

Nutrients in Social Wasp (Hymenoptera: Vespidae, Polistinae) Honey

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ABSTRACT Previous investigators have questioned the temporal occurrence, biochemistry, and nutritional use of honey sometimes present in nests of some social polistine wasps. Honey of species in the genera *Polistes* and *Polybia* contains diverse amino acids. Inositols (alicyclic polyalcohols) also are present in the honey of both genera; quercitol was the most abundant inositol in honey of *Polistes metricus* (Say) from Missouri, but it was not present in honey of *Polybia diguetana* R. du Buysson from Costa Rica. Honey of *P. metricus* from Missouri is similar to that of *Brachygastra mellifica* (Say) because it contained more fructose than glucose. Honey has been seen in *P. metricus* nests in Missouri at all phases of the colony cycle except late preemergence (1st pupae); scattered but suggestive observations indicate that swarm-founding tropical polistine wasps store honey primarily during low activity phases of the colony cycle in locales with alternating wet and dry or subtropical seasons. Although the nutritional value of wasp honey seems clear, the nutritional role of honey in wasp colonies remains unknown.

KEY WORDS *Polistes*, *Polybia*, Epiponini, wasp, honey, amino acid, inositol, quercitol, glucose, fructose

HONEY STORAGE IS well-known in highly social bees such as honey bees (*Apis*: Apinae), stingless bees (e.g., *Melipona*, *Trigona*: Meliponinae), and bumble bees (*Bombus*: Bombinae) (all reviewed by Michener 1974). Honey storage by social polistine wasps (Vespidae: Polistinae) is less well-known, although it was first reported nearly 200 yr ago. Azara (1809, pp. 171-172) drew the attention of European naturalists when he described the collection in Paraguay of a social wasp nest that contained honey. Although Walckenaer (footnote in Azara, 1809, p. 172) and Latreille (1824) believed Azara had mistaken bees for wasps, St. Hilaire's description (1825) of honey in a nest made of paper like those of European wasps confirmed Azara's observations, as reported by Latreille (1824) and reconfirmed by White (1841). These wasps were swarm-founding species, now placed in Epiponini (Carpenter 1982); they have large colonies and build enclosed nests with multiple combs of cells. The biology of Epiponini is reviewed by Jeanne (1991). Honey storage by the epiponine *Brachygastra mellifica* (Say) in south Texas has been described by Schwarz (1929), Bequaert (1932), and Sugden and McAllen (1994).

Polistes (Polistini) are small-colony paper wasps that found nests independently and that build single-

comb, unenclosed nests (biology reviewed by Evans and West Eberhard 1970, Reeve 1991). Honey storage by *Polistes* was reported by Lepageletier (1836, pp. 482-483), Rouget (1873, pp. 196-197), Brongniart (1895), and Marchal (1896). von Ihering (1904) reported observations on honey storage by Brazilian wasps, including species of *Mischocyttarus* (Mischocyttarini), which resemble *Polistes* in most aspects of their biology (reviewed by Gadagkar 1991). Jeanne (1972) describes honey storage by *M. drewseni* de Saussure.

Wheeler (1908) and Rau (1928) provide extensive discussions of honey storage by wasps, especially *Polistes*. Honey of epiponine wasps typically fills half or more of a nest cell that has no egg or larva, and there may be hundreds to thousands of honey-containing cells in a nest at one time. Such honey is sometimes collected for human consumption (e.g., Vellard 1939, Crane 1990). Honey in epiponine nests also may occur as hard crystalline (candied) spheres in nest cells or as small droplets on nest cell walls or, uncommonly, as droplets in interstices of the nest envelope (e.g., *Polybia aequatorialis* Zavattari, S. O'Donnell, personal communication). Honey in nests of *Polistes* and *Mischocyttarus* occurs most often as droplets, each usually $\approx 5 \mu\text{l}$, or less frequently as candied blobs. The honey occurs on the walls of nest cells that are empty or that contain an egg or small larva (J.H.H., unpublished data).

In no case is the honey in wasp nests sited where larvae can feed on it. Only adults can feed directly on it, and it is not known if adult wasps, like honey bees (Winston 1987), regurgitate honey from their crops to larvae, as Marchal (1896) believed to be the case.

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Pathways of nutrient movement among individuals of a social wasp colony can be as intertwined as those of any social insect (Hunt et al. 1987, figure 12.2 in Hunt [1991]), and intracolony movement pathways and primary consumers of honey in wasp colonies remain to be fully documented.

Lepeletier (1836) observed honey in *Polistes* nests during the phase of the colony cycle in which males and reproductive females are produced, and he speculated that the honey must be essential for the development of these reproductives. Marchal (1896) was skeptical, because he found honey in *Polistes* nests only in the preemergence phase. Rouget (1873) saw no natural honey storage at all in *Polistes*, but colonies that were fed sugar stored it as honey at the end of the colony cycle, when there were few if any larvae to be fed, and Rouget saw no use in such end-of-season honey storage. Strassmann (1979) addressed Rouget's concern when she described the use of honey in nests of *Polistes annularis* (L.) in Texas as a source of mid-winter nourishment for gynes (inseminated females that are potential queens of the coming nesting season) that emerged from sheltered hibernacula on warm midwinter days, and she proposed an adaptive role for honey in enhanced gyne survivorship. Hunt (1982, 1991) proposed honey to be 1 of several sources of liquid proteinaceous nourishment that can undergird the origin and elaboration of sociality in Hymenoptera.

Lassaigne (1824) described the color, solubility, and crystalization of the wasp honey collected by Saint-Hilaire (1825). Bertrand (1895) reported that honey collected from a *Polistes* nest did not turn the plane of polarized light to the left, as does honey bee honey. The disaccharide sucrose, which turns polarized light to the right, is a principle carbohydrate of many flower nectars (Maurizio 1975). Honey bees mix nectar with salivary enzymes (Maurizio 1975, White 1975) that cleave sucrose into the monosaccharides glucose and fructose and, concomitantly, cause polarized light to be turned to the left (i.e., the plane of polarized light becomes inverted). Bertrand (1895) concluded that wasp honey must contain primarily sucrose and dextrose (glucose) without levulose (fructose), and he proposed that wasp honey must therefore consist of concentrated nectar without alteration by invertase. However, Santolaya and Gentile (1953) reported 74% reducing sugars, which include glucose and fructose, and 5% sucrose (saccharose) in honey of *Polybia scutellaris* (White) from Cordoba, Argentina. Sugden and McAllen (1994) performed gas chromatographic analyses of major sugars in honey of *Brachgastra mellifica* (Say) from southern Texas, and they reported the probable identity (given here in rank order from most to least) of fructose, "other", glucose, sucrose, stachyose, raffinose, melibiose, and arabinose. Burgett (1974) gave evidence for the presence of glucose oxidase, which can inhibit bacterial activity, in honey of the epiponine *Protonectarina sylberiae* (de Saussure). Crane (1990) said, "it appears likely that both invertase and glucose oxidase systems were developed . . . by all 'honey'-storing Hymenoptera in the Apidae,

Vespidae, and Formicidae." She advocated use of the word "honey" for the stored carbohydrate food of any social insect that has invertase and glucose oxidase systems.

Two studies strongly suggested that honey can be important to nutrient economy in wasps during the active phases of the colony cycle. Rossi and Hunt (1988) placed 5- μ l droplets of slightly dilute *Apis* honey into nests of *P. metricus* beginning soon after nest founding and continuing twice weekly until offspring emergence. Supplemented nests produced 1st offspring earlier than did control colonies, and 1st-emerged offspring of supplemented colonies had a higher percentage of fat than not only control offspring but also foundresses. Dove (1994) repeated the experiment and found that *P. metricus* colonies that received honey supplementation in the preemergence phase attained larger nest sizes and produced more offspring by the end of the season, but the frequency of workers among total offspring was lower than in controls.

It seems reasonable that honey can significantly affect both development and life history of other honey-storing wasps as it does for *P. metricus*. Nutrient analyses of wasp honey can thus be of direct relevance to broad issues in the biology of social wasps. The contents of *Polistes* honey have never been examined, nor are there analyses of amino acids in any wasp honey, although amino acids are well-known constituents of *Apis* honey (e.g., White 1975, Davies 1975) and, as free amino acids, of floral and extrafloral nectars (Baker and Baker 1973). Here we analyze amino acids and inositols in honey of wasps in the genera *Polistes* (Polistini) and *Polybia* (Epiponini) plus major sugars in honey of a *Polistes* species.

Materials and Methods

Study and Collection Sites. Honey-containing nest combs from an active colony of *Polybia occidentalis* (Olivier) and another of *Polybia diguetana* R. du Buysson were collected on 31 January 1984 at Hacienda La Pacifica near Cañas, Guanacaste Province, Costa Rica. Nest combs containing honey were taken from a colony of *P. diguetana* at Monteverde, Puntarenas Province, Costa Rica, on 9 January 1996. Observations of honey in epiponine nests have been made occasionally at other times at these and other Neotropical locations.

Three honey-containing preemergence nests of *Polistes humilis synoecus* de Saussure were collected from December 1988 to February 1989 at the CSIRO Black Mountain Site, Canberra, ACT, Australia. Two honey-containing, preemergence nests of *P. metricus* were collected at Washington University's Tyson Research Center near Eureka, St. Louis County, MO, on 2 May 1988. Three similar *P. metricus* nests were collected on 17 May 1996. Three postemergence *P. metricus* nests were collected on 7 July 1997 and another on 30 July 1997 at the Missouri Botanical Garden's Shaw Arboretum near Gray Summit, Franklin County, MO. Sporadic observations of honey in *P. metricus*

nests have been made over the past 20 yr at Tyson Research Center and Shaw Arboretum. Vouchers specimens of the *Polybia* species and *P. metricus* are in the Museum of Natural History of the University of Missouri-St. Louis.

Amino Acids. The samples of *P. occidentalis* and *P. diguetana* honey collected in 1984 were held in situ in the nest comb under laboratory ambient conditions until May 1988. At that time samples of honey were taken from these combs and from combs of *Polistes humilis* collected a few months earlier, and from *P. metricus* combs, which were freshly collected. For each nest of each species, honey droplets were placed on the walls of a clean glass vial by using a spatula, and the samples were frozen at -80°C until analysis in July 1990. Sample preparation was preceded by suspension of the honey droplets in deionized water. A 10-mg aliquot of each sample was hydrolyzed with 6 M HCl at 110°C for 24 h under a nitrogen atmosphere and then freeze-dried. Samples were derivatized prior to amino acid analysis by adding 100 μl of 2:2:1 ethanol:triethanolamine (TEA): H_2O , drying under a vacuum, and then adding 20 μl of 7:1:1 ethanol:TEA: H_2O :phenylisothiocyanate (PITC) to each sample. The derivatizing mixture was allowed to react for 20 min under a nitrogen atmosphere at 22°C . Samples were dried and reconstituted in a phosphate buffer containing 5 mM sodium phosphate with 6% acetonitrile. Samples and external amino acid standards were quantified with a Waters Associates high-performance liquid chromatography (HPLC) gradient system equipped with a Pico-Tag column (Heinrickson and Meredith 1984, Andersen et al. 1989).

Glucose and Fructose. Nests collected in 1997 were frozen with honey droplets in situ at -20°C within 5 h of collection. For analysis, honey droplets were removed from nests, weighed, and dissolved in deionized water. Honey droplets from the 3 nests collected on 7 July were pooled for 1 sample; droplets from the nest collected on 30 July were pooled for a 2nd sample. Both samples were analyzed enzymatically by the method of Passonneau and Lowry (1993). The reagent consisted of 50 mM Tris buffer at pH 8.1, 2 mM magnesium chloride, 0.5 mM ATP, 0.5 mM NADP, and 0.02% BSA. To measure glucose, 1.0 unit per ml hexokinase and 0.1 unit per ml glucose-6-phosphate dehydrogenase were added to a mix of 0.025 ml of sample in 3.0 ml of reagent, and the increase in NADPH was monitored in a Perkin-Elmer Lambda 3B double-beam spectrophotometer. At the completion of the glucose reaction, fructose was measured by addition of 0.4 units per ml phosphoglucosomerase and spectrophotometric monitoring. The millimolar extinction coefficient of NADPH was taken to be 6.22 at 340 nm.

Inositols. Inositols are alicyclic polyalcohols (polyols) with hydroxyl groups directly attached to the carbons of the ring. Samples collected in 1996 were prepared for analysis by a procedure that removes 99% of the reducing sugars and that retains polyols (Ostlund et al. 1993). A water-diluted sample of honey was shaken with 250 μl of washed Amberlite IR-120(H+) for 45 min; the supernatant was then transferred to a

tube containing 350 μl of Amberlite IRA-440c(OH) and shaken for 2.5 h. Next, the supernatant was passed over a C-18 Sep-Pak (Waters Associates, Milford, MA) that had been washed with methanol and water, and finally the samples were vacuum-dried before derivatization.

Samples were derivatized with pentafluoropropionyl (PFP) groups (Ostlund et al. 1993) by reacting with 40 μl of an acetonitrile solution containing 10% pentafluoropropionic anhydride (PFPA) for 30 min at 65°C . An aliquot was then dried and redissolved in a solution of 0.35% PFPA in acetonitrile prior to chromatography. For derivatization with trimethylsilyl (TMS) groups (Sherman et al. 1970), dried samples were treated with 50% bis(trimethylsilyl) trifluoroacetamide (Regis) in dry pyridine for 1 h at 65°C .

The derivatized samples were analyzed by gas chromatography/mass spectrometry (GC-MS). PFP-derivatized samples were run on a Hewlett-Packard 5988A GC-MS in the negative ion chemical ionization (NICI) mode; TMS-derivatized samples were run on a Hewlett-Packard 5970 GC-MS with electron ionization in the positive ion detection mode. Separation of the PFP-derivatized samples was carried out on a Chirasil-Val fused silica capillary column (25 m by 0.32 mm i.d., Alltech Associates, Deerfield, IL) with temperature programming as follows: 92°C for 1 min, $12^{\circ}/\text{min}$ to 112° , $21^{\circ}/\text{min}$ to 180°C , followed by a bake-out of $70^{\circ}\text{C}/\text{min}$ to 205°C . For the TMS derivatives, separation was performed on a J&W DB-210 column (Alltech) (15 m by 0.32 mm i.d.) with temperature programming of 90°C for 0.5 min followed by $20^{\circ}\text{C}/\text{min}$ change to 200°C where it was held for 7 min for bake-out.

Chemical standards of inositols were obtained as follows: D-*chiro*-inositol from Calbiochem; authentic (+)-quercitol and L-*chiro*-inositol as gifts of Laurens Anderson (University of Wisconsin, Madison); and deuterium-labeled (1,2,3,4,5,6- d_6 -D,L-*chiro*-inositol from Ken Sasaki (Connaught Centre for Biotechnology Research, Toronto). The preparation of the latter compound is described in Sasaki et al. (1987). This material also contained small amounts of d_6 -*myo*-inositol, d_6 -*neo*-inositol, and d_6 -*scyllo*-inositol.

Results

Amino Acids. In the honey of 4 wasp species the number of detected amino acids ranged from 12 (*P. diguetana*) to 17 (*P. metricus* 1) (Table 1). All of the detected amino acids are nutritive. Three of the samples contained 6 of the 10 essential amino acids; 2 samples contained 9, and 2 samples contained all 10. The proportional abundances of amino acids in wasp honey are less equitable than those of typical protein reported by King and Jukes (1969). Wasp honeys had from 2 to 4 amino acids with proportions higher than those of the most common amino acid in typical protein and, concomitantly, from 3 to 9 amino acids with proportions lower than the least abundant amino acid in typical protein.

Table 1. Concentrations (mol%) of total amino acids in the honey of 4 wasps species

	<i>P.h.1</i>	<i>P.h.2</i>	<i>P.h.3</i>	<i>P.m.1</i>	<i>P.m.2</i>	<i>P.d.</i>	<i>P.o.</i>	K-J
Asparagine	11.1	7.6	21.3	33.1	4.1	23.2	15.5	10.8
Glutamine	26.4	9.0	16.9	25.2	40.0	21.3	17.4	9.2
Serine	2.7	3.9	4.4	2.6	2.5	—	2.4	8.5
Glycine	4.6	4.6	6.7	1.3	1.1	1.7	4.2	8.0
Histidine*	—	7.2	3.0	3.5	1.7	—	1.3	3.0
Arginine*	4.1	17.6	2.3	4.6	15.5	1.3	3.1	4.4
Threonine*	4.4	17.4	9.6	7.2	4.3	6.6	9.8	6.5
Alanine	14.3	3.3	14.5	8.7	5.3	24.3	19.3	7.8
Proline	8.5	13.6	10.1	6.4	8.7	10.4	12.2	5.2
Tyrosine*	1.4	2.5	1.8	1.4	2.1	5.2	3.9	3.5
Valine*	1.1	1.5	1.9	1.7	5.0	1.1	1.7	7.1
Methionine*	1.6	1.8	1.4	0.9	0.6	1.5	1.7	1.9
Cysteine	17.5	1.7	1.0	0.4	—	1.1	0.9	—
Isoleucine*	—	—	—	0.7	2.5	—	—	4.0
Leucine*	—	3.2	1.9	1.0	1.4	—	2.8	8.0
Phenylalanine*	—	2.2	1.7	1.0	1.0	—	1.7	4.2
Lysine*	2.4	3.1	1.7	0.4	4.5	2.3	2.0	6.0

P.h., *P. humilis* (3 nests); *P.m.*, *P. metricus* (2 nests); *P.d.*, *P. diguetana*; *P.o.*, *P. occidentalis*. Also given is the composition of average protein, as calculated by King and Jukes (1969). Essential amino acids are marked with an *.

Glucose and Fructose. Table 2 gives the mean of 2 assays for each pooled honey sample as weight, micromoles, and percentage of total sample weight for both glucose and fructose. In both samples fructose constitutes 20% by weight of the total honey. Glucose is much less abundant, at 1–3%.

Inositols. In *P. metricus* honey, the principal inositol of both PFP and TMS derivatives was identified as quercitol (1L-1,2,4/2/5-cyclohexanepentol) by the following criteria. The PFP-derivatized honey samples had 2 principal GC peaks with retention times of 135.25 and 138.00 s and of equal intensity; the authentic (+)-quercitol as the PFP derivative had a single major component GC peak at 134.71 s. The mass spectrum of both of the major GC peaks of the PFP-derivatized honey had major ions at m/z 894 (M^-) and m/z 874 ($M^- - HF$; ion intensity of m/z 874 > 894); the same major ions and relative intensities were observed in the spectrum of the PFP derivative of authentic quercitol. We therefore ascribe the presence of 2 GC peaks identified as quercitol in the honey sample, in contrast to only 1 analogous peak in the authentic (+)-quercitol, to the chiral separatory properties of the Chirasil-Val GC column (Leavitt and Sherman 1982). We suggest that the *P. metricus* honey samples

contained both (+)-quercitol and (–)-quercitol, with the (+) form being the 1st of the pair to elute from the GC column. The *P. diguetana* honey sample contained no quercitol.

The TMS-derivatized *P. metricus* honey revealed a single major peak with retention time of 207.6 s; authentic (+)-quercitol had an analogous single peak eluting at 207.4 s. The presence of a single GC peak as the TMS derivative in both cases is attributed to the achiral nature of the separation on the DB-210 GC column. The mass spectra of both the *P. metricus* honey and the authentic quercitol contained the following ions in decreasing order of intensity: m/z 318, m/z 305, m/z 344, m/z 419, m/z 331, and m/z 329. Of these ions, m/z 344 can be related to the molecular ion m/z 524 (not seen) by the loss of 2 TMSOH moieties, and m/z 419 can be the result of the loss of 1 TMSOH and 1 methyl radical from M^+ . Ions at m/z 318 and 305 are common to the TMS inositols.

Although (+)-quercitol appears to be the major inositol in the *P. metricus* honey samples, retention times and the presence of appropriate mass spectral ions during GC-MS of the PFP and TMS derivatives indicate the presence of small amounts of *myo*-inositol, *L-chiro*-inositol, *D-chiro*-inositol, *neo*-inositol, *scyllo*-inositol, and pinitol (Table 3).

Because the mass spectrum of viburnitol (1L-1,2,4/3,5-cyclohexanepentol) would probably be indistinguishable from that of quercitol, and because we had no viburnitol for use as a gas chromatographic retention time standard, we cannot exclude the possibility that our samples are, or contained, viburnitol. However, this is unlikely considering the retention time identities to quercitol that we observed with 2 different derivatives and 2 different columns.

Discussion

Because our amino acid analyses were of hydrolyzed samples, we cannot conclude whether our amino acid data represent protein, free amino acids, or

Table 2. Glucose and fructose by weight, micromoles, and as percent of total sample weight for 2 pooled samples of *P. metricus* honey

	7 July nests	30 July nest
Total honey		
Weight, mg	8.9	36.2
Glucose		
Weight, mg	0.1	1.1
Micromoles	0.6	6.1
Percent	1.0	3.0
Fructose		
Weight, mg	2.0	7.3
Micromoles	11.1	40.5
Percent	22.0	20.0

Each value is the mean of 2 analyses.

Table 3. Inositols found in honey samples of *P. metricus* (3 nests) and *P. diguetana* and identified as PFP or TMS derivatives

Wasp Derivative	<i>P. met. 1</i>		<i>P. met. 2</i>		<i>P. met. 3</i>		<i>P. diguetana</i>	
	PFP	TMS	PFP	TMS	PFP	TMS	PFP	TMS
Inositol								
<i>chiro-</i>	+	+	+	+	+	+	+	+
<i>neo-</i>	+	+	+	+	+	+	+	+
<i>myo-</i>	+	+	+	+	+	+	+	+
Pinitol	+	+	+	+	+	+	+	+
Quercitol	+	+	+	+	+	+	-	-
<i>scyllo-</i>	+	+	+	+	+	+	+	+

+, Indicates a chromatographic retention time identity with an authentic sample as well as the presence of mass spectral ions that are representative of the inositol derivative. -, Denotes that the inositol was absent or below the detection limits of the analysis.

both. Ingested pollen has been found in several *Polistes* species and in *B. mellifica* (Hunt et al. 1991), but we did not examine wasps or honeys in the current study for the presence of pollen. Pollen could contribute significantly to the amino acid composition of honey, and future investigations of wasp honeys should include attention to this possibility. Differences in the ranked proportional abundances of amino acids among the 3 samples of *P. humilis* honey and between the 2 samples of *P. metricus* honey from the same site and date suggest use of diverse, possibly nonfloral nectar or nonnectar, sources by these small-colony wasps. Similarities of ranked proportional abundances of the more common amino acids in honey of the 2 large-colony *Polybia* species may reflect use of a similar, possibly floral resource, even though the samples represent different dates and sites separated by ≈ 50 km.

The honey of *P. metricus* is similar to that of *B. mellifera* as reported by Sugden and McAllen (1994) because it contains more fructose than glucose. The small available quantities of *P. metricus* honey precluded analyses for sucrose or other major sugars. At this time, we cannot address the sources of the *P. metricus* honey. We strongly agree with Crane (1975) that the question of whether or not wasps do, indeed, have enzymatic cleavage (inversion) of sucrose merits careful reexamination.

Inositols are known from a wide range of plant and animal tissues, are implicated in a wide range of physiological activities, and are apparently universal as phospho-inositides in phospholipids (Posternak 1965). Plants produce inositols, and nearly all plants contain them (Posternak 1965). (+)-Quercitol occurs in various parts, including leaves and twigs, of several species of oak (*Quercus*) (Posternak 1965); various oaks are abundant at Shaw Arboretum where the honey samples we analyzed for inositols were collected. In insects, inositols have been shown to be involved in melanization in *Schistocerca gregaria* (Forsk.) (Dadd 1961), and they are essential for the growth of *Periplaneta americana* L., various plant-feeding Orthoptera, Coleoptera, Homoptera, and several Lepidoptera, although not all insects require them (references in Wigglesworth 1972, Dadd 1973, House

1974). Inositols stimulate feeding in most (herbivorous?) insects (Hanson 1983). In Hymenoptera, inositols are essential for larval development in honey bees (Winston 1987), and a "red ant" had an inositol content of 2,200 γ /g (Posternak 1965). The nutritional role of inositols in the wasps we studied is unknown, but a significant role is probable. Two propositions that can be drawn from the presence of inositols in wasp honey are that the honey may have come, at least in part, from nonfloral sources and that the honey can serve the wasps as a reserve of this potentially important class of nutrients.

Several of the pioneering authors cited in the introduction queried the temporal occurrence of honey in wasp nests. Rau (1928) studied *Polistes* in the same region of Missouri, but not in the same sites used in the current study, and he noted occurrence of honey droplets in *Polistes* nests "throughout the nesting season but in greater abundance toward the end of the summer." Our observations in Missouri have revealed that honey storage in early preemergence *P. metricus* nests (lacking pupae) in May is commonplace. Interestingly, honey seems to be absent from late preemergence nests (with pupae) in June, but it is often found (although sometimes difficult to observe) in nests in July and August from which offspring have emerged. Although the largest *P. metricus* nest studied by Dove (1994) had more abundant honey droplets in early October than any other *Polistes* nest we observed (J.H.H., unpublished data), other active nests at the same site and date had none.

Honey storage by tropical epiponines seems to reach highest levels in the dry season of strongly seasonal locales (Hunt et al. 1987, Kojima 1996) and at high elevation (e.g., Cooper 1993) and latitudinal extremes (Texas and Paraguay) of their distribution. It seems likely that honey storage by epiponines occurs primarily in seasonal sites during times when larvae are few in number.

Investigations describing 2 honey-storing species of the epiponine genus *Brachygastra* at the margins of the genus' distribution in Texas (*B. mellifica*: Sugden and McAllen 1994) and Paraguay (*B. lecheguana*: Azara 1809) have noted that the wasps are not very aggressive (see also Vázquez de Espinosa 1942 [1628]). This contrasts markedly to experience with *B. mellifica* in tropical dry forest regions of Costa Rica and savannas of Venezuela, where it is among the more aggressive epiponine wasps (J.H.H., unpublished data, *vide* R. L. Jeanne), except when a nest is in decline and nearly devoid of both brood and honey (J.H.H., unpublished data).

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