PETITION TO LIST

The rusty patched bumble bee
***Bombus affinis* (Cresson), 1863**

AS AN ENDANGERED SPECIES
UNDER THE U.S. ENDANGERED SPECIES ACT

Female *Bombus affinis* foraging on *Dalea purpurea* at Pheasant Branch Conservancy, Wisconsin, 2012, Photo © Christy Stewart

Submitted by

The Xerces Society for Invertebrate Conservation

Prepared by Sarina Jepsen, Elaine Evans, Robbin Thorp, Rich Hatfield,
and Scott Hoffman Black

January 31, 2013

The Honorable Ken Salazar
Secretary of the Interior
Office of the Secretary
Department of the Interior
18th and C Street N.W.
Washington D.C., 20240

Dear Mr. Salazar:

The Xerces Society for Invertebrate Conservation hereby formally petitions to list the rusty patched bumble bee (*Bombus affinis*) as an endangered species under the Endangered Species Act, 16 U.S.C. § 1531 *et seq.* This petition is filed under 5 U.S.C. 553(e) and 50 CFR 424.14(a), which grants interested parties the right to petition for issue of a rule from the Secretary of the Interior.

Bumble bees are iconic pollinators that contribute to our food security and the healthy functioning of our ecosystems. The rusty patched bumble bee was historically common from the Upper Midwest to the eastern seaboard, but in recent years it has been lost from more than three quarters of its historic range and its relative abundance has declined by ninety-five percent. Existing regulations are inadequate to protect this species from disease and other threats.

We are aware that this petition sets in motion a specific process placing definite response requirements on the U.S. Fish and Wildlife Service and very specific time constraints upon those responses. 16 U.S.C. § 1533(b). We will therefore expect a finding by the Service within 90 days regarding whether our petition contains substantial information to warrant a full status review.

Sincerely,

Sarina Jepsen
Endangered Species Program Director, The Xerces Society for Invertebrate Conservation

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I. EXECUTIVE SUMMARY

The rusty patched bumble bee (*Bombus affinis*) faces an immediate risk of extinction. Recent research has shown a significant reduction in both the range and relative abundance of this species. Although it was historically common from the Upper Midwest to the eastern seaboard, a recent nationwide study estimated that the rusty patched bumble bee had been lost from 87% of its historic range and that its relative abundance had declined by 95% (Cameron *et al.* 2011a; pers. comm. J. Lozier). A second recent study of historic museum specimens and records from contemporary surveys confirmed that this species has been lost from more than 70% of its entire historic range (including Canada) (Colla *et al.* 2012).

The rusty patched bumble bee is threatened with extinction. Possible causes of its decline include pathogens, habitat loss or degradation, pesticide use, and climate change. Reduced genetic diversity, which could be a result of declining, isolated populations caused by any of the aforementioned factors, likely also threatens this species with extinction. Furthermore, existing regulations are wholly inadequate to protect this species.

Pollinators are critical components of our environment and essential to our food security. Insects – and primarily bees – provide the indispensable service of pollination to more than 85% of flowering plants (Renner 1998 *in* Memmott *et al.* 2004, Ollerton *et al.* 2011), contributing to 35% of global food production (Klein *et al.* 2007). Many vitamins and other nutrients essential to human nutrition are found primarily in plants that require insect pollination (Eilers *et al.* 2011), so the loss of pollinators may pose challenges to human nutrition. In Europe, declines in pollinators have been associated with a parallel decline in insect pollinated plants (Biesmeijer *et al.* 2006).

Bumble bees are among the most iconic and well understood group of native pollinators in North America. They are generalist pollinators that play a valuable role in the reproduction of a wide variety of plants, including human food crops such as tomato, squash, melon, blueberry, pepper, cranberry and clover, and numerous wildflowers.

This petition presents information that the rusty patched bumble bee meets multiple criteria of an Endangered Species under the U.S. Endangered Species Act.

II. CANDIDATE BACKGROUND, STATUS, AND LISTING HISTORY

The rusty patched bumble bee has no legal protection under the U.S. Endangered Species Act or any state endangered species statutes. The rusty patched bumble bee (*Bombus affinis*) has never been petitioned for listing under the Endangered Species Act and it has no federal status. Canada lists the rusty patched bumble bee as Endangered, Schedule 1, under the Species At Risk Act (SARA 2010) and provincially as Endangered in Ontario (Ontario Ministry of Natural Resources 2012). NatureServe ranks the rusty patched bumble bee as G1G2, or Critically Imperiled [at very high risk of extinction due to extreme rarity (often 5 or fewer populations), very steep declines, or other factors] / Imperiled [at high risk of extinction due to very restricted range, very few populations (often 20 or fewer), steep declines, or other factors] (NatureServe 2012). The rusty patched bumble bee is listed as imperiled on the Xerces Society's *Red List of Pollinator Insects* *of North America* (Shepherd *et al.* 2005). The Wisconsin Department of Natural Resources, the Connecticut Department of Environmental Protection, and the Michigan Department of Natural Resources list the rusty patched bumble bee as a species of Special Concern, but there are no laws regulating its use, possession, or harvesting (Wisconsin DNR 2011; Connecticut Department of Environmental Protection 2011; Michigan DNR 2012).

III. POPULATION STATUS AND DISTRIBUTION

A. Historic Distribution

Historically, the rusty patched bumble bee was broadly distributed across the eastern United States and Upper Midwest, North to Maine in the U.S. and southern Quebec and Ontario in Canada, south to the northeast corner of Georgia, reaching west to the eastern edges of North and South Dakota at elevations from sea level to circa 6,000 feet (See Figure 1).

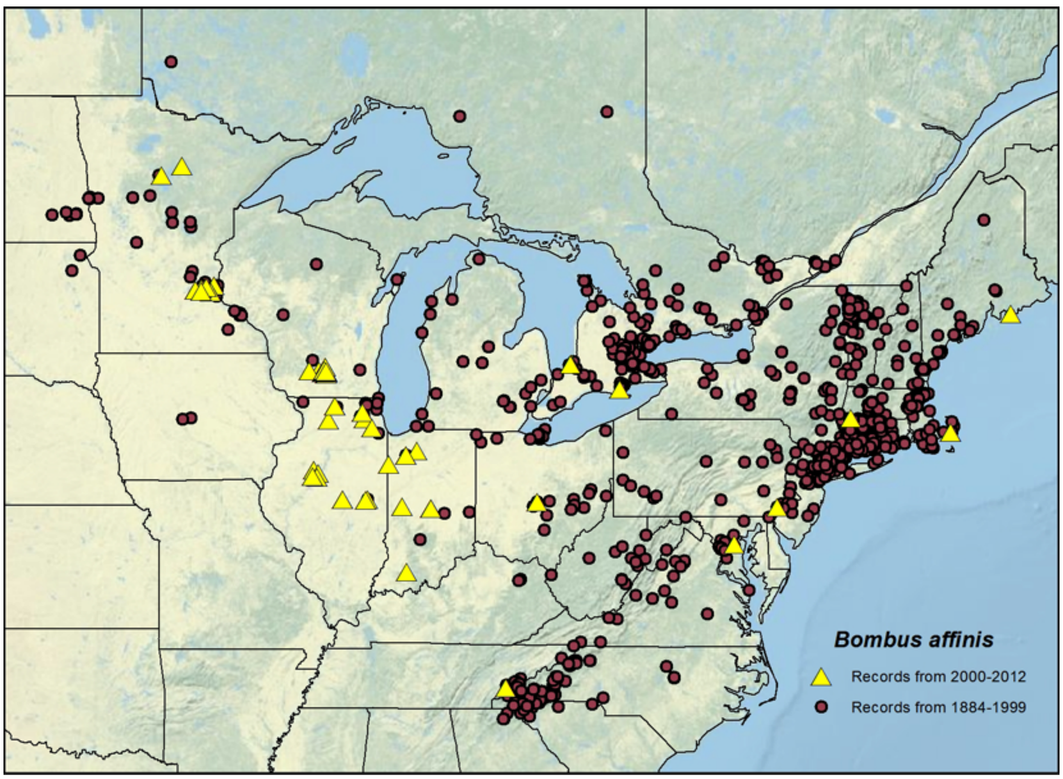

Figure 1. Historic (1884-1999) distribution of the rusty patched bumble bee represented by circles; contemporary (2000-2012) distribution of the rusty patched bumble bee represented by triangles. Note that bumble bee search effort has dramatically increased since 2000 relative to the entire 20th Century (see Figure 1 in Colla *et al.* 2012, which uses the same dataset), and many observers have specifically targeted the rusty-patched bumble bee in recent years. Data for this map is from Colla *et al.* In Prep. Original data sources are listed in Appendix I.

B. Population Status

A 2007-2009 field survey of more than 16,000 bumble bees from throughout the U.S., compared to collections of more than 73,000 historical bumble bee specimens, revealed that the historic range of the rusty patched bumble bee has contracted by an estimated 87% (Cameron *et al.* 2011a). This same study concluded that the relative abundance of the rusty patched bumble bee has declined by 95% (pers. comm. with J. Lozier); the species was only detected at low numbers in three Illinois locations and one Indiana location in the recent survey (Cameron *et al.* 2011a). A separate analysis of nearly 45,000 eastern bumble bee records from museum collections and contemporary surveys considering both Canada and the US concluded that the rusty patched bumble bee has suffered greater than a 70% range decline; the authors classify this species as *Endangered* using modified criteria from the International Union for the Conservation of Nature (Colla *et al.* 2012; IUCN 2001). The relative abundance of the rusty patched bumble bee from 1991-2009 is 87% less than its relative abundance in collections from <1931-2000 (derived from data presented in Figure 1, Colla *et al.* 2012).

A 2004-2006 study of approximately 9,000 bumble bees from 28 sites where the rusty patched bumble bee historically occurred, plus 15 sites within the bee's historic range in eastern North America and eastern Canada found only a single rusty patched bumble bee in southern Ontario, despite numerous reports that the species was historically common (Colla & Packer 2008).

In addition to these three studies, there are multiple local examples of extirpations and decreases in the relative abundance of the rusty patched bumble bee, summarized in Evans *et al.* (2008) and presented below.

1. Midwestern United States

A study comparing records from a contemporary survey to historic records of the rusty patched bumble bee in Illinois revealed that the distribution of this species has decreased by nearly one-third in that state since 2000, with only 67% of its pre-2000 distribution remaining (Grixti *et al.* 2009).

A multi-year survey in northern Indiana that included collecting over 880 individual bumble bees found 25 rusty patched bumble bee specimens out of 217 (12%) in 2001, two out of 451 (0.004%) in 2002, and zero out of 553 in 2003 (R. Jean & P. E. Scott pers. comm. with E. Evans, September 2007).

A survey from 1994-1995 of 464 bumble bees at Long Lake Regional Park in New Brighton, Minnesota found 98 rusty patched bumble bee individuals (Reed 1995; C. Reed, pers. comm. with E. Evans, June 2007). A survey during the summers of 2007 and 2008 at the same park of 593 bumble bees found no rusty patched bumble bees (E. Evans, personal observation, July 2008).

2. Northeastern United States and southeastern Canada

A 2003 survey including over 1,261 bumble bees in New York, where the rusty patched bumble bee was considered historically to be "moderately abundant in the eastern to southern parts of the state…" (Leonard 1928, referenced in Giles & Ascher 2006), failed to find any rusty patched bumble bees (Giles & Ascher 2006). In the same paper, the authors noted that the rusty patched bumble bee is well represented in historical collections from the northeastern U.S.

A study by Colla and Packer (2008) of two sites in southern Ontario comparing a recent collection of nearly 1,200 bumble bees to a historical collection (Macfarlane 1974) of >3,600 bumble bees from the same locations revealed that the rusty patched bumble bee has been extirpated from those sites, despite the fact that it comprised approximately 14% of the 1970s collection. P.Williams reported that the rusty patched bumble bee was formerly abundant in Toronto, Ontario in 1983 but was not been seen during regular surveys in the Toronto area from 2003 to 2008 (pers. comm. with E. Evans, July 2008).

3. Southeastern United States

In a sample of nearly 1,000 bumble bees on the Patuxent National Wildlife Refuge in Maryland from 2002 to 2007, a single rusty patched bumble bee specimen was collected in 2002 and none have been collected since (S. Droege, pers. comm. with E. Evans, Feb. 2008). The same researcher reports that rusty patched bumble bees were numerous in collections in the 1980s in areas near Patuxent National Wildlife Refuge north of Baltimore, Maryland and in northern Delaware (S. Droege, pers. comm. with E. Evans, Feb. 2008).

Since 2000 the rusty patched bumble bee has not been seen in the Great Smoky Mountains National Park in North Carolina and Tennessee, where it was once abundant (A. J. Mayor, pers. comm. with E. Evans, Sept. 2007). Surveys of spring queens in North Carolina consistently found the rusty patched bumble bee from 1995 to 2001, yet between 2002 and 2007, no rusty patched bumble bee queens were found while other bumble bee species were present (R. Jacobson, pers. comm. with E. Evans, Sept. 2007).

C. Current Distribution

Since 2000, there has been substantial recent interest in bumble bees among scientists, naturalists, and the general public, and collection effort has dramatically increased (Colla *et al.* 2012; Figure 1 therein). Our understanding of the current distribution of the rusty patched bumble bee has been greatly informed by a citizen monitoring effort that began in 2008 to specifically target this species and other rare or potentially rare species of bumble bees (Xerces Society 2012).

While recent incidental observations allow us to identify the current distribution of the rusty patched bumble bee, the multiple recent observations of this species in the Midwestern US should not be interpreted as evidence that this species' populations are stable or recovering. Because collection effort has changed over time, and because people have been specifically looking for and documenting occurrences of rare species of bumble bees, it is important to evaluate changes in relative abundance and range size when drawing conclusions about whether a bumble bee species' population is declining, stable, or increasing. In fact, every study that has evaluated these metrics on a nationwide, regional or local scale has found that the rusty patched bumble bee has declined dramatically (Cameron *et al.* 2011a; Colla *et al.* 2012; Colla & Packer

2008).

Since 2000, The rusty patched bumble bee has been observed or collected in Connecticut (Litchfield County), Illinois (Champaign, Cook, DeWitt, Dupage, McHenry, Ogle, Peoria and Winnebago Counties), Indiana (Jasper, Marion, Montgomery, Newton and Starke Counties), Iowa (unknown county), Maryland (Anne Arundel and Prince George Counties), Massachusetts (Barnstable County), Minnesota (Cass, Hennepin, Itasca, Ramsey and Washington Counties), Ontario (Lambton and Norfolk Counties), Tennessee (Blount/Swain County), and Wisconsin (Dane and Iowa Counties) (Table 1).

Table 1. Observations or collections of the rusty patched bumble bee since 2000.

Year	State or Province	County	Location	Reference
2008	Connecticut	Litchfield	Salisbury, 0.15 km E jct. US Highway 44 and Taconid Rd.	coll. Chris T. Maier, American Museum of Natural History 022612, L. Richardson database
2012, 2009	Illinois	Winnebago	Private residence in Rockford	B. Williams, pers. comm. with S. Jepsen, August 2009; L. Richardson database
2009, 2008	Illinois	Peoria	Jubilee College State Park	Cameron et al. 2011a; J. James-Heinz pers. comm. with S. Jepsen, Sept. 2008
2012	Illinois	Dupage	Private Residence in Downers Grove	C. Hlohowskyj, pers. comm. with R. Hatfield, Xerces Society, July 2012
2009	Illinois	Cook	Bluff Spring Fen	Cameron *et al.* 2011a
2009	Illinois	Ogle	Castle Rock SP	Cameron *et al.* 2011a
2007	Illinois	Champaign	Champaign-Urbana	Grixti *et al.* 2009; J. Grixti and C. Favret, pers. comm. with E. Evans, Nov. 2007
2007	Illinois	McHenry	Algonquin	Grixti *et al.* 2009; J. Grixti and C. Favret, pers. comm. with E. Evans, Nov. 2007
2007	Illinois	Peoria	Peoria/Airport region	Grixti *et al.* 2009; J. Grixti and C. Favret, pers. comm. with E. Evans, Nov. 2007
2006	Illinois	DeWitt	Weldon Springs	Grixti *et al.* 2009; J. Grixti and C. Favret, pers. comm. with E. Evans, Nov. 2007
2010, 2009	Indiana	Marion	Daubenspeck Park, W. 89th St. & Ditch Rd., Indianapolis	L. Day, pers. comm. with S. Jepsen, July 2010
2002, 2001	Indiana	Jasper		R. Jean, pers. comm. with E. Evans, Sept. 2007
2002, 2001	Indiana	Newton		R. Jean, pers. comm. with E. Evans, Sept. 2007

2002, 2001	Indiana	Starke		R. Jean, pers. comm. with E. Evans, Sept. 2007
2009	Indiana	Montgomery	Alamo (Hwy 234)	Cameron *et al.* 2011a
2000	Iowa			S. Hendrix and C. Gienapp, pers. comm. with E. Evans, Sept. 2007
2002	Maryland	Prince George's	Patuxent Research Refuge	S. Droege, pers. comm. with E. Evans, Feb. 2008
2001	Maryland	Anne Arundel	Laurel, PWRC	L. Moore, H. Herbers, American Museum of Natural History 022612, L. Richardson database
2009	Massachusetts	Barnstable	Jenkin's Bog, Harwich	M. Notestine, pers. comm. with S. Jepsen, Feb. 2010
2012, 2011, 2010	Minnesota	Hennepin	Lake Harriet Peace Garden, Lyndale Park, Minneapolis	J. Knutson, pers. comm. with S. Jepsen, August 2010; E. Evans, pers. obs., July 2011 and June 2012
2011, 2010	Minnesota	Washington	Private Residence in Newport	J. Knutson, pers. comm. with S. Jepsen, Aug. 2010 and Aug. 2011
2012	Minnesota	Hennepin	Private residence in Minnetonka	H. Holm, pers. comm. with R. Hatfield, Xerces Society, July and August 2012
2012	Minnesota	Itasca	Bowstring Lake (northern end) boat ramp, Itasca	K. Pouliquen, pers. comm. with E. Evans, June 2012
2012	Minnesota	Ramsey	Private Residence in Saint Paul	E. Evans, pers. obs., June and July 2012
2011	Minnesota	Washington	Grove Street Overlook Park, where 10th street hits the Mississippi River, Newport	J. Knutson, pers. comm. with S. Jepsen, Aug. 2011
2002	Minnesota	Cass	3 mi E of Cass Lake	coll. J.R. Powers, Doug Yanega 020312, L. Richardson database
2000	Ohio	Franklin	Blendon Woods, Westerville	Coll. R. Thorn
2009, 2006, 2005	Ontario	Lambton	Pinery Provincial Park	Colla and Packer 2008, email sent to BOMBUS listserv by S. Colla, Aug. 2009; Zuzu Gadallah (L. Richardson database)
2000	Ontario	Norfolk	Manester Tract	Colla 2012
2000	Tennessee	Blount / Swain	Gregory Bald (Great Smoky Mountains National Park)	A.J. Mayor, pers. comm. with E. Evans, Sept. 2007
2012, 2011, 2010, 2009	Wisconsin	Dane	Curtis Prairie, University of Wisconsin Arboretum, Madison	N. Rafferty pers. comm. with S. Jepsen February 2011; Email from R. Thorp to BOMBUS listserv, May 2011; S. Carpenter and M. Murray pers. comm. with Xerces Society staff 2011-2012

2012	Wisconsin	Dane	Pheasant Branch Conservancy and nearby roadside, Middleton	C. Stewart, pers. comm. with R. Hatfield, Xerces Society, July 2012
2012	Wisconsin	Dane	Private residence, Waunakee	C. Stewart, pers. comm. with R. Hatfield, Xerces Society, July 2012
2012	Wisconsin	Dane	Owen Conservation Park, Madison	C. Stewart, pers. comm. with R. Hatfield, Xerces Society, July 2012
2012	Wisconsin	Iowa	Private residence in Barneveld	C. Stewart, pers. comm. with R. Hatfield, Xerces Society, July 2012
2011	Wisconsin	Dane	West Madison Agricultural Research Station, Verona	C. Stewart, pers. comm. with S. Jepsen, August 2011
2006	Wisconsin	Dane	Cross Plains	I. Loser, http://bugguide.net/node/view/80952#93112; pers. comm. with S. Jepsen, Dec. 2007

IV. CURRENT AND POTENTIAL THREATS – SUMMARY OF FACTORS FOR CONSIDERATION

The following factors pose substantial threats to the survival of the rusty patched bumble bee: *A. The present or threatened destruction, modification, or curtailment of its habitat or range; C. Disease or Predation; D. The inadequacy of existing regulatory mechanisms; and E. Other natural or manmade factors affecting its continued existence*. Factor *B. Overutilization for commercial, recreational, scientific, or educational purposes* does not pose a substantial threat to the rusty patched bumble bee. Below we summarize the rationale and available evidence for each factor.

A. The Present or Threatened Destruction, Modification, or Curtailment of its Habitat or Range

The rusty patched bumble bee, like most North American bumble bees, faces general threats from habitat alterations that can interfere with its primary habitat requirements, including: access to sufficient food (nectar and pollen from flowers), nesting sites (such as underground abandoned rodent cavities or above ground in clumps of grasses), and overwintering sites for hibernating queens (undisturbed soil). Like many other bumble bees, the rusty patched bumble bee historically occupied the grasslands of the Upper Midwest and Northeast, which have largely been lost or fragmented by agricultural conversion and urban development or transformed by fire suppression, invasive species, and livestock grazing. Noss *et al.* (1995) consider tall grass prairies an endangered ecosystem because they have declined by 85-98%. Specifically, tall grass prairies east of the Missouri River are considered by these authors to be a critically endangered ecosystem, having declined by more than 98%.

Bumble bee species richness, abundance, and genetic diversity are influenced by the quality of habitat on a landscape level. Isolated patches of habitat may not be sufficient to support bumble bee populations (Hatfield & LeBuhn 2007; Öckinger & Smith 2007), and populations of bumble bees existing in fragmented habitats can also face problems with inbreeding depression (Darvill *et al.* 2006 and 2012; Ellis *et al.* 2006). Specifically, Darvill *et al.* (2012) found that bumble bee populations limited to less than 15 km^2 of habitat were more likely to show signs of inbreeding. Goulson (2010, p.193) suggests that a viable population of bumble bees probably requires approximately 3.3-10 km^2 of suitable habitat. A recent study in the western US found a trend that inbreeding in one species of bumble bee was less common in landscapes with increasing natural woodland cover relative to other landscape types (S. Jha, ESA presentation 2012).

1. Agricultural Intensification

Agricultural intensification is primarily blamed for the decline of bumble bees in Europe (Williams 1986; Carvell *et al.* 2006; Diekotter *et al.* 2006; Fitzpatrick *et al*. 2007; Kosior *et al.* 2007; Goulson *et al.* 2008), and may also pose a significant threat to bumble bees in the US. Increases in farm size and changes in technology and operating efficiency have led to many practices that are detrimental to bumble bees, including loss of hedgerows, weed cover, and legume pastures. The widespread application of the herbicide glyphosate in conjunction with increased planting of genetically modified crops that are tolerant to glyphosate has likely reduced the availability of wildflowers in agricultural field margins (Pleasants & Oberhauser 2012), which otherwise would have been an important resource for the rusty patched bumble bee. Another study in northern Alberta found genetically modified herbicide tolerant canola fields to have fewer wild bees than conventional or organic canola fields (Morandin and Winston 2005). Although the rusty patched bumble bee generally nests one to four feet below ground, reports exist of this species nesting above ground, such as “in an open mowing place on the surface of the ground” (Plath 1922). Bumble bee nests may be at risk of being destroyed by farm machinery (Goulson 2003). The broad scale use of pesticides, including a novel class of systemic pesticides (neonicotinoids), poses a unique threat to the rusty patched bumble bee; this topic is discussed in detail below under Factor *E: Other natural or manmade factors affecting its continued existence*.

Hines and Hendrix (2005) found that bumble bee diversity in Iowa prairies was linked to floral abundance and the presence of grasslands in the surrounding landscape, both of which are reduced in modern agricultural landscapes. The decline of the rusty patched bumble bee and other bumble bees in Illinois from 1940-1960 coincides with a period of major agricultural intensification in the Midwest (Grixti *et al*. 2009). Although some flowering crops provide nectar and pollen resources for bumble bees, which can lead to increased densities of bumble bees and colony growth (Westphal *et al*. 2003, 2009), large monocultures do not necessarily improve the reproductive success of bumble bees (Westphal *et al.* 2009), likely because the resources they provide are typically only available for a short period of time. Colonies need floral resources throughout their colony cycle from early spring to fall (Goulson *et al.* 2008).

2. Livestock Grazing

Ungulate grazing can significantly alter the landscape. Studies have shown that grazing can have both indirect and direct effects on bumble bee populations. Indirect effects include removing floral resources (Morris 1967; Sugden 1985; Kruess and Tscharntke 2002a, 2002b; Vazquez and Simberloff 2003; Hatfield and LeBuhn 2007; Xie *et al.* 2008; Kimoto 2010; Scohier *et al*. 2012) and potentially reducing populations of nesting rodents (e.g. Bueno *et al.* 2011), which in turn may reduce the number of nest sites available to bumble bees. Ungulates can directly affect above ground bumble bee nests by trampling (Sugden 1985). The habitat, type of grazer, as well as the timing, intensity, and length of livestock grazing are all factors that can influence how the practice affects flora and fauna (Carvell 2002; Gibson *et al.* 1992; Sjodin 2007). Numerous studies have found intensive sheep grazing to be particularly detrimental to bumble bee populations (Carvell 2002; Hatfield and LeBuhn 2007; Scohier *et al*. 2012), an effect that is likely due to the selective removal of flowers by sheep.

3. Urban Development

The conversion of the landscape to urban and suburban uses continues to transform and fragment habitat, which has likely had a negative effect on populations of many bumble bee species, including the rusty patched bumble bee. Roads and railroads fragment plant populations and thus restrict the movement of bumble bees (Bhattacharya *et al.* 2003). Recent research in northern California found that the overall area of the landscape covered by pavement had a negative effect on the density of bumble bee nests. In addition, bumble bee colony density was greater in natural oak chaparral than other landscape types, including urban areas (Jha & Kremen 2012). The rusty patched bumble bee has been found in some natural areas within urban environments, such as parks, restored prairies, and other natural areas within the urban centers of Philadelphia, PA, Minneapolis, MN and Madison, WI. Some residential gardens and urban parks can provide valuable floral, and in some cases, nesting and overwintering resources, and may serve as important habitat refuges for bumble bees (Frankie *et al*. 2005; McFrederick & LeBuhn 2006, Goulson *et al.* 2010), even though they may not support the species richness that was found historically (McFrederick & LeBuhn 2006).

4. Fire and Fire Suppression

Historically, occasional fires maintained forbs and grasses within meadows and prairies, and prevented shrubs and trees from encroaching. Fire suppression can lead to extensive changes in vegetation structure, including degradation and loss of grasslands and herbaceous species as the shrub community matures (Panzer 2002; Schultz & Crone 1998). The practice of fire suppression has compromised grassland habitats that formerly supported diverse communities of bumble bees. Forest encroachment not only reduces available bumble bee habitat, but also closes off corridors between meadows, which reduces dispersal and foraging opportunities (Roland & Matter 2007). Continued fire suppression not only results in habitat alteration, but also renders the habitat susceptible to catastrophic, large scale, and high temperature fires due to increases in combustible fuel loads, tree density, and fire intolerant species (Huntzinger 2003). These high intensity fires may be particularly harmful to already vulnerable populations of the rusty patched bumble bee.

Prescribed fire can be a valuable tool in restoring native prairie and meadow plant fauna, which in turn has the potential to benefit bumble bees. However, natural or introduced fire can be detrimental to bumble bee populations if not planned and executed carefully with the life history needs of bumble bees considered. In order to protect bumble bee populations, it is recommended that: burns occur during the winter months, only small sections are burned at a time and no more than one-third of an area be burned each year, a specific area is only burned once every 3-6 years, high intensity fires are avoided, and fire breaks be created so that patches of unburned areas exist as a refuge for bumble bees (Hatfield *et al*. 2012). The only known Canadian population of the rusty-patched bumble bee occurs in a large park in which prescribed burns are staggered (S. Colla, pers. comm. December 2012).

B. Overutilization for Commercial, Recreational, Scientific, or Educational Purposes

To the best of the petitioners' knowledge, the rusty patched bumble bee is not produced or sold commercially. While specimens of female workers or males may still occasionally be collected for research purposes, this activity probably does not pose a threat to the overall survival of the species. However, if a rusty patched bumble bee queen is collected, the entire colony will be effectively eliminated. Collection of queens or large numbers of workers or males from populations that are already small and isolated could threaten the rusty patched bumble bee with extinction, although we have no evidence that this practice is occurring.

C. Disease or Predation

1. Pathogens and Parasites of Bumble Bees

Pathogens and parasites pose a substantial threat to the continued survival of the rusty patched bumble bee. Worldwide, reported pathogens and parasites of bumble bees include: viruses, bacteria, fungi, protozoa, nematodes, hymenopteran and dipteran parasitoids, one lepidopteran parasite, and mites (Acari) (summarized in Schmid-Hempel 2001). Pathogen prevalence and fitness effects in wild North American bumble bees are generally not well understood. The microparasites and macroparasites that have been identified as pathogens of concern to wild North American bumble bees (Cameron *et al.* 2011b, page 16) are discussed below.

a. Microparasites

<u>*Nosema bombi*</u>
Nosema bombi is a microsporidian parasite that infects bumble bees primarily in the malpighian tubules, but also in fat bodies, nerve cells, and sometimes the tracheae (Macfarlane *et al*. 1995). Colonies can appear to be healthy but still carry *N. bombi* (Larsson 2007) and transmit it to other colonies. *N. bombi* can reduce colony fitness, as well as reduce individual reproduction rate and life span in bumble bees (Schmid-Hempel & Loosli 1998; Schmid-Hempel 2001; Colla *et al*. 2006; Otti & Schmid-Hempel 2007, 2008; van der Steen 2008; Rutrecht & Brown 2009). This parasite has been observed recently in wild bumble bees throughout North America (Colla *et al.* 2006; Gillespie *et al.* 2010; Kissinger *et al.* 2011; Cameron *et al.* 2011a; Cordes *et al.* 2012).

Cameron *et al.* (2011a) found a significantly higher prevalence of *N. bombi* in declining North American bumble bee species (*B. occidentalis* and *B. pensylvanicus*). Rusty patched bumble bees were tested but the sample size was so low that the data were excluded from the statistical analyses. However, the authors note that the available data show that this species followed the same infection trend of the other declining species with infected individuals collected at four of five sites, and infections detected in 7 of the 14 individuals collected. *N. bombi* infection was significantly lower in species that have not exhibited recent declines in range and relative abundance.

Crithidia

Crithidia bombi is a trypanosome protozoan that can dramatically reduce bumble bee longevity and colony fitness (Brown *et al.* 2003; Otterstatter & Whidden 2004), interfere with learning among bumble bee foragers (Otterstatter *et al.* 2005), increase ovary development in workers (Shykoff and Schmid-Hempel 1991), and decrease pollen loads carried by workers (Shykoff and Schmid-Hempel 1991).

In the UK, researchers found a higher prevalence of the pathogen *C. bombi* in bumble bee populations with reduced genetic diversity, suggesting that as populations become smaller and lose heterozygosity, the impact of this parasite will increase (Whitehorn *et al.* 2010), pushing already at-risk populations closer to extinction.

Crithidia expoeki is a recently identified protozoan characterized from bumble bees collected in North America (Alaska) and Switzerland (Schmid-Hempel & Tognazzo 2010) that may also present a serious threat to wild populations of the rusty patched bumble bee if moved out of its natural range.

Apicystis bombi

Apicystis bombi is a neogregarine protozoa that has been shown to infect 2.5% of rusty patched bumble bee queens in Ontario, Canada (Macfarlane *et al.* 1995). This parasite is associated with rapid death of infected bumble bee queens early in the season (Macfarlane *et al.* 1995; Rutrecht & Brown 2008). It has also been shown to inhibit ovary development and reduce queen longevity (Rutrecht & Brown 2008). More research is needed to understand causal effects that this parasite has on bumble bees and how this parasite is transmitted. This parasite has been found in commercial bumble bee colonies (Meeus *et al.* 2011), and researchers suggest that this pathogen may have been introduced from Europe to NW Patagonia, Argentina on commercial bumble bees, potentially causing an observed population collapse in a native bumble bee species (Arbetman *et al.* 2012). *Apicyistis bombi* poses a serious potential threat to the continued survival of the rusty patched bumble bee.

RNA viruses

RNA viruses that have historically been considered to be specific to honey bees (*Apis mellifera*), including Israeli acute paralysis virus, black queen cell virus, sacbrood virus, deformed wing virus, and Kashmir bee virus, have been recently detected in wild North American bumble bees foraging near apiaries (Singh *et al.* 2010). Deformed wing virus, which is associated with severe winter losses in honey bees (Highfield *et al.* 2009), was also detected in bumble bees in Germany, and the infected bumble bees displayed the same deformities that are typical of infected honey bees (Genersch *et al.* 2006). To understand the extent of the threat to the rusty patched bumble bee, the prevalence of these viruses in wild populations of bumble bees, as well as their effects on bumble bee fitness, are in urgent need of further study.

b. Macroparasites

Locustacarus buchneri

Bumble bees are infected by mites, including *Locustacarus buchneri*, a species that parasitizes the trachea of bumble bees (Husband & Shina 1970). *Locustacarus buchneri* is associated with reduced foraging and lethargic behavior (Husband & Shina 1970) and a significantly reduced lifespan in male bumble bees (Otterstatter & Whidden 2004). Otterstatter and Whidden (2004) reported that this mite was most prevalent in bumble bees of the subgenus *Bombus sensu stricto* (*B. occidentalis, B. moderatus, B. terricola*) in a study in southwestern Alberta. Although the rusty patched bumble bee was not present at these study sites, it belongs to the same subgenus as the species listed above that were heavily parasitized by *L. buchneri*, and thus may also be particularly susceptible to this parasite.

Sphaerularia bombi

Sphaerularia bombi is an entomopathogenic nematode that infects hibernating bumble bee queens and sterilizes them (Schmid-Hempel 2001). In a literature review, Macfarlane *et al.* (1995) notes that bumble bee queens infected with this parasite in New Zealand colonized new areas at a rate of less than 1% of that of healthy queens. This parasite has been detected in the rusty patched bumble bee (Macfarlane *et al.* 1995) and may pose a threat to the long-term survival of the species.

In summary, a variety of microparasites (*Nosema bombi*, *Crithidia bombi*, *Apicystis bombi*, and RNA viruses) and macroparasites (*Locustacarus buchneri* and *Sphaerularia bombi*) can cause harm to bumble bees and pose a threat to the rusty patched bumble bee.

2. Pathogen Spillover

The spread of pathogens to the rusty patched bumble bee from the domesticated common eastern bumble bee (*Bombus impatiens*) and other species of bumble bees that are currently being developed for commercial use threatens the rusty patched bumble bee with extinction. In addition, RNA viruses from the domesticated honey bee (*Apis mellifera*) can be transmitted to bumble bees at shared flowers (Singh *et al.* 2010), and pose a novel threat to the rusty patched bumble bee.

a. Commercial Bumble Bees

Commercial bumble bees are used primarily to pollinate greenhouse tomatoes, and increasingly to pollinate a wide variety of other greenhouse and open field vegetable and fruit crops in the US and worldwide (Velthius & van Doorn 2006; Koppert 2012). The commercial bumble bee industry has grown dramatically in the past two decades (Velthius & van Doorn 2006), coincident with the growth of the greenhouse tomato industry. From 1985-2005, there has been a 30% increase in fresh tomato consumption in the U.S., with more than one-third of the fresh tomatoes in stores coming from hothouses (compared to a negligible amount in the early 1990s) (Calvin & Cook 2005). Commercial bumble bees often escape greenhouses to forage on nearby plants (Whittington *et al*. 2004; Morandin *et al*. 2001), where they interact with wild bumble bees and have the opportunity to transmit pathogens at shared flowers. Commercially raised bumble bees frequently harbor high pathogen loads (Goka *et al*. 2000; Whittington & Winston 2003; Niwa *et al*. 2004; Colla *et al*. 2006) and the spillover of pathogens from commercial bumble bees in greenhouses to wild, native bumble bees foraging near greenhouses has been documented (Colla *et al*. 2006; Goka *et al.* 2006; Otterstatter & Thomson 2008).

Meeus *et al.* (2011) reviewed the effects of invasive parasites on bumble bee declines. They report that the commercial production of bumble bees has the potential to lead to bumble bee declines in three ways: commercial colonies may have high parasite loads, which could then infect wild bumble bee populations; commercial production may allow higher parasite virulence to evolve, leading to the introduction of parasites that are potentially more harmful to wild bumble bees than naturally occurring parasites; and the global transport of commercial bumble bees can introduce novel parasites to which resident, native bumble bees have not adapted. Pathogens reported from commercial bumble bee colonies worldwide include: *Apicystis bombi*, *Crithidia bombi*, *Locustacarus buchneri*, *Nosema bombi*, black queen cell virus, deformed wing virus, Israeli acute paralysis virus, and Kashmir bee virus (Meeus *et al.* 2011). Commercial bumble bee colonies in North America have tested positive for *Crithidia bombi*, *Nosema bombi*, *Locustacarus buchneri*, deformed wing virus, black queen cell virus, sacbrood virus (Morkeski & Averill 2012; Averill unpublished data), and Israeli acute paralysis virus (Singh *et al.* 2010).

The spillover of the microsporidian parasite *Nosema bombi* from commercial to wild bumble bees has been hypothesized as a cause of the sudden, rapid decline of the rusty patched bumble bee and three other closely related North American bumble bees – Franklin's bumble bee (*Bombus franklini*), the western bumble bee (*Bombus occidentalis*) and the yellow banded bumble bee (*Bombus terricola*) (Thorp and Shepherd 2005; Evans *et al*. 2008). This hypothesis is supported by the timing, speed, and severity of the population declines of the rusty patched bumble bee and its close relatives. In the early 1990s, commercial bumble bee producers brought western bumble bee queens from western North America to European bee rearing facilities, where those bees may have come into contact with pathogens of the commercially produced European buff-tailed bumble bee (*Bombus terrestris*). From 1992-1994, the USDA-APHIS allowed commercial colonies of western bumble bees and common eastern bumble bees (*Bombus impatiens*) to return from European facilities to the U.S. (Flanders *et al*. 2003). In 1997, bumble bee producers reported an outbreak of *Nosema bombi* in laboratory populations of the western bumble bee, and eventually had to stop producing this species commercially (Flanders *et al.* 2003; Velthuis & van Doorn 2006; van Doorn 1998 email to BOMBUS-listserv). Coincident with the crash in commercial colonies of the western bumble bee, researchers noticed that the western bumble bee, the rusty patched bumble bee and their relatives began disappearing from the wild in the late 1990s (Thorp & Shepherd 2005; Evans *et al*. 2008; Thorp *et al.* 2010).

This hypothesis is currently under investigation by Dr. Cameron at the University of Illinois. Her research team has already determined that declining bumble bee species harbor higher levels of *N. bombi* than stable species. They initially determined that *N. bombi* was genetically identical to *N. bombi* found in European bumble bees (Cameron *et al.* 2011a), but a more recent, in-depth analysis by Cordes *et al.* (2012) revealed that North American bumble bees harbor a unique strain of *N. bombi*. The research that has been done to date, however, has been insufficient to determine whether or not a European strain of *N. bombi* was released in the U.S., and if so, whether it led to the decline of the rusty patched bumble bee (Cordes *et al*. 2012).

A recent analysis by Szabo *et al.* (2012) found a significant correlation between vegetable greenhouse density, which was used as a proxy for commercial bumble bee use, and the decline of the yellow banded and American bumble bees, but found no significant correlation between vegetable greenhouse density and the decline of the rusty patched bumble bee. However, this analysis did not address the possibility of an acute pathogen spillover event in which a rapid disease spread through wild populations. Furthermore, the analysis did not include areas where bumble bees are used in open field settings.

In Canada, higher levels of the protozoan parasite *Crithidia bombi* were detected in wild bumble bees foraging near greenhouses that used commercial bumble bees (Colla *et al.* 2006; Otterstatter & Thomson 2008), and it was suggested that this pathogen may be implicated in the sudden, widespread decline observed in North American bumble bees in the subgenus *Bombus sensu stricto*, including the rusty patched bumble bee (Otterstatter & Thomson 2008). However, a more recent analysis of pathogen prevalence in wild bumble bees, including the rusty patched bumble bee, did not find evidence that *Crithidia* infections are involved in the decline of U.S. bumble bee species (Cordes *et al*. 2012).

In Japan, where both Japanese and European bumble bee species are imported from the Netherlands for commercial use, researchers found that commercially raised bumble bees had a higher rate of infestation by the tracheal mite *Locustacarus buchneri* than wild bees. Their findings also suggested that a European strain of this mite has likely invaded native Japanese bumble bee populations. (Goka *et al.* 2001, 2006).

In NW Patagonia, Argentina, the commercial buff-tailed bumble bee (*Bombus terrestris*) was introduced from Europe in 2006. Researchers suggest that the highly pathogenic *Apicystis bombi* hitchhiked on the commercial bumble bees and spread to wild bumble bees, potentially causing the observed population collapse in the world's largest native bumble bee – *Bombus dahlbomii* (Arbetman *et al.* 2012).

In summary, the spillover of pathogens from commercial to wild bumble bees has been documented in Canada and Japan, and suspected in Argentina. The recent decline of the rusty patched bumble bee is hypothesized to have been caused by an exotic fungal pathogen introduced from Europe via commercial bumble bees (Thorp & Shepherd 2005). This hypothesis is still under investigation.

b. Honey Bees

The spillover of RNA viruses from honey bees to bumble bees is a recently identified threat to wild bumble bees, including the rusty patched bumble bee. A number of RNA viruses that were formerly thought to be specific to honey bees have now been reported to infect bumble bees (Genersch *et al.* 2006; Meeus *et al.* 2010; Singh *et al.* 2010; Morkeski & Averill 2012). The virulence of many of these RNA viruses in bumble bees has not yet been evaluated. RNA viruses can be transmitted from honey bees to wild bumble bees when they interact at shared flowers (Singh *et al.* 2010), where infected pollen grains left by honey bees are collected by bumble bees and brought back to the nest. Bumble bees may also be infected by RNA viruses when commercial bumble bee producers use honey bee pollen to rear bumble bee colonies (if the pollen is not treated with radiation). Morkeski & Averill (2012) found what appear to be deformed wing virus and black queen cell virus in colonies of bumble bees from two North American commercial production facilities and Singh *et al.* (2010) found Israeli acute paralysis virus in colonies from one North American commercial bumble bee production facility.

D. The Inadequacy of Existing Regulatory Mechanisms

Existing regulations fail to protect the rusty patched bumble bee from threats it faces from habitat loss or modification, diseases, and pesticides.

1. Existing Regulations are Inadequate to Protect this Species' Habitat

Because the rusty patched bumble bee is not listed under the Endangered Species Act, the habitat essential to its survival is not protected from destruction or adverse modification throughout its range in the US. The rusty patched bumble bee is listed as a Species of Special Concern in Wisconsin, Michigan, and Connecticut, although this designation does not provide habitat protection in these states.

The rusty patched bumble bee primarily occurs in the eastern US, although the northernmost part of its range extends into southern Canada. This species is listed as endangered under Canada's Species At Risk Act (SARA 2010), a designation that protects the rusty patched bumble bee and its habitat where it occurs only on Canadian federal land. Because this designation applies to such a small portion of the total range of the rusty patched bumble bee, this designation is insufficient to provide meaningful protection to the entire species.

2. Existing Regulations are Inadequate to Protect this Species from Disease

Existing regulatory mechanisms fail to protect the rusty patched bumble bee from disease. Although the US Department of Agriculture's Animal and Plant Health Inspection Service (APHIS) prohibits the importation of exotic commercial bumble bee species, such as the European buff-tailed bumble bee (*Bombus terrestris*), it does not have any disease requirements for commercial common eastern bumble bees (*Bombus impatiens*) that are moved throughout North America, leaving the rusty patched bumble bee vulnerable to exposure to diseases from commercial bumble bees. Furthermore, there are no regulations requiring commercial bumble bee producers to irradiate honey bee pollen before feeding it to commercial bumble bees (which is part of the bumble bee rearing process), and recent research has demonstrated that one virus stored in pollen can remain virulent after six months of storage (Singh *et al.* 2010).

In January of 2010, the Xerces Society, Dr. Robbin Thorp, Defenders of Wildlife and the Natural Resources Defense Council petitioned the Secretary of Agriculture and APHIS with a request that they require that all commercial bumble bees transported across state lines be certified as disease-free, citing their authority under the Plant Protection Act, the Honeybee Act and the Animal Health Protection Act (Xerces Society 2010). To date, the petitioners have not received an official response from APHIS.

Individual State Departments of Agriculture within the range of the rusty patched bumble bee do not require that commercial bumble bees entering their state be free of the harmful pathogens that have the potential to be transmitted to wild populations of the rusty patched bumble bee. In addition, there are no state or federal regulatory mechanisms that govern the placement of honey bee colonies; the risks associated with this practice include the potential transmission of honey bee diseases (discussed above in section C. Disease or Predation) and competition for floral resources (discussed below in section E. Other natural or manmade factors affecting its continued existence).

3. Existing Regulations are Inadequate to Protect this Species from Pesticides

Existing regulations regarding the approval of new pesticides and the use of existing pesticides fail to protect bumble bees from exposure to harmful pesticides. The Environmental Protection Agency regulates the approval of new pesticides, and this agency currently does not require that research be done to evaluate the lethal or sublethal effects of insecticides, herbicides or fungicides on bumble bees before those chemicals are approved for use.

Although acute toxicity to honey bees (*Apis mellifera*) is evaluated in the pesticide approval process, honey bees are not adequate surrogates for bumble bees in this process. Because bumble bees have different behaviors and life histories than honey bees (for example, they have smaller colonies that are founded each spring, they forage at different times of the day, and they do not process pollen before feeding it to immature bees), they will have different exposure scenarios and may be more vulnerable to pesticides than honey bees (Thompson & Hunt 1999; Fischer & Moriarty 2011; Osborne 2012).

E. Other Natural or Manmade Factors Affecting its Continued Existence

1. Pesticides

Pesticides are used widely in agricultural, urban, and even natural areas and can exert both lethal and sublethal toxic effects on bumble bees. Foraging bumble bees can be poisoned by pesticides when they absorb toxins directly through their exoskeleton, drink contaminated nectar, gather contaminated pollen, or when larvae consume contaminated pollen. Because bumble bees nest in the ground, they may be uniquely susceptible to pesticides used on lawns or turf (National Research Council 2007). Pesticides applied in the spring, when bumble bee queens are foraging and colonies are small, are likely to be most detrimental to bumble bee populations (Goulson *et al.* 2008). Since males and queens are produced at the end of the colony cycle, sublethal doses of pesticides applied at any time during the bumble bee lifecycle can have substantial adverse effects on subsequent generations. Any application of pesticides can threaten bumble bees, but pesticide drift from aerial spraying can be particularly harmful. One study demonstrated that 80% of foraging bees close to the source were killed, and drift can continue to be dangerous for well over a mile from the spray site (Johansen and Mayer 1990). In Europe, the recent declines in bumble bees have been partially attributed to the use of pesticides (Williams 1986; Thompson and Hunt 1999; Rasmont *et al.* 2006).

The rusty patched bumble bee is threatened by the widespread use of pesticides across its range. Insecticides are designed to kill insects directly and herbicides can indirectly affect bumble bees by removing floral resources. There is very little data available on the effect of fungicides on bumble bees, although a literature review suggests that most active ingredients in fungicides are compatible with commercial bumble bees (Mommaerts & Smagghe 2011).

a. Insecticides

Neonicotinoids

Neonicotinoids are a relatively new class of systemic insecticides that are used widely to combat insect pests of agricultural crops, turfgrass, gardens and pets (Cox 2001). Colla & Packer (2008) suggested that neonicotinoids may be one of the factors responsible for the decline of the rusty patched bumble bee since the use of this class of insecticides began in the US in the early 1990s, shortly before the decline of the rusty patched bumble bee was noticed.

A recent study exposing bumble bees to field-realistic levels of the neonicotinoid imidacloprid found an 85% reduction in the production of new queens and significantly reduced colony growth rates compared to control colonies (Whitehorn *et al.* 2012). The authors suggest that neonicotinoids "may be having a considerable negative impact on wild bumble bee populations across the developed world" (Whitehorn *et al.* 2012). Another study of bumble bees exposed to varying levels of imidacloprid found a dose-dependent decline in fecundity and documented that field realistic levels of this pesticide were capable of reducing brood production by one-third (Laycock *et al.* 2012). The authors speculate that this decline in fecundity is a result of individual bumble bees failing to feed, which raises concerns about the impact of this pesticide on wild bumble bees (Laycock *et al.* 2012). Other toxicity studies have demonstrated that contact exposure of imidacloprid and clothianidin to bumble bees can be very harmful (Marletto *et al.* 2003; Gradish *et al.* 2009; Scott-Dupree *et al.* 2009), and an acute oral dose of imidacloprid is highly toxic to bumble bees (Marletto *et al.* 2003, *In* Hopwood *et al.* 2012). Mommaerts *et al.* (2010) found that chronic exposure of three neonicotinoids to bumble bees was dose dependent, and another study by Incerti *et al.* (2003) found that one third of bumble bees in a flight cage exposed to blooming cucumbers treated with a "field dose" of imidacloprid died within 48 hours (*In* Hopwood *et al.* 2012). A study by Gill *et al.* (2012) examining the effects of the combined exposure of bumble bees to field realistic levels of two pesticides – an imidicloprid and a pyrethroid – found that foraging behavior was impaired, worker mortality increased, and both brood development and colony success were significantly reduced.

Other studies have also documented sublethal effects of neonicotinoids on bumble bees, including: reduced foraging ability (Morandin & Winston 2003); reduced drone production and longer foraging times (Mommaerts *et al.* 2010); reduced foraging activity, reduced food storage and reduced adult survival (Al-Jabr 1999); and lower worker survival and reduced brood production (Tasei *et al.* 2000). (In Hopwood *et al.* 2012).

Neonicotinoids are widely used on agricultural crops that are attractive to pollinators, as well as on horticultural plants and lawns in urban and suburban areas. Thus, this class of insecticide is likely to affect the rusty patched bumble bee. Of particular concern is a finding in a recent review of the impact of neonicotinoid pesticides on pollinating insects which found that products approved for home and garden use may be applied to ornamental and landscape plants and turf grass at significantly higher concentrations (potentially 32 times higher) than the allowable concentration of the same products applied on agricultural crops (Hopwood *et al.* 2012).

Other Insecticides

In forested areas insecticides have been used to control defoliators such as tussock moth, gypsy moth, and spruce budworm. In New Brunswick, Canada, bumble bee populations declined drastically when exposed to fenitrothion (reviewed in Kevan and Plowright 1995) resulting in reduced pollination of nearby commercial blueberries and other plants such as orchids and clovers (Kevan 1975; Plowright *et al.* 1978, 1980). Organophosphate, carbamate, and pyrethroid insecticides have been associated with bee poisonings in food crops (Johansen 1977; Kearns *et al.* 1998). Bumble bee deaths have been reported after application of a pyrethroid insecticide to oilseed rape (Thompson 2001). The use of Spinosad, a commonly used insect neurotoxin, has resulted in reduced worker foraging efficiency when bumble bee larvae are fed with pollen containing this pesticide (Morandin *et al*. 2005). Skyrm (2011) observed significant queen mortality when exposed to low doses of Spinosad. In an examination of the effect of chitin synthesis inhibitors on *Bombus,* Mommaerts *et al.* (2006) found that even at very low concentrations, diflubenzuron and teflubenzuron increased egg mortality and removal of larvae.

<u>Transgenic Plants</u>

Increasing numbers of insecticidal transgenic plants are being used to control pest species, and the effect of most of these transgenic plants on bumble bees is not known (Malone & Pham Delègue 2001). However, there is evidence of negative effects on bumble bees of two compounds that are produced in transgenic plants; the soybean trypsin inhibitor (a protease inhibitor) and *Galanthus nivalis* agglutinin (a lectin) have been shown to reduce bumble bee longevity and reproduction when administered experimentally (Babendreier *et al*. 2008). However, the amount of transgene product expressed in pollen and nectar is still unknown, so it is difficult to determine the impact of these products on bumble bees in the wild.

b. *Herbicides*

Herbicides can be a valuable tool for the control of invasive weed species. However, the use of broad-spectrum herbicides to control weeds can indirectly harm pollinators by decreasing the habitat quality for pollinators through removal of flowers that provide pollen and nectar for existing populations (Williams 1986; Shepherd *et al*. 2003, Pleasants & Oberhauser 2012).

Just as pollinators can influence the plant community, changes in vegetation can have an impact on pollinators (Kearns & Inouye 1997). The broadcast application of a non-selective herbicide can indiscriminately reduce floral resources, host plants, and nesting habitat (Smallidge & Leopold 1997). Bumble bees require consistent sources of nectar, pollen, and nesting material during times adults are active, typically from mid-February to late September in temperate areas. The reduction in resources caused by non-selective herbicide use could cause a decline in bumble bee reproductive success and/or survival rates. Kevan (1999) found that herbicides reduced Asteraceae and Lamiaceae flowers in France, contributing to a decline in bumble bee populations. Kevan (1999) also found that herbicide applications have reduced the reproductive success of blueberry pollinators by limiting alternative food sources that can sustain the insects when the blueberries are not in bloom. Kearns *et al.* (1998) state “herbicide use affects pollinators by reducing the availability of nectar plants. In some circumstances, herbicides appear to have a greater effect than insecticides on wild bee populations… Some of these bee populations show massive declines due to the lack of suitable nesting sites and alternative food plants.”

The use of the herbicide glyphosate (Roundup™) has dramatically increased with the widespread planting of genetically modified glyphosate-tolerant corn and soybeans, which were introduced in 1998 and 1996, respectively (Pleasants & Oberhauser 2012). Increased use of glyphosate in agricultural areas has likely led to the reduced availability of wildflowers in field margins – which otherwise would have been an important resource for the rusty patched bumble bee. Pleasants and Oberhauser (2012) estimate a 58% reduction in milkweed, an important nectar plant for bumble bees, in the Midwestern US from 1999-2010, and suggest that this decline is due to the increased use of glyphosate in corn and soybean fields.

2. Population Dynamics and Structure

Since the rusty patched bumble bee has recently undergone a dramatic decline in range and relative abundance (Cameron *et al.* 2011a; Colla & Packer 2008), genetic factors (including reduced genetic diversity, inbreeding depression, and the method of sex determination utilized by bumble bees) are likely among the most significant threats to the long-term survival of this species (reviewed in Zayed 2009).

a. Declining North American Bumble Bees have lost Genetic Diversity

Recent research indicates that populations of the declining western bumble bee (*Bombus occidentalis*) and American bumble bee (*Bombus pensylvanicus*) have lower genetic diversity compared to populations of co-occurring stable species (Cameron *et al.* 2011a; Lozier *et al.* 2011). While the rusty patched bumble bee was included as a target species in one of these studies (Cameron *et al.* 2011a), it is notable that the research team was unable to find and collect a sufficient number of individuals to include this species in their analysis due to its extreme scarcity within the landscape. Another recent genetic study of the declining American bumble bee found an increase in this species' population structure, suggesting that the American bumble bee has become increasingly isolated over the past four decades (Lozier & Cameron 2009).

It is reasonable to expect that the rusty patched bumble bee may have suffered a similar loss of genetic diversity and increase in population structure, although this has not been examined due to the scarcity of this species.

b. Impacts of Genetic Factors on Bumble Bees

Loss of genetic diversity, which is frequently the result of inbreeding or random drift, can pose significant threats to small, isolated populations of bumble bees (Whitehorn *et al.* 2009). A loss of genetic diversity limits the ability of a population to adapt and reproduce when the environment changes and can lead to an increased susceptibility to pathogens (Altizer *et al.* 2003).

Bumble bees have a single locus complementary sex determination system, meaning that the gender of an individual bee is determined by the number of unique alleles at the sex-determining locus (van Wilgenburg 2006). Normally this gender determination comes through a haplodiploid genetic structure in which female bees are diploids and are produced from fertilized eggs with two different copies of an allele at the sex-determining locus. Most male bees are haploid, and they are produced from unfertilized eggs (with only a single copy of an allele at the sex-determining locus). However, when closely related bumble bees mate, the offspring can have two copies of the exact same allele (or be homozygous) at the sex-determining locus, which causes a diploid male to be produced instead of a diploid female. These diploid males may have reduced viability or may be sterile (van Wilgenburg 2006). When diploid males are able to mate, they produce sterile triploid offspring, which has been found to be negatively correlated with surrogates of bumble bee population size (Darvill *et al.* 2012). Diploid males are produced at the expense of female workers and new queens, and the production of diploid males can reduce colony fitness (including slower growth rates, lower survival, and colonies that produce fewer offspring) in bumble bees (Whitehorn *et al.* 2009). It has been suggested that diploid male production in inbred populations substantially increases the risk of extinction in bumble bee populations compared to other animal taxa (Zayed & Packer 2005).

Inbreeding and loss of genetic diversity can increase parasite prevalence in populations and parasite susceptibility in individuals (Frankham *et al.* 2010 *in* Whitehorn *et al.* 2010). Populations of bumble bees with low genetic diversity have been found to have a higher prevalence of pathogens (Whitehorn *et al.* 2010; Cameron *et al.* 2011a), suggesting that as populations lose genetic diversity, the impact of parasitism will increase and threatened populations will become more prone to extinction.

3. Global Climate Change

Climate change may pose a significant threat to the continued survival of the rusty patched bumble bee. Changes to the climate that are expected to have the most significant effects on bumble bee populations include: increased temperature and precipitation, increased drought, increased variability in temperature and precipitation extremes, early snow melt, and late frost events. These changes may lead to increased pathogen pressure, decreased resource availability (both floral resources and hibernacula), and a decrease in nesting habitat availability due to changes in rodent abundance or distribution (Cameron *et al.* 2011b).

Variability in climate can lead to phenological asynchrony between bumble bees and the plants they use (Memmott *et al.* 2007; Thomson 2010). There is evidence of mismatch between early blooming plants and their bumble bee pollinators (Kudo *et al.* 2004). Early spring is a critical time for bumble bees since that is the time when the foundresses emerge from hibernation and initiate nests. After the fourth-warmest winter on record for the U.S. (Dolce 2012), a rusty patched bumble bee queen emerged from hibernation in Wisconsin in March of 2012 (S. Carpenter pers. comm. with Xerces Society staff, 2012). Prior to this observation, the earliest recorded queens of this species from any region were recorded as emerging in April. Since bumble bees are generalist foragers, they do not require synchrony with a specific plant, but asynchrony could lead to diminished resource availability at times that are critical to bumble bee colony success. For example, as the climate in the Rocky Mountains has become warmer and drier in the past 30 years, researchers have observed a mid-season period of low floral resources, a change which can negatively impact pollinators (Aldridge *et al.* 2011). Furthermore, changes in the distributions of plants visited by bumble bees have been correlated with a changing climate (Forrest *et al.* 2010; Inouye 2008).

J. Kerr (unpublished data) found that interannual climatic variability has increased by a large margin during the period which the rusty patched bumble bee disappeared from much of its historic range. This climatic variability has been most extreme in the areas where the species' populations have apparently been extirpated, and less extreme where the rusty patched bumble bee currently persists.

In modeling studies, Kirilenko and Hanley (2007a, 2007b) predict that the ranges of three bumble bee species will change in size and shift in response to predicted changes in the North American climate. Although the rusty patched bumble bee was not one of the species included in this study, the impact of climatic changes on already vulnerable populations of the rusty patched bumble bee could be potentially severe.

Climate change can also affect the quality of nectar produced by flowers. Pumpkin flowers grown under experimental conditions mimicking predicted climate futures were altered in attractiveness and nutritional quality (Hoover *et al*. 2012). Bumble bees foraging on these plants suffered a 22% reduction in survival. Although this study was based on predicted future conditions, similar effects may be occurring presently at levels that are undetected but may still affect bumble bee populations.

4. Competition with Commercial Honey Bees

Honey bees (*Apis mellifera*) were introduced to eastern North America in the early 1620s. They compete with bumble bees for floral resources. The competitive effects on bumble bees that have been observed include: lower reproductive success, smaller body size, and changes in bumble bee foraging behavior (notably a reduction in pollen gathering) (Evans 2001; Thomson 2004, 2006; Walther-Hellwig *et al.* 2006; Goulson & Sparrow 2009). Honey bee presence reduces the availability of nectar and pollen (Paton 1990, 1996; Wills *et al.* 1990; Horskins & Turner 1999; Anderson 1989; Dafni & Shmida 1996; Dupont *et al.* 2005), and they displace some species of bumble bees when they are foraging in the same area (Walther-Hellwig *et al.* 2006). High density placement of honey bee hives near imperiled populations of the rusty patched bumble bee could threaten the continued survival of this species.

V. TAXONOMIC STATUS

All bumble bees belong to the genus *Bombus* within the family Apidae. The rusty patched bumble bee (*Bombus affinis*) belongs to the subgenus *Bombus* sensu stricto. *Bombus* sensu stricto is well supported as a distinct subgenus (Williams *et. al* 2008). *Bombus affinis* Cresson was first described by Cresson (1863). Its status as a species was upheld by Williams (1998) and more recently by Cameron *et al.* (2007) and Williams *et al.* (2012).

VI. SPECIES DESCRIPTION

A. Queens and Workers

Rusty patched bumble bee queens and workers differ slightly in coloration (an uncommon feature in bumble bees), the primary difference being size and a medial rusty patch present on the second tergal segment on the worker. Queens are 21 to 22 mm in length, 9.5 to 11 mm in breadth (Mitchell 1962). Workers are 11 to 16 mm in length, 5 to 9 mm in breadth (Mitchell 1962). Their hair is entirely black on the head, the bottom of the thorax, and in large part on the legs. The rest of the thorax has mostly yellow hair, with a black area in the middle of the thorax. Their hair is entirely yellow on the first two tergal segments and black on the rest of the abdomen. On workers, there is more black intermixed with yellow near the base of the wings forming somewhat of an interalar band and with black hairs extending rearward in a narrow V that partially bisects the yellow on the scutellum. The second tergal segment has a rusty reddish patch, which is usually located centrally, with yellow hairs around the edges of the segment. See Figure 2 for illustrations of rusty patched bumble bee queens and workers.

B. Males

Rusty patched bumble bee males are 13 to 17.5 mm in length with a breadth of 5 to 7 mm (Mitchell 1962). Their hair is largely black on the head, but with a few pale hairs intermixed near the top of the head. Black hairs sometimes form an obscure band across the middle of the thorax, otherwise the hair on the thorax is largely pale yellowish. The first two tergal segments have pale yellow hair. Like workers, males usually have a reddish patch centrally and anteriorly located on the second tergal segment of the abdomen. The hair on the rest of the abdomen is black. See Figure 2 for an illustration of a rusty patched bumble bee male.

Figure 2. Illustrations of a rusty patched bumble bee queen (left), worker (center), and male (right) by Elaine Evans, The Xerces Society.

VII. BIOLOGY, HABITAT REQUIREMENTS AND POLLINATION ECOLOGY

A. Biology and Habitat Requirements

The rusty patched bumble bee, like other bumble bees, lives in colonies consisting of a queen (foundress) and her offspring, the workers and near the end of the season the reproductive members of the colony, the males and new queens. There is a division of labor among these three types of bees. The foundress is responsible for initiating colonies and laying eggs. Workers are responsible for most food collection, colony defense, and feeding of the young. Males' leave the nest once they reach maturity and their sole function is to mate with queens. New queens remain with the nest until the end of the season when they leave to mate and find a hibernacula.

Colonies are annual, progressing from colony initiation by solitary queens in spring, to production of workers, and finally to production of queens and males. Rusty patched bumble bee queens are one of the earliest species to emerge, with observations as early as March and April (Plath 1922; Mitchell 1962; Milliron 1971; Colla and Dumesh 2010; S. Carpenter pers. comm. with Xerces Society staff, 2012). The foundress begins searching for suitable nesting sites and collects nectar and pollen from flowers to support the production of her eggs, which are fertilized by sperm she has stored since mating the previous fall. In the early stages of colony development, the queen is responsible for all food collection and care of the young. As the colony grows, workers take over the duties of food collection, colony defense, and care of the young. The foundress then remains within the nest and spends most of her time laying eggs. Colonies of the rusty patched bumble bee are considered large compared to other species of bumble bees, producing up to 1,000 workers throughout the season (Macfarlane *et al.* 1994). New queens and males are produced during the later stages of colony development, which is generally from mid-July or August to September (Plath 1922; Milliron 1971; Macfarlane *et al.* 1994). This species may be particularly susceptible to stressors because it emerges so early, but does not produce the next generation until late in the summer. The new queens mate before entering diapause, which is a form of hibernation. At the end of the season, the foundress dies.

Occasionally nests of the rusty patched bumble bee have been observed above ground. However, nests are usually one to four feet below ground in abandoned rodent nests or other cavities (Plath 1922; Macfarlane *et al.* 1994). Thus, nesting sites may be limited by the abundance of rodents. This species has been observed or collected from woodlands, marshes, agricultural landscapes, and, more recently from residential parks and gardens (Colla & Packer 2008; Colla & Dumesh 2010; Xerces Society 2012).

Although little is known about the overwintering habits of rusty patched bumble bee queens, queens of other species frequently dig a few centimeters into soft, disturbed soil and form an oval shaped chamber in which she will spend the duration of the winter. Compost in gardens or mole hills may provide suitable sites for queens to overwinter (Goulson 2010).

Bumble bees are particularly vulnerable to extinction due to their complementary sex determination system and haplodiploid life history (Zayed & Packer 2005), described above in section *IV. CURRENT AND POTENTIAL THREATS – SUMMARY OF FACTORS FOR CONSIDERATION E. Other Natural or Manmade Factors Affecting its Continued Existence; 2. Population Dynamics and Structure; b. Impacts of Genetic Factors on Bumble Bees*.

B. Bumble Bee Pollination Ecology

Bumble bees are generalist foragers, meaning that they gather pollen and nectar from a wide variety of flowering plants. To meet its nutritional needs, the rusty patched bumble bee requires a constant supply of flowers that bloom throughout the duration of the colony life cycle, which is from approximately April to September (Plath 1922; Mitchell 1962; Milliron 1971; Macfarlane *et al.* 1994). Nectar provides bumble bees with carbohydrates and pollen provides them with protein. The amount of pollen available to bumble bee colonies directly affects the number of queens that can be produced (Burns 2004). Since queens are the only bumble bees capable of forming new colonies, pollen availability directly impacts future bumble bee population levels.

The rusty patched bumble bee probably needs floral resources to be located in relative close proximity to its nest sites, as studies of other bumble bee species indicate that they routinely forage within less than one kilometer from their nests (Knight *et al*. 2005; Wolf & Moritz 2008; Dramstad 1996; Osborne *et al*. 1999), although in some cases nearly two kilometers (Walther-Hellwig & Frankl 2000). Colla and Dumesh (2010) suggest that the rusty patched bumble bee is likely dependent upon woodland spring ephemeral flowers, since this bumble bee emerges early in the year and is associated with woodland habitats.

The rusty patched bumble bee is a short-tongued species (Medler 1962) and thus is not able to easily access the nectar in flowers with deep corollas. Short-tongued bees are better suited for pollination of open flowers and those with short corollas, including cranberry (Patten *et al.* 1993).

During collection of pollen and nectar from flowers, bumble bees also transport pollen between flowers, facilitating seed and fruit production. Bumble bees have many qualities that contribute to their suitability as agricultural pollinators. They are able to fly in cooler temperatures and lower light levels than many other bees, which extends their work day and improves the pollination of crops during inclement weather (Corbet *et al.* 1993). They also possess the ability to "buzz pollinate," in which a bee grabs the pollen producing structure of the flower in her jaws and vibrates her wing musculature. This activity causes the flower to vibrate, which in turn dislodges pollen that would have otherwise remained trapped in the flower's anthers (Buchmann 1983). Some plants, including tomatoes and peppers, benefit from buzz pollination. The rusty patched bumble bee has been shown to be an excellent pollinator of cranberry (Cane & Schiffauer 2003) and other important food crops such as plum and apple (Medler & Carney 1963; Mitchell 1962), alfalfa (Holm 1966), and onion for seed production (Caron *et al.* 1975).

In addition to commercially important crops, the rusty patched bumble bee also plays a vital role as a generalist pollinator of native flowering plants, and its loss may have far ranging ecological impacts. An examination of the theoretical effect of removing specialist and generalist pollinators on the extinction of plant species concluded that the loss of generalist pollinators, especially bumble bees, caused the greatest number of plant extinctions (Memmott *et al.* 2004). In Britain and the Netherlands, where multiple pollinators have declined, there is evidence of a parallel decline in the abundance of insect pollinated plants (Biesmeijer *et al.* 2006).

Bombus affinis visits a wide variety of wild plants including: *Abelia grandiflora* (Speight 1967), *Aesculus* spp. (Dieringer 1982; Macfarlane 1974), *Agastache foeniculum* (C. Reed, pers. comm. with E. Evans, July 2008), *Amorpha canadense* (C. Reed, pers. comm. with E. Evans, July 2008), *Asclepias syriaca, A. incarnata, A. verticillata* (Frost 1965; Macior 1965), *Aralia* spp. (Mitchell 1962), *Aster* spp. (Costelloe 1988), *Aquilegia canadensis* (Macior 1978a), *Aureolaria pedicularia* (Stiles 1977), *Berberis* spp. (Macior 1965), *Camassia scilloides* (Macior 1978b), *Carduus* sp. (Macior 1965), *Ceanothus americanus* (Bequaert 1920), *Cercis canadensis* (Fye & Medler 1954), *Chamaedaphne calyculata* (Judd 1966), *Coreopsis major* (Speight 1967), *Crataegus* spp. (Macior 1968), *Dalea purpurea* (C. Reed, pers. comm. with E. Evans, July

2008), *Delphinium tricorne* (Macior 1975), *Dicentra canadensis*, *D. cucullaria* (Macior 1978b), *Echium vulgare* (Macfarlane 1974), *Helianthus* spp. (Fye & Medler 1954; Colla & Packer 2008), *Hydrangea* spp. (Mitchell 1962), *Hydrophyllum* spp. (Macior 1978b, Macfarlane 1974), *Impatiens capensis* (R. Gegear, pers. comm. with E. Evans, May 2008), *Lamium purpureum* (Macior 1978a), *Laportea* spp. (Speight 1967), *Leonurus* sp. (Macior 1965), *Linaria* sp. (Macior 1965), *Lonicera* spp. (Macior 1968), *Lotus corniculatus* (Fye & Medler 1954), *Medicago sativa* (Fye & Medler 1954), *Mertensia virginica* (Macior 1978b), *Monarda* sp. (Macior 1965), *Nepeta* spp. (Macior 1965), *Pedicularis canadensis* (Macior 1978b; Dieringer 1982), *Pedicularis lanceolata* (Costelloe 1988; Macior 1969), *Penstemon grandiflorus* (C. Reed, pers. comm. with E. Evans, July 2008), *Philadelphus* spp. (Speight 1967), *Polymnia* spp. (Speight 1967), *Prunella vulgaris* (Speight 1967), *Prunus* spp. (Fye & Medler 1954), *Pyrus ioensis* (Macior 1968), *Pyrus malus* (Macior 1968), *Ratibida pinnata* (C. Reed, pers. comm. with E. Evans, July 2008), *Rhododendron* spp. (Macfarlane 1974), *Rhus* spp. (Speight 1967), *Ribes* spp. (Macfarlane 1974) *Robinia* spp. (Mitchell 1962), *Rosa* spp. (Macior 1965), *Rubus* spp. (Macfarlane 1974), *Salix* spp. (Medler & Carney 1963), *Sarracenia purpurea* (Ne'eman *et al.* 2006), *Solanum* sp. (Macior 1965), *Solidago* spp. (Mitchell 1962), *Symphytum officinale* (Macfarlane 1974), *Syringia* spp. (Macior 1968), *Syringia vulgaris* (Fye & Medler 1954), *Taraxacum* spp. (Macior 1968), *Trifolium* spp. (Fye & Medler 1954; Macfarlane 1974), *Vaccinium* spp. (Mitchell 1962), *Verbascum* spp. (Macior 1965), *Verbesina occidentalis* (Speight 1967), *Vicia* spp. (Fye & Medler 1954; Macfarlane 1974).

VIII. CONCLUSION

Bumble bees are essential pollinators of crops and wildflowers in agricultural, urban and natural ecosystems. They play an important role in the reproduction of tomato, blueberry, pepper, cranberry, clover, and many other crops. Although the rusty patched bumble bee was historically distributed throughout the Upper Midwest, Northeast and eastern seaboard, recent range-wide studies have estimated that *Bombus affinis* is no longer found in 70-87% of its historic range. Where it does still occur, its relative abundance has declined by 87-95% (Colla *et al.* 2012; Cameron *et al.* 2011a). Declines in North American bumble bees have been associated with increased levels of the pathogen *Nosema bombi* and reduced genetic diversity (Cameron *et al.* 2011a). Habitat loss or degradation, other pathogens, pesticides, climate change, and competition with honey bees also threaten this species with rangewide extinction. When considered individually, each of these factors pose a significant potential threat to the rusty patched bumble bee. However, when considered together, they present a daunting case for the recovery of this animal. In addition, existing regulations are inadequate to protect the rusty patched bumble bee from disease and pesticides, and to protect its habitat. The rusty patched bumble bee should be listed as an Endangered Species under the U.S. Endangered Species Act to prevent global extinction of this once common bumble bee.

IX. REFERENCES CITED

Al-Jabr AM. 1999. Integrated pest management of tomato/potato psyllid, *Paratrioza cockerelli* (Sulc) (Homoptera: Psyllidae), with emphasis on its importance in greenhouse grown tomatoes. PhD dissertation, Deparment of Bioagricultural Sciences and Pest

Management, Colorado State University. Available at: http://www.colostate.edu/Depts/Entomology/theses/sljabr/thesis.html
Aldridge G, DW Inouye, JRK Forrest, WA Barr, and AJ Miller-Rushing. 2011. Emergence of a mid-season period of low floral resources in a montane meadow ecosystem associated with climate change. Journal of Ecology 99(4):905-913.
Altizer S, D Harvell, and E Friedle. 2003. Rapid evolutionary dynamics and disease threats to biodiversity. TRENDS in Ecology and Evolution 18(11):589-596.
Anderson GJ and MK Anderson. 1989. Assaying pollinator visitation to *Solanum* flowers. Solanaceae newsletter 3(1):71.
Arbetman MP, I Meeus, CL Morales, MA Aizen, and G Smagghe. 2012. Alien parasite hitchhikes to Patagonia on invasive bumblebee. Biological Invasions.
Babendreier D, B Reichhart, J Romeis, and F Bigler. 2008. Impact of insecticidal proteins expressed in transgenic plants on bumblebee microcolonies. Entomologia Experimentalis et Applicata 126:148-157.
Bequaert J. 1920. Hymenoptera collected near Boston, Mass., with description of a variety of *Bombus affinis*. Psyche 27:6-12.
Bhattacharya M, RB Primack, and J Gerwein. 2003. Are roads and railroads barriers to bumblebee movement in a temperate suburban conservation area? Biological Conservation 109:37-45.
Biesmeijer JC, SPM Roberts, M Reemer, R Ohlemüller, M Edwards, T Peeters, AP Schaffers, SG Potts, R Kleukers, CD Thomas, J Settele, and WE Kunin. 2006. Parallel declines in pollinators and insect-pollinated plants in Britain and the Netherlands. Science 313:351-354.
Buchmann SL. 1983. Buzz pollination in angiosperms, p. 73-113. In C. E. Jones and R. J. Little, (eds.), Handbook of Experimental Pollination Biology, Van Nostrand Reinhold, New York, NY.
Bueno C, KE Ruckstuh, N arrigo, AN Aivaz, and P Neuhaus. 2011. Impacts of cattle grazing on small-rodent communities: an experimental case study. Canadian Journal of Zoology 90:22-30.
Burns I. 2004. Social development and conflict in the North American bumblebee *Bombus impatiens* Cresson. U of MN. Ph.D. Thesis.
Brown MJF, R Schmid-Hempel, and P Schmid-Hempel. 2003. Strong context-dependent virulence in a host-parasite system: reconciling genetic evidence with theory. Journal of Animal Ecology 72:994-1002.
Calvin L and R Cook. 2005. North American greenhouse tomatoes emerge as a major market force. Amber Waves: US Department of Agriculture Economic Research Service. 8 pages.
Cameron SA, HM Hines, and PH Williams. 2007. A comprehensive phylogeny of the bumble bees (*Bombus*). Biological Journal of the Linnean Society 91:161-188.
Cameron S, Lozier JD, Strange JP, Koch JB, Cordes N, Solter LF, Griswold TL. 2011. Patterns of widespread decline in North American bumble bees. Proceedings of the National Academy of the United States of America 108:662-667.
Cameron S, S Jepsen, E Spevak, J Strange, M Vaughan, J Engler, and O Byers (eds). 2011b. North American Bumble Bee Species Conservation Planning Workshop Final Report. IUCN/SSC Conservation Breeding Specialist Group: Apple Valley, MN. Available

online at:
http://www.cbsg.org/cbsg/workshopreports/26/bumble_bee_conservation_2010.pdf
Cane JH and D Schiffhauer. 2003. Dose-response relationships between pollination and fruiting refine pollinator comparisons for cranberry (*Vaccinium macrocarpon*). American Journal of Botany 90:1425-1432.
Caron DM, RC Lederhouse, and RA Morse. 1975. Insect pollinators of onion in New York State. HortScience 10:273-274.
Carvell C. 2002. Habitat use and conservation of bumblebees (*Bombus* spp.) under different grassland management regimes. Biological Conservation 103:33-49.
Carvell C, DB Roy, SM Smart, RF Pywell, CD Preston, and D Goulson. 2006. Declines in forage availability for bumblebees at a national scale. Biological Conservation 132:481-489.
Colla SR. 2012. The Ecology and Conservation of North American Bumblebees, Ph.D. Thesis York University.
Colla SR, MC Otterstatter, RJ Gegear, and JD Thomson. 2006. Plight of the bumblebee: pathogen spillover from commercial to wild populations. Biological Conservation 129:461-467.
Colla SR and L Packer. 2008. Evidence for the decline of Eastern North American Bumble Bees, with special focus on *Bombus affinis* Cresson. Biodiversity and Conservation 17:1379-1391.
Colla SR and S Dumesh. 2010. The bumble bees of southern Ontario: Notes on natural history and distribution. Journal of the Ecological Society of Southern Ontario, 141:39-68.
Colla SR, F Gadallah, L Richardson, D Wagner and L Gall. 2012. Assessing declines of North American bumble bees (*Bombus* spp.) using museum specimens. Biodiversity and Conservation DOI 10.1007/s10531-012-0383-2.
Colla S, LL Richardson, RW Thorp and PH Williams. *In prep*. Guide to the Bumble Bees of North America. Princeton University Press.
Connecticut Department of Environmental Protection, http://www.ct.gov/dep/cwp/view.asp?A=2702&Q=323478, accessed 17 June 2011.
Corbet SA, M Fussell, R Ake, A Fraser, C Gunson, A Savage, and K Smith. 1993. Temperature and the pollinating activity of social bees. Ecological Entomology 18:17-30.
Cordes N, WF Huang, JP Strange, SA Cameron, TL Griswold, JD Lozier, and LF Solter. 2012. Interspecific geographic distribution and variation of the pathogens *Nosema bombi* and *Crithidia* species in United States bumble bee populations. Journal of Invertebrate Pathology 109:209-216.
Costelloe BH 1988. Pollination ecology of *Gentiana andrewsii*. Ohio Journal of Science 88:132-138.
Cox C. 2001. Insecticide factsheet: Imidacloprid. Journal of Pesticide Reform. 21:15-22.
Cresson, ET. 1863. List of the North American species of *Bombus* and *Apathus*. Proceedings of the Entomological Society of Philadelphia 2:83-116.
Dafni A and A Shmida. 1996. The possible ecological implications of the invasion of *Bombus terrestris* (L.) (Apidae) at Mt. Carmel, Israel. In A Matheson, SL Buchmann, C O'Toole, P Westrich, and IH Williams (eds.). The Conservation of Bees. Academic Press pp. 183-200.

Darvill B, JS Ellis, GC Lye, and D Goulson. 2006. Population structure and inbreeding in a rare and declining bumblebee, *Bombus muscorum* (Hymenoptera: Apidae). Molecular Ecology 15:601-611.

Darvill B, O Lepais, LC Woodall, and D Goulson. 2012. Triploid bumblebees indicate a direct cost of inbreeding in fragmented populations. Molecular Ecology 21, DOI: 10.1111/j.1365-294X.2012.05679.x

Diekotter T, K Walther-Hellwig, M Conradi, M Suter, and R Frankl. 2006. Effects of landscape elements on the distribution of the rare bumblebee species *Bombus muscorum* in an agricultural landscape. Biodiversity and Conservation 15:57-68.

Dieringer G. 1982. The pollination ecology of *Orchis spectabilis* L. (Orchidaceae). Ohio Academy of Science 82:218-225.

Dolce C. 2012. NOAA: Fourth warmest winter on record. Weather.com. Online: http://www.weather.com/outlook/weather-news/news/articles/looking-back-winter-2011-2012_2012-03-07, last accessed 14 January 2013.

Dramstad WE. 1996. Do bumblebees (Hymenoptera: Apidae) really forage close to their nests? J. Insect. Behav. 9:163-182.

Dupont YL, C Damgaard, and V Simonsen. 2011. Quantitative Historical Change in Bumblebee (*Bombus* spp.) Assemblages of Red Clover Fields. PLoS ONE 6(9): e25172. doi:10.1371/journal.pone.0025172

Eilers EJ, C Kremen, SS Greenleaf, AK Garder, and A Klein. 2011. Contribution of pollinator-mediated crops to nutrients in the human food supply. PLoS ONE 6(6):e21363.

Ellis, JS, ME Knight, B Darvill, and D Goulson. 2006. Extremely low effective population sizes, genetic structuring and reduced genetic diversity in a threatened bumblebee species, *Bombus sylvarum* (Hymenoptera: Apidae). Molecular Ecology 15:4375-4386.

Evans, EC. 2001. Competition between European honey bees and native bumblebees: resource overlap and impact on reproductive success. Master's Thesis, University of Minnesota.

Evans, E, Thorp, RW, Jepsen, S, and Black, SH. 2008. Status review of three formerly common species of bumble bee in the Subgenus *Bombus*. Tech. rep., The Xerces Society. http://www.xerces.org/wp-content/uploads/2009/03/xerces_2008_bombus_status_review.pdf

Fischer D and T Moriarty (Eds). 2011. Pesticide risk assessment for pollinators: Summary of a SETAC Pellston workshop. 15-21 January 2011, Pensacola, Florida, USA.

Fitzpatrick, U, TE Murray, RJ Paxton, J Breen, D Cotton, V Santorum, and MJF Brown. 2007. Rarity and decline in bumblebees: a test of causes and correlates in the Irish fauna. Biological Conservation 136:185-194.

Flanders RV, WF Wehling, and AL Craghead. 2003. Laws and regulations on the import, movement and release of bees in the United States. pp. 99-111. In K. Strickler and J. H. Cane (eds.) For Nonnative Crops, Whence Pollinators of the Future? Thomas Say Publications in Entomology: Proceedings. Entomological Society of America, Lanham, MD.

Forrest J, Inouye DW, Thomson JD. 2010. Flowering phenology in subalpine meadows: Does climate variation influence community co-flowering patterns? Ecology 91:431–440.

Frankie, GW, RW Thorp, M Schindler, J Hernandez, B Ertter, and M Rizzardi. 2005. Ecological patterns of bees and their host ornamental flowers in two northern California cities. Journal of the Kansas Entomological Society 78:227-246.

Frankham R, JD Ballou, and DA Briscoe. 2010. Introduction to Conservation Genetics Second Edition, Cambridge University Press, 618 pages.
Franzen, P. 1993 (unpublished). Field study of native bees (bumblebees) *Bombus* species. Fermilab, Batavia, Illinois.
Frost, SW. 1965. Insects and pollinia. Ecology 46:556-558.
Fye, RE and JT Medler. 1954. Spring emergence and floral hosts of Wisconsin bumblebees. Transactions of the Wisconsin Academy of Science, Arts and Letters 43:75-82.
Genersch, E, C Yue, I Fries, and JR de Miranda. 2006. Detection of Deformed wing virus, a honey bee viral pathogen, in bumble bees (*Bombus terrestris* and *Bombus pascuorum*) with wing deformities. Journal of Invertebrate Pathology 91:61-63.
Gibson, C, V Brown, L Losito, and G McGavin. 1992. The response of invertebrate assemblies to grazing. Ecography 15:166-176.
Giles V and JS Ascher. 2006. A Survey of Bees of the Black Rock Forest Preserve, New York (Hymenoptera: Apoidea). Journal of Hymenoptera Research 15:208-231.
Gill RJ, O Ramos-Rodriguez, and NE Raine. 2012. Combined pesticide exposure severely affects individual- and colony-level traits in bees. Nature, DOI:10.1038/nature11585
Gillespie S. 2010. Factors affecting parasite prevalence among wild bumblebees. Ecological Entomology, DOI: 10.1111/j.1365-2311.2010.01234.x
Goka K, Okabe K, Niwa S, Yoneda M. 2000. Parasitic mite infestation in introduced colonies of European bumblebees, *Bombus terrestris*. Japanese Journal of Applied Entomology and Zoology 44(1):47-50.
Goka K, K Okabe, M Yoneda, and S Niwa. 2001. Bumblebee commercialization has caused international migration of parasitic mites. Molecular Ecology 10:2095-2099.
Goka K, Okabe K, Yoneda M. 2006. Worldwide migration of parasitic mites as a result of bumblebee commercialization. Population Ecology 48:285–291.
Goulson D. 2003. Bumblebees, Their Behaviour and Ecology. Oxford University Press, Oxford.
Goulson D. 2010. Bumblebees: Behaviour, ecology and conservation. Second edition. Oxford University Press. 317 pages.
Goulson D, GC Lye, and B Darvill. 2008. Decline and conservation of bumble bees. Annual Review of Entomology 53:191-208.
Goulson D and K Sparrow. 2009. Evidence for competition between honeybees and bumblebees; effects on bumblebee worker size. Journal of Insect Conservation 13(2):177-181.
Goulson D, O Lepais, S O'Connor, JL Osborne, RA Sanderson, J Cussans, L Goffe, and B Darvill. 2010. Effects of land use at a landscape scale on bumblebee nest density and survival. Journal of Applied Ecology 6:1207–1215.
Gradish AE, CD Scott-Dupree, L Shipp, CR Harris, and G Ferguson. 2009. Effect of reduced risk pesticides for use in greenhouse vegetable production on *Bombus impatiens* (Hymenoptera: Apidae). Pest Management Science 66:142-146. DOI 10.1002/ps.1846
Grixti JC, LT Wong, S Cameron and C Favret. 2008. Decline of bumble bees (*Bombus*) in the North American Midwest. Biological Conservation, DOI:10.1016/j.biocon.2008.09.027
Hatfield RG and G LeBuhn. 2007. Patch and landscape factors shape community assemblage of bumble bees, *Bombus* spp. (Hymenoptera: Apidae), in montane meadows. Biological Conservation 139:150-158.
Hatfield R, S Jepsen, E Mader, SH Black and M Shepherd. 2012. Conserving Bumble Bees: Guidelines for Creating and Managing Habitat for America's Declining Pollinators. 32

pp. Portland, OR: The Xerces Society for Invertebrate Conservation. http://xerces.org/bumblebees/guidelines

Highfield AC, A El Nagar, LCM Mackinder, L Noel, MJ Hall, SJ Martin, et al. 2009. Deformed wing virus implicated in overwintering honeybee colony losses. Appl Environ Microbiol 75:7212–7220.

Hines HM and SD Hendrix. 2005. Bumble bee (Hymenoptera: Apidae) diversity and abundance in tallgrass prairie patches: effects of local and landscape floral resources. Environmental Entomology 34:1477-1484.

Holm SN. 1966. The utilization and management of bumble bees for red clover and alfalfa seed production. Annual Review of Entomology 11:155-182.

Hoover SER, JJ Ladley, AA Shchepetkina, M Tisch, SP Gieseg & JM Tylianakis. 2012. Warming, CO2, and nitrogen deposition interactively affect a plant-pollinator mutualism. Ecology Letters 15(3)227-234.DOI:10.1111/j.1461-0248.2011.01729.x

Hopwood J, M Vaughan, M Shepherd, E Mader, and SH Black. 2012. Are Neocicotinoids Killing Bees? A review of research into the effects of neonicotinoid insecticides on bees, with recommendation for action. Tech. rep., The Xerces Society. http://www.xerces.org/neonicotinoids-and-bees/

Horskins K and VB Turner. 1999. Resource use and foraging patterns of honeybees, *Apis mellifera*, and native insects on flowers of *Eucalyptus costata*. Aust. J. Ecol. 24:221–27

Huntizinger M. 2003. Effects of fire management practices on butterfly diversity in the forested western United States. Biological Conservation 113:1-12. DOI:10.1016/S0006-3207(02)00356-7

Husband RW and RN Shina. 1970. A review of the genus *Locustacarus* with a key to the genera of the family Podapolipidae (Acarina), Ann. Entomol. Soc. Am. 63:1152–1162.

Inouye, DW. 1980. The effects of proboscis and corolla tube lengths on patterns and rates of flower visitation by bumblebees. Oecologia 45:197-201.

IUCN. 2001. IUCN Red List Categories and Criteria: Version 3.1. IUCN Species Survival Commission. IUCN, Gland, Switzerland and Cambridge, UK. ii + 30 pp.

Incerti F, L Bortolotti, C Porrini, A Micciarelli Sbrenna, and G Sbrenna. 2003. An extended laboratory test to evaluate the effects of pesticides on bumblebees, preliminary results. Bulletin of Insectology 56:159-164.

Inouye DW. 2008. Effects of climate change on phenology, frost damage, and floral abundance of montane wildflowers. Ecology 89(2):353-362.

Irwin RE and AL Brody. 1999. Nectar-robbing bumble bees reduce the fitness of *Ipomopsis aggregata* (Polemoniaceae). Ecology 80:1703-1712.

Jha S. 2012. Ecological Society of America presentation. Abstract available at: http://eco.confex.com/eco/2012/webprogram/Paper33081.html

Jha S & C Kremen. 2012. Resource diversity and landscape-level homogeneity drive native bee foraging. PNAS. www.pnas.org/cgi/doi/10.1073/pnas.1208682110

Johansen CA. 1977. Pesticides and pollinators. Annual Review of Entomology 22:177-192.

Johansen CA and DF Mayer. 1990. Pollinator Protection: a Bee and Pesticide Handbook. Wicwas Press, Cheshire, Connecticut.

Kearns CA and DW Inouye. 1997. Pollinators, flowering plants, and conservation biology. BioScience 47:297-307.

Kearns CA, DW Inouye, and N Waser. 1998. Endangered mutualisms: the conservation of plant–pollinator interactions. Annual Review of Ecology and Systematics 29:83-112.

Kevan PG. 1999. Pollinators as bioindicators of the state of the environment: species, activity and diversity. Agriculture Ecosystems and Environment 74:373-393.
Kevan PG. 1975. Forest application of the insecticide Fenitrothion and its effect on wild bee pollinators (Hymenoptera: Apoidea) of lowbush blueberries (Vaccinium spp.) in southern New Brunswick, Canada. Biological Conservation 7:301-309.
Kevan PG and RC Plowright. 1995. Impact of pesticides on forest pollination. pp. 607-618 In J. A. Armstrong and W. G. H. Ives (eds.) Forest Insect Pests in Canada. Natural Resources Canada, Canadian Forest Service, Science and Sustainable Development Directorate, Ottawa, Ontario, Canada.
Kimoto C. 2010. Effect of livestock grazing on native bees in a Pacific Northwest bunchgrass prairie. MS Thesis, Oregon State University.
Kirilenko A and RS Hanley. 2007a. Predicting climate change impact on insect biodiversity: comparison of scenarios. pp. 453-457 In Proceedings of the 32nd International Symposium on Remote Sensing of Environment, San Jose, Costa Rica (June 25-29, 2007).
Kirilenko A and RS Hanley. 2007b. Using multiple methods to predict climate change impacts on bumblebees in North America. pp. 42-47, In Proceedings of the International Conference on Environmental Modeling and Simulation.
Kissinger CN, SA Cameron, RW Thorp, B White, and LF Solter. 2011. Survey of bumble bee (*Bombus*) pathogens and parasites in Illinois and selected areas of northern California and southern Oregon. Journal of Invertebrate Pathology 107:220-224.
Klein A, B Vaissiere, J Cane, I Steffan-Dewenter, S Cunningham, C Kremen, et al. 2007. Importance of pollinators in changing landscapes for world crops. Proceedings of the Royal Society B: Biological Sciences, 274:303-313.
Knight ME, Martin AP, Bishop S, Osborne JL, Hale RJ, Sanderson A, Goulson D (2005) An interspecific comparison of foraging range and nest density of four bumblebee (*Bombus*) species. Molecular Ecology 14:1811–1820.
Koppert, http://www.koppert.com/pollination/vegetable-crops/, http://www.koppert.com/pollination/fruit-crops/, accessed July 2012.
Kosior A, W Celary, P Olejniczak, J Fijat, W Krol, W Solarz, and P Plonka. 2007. The decline of the bumble bees and cuckoo bees (Hymenoptera: Apidae: Bombini) of western and central Europe. Oryx 41:79-88.
Kruess A and T Tscharntke. 2002a. Contrasting responses of plant and insect diversity to variation in grazing intensity. Biological Conservation 106:293-302.
Kruess A and T Tscharntke. 2002b. Grazing intensity and the diversity of grasshoppers, butterflies, and trap-nesting bees and wasps. Conservation Biology 16:1570-80.
Kudo G, Y Nishikawa, T Kasagi & S Kosuge. 2004. Does seed production of spring ephemerals decrease when spring comes early? Ecological Research 19:255–259.
Larsson RJI. 2007. Cytological variation and pathogenicity of the bumble bee parasite *Nosema bombi* (Microspora, Nosematidae). Journal of Invertebrate Pathology 94:1-11.
Laycock I, KM Lenthall, AT Barratt, and JE Cresswell. 2012. Effects of imidacloprid, a neonicotinoid pesticide, on reproduction in worker bumble bees (*Bombus terrestris*). Ecotoxicology, DOI 10.1007/s10646-012-0927-y.
Leonard MD. 1928. A list of the insects of New York, with a list of the spiders and certain other allied groups. Cornell University Agricultural Experiment Station Memoir 101:5-1121.

Lozier JD and SA Cameron. 2009. Comparative genetic analyses of historical and contemporary collections highlight contrasting demographic histories for the bumble bees *Bombus pensylvanicus* and *B. impatiens* in Illinois. Molecular Ecology 18:1875-1886, DOI:10.1111/j.1365-294X.2009.04160.x

Lozier JD, JP Strange, IJ Stewart, and SA Cameron. 2011. Patterns of range-wide genetic variation in six North American bumble bee (Apidae: *Bombus*) species. Molecular Ecology 20:4870-4888, DOI: 10.1111/j.1365-294X.2011.05314.x

Macfarlane RP. 1974. Ecology of Bombinae (Hymenoptera: Apidae) of Southern Ontario, with emphasis on their natural enemies and relationships with flowers. PhD Thesis, University of Guelph, Guelph.

Macfarlane RP, KD Patten, LA Royce, BKW Wyatt, and DF Mayer. 1994. Management potential of sixteen North American bumble bee species. Melanderia 50:1-12.

Macfarlane RP, JJ Lipa, and HJ Liu. 1995. Bumble bee pathogens and internal enemies. Bee World 76:130-148.

Macior LW. 1965. Insect adaptation and behavior in *Asclepias* pollination. Bulletin of the Torrey Botanical Club 92:114-126.

Macior LW. 1968. *Bombus* (Hymenoptera, Apidae) queen foraging in relation to vernal pollination in Wisconsin. Ecology 49:20-25.

Macior LW. 1969. Pollination adaptation in *Pedicularis lanceolata*. American Journal of Botany 56:853-859.

Macior LW. 1975. The pollination ecology of *Delphinium tricorne* (Ranunculaceae). American Journal of Botany 62:1009-1016.

Macior LW. 1978a. Pollination interactions in sympatric *Dicentra* species. American Journal of Botany 65:57-62.

Macior LW. 1978b. Pollination ecology of vernal angiosperms. Oikos 30:452-460.

Malone LA and M Pham-Delègue. 2001. Effects of transgene products on honey bees (*Apis mellifera*) and bumblebees (*Bombus* sp.). Apidologie 32:287-304.

Maloof JE. 2001. The effects of a bumble bee nectar robber on plant reproductive success and pollinator behavior. American Journal of Botany 88:1960-1965.

Marletto F, A Patetta, and A Manino. 2003. Laboratory assessment of pesticide toxicity to bumblebees. Bulletin of Insectology 56:155-158.

McFrederick QS and G LeBuhn. 2006. Are urban parks refuges for bumble bees *Bombus* spp. (Hymenoptera : Apidae)? Biological Conservation 129:372-382.

Meeus I, JMF Brown, DC De Graaf, and G Smagghe. 2011. Effects of invasive parasites on bumble bee declines. Conservation Biology 25(4):662-671.

Medler JT. 1962. Morphometric studies on bumble bees. Annals of the Entomological Society of America 55:212-218.

Medler JT and DW Carney. 1963. Bumblebees of Wisconsin (Hymenoptera: Apidae). Research Bulletin, University of Wisconsin, Agricultural Experiment Station 240:47 pp.

Memmott J, NM Waser, and MV Price. 2004. Tolerance of pollination networks to species extinctions. Proceedings of the Royal Society B 271:2605-2611.

Memmott J, PG Craze, NM Waser, and MV Price. 2007. Global warming and the disruption of plant-pollinator interactions. Ecology Letters 10:710-717.

Michigan Department of Natural Resources. 2012. http://mnfi.anr.msu.edu/data/specialanimals.cfm/

Milliron HE. 1971. A monograph of the western hemisphere bumblebees (Hymenoptera: Apidae; Bombinae). I. The genera *Bombus* and *Megabombus* subgenus *Bombias*. Memoirs of the Entomological Society of Canada 82:1-80.

Mitchell TB. 1962. Bees of the Eastern United States. Vol. II. North Carolina Agricultural Experiment Station Technical Bulletin 152:1-557.

Mommaerts V, G Sterk, and G Smagghe. 2006. Hazards and uptake of chitin synthesis inhibitors in bumblebees *Bombus terrestris*. Pest Management Science 62:752-758.

Mommaerts, V, S Reynders, J Boulet, L Besard, G Sterk and G Smagghe. 2010. Risk assessment for side-effects of neonicotinoids against bumblebees with and without impairing foraging behavior. Ecotoxicology 19:207-215.

Mommaerts V and G Smagghe. 2011. Side-effects of pesticides on the pollinator *Bombus*: An overview. In Pesticides in the Modern World, M. Stoytcheva (Ed.), pp. 507-552.

Morandin LA, Laverty TM, Kevan PG, Khosla S, Shipp L. 2001. Bumble bee (Hymenoptera : Apidae) activity and loss in commercial tomato greenhouses. Canadian Entomology 133(6):883-93.

Morandin LA and ML Winston. 2003. Effects of novel pesticides on bumble bee (Hymenoptera: Apidae) colony health and foraging ability. Environmental Entomology 32(3):555-563.

Morandin LA, ML Winston, MT Franklin, and VA Abbott. 2005. Lethal and sub-lethal effects of spinosad on bumble bees (*Bombus impatiens* Cresson). Pest Management Science 61:619-626, DOI: 10.1002/ps.1058

Morkeski A and A Averill. February 20, 2012. Wild bee status and evidence for pathogen spillover with honey bees. CAP Updates: 12. Oregon State University Extension. Available online: http://www.extension.org/pages/30998/wild-bee-status-and-evidence-for-pathogen-spillover-with-honey-bees, Accessed January 2013.

Morris M. 1967. Differences between invertebrate faunas of grazed and ungrazed chalk grassland. I. Responses of some phytophagous insects to cessation of grazing. Journal of Applied Ecology 4:459-474.

National Research Council. 2007. Status of Pollinators in North America. The National Academies Press, Washington, DC.

NatureServe. 2012. NatureServe Explorer: An online encyclopedia of life [web application]. Version 7.1. NatureServe, Arlington, Virginia. Available http://www.natureserve.org/explorer. Accessed: November 16, 2012.

Niwa S, H Iwano, S Asada, M Matsuurra, and K Goka. 2004. A microsporidian pathogen isolated from a colony of the European bumblebee, *Bombus terrestris*, and infectivity on Japanese bumblebee. Japanese Journal of Applied Entomology and Zoology 48:60-64 (in Japanese with English abstract).

Noss RF, ET LaRoe, and JM Scott. 1995. Endangered ecosystems of the United States: A preliminary assessment of loss and degradation. Washington, D.C.: U.S. Dept. of the Interior, National Biological Service. 58 pages.

Öckinger E and HG Smith. 2007. Semi-natural grasslands as population sources for pollinating insects in agricultural landscapes. Journal of Applied Ecology 44:50-59.

Ollerton J, R Winfree, and S Tarrant. 2011. How many flowering plants are pollinated by animals. Oikos 120:321-326. DOI: 10.1111/j.1600-0706.2010.18644.x

Ontario Ministry of Natural Resources. Species at Risk in Ontario (SARO) list. Available at: http://www.mnr.gov.on.ca/en/Business/Species/2ColumnSubPage/276722.html (last accessed 20 December 2012).

Osborne JL, SJ Clark, RJ Morris, IH Williams, JR Riley, AD Smith, DR Reynolds, and AS Edwards. 1999. A landscape-scale study of bumble bee foraging range and constancy, using harmonic radar. J. Appl. Ecol. 36:519-533.
Osborne JL. 2012. Bumblebees and pesticides. Nature: News & Views, DOI:10.1038/nature11637.
Otterstatter MC and TL Whidden. 2004. Patterns of parasitism by tracheal mites (*Locustacarus buchneri*) in natural bumble bee populations. Apidologie 35:351-357.
Otterstatter MC, RJ Gegear, SR Colla, and JD Thomson. 2005. Effects of parasitic mites and protozoa on the flower constancy and foraging rate of bumble bees. Behavioral Ecology and Sociobiology 58:383-389, DOI:10.1007/s00265-005-0945-3.
Otterstatter MC and JD Thomson. 2008. Does pathogen spillover from commercially reared bumble bees threaten wild pollinators? PLoS ONE 3, http://www.plosone.org/doi/pone.0002771
Otti O and P Schmid-Hempel. 2007. *Nosema bombi*: A pollinator parasite with detrimental fitness effects. Journal of Invertebrate Pathology 96:118–124.
Otti O and P Schmid-Hempel. 2008. A field experiment on the effect of *Nosema bombi* in colonies of the bumblebee *Bombus terrestris*. Ecological Entomology 33:577–582.
Panzer R. 2002. Compatibility of prescribed burning with the conservation of insects in small, isolated prairie reserves. Conservation Biology 16(5)1296-1307.
Paton DC. 1990. Budgets for the use of floral resources in mallee heath. In The Mallee Lands: A Conservation Perspective, ed. JC Noble, PJ Joss, GK Jones, pp. 189–93. Melbourne: CSIRO.
Paton DC. 1996. Overview of feral and managed honeybees in Australia: distribution, abundance, extent of interactions with native biota, evidence of impacts and future research. Canberra: Aust. Nat. Conserv. Agency.
Patten KD, CH Shanks, and DF Mayer. 1993. Evaluation of herbaceous plants for attractiveness to bumble bees for use near cranberry farms. Journal of Apicultural Research 32:73-79.
Peat J, J Tucker, and D Goulson. 2005. Does intraspecific size variation in bumblebees allow colonies to efficiently exploit different flowers? Ecological Entomology 30:176-181.
Plath OE. 1922. Notes on the nesting habits of several North American bumblebees. Psyche 29(5-6):189-202.
Pleasants JM and KS Oberhauser. 2012. Milkweed loss in agricultural fields because of herbicide use: effect on the monarch butterfly population. Insect Conserv. Diver. DOI: 10.1111/j.1752-4598.2012.00196.x.
Plowright RC, BA Pendrel, and IA McLaren. 1978. The impact of aerial fenitrothion spraying upon the population biology of bumble bees (*Bombus* Latr: Hym.) in south-western New Brunswick. Canadian Entomologist 110:1145-1156.
Plowright RC, JD Thomson, and GR Thaler. 1980. Pollen removal in *Cypripedium acaule* (Orchidae) in relation to aerial fenitrithion spraying in New Brunswick. Canadian Entomologist 112:765-769.
Plowright CMS and RC Plowright. 1997. The advantage of short tongues in bumble bees (*Bombus*): analyses of species distributions according to flower corolla depth, and of working speeds on white clover. Canadian Entomologist 129:51-59.
Rasmont P, A Pauly, M Terzo, S Patiny, D Michez, S Iserbyl, Y Barbier, and E Haubruge. 2006. The survey of wild bees (Hymenoptera, Apoidea) in Belgium and France. p.18 In Status

of the World's Pollinators. Food and Agriculture Organization of the United Nations, Rome.
Reed CC. 1995. Species richness of insects on prairie flowers in southeastern Minnesota. Proceedings of the 14th North American Prairie Conference: 103-115.
Renner SS. 1998. Effects of habitat fragmentation on plant-pollinator interactions in the tropics. In Dynamics of tropical communities (Ed. D.M. Newbery, H.H.T. Prins & N.D. Brown), pp. 339-360. Oxford: Blackwell Science.
Roland J and SF Matter. 2007. Encroaching forests decouple alpine butterfly population dynamics. Proceedings of the National Academy of Sciences 104:13702-13704.
Rutrecht ST, Brown MJF. 2008. Within colony dynamics of Nosema bombi infections: disease establishment, epidemiology and potential vertical transmission. Apidologie 39:504-514.
Rutrecht ST, Brown MJF. 2009. Differential virulence in a multiple host parasite of bumble bees: resolving the paradox of parasite survival? Oikos 118:941-949.
SARA (Species At Risk Act). April 2010. http://www.sararegistry.gc.ca/species/speciesDetails_e.cfm?sid=1081
Schmid-Hempel P. 2001. On the evolutionary ecology of host-parasite interactions: addressing the question with regard to bumblebees and their parasites. Naturwissenschaften 88:147-158.
Schmid-Hempel P and R Loosli. 1998. A contribution to the knowledge of *Nosema* infections in bumble bees, *Bombus* spp. Apidologie 29:525-535.
Schmid-Hempel R and M Tognazzo. 2010. Molecular divergence defines to distinct lineages of Crithidia bombi (Trypanosomatidae), parasites of bumblebees. The Journal of Eukaryotic Microbiology 57(4):337-345.
Schultz CB and EE Crone. 1998. Burning prairie to restore butterfly habitat: A modeling approach to management tradeoffs for the Fender's blue. Restoration Ecology 6(3):244-252.
Scohier A, A Ouin, A Farruggia, and B Dumont. Is there a benefit of excluding sheep from pastures at flowering peak on flower-visiting insect diversity? Journal of Insect Conservation, DOI 10.1007/s10841-012-9509-9
Scott-Dupree CD, L Conroy, and CR Harris. 2009. Impact of currently used or potentially useful insecticides for canola agroecosystems on *Bombus impatiens* (Hymenoptera: Apidae), *Megachile rotundata* (Hymenoptera: Megachilidae), and *Osmia lignaria* (Hymenoptera: Megachilidae). Journal of Economic Entomology 102:177-182.
Shepherd M, SL Buchmann, M Vaughan, and SH Black. 2003. Pollinator Conservation Handbook. The Xerces Society, Portland, Oregon.
Shepherd MD, DM Vaughan, and SH Black (Eds). Red List of Pollinator Insects of North America. CD-ROM Version 1 (May 2005). Portland, OR: The Xerces Society for Invertebrate Conservation.
Shykoff JA and P Schmid-Hempel. 1991. Parasites delay worker reproduction in bumblebees: consequences for eusociality. Behavioral Ecology 2(3):242-248.
Singh R, AL Levitt, EG Rajotte, EC Holmes, N Ostiguy, D vanEngelsdorp, WI Lipkin, CW dePamphilis, AL Toth, and DL Cox-Foster. 2010. RNA Viruses in Hymenopteran Pollinators: Evidence of Inter-Taxa Virus Transmission via Pollen and Potential Impact on Non-Apis Hymenopteran Species. PLoS ONE 5(12): e14357. doi:10.1371/journal.pone.0014357.

Sjodin NE. 2007. Pollinator behavioural responses to grazing intensity. Biodiversity Conservation 16:2103-2121.

Skyrm KM. 2011. Environmental impacts on native bumble bee pollinators in an agricultural landscape of western Oregon. Ph. D. Dissertation in Entomology. Oregon State University.

Smallidge PJ and DJ Leopold. 1997. Vegetation management for the maintenance and conservation of butterfly habitats in temperate human-dominated habitats. Landscape and Urban Planning 38:259-280.

Speight DL. 1967. The bumblebees (Hymenoptera, Apidae, Bombinae) of east Tennessee. Master's Thesis, University of Tennessee, Knoxville, TN.

Stiles EW. 1977. Foraging behavior of bumblebees (*Bombus impatiens*, *Bombus vagans*, *Bombus affinis*) on false foxglove (*Aureolaria pedicularia*). Journal of the New York Entomological Society 85:249-252.

Sugden EA. 1985. Pollinators of *Astragalus monoensis* Barneby (Fabaceae) - New host records - Potential impact of sheep grazing. Great Basin Naturalist 45:299-312.

Szabo ND, SR Colla, DL Wagner, LF Gall and JT Kerr. 2012. Do pathogen spillover, pesticide use, or habitat loss explain recent North American bumblebee declines? Conservation Letters 5(3):232-239.

Tasei JN, J Lerin, and G Ripault. 2000. Sub-lethal effects of imidacloprid on bumblebees, *Bombus terrestris* (Hymenoptera: Apidae), during a laboratory feeding test. Pest Management Science 56:784-788.

Thompson HM. 2001. Assessing the exposure and toxicity of pesticides to bumblebees *Bombus* sp.). Apidologie 32:305-321.

Thompson HM and LV Hunt. 1999. Extrapolating from honeybees to bumblebees in pesticide risk assessment. Ecotoxicology 8:147–166.

Thomson D. 2004. Competitive interactions between the invasive European honey bee and native bumble bees. Ecology 85:458-470.

Thomson DM. 2006. Detecting the effects of introduced species: a case study of competition between *Apis* and *Bombus*. Oikos 114:407-418.

Thomson JD. 2010. Flowering phenology, fruiting success and progressive deterioration of pollination in an early-flowering geophyte. Philosophical Transactions of the Royal Society B: Biological Sciences 365:3187-3199.

Thorp RW and MD Shepherd. 2005. Subgenus *Bombus*. Latreille, 1802 (Apidae: Apinae: Bombini). In Shepherd, M. D., D. M. Vaughan, and S. H. Black (eds.) Red List of Pollinator Insects of North America. CD-ROM Version 1 (May 2005). Portland, OR: The Xerces Society for Invertebrate Conservation. http://www.xerces.org/Pollinator_Red_List/Bees/Bombus_Bombus.pdf

Thorp RW, S Jepsen, SF Jordan, E Evans and SH Black. 2010. Petition to list Franklin's bumble bee as an Endangered Species under the U.S. Endangered Species Act, Submitted by The Xerces Society for Invertebrate Conservation. http://www.xerces.org/wp-content/uploads/2010/06/bombus-franklini-petition.pdf

van Der Steen JJM. 2008. Infection and transmission of *Nosema bombi* in *Bombus terrestris* colonies and its effect on hibernation, mating and colony founding. Apidologie 39:273-282, DOI: 10.1051/apido:2008006 van Wilgenburg E, G Driessen, and LW Beukeboom. 2006. Single locus complementary sex determination in Hymenoptera: an “unintelligent” design? Frontiers in Zoology 3(1), http://www.frontiersinzoology.com/content/3/1/1

Vazquez DP and D Simberloff. 2003. Changes in interaction biodiversity induced by an introduced ungulate. Ecology Letters 6:1077-1083.

Velthius HHW and A van Doorn. 2006. A century in advances in bumblebee domestication and the economic and environmental aspects of its commercialization for pollination. Apidologie 37:421-451.

Walther-Hellwig K and R Frankl. 2000. Foraging distances of *Bombus muscorum*, *Bombus lapidarius*, and *Bombus terrestris* (Hymenoptera, Apidae). Journal of Insect Behavior 13:239–246.

Walther-Hellwig K, G Fokul, R Frankl, R Buchler, K Ekschmitt, and V Wolters. 2006. Increased density of honeybee colonies affects foraging bumblebees. Apidologie 37:517-532, http://dx.doi.org/10.1051/apido:2006035

Wolf S and RFA Moritz. 2008. Foraging distance in *Bombus terrestris* (Hymenoptera: Apidae). Apidologie 38:419-427.

Walton R, D Huie and S Sheehan. Plant-pollinator associations in reconstructed prairies at Fermilab. Chicago Wilderness Journal 3(2):24-31.

Westphal C, I Steffan-Dewenter, and T Tscharntke. 2003. Mass flowering crops enhance pollinator densities at a landscape scale. Ecology Letters 6:961-965. DOI 10.1046/j.1461-0248.2003.00523.x

Westphal C, I Steffan-Dewenter, and T Tscharntke. 2009. Mass flowering oilseed rape improves early colony growth but not sexual reproduction of bumblebees. Journal of Applied Ecology 46:187-193. DOI 10.1111/j.1365-2664.2008.01580.x

Whitehorn PR, MC Tinsley, MJF Brown, B Darvill, and D Goulson. 2009. Impacts of inbreeding on bumblebee colony fitness under field conditions. BMC Evolutionary Biology 9:152, http://www.biomedcentral.com/1471-2148/9/152

Whitehorn PR, MC Tinsley, MJF Brown, B Darvill, and D Goulson. 2010. Genetic diversity, parasite prevalence and immunity in wild bumblebees. Proceedings of the Royal Society B, DOI:10.1098/rspb.2010.1550

Whitehorn P, S O'Connor, F Wackers, and D Goulson. 2012. Neonicotinoid pesticide reduces bumble bee colony growth and queen production. Science, 10.1126/science.1215025.

Whittington R, ML Winston, C Tucker, and AL Parachnowitsch. 2004. Plant-species identity of pollen collected by bumblebees placed in greenhouses for tomato pollination. Canadian Journal of Plant Science 599-602.

Williams PH. 1986. Environmental change and the distribution of British bumble bees (*Bombus* Latr.). Bee World 67:50-61.

Williams PH. 1998. An annotated checklist of bumble bees with an analysis of patterns of description (Hymenoptera: Apidae, Bombini). Bulletin of the Natural History Museum, London (Ent.) 67:79-152. Updated at: www.nhm.ac.uk/research-curation/projects/bombus/

Williams PH, SA Cameron, HM Hines, B Cederberg, and P Rasmont. 2008. A simplified subgeneric classification of the bumblebees (genus *Bombus*). Apidologie 39:46-74.

Williams PH, MJF Brown, JC Carolan, J An, D Goulson, A Murat Ayetkin, LR Best, AM Byvaltsev, B Cederberg, R Dawson, J Huang, M Ito, A Monfared, RH Raina, P Schmid-Hempel, CS Sheffield, P Sima, and Z Xie. 2012. Unveiling cryptic species of the bumblebee subgenus *Bombus* s. str. Worldwide with COI barcodes (Hymenoptera: Apidae). Systematics and Biodiversity, http://dx.doi.org/10.1080/14772000.2012.664574

Wills RT, Lyons MN, Bell DT. 1990. The European honey bee inWestern Australian kwongan: foraging preferences and some implications for management. Proc. Ecol. Soc. Aust. 16:167–76

Wisconsin Department of Natural Resources, 17 June 2011, http://www.dnr.state.wi.us/org/land/er/biodiversity/index.asp?mode=detail&Grp=50

Xerces Society. 2010. Petition before the US Department of Agriculture Animal and Plant Health Inspection Service. Submitted by The Xerces Society, January 12, 2010. http://www.xerces.org/petition/xerces-bumblebee-petition-to-aphis.pdf

Xerces Society. 2012. Database of records from Bumble Bee Citizen Monitoring Project (2008-2012).

Xie Z, P Williams, and Y Tang. 2008. The effect of grazing on bumblebees in the high rangelands of the eastern Tibetan Plateau of Sichuan. Journal of Insect Conservation 12(6):695-703.

Zayed A and L Packer. 2005. Complementary sex determination substantially increases extinction proneness of haplodiploid populations. Proceedings of the National Academy of Sciences 102:10742-10746.

Zayed A. 2009. Bee genetics and conservation. Apidologie 40(3):237-262.

X. PERSONAL COMMUNICATION

A. J. Mayor, pers. comm. with E. Evans, Sept. 2007

B. Williams, pers. comm. with S. Jepsen, August 2009

C. Hlohowskyj, pers. comm. with R. Hatfield, July 2012

C. Reed, pers. comm. with E. Evans June 2007, July 2008

Christy Stewart, pers. comm. with S. Jepsen, Aug 2011 and with R. Hatfield July 2012

Colin Stewart, APHIS, pers. comm. with S. Jepsen, Nov. 2010

H. Holm, pers. comm. with R. Hatfield, July and August 2012

I. Loser, pers. comm. with S. Jepsen, Dec. 2007; http://bugguide.net/node/view/80952#93112

J. Grixti and C. Favret, pers. comm. with E. Evans Nov 2007

J. James-Heinz, pers. comm. with S. Jepsen, Sept. 2008

J. Knutson, pers. comm. with S. Jepsen, Aug. 2010 and Aug. 2011

J. Lozier, pers. comm. with S. Jepsen, Oct. 2012

K. Pouliquen, pers. comm. with E. Evans, June 2012

L. Day, pers. comm. with S. Jepsen, July 2010

M. Notestine, pers. comm. with S. Jepsen, Feb. 2010

N. Rafferty pers. comm. with S. Jepsen February 2011

P. Williams, pers. comm. with E. Evans, July 2008

R. Gegear, pers. comm. with E. Evans, May 2008

R. Jacobson, pers. comm. with E. Evans, Sept 2007

R. Jean and P.E. Scott, pers. comm. with E. Evans, Sept. 2007

R. Malfi, pers. comm. with S. Jepsen, July 2010

R. Thorp, Email to BOMBUS listserv, May 2011
S. Carpenter and M. Murray, pers. comm. with Xerces Society staff 2011-2012
S. Colla, email to BOMBUS listserv, Aug. 2009
S. Colla, pers comm. with S. Jepsen, Dec. 2012
S. Droege, pers. comm. with E. Evans, Feb. 2008
S. Hendrix and C. Gienapp, pers. comm. with E. Evans, Sept. 2007
S. Sheehan, pers. comm. with E. Evans, March 2008

XI. APPENDIX I

Original data providers contributing to Colla *et al.* in prep., illustrated in Figure 1, include:

American Museum of Natural History [NSF Grant #NSF-DBI#0956388 (PI Ascher)]; collections from AMNH, CAES, GSC, JSA, NYSM, RUAC, SIIS, UCD, UCMS, UMA, and UNHP
Cameron *et al.* 2011
Sheila Colla
Elaine Evans
Zuzu Gadallah
Doug Gill (via Sam Droege), Smithsonian
Guelph
Bernd Heinrich
Kansas SEMC
McGill University
Milliron 1970-1973
NCSU
OSUC
Peabody Yale U (012912)
Leif Richardson; UVM Zadock Thomson Museum, Dartmouth Teaching Collection
Cory Sheffield
Ethan Temeles
Paul Williams
www.bugguide.net
Xerces Society Citizen Monitoring Project
Doug Yanega (020312); Essig Museum Berkeley, Los Angeles County Museum, UC Riverside Entomology