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Article title: Effect of climate, anther morphology, and pollination syndrome on pollen availability in *Penstemon*

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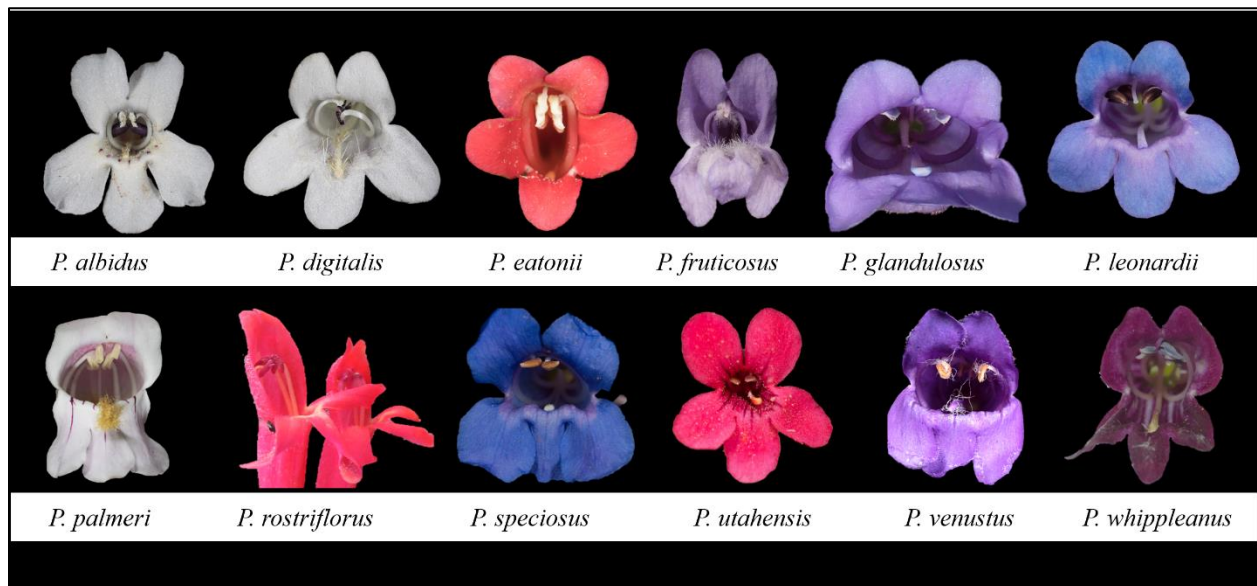


Fig. S1 Flower morphology of the *Penstemon* species included in this study. Picture Credit: Andrea D. Wolfe (*P. albidus*, *P. digitalis*, *P. eatonii*, *P. fruticosus*, *P. leonardii*, *P. palmeri*, *P. speciosus*, *P. utahensis*, and *P. venustus*) and Rosa A. Rodríguez-Peña (*P. glandulosus*, *P. rostriflorus*, and *P. whippleanus*).



Fig. S2 Design and usage of the open-top chambers.

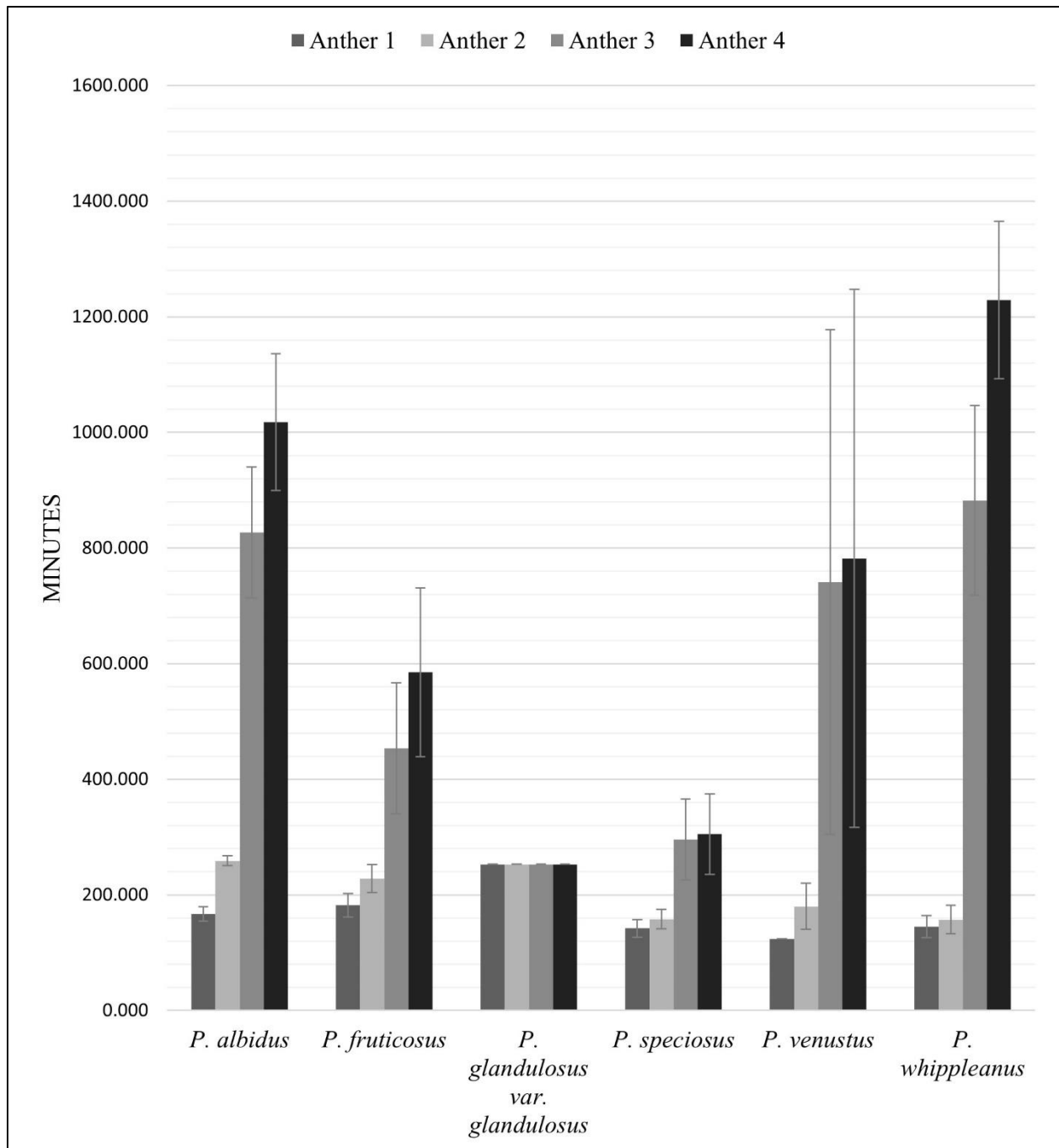


Fig. S3 Anther dehiscence time by species collected in 2018. All these penstemons are Hymenoptera-pollinated. Notice the short anther dehiscence time of *P. g. glandulosus* compared to, say, *P. albidus*. The error bars represent the standard error.

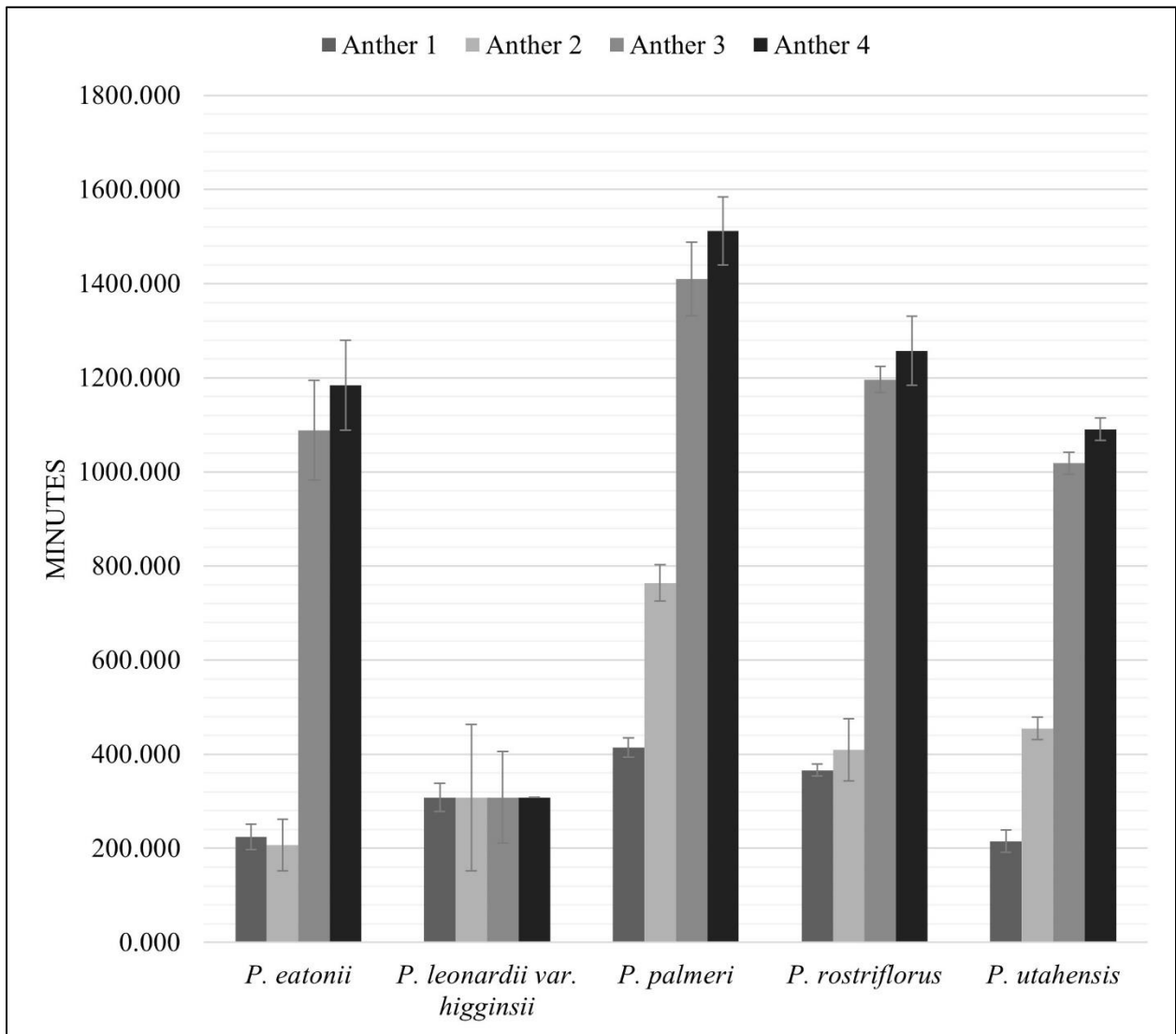


Fig. S4 Anther dehiscence time by species collected in 2019. *Penstemon eatonii*, *P. rostriflorus*, and *P. utahensis* are pollinated by birds, and *P. palmeri* and *P. leonardii var. higginsii* show features of bee pollination syndrome. The error bars represent the standard error.

Table S1 Hedge’s g effect size between pair of species pollinated by Hymenoptera. ** $P < 0.01$ and * $P < 0.05$. The P -value represents the significance level of the comparison between species means of anther dehiscence time using the Gp family of tests as implemented in the R package “survival”.

Species/Anthers	<i>P. albidus</i>	<i>P. fruticosus</i>	<i>P. glandulosus</i>	<i>P. speciosus</i>	<i>P. venustus</i>
Anther 1					
<i>P. fruticosus</i>	-0.252				
<i>P. glandulosus</i>	-1.662	-1.020			
<i>P. speciosus</i>	0.388	0.556	1.517		
<i>P. venustus</i>	0.855	0.870	NA	0.252	
<i>P. whippleanus</i>	0.381	0.546	1.641	-0.042	-0.333
Anther 2					
<i>P. fruticosus</i>	0.520				
<i>P. glandulosus</i>	0.161	-0.301			
<i>P. speciosus</i>	1.558**	0.860*	1.180		
<i>P. venustus</i>	2.047*	0.591	1.298	-0.277	
<i>P. whippleanus</i>	1.671*	0.841	1.121	0.012	0.275
Anther 3					
<i>P. fruticosus</i>	0.833**				
<i>P. glandulosus</i>	1.195*	0.513			
<i>P. speciosus</i>	1.314**	0.448	-0.130		
<i>P. venustus</i>	0.176	-0.704	-0.791	-1.279	
<i>P. whippleanus</i>	-0.107	-0.877	NA	-1.373	-0.246
Anther 4					
<i>P. fruticosus</i>	0.861				
<i>P. glandulosus</i>	1.524*	0.657			
<i>P. speciosus</i>	1.444**	0.706	-0.160		
<i>P. venustus</i>	0.461	-0.379	-0.804	-1.371	
<i>P. whippleanus</i>	-0.430*	-1.318*	-2.069	-2.416**	-0.912

Table S2 Hedge’s g effect size between pair of subgenera. The P-value represents the significance level of the comparison between subgenus means of anther dehiscence time using the G_p family of tests as implemented in the R package “survival”.

Subgenera	<i>Dasanthera</i>	<i>Habroanthus</i>	<i>Penstemon</i>
Anther 1			
<i>Dasanthera</i>			
<i>Habroanthus</i>	0.538		
<i>Penstemon</i>	0.452	-0.187	
<i>Saccanthera</i>	0.287	-0.267	-0.132
Anther 2			
<i>Dasanthera</i>			
<i>Habroanthus</i>	1.144*		
<i>Penstemon</i>	0.477	-0.723*	
<i>Saccanthera</i>	0.591	-0.583	0.305
Anther 3			
<i>Dasanthera</i>			
<i>Habroanthus</i>	0.449		
<i>Penstemon</i>	-0.820**	-1.238**	
<i>Saccanthera</i>	-0.310	-0.811	0.517
Anther 4			
<i>Dasanthera</i>			
<i>Habroanthus</i>	0.706		
<i>Penstemon</i>	-1.029**	-1.827**	
<i>Saccanthera</i>	-0.040	-0.843	0.977

Table S3 Hedge’s g effect size between pair of species sampled in Utah. ** $P < 0.01$ and * $P < 0.05$. The P -value represents the significance level of the comparison between species means of anther dehiscence time using the Gp family of tests as implemented in the R package “survival”. Three of these species are pollinated by birds: *P. eatonii* (Subgenus *Habroanthus*), *P. rostriflorus* (Subgenus *Saccanthera*), and *P. utahensis* (Subgenus *Penstemon*).

Species/Anthers	<i>P. eatonii</i>	<i>P. palmeri</i>	<i>P. rostriflorus</i>	<i>P. utahensis</i>
Anther 1				
<i>P. palmeri</i>	-1.349			
<i>P. rostriflorus</i>	-0.964*	0.418		
<i>P. utahensis</i>	0.268	2.146**	1.453**	
<i>P. leonardii</i> var. <i>higginsii</i>	-0.490	1.195	0.498	-1.025
Anther 2				
<i>P. palmeri</i>	-1.311*			
<i>P. rostriflorus</i>	-0.411	1.169*		
<i>P. utahensis</i>	-0.352	0.648*	-0.117	
<i>P. leonardii</i> var. <i>higginsii</i>	-0.004	1.022*	0.459	0.316
Anther 3				
<i>P. palmeri</i>	-0.686			
<i>P. rostriflorus</i>	-0.234	0.502		
<i>P. utahensis</i>	0.187	1.793**	0.548**	
<i>P. leonardii</i> var. <i>higginsii</i>	1.524*	3.956**	2.018**	3.644
Anther 4				
<i>P. palmeri</i>	-0.807*			
<i>P. rostriflorus</i>	-0.164	0.682*		
<i>P. utahensis</i>	0.185*	0.875**	0.344*	
<i>P. leonardii</i> var. <i>higginsii</i>	1.913**	68.786**	2.318**	1.521*

Table S4 Hedge’s g effect size between pair of treatments. ** $P < 0.01$ and * $P < 0.05$. The P -value represents the significance level of the comparison between treatment means of anther dehiscence time using the Gp family of tests as implemented in the R package “survival”. Species: *P. digitalis*

Treatments/anthers	Control	HH
Anther 1		
HH	-0.386**	
LH	0.035	0.417**
Anther 2		
HH	-0.386**	
LH	-0.012	0.366**
Anther 3		
HH	-0.526**	
LH	0.327**	0.812**
Anther 4		
HH	-0.530**	
LH	0.288*	0.776**

Notes S1

Studied species additional description

We sampled one species in the subgenus *Dasanthera* Raf., *P. fruticosus* Greene. This group has anthers that dehisce from tip-to-tip and across the connective and are covered in wooly hair (type-A anthers, Fig. 1). The function of the hair has not been conclusively studied but probably reduces pollen waste to non-pollinating insects. This species has typical bee-pollinated flowers that are pink or lavender with a large mouth opening (Fig. S1). It occurs in the Pacific Northwest and Canada where it grows in exposed rocky slopes from the foothills to mountain tops (Strickler, 1997).

From the subgenus *Habroanthus* Crosswh., one Hymenoptera-pollinated species, *P. speciosus* Douglas, and a bird-pollinated species, *P. eatonii* A. Gray, were sampled. This subgenus is characterized by anthers that dehisce at the tips only (type-B anther, Fig. 1). *Penstemon speciosus* has a blue corolla and the corolla mouth can be light purple or white presumably to guide pollinators. It is primarily pollinated by wasps from the genus *Pseudomasaris* Ashmead (Vespidae). On the other hand, *P. eatonii* has a red-orange and tubular corolla that are pollinated mostly by hummingbirds. It can be found in Western United States from California to Nevada, Arizona, Utah, and Colorado.

Penstemon albidus Nutt., *P. digitalis* Nutt., *P. palmeri* A. Gray, *P. utahensis* Eastw., and *P. whippleanus* A. Gray belong to subgenus *Penstemon* Schmidel with anthers that dehisce from end-to-end and across the connective (type-C anthers, Fig. 1). *Penstemon albidus* is widely distributed in the Great Plains. The corolla is white or light pink, short, and wider at the corolla mouth (Fig. A1). The lower part of the corolla mouth has pink lines inferred to be nectar guides. *Penstemon digitalis* naturally occurs east of the Rocky Mountains from the Great Plains to Maine and Quebec (Nold 1999). It has white corolla, sometimes with pink lines, and it is pollinated by bees. In Ohio, it can be found naturally growing throughout the state, but it is also cultivated in metropolitan parks for restoration purposes. *Penstemon palmeri* is a short-lived perennial plant (Meyer and Kitchen 1992) with highly aromatic flowers. The

corolla has a large throat and mouth and a very short and narrow tube. The flowers are light pink and the staminode is exerted with copious yellow hairs. This species occurs in the south-western USA in arid environments (Meyer and Kitchen 1992). *Penstemon utahensis* is a Utah endemic with red flowers, a very narrow corolla mouth, and a flower tube typical of a bird syndrome; however, the corolla limbs project radially, similar to species pollinated by bees. Nold (1999) suggested that this species might not have completely adapted to bird pollination. On the other hand, Wilson et al. (2007) considered this species to be hummingbird pollinated. *Penstemon whippleanus* typically has a dark purple corolla (Fig. A1), but there are three less common color morphs: white, yellow, and blue (Fig. A1). This species grows in subalpine and alpine rocky slopes and meadows of Colorado, Utah, and Wyoming, but it also occurs in New Mexico and Arizona (Strickler, 1997; Nold, 1999).

Finally, we sampled three bee-pollinated species from the subgenus *Saccanthera* Benth., *P. glandulosus* Douglas ex Lindl., *P. leonardii* var. *higginsii* Neese, and *P. venustus* Douglas, and one bird-pollinated, *P. rostriflorus*. These species have anthers that dehisce from a pore-like structure at the connective, forming a sac (type-D anthers, Fig. 1). The corolla of *P. glandulosus* is light purple or pink with a wide and flattened mouth and the leaves are glandular and pubescent. It inhabits rocky hillsides with scattered vegetation in Idaho and Washington (Strickler 1997). *Penstemon venustus* has purple flowers with a somewhat circular corolla mouth and is hairy where the anthers are attached to the filament. It occurs naturally in Idaho, Oregon, and Washington. *Penstemon rostriflorus* Kellogg has red-orange and tubular flowers typical of hummingbird-pollinated plants (Fig. S1). It can be found from California to Colorado, Nevada, New Mexico, and Utah.