Healing Properties of Honey

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Introduction:

Honey has been used effectively in the treatment of burns as well as acute, chronic and persistently infected wounds. Peroxide levels in honey, generated by the oxidation of glucose by glucose oxidase, an enzyme derived from nectar is our indicator of healing properties.

Nectar, pollen and bee chemicals all combine to produce honey. Its characteristic flavor, color and aromaticity is linked to nectar source. The phytochemicals, in conjunction with bee chemicals, contribute to the its potential healing properties. As a refresher, let us remind you of the key components of honey that affect healing.

Honey is essentially a sugar matrix combining numerous healing components. The sugars themselves aid in controlling the wound environment to guard against infection. Honey's acidity has a bactericidal effect which is coupled with the Reactive Oxygen Species, such as hydrogen peroxide, that also arrest the growth of pathogens. Anti-oxidants have become widely accepted as suppressing age-related diseases and they are present in honey as flavonoids and phenolic acids. Finally, Bee defensin-1, an antibiotic peptide synthesized in the hypopharyngeal glands of cleaner and forager worker bees, is effective in treating biofilm infections.

Floral sources vary and whether that source is mono-floral or multi-floral affects the healing characteristics as does geography and climate. This article examines the honeys collected from different geographical regions across multiple years and in some cases, from colonies in the same year. To maintain the healing properties, honey samples were never heated above hive temperature. The GPS coordinates of the hives are known, the date of collection and their experimentally determined peroxide values are known. In this article some of the samples from 2018, 2019 and 2020 will be discussed. Selected samples from 2018 and 2019 had pollen analysis performed by mellisopalynology.

Pollen Content by Geographic Region - 2018:

Ten hives in various locations across Ontario had their peroxide values determined and pollen analysis performed. **Figure 1** shows the location of the 10 hives.



Figure 1 - Peroxide Value by Hive Code, Latitude and Longitude (2017-2018)

The center of the circle marks the GPS coordinates of the hive and the color of the circle identifies the hive (see Legend). The size of the circle indicates the absolute or averaged peroxide value as some hives (e.g. BCR) were sampled through multiple seasons. The largest circle (GARLOW) with a value of 15 mg/L, is the highest peroxide value obtained from 400+ samples tested. A value of 5 or more is significant but typical values are between 0 and 2. **Table 1** shows the Hive code, the peroxide values, date sampled and the major suspended pollen species (top 5-6 by %). Note the peroxide values of WR and 2 BCR hives with values of 7.5, all were sampled in August.

			Clovers							
Hive Code	PEROXIDE	Season	(4)	Soybean	Trefoil	Crucifer	Buckthorn	Boneset	Willow	Other
										58.5 Lily Family;
вн	5	SUM0	10.1	13.9	1.6	0.3	4.4			6.0 Virginia Creeper
GARLOW	15	FALL	13.7	12.2	13.2		1.3	50.2		
WR	7.5	SUM2	33.0	37.5	2.3	13.0	1.0	6.6		
BCR	5	SPR	2.4			2.7	76.9	0.3	14.1	
BCR	2	SUM0	23.5	31.5	18.6	4.6	12.0		4.9	
BCR	7.5	SUM1	11.9	49.8	7.5	0.3	0.3	12.3		6.8 Purple Loosestrife
BCR	7.5	SUM2	6.8	78.8	1.6	2.8	1.9	4.4		
Manitoulin	5	SUM2	0.9		70.3			19.2		
										38.0 Maple;
										6.4 Strawberry;
Sloss	5	SUM1	23.4		1.8	5.3		4.1	5.8	6.4 Holly
Ferguson	0	FALL	10.1	4.1	13.3	14.3	12.2	30.6		11.2 Buckwheat
										10.7 Apple;
Overton	0	SPR	6.9	2.1	6.0	23.3	7.5	5.4	22.1	5.4 Sumac
										30.0 Cow Parsley;
Walker-b	0	SPR	2.1		0.8	5.1	18.6		21.1	9.3 Apple
Walker-a	5	SUM2	5.4	9.2	39.9				0.9	42.6 Basswood

Table 1 – Seasons SUM0, SUM1 and SUM2 refer to Early, Mid and Late Summer

Why are the peroxide values so high? Was this a result of geography, climate, time of year or floral source? Suspended pollen analysis performed on these honey samples revealed a diverse pollen profile. The range was from 9-20 distinct pollen types, some at less than 1%. **Figure 2** shows the main pollen components of honey samples with non-zero peroxide values. The hives are described by colored rectangles with the size of the inner rectangular structure being correlated to the pollen percentage. This gives a graphical indication of the data in Table 1, to correlate pollen with higher peroxide levels.



Figure 2 – Hive Codes are listed, as are top pollen types.

What does **Figure 2** tell us about peroxide strength and pollen source? Soybean, Trefoil, Buckthorn, Clover and Boneset all appear consistently among the high peroxide level hives but individual cases indicate Maple, Basswood and Lily may contribute as well. What is clear is that the higher peroxide values did coincide with honey sampling later in the season, August on into fall. We will look more closely at that trend across three years.

Pollen analysis needs to be coupled to hive site data. These hives are geographically well separated. Manitoulin, collected from the island of the same name consists mainly of Trefoil and Boneset. Trefoil refers to Bird's Foot Trefoil and Boneset is a mixture of boneset, golden rod, and aster (the pollen grains are similar and the plants bloom at the same time). This mixture will be referred to as Boneset. Sloss is the northern most location of the hives examined, its major pollens are Mountain Maple and White Clover. Walker-a and Walker-b are from the same bee keeper. Over 80% of Walker-a is made up of Basswood and Bird's Foot Trefoil. Certification of Basswood requires only 15 % basswood pollen and this sample has 42.6 %. Walker-b, Overton and Ferguson are not represented in **Figure 2 as** their peroxide values

became zero over time. Although Walker-b was harvested during blueberry season, minimal blueberry pollen (4.2%) is present. The most abundant pollens in Walker-b are Cow Parsley, Willow and Buckthorn. The Overton honey sample had the largest diversity of pollen types (20) with Crucifer, Willow and Apple making up ~50%. Crucifer, a combined category of cabbage, canola and the mustard family, cannot be differentiated by pollen morphology. Ferguson honey, with the distinctive color and taste of buckwheat contains little buckwheat pollen (11.2%).

The remaining hives; GARLOW, BH, WR and BCR are near each other and were examined more closely for 2019 and 2020. GARLOW is surrounded by mostly forest and scrub and is a bit swampy. It gave a very large peroxide value and is >50 % Boneset, with Trefoil at 13.2 % and Soybean at 12.2 %. BH is located on a Lavender farm, consisting of several varieties over 4 acres. Most of the pollen grains (58.5 %) were from the Lily family but it was difficult to identify unequivocally, the onion family and asparagus are also likely. Lavender is a bee favorite, but for nectar not pollen. It is worth emphasizing that pollen analysis, while valuable does not reveal the available nectar sources. WR consisted mainly of Soybean (37.5 %) along with a significant amount of Red Clover (23.7 %) and Crucifer (13 %). Based on pollen analysis, four types of clover have been identified. White clover (sometimes called Dutch clover) is one of the most preferred flowers for honey bees. It's a perennial that provides a great nectar source and consequently, much honey. Sweet Clover is a biennial and has a long honey flow. Long thought a weed it has proven itself a great soil builder. Alsike Clover has an extended period of bloom and yields a mild, white honey. The name Alsike was coined by Linnaeus (a Swedish botanist from the 18th century) after a town in Sweden, he also was first to use the term nectar. Red clover, is a short-lived perennial favored by bumblebees. The shorter tongue of the honey bee cannot always reach the nectar and the theologically explanation was encapsulated by the German folk expression; The honey bee was forbidden the red clover because she didn't keep Sunday. However, we have managed to consistently find evidence of pollen from all four clovers.

BCR is located in a region of commercial agriculture, devoted mainly to Alfalfa and pasture land. There is forest, scrub and fallow land nearby. **Table 1** revealed that Hive BCR was

sampled 4 times in 2018 in Spring through Summer. **Figure 2** contained overall pollen counts but four distinct sampling events allow us to slice the information even finer. Mellisopalynology was performed and selected pollen species are shown in **Figure 3**.



Figure 3 - Selected Pollens by Percent through 2018 (Hive BCR)

Table 1 shows the most abundant pollen data, but Figure 3 reflects seasonal trends for BCR. Bird's Foot Trefoil follows a decreasing trend from SUM0 to SUM2, whereas Soybean increases through the summer. Boneset is identified in August and both Willow and Buckthorn follow a decreasing trend following a spring peak. Alfalfa has a small presence in summer (3.1 %). The Clovers are present throughout the summer months with both Sweet and White Clover peaking in early summer and tapering off.

Pollen Content by Geographic Region - 2019:

By focusing on BCR and geographically similar hives other trends are revealed. Pollen analysis was performed on honey samples from BH, SWIFT, NORTON and BCR (2019 season). **Table 2** shows honey peroxide values, when sampling occurred, and the most abundant pollens. All had significant peroxide values, particularly NORTON. Ideally, a correlation is established between peroxide value and pollen species.

Hive			Clovers					
Code	PEROXIDE	Season	(4)	Soybean	Trefoil	Basswood	Boneset	Other
ВН	5	SUM0	10.6	3.3	1.5	15.6		62.9 Lily Family
ВН	5	SUM1	8.9	4.0	0.9	64.4	0.4	19.1 Lily Family
BH	2	SUM3	19.1	4.7	0.3	2.2	4.1	61.1 Lily Family
SWIFT	5	SUM3	32.2	37.8	6.9	4.3	1.3	7.2 Sunflower type
NORTON	5	SUM2	25.7	49.0	12.2	4.5		
NORTON	7.5	SUM3	17.3	37.0	22.2	4.2	5.1	7.1 Sunflower type
NORTON	7.5	FALL	33.3	9.9	17.1	0.8	29.4	5.2 Sunflower type
BCR	2	SUM1	19.9	28.2	4.5	36.5		
BCR	5	SUM2	22.8	43.6	9.3	6.4		8.6 Buckthorn
BCR	2	SUM3	28.0	26.3	3.9	12.7	3.1	7.5 Knapweed
BCR	5	FALL	16.7	11.7	3.4	3.0	52.7	

Table 2 - SUM0 to SUM2 are early, mid, late summer, SUM 3 is early September

As with 2018, pollen analysis revealed significant diversity of pollen types (range 12-22) many in amounts of less than 1 %. **Figure 4** presents the pollens which are significant (> 5% abundant) and common across hives. Buckthorn and Knapweed were left off the plot as they had a single occurrence.

The same pollen sources as 2018 (Soybean, Trefoil, Clover and Boneset) are seen in **Figure 4**. There is a consistent correlation between these pollen types and high peroxide values (Buckthorn was mostly absent in 2019). Hive specific results, such as BH, the lavender farm, showed Lily and Basswood as the majority types. The pollen values are absolute value for SWIFT and average values for the remaining hives.



Figure 4 - Top Pollen Types (> 5%) by Hive

Figure 5 shows seasonal trends (2019) for pollen from BCR honey sampling. Basswood is strong early, Soybean and Trefoil peak in August, Buckthorn and Total Clover have a presence throughout the year, and finally Boneset and Knapweed are late in the season. Peroxide values are higher in August and October. This would indicate that Soy and Trefoil are important peroxide contributors, as is Boneset and Buckthorn. Clover is strong throughout.



Birds Foot Trefoil
 Soybean
 Boneset
 Basswood
 Clovers
 Buckthorn
 Knapweed

Figure 5 - Selected Pollens by Percent through 2019 (BCR)

NORTON honey samples are examined seasonally in **Figure 6**. Soybean peaks in August, Trefoil in September and Boneset in October. Sunflower is significant in the autumn months while the presence of Clover is constant. The August honey sample had a peroxide of 5 with both September and October being even stronger (7.5). This is consistent with BCR (**Figure 5**) in that Soy and Trefoil are important, as is Boneset and of course Clover.



Figure 6 - Selected Pollens by Percent through 2019 (NORTON)

Figure 7 presents pollen data for July through September at the Lavender farm (BH). Here July and August have peroxide values of 5 with September dropping to a 2. The usual contributors to high peroxide values (Trefoil, Soybean and Boneset) are present but in much smaller quantities. The three most significant contributors are Basswood, Clovers and Lily. The fact that the September sample has a peroxide value of 2 with a substantial amount of Lily pollen sheds doubt on its contribution to elevated peroxide levels. Lastly, we should discuss SWIFT. This hive, sampled in September, had a peroxide value of 5. Its relative pollen amounts are represented in **Figure 4**. Soybean, Trefoil and Clovers made up ~77 % of the pollen with the remainder being mainly Sunflower and Basswood.



Figure 7 - Selected Pollens by Percent through 2019 (BH)

Pollen data from 2020 are not available at this time. Our data would indicate that the presence of Soybean, Birds Foot Trefoil, Boneset/Golden Rod/Aster and the Clovers are consistent with a healing honey (high peroxide). Other pollens (Lily and Basswood) may yield higher peroxide values but the data has inconsistencies. Nectar may be contributing factor. At present, only pollen analysis has been performed and it is not as comprehensive as desired. However, there are a substantial number of peroxide analyses which leads into the following discussion.

Peroxide Values across Three Years:

The trend observed for pollen can be correlated to season. Thus, there is value in sampling honey multiple times through the year and measuring the peroxide value. This is less expensive than pollen analysis and may show some useful trends.

For the years 2018 through 2020, the peroxide levels from multiple honey samples (5-6) from 10 of the 120 Cool Creek Apiaries hives were determined. **Figure 8** shows their locations.



Figure 8 - Location by GPS of Cool Creek Hives

Examining all the peroxide data by year and month reveals some general trends. **Figure 9a** and **9b** show the average peroxide value by year and month. Yearly data shows a decrease in peroxide values from 2018 on. The monthly breakdown is consistent with what was mentioned earlier; the higher peroxide values are associated with those plants blooming in the latter season, peaking in October.





Figures 9a and 9b – Average Peroxide Value by Year and Month

It is instructive to look at individuals hives and how their peroxide values change through the year. **Figures 10a, 10b** and **10c** reveal which hives show similar trends across the three years.



Figure 10a – Average Peroxide by Month for three of ten Hives

Figure 10a highlights the hives with a tendency to high peroxide values early in the year; BH, KELLAM and MG. The peroxide counts peak in June/July although they do show solid values in October as well. KELLAM and MG are the two westernmost hives and BH is the furthest south. **Figure 10b** shows the results for BCR, BOSTON and LBC, all of which peak in August, again with a strong showing in October. They are the most central cluster of the ten hives.



Figure 10b – Average Peroxide by Month for three of ten Hives

Finally **Figure 10c** reveals the four hives that have their strongest values in September and October. Those are GARLOW, LITTLE, NORTON and SWIFT. Late season honeys do indeed have the strongest peroxide values. You will note that these hives are eastern most "cluster" and three are within 10 km.



Figure 10c – Average Peroxide by Month for four of ten Hives

The geographical trends noted from the three hive "clusters" (Figures 10a-10c) are significant. These ten hives are in a region of ~150 square kilometers. The peroxide values are averaged over 3 years and are still similar for each "cluster". The hives in a cluster are within 5-10 km and presumably have similar flora for foraging. The differences between clusters are likely due to local plantings and certainly the lavender farm is an outlier. Differences are observed by season, by year and by location.

Conclusions:

Peroxide values have been determined across 3 years and the pollen content across 2 years. It is instructive to assess the presence of different pollen, as there are annual changes associated with weather or a farmer's whim. Seasonal changes occur in hives at a specific location, there are differences across yearly weather patterns and from geographical location. The individuality of honey samples is hardly surprising. Sampling honey once a year makes

interpretation difficult but honey harvested multiple times a year has shown some useful trends.

Pollen analysis confirmed seasonal trends and some effects of geography. Not surprisingly, early blooming plant pollen was found in honey harvested in spring and early summer. Honey from later in the year revealed a similar phenomenon. Our indicator for healing honeys is the level of peroxide. Peroxide is formed by the oxidation of glucose by glucose oxidase, which means nectar plays a role. Plants that produce both pollen and nectar that is favoured by honeybees will be present in the data. Plants in which only nectar is harvested are not observed. Thus, one has to be cautious in linking peroxide to pollen grains identified using melissopalynology.

Nectar evaluation is typically done by sensory analysis - taste, smell, color. The beekeeper (and the consumer) use taste and olfactory analysis to verify the honey type. "If it tastes like Basswood honey, it is Basswood honey". Flavonoids and phenolics lead to the aroma and taste associated with specific floral sources. Collecting and identifying honey's aroma is problematic, although we are developing a method to do just that. Regardless of whether the peroxide value is tied to pollen or nectar we can unequivocally state that it is highest in the autumn honeys.

We could not have done this without the generous contributions from over 70 beekeepers. Thank you to all who contributed to our study and especially those used in this article; Cool Creek Apiaries, Ferguson Apiaries, Creekside Farm, Walker Apiaries and I. W. Sloss

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