley J, LeBlanc B, Wardell G, Ahumada-Segura F et al. (2009) Feeding essential oils and 2-heptanone in sugar syrup and liquid protein diets to honey bees (*Apis mellifera* L.) as potential Varroa mite (*Varroa destructor*) controls. Journal of Apicultural Research 48(4): 256-262.

PROBIOTICS AND PREBIOTICS

Bonilla-Rosso G, Engel P. (2018) Functional roles and metabolic niches in the honey bee gut microbiota. Current Opinion in Microbiology 43: 69-76.

Borges D, Guzman-Novoa E, Goodwin PH. (2021) Effects of Prebiotics and Probiotics on Honey Bees (*Apis mellifera*) Infected with the Microsporidian Parasite *Nosema ceranae*. Microorganisms 9(3): 481.

Daisley BA, Pitek AP, Chmiel JA, Al KF, Chernyshova AM et al. (2020) Novel probiotic approach to counter Paenibacillus larvae infection in honey bees. The ISME Journal 14: 476–491.

Daisley BA, Pitek AP, Torres C, Lowery R, Adair BA et al. (2023) Delivery mechanism can enhance probiotic activity against honey bee pathogens. The ISME Journal. <u>https://doi.org/10.1038/s41396-023-01422-z</u>

Mazzei M, Fronte B, Sagona S, Carrozza ML, Forzan M et al. (2016) Effect of 1,3-1,6 β-Glucan on Natural and Experimental Deformed Wing Virus Infection in Newly Emerged Honeybees (*Apis mellifera ligustica*). PLoS ONE 11:e0166297.

Motta EVS, Powell JE, Leonard SP, Moran NA (2022) Prospects for probiotics in social bees. Philosophical Transactions of the Royal Society B. 377: 20210156.

Vetvicka V, Fernandez-Botran (2018) R β -Glucan and parasites. Helminthologia 55: 177–184.

GAMMA IRRADIATION OF POLLEN

Hidalgo EA, Hernandez-Flores JL, Moreno VDA, López MR, Gómez SR et al. (2020) Gamma irradiation effects on the microbial content in commercial bee pollen used for bumble bee mass rearing. Radiation Physics and Chemistry 168: 108511.

Strange JP, Tripodi AJ, Huntzinger C, Knoblett J, Klinger E et al. (2023) Comparative analysis of 3 pollen sterilization methods for feeding bumble bees. Journal of Economic Entomology 116(3): 662–673.

GENERAL BEE NUTRITION, COLONY MAINTENANCE, BEEKEEPING COSTS AND SUPPLEMENTAL FEEDING

<u>Best Management Practices for Hive Health</u> – A Guide for Beekeepers by Honey Bee Health Coalition

Bonoan RE, O'Connor LD, Starks PT (2018) Seasonality of honey bee (*Apis mellifera*) micronutrient supplementation and environmental limitation. Journal of Insect Physiology 107: 23-28.1

Chakrabarti P, Morré JT, Lucas HM, Maier CS, Sagili RR (2019) The omics approach to bee nutritional landscape. Metabolomics 15(10): 127.

Corby-Harris V, Bennett MM, Deeter ME, Snyder L, Meador C et al. (2021) Fatty acid homeostasis in honey bees (*Apis mellifera*) fed commercial diet supplements. Apidologie 52: 1195-1209.

De Groot AP (1952). Amino Acid Requirements for Growth of the Honeybee (*Apis mellifera* L.). Cellular and Molecular Life Sciences 8(5): 192-194.

Lau PW, Nieh JC (2016) Salt preferences of honey bee water foragers. Journal of Experimental Biology 219(6): 790-796.

Melathopoulos A (2005) <u>Milling pollen does not make patty supplements less attractive to honey bees:</u> <u>a challenge to the chocolate chip theory</u>.

<u>National Agricultural Statistics Service (NASS)</u>, Agricultural Statistics Board, United States Department of Agriculture (USDA) website

Paoli PP, Donley D, Stabler D, Saseendranath A, Nicolson SW et al. (2014) Nutritional balance of essential amino acids and carbohydrates of the adult worker honeybee depends on age. Amino Acids 46: 1449-1458.

Schmidt JO, Hanna A (2006) Chemical Nature of Phagostimulants in Pollen Attractive to Honeybees. Journal of Insect Behavior 19: 521–532.

Topitzhofer E, Breece C, Wyns D, Sagili RR (2020) Honey Bee Colony Maintenance Expenses: Supplemental Feed, Requeening and Medication. Pacific Northwest Extension Publishing PNW 745.

Tsuruda JM, Chakrabarti P, Sagili RR (2021) Honey Bee Nutrition. Veterinary Clinics of North America: Food Animal Practice 37: 505–519.

COMPARING POLLEN SUBSTITUTES

Hoover SE, Ovinge LP, Kearns JD (2022) Consumption of Supplemental Spring Protein Feeds by Western Honey Bee (Hymenoptera: Apidae) Colonies: Effects on Colony Growth and Pollination Potential. Journal of Economic Entomology 115(2): 417-429.

Manaswi A, Noordyke E, Prouty C, Ellis JD (2023) Western honey bee (*Apis mellifera* L.) attraction to commercial pollen substitutes and wildflower pollen in vitro. Journal of Applied Entomology 00: 1-4.

Noordyke ER, Ellis JD (2021) Reviewing the Efficacy of Pollen Substitutes as a Management Tool for Improving the Health and Productivity of Western Honey Bee (*Apis mellifera*) Colonies. Frontiers in Sustainable Food Systems 5: 772897.

Noordyke ER, van Santen E, Ellis JD (2021) Tracing the Fate of Pollen Substitute Patties in Western Honey Bee (Hymenoptera: Apidae) Colonies. Journal of Economic Entomology 114(4): 1421-1430.

Saffari A, Kevan PG, Atkinson JL (2010) Palatability and consumption of patty-formulated pollen and pollen substitutes and their effects on honeybee colony performance. Journal of Apicultural Science 54(2): 63-71.

Standifer LN, Moeller FE, Kauffeld NM, Herbert EW Jr., Shimanuki H (1977) Supplemental Feeding of Honey Bee Colonies. United States Department of Agriculture Information Bulletin No. 413.