CHAPTER TWO

Current trends in the oxidative stress and ageing of social hymenopterans

Hongmei Li-Byarlay[a,b,*] and Xaryn L. Cleare[a]

[a]Department of Agricultural and Life Science, Central State University, Wilberforce, OH, United States
[b]Agricultural Research and Development Program, Central State University, Wilberforce, OH, United states
*Corresponding author: e-mail address: hli-byarlay@centralstate.edu

Contents

Abstract

Social hymenopteran insects, including wasps, bees, and ants, are amazing insects with a versatile range of physiological and behavioural plasticity. The life span of ageing among different castes can be different from weeks to years. This plasticity is controlled by environmental and genetic factors. Oxidative stress is one of the mechanisms to explain the plasticity of ageing. In this report, we will review the molecular mechanisms underlying oxidative stress, how oxidative stress is associated with caste differentiation, epigenetics, energy and metabolic process, environmental and pesticide contamination in social insects. Future research is still needed to reveal new mechanisms of oxidative stress and ageing in social insects.

Advances in Insect Physiology, Volume 59
ISSN 0065-2806
https://doi.org/10.1016/bs.aiip.2020.09.002

Abbreviation

ATP	adenosine triphosphate
CAT	catalase
CYP	cytochrome P450
GPX	glutathione peroxidase
GR	glutathione reductase
GSH	glutathione
GSSG	glutathione disulphide
GST	glutathione S-transferases
GTPX	glutathione peroxidase homologue with thioredoxin peroxidase activity
IGF-1	insulin/insulin-like growth factor 1
JH	juvenile hormone
MDA	malondialdehyde
mGPDH	mitochondrial glycerol 3-phosphate dehydrogenase
OxPhos	oxidative phosphorylation
POX	peroxidase
ROS	reactive oxygen species
SOD	superoxide dismutase
TOR	target of rapamycin
TPX	thioredoxin peroxidase
Vg	vitellogenin

1. Introduction

Oxidative stress is characterized as a condition caused by cellular disruption of the prooxidant-antioxidant ratio (Finkel and Holbrook, 2000; Sohal and Weindruch, 1996) and involves multiorgan failure pathogenesis. Reactive oxygen species (ROS) are superoxide anions, hydroxyl radical, hydrogen peroxide, all intermediate in O_2 to H_2O reduction and derived from redox-active chemicals in living cells (Schieber and Chandel, 2014), which are by-products of oxidative phosphorylation pathway in the mitochondria (Li et al., 2008). When the homeostasis balance fails to be maintained in cells, overwhelmed accumulation of ROS can cause cellular damage such as protein oxidation, DNA damage and mutations, and membrane lipid peroxidation, which aid in ageing, carcinogenesis, and death of cells (Lennicke and Cocheme, 2020; Metcalfe and Alonso-Alvarez, 2010; Schieber and Chandel, 2014; Terman and Brunk, 2006). Conversely, antioxidants are the substances that scavenge and secure the various biomolecules, including DNA, protein, carbohydrates, lipids that combat oxidative

stress (Terman and Brunk, 2006). However, new evidence argues that ageing may not be caused by the accumulation of ROS (Blagosklonny, 2008; Brink et al., 2009; Parker, 2010), instead of by changes in developmental pathways as based on the evidence in *C. elegans* (Budovskaya et al., 2008). Empirical studies support that effects from higher system levels are more likely to drive the process of ageing than ROS or macromolecular damage accumulation (Blagosklonny, 2008; Budovskaya et al., 2008; Parker, 2010), which will not be covered here.

Research on ageing and oxidative stress has increased significantly in the past century because of improved health control and quality of human life (Crimmins, 2015). Studies to investigate the molecular mechanism of ageing and to use insect models are also increasing since the 1980s when we searched in the scientific literature (Fig. 1A). *Drosophila melanogaster*, for example, is a model insect system for ageing and oxidative stress studies (Li et al., 2008, 2009; Li-Byarlay et al., 2014; Partridge et al., 2011; Radyuk et al., 2001; Rascon and Harrison, 2010; Strycharz et al., 2013). Social insects in the order Hymenoptera (wasps, bees, and ants) have a complicated biological system which is fascinating for studies of physiology, behavioural ecology, and evolution (De Wilde and Beetsma, 1982; Hölldobler and Wilson, 2009; Kapheim et al., 2015; Michener, 1974; Wilson, 1979). Besides the order Hymenoptera, termites in the order Blattodea are social insects and a great model system to study ageing and oxidative stress as well (Kuhn and Korb, 2016). This review is focusing on social hymenopterans. Social insects have been studied since 1895, but around the last century, a paramount of impact occurred with research on social insects (Fig. 1B). The life span of these social

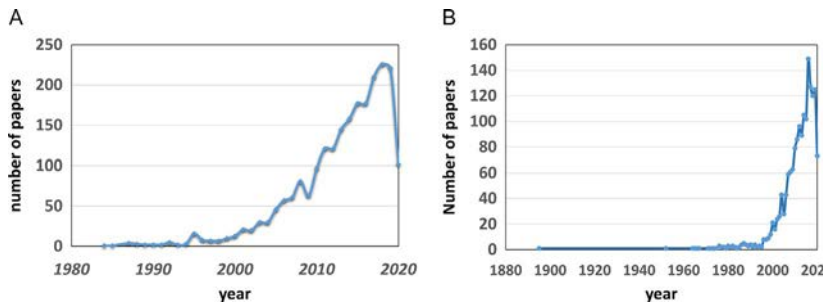

Fig. 1 The overall trend in (A) insects and oxidative stress and (B) social insects-related publications. A dramatic increase was detected in the number of publications when searched keywords of "insects/oxidative stress" or "social insects" in the title or abstract of PubMed database (searched in August 2020).

Table 1 Total life span length of different castes in social Hymenoptera.

Caste	Paper wasp (*Polistes* spp.)	Honey bee (*Apis mellifera*)	Leafcutter ant (*Atta*, *Acromyrmex*)
Queen/ reproductive	1 year	1–5 years	10–20 years
Worker/ nonreproductive	2–3 weeks	5–6 weeks/summer 17–21 weeks/winter	3–5 weeks

insects ranges from a few weeks to a few decades, which is controlled by the environmental and genetic factors (Rueppell, 2009). This wide range of variation of plasticity in ageing or senescence (Table 1) enable scientists to use social insects as unique models to study the molecular and physiological mechanisms underlying developmental plasticity (Keller and Jemielity, 2006; Lucas and Keller, 2020). However, gaps of knowledge and questions of how to use social insects to study ageing and oxidative stress have remained to be addressed.

Several classical hypotheses for ageing from an evolutionary perspective are (1) mutation accumulation hypothesis, which argued that ageing might result from accumulative effects of linkage and mutation (Medawar, 1952), (2) antagonistic pleiotropy hypothesis, which argues that natural selection will act to minimize the rate of ageing (Williams, 1957), and (3) ageing may be due to an energy-saving approach of reduced error control in somatic cells (Kirkwood, 1977). However, these hypotheses may not apply to social insects directly because colonies of social insects consist of different castes (reproductive vs nonproductive), different aged (young and old) individuals, and different tasks (a division of labour). In addition, natural selection may not act on individuals as classical models, but instead on the whole colony (Avalos et al., 2020; Kramer et al., 2016).

In this review, we summarize the previous and current proceedings addressing the research of oxidative stress and ageing from the perspectives of social evolution and caste determination, epigenetics, environmental factors, and underlying molecular and cellular mechanisms in these social hymenopterans, and coping mechanisms of oxidative stress.

2. Molecular mechanisms of oxidative stress in social insects

Several molecular and cellular mechanisms are linked to the process of ageing and oxidative stress. The vitellogenin (Vg), TOR (target of rapamycin) pathway, IGF-1 (insulin/insulin-like growth factor 1)

signalling, ILP (insulin-like peptide), transposable elements, and hormones are the main molecular and cellular mechanisms reported in social insects (Amdam, 2010; Elsner et al., 2018; Gems and Partridge, 2013; Heinze and Schrempf, 2008; Parker, 2010; Partridge et al., 2011; Tatar et al., 2003).

Reproductive proteins have been proven to protect against oxidative stress. Vg as an egg yolk precursor protein associated with lipid storage and transport protects workers against oxidative stress (Amdam et al., 2012; Salmela et al., 2016; Seehuus et al., 2006b; Zhang et al., 2017a). Juvenile hormone (JH) as a key factor for insect development is also associated with oxidative stress and ageing in social insects. In short-lived workers, nurse bees are stress-resistant with high Vg and low JH, whereas forager bees are more susceptible to stress with high JH and low Vg (Flatt et al., 2013; Keller and Jemielity, 2006). JH is related to the fecundity and longevity trade-off (Kuhn and Korb, 2016). In nonsocial insects, there is a trade-off between reproduction and longevity. Reproduction can decrease the lifespan of the organism. But in social insects, this is being reshaped with a much longer lifespan in the reproductive caste than the nonreproductive caste (Kuhn et al., 2019). From the physiology perspective, IGF affects the production of JH, and JH regulates the production of Vg, which is directly associated with reproduction in both reproductive and nonreproductive castes (Ament et al., 2008). The IGF-JH-Vg circuit directly regulates reproduction, which is also coordinated with growth (development), cellular repair, and maintenance (ROS and oxidative stress), homeostasis, and energy metabolism.

The accumulated oxidative damage or the level of oxidative stress is determined by the homeostasis balance between the ROS occurring in the cell, and the antioxidant defence (Finkel and Holbrook, 2000). Key enzymes act as antioxidants against ROS are superoxide dismutases (SODs), catalases (CATs), and peroxidases (POXs). CATs break down hydrogen peroxide into oxygen and water. POXs catalyse a chemical reaction to reduce hydrogen peroxide to water, which reduces TRX or GSH (Radyuk et al., 2001). Insects also contain additional enzymes that can act as POXs, which are TPXs, GTPX, GSTs (Corona and Robinson, 2006; Li et al., 2008; Weinhold et al., 1990). SOD in bees and ants plays a major role in cellular stress responses and antioxidant processes, but may not be related to queen longevity (Jia et al., 2014; Parker et al., 2004). SOD, GST, and CAT may affect the spermathecal and sperm storage in queens (Collins et al., 2004; Weirich et al., 2002). In Eastern honey bees, *Apis cerana*, the function of the GST gene was validated as an antioxidant (Liu et al., 2016; Yan et al., 2012; Zhao et al., 2019).

The end of chromosomes, telomeres, are considered to determine the limitation of replicates in somatic cells of vertebrates, and are closely related to ageing (Hall et al., 2004; Harley et al., 1990; Lopez-Otin et al., 2013). Shortened telomeres result in limited growth of cells and ageing of cells in honey bees, bumblebee queens, and males of *Lasius niger* ants (Criscuolo et al., 2018; Jemielity et al., 2007; Korandova and Frydrychova, 2016; Koubova et al., 2019). Reasons for telomere shortening are the DNA damage by ROS and oxidative stress and the limited ability of DNA replication at the end of the DNA strand (Lingner et al., 1995; Passos and Von Zglinicki, 2006). Additional genomic analysis in honey bees showed the queen caste has higher telomerase gene expression than workers (Grozinger et al., 2007; Korandova and Frydrychova, 2016).

Compared to *Drosophila,* which is a classic genetic system with mutants and transgenic tools, social insects are not well established yet for tissue or gene-specific manipulations. But in recent years RNA interference via functional genomics, CRISPR genome editing tools, and improved genomes of social insects (Değirmenci et al., 2020; Elsik et al., 2014; Favreau et al., 2018; Kohno et al., 2016; Li-Byarlay et al., 2013; Trible et al., 2017) can be utilized for decoding molecular mechanism for oxidative stress and ageing. With these tools, mechanistic studies are potentially possible to understand the nature of ageing.

3. Oxidative stress and castes

3.1 Oxidative stress, ageing, and castes

Social insects in the order Hymenoptera live in a society with different castes, including a reproductive female caste—queens, nonreproductive female caste—workers, and male reproductive caste—males (Table 1). Among social bees, the honey bee serves as an excellent model for various areas of basic biological research (Amdam and Rueppell, 2006; Page et al., 2002; Robinson et al., 2008). Honey bees as eusocial insects live in either natural or managed colonies as extended families with one reproductive queen and typically 10,000–50,000 female sterile worker bees that perform all nonreproductive tasks (Winston, 1987) (Fig. 2). Males are only produced for the active season (spring, summer, and fall) of each year to mate with virgin queens from other colonies during mating flights. Young adult workers, as in-hive nurses, care for the young (eggs, larvae, and prepupae), build wax cells, clean hives and help store food, and feed the queen. By contrast, the older workers forage for nectar, pollen, and water as food for their hive. Nectar is processed into

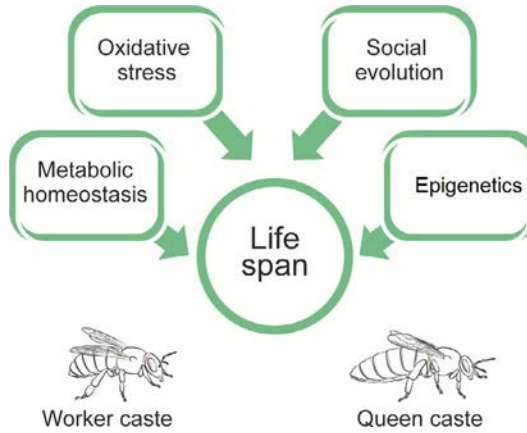

Fig. 2 Different factors for ageing and caste difference in social honey bees.

honey as a stored long-term food reserve, while pollen is converted to proteinaceous jelly that is mainly fed to the queen and the developing larvae (Winston, 1987).

Among all the different castes (queen, worker, and drone), there is a striking difference in their lifespan. Typically, a queen can live up to 3–5 years in a colony, but the workers only live about 6–8 weeks during the active season. During the winter, worker bees, or *diutinus* workers, are physiologically distinct from summer workers and can live for several months, but still have a shorter life span than a queen. This striking difference between castes (reproductive vs nonreproductive) make social honey bees a great model system to study ageing of social insects (Amdam and Rueppell, 2006; Corona et al., 2005; Rueppell, 2009).

When comparing queen and worker honey bees, the gene expression of antioxidants showed a decrease with age in queens, but the mitochondrial genes affecting ROS production may be associated with caste-specific differences in lifespan (Corona et al., 2005). Another study examined nine genes in different castes of the ant *Formica exsecta* that were infected with a fungus and found clear differences in their immune response between different castes (Stucki et al., 2017).

The theory of disposable soma proposed that more resources are allocated for the germline than the somatic cells. The reproductive queen caste and nonreproductive worker caste can be considered as the germ line and somatic cells of a colony. In the ants, *Temnothorax americanus and*

Temnothorax longispinosus, workers from both species have increased expression of genes linked to oxidation-reduction processes (Gstöttl et al., 2020). While queens of *T. longispinosus* avoid DNA degradation, queens of *T. americanus* invest in the metabolism of trehalose (Lucas et al., 2016). Ultimately, these variations are compatible with the hypothesis that greater expenditure in somatic repair would contribute to longer longevity (Lucas et al., 2016).

Gene expression measurements of juvenile queens indicated lower ROS levels, lower superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) activities, and higher thioredoxin reductase (TR) activity than mature queens, which suggested antioxidant activity in fat cells and lipid increase in queens (Hsieh and Hsu, 2013). Furthermore, in research performed in the ant, *Temnothorax rugatulus*, young queens have invested in immunity and tolerance to environmental and physiological stress, and older queens have invested in antiageing processes via overproduction of antioxidants (Negroni et al., 2019).

In addition, nutrition difference between royal jelly (fed to queen bee) and worker jelly (fed to worker bees) may allow queen bees to have peroxidation resistant-membranes (Haddad et al., 2007). In the honey bee, mechanical senescence has been found to decrease the mortality rate (Kojić et al., 2019), likely because it causes an increased workload and increased metabolism (Neukirch, 1982). In fact, honey bee researchers have found that many specific classes of genes correlated with survival in other organisms are substantially more regulated in queens (Grozinger et al., 2007). Analysis reveals that phospholipids of queen honey bees (and therefore their membranes) have more monounsaturated and fewer polyunsaturated fatty acids than worker bees; therefore the queen is substantially more immune to peroxidative damage (Haddad et al., 2007). Moreover, the expression of vitellogenin indicates an age-related drop in workers (Hartfelder and Engels, 1998).

Drones are male honey bees that can live for up to 55 days, with an average of 30 days (Metz and Tarpy, 2019; Rueppell et al., 2005; Seeley and Mikheyev, 2003). Under oxidative stress induced by paraquat, drones with higher resistant and longer life span were considered to be more tolerant of the oxidative damage than short-lived drones (Li-Byarlay et al., 2016). Additionally, research shows the major development in the spermatheca of mated queens throughout three enzymes (CAT, GST, and SOD) indicates they are tasked with providing long-term defence against ROS (Weirich et al., 2002).

3.2 Oxidative stress and aging within caste

Within the nonreproductive caste of social insects, a division of labour (Robinson, 1992) and a considerable plasticity in life span occurs in workers. For example, young worker bees, nurse bees, work in the dark hive and perform hive-related tasks such as brood care, building cells, and cleaning etc., then switch to foraging after about 3 weeks of adult life to collect pollen, nectar, and water for the colony. This behavioural maturation also increases the external mortality risk during foraging trips (Rueppell, 2009). Previous research indicates that young workers have higher amounts of reactive oxygen, greater levels of superoxide and thioredoxin reductase, and lower catalase and glutathione peroxidase relative to mature workers (Hsu and Hsieh, 2014). Furthermore, with the accumulation of the above enzymes, it is suggested that with ageing oxidative stress is reduced in the trophocytes or fat body of workers (Hsu and Hsieh, 2014). Foragers may have the highest accumulation of protein carbonylation (a type of protein oxidation that can be promoted by ROS), as it produces more ROS than nurse bees and diutinus workers (Seehuus et al., 2006a). Foraging is a dangerous activity linked to a high rate of mechanical senescence, as well as repeated exposure to pathogens and ultraviolet light (Kefuss and Nye, 1970). Proteomic research in the black garden ant, *Lasius niger*, showed a difference in protein profiles between foragers and nest-workers (Quque et al., 2019). In the worker ants, the proteins associated with energy metabolism were more abundant. Research on the ant *T. longispinosus* indicates that genes with higher expression in brood care workers relative to foragers and young workers display enriched functions linked to various repair mechanisms, including DNA integrity, oxidative stress response, and immunocompetence, suggesting heavy expenditure over a long life span (Kohlmeier et al., 2019).

4. Epigenetics, ageing, and oxidative stress in social insects

Epigenetic signals such as DNA methylation, histone modifications, chromatin modifications, transcription factors, and noncoding RNAs play a critical role in cell and gene regulations (Bird, 2007; Jirtle and Skinner, 2007; Wolffe and Matzke, 1999). DNA methylation is associated with the regulation of ageing-related genes (Yan et al., 2015). For example, Vg is one of the target genes of JH, which is regulated by the IIS pathway.

Vg can function as an antioxidant against oxidative stress in honey bee workers (Seehuus et al., 2006b). DNA methylation may modulate the expression of Vg and have an impact on the lifespan of worker bees (Cardoso-Júnior et al., 2018). When compared to expression patterns of different castes of honey bees, DNA methylation is also associated with their differential expressed genes, including ILP-2, TOR, Vg, yellow/major royal jelly protein family, translation initiation factor, and hexamerin family (Elango et al., 2009). DNA methylation can also affect alternative splicing as a mechanism of gene regulation (Li-Byarlay, 2016; Li-Byarlay et al., 2013). In human studies, alternative splicing may be associated with ageing and neurodegeneration (Harries et al., 2011; Tollervey et al., 2011).

Histone modifications are another potential mechanism to affect ageing. In worker ants of *Camponutus floridanus*, histone acetylation can regulate their behaviour (Simola et al., 2013). The chromatin modification, such as polycomb and trithorax, can regulate the life span of insects (Siebold et al., 2010). Interestingly during caste determination, genes may be regulated by DNA methylation, as well as chromatin accessibility and histone modifications (Bonasio et al., 2012; Herb et al., 2012).

Oxidative stress can cause DNA damage in the cells, which eventually leads to ageing (Metcalfe and Alonso-Alvarez, 2010; Shames et al., 2007). Indeed, DNA methylation is associated with DNA repair (Cuozzo et al., 2007) based on in vitro evidence. A lot of cancer research has shown interactions between DNA methylation and oxidative stress (Donkena et al., 2010; Franco et al., 2008; Zhang et al., 2017b). However, there is a gap of knowledge as to how DNA methylation and oxidative stress relate in the models of social insects.

5. Oxidative stress and metabolic process of social insects

5.1 Metabolic studies in bees

One classical theory of ageing is the "rate-of-living" hypothesis, indicating faster metabolism accelerates the ageing process (Pearl, 1928), which suggests that organisms may age faster with a faster metabolism. The underlying cause could be a limitation of energy or function in physiology (Neukirch, 1982). ROS are generated as by-products of oxidative phosphorylation pathway and energy metabolism in the mitochondria (Schieber and Chandel, 2014). The main role of mitochondria is to produce most of the cellular ATP by oxidative phosphorylation (>90%) (Boelsterli, 2007).

Substrates such as pyruvate and Kreb cycle intermediates are oxidized by subsequent dehydrogenases that decrease oxidized adenine dinucleotide nicotinamide (NAD^+) or flavin adenine dinucleotide (FAD). Transport by an electron from the oxidation of NAD (NADH) and reduced FAD (FADH2) to O_2 is directly related to ATP synthesis. It happens by protein-binding redox centres involving, complex I (Q reductase), II (coenzyme succinate Q reductase), III (coenzyme cytochrome *c* reductase), and then IV (cytochrome *c* oxidase). This happens through the redox centres that bind to proteins. The power generated by this transport is converted by a proton pump into an electrochemical hydrogen (H^+) gradient via the inner mitochondrial membrane. Alternatively, ATP synthesis is compulsorily aerobic, and much of the ATP synthesis needed to drive muscle contraction is derived from mitochondrial OxPhos (Rothe and Nachtigall, 1989; Sacktor, 1976). This gradient's electrochemical potential is then used for the synthesis of ATP by Complex V (ATPase) (Alberts et al., 2002; Li et al., 2017; Schmidt et al., 2001). Genes of the OxPhos pathway affect mitochondria and nuclear function together to support the production of ATP in cells and modulate social behaviour of social honey bees (Li-Byarlay et al., 2014; Shadel and Horvath, 2015). Incompatible mitochondrial and nuclear genes can decrease the production capacity of cellular ATP and lead to oxidative stress, causing a wide range of problems including developmental malformations (Bar-Yaacov et al., 2012). Even though most of the oxygen is reduced to water, a very small portion of it (5%) is reduced to a superoxide anion (Muller, 2000). In a variety of human chronic disease states, oxidative stress was typically grouped under the banner of the metabolic syndrome and is believed to contribute to the ageing cycle (Frisard and Ravussin, 2006).

The level of oxidative stress in different tissue types may differ due to the oxygen consumption, levels of antioxidant, and potential repair mechanisms (Li-Byarlay et al., 2016; Simone-Finstrom et al., 2016). The muscle cells with high metabolism have a possible high risk of ROS, which can outperform antioxidant investments (Ji et al., 1998). Social insects as flying insects are good model systems to study the role of energy metabolism, oxidative stress, and ageing (Hedges et al., 2019; Margotta et al., 2018; Orčić et al., 2017; Suarez et al., 2000). A previous study showed worker honey bees with more flight experience displayed more oxidative DNA damage and less antioxidants than those bees with less flight experience (Margotta et al., 2018). Flight muscles in honey bees may produce high levels of ROS which could be responsible for behavioural senescence and reduced lifespan due to declines in antioxidant activity (Williams et al., 2008).

A higher protein expression of SOD as antioxidant defence was detected in mature foragers compared to nonforaging hive bees because of highly oxidative muscles (Schippers et al., 2006). Heat shock proteins such as hsp70 can be induced by flight and oxidative damage to flight muscles (Elekonich, 2009). More studies are needed to measure the enzyme activities of the electron transport chain and antioxidants in flying social insects.

Social bees and ants with small body size have higher metabolic rates in the brain and brain structures also change during ageing (Kern and Wegener, 1984; Kraft et al., 2019; Seid et al., 2005, 2008). The nervous system in worker bees may cost a significant portion of the bees bodily metabolism and energy, which may be similar to mammals (Aiello and Wheeler, 1995; Giraldo and Traniello, 2014; Li-Byarlay et al., 2014). Gene expressions of cytochrome C and Clk-1 (clock-1) in queen bees, which are associated with electron and proton transfer between complexes of OxPhos pathway, were higher than workers (Corona et al., 2005). The honey bee brain has more plasticity in gene expression than flight muscle; therefore, it may have more capacity for stress mitigation (Margotta et al., 2012). When the corpora allata were removed, honey bees slowed their ground speed during orientation flights and had a decreased metabolic rate (Sullivan et al., 2003).

The honey bee colony relies on utilizing organismic and cellular-molecular processes to respond when hive conditions vary from a homeostasis level. Therefore, cellular stress protein response (heat shock proteins, HSP) is an essential component for a systemic response that provides resilience to organism stress (Feder and Hofmann, 1999). Rapid flight-related aggregation of ROS in older flight-capable hive bees and foraging bees results in elevated HSP70 levels due to protein damage and declining antioxidants with age (Elekonich, 2009).

5.2 Ants and wasps

Compared to studies on the metabolic process and oxidative stress in bees, there are only a few studies in ants and wasps. When the oxygen supply is insufficient in cells, insects experience the condition of hypoxia, which can reduce the life span (Sohal and Weindruch, 1996). One example is the carpenter ant, *Camponotus vicinus*, with utilizes discontinuous gas exchange and spends a significant part of their life cycle underground (Harrison et al., 2006; Lighton and Garrigan, 1995). Gene expression in ant colonies of *Temnothorax longispinosus* was shown to be associated with behavioural

differentiation (nurse vs forager), age (young vs old), and fertility (fertile vs infertile). Enriched gene ontology concepts were largely represented by multiple processes associated with metabolism, catabolism, and anion transport (Kohlmeier et al., 2019). The reduced commitment of foragers and aged workers in antiageing processes, therefore, confirms the characterization of the usually aged foragers as a disposable caste (Porter and Jorgensen, 1981). Lipids or fatty acids are stored in cells as supply of energy for oxidative metabolism (Arrese and Soulages, 2010). When receiving lipid supplements, a reduction of lifespan as measured in groups of ants occurred (Vieira and Bueno, 2015). Unlike bees that forage for plant nectar and pollen, social wasps forage for animal protein and carbohydrates (Richter, 2000). Therefore, wasps are likely to have a metabolic cost of elevated production of ROS due to higher mitochondrial glycerol 3-phosphate dehydrogenase (mGPDH) activity (Hedges et al., 2019).

6. Environmental factors

6.1 Insecticides and environmental pollution

Besides genetic programming, a major cause of oxidative stress and ageing in social insects are environmental factors including pesticide or chemical contamination in the habitat, food and foraging resource, nutrition, temperature, and other related factors, which are considered to be the abiotic stress for social hymenopterans (Chakrabarti et al., 2015; Goulson et al., 2015; Lopez-Uribe et al., 2020; Neuwirthová et al., 2019). Environmental and ecological factors inducing oxidative stress in social honey bees can be colony habitat change by human management, pesticides exposure, UV light, parasitic and pathogenic stresses, and predation variables.

Insecticides such as fipronil, flupyradifurone, chlorpyrifos, and neonicotinoids can induced oxidative stress and produce ROS in the midgut and brains of honey bees (Chakrabarti et al., 2015, 2020; Decourtye et al., 2004; Gauthier et al., 2018; Ma et al., 2019; Paris et al., 2017; Roat et al., 2014; Shafiqur et al., 2012). Oxidative damage caused by chlorpyrifos in the honey bee nervous system resulted in the build-up of toxic hydroxide ions, which could provide a possible mechanism for chlorpyrifos neurotoxicity (Shafiqur et al., 2012). A common neonicotinoid pesticide imidacloprid in chronic and sublethal exposure, together with the broadly distributed parasite *Nosema ceranae* caused physiological changes observed in queens. Increase in enzyme activities of catalase [CAT] and glutathione S-transferase [GST] were associated with protective responses to xenobiotics

and oxidative stress (Dussaubat et al., 2016). The GSTs groups Delta and Epsilon are special to insects and comprise the bulk of GSTs correlated with insecticidal detoxification.

Herbicides such as paraquat and atrazine are often used as an inducer for oxidative stress in social insects. The treatment of paraquat changed the expression of several antioxidant and detoxification-related genes, as well as a spike in pathogen titres (de Mattos et al., 2018). Biochemical and molecular markers of oxidative stress response indicated atrazine compromised the health of bee colonies (Williams, 2016). Fungicides such as acetamiprid and propiconazole increased oxidative stress and mortality with cellular damage (Han et al., 2019). The fungicide chlorothanolin affected the differential transcriptional expression of oxidative chromatin-related genes and proteins, detoxification enzymes, acetylcholine receptor alpha 1, and endocrine regulation in honey bees (Christen et al., 2019).

Certain miticides have potential risks to bees and can disrupt the homeostasis of bee colonies under chronic conditions (Qi et al., 2020). Research data have also shown that fluvalinate, a synthetic pyrethroid chemical compound as a miticide, has led to an increase in GST activity and a decrease in AChE activity in emerging and nurse bees compared with controls (Rouibi et al., 2016). The above mentioned pesticides presented here are a few examples to show their effects on the oxidative phosphorylation pathway, which generated ROS for increasing oxidative stress in social insects. Almost all the previous studies are in honey bees due to their important ecological roles as pollinators. More research in the future is needed to reveal the effect of pesticides on the physiology of social wasps and ants.

Environmental pollution of selenium, heavy metals, and airborne fine particulates can affect oxidation stress in social insects (Alburaki et al., 2019; Feldhaar and Otti, 2020). Furthermore, orally ingested metals (Al, Pb and Cd) at concentrations found in the environment altered the metallothionein-like protein (MTLP) (Gauthier et al., 2016).

6.2 Environmental changes

Environmental changes in habitat and foraging area can become a new normal when managed pollinators such as honey bees are used for pollination service. The long transportation or migration of bee colonies on trucks from the East coast to the West coast of the United States creates changes in the colony environment for foraging and food resources. This long distance transportation significantly reduced the life span of honey bee workers via

oxidative stress in lipid and protein damage (Simone-Finstrom et al., 2016). Effects of transportation on bees (24-h after a 3-day trip) led to a reduction in the size of the hypopharyngeal gland acini which are essential for brood food production in nurse bees and larval nutrition (Ahn et al., 2012). Higher oxidative stress was detected in commercially managed honey bee colonies than that of traditionally managed colonies (Taric et al., 2020), which showed evidence of anthropogenic influence. Long-term greenhouse pollination reduces the detoxification potential and thus causes oxidative stress in honey bees by generating more protein carbonyl during greenhouse pollination (Morimoto et al., 2011). In addition, an ecological condition such as predation by wasps affects oxidative lipid damage, the activity of antioxidants as well as levels of mitochondrial-related enzymes under natural conditions (Leza et al., 2019).

6.3 Heat stress

Heat stress in colonies during the beekeeping practice of long distance transport caused the loss of sperm viability and changes in enzyme expressions to alleviate oxidative stress in honey bee queens (McAfee et al., 2020). Heat stress also caused changes in life span and expression changes in genes of worker bees associate with oxidative stress (Bordier et al., 2017; Li et al., 2019). Also, urbanization can also increase pathogen pressure on managed honey bees (Youngsteadt et al., 2015). Research suggests that high temperatures can adversely affect bees not only on their survival and water loss but also in stimulating oxidative stress (Helmer et al., 2015). Interestingly, fluctuation of temperatures prevent chill injury but did not the damage caused by oxidative stress or antioxidant capacity in the alfalfa leafcutting bee, *Megachile rotundata* (Torson et al., 2019).

6.4 Nutrition

Nutrition input, such as pollen or dietary conditions, also plays a pivotal role in oxidative stress levels of social insects (Freitak et al., 2014; Huang, 2012). Feeding bees with a poor quality pollen was shown to decrease the resistance of bees to microsporidians, *Nosema apis*, an increase in pesticide sensitivity, and an increased titre of bee virus (Huang, 2012). On the other hand, pollen substitute with a protein level of approximately 30% was identified as an excellent diet for antioxidant enzymatic activity (Li et al., 2012). Dietary supplementation with vitamin C (ascorbic acid) increased total antioxidant status (TAS), glutathione content, and activity of four antioxidant enzymes:

SOD, POX, CAT, and GST (Farjan et al., 2012). Propolis collected by the native stingless bees (Meliponinae) was a strong source of natural antioxidants to be used in the hive (Araujo et al., 2016). Furthermore, research shows that bee pollen consists of flower pollen mixed with bee digestive enzymes and preserved with some honey and nectar, contained high antioxidant activity due to the presence of polyphenols and flavonoids which are beneficial for the hive, thus aiding against oxidative stress (Fadzilah et al., 2017). Future studies are very much needed for additional knowledge on the role of nutrients against oxidative stress on other social Hymenoptera.

6.5 Parasites and pathogens

Parasites and pathogens as ecological factors of the colonies, can cause oxidative stress as well. By comparing honey bees infected with *Varroa destructor* mites, the parasitization by mites in the colony brood caused a substantial loss in their protein content and antioxidant enzymes (Badotra et al., 2013). In addition, significantly high activity of three antioxidant enzymes isolated from *Varroa destructor*-infested drone prepupae tissues suggests that oxidative stress may be one of the pathogenic pathways of varroosis. The average activity of superoxide dismutase (SOD), glutathione peroxidase (GPX) and plasma protein ceruloplasmin (CP) in infested prepupae is about 2–4 times higher than levels in mite-free prepupae (Lipiński and Żółtowska, 2005). Moreover, research indicates, in the presence of a parasite (*N. ceranae*) alone, the ROS damage was decreased. However, the combination of *N. ceranae* and fipronil pesticide reveals an increasing level of oxidative damage and suggests increased toxicity to fipronil in infected bees (Paris et al., 2017). Researchers analysed the impact of *N. ceranae* on queen physiology and found that *N. ceranae* infection did not affect the fat body content but altered the vitellogenin titre (a fertility and longevity indicator), total antioxidant capacity, and queen mandibular pheromones, all of which increased significantly in *Nosema*-infected queens (Alaux et al., 2010). Overall, environmental factors can significantly affect the levels of oxidative stress and ageing of social bees and ants.

7. Conclusion and future directions

The phenomenon of ageing is a complicated process affected by physiology, social evolution, development, ecological factors, and environmental factors. Mechanistically and phenotypically, oxidative stress is highly relevant to ageing, either from genetic or environmental factors. Our report

synthesized the current research status of oxidative stress and ageing among social wasps, bees, and ants. Most research was performed in the species with economic and ecological importance. We also addressed the molecular and cellular mechanisms underlying oxidative stress and antioxidant gene families. Future investigations with new genomic tools such as CRISPR genome editing and high-throughput functional protein analysis will be expected to draw a more complete picture of how the ageing process works in social insect physiology.

Acknowledgements

We thank the funding support from the USDA NIFA Evans-Allen grant (NI181445XXXXG007), USDA SARE NCR grant, USDA AFRI Pollinator Health Seed Grant, NSF RIA Grant, and Central State University Agricultural Research and Development Program. We also thank Rachelle Adams at Ohio State University for discussions and Susan Balfe for the art design of Fig. 2.

References

Ahn, K., Xie, X., Riddle, J., Pettis, J., Huang, Z.Y., 2012. Effects of long distance transportation on honey bee physiology. Psyche J. Entomol. 193029.

Aiello, L.C., Wheeler, P., 1995. The expensive-tissue hypothesis: the brain and the digestive system in human and primate evolution. Curr. Anthropol. 36, 199–221.

Alaux, C., Folschweiller, M., McDonnell, C., Beslay, D., Cousin, M., Dussaubat, C., Brunet, J.L., Le Conte, Y., 2010. Pathological effects of the microsporidium *Nosema ceranae* on honey bee queen physiology (*Apis mellifera*). J. Invertebr. Pathol. 106, 380–3865.

Alberts, B., Johnson, A., Lewis, J., Raff, M., Roberts, K., Walter, P., 2002. The Mitochondrion. Molecular Biology of the Cell, fourth ed. Garland Science.

Alburaki, M., Smith, K.D., Adamczyk, J., Karim, S., 2019. Interplay between selenium, selenoprotein genes, and oxidative stress in honey bee *Apis mellifera* L. J. Insect Physiol. 117, 103891.

Amdam, G.V., 2010. Social context, stress, and plasticity of aging. Aging Cell 10, 18–27.

Amdam, G.V., Rueppell, O., 2006. Models of aging in honey bee workers. In: Conn, P.M. (Ed.), Handbook of Models for Human Aging. Academic Press, San Diego, CA, pp. 267–276.

Amdam, G.V., Fennern, E., Havukainen, H., 2012. Vitellogenin in honey bee behavior and lifespan. In: Galizia, C.G., Eisenhardt, D., Giurfa, M. (Eds.), Honey Bee Neurobiology and Behavior. Springer, pp. 17–29.

Ament, S.A., Corona, M., Pollock, H.S., Robinson, G.E., 2008. Insulin signaling is involved in the regulation of worker division of labor in honey bee colonies. Proc. Natl. Acad. Sci. U. S. A. 105, 4226–4231.

Araujo, K.S.D., dos Santos, J.F., Sato, M.O., Finco, F., Soares, I.M., Barbosa, R.D., Alvim, T.D., Ascencio, S.D., Mariano, S.M.B., 2016. Physicochemical properties and antioxidant capacity of propolis of stingless bees (Meliponinae) and *Apis* from two regions of Tocantins, Brazil. Acta Amazon. 46, 61–68.

Arrese, E.L., Soulages, J.L., 2010. Insect fat body: energy, metabolism, and regulation. Annu. Rev. Entomol. 55, 207–225.

Avalos, A., Fang, M., Pan, H., Lluch, A.R., Lipka, A.E., Zhao, S.D., Giray, T., Robinson, G.E., Zhang, G., Hudson, M.E., 2020. Genomic regions influencing aggressive behavior in honey bees are defined by colony allele frequencies. Proc. Natl. Acad. Sci. U. S. A. 117, 17135–17141.

Badotra, P., Kumar, N.R., Harjai, K., 2013. *Varroa* causes oxidative stress in *Apis mellifera*. L. J. Glob. Biosci. 2, 199–201.

Bar-Yaacov, D., Blumberg, A., Mishmar, D., 2012. Mitochondrial-nuclear co-evolution and its effects on OXPHOS activity and regulation. BBA Gene Regul. Mech. 1819, 1107–1111.

Bird, A., 2007. Perceptions of epigenetics. Nature 447, 396–398.

Blagosklonny, M.V., 2008. Aging ROS or TOR. Cell Cycle 7, 3344–3354.

Boelsterli, U., 2007. Disruption of mitochondrial function and mitochondria-mediated toxicity. In: Boelsterli, U.A. (Ed.), Mechanistic Toxicology: The Molecular Basis of How Chemicals Disrupt Biological Targets, second ed. CRC Press, Boca Raton, FL, pp. 357–389.

Bonasio, R., Li, Q., Lian, J., Mutti, N.S., Jin, L., Zhao, H., Zhang, P., Wen, P., Xiang, H., Ding, Y., Jin, Z., Shen, S.S., Wang, Z., Wang, W., Wang, J., Berger, S.L., Liebig, J., Zhang, G., Reinberg, D., 2012. Genome-wide and caste-specific DNA methylomes of the ants *Camponotus floridanus* and *Harpegnathos saltator*. Curr. Biol. 22, 1755–1764.

Bordier, C., Suchail, S., Pioz, M., Devaud, J.M., Collet, C., Charreton, M., Le Conte, Y., Alaux, C., 2017. Stress response in honey bees is associated with changes in task-related physiology and energetic metabolism. J. Insect Physiol. 98, 47–54.

Brink, T.C., Demetrius, L., Lehrach, H., Adjaye, J., 2009. Age-related transcriptional changes in gene expression in different organs of mice support the metabolic stability theory of aging. Biogerontology 10, 549–564.

Budovskaya, Y.V., Wu, K., Southworth, L.K., Jiang, M., Tedesco, P., Johnson, T.E., Kim, S.K., 2008. An elt-3/elt-5/elt-6 GATA transcription circuit guides aging in *C. elegans*. Cell 134, 291–303.

Cardoso-Júnior, C.A.M., Guidugli-Lazzarini, K.R., Hartfelder, K., 2018. DNA methylation affects the lifespan of honey bee (*Apis mellifera* L.) workers—evidence for a regulatory module that involves vitellogenin expression but is independent of juvenile hormone function. Insect Biochem. Mol. Biol. 92, 21–29.

Chakrabarti, P., Rana, S., Sarkar, S., Smith, B., Basu, P., 2015. Pesticide-induced oxidative stress in laboratory and field populations of native honey bees along intensive agricultural landscapes in two Eastern Indian states. Apidologie 46, 107–129.

Chakrabarti, P., Carlson, E.A., Lucas, H.M., Melathopoulos, A.P., Sagili, R.R., 2020. Field rates of Sivanto (TM) (flupyradifurone) and Transform (R) (sulfoxaflor) increase oxidative stress and induce apoptosis in honey bees (*Apis mellifera* L.). PLoS One 15, e0233033.

Christen, V., Krebs, J., Fent, K., 2019. Fungicides chlorothanolin, azoxystrobin and folpet induce transcriptional alterations in genes encoding enzymes involved in oxidative phosphorylation and metabolism in honey bees (*Apis mellifera*) at sublethal concentrations. J. Hazard. Mater. 377, 215–226.

Collins, A.M., Williams, V., Evans, J.D., 2004. Sperm storage and antioxidative enzyme expression in the honey bee *Apis mellifera*. Insect Mol. Biol. 13, 141–146.

Corona, M., Robinson, G.E., 2006. Genes of the antioxidant system of the honey bee: annotation and phylogeny. Insect Mol. Biol. 15, 687–701.

Corona, M., Hughes, K.A., Weaver, D.B., Robinson, G.E., 2005. Gene expression patterns associated with queen honey bee longevity. Mech. Ageing Dev. 126, 1230–1238.

Crimmins, E.M., 2015. Lifespan and health span: past, present, and promise. Gerontologist 55, 901–911.

Criscuolo, F., Smith, S., Zahn, S., Heidinger, B.J., Haussmann, M.F., 2018. Experimental manipulation of telomere length: does it reveal a corner-stone role for telomerase in the natural variability of individual fitness? Philos. Trans. R Soc. Lond. B Biol. Sci. 373, 20160440.

Cuozzo, C., Porcellini, A., Angrisano, T., Morano, A., Lee, B., Di Pardo, A., Messina, S., Iuliano, R., Fusco, A., Santillo, M.R., 2007. DNA damage, homology-directed repair, and DNA methylation. PLoS Genet. 3, e110.

de Mattos, I.M., Soares, A.E.E., Tarpy, D.R., 2018. Mitigating effects of pollen during paraquat exposure on gene expression and pathogen prevalence in *Apis mellifera* L. Ecotoxicology 27, 32–44.

De Wilde, J., Beetsma, J., 1982. The physiology of caste development in social insects. Adv. Insect Physiol. 16, 167–246.

Decourtye, A., Armengaud, C., Renou, M., Devillers, J., Cluzeau, S., Gauthier, M., Pham-Delègue, M.-H., 2004. Imidacloprid impairs memory and brain metabolism in the honey bee (*Apis mellifera* L.). Pestic. Biochem. Physiol. 78, 83–92.

Değirmenci, L., Geiger, D., Ferreira, F.R., Keller, A., Krischke, B., Beye, M., Steffan-Dewenter, I., Scheiner, R., 2020. CRISPR/Cas9 mediated mutations as a new tool for studying taste in honey bees. bioRxiv.

Donkena, K.V., Young, C.Y., Tindall, D.J., 2010. Oxidative stress and DNA methylation in prostate cancer. Obstet. Gynecol. Int. 2010, 302051.

Dussaubat, C., Maisonnasse, A., Crauser, D., Tchamitchian, S., Bonnet, M., Cousin, M., Kretzschmar, A., Brunet, J.-L., Le Conte, Y., 2016. Combined neonicotinoid pesticide and parasite stress alter honey bee queens' physiology and survival. Sci. Rep. 6, 31430.

Elango, N., Hunt, B.G., Goodisman, M.A., Soojin, V.Y., 2009. DNA methylation is widespread and associated with differential gene expression in castes of the honey bee, *Apis mellifera*. Proc. Natl. Acad. Sci. U. S. A. 106, 11206–11211.

Elekonich, M.M., 2009. Extreme thermotolerance and behavioral induction of 70-kDa heat shock proteins and their encoding genes in honey bees. Cell Stress Chaperones 14, 219–226.

Elsik, C.G., Worley, K.C., Bennett, A.K., Beye, M., Camara, F., Childers, C.P., de Graaf, D.C., Debyser, G., Deng, J., Devreese, B., Elhaik, E., Evans, J.D., Foster, L.J., Graur, D., Guigo, R., Hoff, K.J., Holder, M.E., Hudson, M.E., Hunt, G.J., Jiang, H., Joshi, V., Khetani, R.S., Kosarev, P., Kovar, C.L., Ma, J., Maleszka, R., Moritz, R.F.A., Munoz-Torres, M.C., Murphy, T.D., Muzny, D.M., Newsham, I.F., Reese, J.T., Robertson, H.M., Robinson, G.E., Rueppell, O., Solovyev, V., Stanke, M., Stolle, E., Tsuruda, J.M., Van Vaerenbergh, M., Waterhouse, R.M., Weaver, D.B., Whitfield, C.W., Wu, Y., Zdobnov, E.M., Zhang, L., Zhu, D., Gibbs, R.A., On Behalf of Honey Bee Genome Sequencing Consortium, 2014. Finding the missing honey bee genes: lessons learned from a genome upgrade. BMC Genomics 15, 86.

Elsner, D., Meusemann, K., Korb, J., 2018. Longevity and transposon defense, the case of termite reproductives. Proc. Natl. Acad. Sci. U. S. A. 115, 5504–5509.

Fadzilah, N.H., Jaapar, M.F., Jajuli, R., Omar, W.A.W., 2017. Total phenolic content, total flavonoid and antioxidant activity of ethanolic bee pollen extracts from three species of Malaysian stingless bee. J. Apic. Res. 56, 1287996.

Farjan, M., Dmitryjuk, M., Lipinski, Z., Biernat-Lopienska, E., Zoltowska, K., 2012. Supplementation of the honey bee diet with vitamin C: The effect on the antioxidative system of *Apis mellifera carnica* brood at different stages. J. Apic. Res. 51, 263–270.

Favreau, E., Martinez-Ruiz, C., Santiago, L.R., Hammond, R.L., Wurm, Y., 2018. Genes and genomic processes underpinning the social lives of ants. Curr. Opin. Insect Sci. 25, 83–90.

Feder, M.E., Hofmann, G.E., 1999. Heat-shock proteins, molecular chaperones, and the stress response: evolutionary and ecological physiology. Annu. Rev. Physiol. 61, 243–282.
Feldhaar, H., Otti, O., 2020. Pollutants and their interaction with diseases of social Hymenoptera. Insects 11, 11030153.
Finkel, T., Holbrook, N.J., 2000. Oxidants, oxidative stress and the biology of ageing. Nature 408, 239–247.
Flatt, T., Amdam, G.V., Kirkwood, T.B.L., Omholt, S.W., 2013. Life-history evolution and the polyphenic regulation of somatic maintenance and survival. Q. Rev. Biol. 88, 185–218.
Franco, R., Schoneveld, O., Georgakilas, A.G., Panayiotidis, M.I., 2008. Oxidative stress, DNA methylation and carcinogenesis. Cancer Lett. 266, 6–11.
Freitak, D., Bos, N., Stucki, D., Sundström, L., 2014. Inbreeding-related trade-offs in stress resistance in the ant *Formica exsecta*. Biol. Lett. 10, 20140805.
Frisard, M., Ravussin, E., 2006. Energy metabolism and oxidative stress. Endocrine 29, 27–32.
Gauthier, M., Aras, P., Jumarie, C., Boily, M., 2016. Low dietary levels of Al, Pb and Cd may affect the non-enzymatic antioxidant capacity in caged honey bees (*Apis mellifera*). Chemosphere 144, 848–854.
Gauthier, M., Aras, P., Paquin, J., Boily, M., 2018. Chronic exposure to imidacloprid or thiamethoxam neonicotinoid causes oxidative damages and alters carotenoid-retinoid levels in caged honey bees (*Apis mellifera*). Sci. Rep. 8, 16274.
Gems, D., Partridge, L., 2013. Genetics of longevity in model organisms: debates and paradigm shifts. Annu. Rev. Physiol. 75, 621–644.
Giraldo, Y.M., Traniello, J.F.A., 2014. Worker senescence and the sociobiology of aging in ants. Behav. Ecol. Sociobiol. 68, 1901–1919.
Goulson, D., Nicholls, E., Botias, C., Rotheray, E.L., 2015. Bee declines driven by combined stress from parasites, pesticides, and lack of flowers. Science 347, 1255957.
Grozinger, C.M., Fan, Y.L., Hoover, S.E.R., Winston, M.L., 2007. Genome-wide analysis reveals differences in brain gene expression patterns associated with caste and reproductive status in honey bees (*Apis mellifera*). Mol. Ecol. 16, 4837–4848.
Gstöttl, C., Stoldt, M., Jongepier, E., Bornberg-Bauer, E., Feldmeyer, B., Heinze, J., Foitzik, S., 2020. Comparative analyses of caste, sex, and developmental stage-specific transcriptomes in two *Temnothorax* ants. Ecol. Evol. 10, 4193–4203.
Haddad, L.S., Kelbert, L., Hulbert, A.J., 2007. Extended longevity of queen honey bees compared to workers is associated with peroxidation-resistant membranes. Exp. Gerontol. 42, 601–609.
Hall, M.E., Nasir, L., Daunt, F., Gault, E.A., Croxall, J.P., Wanless, S., Monaghan, P., 2004. Telomere loss in relation to age and early environment in long-lived birds. Proc. R. Soc. B Biol. Sci. 271, 1571–1576.
Han, W., Yang, Y., Gao, J., Zhao, D., Ren, C., Wang, S., Zhao, S., Zhong, Y., 2019. Chronic toxicity and biochemical response of *Apis cerana cerana* (Hymenoptera: Apidae) exposed to acetamiprid and propiconazole alone or combined. Ecotoxicology 28, 399–411.
Harley, C.B., Futcher, A.B., Greider, C.W., 1990. Telomeres shorten during aging of human fibroblasts. Nature 345, 458–460.
Harries, L.W., Hernandez, D., Henley, W., Wood, A.R., Holly, A.C., Bradley-Smith, R.M., Yaghootkar, H., Dutta, A., Murray, A., Frayling, T.M., 2011. Human aging is characterized by focused changes in gene expression and deregulation of alternative splicing. Aging Cell 10, 868–878.
Harrison, J., Frazier, M.R., Henry, J.R., Kaiser, A., Klok, C.J., Rascon, B., 2006. Responses of terrestrial insects to hypoxia or hyperoxia. Respir. Physiol. Neurobiol. 154, 4–17.

Hartfelder, K., Engels, W., 1998. Social insect polymorphism: hormonal regulation of plasticity in development and reproduction in the honey bee. Curr. Top. Dev. Biol. 40, 45–77.
Hedges, C.P., Wilkinson, R.T., Devaux, J.B., Hickey, A.J., 2019. Hymenoptera flight muscle mitochondrial function: Increasing metabolic power increases oxidative stress. Comp. Biochem. Physiol. A Mol. Integr. Physiol. 230, 115–121.
Heinze, J., Schrempf, A., 2008. Aging and reproduction in social insects—a mini-review. Gerontology 54, 160–167.
Helmer, S.H., Kerbaol, A., Aras, P., Jumarie, C., Boily, M., 2015. Effects of realistic doses of atrazine, metolachlor, and glyphosate on lipid peroxidation and diet-derived antioxidants in caged honey bees (*Apis mellifera*). Environ. Sci. Pollut. Res. 22, 8010–8021.
Herb, B.R., Wolschin, F., Hansen, K.D., Aryee, M.J., Langmead, B., Irizarry, R., Amdam, G.V., Feinberg, A.P., 2012. Reversible switching between epigenetic states in honey bee behavioral subcastes. Nat. Neurosci. 15, 1371–1373.
Hölldobler, B., Wilson, E.O., 2009. The Superorganism: The Beauty, Elegance, and Strangeness of Insect Societies. WW Norton & Company.
Hsieh, Y.-S., Hsu, C.-Y., 2013. Oxidative stress and anti-oxidant enzyme activities in the trophocytes and fat cells of queen honey bees (*Apis mellifera*). Rejuvenation Res. 16, 295–303.
Hsu, C.-Y., Hsieh, Y.-S., 2014. Oxidative stress decreases in the trophocytes and fat cells of worker honey bees during aging. Biogerontology 15, 129–137.
Huang, Z., 2012. Pollen nutrition affects honey bee stress resistance. Terr. Arthropod Rev. 5, 175–189.
Jemielity, S., Kimura, M., Parker, K.M., Parker, J.D., Cao, X., Aviv, A., Keller, L., 2007. Short telomeres in short-lived males: what are the molecular and evolutionary causes? Aging Cell 6, 225–233.
Ji, L.L., Leeuwenburgh, C., Leichtweis, S., Gore, M., Fiebig, R., Hollander, J., Bejma, J., 1998. Oxidative stress and aging: role of exercise and its influences on antioxidant systems. Ann. N. Y. Acad. Sci. 854, 102–117.
Jia, H.H., Sun, R.J., Shi, W.N., Yan, Y., Li, H., Guo, X.Q., Xu, B.H., 2014. Characterization of a mitochondrial manganese superoxide dismutase gene from *Apis cerana cerana* and its role in oxidative stress. J. Insect Physiol. 60, 68–79.
Jirtle, R.L., Skinner, M.K., 2007. Environmental epigenomics and disease susceptibility. Nat. Rev. Gen. 8, 253–262.
Kapheim, K.M., Pan, H.L., Li, C., Salzberg, S.L., Puiu, D., Magoc, T., Robertson, H.M., Hudson, M.E., Venkat, A., Fischman, B.J., Hernandez, A., Yandell, M., Ence, D., Holt, C., Yocum, G.D., Kemp, W.P., Bosch, J., Waterhouse, R.M., Zdobnov, E.M., Stolle, E., Kraus, F.B., Helbing, S., Moritz, R.F.A., Glastad, K.M., Hunt, B.G., Goodisman, M.A.D., Hauser, F., Grimmelikhuijzen, C.J.P., Pinheiro, D.G., Nunes, F.M.F., Soares, M.P.M., Tanaka, E.D., Simoes, Z.L.P., Hartfelder, K., Evans, J.D., Barribeau, S.M., Johnson, R.M., Massey, J.H., Southey, B.R., Hasselmann, M., Hamacher, D., Biewer, M., Kent, C.F., Zayed, A., Blatti, C., Sinha, S., Johnston, J.S., Hanrahan, S.J., Kocher, S.D., Wang, J., Robinson, G.E., Zhang, G.J., 2015. Genomic signatures of evolutionary transitions from solitary to group living. Science 348, 1139–1143.
Kefuss, J.A., Nye, W.P., 1970. The influence of photoperiod on the flight activity of honey bees. J. Apic. Res. 9, 133–139.
Keller, L., Jemielity, S., 2006. Social insects as a model to study the molecular basis of ageing. Exp. Gerontol. 41, 553–556.
Kern, M., Wegener, G., 1984. Age affects the metabolic-rate of insect brain. Mech. Ageing Dev. 28, 237–242.
Kirkwood, T.B., 1977. Evolution of ageing. Nature 270, 301–304.

Kohlmeier, P., Alleman, A.R., Libbrecht, R., Foitzik, S., Feldmeyer, B., 2019. Gene expression is more strongly associated with behavioural specialization than with age or fertility in ant workers. Mol. Ecol. 28, 658–670.

Kohno, H., Suenami, S., Takeuchi, H., Sasaki, T., Kubo, T., 2016. Production of knockout mutants by CRISPR/Cas9 in the European honey bee, *Apis mellifera* L. Zoolog. Sci. 33, 505–513.

Kojić, D., Purać, J., Nikolić, Č., Orčić, D., Vujanović, D., Iljević, K., Vukašinović, E., Blagojević, D., 2019. Oxidative stress and the activity of antioxidative defense enzymes in overwintering honey bees. Entomol. Gen. 39, 33–44.

Korandova, M., Frydrychova, R.C., 2016. Activity of telomerase and telomeric length in *Apis mellifera*. Chromosoma 125, 405–411.

Koubova, J., Jehlik, T., Kodrik, D., Sabova, M., Sima, P., Sehadova, H., Zavodska, R., Frydrychova, R.C., 2019. Telomerase activity is upregulated in the fat bodies of pre-diapause bumblebee queens (*Bombus terrestris*). Insect Biochem. Mol. Biol. 115, 103241.

Kraft, N., Spaethe, J., Rossler, W., Groh, C., 2019. Neuronal plasticity in the mushroom-body calyx of bumble bee workers during early adult development. Dev. Neurobiol. 79, 287–302.

Kramer, B.H., van Doorn, G.S., Weissing, F.J., Pen, I., 2016. Lifespan divergence between social insect castes: challenges and opportunities for evolutionary theories of aging. Curr. Opin. Insect Sci. 16, 76–80.

Kuhn, J.M.M., Korb, J., 2016. Editorial overview: social insects: aging and the re-shaping of the fecundity/longevity trade-off with sociality. Curr. Opin. Insect Sci. 16, vii–x.

Kuhn, J.M.M., Meusemann, K., Korb, J., 2019. Long live the queen, the king and the commoner? Transcript expression differences between old and young in the termite *Cryptotermes secundus*. PLoS One 14, e0210371.

Lennicke, C., Cocheme, H.M., 2020. Redox signalling and ageing: insights from *Drosophila*. Biochem. Soc. Trans. 48, 366–376.

Leza, M., Herrera, C., Marques, A., Roca, P., Sastre-Serra, J., Pons, D., 2019. The impact of the invasive species *Vespa velutina* on honey bees: a new approach based on oxidative stress. Sci. Total Environ. 689, 709–715.

Li, H.M., Buczkowski, G., Mittapalli, O., Xie, J., Wu, J., Westerman, R., Schemerhorn, B.J., Murdock, L.L., Pittendrigh, B.R., 2008. Transcriptomic profiles of *Drosophila melanogaster* third instar larval midgut and responses to oxidative stress. Insect Mol. Biol. 17, 325–339.

Li, H.M., Sun, L., Mittapalli, O., Muir, W.M., Xie, J., Wu, J., Schemerhorn, B.J., Sun, W., Pittendrigh, B.R., Murdock, L.L., 2009. Transcriptional signatures in response to wheat germ agglutinin and starvation in *Drosophila melanogaster* larval midgut. Insect Mol. Biol. 18, 21–31.

Li, C.C., Xu, B.H., Wang, Y.X., Feng, Q.Q., Yang, W.R., 2012. Effects of dietary crude protein levels on development, antioxidant status, and total midgut protease activity of honey bee (*Apis mellifera ligustica*). Apidologie 43, 576–586.

Li, Y.Y., Zhang, R., Liu, S.L., Donath, A., Peters, R.S., Ware, J., Misof, B., Niehuis, O., Pfrender, M.E., Zhou, X., 2017. The molecular evolutionary dynamics of oxidative phosphorylation (OXPHOS) genes in Hymenoptera. BMC Evol. Biol. 17, 269.

Li, X., Ma, W., Shen, J., Long, D., Feng, Y., Su, W., Xu, K., Du, Y., Jiang, Y., 2019. Tolerance and response of two honey bee species *Apis cerana* and *Apis mellifera* to high temperature and relative humidity. PLoS One 14, e0217921.

Li-Byarlay, H., 2016. The function of DNA methylation marks in social insects. Front. Ecol. Evol. 4, 57.

Li-Byarlay, H., Li, Y., Stroud, H., Feng, S., Newman, T.C., Hou, K.K., Kaneda, M., Wickline, S.A., Jacobsen, S.E., Ma, J., Robinson, G.E., 2013. RNA interference knockdown of DNA methyl-transferase 3 affects gene alternative splicing in the honey bee. Proc. Natl. Acad. Sci. U. S. A. 110, 12750.

Li-Byarlay, H., Rittschof, C.C., Massey, J.H., Pittendrigh, B.R., Robinson, G.E., 2014. Socially responsive effects of brain oxidative metabolism on aggression. Proc. Natl. Acad. Sci. U. S. A. 111, 12533–12537.
Li-Byarlay, H., Huang, M.H., Simone-Finstrom, M., Strand, M.K., Tarpy, D.R., Rueppell, O., 2016. Honey bee (*Apis mellifera*) drones survive oxidative stress due to increased tolerance instead of avoidance or repair of oxidative damage. Exp. Gerontol. 83, 15–21.
Lighton, J.R.B., Garrigan, D., 1995. Ant breathing–testing regulation and mechanism hypotheses with hypoxia. J. Exp. Biol. 198, 1613–1620.
Lingner, J., Cooper, J.P., Cech, T.R., 1995. Telomerase and DNA end replication—no longer a lagging-strand problem. Science 269, 1533–1534.
Lipiński, Z., Żółtowska, K., 2005. Preliminary evidence associating oxidative stress in honey bee drone brood with *Varroa destructor*. J. Apic. Res. 44, 126–128.
Liu, S.C., Liu, F., Jia, H.H., Yan, Y., Wang, H.F., Guo, X.Q., Xu, B.H., 2016. A glutathione S-transferase gene associated with antioxidant properties isolated from *Apis cerana cerana*. Sci. Nat. 103, 43.
Lopez-Otin, C., Blasco, M.A., Partridge, L., Serrano, M., Kroemer, G., 2013. The hallmarks of aging. Cell 153, 1194–1217.
Lopez-Uribe, M.M., Ricigliano, V.A., Simone-Finstrom, M., 2020. Defining pollinator health: a holistic approach based on ecological, genetic, and physiological factors. Annu. Rev. Anim. Biosci. 8, 269–294.
Lucas, E.R., Keller, L., 2020. The co-evolution of longevity and social life. Funct. Ecol. 34, 76–87.
Lucas, E.R., Privman, E., Keller, L., 2016. Higher expression of somatic repair genes in long-lived ant queens than workers. Aging 8, 1940.
Ma, C., Zhang, Y., Sun, J., Imran, M., Yang, H., Wu, J., Zou, Y., Li-Byarlay, H., Luo, S., 2019. Impact of acute oral exposure to thiamethoxam on the homing, flight, learning acquisition and short-term retention of *Apis cerana*. Pest Manag. Sci. 75, 2975–2980.
Margotta, J.W., Mancinelli, G.E., Benito, A.A., Ammons, A., Roberts, S.P., Elekonich, M.M., 2012. Effects of flight on gene expression and aging in the honey bee brain and flight muscle. Insects 4, 9–30.
Margotta, J.W., Roberts, S.P., Elekonich, M.M., 2018. Effects of flight activity and age on oxidative damage in the honey bee, *Apis mellifera*. J. Exp. Biol. 221, 183228.
McAfee, A., Chapman, A., Higo, H., Underwood, R., Milone, J., Foster, L.J., Guarna, M.M., Tarpy, D.R., Pettis, J.S., 2020. Vulnerability of honey bee queens to heat-induced loss of fertility. Nat. Sustain. 3, 367–376.
Medawar, P.B., 1952. An Unsolved Problem of Biology. H.K. Lewis, London.
Metcalfe, N.B., Alonso-Alvarez, C., 2010. Oxidative stress as a life-history constraint: the role of reactive oxygen species in shaping phenotypes from conception to death. Funct. Ecol. 24, 984–996.
Metz, B.N., Tarpy, D.R., 2019. Reproductive senescence in drones of the honey bee (*Apis mellifera*). Insects 10, 10010011.
Michener, C.D., 1974. The Social Behavior of the Bees: A Comparative Study. vol. 73. Harvard University Press.
Morimoto, T., Kojima, Y., Toki, T., Komeda, Y., Yoshiyama, M., Kimura, K., Nirasawa, K., Kadowaki, T., 2011. The habitat disruption induces immune-suppression and oxidative stress in honey bees. Ecol. Evol. 1, 201–217.
Muller, F., 2000. The nature and mechanism of superoxide production by the electron transport chain: its relevance to aging. J. Am. Aging Assoc. 23, 227–253.
Negroni, M.A., Foitzik, S., Feldmeyer, B., 2019. Long-lived *Temnothorax* ant queens switch from investment in immunity to antioxidant production with age. Sci. Rep. 9, 7270.
Neukirch, A., 1982. Dependence of the life-span of the honey bee (*Apis mellifica*) upon flight performance and energy-consumption. J. Comp. Physiol. 146, 35–40.

Neuwirthová, N., Trojan, M., Svobodová, M., Vašíčková, J., Šimek, Z., Hofman, J., Bielská, L., 2019. Pesticide residues remaining in soils from previous growing season (s)-can they accumulate in non-target organisms and contaminate the food web? Sci. Total Environ. 646, 1056–1062.

Orčić, S., Nikolić, T., Purać, J., Šikoparija, B., Blagojević, D.P., Vukašinović, E., Plavša, N., Stevanović, J., Kojić, D., 2017. Seasonal variation in the activity of selected antioxidant enzymes and malondialdehyde level in worker honey bees. Entomol. Exp. Appl. 165, 120–128.

Page, R.E., Gadau, J., Beye, M., 2002. The emergence of hymenopteran genetics. Genetics 160, 375–379.

Paris, L., Roussel, M., Pereira, B., Delbac, F., Diogon, M., 2017. Disruption of oxidative balance in the gut of the western honey bee *Apis mellifera* exposed to the intracellular parasite *Nosema ceranae* and to the insecticide fipronil. J. Microbial. Biotechnol. 10, 1702–1717.

Parker, J.D., 2010. What are social insects telling us about aging? Myrmecol. News 13, 103–110.

Parker, J.D., Parker, K.M., Sohal, B.H., Sohal, R.S., Keller, L., 2004. Decreased expression of Cu–Zn superoxide dismutase 1 in ants with extreme lifespan. Proc. Natl. Acad. Sci. U. S. A. 101, 3486–3489.

Partridge, L., Alic, N., Bjedov, I., Piper, M.D.W., 2011. Ageing in *Drosophila*: The role of the insulin/Igf and TOR signalling network. Exp. Gerontol. 46, 376–381.

Passos, J.F., Von Zglinicki, T., 2006. Oxygen free radicals in cell senescence: are they signal transducers? Free Radic. Res. 40, 1277–1283.

Pearl, R., 1928. The Rate of Living. University Press, London.

Porter, S.D., Jorgensen, C.D., 1981. Foragers of the harvester ant, *Pogonomyrmex owyheei*: a disposable caste? Behav. Ecol. Sociobiol. 9, 247–256.

Qi, S., Zhu, L., Wang, D., Wang, C., Chen, X., Xue, X., Wu, L., 2020. Flumethrin at honey-relevant levels induces physiological stresses to honey bee larvae (*Apis mellifera* L.) in vitro. Ecotoxicol. Environ. Safety 190, 110101.

Quque, M., Benhaim-Delarbre, M., Deneubourg, J.L., Sueur, C., Criscuolo, F., Bertile, F., 2019. Division of labour in the black garden ant (*Lasius niger*) leads to three distinct proteomes. J. Insect Physiol. 117, 103907.

Radyuk, S.N., Klichko, V.I., Spinola, B., Sohal, R.S., Orr, W.C., 2001. The peroxiredoxin gene family in *Drosophila melanogaster*. Free Radic. Biol. Med. 31, 1090–1100.

Rascon, B., Harrison, J.F., 2010. Lifespan and oxidative stress show a non-linear response to atmospheric oxygen in *Drosophila*. J. Exp. Biol. 213, 3441–3448.

Richter, M.R., 2000. Social wasp (Hymenoptera: Vespidae) foraging behavior. Annu. Rev. Entomol. 45, 121–150.

Roat, T.C., dos Santos-Pinto, J.R.A., dos Santos, L.D., Santos, K.S., Malaspina, O., Palma, M.S., 2014. Modification of the brain proteome of Africanized honey bees (*Apis mellifera*) exposed to a sub-lethal doses of the insecticide fipronil. Ecotoxicology 23, 1659–1670.

Robinson, G.E., 1992. Regulation of division of labor in insect societies. Annu. Rev. Entomol. 37, 637–665.

Robinson, G.E., Fernald, R.D., Clayton, D.F., 2008. Genes and social behavior. Science 322, 896–900.

Rothe, U., Nachtigall, W., 1989. Flight of the honey bee. 4. Respiratory quotients and metabolic rates during sitting, walking and flying. J. Comp. Physiol. B Biochem. Sys. Environ. Physiol. 158, 739–749.

Rouibi, A., Bouchema, W.-F., Loucif-Ayad, W., Achou, M., Soltani, N., 2016. Risks assessment of two acaricides (fluvalinate and oxalic acid) in *Apis mellifera intermissa* (Hymenoptera, Apidae): acetylcholinesterase and glutathione S-transferase activities. J. Entomol. Zool. Stud. 4, 503–508.

Rueppell, O., 2009. Aging of social insects. In: Gadau, J., Fewell, J. (Eds.), Organization of Insect Societies. Harvard University Press, Cambridge, pp. 51–73.
Rueppell, O., Fondrk, M.K., Page Jr., R.E., 2005. Biodemographic analysis of male honey bee mortality. Aging Cell 4, 13–19.
Sacktor, B., 1976. Biochemical adaptations for flight in the insect. Biochem. Soc. Symp. 41, 111–131.
Salmela, H., Stark, T., Stucki, D., Fuchs, S., Freitak, D., Dey, A., Kent, C.F., Zayed, A., Dhaygude, K., Hokkanen, H., 2016. Ancient duplications have led to functional divergence of vitellogenin-like genes potentially involved in inflammation and oxidative stress in honey bees. Genome Biol. Evol. 8, 495–506.
Schieber, M., Chandel, N.S., 2014. ROS function in redox signaling and oxidative stress. Curr. Biol. 24, R453–R462.
Schippers, M.-P., Dukas, R., Smith, R., Wang, J., Smolen, K., McClelland, G., 2006. Lifetime performance in foraging honey bees: behaviour and physiology. J. Exp. Biol. 209, 3828–3836.
Schmidt, T.R., Wu, W., Goodman, M., Grossman, L.I., 2001. Evolution of nuclear- and mitochondrial-encoded subunit interaction in cytochrome C oxidase. Mol. Biol. Evol. 18, 563–569.
Seehuus, S.-C., Krekling, T., Amdam, G.V., 2006a. Cellular senescence in honey bee brain is largely independent of chronological age. Exp. Gerontol. 41, 1117–1125.
Seehuus, S.-C., Norberg, K., Gimsa, U., Krekling, T., Amdam, G.V., 2006b. Reproductive protein protects functionally sterile honey bee workers from oxidative stress. Proc. Natl. Acad. Sci. U. S. A. 103, 962–967.
Seeley, T., Mikheyev, A., 2003. Reproductive decisions by honey bee colonies: tuning investment in male production in relation to success in energy acquisition. Insectes Soc. 50, 134–138.
Seid, M.A., Harris, K.M., Traniello, J.F.A., 2005. Age-related changes in the number and structure of synapses in the lip region of the mushroom bodies in the ant *Pheidole dentata*. J. Comp. Neurol. 488, 269–277.
Seid, M.A., Goode, K., Li, C., Traniello, J.F.A., 2008. Age- and subcaste-related patterns of serotonergic immunoreactivity in the optic lobes of the ant *Pheidole dentata*. Dev. Neurobiol. 68, 1325–1333.
Shadel, G.S., Horvath, T.L., 2015. Mitochondrial ROS signaling in organismal homeostasis. Cell 163, 560–569.
Shafiqur, R., Rehman, S., Waliullah, M.I.S., 2012. Chlorpyrifos-induced neuro-oxidative damage in bee. Toxicol. Environ. Health Sci. 4, 30–36.
Shames, D.S., Minna, J.D., Gazdar, A.F., 2007. DNA methylation in health, disease, and cancer. Curr. Mol. Med. 7, 85–102.
Siebold, A.P., Banerjee, R., Tie, F., Kiss, D.L., Moskowitz, J., Harte, P.J., 2010. Polycomb repressive complex 2 and trithorax modulate *Drosophila* longevity and stress resistance. Proc. Natl. Acad. Sci. U. S. A. 107, 169–174.
Simola, D.F., Ye, C., Mutti, N.S., Dolezal, K., Bonasio, R., Liebig, J., Reinberg, D., Berger, S.L., 2013. A chromatin link to caste identity in the carpenter ant *Camponotus floridanus*. Genome Res. 23, 486–496.
Simone-Finstrom, M., Li-Byarlay, H., Huang, M.H., Strand, M.K., Rueppell, O., Tarpy, D.R., 2016. Migratory management and environmental conditions affect lifespan and oxidative stress in honey bees. Sci. Rep. 6, 32023.
Sohal, R.S., Weindruch, R., 1996. Oxidative stress, caloric restriction, and aging. Science 273, 59–63.
Strycharz, J.P., Lao, A., Li, H., Qiu, X., Lee, S.H., Sun, W., Yoon, K.S., Doherty, J.J., Pittendrigh, B.R., Clark, J.M., 2013. Resistance in the highly DDT-resistant 91-R strain of *Drosophila melanogaster* involves decreased penetration, increased metabolism, and direct excretion. Pestic. Biochem. Physiol. 107, 207–217.

Stucki, D., Sundstrom, L., Freitak, D., 2017. Caste-specific expression of constitutive and *Beauveria bassiana* induced immunity in the ant *Formica exsecta* (Hymenoptera: Formicidae). Myrmecol. News 25, 83–93.

Suarez, R.K., Staples, J.F., Lighton, J.R.B., Mathieu-Costello, O., 2000. Mitochondrial function in flying honey bees (*Apis mellifera*): respiratory chain enzymes and electron flow from complex III to oxygen. J. Exp. Biol. 203, 905–911.

Sullivan, J.P., Fahrbach, S.E., Harrison, J.F., Capaldi, E.A., Fewell, J.H., Robinson, G.E., 2003. Juvenile hormone and division of labor in honey bee colonies effects of allatectomy on flight behavior and metabolism. J. Exp. Biol. 206, 2287–2296.

Taric, E., Glavinic, U., Vejnovic, B., Stanojkovic, A., Aleksic, N., Dimitrijevic, V., Stanimirovic, Z., 2020. Oxidative stress, endoparasite prevalence and social immunity in bee colonies kept traditionally vs. those kept for commercial purposes. Insects 11, 266.

Tatar, M., Bartke, A., Antebi, A., 2003. The endocrine regulation of aging by insulin-like signals. Science 299, 1346–1351.

Terman, A., Brunk, U.T., 2006. Oxidative stress, accumulation of biological 'garbage', and aging. Antioxid. Redox Signal. 8, 197–204.

Tollervey, J.R., Wang, Z., Hortobágyi, T., Witten, J.T., Zarnack, K., Kayikci, M., Clark, T.A., Schweitzer, A.C., Rot, G., Curk, T., 2011. Analysis of alternative splicing associated with aging and neurodegeneration in the human brain. Genome Res. 21, 1572–1582.

Torson, A.S., Yocum, G.D., Rinehart, J.P., Nash, S.A., Bowsher, J.H., 2019. Fluctuating thermal regimes prevent chill injury but do not change patterns of oxidative stress in the alfalfa leafcutting bee, *Megachile rotundata*. J. Insect Physiol. 118, 103935.

Trible, W., Olivos-Cisneros, L., McKenzie, S.K., Saragosti, J., Chang, N.-C., Matthews, B.J., Oxley, P.R., Kronauer, D.J., 2017. Orco mutagenesis causes loss of antennal lobe glomeruli and impaired social behavior in ants. Cell 170, 727–735.

Vieira, A.S., Bueno, O.C., 2015. Mitochondrial and peroxisomal population in post-pharyngeal glands of leaf-cutting ants after lipid supplementation. Micron 68, 8–16.

Weinhold, L., Ahmad, S., Pardini, R., 1990. Insect glutathione-S-transferase: a predictor of allelochemical and oxidative stress. Comp. Biochem. Physiol. B Comp. Biochem. 95, 355–363.

Weirich, G.F., Collins, A.M., Williams, V.P., 2002. Antioxidant enzymes in the honey bee, *Apis mellifera*. Apidologie 33, 3–14.

Williams, G.C., 1957. Pleiotropy, natural selection, and the evolution of senescence. Evolution 11, 398–411.

Williams, J.R., 2016. Biomarkers of Oxidative Stress in Atrazine-Treated Honey Bees: A Laboratory and In-Hive study. Thesis, Virginia Tech.

Williams, J.B., Roberts, S.P., Elekonich, M.M., 2008. Age and natural metabolically-intensive behavior affect oxidative stress and antioxidant mechanisms. Exp. Gerontol. 43, 538–549.

Wilson, E.O., 1979. The evolution of caste systems in social insects. Proc. Am. Philos. Soc. 123, 204–210.

Winston, M.L., 1987. The Biology of the Honey Bee. Harvard University Press, Cambridge, MA.

Wolffe, A.P., Matzke, M.A., 1999. Epigenetics: regulation through repression. Science 286, 481–486.

Yan, H.R., Meng, F., Jia, H.H., Guo, X.Q., Xu, B.H., 2012. The identification and oxidative stress response of a zeta class glutathione S-transferase (GSTZ1) gene from *Apis cerana cerana*. J. Insect Physiol. 58, 782–791.

Yan, H., Bonasio, R., Simola, D.F., Liebig, J., Berger, S.L., Reinberg, D., 2015. DNA methylation in social insects: how epigenetics can control behavior and longevity. Annu. Rev. Entomol. 60, 435–452.

Youngsteadt, E., Appler, R.H., Lopez-Uribe, M.M., Tarpy, D.R., Frank, S.D., 2015. Urbanization increases pathogen pressure on feral and managed honey bees. PLoS One 10, e0142031.

Zhang, W.X., Liu, Z.G., Zhu, M., Ma, L.T., Wang, Y., Wang, H.F., Guo, X.Q., Xu, B.H., 2017a. Molecular cloning, expression and oxidative stress response of the vitellogenin gene (AccVg) from *Apis cerana cerana*. Apidologie 48, 599–611.

Zhang, Y.W., Wang, Z., Xie, W., Cai, Y., Xia, L., Easwaran, H., Luo, J., Yen, R.-W.C., Li, Y., Baylin, S.B., 2017b. Acetylation enhances TET2 function in protecting against abnormal DNA methylation during oxidative stress. Mol. Cell 65, 323–335.

Zhao, W.C., Chao, Y.Z., Wang, Y., Wang, L.J., Wang, X.X., Li, H., Xu, B.H., 2019. Role of AccMGST1 in oxidative stress resistance in *Apis cerana cerana*. Cell Stress Chaperones 24, 793–805.