SALIVA COLLECTION AND QUANTIFICATION FROM ADULT BUTTERFLIES (LEPIDOPTERA)[1]

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The coilable proboscis is possessed by over 99% of lepidopteran species and is a key innovation contributing to the success of Lepidoptera as the second largest insect order (Krenn, 2010; Kristensen, 1984; Pogue, 2009). Study of the structure and function of the proboscis is applicable to lepidopteran systematics, natural history (Krenn, 2010), and to the design of microfluidic devices (Tsai et al., 2011; Monaenkova, 2012).

Saliva plays an important role in proboscis function (Krenn, 2010). Saliva is exuded from the food canal of the proboscis through the tip and through slits between the plate-like dorsal legulae and hook-like ventral legulae. The cuticular surface of the proboscis varies along its length, and has microchannels and grooves which could direct saliva between dorsal and ventral sides (Lehnert et al., 2013). Saliva appears as droplets on the surface of the proboscis during assembly (Krenn, 1997), repair (Lehnert et al., 2014), and feeding (Lehnert et al., 2014).

Methods used for collection of butterfly saliva have included rinsing the proboscis with water or buffer (Eberhard et al., 2007), or prompting saliva deposition onto glass (Tokarev et al., 2013). In this paper I describe a new procedure for collecting the saliva of adult butterflies directly from the proboscis into capillary tubes, and I report the volumes of saliva collected from different species. I also report the appearance of a yellow liquid exuded from the proboscis by some lepidopteran adults.

MATERIALS AND METHODS

Butterflies and moths of the families Nymphalidae, Papilionidae, Pieridae, Lycaenidae, Hesperiidae, and Sphingidae were collected at the Clemson University Insectary, Clemson, SC, and other locations in Pickens and Anderson Counties, South Carolina, from August 2013 through June 2014. Adults were maintained indoors at 22-27° and 40-70% relative humidity, with photoperiod of 10:14 (L:D) h, or refrigerated at 4°C. Chrysalises of *Danaus plexippus* (L.) (Nymphalidae), obtained from Shady Oak Butterfly Farm, Brooker, FL, were reared in the laboratory at 16-28°C and 30-81% relative humidity, with photoperiod of 10:14 (L:D) h.

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