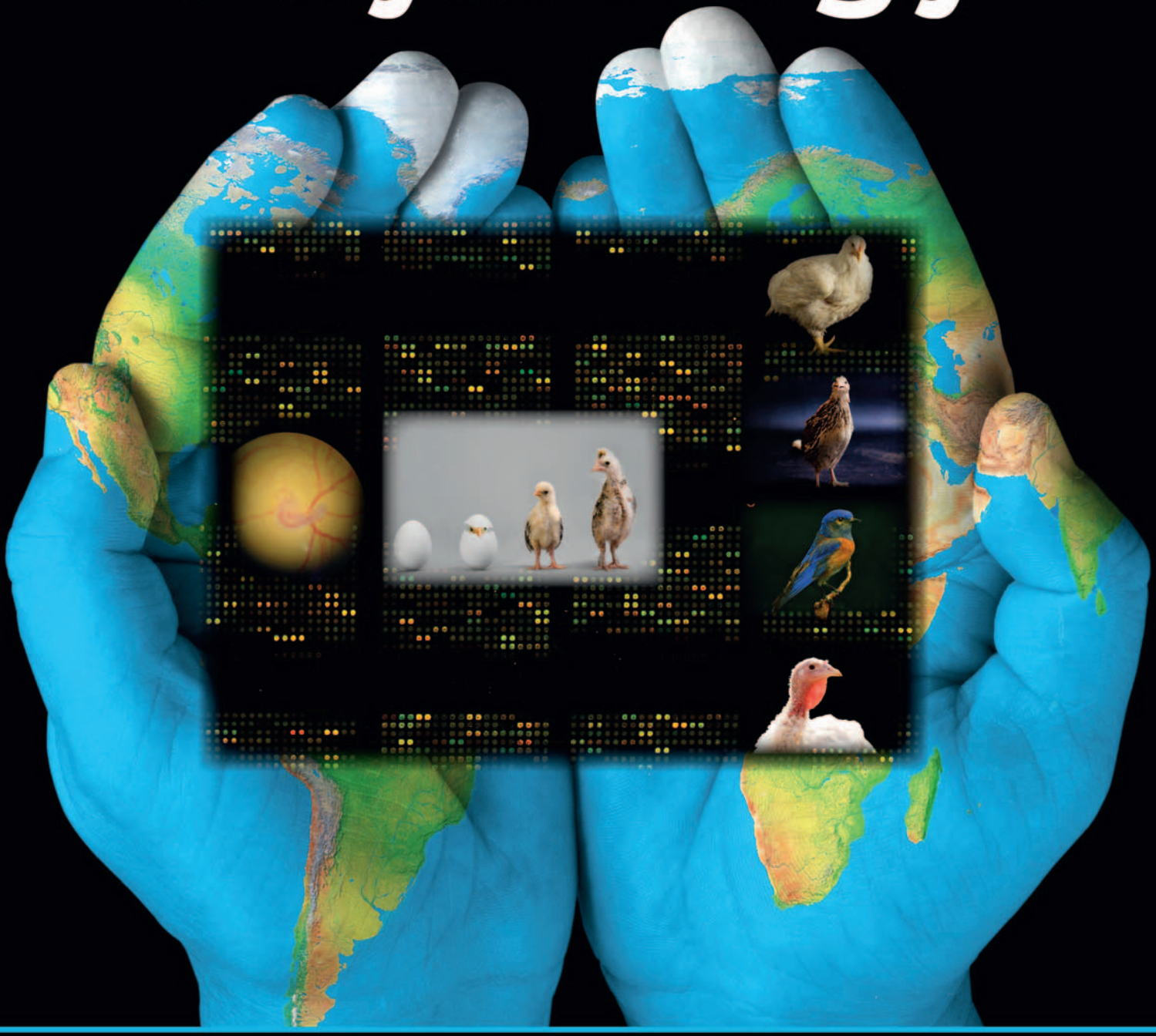
Sturkie's Avian Physiology

Seventh Edition



Edited by

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Chapter 47

Physiological challenges of migration

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Abbreviations

ACTH Adrenocorticotropic hormone
AGRP Agouti-related peptide
AR Androgen receptor
AVT Arginine vasotocin
C-fos Fos Proto-Oncogene, an early immediate gene
CART Cocaine - and amphetamine-regulated transcript protein
CCK Cholecystokinin
CCO Cytochrome-c oxidase
CK Creatine kinase
CPT Carnitine palmitoyltransferase
CS Citrate synthase
DHT Dihydrotestosterone
DIO2 Type II iodothyronine deiodinase
DIO3 Type III iodothyronine deiodinase
ELHS Emergency life history stage
FFA Free fatty acids
FSH Follicle-stimulating hormone
GH Growth Hormone
GnIH Gonadotropin-inhibitory hormone
GnRH Gonadotropin-releasing hormone
GR Glucocorticoid receptor
GtH Gonadotropin
HOAD Hydroxyacyl CoA dehydrogenase
HPA Hypothalamic-pituitary-adrenal axis
HPG Hypothalamic-pituitary-gonad axis
ICV Intracerebroventricular
IGF-1 Insulin-like growth factor-1
IP Intraperitoneal
LH Luteinizing hormone
MDH Malate dehydrogenase
MR Mineralocorticoid receptor
MSH Melanocyte-stimulating hormone
NPY Neuropeptide Y
PFK Phosphofructokinase
PK Pyruvate kinase
POMC Proopiomelanocortin
PPAR Perixisome proliferator-activated nuclear receptors
PSM Plant secondary metabolites
PUFA Polyunsaturated fatty acids
PRL Prolactin
T Testosterone
T3 Triiodothyronine
T4 Thyroxine
TSH Thyroid-stimulating hormone
VA opsin Vertebrate ancient opsin photopigment

47.1 Introduction

Approximately 19% of the ~10,000 species of birds in the world migrate on a regular basis (Kirby et al., 2008), and this is closer to 50% of species if only birds that breed at latitudes above 35° north and south are considered (Newton and Dale, 1996a, 1996b). The physiological feat of migration has been equated to such athletic feats in mammals as running 130 consecutive marathons (semipalmated sandpiper, *Calidris pusilla,* migrates ~ 5000−7500 km nonstop; Anderson et al., 2019) or completing three trips to the moon and back in a lifetime (arctic tern, *Sterna paradisaea,* migrates ~80,000 km a year over a 30 year lifespan; Egevang et al., 2010). The strategies migratory birds use to complete these annual peregrinations vary among bird species with some birds completing their long-distance migration in one, nonstop flight while most fly intermittently, taking advantage of stopover sites along their routes to rest and refuel. Regardless of strategy, all avian migrants face the same physiological challenges while aloft, albeit to different extents, including: (1) burn fat at a very high rate while fasting and combatting the negative effects of lipid peroxidation, (2) maintain adequate hydration while not drinking and losing water due to elevated breathing rates, (3) obtain adequate oxygen while oxidizing fat for fuel, and especially when flying at high altitudes with low-oxygen content, (4) avoid becoming too hot (heat gained from metabolism and the environment) or too cold (heat loss to the environment), and (5) meeting these challenges under a seasonal time constraint. Additionally, birds that stop to rest

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and refuel at stopover sites have the added physiological challenge of rapidly regaining energy stores with initially reduced gut size and function. Clearly, the physiological challenges associated with long-distance flight are significant and require specialized morphological and physiological adaptations and acclimatizations.

The energy costs of bird flight are substantial and have been the focus of ornithological research since the 1960's when Greenewalt (1960) recorded hummingbirds flying in front of a fan and examined changes in flight speed with changes in fan speeds. The ability of researchers to fly birds in a controlled environment led to significant breakthroughs not only in the field of aerodynamics but in the understanding of the physiology of flight as well. For example, classic studies of birds flying in wind-tunnels have shown that forward flapping flight is energetically more costly than running or swimming per gram body mass (Butler, 1991; Bishop and Butler, 2015). Importantly, the advantage of flying on migration, instead of running and swimming, is revealed when travel distance is considered. Specifically, energy required per gram body mass to travel a given distance (i.e., the cost of transport) is considerably less for a flying migrant compared to a running or swimming migrant (Tucker, 1970; Schmidt-Nielsen, 1972; Bishop and Butler, 2015). With an increase in technology and the ability to track migrating birds throughout the globe, we now know the physiology of migratory flights of free-living birds to be a highly complex, synergistic process between an organism and its ever-changing environment.

In this chapter, we will review, within the context of the migration life history of birds, the neuroendocrine and physiological adaptations that allow birds to meet the challenges of long-duration flights, the preparations they implement to begin their journeys, and the in-flight adjustments they make in order to successfully complete their journeys. We will conclude with a discussion about the flexible phenotypes of birds in relation to how they physiologically respond to the challenges of migratory flight in a dynamic seasonal environment, and some of the important mysteries that remain in terms of the physiology of birds during migration.

47.2 Adaptations of birds for long-duration migratory flights

Flapping flight requires both lift to stay aloft and thrust to move forward through the air (Tobalske, 2016). Birds have evolved feathers and wings that enable them to fly because their collective shape produces lift as air flows over and around them. Combine feathers and wings with a skeletal system that is both strong yet light (hollow bones), and a muscular system that provides the power and endurance, and you have the anatomical skeletal-muscular foundation for the evolution of long-distance migration of birds. Bishop and Butler (2015) provide a thorough description of the biomechanics of bird flight, and how the muscles of birds produce the power required for flight, in general, and the long-duration flapping flight used by birds in migration, in particular. Most pertinent to our review of the physiology of long-distance migration in birds is that (a) self-powered flight is the most demanding form of locomotion in terms of its energy cost per unit time—thus, fuel storage and use is critical; (b) on the other hand, flight is much more efficient than running or swimming. In short, the cost of transport for a flyer (in units of energy required for a bird of a given mass to travel a given distance) is appreciably less than that of a runner or a swimmer—thus, overcoming the fuel storage and use challenges of long-duration flapping flight allows migratory birds to travel far for their size.

47.2.1 Cardiovascular and respiratory general adaptations

The energetic costs of flapping flight have driven adaptive responses in the cardiovascular and respiratory systems of birds. In general, birds have larger hearts, lower resting heart rates, higher exercising heart rates, and higher $\dot{V}O_2$ max than nonflying mammals of similar size (Bishop and Butler, 2015). Adaptations in the vascular system of birds such as increased number of capillaries in each muscle fiber along with mitochondria that are in closer proximity to capillaries allows for efficient exchange of respiratory gasses (Powell, 2015). In addition, the structure of the avian respiratory system contributes to a bird's ability to efficiently exchange large volumes of gas as is needed during flight. Taken together the design of air sacs increases ventilation, the increased surface area in the lungs, unidirectional flow across the respiratory surface, and increased cross-current gas exchange with the blood all contribute to a bird's ability to obtain oxygen, supply oxygen to active tissue, and remove carbon dioxide from the blood at a greater rate than mammals of similar size (Dzialowski and Crossley, 2015; Scott et al., 2015). In general, birds are also more tolerant of hypoxia than mammals at least in part because of differences in their sensitivity to hypocapnia (low-partial pressure of arterial blood carbon dioxide (Scott et al., 2015)). Hypocapnia sensitivity of cerebral blood vessels is one of the main mechanisms in which mammals limit hyperventilation, thereby decreasing oxygen delivery to the brain, and likely limiting overall energetic output (Ogoh and Ainslie, 2009; Ainslie and Ogoh, 2010). Migratory bird insensitivity to hypocapnia especially in cerebral blood vessels allows for extreme hyperventilatory rates without blood flow interference to the brain (Faraci, 1991). In turn, this hyperventilation allows for decreased arteriole partial pressure of carbon dioxide subsequently increasing arteriole partial pressure of oxygen due to the Bohr effect (Powell, 2015). Thus, birds are able to maintain

high-oxygen delivery to tissues even in low-oxygen environments such as those at high altitudes. Additional vascular adaptations that enable birds to meet the high-oxygen demands of long-duration flight while retaining essential blood solute concentrations include an overall higher hematocrit level and greater hemoglobin concentrations (Minias et al., 2013; Bishop and Butler, 2015) compared to nonmigratory birds or mammals and the maintenance of plasma levels throughout a long flight regardless of body mass changes (Carmi et al., 1993). Monthly comparison of hematocrit of White-crowned sparrows (*Zonotrichia leucophrys gambelli*) revealed that migrant subspecies were consistently greater than residents, a difference that increased 10−15% further during both seasonal stages of migration (Krause et al., 2016). Interestingly, recent experiments that directly manipulated hematocrit in short-distance migratory Yellow-rumped warblers (*Setophaga coronata*) found that birds with elevated hematocrit performed better at simulated high altitudes (in a wind tunnel) but not at lower altitudes (Yap et al., 2018); many more such manipulative experiments are needed to better understand how variation in hematocrit affects migratory flight performance.

47.2.2 Metabolism and damage control

Mitochondria generate the cellular energy required by birds (and all animals) to fuel their basal metabolism as well as that required for all activity including flapping flight (Bottje, 2015). Associated with this role as powerhouse of the cell, mitochondria are also the major site for generation of reactive species (RS)—thus an effective antioxidant protection system has evolved to reduce or avoid the negative physiological effects of RS production, the components of which are shared by all eukaryotes (Bottje, 2015; Cooper-Mullin and McWilliams, 2016; Skrip and McWilliams, 2016). Most pertinent to our review of the physiology of long-distance migration in birds is that (a) increased metabolism, like that required for long-duration flapping flight, is associated with increased RS production which may overwhelm the antioxidant system and cause oxidative imbalance or "stress" and thus damage to, for example, lipids, proteins, and DNA in cells; (b) repair of damaged structures is energetically expensive—thus, understanding how the antioxidant system of migrating birds contends with RS production is as important as understanding how the power for long-duration flight is generated.

How birds overcome the potential constraints of redox imbalance (so-called "oxidative stress") associated with their relatively high baseline and active metabolic rates has been the subject of much conjecture (Buttemer et al., 2010; Munshi-South and Wilkinson, 2010; Jimenez et al., 2019), although recently a key adaptation of the master antioxidant response of birds has been revealed (Castiglione et al., 2020). The transcription factor NF-E2-related factor 2 (NRF2) is the major cellular pathway for regulating the antioxidant response of metazoans against oxidative imbalance within cells. Under steady-state conditions, KEAP1 binds NRF2 and keeps it at low levels in the cell. Under conditions of increased metabolic rate and thus RS production, KEAP1 repression is relaxed allowing NRF2 to accumulate, bind to an antioxidant response element, and activate expression of a host of antioxidant target genes. Castiglione et al. (2020) demonstrate that a mutation in the KEAP1 coding sequence, likely caused by intrachromosomal rearrangement, in the Neoaves ancestor has led to constitutive activation of the NRF2 master antioxidant response in living birds, and this enhanced response lowers the risk of macromolecular oxidative damage and thus a decrease in oxidative stress.

47.2.3 Immunity

The avian immune system is broadly similar between mammals and birds although most of what we know about "birds" is based on studies of domesticated chickens (reviewed in Schat et al., 2014; Kaiser and Balic, 2015). Birds and mammals have both an innate and adaptive immune response and the latter includes both cell-mediated (T-cell dependent, developed in the thymus) and humoral (B-cell dependent, developed in the Bursa of Fabricius) immune responses—note that in mammals, the B-cells are derived from bone marrow. Given the high costs of maintaining the immune system (Klasing, 2004; Hasselquest and Nilsson, 2012), birds during migration may reduce immune function to spare these costs (Hasselquest, 2007; Buehler and Piersma, 2008) or since they may encounter more pathogens they may instead upregulate immune function (Møller and Erritzøe, 1998; Buehler et al., 2010) or they could reallocate resources and so maintain immune function (Lee, 2006). The balance of evidence to date at least in terms of innate immune function suggests that it may be compromised during migration in birds (Owen and Moore, 2006; Hegemann et al., 2012; Nebel et al., 2012; Eikenaar and Hegemann, 2016) although many more such studies are needed.

47.2.4 Sensory systems and navigation

Migratory birds must be able to orient and navigate during migration, and birds have evolved several key sensory mechanisms that enable successful navigation (for example, see Mouritsen, 2015). However, this topic is beyond the scope of this chapter. Several recent reviews provide excellent entry into this realm of bird migration physiology (Berthold et al., 2003; Wiltschko and Wiltschko, 2009; Mouritsen, 2018; Muheim et al., 2018).

47.2.5 Endocrine system and the environment

Migration represents the morphological, behavioral, and physiological adaptations to seasonal environments—locations with transient resources sufficient to support breeding at one time of the year and another at a distance for survival, thus creating seasonal movements in spring and autumn. To take advantage of seasonal productivity in support of breeding and rearing young (Baker, 1938; Perrins, 1970), migratory species must maintain synchrony with the environment by relying on environmental cues in conjunction with endogenous rhythms to coordinate patterns of movement and stasis.

The endocrine system is the crucial link integrating the environmental cues with behavioral and physiological cycles that contribute to seasonal migrations. Whereas, the endocrine regulation of reproduction of Aves has been thoroughly studied, less focus and progress has been made on migration. To understand why, it is valuable to consider what migration is. It is a complicated biological process with a variety of forms extending from obligate migration that occurs on a strict time table relying on seasonally reliable resources necessary for breeding and survival to facultative forms, i.e., nomadic and irruptive movements of species that respond reactively to resources that vary unpredictably (Watts et al., 2018). Furthermore, migrations occur over multiple seasons and generations incorporating great distances, various environments, habitats, and climate. Together such variables make for a difficult system to study in its entirety. One approach that has contributed to understanding the relationship of the environment and endocrine systems is Finite State Machine Theory (Jacobs and Wingfield, 2000; Ramenofsky and Wingfield, 2007) by focusing on the unique conditions of each life history stage. For example, the life history of a calendar migrant includes two migration stages—vernal and autumnal—that occur at separate times of the year under differing environmental conditions and regulated by unique neuroendocrine mechanisms. Each stage is composed of three sequential phases—(1) development, the initiation of molecular, biochemical, and genetic processes; (2) mature capability, where all morphological, physiological, and behavioral traits are expressed and actual migration begins; and (3) termination, the disorganization of the migratory traits and beginning of the next life history stage (Figure 47.1). Each phase is regulated by the environmental-endocrine nexus that affects its timing and progression. Current knowledge of the mechanisms regulating the two stages are not thoroughly defined but presenting migration in this format provides an integrated focus for future studies.

47.3 Endocrinology of migration

47.3.1 Vernal stage

47.3.1.1 Synchronization with the environment: initial predictive cue

Though not thoroughly known for all migratory species, a general consensus from many is the cue response to the vernal increase in daylength or photostimulation (Rani and Kumar, 2014; Hahn et al., 2015). The mechanism involves coincidence of the light phase of the photoperiod occurring during the peak of the circadian rhythm of photosensitivity that takes place for most species between 12 and 15 h after dawn. The reaction resides in the extraretinal photoreceptors of the basal hypothalamus in association with the putative photopigments VA opsin and Rod opsin which are

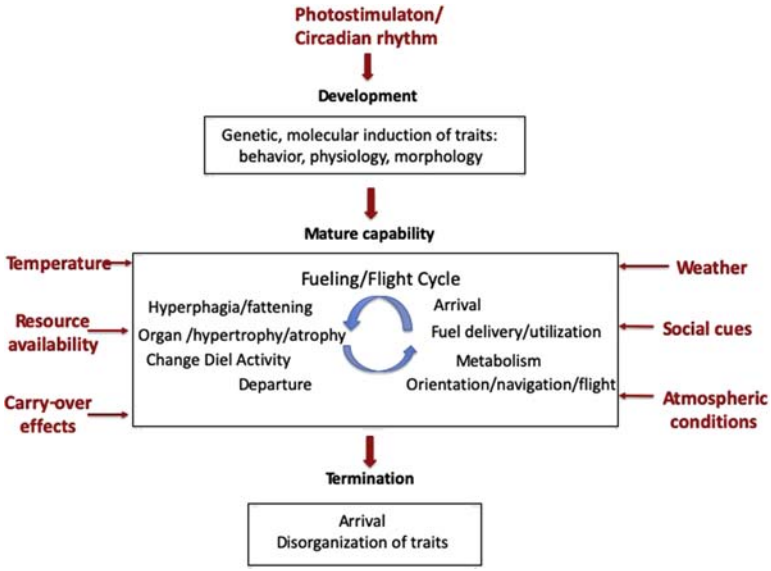

FIGURE 47.1 Three phases of the migration life history stage. Initial predictive cue of photostimulation induces the development phase and continues its effect throughout the entire stage. Local environmental and conditional cues (*red font*) affect the rate of progression through the substages of mature expression and termination.

co-expressed in the gonadotropin-releasing hormone (GnRH) and arginine vasotocin neurons (Follett et al., 1974, 1998; Meddle et al., 1997; Rastogi et al., 2013). Evidence of photostimulation was demonstrated in the First-day Release Model in which activation of the early immediate gene (*c-fos*) was detected 18 h after dawn of the first long day indicating cellular activity in the basal tuberal hypothalamus of Japanese quail (*Coturnix c. Japonica*) (Meddle et al., 1997). As a result, plasma levels of luteinizing hormone from the anterior pituitary could be detected at 22 h after dawn that in turn induced synthesis and secretion of testosterone from the gonads affecting both migration and breeding life history stages. Given that one cue, photostimulation, initiates both migration and breeding life history stages, the question becomes how are they separated to occur sequentially? Photostimulating white-crowned sparrows with low-penetration green light Wang et al. (2013) was able to separate photoperiodic responses of the migration and breeding stages. Birds developed migratory traits of mass increase, fattening, and nocturnal migratory restlessness but without gonadal recrudescence. Separation of the neuroendocrine control over initiation of migration and breeding have further support from the electrolytic lesion studies in the distinct locations of the hypothalamus in both White-throated, *Zonotrichia albicollis*, and white-crowned sparrows that demonstrated separate pathways for migratory fattening and restlessness versus gonadal development (Kuenzel and Helms, 1967; Stetson and Erickson, 1972; Yokoyama, 1976). Taken together these results suggest separate locations for photoreception and transduction for the two stages: one regulating the migratory life history and the other a more attenuated response of gonadal development and eventual breeding.

47.3.1.2 Developmental phase: hyperphagia and fattening

The initial response to photostimulation is increased food intake—hyperphagia—that leads to elevated body mass and fat stores providing fuel, lipid, and protein to support migratory flight (Ramenofsky, 1990; Lindström and Piersma, 1993). Unlike other homeostatic processes, the alterations of body mass during migration is regarded as rheostasis where the *set point* of body mass is defended but fluctuates throughout the fueling flight cycles of the stage (Mrosovsky, 1990; Jenni et al., 2000; Cornelius et al., 2013; Boswell and Dunn, 2015). The hormone action of these processes is not thoroughly understood for most migratory species but the basic principles are drawn from migratory European quail (*Coturnix coturnix*) and domesticated fowl that focus on the gene expression of opposing sets of neurons of the central melanocortin system in the arcuate nucleus of the mediobasal hypothalamus and considered the control center for body weight (Boswell, 2005; Boswell and Dunn, 2015) (Figure 47.2). Two neuropeptides synthesized in the arcuate nucleus—neuropeptide Y (NPY) and agouti-relate protein (AGRP)—act to promote feeding and mass gain. Opposing these actions are the interconnected neurons that express pro-opiomelanocortin (POMC) and cocaine-and amphetamine-regulated transcript (CART) genes. Given its basal position in the hypothalamus, the arcuate nucleus sits adjacent to the third ventricle and median eminence where the neurons are exposed to circulating levels of proteins, metabolites, and hormones that provide peripheral information. The arcuate nucleus expresses receptors for such metabolic compounds as cholecystokinin (CCK), ghrelin, corticosterone, insulin, and leptin and may serve as interpreters of metabolic state with regulatory capacity to promote or inhibit food intake via the NPY, AGRP or POMC, and CART pathways. Empirical studies using intracerebroventricular (ICV) injections into White-crowned sparrows report that while NPY stimulated food intake, the octapeptide CCK inhibited offering further support for the diverse pathways of control for feeding in a migratory bird (Richardson et al., 1992, 1993).

47.3.1.3 Thyroid hormones

Thyroid hormones play broad regulatory roles in avian physiology including metabolism, thermoregulation, breeding, molt, and migration (Groscolas and Leloup, 1986; Jenni-Eiermann et al., 2002; Zeng et al., 2013). In older literature, migratory fattening and restlessness were associated with elevated activity of the thyroid gland (Wingfield et al., 1990). Additionally, measurements of

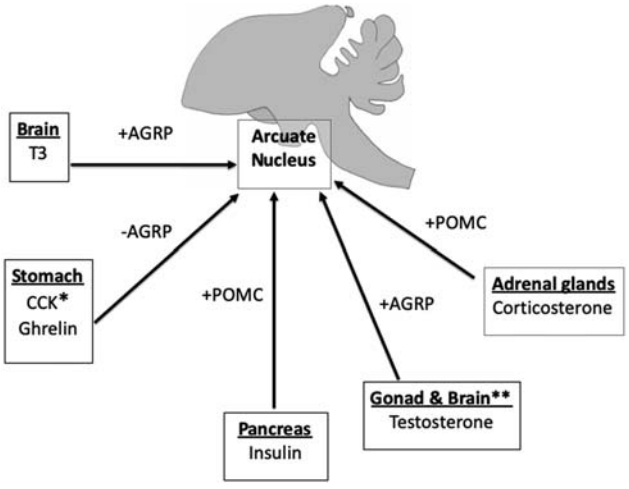

FIGURE 47.2 Suggested model for the endocrine influences on food intake via neuropeptides in the arcuate nucleus of the avian hypothalamus, * Synthesized both in gut and brain, ** possible pathway. *Adapted from Boswell and Dunn (2015).*

plasma levels of thyroxine (T4) and triiodothyronine (T3) were increased during the early stages of vernal migration (Chandola and Pathak, 1980; Smith, 1982; Pant and Chandola-Saklani, 1993). More recently, chemical inhibition of thyroid hormone disrupted prenuptial molt, fattening, mass gain, flight muscle hypertrophy, expression of migratory restlessness, and gonadal recrudescence while administration of T4 reversed these effects (Pérez et al., 2016, 2018). These results indicate a major role for thyroid hormones across the vernal stage.

Regarding photostimulation, thyroid hormones are considered central to molecular mechanisms regulating breeding (Rani and Kumar, 2014, Boswell and Dunn, 2017; Pérez et al., 2018). For a number of seasonally breeding birds, photoperiod is linked to release of thyroid-stimulating hormone (TSH) from the pars tuberalis of the pituitary gland. TSH stimulates expression of the biosynthetic enzyme Type II iodothyronine deiodinase to direct conversion of T4 to the biologically active T3 in the mediobasal hypothalamus that, in turn, stimulates release of GnRH from the median eminence to initiate breeding (Hahn et al., 2015; Nishiwaki-Ohkawa and Yoshimura, 2016). Evidence is accruing in Siberian hamsters and domestic chicks that the hypothalamic T3 may also exert effects on food intake and fattening through the AGRP neurons of the arcuate nucleus (Boswell and Dunn, 2017) (Figure 47.2). Whether these effects take place with photostimulation during migration are unknown but currently under investigation in a First-day Release Model with acute photostimulation of short-day white-crowned sparrows that will test directly whether long-day exposure induces expression of AGRP and increases food intake in the vernal stage.

47.3.1.4 Testosterone

Additionally, testosterone is implicated in regulating spring fattening. In a number of species, low levels of testosterone circulating during the winter stage are necessary for expression of hyperphagia and fattening in both sexes in the vernal but not the autumnal stages (Schwabl and Farner, 1989; Wingfield et al., 1990; Deviche, 1995; Ramenofsky and Nemeth, 2014), a condition challenged by others (Weise, 1967; Boswell et al., 1993). The mechanism by which testosterone could influence migratory fattening is through the androgen receptor (AR) on the arcuate nucleus stimulating the AGRP/NPY/feeding center pathways, as documented in mammals (Diano et al., 1997) (Figure 47.2). The timing of this effect is not known but could vary across migratory species.

47.3.1.5 Insulin and glucagon

The pancreas is the source of 3 proteins, insulin, glucagon, and pancreatic polypeptide; the former two are considered major regulators of food intake in birds acting directly and/or through their effects on carbohydrate and lipid metabolism (Hazelwood, 2000). Only a few studies have investigated these in migrating birds (Goodridge, 1964; Totzke et al., 1997; Cornelius et al., 2013). What is known is that insulin and glucagon have opposite effects on plasma glucose that in turn may influence food intake. Surprisingly, intramuscular injections of either glucagon or insulin decreased feeding in captive white-crowned sparrows (Boswell et al., 1995). Plasma glucose is exceedingly high in birds even though plasma insulin falls within the normal range suggesting peripheral insensitivity to insulin (Sweazea et al., 2006). ICV administration of insulin increased feeding indicating a marked diversion from mammals and suggesting central control of food intake by insulin (Boswell et al., 1995). Besides its effect on carbohydrate metabolism, glucagon has a biphasic effect on lipid production. At elevated levels, glucagon promotes lipolysis enhancing release of free fatty acids (FFAs), a source of energy, into the circulation. At lower range, this is reversed with increased lipogenesis and fat storage (Goodridge, 1964). Testing the effects of metabolites on feeding reported a predominant sensitivity to lipid as opposed to carbohydrate, a reasonable conclusion given the major role that lipid plays during migration (Boswell et al., 1995). Taken together, it is suggestive that glucagon levels cycle throughout migration with lowest levels occurring during the migratory fattening to support lipogenesis countered by elevated levels during flight promoting lipolysis making fatty acids available for oxidation (Goodridge, 1964). More comprehensive studies on the pancreatic peptides are needed.

47.3.1.6 Glucocorticoids (Box 47.1)

Corticosterone in birds has long been associated with hyperphagia, fattening, and stress physiology in migrants, but results across numerous studies and species have not revealed consistent patterns (Landys et al., 2006; Ramenofsky, 2011; Wagner et al., 2014; Eikenaar, 2017). One of two views contributing to these divisions is the Migration Modulation Hypothesis (Holberton et al., 1996) that addresses stress physiology with the hypothesis that neotropical migrants would not show the expected stress response to capture and handling with two predictions: (1). baseline levels of corticosterone remain elevated during migration to promote hyperphagia and lipogenesis, (2). suppression of the stress response to avoid the deleterious effects of catabolism on flight muscle (Long and Holberton, 2004). The alternative view is that most migrants display a robust stress response throughout migration with plasma levels varying seasonally across the stages and in healthy birds within physiological range (Level B (Box 47.1)). Corticosterone concentration is

BOX 47.1 Aligning corticosterone, physiological state, and allostasis.

Corticosterone, the predominant adrenal steroid of birds, is associated with many aspects of the migration life history stage including behavior, metabolism, and allostasis—measure of energy requirements over time (Blas, 2015; Ramenofsky, 2011; Landys-Ciannelli et al., 2002; Romero et al., 1997). Corticosterone acts in concert with other metabolic hormones such as CCK and insulin by regulating availability of energy in the forms of circulating FFAs and glucose to maintain homeostasis (Eikenaar, 2017). To fully appreciate the multiple functions and levels of control of corticosterone, we refer to the hypothalamus-pituitary-adrenal (HPA) axis by considering its actions at three physiological states and hormone levels. Level A is considered the basic homeostasis that maintains life (Romero et al., 2009). Level B involves the activation of the HPA for fluctuations in response to predictable, daily or seasonal demands for energy, allostatic load, across the annual life history stages. Level C, further activation of the HPA that tend to be transitory as the individual reacts to an acute and unpredictable event or stressor. The induced behavioral response is to move away or adopt an emergency life history stage (ELHS) to alleviate effects of the stress (Romero and Wingfield, 2016). To assess function of the HPA axis, two measurements of circulating corticosterone are collected: the first is the baseline measure collected within the first 3 min of capture and reflecting the current and unprovoked energetic state of the organism. The second sample is the acute or Level C collected at the peak of the response to capture and handling indicating the maximal response of the corticosterone synthesis and secretion (Figure 47.3).

regulated by the enzyme 11β-hydroxy steroid dehydrogenase, and it acts through two cellular receptors, the high-affinity mineralocorticoid receptor (MR) and low-affinity glucocorticoid receptor (GR). Considering these multiple levels of control, corticosterone is regarded as a pleiotropic hormone with multiple effects acting in permissive roles with critical metabolic compounds that affect energy balance and allostasis including: insulin, insulin-like growth factor-1, adipose tissue lipoprotein lipase, low-density lipoprotein, FFA, NPY action (Savard et al., 1991; Remage-Healey and Romero, 2001; Landys et al., 2004a, 2006; Blas, 2015; Mishra et al., 2017), and growth hormone (GH) (Tsipoura et al., 1999). All of which contribute to the complexity and often confusion over corticosterone's action. Taking a more comprehensive view of the interactions of hormones and metabolic compounds in terms of allostasis and stress at each substage may provide a more complete assessment of the role of this steroid in migration.

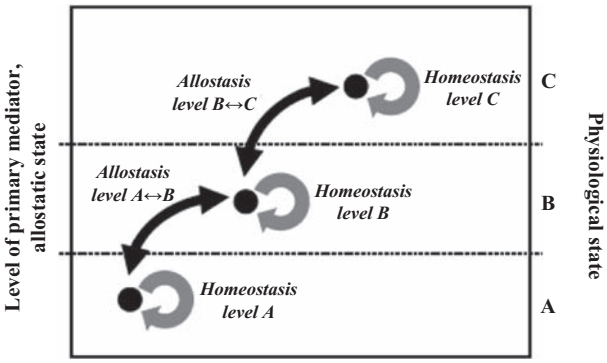

FIGURE 47.3 Description of state levels of corticosterone in relation to homeostasis, allostasis, and physiological state. *With permission from Landys et al. (2006).*

47.3.1.7 Leptin

The protein hormone leptin is known as a strong regulator of energy balance in mammals synthesized almost exclusively in the adipose tissue and signaling presence or deposition of fat—positive energy balance (Zhang et al., 1997). Thus it is considered a hormone with autocrine, paracrine, and endocrine functions promoting homeostasis of energy stores and has been coined an *adipostat* in mammals regulating food intake according to energy balance—fat content (Friedman-Einat et al., 2014; Friedman-Einat and Seroussi, 2019). Recent identification in birds of a leptin ortholog and characterization of leptin activity has questioned whether it functions either as a hormone or an *adipostat*. The leptin ortholog shows no expression in adipose tissue but is found in brain, adrenal gland, and gonad. So far, the avian leptin receptor LEPR is expressed only in the pituitary. Given these diversions from mammals, it is unlikely that avian leptin is acting as an homeostatic adipostat and offers preliminary support for the Rheostasis Model during migration that would allow for greater range of body mass and fat stores (Cornelius et al., 2013; Friedman-Einat et al., 2014; Watts et al., 2018).

47.3.1.8 Ghrelin

Acting in opposition to leptin, this 28-amino-acid peptide is secreted from the stomach and other digestive organs of birds when body condition is low. Ghrelin is reported to be a GH-releasing factor in chickens and mammals that relays information to the arcuate nucleus of nutritional state. In rats, both intraperitoneal (IP) and ICV injections of ghrelin increased food intake. This effect is most apparent during fasting with ghrelin acting on the NPY/agouti-related peptide (AGRP) neurons in the arcuate nucleus (Nakazato et al., 2001). In quail, plasma ghrelin was elevated in fasted

birds but dropped once feeding commenced. The results obtained with injections were less clear; at low concentrations, IP injections enhanced feeding but at greater dosages both IP and ICV injections decreased feeding suggesting a biphasic effect. Other studies in which ghrelin was administered reported that food intake in neonatal chicks was inhibited (Kaiya et al., 2009). Although presently controversial in domestic fowl, these results suggest that ghrelin could regulate feeding during migration, particularly when birds are lean, as they would be following flight. In a recent study, lean Garden warblers, *Silvia borin* captured in the morning upon arrival at a stopover site on the Mediterranean Island of Ponza measured elevated plasma levels of the acylated ghrelin (Goymann et al., 2017). After refueling throughout the day in captivity, injections of the unacylated form of ghrelin decreased food intake and increased migratory restlessness at night pointing to either a biphasic effect on behavior of hormone concentration or different responses to the chemical forms of the hormone. By contrast, plasma ghrelin was not associated with nocturnal departure in fully fattened European blackbirds (*Turdus merula*) at an autumn stopover (Eikenaar et al., 2018b). These outcomes may represent species or seasonal differences calling for a more complete understanding of ghrelin's role during migration.

47.3.1.9 Prolactin

Understanding prolactin's role in migratory feeding and fattening has been problematic. Administration of this pituitary protein increases fattening in a number of migratory species (Meier and Farner, 1964; Yokoyama, 1976). Also, central effects of prolactin have been identified in specific sites of the hypothalamus of migratory and nonmigratory birds (Yokoyama, 1976; Buntin et al., 1993). Prolactin levels, like other pituitary hormones, vary seasonally as photostimulation induces secretion of vasoactive intestinal peptide, the stimulatory hypothalamic neuropeptide that induces production and secretion of prolactin from the pituitary (Figure 47.4). This information led to the hypothesis that prolactin could be acting in conjunction with gonadal steroids and corticosterone that were also elevated with photostimulation to induce migratory fattening and restlessness. However, questions have been raised in a number of migrants as the photoinduced peak of prolactin corresponds only weakly with onset of feeding, fattening, and migratory restlessness (Hall and Gwinner, 1987; Schwabl et al., 1988; Holberton and Dufty, 2005). Timing of peak prolactin levels appears to align its function with the autumnal stages of refractoriness and postnuptial molt (Dawson et al., 2001). So, even though prolactin injections increase fattening in birds, prolactin function in relation to migratory fattening is far from resolved.

47.3.1.10 Hormones of flight muscle hypertrophy

In addition to fueling, the pectoralis flight muscles undergo hypertrophy in numerous migratory species (Fry et al., 1972; Driedzic et al., 1993; Jehl, 1997; Bauchinger et al., 2005; Banerjee and Chaturvedi, 2016) during migration that entails increased size of the muscle fibers serving to enhance contractile force and flight performance (Marsh, 1984; Gaunt et al., 1990; Evans et al., 1992; Vezina et al., 2021) (Ramenofsky et al., in prep). Though not well

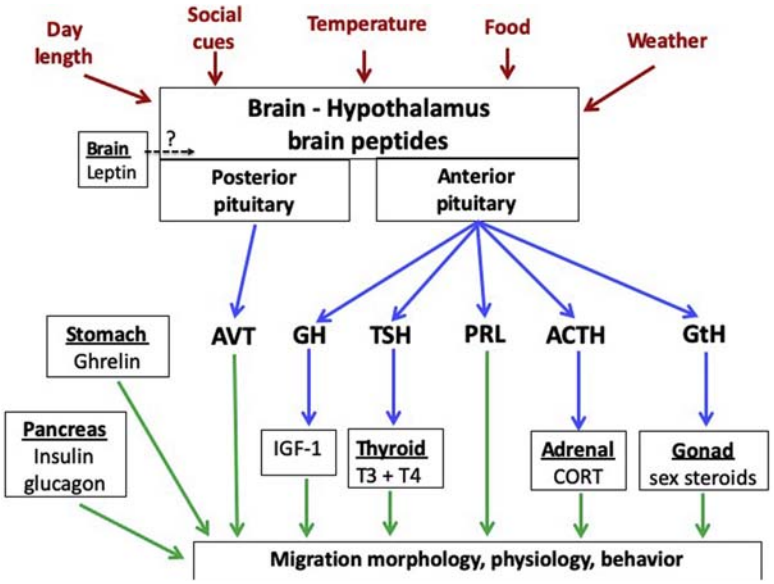

FIGURE 47.4 Flow chart of the environmental cues on the neuroendocrine—endocrine system influencing migratory morphology, physiology and behavior. *Red arrows*—environmental cues influence the activity of hypothalamic brain peptides, *Blue arrows*—pituitary hormones regulated by the brain peptides affect peripheral endocrine glands. *Green arrows*—hormones of the endocrine glands regulate migratory functions. Role of leptin at present is uncertain (Friedman-Einat and Seroussi, 2019).

understood in migratory birds, anabolic steroids—testosterone and 5α-dihydrotestosterone (DHT)—are likely candidates for regulating phenotypic changes given reports of AR gene increasing expression in flight muscles of Golden-collared manakin (*Manacus vitellinus*) during sexual displays (Fuxjager et al., 2012). Furthermore, elevations of androgen signaling via DHT, AR mRNA and the androgen dependent gene, insulin-like growth factor 1, showed increased levels of expression in White-crowned sparrows at spring departure when flight muscles were enlarging (Pradhan et al., 2018). Regulation of muscle hypertrophy could be a function of vernal photostimulation that activates androgen secretion and gonadal recrudescence (Blanchard and Erickson, 1949; Wingfield et al., 1990; Bauchinger et al., 2007). Or it is possible that testosterone secreted during the winter stage has an early priming effect on muscle as it does on fattening (Wingfield et al., 1990), something that is unknown at present. Nevertheless, the circulating levels of androgen are low as birds prepare for vernal departure suggesting intramuscular cell signaling pathways may direct muscle remodeling and thus avoiding the systemic and behavioral effects if levels of androgen were elevated at this substage (Bauchinger et al., 2007; Pradhan et al., 2018). In addition to androgen, corticosterone is implicated in flight muscle hypertrophy at vernal departure. Elevation of baseline corticosterone was associated with increased expression of the high-affinity MRs expression in White-crowned sparrows (Pradhan et al., 2019). This relationship is thought to promote the anabolic stimulation of the muscle fibers, contributing to hypertrophy. Such effects are not known for the autumnal stage.

In addition to the phenotypic changes orchestrated by the endocrine system, integral components of the muscle fibers alter during this vernal substage. Myosin heavy chain (MyHC) expression is associated with the muscle's mechanical capabilities (Reiser et al., 1996). Changes in the expression of the MyHC isoform during the development phase may represent an adaptation for upgrading the muscle mechanics for impending long-distance flight (Velten et al., 2016). Again, hormonal mechanisms regulating these molecular processes are not known but deserve further study during the autumnal stage.

47.3.1.11 Mature expression—hormones of the fueling and flight cycle

Stopover in both the vernal and autumnal stages provides the clearest example of the fueling and flight cycle as flight temporarily ceases upon arrival for rest, fueling, and then departure (Delingat et al., 2006, 2009) offering an excellent opportunity for tracking metabolic hormones. During the 4500 km journey from Africa to a stopover site along the northern Dutch coast in spring, Bar-tail godwits (*Limosa lapponica*) cease travel for approximately one month to rest, continue prenuptial molt and refuel before continuing northward to the breeding grounds. One indication that baseline corticosterone levels align with allostatic load was noted initially by Landys-Ciannelli et al. (2002). Birds arrived in lean condition having lost on average 55.3% from their African departure body mass with elevated baseline levels of corticosterone suggesting a heightened allostatic load resulting from the "wear and tear" of a long continuous flight. Similarly, elevated baseline levels are reported in recently arrived Semipalmated sandpipers at a vernal stopover in Delaware Bay (Tsipoura et al., 1999), Gray Catbirds (*Dumetella carolinensis*) along the Gulf Coast of the United States (DeSimone et al., 2020), and Garden warblers on the Mediterranean Island of Ponza (J. Wingfield and M. Ramenofsky, in preparation). Measurements of body mass of the sandpipers throughout the period of arrival varied over a range of 10 g indicating the different stages of recovery of birds across the flock. Over this period plasma GH was negatively correlated with mass. Given the lipolytic functions of GH, the data suggest hormone action would supply fatty acids to meet the energetic demands of flight and arrival until sufficient refueling achieved positive energy balance observed in the heavier birds of the flock (Tsipoura et al., 1999). During the refueling phase of the godwits, the baseline corticosterone declined 55% from arrival. The corticosterone response levels of both the refueling and arriving birds were similar, however the peak for the arriving birds reached their maxima more rapidly suggesting a heightened sensitivity of negative feedback possibly related to allostasis and/or CNS functions required for flight (Landys-Ciannelli et al., 2002; Eikenaar et al., 2013; Romero and Wingfield, 2016). Once refueling achieved positive energy balance, godwits became aphagic while digestive organs atrophy to conserve body mass and metabolic costs for the ensuing flight (Piersma, 1998; McWilliams and Karasov, 2001). Baseline corticosterone increased around the time of departure and has been associated with the upregulation of HPA for regulating metabolic and behavioral functions for impending flight and organ remodeling (Reneerkens et al., 2002b; Eikenaar et al., 2013, 2018a; Pradhan et al., 2019). However, such upregulation was not identified in the short-hop migrant, Gray catbird (DeSimone et al., 2020) suggesting a divergence in preparation for flight depending upon species and migratory strategy. Nonetheless, studies of both captive and free-living migrants offer further support for the association of corticosterone with expression of migratory flight in the vernal and autumnal stages (Schwabl et al., 1991; Ramenofsky et al., 1999; Landys et al., 2004b; Falsone et al., 2009; Eikenaar et al., 2014; Ramenofsky and Wingfield, 2017). Taken together the results from these studies offer insight into the effects

of corticosterone and GH, providing endocrine support for the behavioral and metabolic processes during flight, at arrival, and preparation for departure during stopover.

47.3.1.12 Termination: arrival biology

Reaching the breeding grounds heralds the termination phase that includes the substage of arrival biology (Wingfield et al., 2004). This substage requires flexibility and coordination of morphology, behavior, and physiology given the quixotic environmental conditions birds may face in early spring. Observational studies note that migratory behavior changes as birds approach breeding areas and begin to search widely for optimal habitats (Hahn et al., 1995). Studies of captive birds, report changes in intensity of migratory restlessness and orientation near the conclusion of the migratory period, suggesting transition to more wandering and searching behaviors expressed even in captivity (Helms, 1963; Wiltschko et al., 1980; Ramenofsky et al., 2003). Elevated corticosterone during arrival has been proposed to support these behaviors (Ramenofsky, 2011, 2012; Romero and Wingfield, 2016) providing examples of the pleiotropic effects of corticosterone influenced by its multiple sites of control at the enzymatic, receptor, and state levels.

Field studies document that winter conditions may prevail at high altitudes and latitudes after birds arrive with storms likely forcing delay of the onset of breeding (Morton, 2002). In these cases, migrants retain elevated levels of corticosterone that promote escape to refuges until conditions on the breeding grounds improve (Reneerkens et al., 2002a; Hahn et al., 2004; Ramenofsky and Wingfield, 2007; Cornelius et al., 2013; Ramenofsky and Wingfield, 2017).

Throughout the vernal stage, hypothalamic-pituitary-gonad (HPG) axis function progresses with increasing levels of gonadotrophin (GtH), testosterone, and DHT, so males are ready to breed once females arrive and environmental conditions allow (Wingfield and Farner, 1978a; O'Reilly and Wingfield, 1995; Ramenofsky and Wingfield, 2006; Covino et al., 2015, 2017; Lymburnera et al., 2016). For females, the story differs as the final stages of oogenesis, yolk deposition, egg laying, and behaviors regulated by estrogen and progesterone are held in check under the regulation of the hypothalamic peptide gonadotropin-inhibiting hormone (GnIH) (Tsutsui et al., 2000; Bentley et al., 2009). This peptide is thought to act as a "brake" on reproduction by its central effects on sexual behavior and hypophysiotropic control of steroidogenesis and ovulation (Wingfield et al., 2016). Under extreme conditions, interactions of elevated corticosterone and GnIH may serve to extend the more mobile or nomadic phase of arrival biology until conditions improve sufficiently for GnIH levels to decline and activation of the breeding stage to proceed. Regulation of GnIH function is not well understood, but environmental conditions appear to play a major role guiding synchronization of egg laying with the potential peak of food abundance (Perrins, 1970; Ramenofsky and Wingfield, 2017). Onset of breeding marks the termination of the vernal migratory stage.

47.3.2 Autumnal stage

47.3.2.1 Development phase

In many ways, autumnal migration appears similar to that of the vernal stage, as it can be divided into the same three phases. The traits are similar that include hyperphagia, fattening, organ remodeling, fueling flight cycles, etc. But major differences exist that provide important clues to the endocrine regulation and biological significance of each stage. For example, the recognized mechanisms that regulate activation of the vernal stage are not operating in the autumnal stage. Daylength is decreasing rather than increasing and many migrants are known to be photorefractory with a loss of responsiveness of reproductive development to long photoperiods (Dawson et al., 2001). The HPG regresses with circulating levels of GtHs and gonadal hormones decline to basal precluding further breeding attempts late in the season. For most species, both baseline and response levels of corticosterone have dropped from elevated breeding titers.

47.3.2.2 Thyroid hormones

A number of seminal studies of captive birds have identified that the rate of increase of vernal photoperiods during photostimulation exert latent effects on the timing of photorefractoriness followed by the autumnal events of postnuptial molt and migration (Farner and Follett, 1966; Gavrilov and Dolnik, 1974; Dolnik, 1980; Moore et al., 1982, 1983). Thyroid hormones are considered central by playing organizational roles for both vernal and autumnal stages during photostimulation (Dawson et al., 2001; Pérez et al., 2016, 2018). Timing of the thyroid organizational effects is considered the basis for the separate pathways of control for the two stages (Wilson and Reinert, 1996, 1999). A further understanding is now needed for the hormonal and genomic mechanisms underlying the potential roles for thyroid hormones during autumnal migration.

47.3.2.3 Androgen

Unlike in spring, plasma levels of androgen in the autumn are basal and probably not capable of activating feeding centers in the hypothalamus. Furthermore castration during the previous winter months had no effect on autumnal fattening in white-crowned sparrows (Wingfield et al., 1990). However, both androgen and estrogen are

synthesized locally in the brain and affect autumnal behavior (Soma et al., 2002; Pradhan et al., 2010). So, it is possible that neurosteroids (steroid synthesized in the brain) could influence the arcuate nucleus via AGRP as in spring (Boswell and Dunn, 2015) to affect autumnal hyperphagia and fattening. Whether the molecular pathways associated with vernal flight muscle hypertrophy operate during the autumnal stage are unknown at present but represent intriguing possibilities.

47.3.2.4 Glucagon

One of a very few studies investigating glucagon in migratory birds during the autumnal stage in comparison with winter identified a decrease in plasma levels of glucagon and FFA in captive Red-winged blackbirds (*Agelaius phoeniceus*) while fattening for autumnal migration (Hintz, 2000). Although a surprising result, Goodridge (1964) suggested previously that seasonal patterns of circulating levels of glucagon could contribute to fluctuations in adiposity with elevated levels promoting lipolysis to mobilize energy to fuel such activities as thermogenesis during winter months and lower titers to enhance fat storage for migration. Further studies are needed to more clearly define the roles of glucagon during the autumn stage.

47.3.2.5 Mature expression—hormones of the fueling and flight cycle

Field studies focusing on the transition from breeding to autumnal departure identified that birds express individual schedules that vary according to investment and timing of breeding, molt, social interactions, age, body condition as well as local weather and wind direction (Runfeldt and Wingfield, 1985; Morton and Pereyra, 1994; Bonier et al., 2007; Sjoberg et al., 2015; Chmura et al., 2020). This asynchrony across individuals deviates markedly from the highly synchronous schedules birds express in spring driven by the initial predictive cues and strong selective pressure for birds to arrive on the breeding grounds as early as feasibly possible for reproductive advantage (Smith and Moore, 2005). Investigating the differences may help to further identify the regulatory distinctions of the two stages but very few studies have focused on seasonal comparisons at time of departure (Wingfield and Farner, 1978a).

Among the overland migrant populations of White-crowned Sparrow, body mass and fat scores reached their annual peak just prior to departure, a time when plasma levels of baseline and response corticosterone are comparable with the vernal and autumnal values but at significantly lower levels than the annual peaks reached upon arrival on the breeding grounds and during early stages of breeding (Wingfield and Farner, 1978a; 1978b; O'Reilly and Wingfield, 1995; Krause et al., 2021). By contrast, in trans-hemispheric migrant Barn swallow (*Hirundo rustica*), the baseline levels of corticosteron prior to spring departure exceeded the values in autumn, while the response levels of autumn rose above vernal values (Raja-Aho et al., 2013). Body condition of spring birds was poor compared to autumn leading to question the role of corticosterone at the time of departure. Given the disparate results across the two species it is clear that additional studies of the physiological condition and endocrine state of birds at departure in the vernal and autumnal stages require further investigations.

47.3.2.6 Stopover: arrival

As in spring, the autumnal stopover substage provides advantageous views of the behavioral, physiological, and endocrinological states upon arrival and at departure. A pattern of elevated baseline and response corticosterone levels has been recorded in some but not all migratory species with greater titers measured in vernal than in autumnal stages (O'Reilly and Wingfield, 2003; Romero et al., 1997; Loshchagina et al., 2018; Bauer et al., 2019). When seasonal differences are detected it is suggested that allostatic load of vernal migration for these species exceeds that of autumn. Impending breeding, unpredictable and unfavorable climatic conditions and less reliable sources of food are some of the issues migrants face at higher altitudes and latitudes in spring. In species that cross extreme barriers (oceans and deserts) with routes that are comparable in both seasons, the baseline levels corticosterone may not vary to any great extent. However another mechanism for differential regulation of corticosterone is presented in Dark-eyed junco sampled during spring and autumn stopovers (Bauer et al., 2019). Neither baseline nor response corticosterone varied seasonally but negative feedback was weaker in spring than autumn suggesting a greater response to environmental purturbations. Surprisingly, these findings diverge from the increased sensitivity to capture in the spring arriving Bar-tailed godwits in comparison with refueling birds and possibly suggests a species or migratory strategy difference.

Thus, a more consistent theme arising from a variety of species captured during autumnal migration indicate only moderate elevations of baseline corticosterone, suggesting an allostatic load that falls substantially below stress-induced values Level C (Schwabl et al., 1991; Gwinner et al., 1992; Falsone et al., 2009). At such titers, corticosterone may serve to support metabolic processes supplying energy required for endurance flight in terms of protein-mediated transport of fatty acids into flight muscle and high-oxidative capacity (Lundgren and Kiessling, 1985; McFarlan et al., 2009; Price et al., 2010). Yet, a few extreme cases are reported of birds captured in flight in nearly starved condition with depleted stores of fat, atrophied flight muscle, and baseline corticosterone titers that

reach well into the realm of the acute response to stress Level C (Box 47.1). Fat stores are considered key regulators of systemic protein catabolism that can affect allostatic load, plasma corticosterone, and potentially survival (Schwabl et al., 1991; Jenni and Jenni-Eiermann, 1992; Schwilch et al., 2002; Falsone et al., 2009). Such elevated baseline levels indicate that an individual has entered the ELHS in which corticosterone metabolically promotes protein catabolism and directs a change in behavior from landing to foraging enabling recovery as long as sufficient resources can be accessed (Romero and Wingfield, 2016).

47.3.2.7 Stopover: departure

At departure from stopover sites, birds are metabolically ready for flight with elevated body mass, fat deposits, circulating triglyceride levels, and hypertrophied flight muscle (Sandberg et al., 2002; Fusani et al., 2009; Goymann et al., 2010; Covino and Holberton, 2011). Additionally, baseline levels of corticosterone correspond with orientation at take off and environmental cues conducive for flight namely favorable winds (Lohmus et al., 2003; Eikenaar and Schmaljohann, 2015; Eikenaar et al., 2017b, 2018a). In captive migrants, expression of nocturnal migratory restlessness coincides with increased baseline corticosterone (Ramenofsky and Wingfield, 2017). Taken together, departure involves the interactions of the environment, endocrine system, physiological state, and motivation to fly. The causal relationship of these variables remains to be elucidated fully.

47.3.2.8 Termination—arrival

Generally, arrival at the overwintering sites marks the end of autumnal migration with the loss of such traits as hyperphagia, fattening, flight muscle hypertrophy, and nocturnal restlessness (Romero et al., 1997; Ramenofsky, 2011). For the most part overwintering sites provide sufficient resources for survival but competition for access to roosts sites, quality food, and protection from predators can be intense given presence of resident birds as well as potential influx of breeding birds from the opposite hemisphere (Newton, 2008). Unlike the vernal stage, autumnal arrival is less synchronized, more facultative, and birds may not settle for weeks (Watts et al., 2018). To avoid competition, some cohorts arrive early or continue movement to locate in areas where food is more abundant and environmental conditions less harsh, as noted in the differential migration patterns (Ketterson and Nolan, 1976; Terrill, 1990; Newton, 2008). Given the degree of competition for resources at this stage, it is surprising that levels of androgen and corticosterone levels are basal (Wingfield and Farner, 1978a, b; Romero et al., 1997; Krause et al., 2021). However, the prospects that neurosteroids may be influencing behavior is a possibility but unknown to date. Also, involvement of the metabolic hormones is unclear emphasizing the need for more research during the autumnal stage.

47.3.3 Conclusions to endocrine system

Comparing vernal and autumnal life history stages of migration offers insight into the adaptations of each providing a fuller appreciation of the migratory processes as a whole. In general, there are common traits that support movement in either season that include an engine (muscle) for power, fuel supplies, oriented movement, and a clock (Piersma et al., 2005; Ramenofsky and Wingfield, 2007; Dingle, 2014). Specifically, the two stages and their substages diverge in terms of the environmental conditions encountered, types of available resources, reproductive states, endocrine status, age groups participating, and in many cases the migratory routes. These differences are the most revealing as they convey individual and/or population responses to the selective pressures of the environment, season, fitness, and survival supporting the premise that the two migratory stages are distinct adaptations. But before any conclusions can be fully drawn, more thorough coverage of the endocrinology of the autumnal stage is needed in addition to the studies that track plasma levels of hormones over the course of each stage. Together these points emphasize more work is needed that focuses on the molecular and genomic aspects of the endocrine system, receptor expression, and the metabolic enzymes all playing key roles in determining hormone action during both the vernal and autumnal stages as reported by (Sharma et al., 2018). Such integrative studies will provide a more complete understanding of how the environment influences migration and how individuals will cope with the challenges of climate change.

47.4 Physiological aspects of migratory preparation and long-duration flight: fueling/flight cycle

47.4.1 Introduction

Given that the vernal and autumnal migration stages occur at separate times of the year under differing environmental conditions and evidence to date suggests that the two stages are regulated by unique neuroendocrine mechanisms, we would expect some important differences in the physiology of birds during the two migration stages. In general, physiological ecologists who study migration in birds pay attention to the season in which birds are studied, but few studies have focused on how the physiology of migratory birds differs between vernal and autumnal migration. This paucity of information about the mechanistic underpinnings of physiology during the two migration stages has led us to

organize the following section around several themes (feeding and diet selection, fuel storage and fat quality, fuel use and oxidative balance) that we assume to be relatively common to both migration stages, but that may be produced by different gene-to-physiology-to-behavior mechanisms.

47.4.2 Feeding

47.4.2.1 Hyperphagia

During spring and fall migration, migratory birds increase their daily food intake (e.g., hyperphagia) in an effort to increase energy stores (fattening) to fuel their migratory flights (Odum, 1960; King and Farner, 1965). These periods of intensive feeding and fattening can last a few days to several weeks (Schaub and Jenni, 2001; Chernetsov and Mukhin, 2006; O'Neal et al., 2018), and the duration is influenced by a variety of ultimate and proximate factors such as migration strategy (e.g., long-distance, short-distance), weather, food availability, predation risk, endogenous cycles, and physiological state (Jenni and Schaub, 2003; Fusani et al., 2009; Goymann et al., 2010; Guillemette et al., 2012; Hou and Welch, 2016). For many species, the total duration of migration is composed mostly of time spent at stopovers and not in flight itself (Wikelski et al., 2003). Thus, much of the hyperphagic periods of most birds are at stopover sites. For several species of dabbling ducks, for example, the average stopover duration was 28 days and was heavily correlated with habitat quality and the availability of food (O'neal et al., 2018). In contrast, the primary predictor of stopover duration in several species of songbirds was dependent upon the amount of fat reserves when all other factors such as weather, predation, and food availability were equal or controlled (Fusani et al., 2009; Goymann et al., 2010; Smith and McWilliams, 2014) although this generalization is based almost entirely on correlational rather than empirical experiments (Smith and McWilliams, 2014).

47.4.2.2 Balancing the energy costs of hyperphagia

Hyperphagia can inherently increase daily energy expenditure (DEE) due to the additional time required to search for and process food as well as maintain more metabolically active tissue (reviewed in Lindström, 2003). Hyperphagic birds must compensate for increased foraging costs in order to rapidly gain body stores. Thus, migratory birds exhibit behavioral and physiological adjustments during hyperphagia that help to promote fattening while keeping their DEE similar to or below nonhyperphagic periods. For example, premigratory common eiders, *Somateria mollissima*, foraged three times more than postmigration periods yet their DEE remained the same because they reduced their heart rate during inactive times and decreased their time spent flying (Guillemette et al., 2012). Several species of migratory birds ranging in size from hummingbirds (Carpenter and Hixon, 1988) to small passerines (Wojciechowski and Pinshow, 2009) to large geese (Butler and Woakes, 2001) reduce their body temperature and so reduce energy expenditure prior to migratory flight regardless of food availability, weather conditions, or degree of fat storage.

47.4.2.3 Fasting and refeeding during migration

During migration, many birds alternate between short-term fasting while flying and intense refeeding at stop over sites. This cycle of alternating feeding and fasting influences gut morphometrics, fattening rates, and subsequently the pace of migration (Karasov and McWilliams, 2005). It is well established that the guts of migrating birds atrophy after just 1 or 2 days of fasting, and this decreased digestive capacity influences the rate at which birds can increase feeding rate and uptake of nutrients upon arrival at stopover sites (Diamond et al., 1986; Karasov and Pinshow, 2000; McWilliams and Karasov, 2005). Thus, the phenotypic flexibility in the gut of migratory birds plays a significant role in their ability to immediately refeed and fatten. Several species of migratory birds increase gut length, mass, and volume in response to increases in food intake in preparation for migratory flight (Dykstra and Karasov, 1992; McWilliams et al., 1999; McWilliams and Karasov, 2001, McWilliams and Karasov, 2005). However, digestive enzyme activity and nutrient uptake per unit of tissue did not change with an increase in food intake in several species of fasted passerines (reviewed in McWilliams and Karasov, 2005). Thus, the relevant phenotypic flexibility seems to be primarily in the size of the gut and not the chemical aspects of digestive efficiency. The digestive organ sizes of fasted birds appear to increase within one to six days upon increasing food intake (Dekinga et al., 2001; McWilliams and Karasov, 2001) and the activity of digestive enzymes, nutrient transporters, and absorption rates take 2–3 days to recover after fasting (Karasov and Hume, 1997; Karasov and Pinshow, 2000). This delay in recovery can impact the overall time it takes for a bird to become ready to migrate and thus slow the overall pace of migration.

47.4.2.4 Diet selection during migration

Many migratory bird species display seasonal shifts in diets associated with migration. For example, several passerines switch from a primarily insectivorous diet to a more frugivorous diet (Herrera, 1984; Bairlein and Gwinner, 1994; Parrish, 1997, 2000), and some artic nesting

shorebirds switch from a diet of primarily terrestrial invertebrates to aquatic invertebrates during fall migration (Schwemmer et al., 2016). This dietary flexibility has been attributed in part to the changes in food availability linked with season (Martin and Karr, 1986; Tsipoura and Burger, 1999). For example, migratory songbirds locally congregated where fruit abundance was high (Smith and McWilliams, 2014), they consistently chose foraging sites with higher fruit availability than insect availability during migration (Martin and Karr, 1986), and migrating shorebirds chose stopover sites along the shores of the eastern United States in conjunction with horseshoe crab spawning (Tsipoura and Berger, 1999). However, food abundance does not entirely explain diet switching in birds during migration as a multitude of factors play into diet choice (see Murphy, 1994 for complete review) including the need to rapidly satisfy the elevated daily energy and nutrient demands associated with migration while eating foods that are typically nutritionally unbalanced.

Birds seem capable of discriminating between foods based on relatively small differences in key nutrients, and these nutrient-based preferences may change with nutrient demands although the evidence for this is less convincing. As an example, four species of tanagers, Blue Dacnis, *Dacnis cayana*, Flame-crested tanager, *Tachyphonus cristatus*, Green Honeycreeper, *Chlorophanes spiza,* and Short-billed tanager, *Cyanerpes nitidus*, were shown to detect differences of 1% in sugar concentration and 2% in lipid concentration in available diets (Schaefer et al., 2003). In addition, these tanagers were able to distinguish between diets that differed in protein type (casein vs. bovine serum albumin) while protein concentration remained constant (Schaefer et al., 2003). Furthermore, white-crowned sparrows preferred semisynthetic diets that differed only in certain amino acids, and their preferences for sulfur amino acids needed for feather growth increased during times of molt (Murphy and King, 1987). Yellow-rumped warblers offered several diets that varied in nutrient and energy content ate a combination of available diets that allowed them to satisfy both their energy and protein demands during nonmigratory as well as migratory periods (Marshall et al., 2016). Whether or not birds have preferences for specific amino acids during the migratory period has not yet been determined. Likewise, many species of migratory birds prefer diets containing certain types of fats (i.e., unsaturated over saturated) and certain amounts or ratios of fatty acids in their diets (Pierce and McWilliams, 2014). There is some evidence that these bird preferences for specific fats remain similar across seasons. For example, red-eyed vireos, *Vireo olivaceous*, preferred diets with more 18:1n9 than diets with more 18:0 or 16:0, and this diet preference did not change between nonmigratory and migratory periods (Pierce and McWilliams, 2005). In sum, migratory birds can discriminate between diets based on certain nutritionally relevant components such as carbohydrates, amino acids, and fatty acids, although whether these nutrient preferences change with nutrient demands is not well documented.

Plant secondary metabolites (PSMs) may influence diet choice in migratory birds somewhat independently of the nutrient composition of foods. PSMs have been shown to increase fruit consumption in some passerines (Cipollini and Stiles, 1993; Bairlein and Simons, 1995) while decreasing consumption in others (Cipollini and Levey, 1997; Levey and Cipollini, 1998). Cedar waxwings, *Bombycilla cedrorum*, chose to eat the energy-rich fruit of *Viburnum opulus*, only when catkin proteins were simultaneously available as a protein source to help produce buffers against the acidic high-PSM fruits (Witmer, 2001). In addition, dietary PSM increased energy intake, intestinal mass, and BMR of rufous-collard sparrows, *Zonotrichia capensis*, but not the common diuca-finch, *Diuca diuca* (Barceló et al., 2016). The impact of the wide diversity of PSM on the diet choices of birds during migration and their ability to regain fuel stores remains a fruitful but largely unexplored question.

47.4.3 Fuel storage

47.4.3.1 Fats are the primary fuel

Birds fuel their high-intensity endurance exercise such as migratory flights primarily by metabolizing fat along with a small amount of protein (McWilliams et al., 2004; Guglielmo, 2010, 2018; Jenni-Eiermann, 2017). Migratory birds use primarily fat to fuel their long-duration flights because fat provides substantially more energy than an equal mass of either carbohydrate or protein (Blem, 1990) which makes it an ideal fuel for weight-economizing migratory birds (Bishop and Butler, 2015; Braun, 2015). Thus, in preparation for long-duration flights birds store appreciable energy mostly as fat (90+% of body mass increase) as well as key nutrients including protein (Jenni and Jenni-Eiermann, 1998; McWilliams et al., 2004; Bishop and Butler, 2015; Guglielmo, 2018) and antioxidants (Skrip et al., 2015a; Cooper-Mullin and McWilliams, 2016). The resulting hypertrophy of muscles and accumulation of substantial visceral, subcutaneous, and intramuscular fat stores has been long-known and well-studied (Blem, 1976, 1990; Bishop and Butler, 2015).

Given that storage of energy and key nutrients is necessary for all migratory birds, and especially those that travel long distances without the opportunity to stopover (Piersma, 1998), there is a type of "limiting factor" worth mentioning here—there are clear limits to how much storage mass can be carried for birds that also must fly. In general, the scaling of energy required to fly to that available from the muscles given their size declines as body size

increases (Pennycuick, 1975). This means that birds as heavy as ostriches cannot become airborne even if they had the largest of wings (Norberg, 1990; Marden, 1994). Such biomechanical constraints (i.e., high-wing loading expressed as grams body mass/cm^2 wing area) associated with flight have been invoked to explain why, for example, swans and ducks require a running takeoff to become airborne and fly (Lovvorn and Jones, 1994), the flightlessness of steamer ducks (Livezey and Humphrey, 1986), and why there are relatively few bird species that fly and eat only plants due to gut size limitations of herbivory (Dudley and Vermeij, 1992; McWilliams, 1999). However, such biomechanical constraints also apply to individuals that dramatically increase their energy and nutrient reserves or gut mass during key times of the annual cycle such as migration (Guillemette, 1994; Piersma, 1998; McWilliams, 1999), while not changing the size of their flight apparatus. For example, the accumulation of large body reserves during the prelaying period results in some female common eider unable to take off (Guillemette and Ouellet, 2005). Such thresholds for flightlessness seem best explained by biomechanical models that integrate wing morphology, muscle mass, and maximum power output required for take off (Marden, 1994). Thus, limitations associated with the power requirements of flapping flight constrain the extent of energy and nutrient storage that can be accumulated especially for larger birds.

47.4.3.2 Physiological challenges associated with fatty acids as fuels

Fats may be ideal fuels for weight-economizing migratory birds because of their high-energy density; however, the metabolism of stored fats requires protein-mediated transport at most every step of the way from adipocyte to mitochondria (Figure 47.5) which can inhibit the very high rates of fat metabolism required for flight (McWilliams et al., 2004; Guglielmo, 2018). Birds are able to enhance their use of stored fats during migratory flights in several important ways (McWilliams et al., 2004; Price, 2010; Guglielmo, 2018). First, they may change the "quality" of their fat stores to enhance the rate of mobilization and oxidation. Fatty acid composition of stored fat changes in birds during migration primarily in response to diet and to preferential metabolism of certain fatty acids (Pierce and McWilliams, 2005; Pierce and McWilliams, 2014) and such changes in fat quality can affect flight performance during long-duration flights (Price, 2010; Guglielmo, 2018; Carter et al., 2020; McWilliams et al., 2020). Second, birds may increase the membrane proteins involved in transporting lipids into cells from the carriers in the blood plasma and the activity of cellular enzymes and so increase the oxidative capacity to use fatty acids (Guglielmo, 2010, 2018). Third, the acquisition of fat stores in birds during migration is associated with a coincident increase in antioxidant capacity to protect against oxidative damage caused by fat storage and use during long-duration flights (Skrip et al., 2015a; Cooper-Mullin and McWilliams, 2016; Skrip and McWilliams, 2016).

47.4.3.3 Fat quality is dynamic, affected mostly by diet, and changes seasonally during migration

The fatty acid composition of natural foods eaten by birds in migration can be quite variable (Pierce and McWilliams, 2014), migratory birds discriminate between foods based

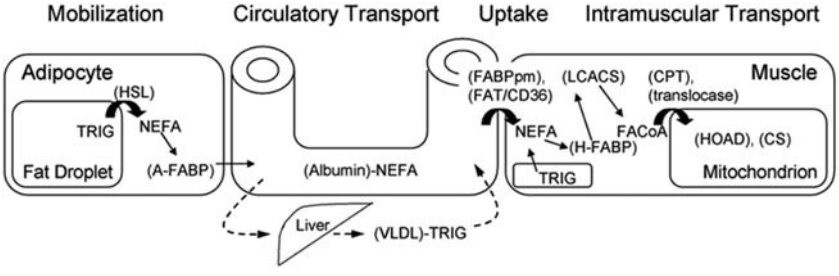

FIGURE 47.5 Lipid transport and oxidation in a bird during exercise while postabsorptive (from Price, 2010). Adipocyte triacylglycerol (TRIG) is hydrolyzed to nonesterified fatty acids (NEFAs) and glycerol through the action of hormone sensitive lipase (HSL). The extremely low-aqueous solubility of fatty acids requires the action of soluble protein carriers at every step of transport. These lipid transporters as well as key enzymes are shown in parentheses. Transport of NEFA within the adipocyte is accomplished by adipocyte fatty acid binding protein (A-FABP). Once exported from the adipocyte, NEFA are bound by plasma albumin and carried to the muscles. In addition to using fat stores as a source of fatty acids, the liver can synthesize many fatty acids de novo or use plasma NEFA to, by esterification to glycerol, produce TRIG packaged into very low-density lipoprotein (VLDL) for release into circulation. Note that the use of plasma VLDL as a source of fatty acids for working muscles requires hydrolysis by lipoprotein lipase (LPL, not shown) at the capillary endothelium (Ramenofsky, 1990; McWilliams et al., 2004). Uptake of fatty acids from circulation into muscle is facilitated by two important fatty acid transport proteins, fatty acid translocase (FAT/CD36) and a plasma membrane fatty acid binding protein (FABPpm). Once in the muscle cells, NEFAs are bound and intracellularly transported by heart-type fatty acid binding protein (H- FABP) which also serves to increase the rate of removal of NEFA from the membrane surface. In addition to these fatty acids sourced from adipocytes and liver, TRIG can also be directly stored within muscle and be hydrolyzed to NEFA and glycerol. Intracellular NEFA are converted by long chain acyl-CoA synthetase (LCACS) to acyl Co-A (FACoA). Uptake of FACoA into the muscle mitochondrion requires several biochemical conversions carried out by two forms of carnitine palmitoyltransferase (CPT) as well as a translocase. Once in the mitochondrial matrix, FACoA enters the β-oxidation pathway and citric acid cycle where key enzymes, including 3-hydroxyacyl-CoA dehydrogenase (HOAD) and citrate synthase (CS), lead to the aerobic production of CO_2, H_2O, and ATP.

on fatty acid composition (Bairlein, 1991; Zurovchak, 1997; Boyles, 2011; Pierce and McWilliams, 2014; Rios et al., 2014), and fatty acid composition of diet primarily determines that of stored fat at least for the mid-chain length fatty acids (e.g., 16:1 and 18:1, no. carbons:no. double bonds) and the essential fatty acids (e.g., 18:2n-6, 18:3n-3 along with their dominate elongation products, 20:4n-6 and 22:6n-3, respectively) with selective metabolism of certain fatty acids playing a possibly important but minor role (Blem, 1976; Pierce et al., 2005; Price and Guglielmo, 2009; Price, 2010). Note that conventional fatty acid nomenclature includes reference to the position of the double bonds relative to the methyl end of the molecule (e.g., n-6 refers to the first double bond at the sixth carbon from this end; n-6 is also referred to as omega-6 or ω-6) because the methyl end is not subject to elongation and desaturation and determines nutritional essentiality (Klasing, 1998). The implication is that migratory birds can select diets so as to achieve a certain fatty acid composition of their stored fat and muscle membranes and thereby satisfy the changing energy and nutritional demands across seasons, including that associated with endurance exercise during migration (Heitmeyer and Fredrickson, 1990; Conway et al., 1994; Bairlein, 1996; Maillet and Weber, 2006; Weber, 2009). Interestingly, behavioral preferences for certain dietary fatty acids (i.e., 18:1 and 18:2 over 18:0) were consistent across migration and nonmigration periods of the annual cycle (Pierce et al., 2004) and more recent studies suggest that migratory birds consistently prefer a 2:1 ratio of 18:1 to 18:2 (Boyles, 2011; Pierce and McWilliams, 2014). In sum, any observed seasonal changes in fatty acid composition of migratory birds are likely due primarily to diet and not to some endogenous seasonal change in diet preference for certain fatty acids or selective metabolism, although many more such studies are clearly needed.

In contrast to the composition of fat stores, the fatty acid composition of muscle and mitochondrial membranes is less affected by diet (Abbott et al., 2012). However, the apparently more consistent membrane composition conceals the fact that 95% of relevant membrane-bound fatty acids (i.e., 16:0, 18:2n-6) turn over on average every 10−17 days in volant birds, and at a faster rate than those in fat stores (Carter et al., 2019). Some studies have measured fatty acid composition of whole plasma [e.g., (Jensen et al., 2020)] or plasma fractions [i.e., NEFA, neutral lipids, phospholipids (Guglielmo et al., 2002c)] although we ignore such information here because seasonal changes in fatty acid composition of circulating lipids primarily reflect changes in feeding rate and diet composition rather than what is necessarily used during migratory flights when birds are fasting and must rely on stored fats (Guglielmo et al., 2002b, 2002c).

Seasonal changes in the fatty acid composition of fat stores in free-living migratory birds suggest that only a few fatty acids may be ecologically relevant and affect exercise performance (Pierce and McWilliams, 2014; Guglielmo, 2018), although given the wide variety of diets eaten by free-living birds during migration, and the relatively few such studies, such interspecific comparisons produce somewhat complicated trends (Table 47.1). In general, the 16- and 18-carbon fatty acids predominate (usually >75% of lipid stores) and the most common forms are usually the saturated 16:0 (no double bonds) and the monounsaturated 18:1n-9. During migration, 16:0 and 18:1n-9 still predominate although 16:1n-7 and two polyunsaturated fatty acids (PUFAs) considered essential for birds, mostly linoleic acid (18:2n-6) and small amounts of alpha-linolenic acid (18:3n-3), are often moderately abundant (up to 40%, but usually <20%) in the fat stores of landbirds. The fatty acid composition of fat stores of marine birds, mostly shorebirds (Napolitano and Ackman, 1990; Egeler and Williams, 2000; Maillet and Weber, 2006; Guglielmo, 2018) and waterfowl (Thomas and George, 1975; Heitmeyer and Fredrickson, 1990) studied to date, also includes much 16:0 and 18:1n-9 but can include up to ca. 20% of longer-chain 18:2n-6 and smaller amounts of 18:3n-3 PUFA (Table 47.1).

Seasonal changes in the fatty acid composition of flight muscle membrane phospholipids in free-living migratory birds confirm that only a few fatty acids may be ecologically relevant and affect exercise performance although much more information is needed (Pierce and McWilliams, 2005; Price, 2010; Guglielmo, 2018). Muscle membrane phospholipids are usually comprised of more saturated fatty acids (notably 16:0 and 18:0), less long-chain monounsaturated fatty acids (notably 18:1n-9), and more longer-chain PUFA (notably 18:2n-6, 20:4n-6, and 20:5n-3, 22:6n-3) compared to the fatty acid composition of fat stores (e.g., western sandpipers *Calidris mauri* (Egeler and Williams, 2000; Guglielmo et al., 2002c), white-throated sparrows (Klaiman et al., 2009), and semipalmated sandpipers (Maillet and Weber, 2006). Fatty acid composition of muscle membrane phospholipids changed seasonally in the two species studied to date (Table 47.1) with n-3 PUFAs (mostly 22:6n-3) decreasing and n-6 PUFAs (mostly 20:4n-6) coincidentally increasing in white-throated sparrows (Klaiman et al., 2009), whereas in western sandpipers, the n-3 PUFAs (mostly 22:5n-3 and 22:6n-3) increased and n-6 PUFAs (mostly 20:4n-6) coincidentally decreased (Guglielmo et al., 2002c), and there was a modest increase in 18:0 during migration in both species. Klaiman et al. (2009) suggest that these differences between species in the reciprocal change in long-chain n-3 and n-6 fatty acids in muscle phospholipids during migration were because of diet changes (to more enriched n-3

TABLE 47.1 Fatty acid composition (%) of subcutaneous fat (including neutral lipids) or whole carcass or muscle phospholipids (structural membranes) for migratory birds captured during both the nonmigration (i.e., breeding or winter) and migration (fall or spring migration) period of the annual cycle.

		Fatty acid composition of tissue(s)							Hypothesized patterns					
Species	Season	14:0	16:0	16:1	18:0	18:1n-9	18:2n-6	18:3n-3	% unsaturated	18:1 / 18:2	16:1&18:1 / 18:2	n-6[a] / n-3	Tissue	Source
Slate-colored Junco[b] (*Junco hyemalis*)	Spring migration	0.6	19.2	3.1	7.3	33.7	31.1		67.9	1.1	1.2		Whole animal	1
			↑			↑	↓		-	-	-			
	Winter	0.7	14.3	3.1	6.2	27.5	41.8		72.4	0.7	0.7		Whole animal	1
White-crowned Sparrow (*Zonotrichia leucophrys*)	Spring migration	31.8	16.6	4.2	5.8	24.4	10.6	1.6	39.2	2.3	2.7	6.6	Whole animal	2
		↑	-	-	-	-	↓	-	-	-	-	-		
	Winter	1.5	22.3	2.8	11.6	31.1	29.3	1.0	63.2	1.1	1.2	29.3	Whole animal	2
Wood Thrush (*Hylocichla mustelina*)	Fall migration		15.2	2.4	7.6	61.6	7.6	1.3	71.6	8.1	8.4	2.6	Whole animal	3
			↓		↓	↑	↓	↓	-	-	-	-		
	Breeding		24.0	4.7	13.8	34.4	12.0[c]	3.9	55.0	2.9	3.3	2.3	Whole animal	3
Red-eyed Vireo (*Vireo olivaceous*)	Fall migration		17.5	28.0	2.5	32.8	16.1	0.2	76.9	2.0	3.8	80.5	Subcutaneous fat	4
			↓	↑		↓		↓	-	-	-	-		
	Breeding		29.9	2.1	6.9	39.7	14.4	4.2	56.2	2.8	2.9	3.4	Subcutaneous fat	4
White-throated Sparrow (*Zonotrichia albicollis*)	Fall & Spring migration[d]		13.5	2.1	5.9	34.0	38.0	4.1	74.1	1.0	1.1	9.3	Subcutaneous fat	5
			↓	↓			↑		↑	↓	↓			
	Winter		21.5	5.0	7.2	32.8	24.9	3.4	62.7	1.5	1.7	7.3	Subcutaneous fat	5
	Fall & Spring migration[d]		18.0		25.5	6.0	15.0[e]	23.0[f]	51.5	0.4	0.4	0.9	Muscle phospholipids	5
					↑		↑	↓				↑		
	Winter		22.1		22.0	5.9	8.1[e]	27.9[f]	50.0	0.7	0.7	0.5	Muscle phospholipids	5
Western Sandpiper[g] (*Calidris mauri*)	Spring migration	4.0	33.8	11.5	10.9	29.6	2.2	1.9	47.3	13.5	18.7	1.0	Subcutaneous fat	6
			↓		↓	↑			↑	-	-	-		
	Winter	6.0	36.3	8.1	17.6	18.7	2.0	2.0	34.8	9.3	13.4	0.9	Subcutaneous fat	6
Western Sandpiper[g]	Spring migration		13.8	0.6	25.7	11.9	22.3[h]	20.3[i]	59.6	6.6	6.9	1.6[j]	Muscle phospholipids	7
					↑	↑	↓	↑						
	Winter		16.4	0.2	21.4	10.4	29.2[h]	14.6[i]	61.0	8.6	8.8	2.4	Muscle phospholipids	7
Canada Goose (*Branta canadensis*)	Spring migration	0.3	24.7	4.1	4.9	45.7	19.1	0.9	68.9	2.4	2.6	22.3	Subcutaneous fat	8
						↓	↑		-	-	-	-		
	Breeding	0.2	24.3	2.5	5.1	54.5	12.8	0.6	69.8	4.3	4.5	23.3	Subcutaneous fat	8
Mallard	Spring migration	1.0	17.1	2.9	6.0	51.9	19.1	2.1	74.9	2.7	2.9	9.1	Peritoneal fat	9
			↓	↓		↑			-	-	-	-		
	Winter	1.3	21.0	3.4	7.0	44.8	18.8	3.4	68.3	2.4	2.6	5.5	Peritoneal fat	9

Only studies that measured fatty acid composition in two or more seasons (e.g., migration, nonmigration periods) are included. Fatty acid nomenclature is number of carbon atoms to number of double bonds, and the location of the first double bond from the methyl end of the fatty acid (e.g., 18:2n-6 has 18 carbons in its backbone, two double bonds, and the first double bond is at the sixth carbon from the methyl end). Fatty acids that were sometimes detected but represent <3% across all studies are excluded (i.e., 18:1n-7, 20:0, 20:3n-6, 20:5n-3, 22:5n-3) except where noted. Given the fatty acid composition of tissues, several hypotheses have been proposed to explain the pattern of change in composition during

Continued

TABLE 47.1 Fatty acid composition (%) of subcutaneous fat (including neutral lipids) or whole carcass or muscle phospholipids (structural membranes) for migratory birds captured during both the nonmigration (i.e., breeding or winter) and migration (fall or spring migration) period of the annual cycle.—cont'd

migration. Arrows denote significant changes in fatty acid composition during migration as reported by the original authors (lack of an arrow or dash indicates no significant difference between migration vs. nonmigration periods for a given species). Dashes between migration versus nonmigration rows for a given species indicate no statistical comparison was reported in the original study.

[1] Bower and Helms, 1968.
[2] Morton and Liebman, 1974.
[3] Conway et al., 1994.
[4] Pierce and McWilliams, 2005.
[5] Klaiman et al., 2009.
[6] Egeler and Williams, 2000.
[7] Guglielmo et al. (2002a, b, c).
[8] Thomas and George, 1975.
[9] Heitmeyer and Fredrickson, 1990.

[a] Combined n-6 fatty acids (18:2n-6, 20:4n-6) and n-3 fatty acids (18:3n-3, 20:5n-3, 22:5n-3, 22:6n-3).
[b] Proportion of 18:3n-3 was not reported by season.
[c] in addition, 20:4n-6, another long-chain n-6 PUFA, comprised 3.9% and 2.0% of total fatty acid composition in breeding versus fall migration periods, respectively.
[d] Birds were sampled in both a fall and spring migration period, but combined here (average presented) because differences between these migration periods were relatively uncommon (exceptions for common fatty acids (>2% of total lipids): 18:0 and 18:2n-6 in muscle phospholipids, 18:3n-3 in adipose, and 18:2n-6 in intramuscular neutral lipids were different between fall versus spring migration).
[e] In addition, 20:4n-6, another long-chain n-6 PUFA, comprised 8.1% and 7.5% of total fatty acid composition in winter versus migration (fall and spring) periods, respectively.
[f] Proportion of 22:6n-3, another long-chain n-3 PUFA, is presented because 18:3n-3 comprised <0.5% of total fatty acid composition.
[g] Males and females were separately measured during winter (December in Panama), spring migration (May in British Columbia), and fall migration (July in British Columbia), but the sexes were averaged here. Fatty acid composition of stored fat (Egeler and Williams, 2000) and pectoralis muscle phospholipids (Guglielmo et al., 2002a, b, c) during spring and fall migration were not significantly different so only fall migration is presented.
[h] Proportion of 20:4n-6, another long-chain n-6 PUFA, is presented because 18:2n-6 comprised <1.9% of total fatty acid composition and unlike 20:4n-6 did not change seasonally.
[i] proportion of the combined longer-chain n-3 PUFAs (i.e., 20:5 + 22:5 + 22:6) is presented because 18:3n-3 comprised <0.5% of total fatty acid composition. Significant seasonal changes were detected in only 22:5n-3 and 22:6n-3, and not 18:3n-3 or 20:5n-3.
[j] The n-6/n-3 ratio for birds in fall migration was similar to that during spring migration (presented here), but was significantly lower (1.0) than during winter.

marine prey for sandpipers, to more enriched n-6 seeds for sparrows). Clearly more such studies are needed to determine if the general pattern of reciprocal change in long-chain n-3 and n-6 PUFAs in flight muscle membrane phospholipids is robust across birds species with different migration strategies that inhabit terrestrial and/or marine ecosystems.

47.4.3.4 Physiology of protein storage and flight muscle preparation

Lean mass typically accounts for approximately one quarter of the body mass gain in passerine birds prior to migration (Klaassen and Biebach, 1994; Klaassen et al., 1997; Bauchinger and Biebach, 2001) and on average ~50% of body mass gain in several species of waders (reviewed in Lindström and Piersma, 1993). Much of the lean mass gain observed in free-living birds has been attributed to increases in the size of the gut and organs associated with flight, i.e., heart and pectoralis (flight) muscle (Marsh, 1984; Dietz et al., 1999; reviewed in Lindström and Piersma, 1993; McWilliams et al., 2004). Several hypotheses have been put forth to explain the increase in lean mass and muscle hypertrophy prior to migration including the increased power needed for flight with a larger body mass (Marsh, 1984), as a source for gluconeogenesis (Jenni and Jenni-Eiermann, 1998; Guglielmo et al., 2017) and a storage mechanism for water (Gerson and Guglielmo, 2011) (see Protein Use section below). For pectoralis (flight) muscle in particular, studies suggest the increase in size of the pectoralis muscle is the result of a concomitant increase in body size (Marsh, 1981), a result of power training before flight (Marsh and Storer, 1981), or of an endogenously regulated process that occurs independent of training or size changes (Evans et al., 1992; Bishop et al., 1998; Dietz et al., 1999).

In addition to an overall mass increase in flight muscle, the mass-specific activities of key catabolic enzymes related to the oxidation of fatty acids, overall aerobic/anaerobic capacity, burst power, and glycolytic capacity increase in flight muscle prior to migration (Marsh, 1981; Lundgren and Kiessling, 1985; Banerjee and Chaturvedi, 2016). For example, hydroxyacyl-CoA-dehydrogenase (HOAD) concentration almost doubles in the flight muscle of gray catbirds (Marsh, 1981), and CS and HOAD activity increases in reed warblers, *Acrocephalus scirpaceus*, during fattening prior to migration (Lundgren and Kiessling, 1985). Additionally, Evans et al. (1992) showed that the percentage volume of mitochondria in muscle fibers in Dunlin, *Calidris alpina*, and Sanderlings, *Calidris alba*, increased from winter to spring migration suggesting an increased aerobic capacity of the flight muscle in preparation for migration. Banerjee and Chaturvedi (2016) concluded that the pectoral muscle of premigratory, red-headed buntings, *Emberiza bruniceps*, showed significant increases in aerobic and anaerobic capacity (CS, CCO, and MDH activity), increased fatty acid oxidation capacity [increased carnitine palmitoyltransferase (CPT) and HOAD activity], increased glycolytic capacity (increased PFK and PK activity), and increased burst power (CK activity) compared to nonmigratory buntings (Banerjee and Chaturvedi, 2016). Thus, although the dynamics of fat stores are more extreme than that of body protein, the turnover and dynamics of body protein has important functional significance (Bauchinger and McWilliams, 2012; Bauchinger and McWilliams, 2010; Carter et al., 2019).

47.4.4 Fuel use

47.4.4.1 Patterns of change in fatty acid composition of birds during migratory stage: proposed hypotheses

Understanding how the fatty acid composition of fat stores in free-living birds changes during migration periods provides a foundation upon which to generate ecologically relevant hypotheses, informed by comparative physiology, about how fatty acid composition can affect exercise performance of migratory birds. Given that specific unsaturated fatty acids are preferentially used and more rapidly mobilized during metabolism over saturated fatty acids (Leyton et al., 1987; Raclot and Groscolas, 1995; McKenzie et al., 1998; McKenzie, 2001; Raclot, 2003; Price et al., 2008), which compensates for their relatively lower energy content and potential for ATP production (Weber, 2009; Price, 2010), one proposed hypothesis is that the proportion of unsaturated fatty acids in fat stores will increase during migration. Both studies that directly tested this hypothesis (Table 47.1) found the predicted increase in unsaturation of fat stores (Egeler and Williams, 2000; Klaiman et al., 2009), and three of the other six species studied (wood thrush, *Hylocichla mustelina*, red-eyed vireos, and mallard, *Anas platyrhynchos*) support this hypothesis. Two of the three species (slate-colored junco, *Junco hyemalis*, Canada goose, *Branta canadensis*) had a relatively similar proportion of unsaturated fatty acids in their fat stores (Table 47.1) which led Guglielmo (2018) to conclude that the most general pattern observed to date was that the proportion of unsaturated fatty acids in fat stores of migratory birds increased or stayed the same during migration (the only exception thus far is the white-crowned sparrow which had unusually high proportions of 14:0 in its fat stores during migration).

The other alternative hypotheses that have been proposed to explain the patterns in the fatty acid composition of birds during migration have focused on the essential fatty acids (18:2n-6 and 18:3n-3) and their elongation products (e.g., 20:4n-6 and 22:6n-3,

respectively). Blem (1980) suggested that the ratio of 18:1 to 18:2 was higher in migratory birds. Results from five of the eight species studied to date (slate-colored junco, white-crowned sparrow, wood thrush, western sandpiper, and mallard) support this alternative hypothesis (Table 47.1). Pierce and McWilliams (2005) noted that the ratio of long-chain monounsaturated fatty acids (16:1, 18:1) to an essential long-chain PUFA (18:2) increased during migratory periods for most of the six species studied up to that point in time. Results from six of the eight species presented in Table 47.1 support this alternative hypothesis (Canada goose and white-throated sparrows were the exceptions). The potential importance of a certain ratio of n-3 and n-6 PUFAs to physiological function is long standing and is known to be strongly influenced by diet (Hulbert et al., 2005). For four of the species studied, n-3 PUFAs during both migration and nonmigration periods were <2% of all fatty acids in the whole animal (junco, white-crowned sparrow) or in separated fat stores (western sandpiper, Canada goose), and none of these four species showed the expected increase in n-6/n-3 ratio during the migration period (Table 47.1). For the other four species with more substantial n-3 fatty acids in their tissues (whole animal for wood thrush, fat stores for red-eyed vireo, white-throated sparrow, and mallard), the ratio of n-6/n-3 PUFAs consistently increased during migration compared to nonmigration periods (Table 47.1). As noted above, the changes in muscle phospholipids of birds during migration are modest compared to those observed in fat stores. The general pattern from the two studies conducted to date of reciprocal change between n-6 and n-3 PUFAs in muscle phospholipids and to a lesser extent 18:0 and 16:0 (but not fat stores; Table 47.1), provides little to no consistent support for any of the proposed hypotheses above.

Thus, the principle pattern that emerges from these few studies (Table 47.1) is that (1) there are seasonal changes in fatty acid composition of fat stores and muscle phospholipids that are associated with migration. For fat stores, the observed changes include (a) the proportion of unsaturated fatty acids either remain the same or increase during migration and (b) in particular, the relative amounts of the n-6 and n-3 PUFAs increase during migration (at least for those bird species with >2% n-3 PUFAs in their fat stores). For muscle phospholipids, reciprocal changes between primarily n-6 and n-3 PUFAs occur during migration although the direction of change differs between the two species studied to date. (2) These seasonal changes in fatty acid composition of fat stores and muscle phospholipids seem primarily driven by seasonal changes in diet, and the extent to which diet affects membrane composition is more modest than that of fat stores. Clearly, more studies are needed that systematically measure fatty acid composition of both fat stores (neutral lipids) and membranes (phospholipids) in several tissues [e.g., (McCue et al., 2009)] in a wide variety of migratory birds during migratory and nonmigratory seasons before these trends are confirmed and considered broadly applicable.

47.4.4.2 The oxidative costs of burning fat as fuel

Regulating oxidative balance is important for all air-breathing organisms such as birds because reactive pro-oxidant molecules can cause considerable cellular damage and affect health, longevity, and performance (Halliwell and Gutteridge, 1999), and PUFAs are especially susceptible to lipid peroxidation (Hulbert et al., 2007; Montgomery et al., 2012). This has led to a rich literature relating variation in membrane fatty acid composition to size-related variation in metabolic rate in mammals and birds (Hulbert and Else, 1999, 2000) and in turn to variation in reactive species (RS) production, life span, and aging (Harman, 1956; Speakman, 2005; Speakman and Selman, 2011; Speakman and Garratt, 2013; Herborn et al., 2016). In this context, birds have often been portrayed as exceptional vertebrates in that they display relatively high-metabolic rates, with the associated increased RS production, yet they are remarkably long-lived compared to mammals (Buttemer et al., 2010; Munshi-South and Wilkinson, 2010; Barja, 2014; Jimenez et al., 2019). Furthermore, birds use of fats as their primary fuel during high-intensity endurance exercise such as migratory flights (Jenni and Jenni-Eiermann, 1998; Guglielmo, 2010) has potential acute oxidative costs because fats, and especially PUFAs, are highly susceptible to oxidative damage (Cooper-Mullin and McWilliams, 2016; Skrip and McWilliams, 2016). As noted earlier, an important adaptation in Neoaves has led to the constitutive activation of the NRF2 master antioxidant response which lowers the risk of macromolecular oxidative damage in birds other than the fowl (e.g., chicken, ducks, geese (Castiglione et al., 2020). This constitutive activation of the antioxidant response may require birds to maintain a more robust and costly endogenous antioxidant system, although consumption of dietary antioxidants could reduce or alleviate these costs.

One of the challenges of studying how the antioxidant system of birds responds to the physiological challenges of migration is that the primary instigator of the oxidative challenge, increased RS production with increased metabolism, is not yet measurable in whole organisms (Costantini, 2014; Cooper-Mullin and McWilliams, 2016; Skrip and McWilliams, 2016). This must be inferred from measurements of key components of the antioxidant system (e.g., upregulation of antioxidant enzymes implies a response to increased RS production) and ideally simultaneous measures of RS-associated damage (e.g., upregulation

of lipid peroxidation products implies increased RS production that are insufficiently quenched by the antioxidant system). Another challenge of studying phenotypic flexibility in the antioxidant system of migratory birds is that the major site for generation of RS is the mitochondria which will vary in density and activity across tissues (Costantini, 2019) as well as with exercise and age (Cooper-Mullin and McWilliams, 2016). In addition, RS can act as both damaging molecules, as discussed above, and also as signaling molecules that can stimulate the endogenous antioxidant system (Halliwell and Gutteridge, 1999; Costantini, 2014, 2019). There are also a multitude of tests available to probe the response of the antioxidant system to ecologically relevant challenges which too often complicates comparisons across studies (Costantini, 2016, 2019; Skrip and McWilliams, 2016). Consequently, integrative studies (from mitochondria to cells to tissues to whole organism) that are also comparative (e.g., multiple tissues within the same individuals, migration state vs. nonmigration periods, multiple species that differ in migration strategy) are required to fully understand the antioxidant system of migratory birds within an ecological context. Given such integrative and comparative studies of migratory birds are largely lacking, evidence to date provides a more piecemeal view of how the antioxidant system of migratory birds changes during migration.

Does fattening and storage of oxidatively vulnerable PUFAs in preparation for migration increase RS production and an associated antioxidant response? Blackpoll warblers, *Dendroica striata*, and red-eyed vireos at a New England stopover site during fall migration (Skrip et al., 2015a), and garden warblers and barn swallows at a Mediterranean coastal stopover site during spring migration (Costantini et al., 2007), simultaneously increased their fat stores, nonenzymatic antioxidant capacity (plasma OXY), and oxidative damage (plasma d-ROMs) suggesting that fattening may incur oxidative costs and that the balance of capacity and damage may be condition-dependent. Captive-reared Northern wheatears, *Oenanthe pileata*, that were photostimulated into "migration state" in fall and that were rapidly refueling and hyperphagic increased their nonenzymatic antioxidant capacity (plasma OXY), consistent with the field studies described above, but not lipid peroxidation (red blood cell MDA) compared to control (*ad libitum*-fed) birds (Eikenaar et al., 2016). Several experimental and field studies suggest that the quality of fats consumed and stored in migratory birds affects circulating markers of oxidative damage. For example, European starlings, *Sturnus vulgaris*, fed for many months on diets with more or less n-6 PUFA had fuel stores composed of more or less n-6 PUFA, and those composed of more n-6 PUFA had consistently higher levels of plasma markers of oxidative damage (McWilliams et al., 2020). White-throated sparrows that consumed diets with more PUFA had increased circulating oxidative damage (d-ROMs) although when given choices between diets that differed only in available antioxidants (vitamin E) these birds did not prefer antioxidant-rich diets when consuming more dietary PUFA (Alan and McWilliams, 2013). Common blackbirds captured at a stopover site in northern Germany had a higher plasma fatty acid peroxidation index, higher nonenzymatic antioxidant capacity (OXY), but similar lipid peroxidation compared to sympatric resident blackbirds (Eikenaar et al., 2017a). The lack of correspondence between plasma fatty acid composition and that of fat stores and cell membranes in the same individuals (as described earlier) may explain why oxidative damage did not change with plasma peroxidation index in this latter study. Hudsonian godwits, *Limosa haemastica*, preparing for a very long-duration migratory flight increased total antioxidant capacity and reduced oxidative damage (TBARS) but did not change metabolic enzyme activity (Gutiérrez et al., 2019). In sum, migratory birds seem to build some component of their antioxidant capacity concomitantly with fat stores and increased oxidative damage may be an inevitable consequence of increasing (or maintaining more) fat stores, especially if they are composed of mostly PUFA.

During migratory flight, do birds upregulate antioxidant capacity and thus avoid the energetically expensive repair of damaged structures? In two recent reviews, Cooper-Mullin and McWilliams, 2016 and Skrip and McWilliams, 2016 reported that free-living birds during migration, or captive birds that flew for certain periods of time, often exhibit an increase in damage to lipids and/or proteins despite an often upregulated antioxidant system composed of antioxidant enzymes, nonenzymatic sacrificial molecules, and dietary antioxidants. For example, homing pigeons, *Columba livia*, that flew further (200 km) had more oxidative damage and less serum nonenzymatic antioxidant capacity than those that flew less far (60 km) when blood was sampled w/in 15 min of returning to the roost (Costantini et al., 2008). European robins, *Erithacus rubecula,* caught during nocturnal migratory flight had elevated markers of circulating oxidative damage to proteins (protein carbonyls) compared to resting robins even though the former also had higher circulating levels of glutathione peroxidase (GPx), an enzymatic antioxidant (Jenni-Eiermann and Jenni, 1991; Jenni-Eiermann et al., 2014). In contrast, migratory common blackbirds had higher nonenzymatic antioxidant capacity (OXY) but similar lipid peroxidation compared to sympatric resident blackbirds (Eikenaar et al., 2017a). Captive zebra finches, *Taeniopygia guttata*, flown for 2 h/day for many weeks increased the coordination between the enzymatic (GPx) and nonenzymatic (OXY) components of the antioxidant system while oxidative damage remained low and similar to sedentary finches (Cooper-Mullin et al., 2019). Plasma

markers of oxidative damage did not change in European starlings over the course of daily flying for several weeks, nor before and after a single long flight, in part because of upregulation of components of their antioxidant system (Frawley et al., 2021; McWilliams et al., 2020). Thus, upregulation of the antioxidant system and, at least in one case, increased coordination of key components of the antioxidant system occurs during repeated long-duration flights, and in some but not all cases, this seems to avoid the potential increase in oxidative damage with exercise. These studies also provide an important reminder that the response of individuals to an oxidative challenge involves an interacting, multicomponent antioxidant system (Costantini, 2019).

Given that some oxidative damage may occur during a migratory flight or enforced exercise, are migratory birds able to recover from this damage in an appropriately short period of time? Migratory birds at stopover sites seem to be able to in part recover from any oxidative damage that may have occurred during migration. For example, European robins that rested during the day had lower protein damage levels (protein carbonyls) than those caught during active nocturnal flight presumably because they had time to remove or repair damaged proteins (Jenni-Eiermann and Jenni, 1991). Individual Garden Warblers sampled repeatedly at a spring stopover reduced circulating lipid peroxidation levels over time (Skrip et al., 2015a). Thus, if oxidative damage occurs, then migratory birds seem capable of repairing this damage in a relatively reasonable amount of time (i.e., days not weeks) given their usual stopover durations, although many more such studies are needed that track recovery of individuals after long-duration flights.

Does consumption of dietary antioxidants affect the upregulation of the endogenous antioxidant system associated with migratory flights? In theory, the costs of upregulating the endogenous antioxidant system could be reduced if dietary antioxidants could quench RS producing during flight (Costantini et al., 2010a; Pamplona and Costantini, 2011; Costantini, 2014). Recent experimental evidence demonstrated for the first time that an ingested fat-soluble dietary antioxidant (Vitamin E) was absorbed and transported to the mitochondria in the flight muscles of a songbird within 22.5 h, but only if the birds were regularly flying each day (Cooper-Mullin et al., 2021). Thus, actively migrating songbirds that ingest dietary antioxidants during the day at a stopover, in this case fat-soluble antioxidants, would have these available in their mitochondria to protect against RS produced during subsequent nocturnal flight(s). Several experimental studies have directly manipulated the amount and type of dietary antioxidant(s) available to migratory birds, or captive birds exposed to regular exercise, and then measured the response of the antioxidant system often in relation to some ecologically relevant challenge (e.g., flight, high-PUFA diets). For example, European starlings fed diets supplemented or not with anthocyanins had similar plasma nonenzymatic antioxidant capacity (OXY) and oxidative damage (d-ROMs), but circulating uric acid was higher in individuals fed the low-antioxidant diet (Frawley et al., 2021). Likewise, zebra finches fed more or less anthocyanins and exercised (daily 2-h flying for months) or not had similar plasma nonenzymatic antioxidant capacity (OXY), antioxidant enzyme activity in liver (GPx, catalase, superoxide dismutase) and oxidative damage (d-ROMs); plasma uric acid was not measured in this study (Skrip et al., 2016). The suggestion from these studies is that when birds consume too little dietary antioxidants, this may not directly affect upregulation of antioxidant enzymes but may increase protein catabolism and thus circulating uric acid when flying for long durations and/or consuming high-PUFA diets. There is also evidence that consumption and physiological use of dietary antioxidants in birds may depend on their nutritional needs, energy demands, and type of dietary antioxidant (Costantini et al., 2010b; Beaulieu and Schaefer, 2013). Clearly, much more work is needed to adequately understand the role of dietary antioxidants in enabling migratory birds to maintain oxidative balance.

47.4.4.3 Carry over effects from winter to breeding

For the many birds that breed in seasonal environments, migration to more benign areas during winter has evolved along with the need to acquire energy and nutrient stores to fuel migration and, in the spring, also prepare for breeding. Life-history theory predicts that major annual cycle events such as breeding and migration are separated in time (and often space) to effectively distribute the total costs over time and to maximize the benefit-to-cost ratio given seasonal changes in resources (Lack, 1954; Dingle, 1996). However, such temporally separated events are not thereby independent in that, for example, condition of animals during one annual cycle event (e.g., in winter or on migration) can affect subsequent events (e.g., migration, reproduction). These so-called carry-over effects (Marra et al., 1998; Holmes, 2007; Harrison et al., 2011; Mitchell et al., 2011; Drake et al., 2013; Paxton and Moore, 2015) reveal links between annual cycle events that affect populations and individual fitness (Harrison et al., 2011). It has long been recognized that the extent of nutrient acquisition during spring, often prior to initiating migration, largely determines reproductive success in many waterfowl species (Ryder, 1970; Drent and Daan, 1980; Alisauskas and Ankney, 1992). As such, waterfowl provide many examples of the importance of carry-over effects (Harrison et al., 2011), the acquisition and storage of nutrient reserves at

one time or place (e.g., during the nonbreeding period) that are then used as resources at a later time and place (e.g., for breeding) (Warren et al., 2013; Sedinger and Alisauskas, 2014). Knowing the location and timing of nutrient storage during the nonbreeding period often has important conservation implications (Raveling and Heitmeyer, 1989; Abraham et al., 2005; Stafford et al., 2014; Alisauskas and Devink, 2015). Despite the importance of carry-over effects especially in migratory animals, a major gap in our understanding of these effects involves identifying the proximate causal links. All previous work has focused on how energy in the form of fat, protein, or carbohydrate stores may provide such linkages, whereas no previous studies have focused on antioxidants even though they have been proposed as an important currency that underpins life history tradeoffs (Catoni et al., 2008; Alonso-Alvarez et al., 2010) and potentially carry-over effects (Harrison et al., 2011).

Dietary antioxidants are likely an important, yet understudied, currency for carry-over effects in birds especially during spring migration for primarily three reasons. First, dietary antioxidants play important roles during the reproductive season in that breeding effort of birds promotes oxidative damage (Alonso-Alvarez et al., 2010) and domestic fowl supplemented with dietary fat-soluble antioxidants gain fitness advantages (Koutsos et al., 2003; Royle et al., 2003; McGraw et al., 2005; Williamson et al., 2006). Additionally, developing egg embryos are protected from external oxidative challenges if the females are able to deposit antioxidants in the yolk (Watson et al., 2018). Consistent with this important role of dietary antioxidants during reproduction, European starlings fed high-anthocyanin diets during spring had more elevated testosterone and more sustained breeding behaviors compared to individuals fed less antioxidants (Carbeck et al., 2018). Second, the increased metabolic rate associated with long-duration migratory flights produces pro-oxidants that must be quenched to avoid damage (Costantini, 2008; Costantini et al., 2007, 2008), and for spring migrating birds, this may reduce the antioxidant capacity available for subsequent reproduction. Importantly, dietary anthocyanins can support the metabolic function of glucocorticoids (corticosterone) and so control potential negative effects of excessive secretion of these hormones especially during long-duration flights (Casagrande et al., 2020). Third, fat-soluble antioxidants can be deposited in fat stores and so migratory birds could strategically store antioxidants prior to departure (Costantini et al., 2007), and balance their use during migratory flights so as to retain enough for investment into eggs that can subsequently enhance offspring survival and fitness (Blount et al., 2003; McGraw et al., 2005; Skrip et al., 2016). Clearly, much more study is needed to determine how female migratory birds contend with the oxidative costs of migratory flights in spring while also preparing for subsequent breeding and egg laying.

47.4.4.4 Fat quality matters

In theory, selectively eating and hence storing certain long-chain unsaturated fatty acids may be advantageous because (1) such fatty acids may be preferentially mobilized and metabolized more quickly (fuel hypothesis); (2) such fatty acids may affect composition and key functions of lipid-rich cell membranes (membrane hypothesis); and (3) such fatty acids may stimulate key facets of aerobic metabolism such as stimulating expression of genes involved in fatty acid oxidation (signal hypothesis) (Figure 47.6). Below we briefly discuss aspects of each of these hypotheses and how they may relate to the observed effect on exercise performance of migratory birds. Other reviews should be consulted for more thorough discussion of these hypotheses and the evidence (Weber, 2009; Guglielmo, 2010; Price, 2010; Pierce and McWilliams, 2014; Guglielmo, 2018). Although we separately describe these three hypotheses for convenience, they are not mutually exclusive.

The fuel hypothesis states that enhanced exercise performance occurs because certain fatty acids are more quickly mobilized and metabolized, and this can occur in several ways (Figure 47.5): selective mobilization from adipocytes, enhanced transport to target tissues, selective uptake by and transport within muscle cells, and/or enhanced oxidation and production of ATP (McWilliams et al., 2004, Price, 2010; Guglielmo, 2018). Several studies across taxa have shown that mobilization of shorter chain SFAs and fatty acids with more double bonds (e.g., 18:2n-6) was more rapid from fat stores in rats, fish, and birds (Raclot and Groscolas, 1995; Sidell et al., 1995; Hulbert et al., 2005; Price et al., 2008; Guglielmo, 2018). Isotope tracer studies that track ingested nutrients to breath CO_2 confirm that long-chain unsaturated fatty acids are oxidized more rapidly than their saturated counterparts (e.g., 18:1 vs. 18:0) (McCue et al., 2010). Once mobilized from fat stores, fatty acids require solubilization and intracellular translocation by the family of fatty acid binding proteins (FABPs) such that an increase in the number and action of FABPs present in the cell membranes and cytosols of muscle cells increases oxidative capacity (Guglielmo et al., 1998, Guglielmo et al., 2002a; McFarlan et al., 2009; Guglielmo, 2010, 2018). Furthermore, within flight muscle cells, the rate of CPT catalysis (Figure 47.5) was relatively higher for fatty acids with more double bonds compared to their saturated forms (e.g., 18:2 and 18:3 vs. 18:0) (Price et al., 2011; Guglielmo, 2018). This evidence supports the fuel hypothesis in that certain fatty acids such as 18:2n-6

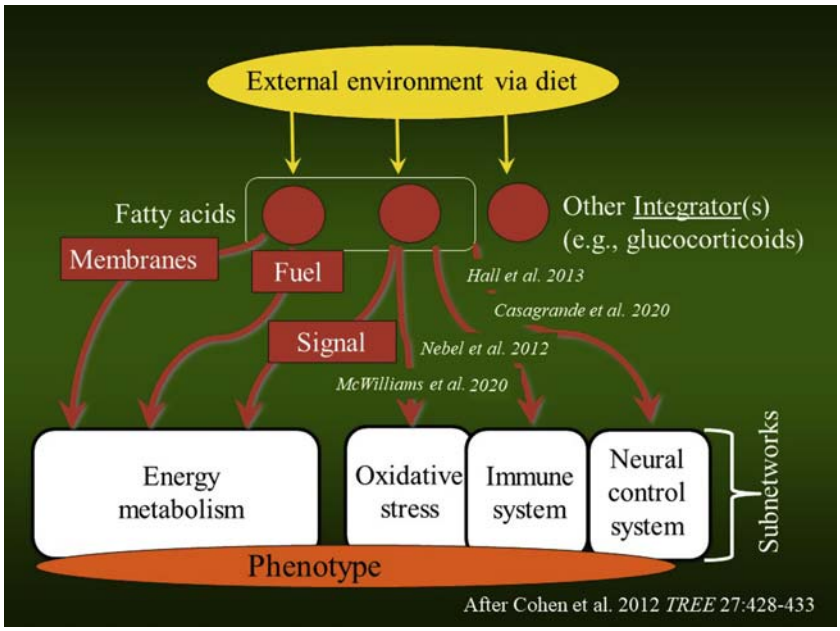

FIGURE 47.6 A simplified schematic of a physiological regulatory network that indicates how certain dietary fatty acids serve as key "integrators" that interact with multiple systems (subnetworks) and each other, thereby ensuring an appropriate match between phenotype and environmental conditions. Dietary fatty acids have been shown to influence the immune system (Nebel et al., 2012), neurogenesis (Hall et al., 2013), and neuroendocrine control of metabolism (Casagrande et al., 2020), and oxidative status (McWilliams et al., 2020). The three hypotheses (membrane, fuel, and signal), and the evidence that supports or refutes them are described in the text. *After Cohen et al. (2012).*

are metabolized more quickly at many steps of fatty acid oxidation although whether this is primarily responsible for enhanced exercise performance during migration periods is not yet clear (Guglielmo, 2010; Price, 2010) and discussed in the next section.

The membrane hypothesis states that the fatty acid composition of membrane phospholipids can effect key aspects of membrane structure and function (Hulbert and Else, 1999; 2000; Hulbert et al., 2005) and thus exercise performance of an organism (Valencak et al., 2003; Maillet and Weber, 2006; Maillet and Weber, 2007; Weber, 2009; Price, 2010). As described above, fatty acid composition of diet influences the phospholipid composition of muscle membranes in vertebrates, and this effect of diet on membrane composition is especially strong for dietary n-3 and n-6 compared to other fatty acids (e.g., shorter-chained saturated fatty acids (Hulbert et al., 2005; Maillet and Weber, 2006; 2007). Accordingly, the fatty acid composition of cell and subcellular membrane phospholipids converged with that of diet in migrating shorebirds that consumed marine invertebrates laden with n-3 PUFAs (Maillet and Weber, 2006, 2007). An increase in membrane PUFAs has potential functional importance for exercising animals in that (1) n-3 and/or n-6 PUFAs increase the fluidity and permeability of cell membranes (Stillwell and Wassall, 2003; Weber, 2009) and (2) n-3 and/or n-6 PUFAs are known to influence the activity of membrane-bound proteins and enzymes (e.g., UCP and -ATPases) which could affect efficiency of aerobic respiration (Hulbert and Else, 2000; Infante et al., 2001; Hulbert et al., 2005; Maillet and Weber, 2006, 2007; Gerson et al., 2008).

The signal hypothesis states that certain dietary fatty acids (most notably the n-3 and n-6 PUFAs) directly act as natural ligands for receptors that regulate the expression of genes associated with lipid metabolism (McClelland, 2004; Bordoni et al., 2006; Maillet and Weber, 2006; 2007; Weber, 2009). In particular, PUFA directly bind to and regulate the activity of peroxisome proliferator-activated nuclear receptors (PPARs) which are well known to stimulate expression of genes involved in fatty acid oxidation (Hochachka and Somero, 2002; Pawar et al., 2002; Zhang et al., 2004; Narkar et al., 2008; Weber, 2009), although the specific mechanism of action depends on the type of PPAR (Feige et al., 2006). PPAR α and β stimulate fatty acid oxidation and transport whereas PPAR γ is involved in lipid storage and adipocyte differentiation (Desvergne and Wahli, 1999; Nagahuedi et al., 2009). PPARs exhibit high affinity for n-3 PUFAs (Bordoni et al., 2006) and the upregulation of rate-limiting enzymes involved in aerobic metabolism by certain fatty acids such as 18:2n-6 (Sidell et al., 1995; Egginton, 1996; McKenzie, 2001) enables more rapid production of ATP during exercise (Guglielmo et al., 1998, 2002a; McWilliams et al., 2004). The effects of PUFAs on gene expression depend largely on the cellular concentration of PUFA, although it is not yet clear if a specific amount or ratio of dietary n-3 and n-6 fatty acids is optimal (Bordoni et al., 2006). The direct binding of certain dietary PUFAs to PPARs that then activate genes regulating lipid metabolism provides an example of "natural doping" whereby exercising animals can eat certain PUFAs, upregulate key aspects of lipid metabolism, and so enhance their exercise performance (Maillet and Weber, 2006, 2007, Weber, 2009). Such natural doping on n-3

PUFAs has been proposed for semipalmated sandpipers as they pause at key stopover sites during fall migration (Maillet and Weber, 2007). Whether this is a most interesting but unusual case in migratory birds deserves further investigations. In addition, these same essential PUFAs are used to build eicosanoids (e.g., prostaglandins, thromboxanes, leukotrienes, and lipoxins) that act locally on the cells that produce them or are nearby, and in this sense they are classified as hormones with signaling properties. At present, the importance of eicosanoids in birds during migration has not been directly studied and only proposed as an explanation (Klaiman et al., 2009; Carter et al., 2020), although their involvement in regulating a wide variety of physiological systems (e.g., immunity, development and growth, thermoregulation, and oviposition) suggests some attention may be worthwhile.

47.4.4.5 Testing the fuel, membrane, and signal hypotheses

Empirical evidence somewhat supports all of these alternative hypotheses although the weight of evidence to date seems to favor the fuel or signal hypotheses. The fuel hypothesis has been invoked to explain how fatty acid composition affects performance in rats, lizards, fish, and more recently migratory birds (Leyton et al., 1987; Geiser and Learmonth, 1994; Raclot and Groscolas, 1995; McKenzie et al., 1998; Wagner et al., 2004; Pierce et al., 2005; Price et al., 2008; Price and Guglielmo, 2009; Petersson et al., 2010). Our own recent work and that of colleagues has confirmed that songbirds with stored fat composed of more n-6 PUFA have improved exercise performance during short-term intense exercise (Pierce et al., 2005; Price and Guglielmo, 2009; Price, 2010). Price and Guglielmo (2009) used a cleverly designed sequence of feeding and fasting protocols to produce white-throated sparrows with fat stores and muscle membranes composed of different fatty acids. Consequently, they were able to demonstrate that the enhanced exercise performance of sparrows was associated with the fatty acid composition of fat stores rather than muscle membranes. The implied mechanism for the enhanced performance, in this case higher peak metabolic rates, included both faster mobilization rates of n-6 fatty acids from adipose as well as selective uptake of fatty acids into muscle cells and intramyocyte transport (Price and Guglielmo, 2009; Price, 2010). As correctly pointed out by Price (2010), a crucial untested assumption of this proposed mechanism for producing higher peak metabolic rates is that rate of fatty acid supply, and not some other factor such as oxygen supply or oxidative capacity, is the physiological limitation responsible for these whole-animal changes in exercise performance. In vivo studies that examine *selective* uptake and intracellular transport rates of certain fatty acids in muscle cells during exercise would be quite informative, especially if combined with diet and feeding regime manipulations that allow the effect of fatty acid composition of fat stores to be isolated from that of the other mechanisms.

Birds during migration, however, may be optimizing energy efficiency rather than maximizing metabolic rate and so may not need to maximize rate of fatty acid supply, as is often assumed. Energy efficiency in this case refers to flying further for a given amount of fuel energy or flying as far but expending less fuel energy. A possible mechanism for such enhanced efficiency that is consistent with the fuel hypothesis includes, for example, the documented higher transport rates of unsaturated fatty acids reducing the energy needed for transport of fatty acids (Price, 2010). Evidence in support of this enhanced flight efficiency idea comes from a study of European starlings from a migratory population in southern Germany that were fed for many months specially formulated diets that differed only in the amount of n-6 and n-3 fatty acids—the fatty acid composition of their fat stores was likewise different as expected. During the fall migration period, and after several weeks of flight-training in a wind tunnel, starlings composed of more n-6 (and n-3) PUFA used substantially less energy (ca. 11%) to fly the same distance (260 km) and duration (6 h) than those composed of more monounsaturated fatty acids (McWilliams and Pierce, 2006; McWilliams et al., 2020). This experimentally demonstrated that migratory birds can enhance fuel economy when composed of more n-6 PUFAs, a result consistent with the fuel hypothesis although it does not by itself definitively refute the other proposed hypotheses. As discussed further below, this energy savings gained during a long flight by starlings composed of more n-6 PUFA came at the long-term cost of higher oxidative damage in the n-6 PUFA-fed birds (McWilliams et al., 2020). This may explain why migratory birds seasonally shift their diet to increase consumption of n-3 and/or n-6 PUFAs during migration but then reduce their consumption of long-chain PUFAs during nonmigration periods.

Several lines of evidence suggest a link between the n-3 and/or n-6 PUFA content in membrane phospholipids and exercise performance. For example, maximal running speed in 36 species of mammal was strongly related to the n-6 PUFA content of their muscle membrane phospholipids (Ruf et al., 2006), n-6 PUFA in muscle membranes of rats was positively associated with their exercise performance (Ayre and Hulbert, 1997), and regular exercise increased PUFA content of muscle membranes in humans (Andersson et al., 2000). Also, PUFA-rich membranes have been found in various muscle types having high-aerobic capacity such as the pectoralis muscle in hummingbirds and the shaker muscle in rattlesnakes (Infante et al., 2001). Along with high levels of n-3 PUFAs, these muscle membranes had increased Ca^{2+}-ATPase activity

which may play a significant role in metabolism during exercise (Infante et al., 2001; Ruf et al., 2006). Furthermore, the activity of Na+/K+ -ATPase enzyme in the tissue membranes of birds and mammals has been correlated with the amount of 22:6n-3 in the membrane suggesting a causal link between certain types of PUFA and metabolic rate (Wu et al., 2001, 2004; Turner et al., 2003).

Studies of migratory birds provide some evidence in support of the membrane hypothesis in terms of effects of diet composition on membrane composition and in turn on cellular-level fat metabolism, although the evidence was not consistent (e.g., Maillet and Weber, 2007; Nagahuedi et al., 2009; Guglielmo, 2010; Dick and Guglielmo, 2019). Most studies to date that related membrane composition to whole-animal performance refuted the membrane hypothesis. For example, as discussed above, when Price and Guglielmo (2009) used a sequence of feeding and fasting protocols to produce white-throated sparrows with fat stores and muscle membranes composed of different fatty acids, they found that the enhanced performance (i.e., peak metabolic rate) was associated with the fatty acid composition of fat stores rather than muscle membranes. Dick and Guglielmo (2019) used diet manipulations to produce groups of yellow-rumped warblers that differed in the MUFA, n-3, and n-6 composition of flight muscle membranes. They found effects of membrane composition on activity of several flight muscle oxidative enzymes in warblers but not on multiple whole-animal performance measures including BMR, peak metabolic rate, and energy expenditure and duration of wind tunnel flights. Carter et al. (2020) fed European Starlings one of two diets that differed primarily in 18:2n-6 (reciprocally replaced with 16:0) and this in turn produced starlings with corresponding differences in fatty acid composition of fat stores and muscle membranes that were consistent over the 4-month experiment (Carter et al., 2020). Carter et al. (2020) found effects of membrane composition on activity of several flight muscle oxidative enzymes (Carter et al., 2021) but also on multiple whole-animal performance measures including BMR, peak metabolic rate, and rate of energy expenditure during >2-h wind tunnel flights. Specifically, birds with higher concentrations of 18:2n-6 in membranes and fat stores had higher BMR and peak metabolic rates, although this pattern was evident early in the fall and not later in the fall experiment. The change through time in performance measures but not membrane composition led Carter et al. (2020) to conclude that their results were most consistent with the signal hypothesis rather than the membrane hypothesis. Thus, evidence to date supports the membrane hypothesis in the sense that key metabolic enzymes and other suborganismal measures are affected by fatty acid composition of membranes; however, these studies have not consistently found the predicted effects on whole-animal exercise performance given differences in membrane composition alone.

The natural doping hypothesis proposes that birds (in this case shorebirds) during migration enhance their exercise performance by selecting high n-3 diets (marine prey such as *Corophium volutator* in the case of semipalmated sandpipers) and thereby (a) incorporate these PUFAs into membrane phospholipids and affect membrane function (membrane hypothesis), and (b) (signal hypothesis) binding of n-3 PUFAs to PPARs that then activate the expression and activity of key genes and enzymes that instigate the metabolic pathways involved in enhancing the oxidative metabolism of fatty acids (Maillet and Weber, 2006, 2007, Weber, 2009). Some support for the mechanisms proposed by the signal hypothesis comes from recent studies of Gray catbirds sampled throughout their annual cycle. During premigratory fattening periods, catbirds had increased mRNA expression of certain PPARs and key target genes (Corder et al., 2016; DeMoranville et al., 2019), and these PPAR isoforms from catbirds were activated by certain fatty acids (i.e., 18:1, 20:5n-3) tested on mammal cell lines (Hamiliton et al., 2018). Whether dietary n-3 fatty acids (and n-6 PUFA) act as important natural ligands and so determine these seasonal changes in PPAR expression in migratory catbirds remains unknown (Corder et al., 2016; DeMoranville et al., 2019). In one of the most comprehensive and relevant tissue-to-whole animal experimental studies of migratory birds to date, yellow-rumped warblers fed more n-3 PUFAs and that had membranes composed of more n-3 PUFA *decreased* PPAR-β mRNA abundance, activity of muscle oxidative enzymes, and overall oxidative capacity, and this was not associated with any whole-animal performance effects, in direct opposition to the natural doping, signal hypothesis (Dick and Guglielmo, 2019). The potential to enhance whole-animal performance as proposed by the signal hypothesis may also pertain to the essential n-6 PUFAs. As noted earlier, Carter et al. (2020) concluded that the results from their study of European starlings that were fed and composed of more n-6 PUFAs, and then flown for long durations in a wind tunnel, were most consistent with the signal hypothesis. These same starlings composed of more 18:2n-6 increased the expression of PPARα in the liver and LPL in the pectoralis; however, this occurred only in starlings that flew daily for 2 weeks in the wind tunnel and not untrained birds fed the same diet, and no such effect of diet was observed on the expression of other PPAR transcription factors, PGC-1 coactivators, and key metabolic genes (i.e., CD36, MCAD, CS, PLIN2, and avUCP) in the pectoralis muscle and the liver (DeMoranville et al., 2020). The dependency of the signaling effect of n-6 PUFA on flight training suggests something more complex than a simple direct diet influence.

The three proposed hypotheses (fuel, membrane, and signal) are not mutually exclusive, and each has garnered some support. Given that the use of fats as fuel requires storage, protein transport at many steps of the ways, up- and downregulation of metabolic enzymes (Figure 47.5), and that dietary fatty acids affect the composition of stored fat as well as membranes and can act as signaling molecules, we should not be surprised that this multifaceted fat metabolism system can be affected by diet and exercise in multiple ways. We know from both experimental and correlative studies that the fatty acid composition of diet largely determines that of fat stores and muscle membranes and that there is seasonal variation in the composition of the fat stores and membranes. Thus, the study of how dietary fatty acids affect fat metabolism at large as well as exercise performance in birds during migration presents a very tractable and ecologically relevant opportunity to test these various hypotheses. Especially intriguing and potentially informative are recent transcriptomic analyses (Dick, 2017) that revealed upregulation of key fatty acid metabolism pathways in captive yellow-rumped warblers in fall (vs. winter) while 45 other metabolic pathways were downregulated including those related to muscle growth, inflammation and immune function, and hormone signaling. The extreme challenges of migration may require such metabolic tradeoffs (Guglielmo, 2018), as has also been suggested in terms of an energy savings-oxidative cost tradeoff for birds during migration (McWilliams et al., 2020).

47.4.4.6 Fatty acid transport really matters

Metabolizing fatty acids at very high rates is quite difficult compared to the alternative, more soluble, carbohydrate and protein fuels used predominately by mammals during exercise (Guglielmo, 2010, 2018). As discussed in the next section, fatty acid oxidation in the flight muscles of migratory birds is enhanced by increasing the activity of key mitochondrial enzymes (e.g., HOAD and CPT) although the primary metabolic limitation seems more related to fatty acid transport through the circulation and its uptake by muscle cells (Guglielmo, 2010, 2018). The hypothesized mechanisms for effective and adequately rapid transport of fatty acids from fat stores into muscle cells in migratory birds has been extensively discussed elsewhere (McWilliams et al., 2004; Guglielmo, 2018) and above. In brief, migratory birds in general and birds during active migration, in particular, rely somewhat on augmented circulatory pathways to enhance fatty acid utilization, but the more important determinants of fatty acid flux seems to occur at the muscle cell level—specifically, protein-mediated transport of fatty acids across the muscle cell membrane [e.g., fatty acid translocase (FAT/CD36), plasma membrane fatty acid binding protein (FABPpm)] and within cells [e.g., heart-type fatty acid binding protein (H-FABP)] seems most likely to limit the overall rate of fatty acid metabolism in migratory birds (Figure 47.5). See Bishop and Butler (2015) for a more thorough treatment of this topic.

47.4.4.7 Protein use during flight and water balance

It is well established that birds catabolize lean mass as a protein source during flight with significant reductions in pectoralis muscle, leg muscle, digestive organs, and liver masses among other organs (Åkesson et al., 1992; Bauchinger and Biebach, 1998, 2001; Battley et al., 2000). Unlike lipids and carbohydrates, birds have no specific storage form for proteins (except very small endogenous amino acid pools). Proteins are stored in functional tissue such as flight muscle, digestive organs including the liver, and even skin (Marsh, 1984; Bauchinger and Biebach, 1998; Battley et al., 2000). Thus, the use of protein during flight causes a concomitant loss of structure and/or function. In red knots, *Calidris tenuirostris*, all organs (flight muscle, leg muscle, intestines, liver, kidney, stomach, heart, spleen, and skin) except brain and lungs decreased in mass during their long-duration flight from Australia to China (Battley et al., 2000). This lean mass loss in organs coincided with a 40% decrease in mass-specific BMR after migration suggesting a significant functional consequence in the organs contributing to BMR (Battley et al., 2000).

Several hypotheses explain why birds utilize protein during long-duration flights including the need for proteins as an adaptive reduction in muscle mass to conserve energy (Pennycuick, 1998), as intermediaries in the citric acid (Kreb's) cycle (Jenni and Jenni-Eiermann, 1998; Jenni-Eiermann, 2017), in tissue maintenance and repair (Lindström and Piersma, 1993; Guglielmo et al., 2001), and as a water source (Gerson and Guglielmo, 2011; Jenni-Eiermann, 2017; Groom et al., 2019). It is well understood that exercising mammals use proteins as a fuel source for gluconeogenesis and as intermediaries in the citric acid cycle (Aragón, 1981; Dohm, 1986). Jenni and Jenni-Eiermann (1998) concluded that the use of protein for the maintenance of citric acid cycle intermediaries is important in permitting the high rates of fatty acid oxidation required to meet the increased metabolic demands of long-duration flight in migratory birds. Guglielmo et al. (2001) found elevated plasma creatine kinase (CK) in migrating western sandpipers, and bar-tailed godwits as compared to nonmigrants in the same year suggesting that birds undergo a moderate amount of flight-induced muscle damage during migration and therefore must repair this damage before continuing their migration. However, the extent to which this damage impacts the recovery time at stopover sites and ultimately the pace of migration is still unclear.

Models of water use during flight suggest that water may be an important factor in limiting flight duration (Carmi et al., 1992; Klaassen, 1996). Protein catabolism provides 5.9 times more water per kilojoule than lipid catabolism (Jenni and Jenni-Eiermann, 1998) and thus can provide an invaluable source of water for fasting, flying birds (Gerson and Guglielmo, 2011; Jenni-Eiermann, 2017; Groom et al., 2019). The rate of protein catabolism during flight has been shown to be directly related to ambient relative humidity. Gerson and Guglielmo (2011) found that Swainson's thrushes *Catharus ustulatus* flown in a wind-tunnel at moderately low-ambient humidity had greater lean mass loss, increased metabolic water production, and higher uric acid levels (indicative of protein catabolism) than birds flown in moderately high humidity. Additionally, Groom et al. (2019) found that lean mass loss in birds exposed to low and high-ambient humidity was independent of metabolic rate in Swainson's thrushes with flown birds having similar protein catabolism as birds at rest. Birds have been shown to be effective at controlling blood plasma volumes regardless of thermal dehydration or dehydration incurred during flight (Carmi et al., 1993). Gerson and Guglielmo (2011) suggest that birds may utilize the protein-for-water strategy in an effort to maintain osmotic homeostasis since the plasma osmolarity of the thrushes was unaffected by ambient humidity during flight.

Although several species of birds have been shown to be sensitive to dehydration in ambient temperatures below 20°C in wind-tunnel studies (Torre-Bueno, 1978; Giladi and Pinshow, 1999; Engel et al., 2006), studies of trans-Saharan migrants suggest that energy savings may supersede any limitations set by water loss as migrants fly at higher ambient temperatures and lower humidity than expected given our current understanding from wind tunnel studies (Schmaljohann et al., 2008, 2009). Notably, access to drinking water may be a factor in a bird's decision to depart a stopover site and continue migration. For example, Garden warblers at a spring migration stopover site in the Mediterranean Sea that were captured with large fat stores and then given no access to water, were more likely to depart that night (i.e., had the greatest zugunruhe activity) compared to warblers with similar fat loads and access to water as well as those with less fat loads regardless of access to water (Skrip et al., 2015b). The degree to which freely available water influences departure of birds from a stopover site deserves further study, especially given the impact of climate change on worldwide water distribution.

The amount of protein used during long flights and the sources of these proteins appears to also be influenced in part by the level of fat storage, at least in passerines. Garden warblers, pied flycatchers *Ficedula hypoleuca*, Willow warblers *Phylloscopus trochilus*, and Barn swallows consumed protein primarily from flight muscle when fat stores comprised greater than 20% of total dry body mass. However, when fat stores reached critically low levels (i.e., 5–10% of total dry body mass), protein catabolism greatly increased and the protein mass of all organs decreased with significant reductions in the digestive organs as well as flight muscle masses (Schwilch et al., 2002). This pattern of protein use suggests an adaptive response in birds to the interaction of fuel use, energy requirements, and flight capability. When fat stores are adequate, reduction in flight muscle coincides with reductions in overall body mass (thus conserving energy) without compromising flight capability. However, when fat stores are nearing depletion, protein requirements increase and so birds rely more heavily on protein stores in nonuse organs such as the digestive tract as well as flight muscle. Although this is an adaptive response to meeting the energetic needs of the bird during flight, it appears to be at the detriment of flight capability.

47.4.4.8 Rebuilding protein stores after flight

The loss of proteins from structural and functional tissue during flight naturally requires that birds replenish what has been lost prior to their next flight. Birds arriving at a stopover site replenish protein reserves first before storing fat (Atkinson et al., 2007). For example, mass gain in red knots at Delaware Bay, USA, comprised ~15% fat when birds were below 133 g and ~84% fat above 133 g suggesting that birds have a critical body mass level where they must first rebuild protein stores before they are able to begin refattening (Atkinson et al., 2007). Since much of the protein used during flight is taken from digestive organs, fasted birds must rebuild their guts before returning to maximum digestive capacity and food intake (McWilliams and Karasov, 2001; Gannes, 2002; Pierce and McWilliams, 2004; Muñoz-Garcia et al., 2012). Passerine birds require 1–3 days for their digestive system to fully recover from fasting (Karasov and Pinshow, 2000; McWilliams and Karasov, 2001; Gannes, 2002) and this recovery may be diet-dependent (Pierce and McWilliams, 2004; Muñoz-Garcia et al., 2012). For example, fasted and food restricted white-throated sparrows fed a fruit diet (low protein) were unable to increase food intake rates to pre-fasted levels and thus regain body stores in three days of refeeding whereas sparrows fed a grain diet (high protein) were able to fully recover food intakes to prefast levels and regain energy stores in 24 h (Pierce and McWilliams, 2004). Munoz-Garcia et al. (2012) fed blackcaps, *Sylvia atricapilla*, diets containing 3% (low) and 20% (high) protein with labeled leucine to determine the extent to which these birds relied on dietary (exogenous) or endogenous sources of protein to rebuild their guts. Blackcaps fed a low-protein diet incorporated less exogenous protein into their tissue than blackcaps fed the high-protein diet. Interestingly, blackcaps incorporated more

exogenous protein into their gut than their pectoral muscle but were unable to rebuild their intestines at a similar rate as the blackcaps fed a high-protein diet (Muñoz-Garcia et al., 2012). However, the rate of rebuilding of protein stores (i.e., gut tissue) in migrating blackcaps was influenced by the availability of drinking water when the water content of food was low (Mizrahy et al., 2011). Blackcaps with limited access to drinking water and fed food with low-water content had no increase in lean mass nor fat mass during a simulated stopover period whereas birds provided with unlimited access to drinking water and/or food with high-water content were able to gain lean and fat mass (Mizrahy et al., 2011). This finding, along with that of Skrip et al. (2015) discussed above, suggests that freely available water at stopover sites may play a much larger role than previously thought in enabling birds to rebuild energy and protein stores and complete migration. A full understanding of the interaction between the protein-for-water strategy and the availability of drinking water at stopover sites deserves further investigation.

47.4.5 Temperature regulation during flight

Much of what is known about the body temperature fluctuations of birds during flight has been from wind tunnel studies of captive birds in short duration flights (Aulie, 1971; Torre-Bueno, 1976; Hudson and Bernstein, 1981). For example, pigeons increase their core body temperature (hyperthermia) during short-duration wind tunnel flights at ambient temperatures between 25 and 29C (Aulie, 1971), and this hyperthermia seems to limit the length of flight bouts in pigeons and European starlings (Aulie, 1971; Torre-Bueno, 1976). More recently, studies on wild birds using implanted data loggers have found that hyperthermia may play a much greater role in limiting the duration of migratory flights than previously assumed (Guillemette et al., 2016, 2017); however, this may only be relevant for the larger aquatic species with continuous, fast-flapping flight (Guillemette et al., 2017). Common eiders allow their body temperatures to rise on average 1C by the end of a typical flight and spend a significant portion (36%) of their daily time budget behaviorally cooling their body temperatures from previous flight bouts (Guillemette et al., 2017). Thus, eiders are heat dumping in between migratory flights. Less is known about temperature regulation to avoid hyperthermia during migratory flight in passerines as there are no studies to our knowledge that have used data loggers to track migratory temperature fluctuations in songbirds. Given the few studies to date on free-living birds, it would be beneficial to investigate the role of hyperthermia in regulating the duration of flight in migratory birds, especially in light of climate change.

47.4.6 Flight at high altitude

Birds are preadapted to low-oxygen conditions since the design of their respiratory and circulatory systems makes their oxygen uptake highly efficient, although several aspects of their physiology enable regular flying at high altitude during migration (see Bishop and Butler, 2015 for a thorough treatment of this topic). For example, birds such as bar-headed geese, *Anser indicus*, that fly regularly at high altitudes produce more hemoglobin, more red blood cells, and more myoglobin (the hemoglobin equivalent in muscle cells) as they prepare for migration which enhances their blood's capacity to bind and transport oxygen (Scott and Milsom, 2006). In addition, relatively larger lungs, and more and deeper penetrating blood capillaries in their heart and flight muscles compared to other geese further facilitates oxygen transport. Bar-headed geese also have a special form of an enzyme (nicknamed COX) involved in energy production within their muscle cells (Scott et al., 2011).

47.5 Beyond systems

Migration is one of the most complicated life history stages given the diverse habitats traversed, distances covered, multiple routes, and daily fluctuations of climatic conditions to which birds are exposed in vernal and autumnal stages. To be successful, this life history stage requires flexibility of morphology, physiology, and behavior in tight coordination with environmental information that initially regulates preparation for the stage and subsequently influences the timing, duration, and amplitude or intensity of the response throughout the fueling and flight cycle and eventually termination. The endocrine system plays major roles integrating environmental information with the molecular and cellular physiology as presented here. But, questions remain including: (1) how does seasonal variation of resources and routes affect fueling and flight, (2) does the endocrine status of vernal and autumnal migrants affect their responses to the conditions of specific habitats *en route*, and (3) are the apparent seasonal differences in the endocrine pathways exerting similar effects on fueling or are there multiple ways in which such regulatory systems have evolved?

A fundamental aspect of the life history of migratory birds is the ability to flexibly modify their physiology to satisfy the changing demands associated with migration. Such physiological flexibility is possible only because the network structure of molecules and regulatory relationships that maintain and adjust homeostasis (i.e., the Physiological Regulatory Network, PRN *structure*) can remain the same while the concentrations and relative strengths of the

relationships (i.e., the PRN *state*) can change depending on conditions (e.g., age, seasons, and migration; Cohen et al., 2012). In this chapter, we have outlined key components of the PRN *structure* (e.g., Figure 47.6) and how the PRN *state* is adjusted within individuals during migration. However, our review also makes clear that there remain major gaps in our understanding of both (a) the network of specific molecules and regulatory relationships that maintain and adjust homeostasis (i.e., PRN *structure*) and especially (b) how the concentrations of key molecules and the relative strengths of certain regulatory relationships change with the context and the conditions of vernal and autumnal migration (i.e., PRN *states*). For example, the biochemical mechanisms involved in using fats as fuel, or those responsible for maintaining redox homeostasis and limiting oxidative damage, are usually considered distinct physiological systems (i.e., separate PRN *structures*) that dynamically respond to changing conditions. However, we have emphasized here that there are physiological linkages between fat metabolism and oxidative status that require a more systems-level, integrative approach to fully understand how migratory birds respond to their changing environment(s), and the conditions of vernal and autumnal migration. Furthermore, phenotypic flexibility of physiological traits requires that the capacity of a physiological system is matched to the prevailing demand but can be modulated in response to changes in demand as to provide some limited excess capacity ("enough but not too much") (Diamond, 1991; Hammond and Diamond, 1997; McWilliams and Karasov, 2014). The level of so-called "spare capacity" and the full extent of phenotypic flexibility and its required time course provide important insights into constraints on whole-animal performance, diet diversity, and ecological niche (Piersma and Gils, 2011; McWilliams and Karasov, 2014). Given this systems-level perspective, the following related questions remain outstanding: (1) for a given physiological system (e.g., fat metabolism, oxidative status, and immune function), what is the PRN *structure* for migratory birds and how does PRN *state* change within individuals during migration, (2) what are the key physiological linkages between these systems that influence the limits to animal performance, and (3) what is the time course for such phenotypic flexibility of key physiological traits given that birds during migration are often time-limited?

References

Abbott, S.K., Else, P.L., Atkins, T.A., Hulbert, A.J., 2012. Fatty acid composition of membrane bilayers: importance of diet polyunsaturated fat balance. Biochim. Biophys. Acta 1818, 1309–1317.

Abraham, K.F., Jefferies, R.L., Alisauskas, R.T., 2005. The dynamics of landscape change and snow geese in mid-continent North America. Global Change Biol. 11, 841–855.

Ainslie, P.N., Ogoh, S., 2010. Regulation of cerebral blood flow in mammals during chronic hypoxia: a matter of balance. Exp. Physiol. 95, 251–262.

Åkesson, S., Karlsson, L., Pettersson, J., Walinder, G., 1992. Body composition and migration strategies: a comparison between robins (*Erithacus rubecula*) from two stop-over sites in Sweden. Vogelwarte 36, 188–195.

Alan, R.R., McWilliams, S.R., 2013. Oxidative stress, circulating antioxidants, and dietary preferences in songbirds. Comp. Biochem. Physiol. 164B, 185–193.

Alisauskas, R.T., Ankney, C.D., 1992. The cost of egg laying and its relationship to nutrient reserves in waterfowl. In: Batt, B.D.J., Afton, A.D., Anderson, M.G., Ankney, C.D., Johnson, D.H., Kadlec, J.A., Krapu, G.L. (Eds.), Ecology and Management of Breeding Waterfowl. University of Minnesota Press, Minneapolis.

Alisauskas, R.T., Devink, J.-M., 2015. Breeding costs, nutrient reserves, and cross-seasonal effects. In: Savard, J.-P.L., Derksen, D.V., Esler, D., Eadie, J.M. (Eds.), Ecology and Conservation of North American Sea Ducks. CRC Press, Boca Raton, FL.

Alonso-Alvarez, C., Perez-Rodriguez, L., Garcia, J.T., Vinuela, J., Mateo, R., 2010. Age and breeding effort as sources of individual variability in oxidative stress markers in a bird species. Physiol. Biochem. Zool. 83, 110–118.

Anderson, A.M., Duijns, S., Smith, P.A., Friis, C., Nol, E., 2019. Migration distance and body condition influence shorebird migration strategies and stopover decisions during southbound migration. Front. Ecol. Evol. 7.

Andersson, A., Sjodin, A., Hedman, A., Olsson, R., Vessby, B., 2000. Fatty acid profile of skeletal muscle phospholipids in trained and untrained young men. Am. J. Physiol. Endocrinol. Metabol. 279, 744–751.

Aragón, J.J., 1981. Replenishment of citric acid cycle intermediates by the purine nucleotide cycle in rat skeletal musle. In: RW Estabroook, P.S. (Ed.), Biological Cycles: Current Topics in Cellular Regulation. Academic Press, London.

Atkinson, P.W., Baker, A.J., Bennett, K.A., Clark, N.A., Clark, J.A., Cole, K.B., Dekinga, A., Dey, A., Gillings, S., González, P.M., 2007. Rates of mass gain and energy deposition in red knot on their final spring staging site is both time-and condition-dependent. J. Appl. Ecol. 44, 885–895.

Aulie, A., 1971. Body temperatures in pigeons and budgerigars during sustained flight. Comp. Biochem. Physiol. Physiol. 39, 173–176.

Ayre, K.J., Hulbert, A.J., 1997. Dietary fatty acid profile affects endurance in rats. Lipids 32, 1265–1270.

Bairlein, F., 1991. Nutritional adaptations to fat deposition in the long-distance migratory Garden Warbler (*Sylvia borin*). In: Proceedings of the 20th International Ornithological Congress. Christ-church, NZ.

Bairlein, F., 1996. Fruit-eating in birds and its nutritional consequences. Comp. Biochem. Physiol. 113A, 215–224.

Bairlein, F., Gwinner, E., 1994. Nutritional mechanisms and temporal control of migratory energy accumulation in birds. Annu. Rev. Nutr. 14, 187–215.

Bairlein, F., Simons, D., 1995. Nutritional adaptations in migrating birds. Israel J. Ecol. Evol. 41, 357–367.

Baker, J.R., 1938. The evolution of breeding seasons. In: Debeer, G.R. (Ed.), Evolution: Essays on Aspects of Evolutionary Biology. Clarendon Press, Oxford, U.K.

Banerjee, S., Chaturvedi, C.M., 2016. Migratory preparation associated alterations in pectoralis muscle biochemistry and proteome in Palearctic–Indian emberizid migratory finch, red-headed bunting, *Emberiza bruniceps*. Comp. Biochem. Physiol. D 17, 9–25.

Barceló, G., Ríos, J.M., Maldonado, K., Sabat, P., 2016. Energetic costs and implications of the intake of plant secondary metabolites on digestive and renal morphology in two austral passerines. J. Comp. Physiol. B 186, 625–637.

Barja, G., 2014. The mitochondrial free radical theory of aging. Prog. Mol. Biol. Transl. Sci. 127, 1–27.

Battley, P.F., Piersma, T., Dietz, M.W., Tang, S., Dekinga, A., Hulsman, K., 2000. Empirical evidence for differential organ reductions during trans–oceanic bird flight. Proc. R. Soc. Lond. Ser. B Biol. Sci. 267, 191–195.

Bauchinger, U., Biebach, H., 1998. The role of protein during migration in passerine birds. Biol. Conserv. della Fauna 102, 299–305.

Bauchinger, U., Biebach, H., 2001. Differential catabolism of muscle protein in garden warblers (*Sylvia borin*): flight and leg muscle act as a protein source during long-distance migration. J. Comp. Physiol. B 171, 293–301.

Bauchinger, U., McWilliams, S.R., 2010. Extent of phenotypic flexibility during long-distance flight is determined by tissue-specific turnover rates: a new hypothesis. J. Avian Biol. 41, 1–7.

Bauchinger, U., McWilliams, S.R., 2012. Tissue-specific mass changes during fasting: the protein turnover hypothesis. In: McCue, M. (Ed.), The comparative physiology of fasting and starvation. Springer-Verlag, Berlin, pp. 193–206.

Bauchinger, U., Van't hof, T., Biebach, H., 2007. Testicular development during long-distance spring migration. Horm. Behav. 51, 295–305.

Bauchinger, U., Wohlmann, A., Biebach, H., 2005. Flexible remodeling of organ size during spring migration of the garden warbler (*Sylvia borin*). Zoology 108, 97–106.

Bauer, C.M., Graham, J.L., Greives, T.J., 2019. Corticosterone negative feedback is weaker during spring vs. autumn migration in a songbird (Junco hyemalis). Gen. Comp. Endocrinol. 280, 36–42.

Beaulieu, M., Schaefer, H.M., 2013. Rethinking the role of dietary antioxidants through the lens of self-medication. Anim. Behav. 86, 17–24.

Bentley, G.E., Ubuka, T., Mcguire, N.L., Calisi, R., Perfito, N., Kriegsfeld, L.J., Wingfield, J.C., Tsutsui, K., 2009. Gonadotrophin-inhibitory hormone: a multifunctional neuropeptide. J. Neuroendocrinol. 21, 276–281.

Berthold, P., Gwinner, E., Sonnenschein, E. (Eds.), 2003. Avian Migration. Spring-Verlag, Berlin, Germany.

Bishop, C.M., Butler, P.J., 2015. Flight. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Bishop, C.M., Butler, P.J., El Haj, A.J., Egginton, S., 1998. Comparative development in captive and migratory populations of the barnacle goose. Physiol. Zool. 71, 198–207.

Blanchard, B.D., Erickson, M.M., 1949. The Cycle of the Gambel Sparrow, vol. 47. University of California Publications in Zoology, pp. 255–318.

Blas, J., 2015. Stress in birds. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London, pp. 769–805.

Blem, C.R., 1976. Patterns of lipid storage and utilization in birds. Am. Zool. 16, 671–684.

Blem, C.R., 1980. The energetics of migration. In: Gauthreaux Jr., S.A. (Ed.), Animal Migration, Orientation, and Navigation. Academic Press, New York.

Blem, C.R., 1990. Avian energy storage. In: Power, M. (Ed.), Current Ornithology. Plenum Press, New York.

Blount, J.D., Metcalfe, N.B., Arnold, K.E., Surai, P.F., Devevey, G.L., Monaghan, P., 2003. Neonatal nutrition, adult antioxidant defences and sexual attractiveness in the zebra finch. Proc. Roy. Soc. B 270, 1691–1696.

Bonier, F., Martin, P.R., Jensen, J.P., Butler, L.K., Ramenofsky, M., Wingfield, J.C., 2007. Pre-migratory life history stages of juvenile arctic birds: costs, constraints, and trade-offs. Ecology 88, 2729–2735.

Bordoni, A., Nunzio, M.D., Danesi, F., Biagi, P.L., 2006. Polyunsaturated fatty acids: from diet to binding to PPARS and other nuclear receptors. Genes Nutr. 1, 95–106.

Boswell, T., 2005. Regulation of energy balance in birds by the neuroendocrine hypothalamus. J. Poultry Sci. 42, 161–181.

Boswell, T., Dunn, I.C., 2015. Regulation of the avian central melanocortin system and the role of leptin. Gen. Comp. Endocrinol. 221, 278–283.

Boswell, T., Dunn, I.C., 2017. Regulation of agouti-related protein and pro-opiomelanocortin gene expression in the avian arcuate nucleus. Front. Endocrinol. 8, 75–86.

Boswell, T., Hall, M.R., Goldsmith, A.R., 1993. Annual cycles of migratory fattening, reproduction and moult in European quail (*Coturnix coturnix*). J. Zool. Lond. 231, 627–644.

Boswell, T., Richardson, R.D., Seeley, R.J., Ramenofsky, M., Wingfield, J.C., Friedman, M.I., Woods, S.C., 1995. Regulation of food intake by metabolic fuels in white-crowned sparrows. Am. J. Physiol. Regul. Integr. Comp. Physiol. 269, R1462–R1468.

Bottje, W., 2015. Mitochondrial physiology. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Bower, E.B., Helms, C.W., 1968. Seasonal variation in fatty acids of the slate-colored junco (*Junco hyemalis*). Physiol. Zool. 41 (2), 157–168.

Boyles, M., 2011. Seasonal Diet Preferences for Fatty Acids Differ between Species of Migratory Passerine, are Affected by Antioxidant Level, and Relate to the Fatty Acid Composition of Wild Fruits (Masters in Environmental Science). University of Rhode Island.

Braun, E.J., 2015. Physiological challenges of migration. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Buehler, D.M., Piersma, T., 2008. Traveling on a budget: predictions and ecological relevance for bottlenecks in the annual cycle of long-distance migrants. Phil. Trans. Biol. Sci. 363, 247–266.

Buehler, D.M., Tieleman, B.I., Piersma, T., 2010. How do migratory species stay healthy over the annual cycle? A conceptual model for immune function and for resistance to disease. Integr. Comp. Biol. 50, 346–357.

Buntin, J.D., Ruzycki, E., Witebsky, J., 1993. Prolactin receptors in dove brain: autoradiographic analysis of binding characteristics in discrete brain regions and accessibility to blood-borne prolactin. Neuroendocrinology 57, 738.

Butler, P., 1991. Exercise in birds. J. Exp. Biol. 160, 233–262.

Butler, P.J., Woakes, A.J., 2001. Seasonal hypothermia in a large migrating bird: saving energy for fat deposition? J. Exp. Biol. 204, 1361–1367.

Buttemer, W.A., Abele, D., Costantinit, D., 2010. From bivalves to birds: oxidative stress and longevity. Funct. Ecol. 24, 971–983.

Carbeck, K.M., DeMoranville, K.J., D'amelio, P.B., Goymann, W., Trost, L., Pierce, B., Bryla, A., Dzialo, M., Bauchinger, U., McWilliams, S.R., 2018. Environmental cues and dietary antioxidants affect breeding behavior and testosterone of male European starlings (*Sturnus vulgaris*). Horm. Behav. 103, 36–44.

Carmi, N., Pinshow, B., Horowitz, M., Bernstein, M.H., 1993. Birds conserve plasma volume during thermal and flight-incurred dehydration. Physiol. Zool. 66, 829–846.

Carmi, N., Pinshow, B., Porter, W.P., Jaeger, J., 1992. Water and energy limitations on flight duration in small migrating birds. Auk 109, 268–276.

Carpenter, F.L., Hixon, M.A., 1988. A new function for torpor: fat conservation in a wild migrant hummingbird. Condor 90, 373–378.

Carter, W.A., Bauchinger, U., McWilliams, S.R., 2019. The importance of isotopic turnover for understanding key aspects of animal ecology and nutrition. Diversity 11, 1–20.

Carter, W.A., DeMoranville, K.J., Pierce, B.J., McMilliams, S.R., 2021. Dietary linoleic acid, antioxidants, and flight training influence the activity of oxidative enzymes in European Starlings (*Sturnus vulgaris*). J. Comp. Physiol. B Biochem. Syst. Environ. Physiol. 191, 357–370.

Carter, W.A., DeMoranville, K.J., Pierce, B.J., McWilliams, S.R., 2020. The effects of dietary linoleic acid and hydrophilic antioxidants on basal, peak, and sustained metabolism in flight-trained European starlings. Ecol. Evol. 10, 1552–1566.

Carter, W.A., Whiteman, J.P., Cooper-Mullin, C., Newsome, S.D., McWilliams, S.R., 2019. Dynamics of individual fatty acids in muscle fat stores and membranes of a songbird and its functional and ecological importance. Physiol. Biochem. Zool. 92, 239–251.

Casagrande, S., Demoranville, K.J., Trost, L., Pierce, B.J., Bryla, A., Dzialo, M., Sadowska, E.T., Bauchinger, U., McWilliams, S.R., 2020. Dietary antioxidants attenuate the endocrine stress response during long-duration flight of a migratory bird. Proc. Roy. Soc. B 287, 20200744.

Castiglione, G.M., Xu, Z., Zhou, L., Duh, E., 2020. Adaptation of the master antioxidant response connects metabolism, lifespan and feather development pathways in birds. Nat. Commun. 11, 2476.

Catoni, C., Peters, A., Schaefer, H.M., 2008. Life history trade-offs are influenced by the diversity, availability and interactions of dietary antioxidants. Anim. Behav. 76, 1107–1119.

Chandola, A., Pathak, V., 1980. Premigratory increase in circulating triiodothyronine/thyroxine ratio in the red-headed bunting (*Emberiza brunicaps*). Gen. Comp. Endocrinol. 42, 39–46.

Chernetsov, N., Mukhin, A., 2006. Spatial behavior Of European robins during migratory stopovers: a telemetry study. Wilson J. Ornithol. 118, 364–373, 10.

Chmura, H.E., Krause, J.S., Perez, J.H., Ramenofsky, M., Wingfield, J.C., 2020. Autumn migratory departure is influenced by reproductive timing and weather in an Arctic passerine. J. Ornith. 161, 779–791.

Cipollini, M.L., Levey, D.J., 1997. Secondary metabolites of fleshy vertebrate-dispersed fruits: adaptive hypotheses and implications for seed dispersal. Am. Nat. 150, 346–372.

Cipollini, M.L., Stiles, E.W., 1993. Fruit rot, antifungal defense, and palatability of fleshy fruits for frugivorous birds. Ecology 74, 751–762.

Cohen, A.A., Martin, L.B., Wingfield, J.C., Mcwilliams, S.R., Dunne, J.A., 2012. Physiological regulatory networks: ecological roles and evolutionary constraints. Trends Ecol. Evol. 27, 428–435.

Conway, C.J., Eddleman, W.R., Simpson, K.L., 1994. Seasonal changes in fatty acid composition of the wood thrush. Condor 96, 791–794.

Cooper-Mullin, C., Carter, W.A., Amato, R.S., Podlesak, D., McWilliams, S.R., 2021. Dietary antioxidants reach the mitochondria in the flight muscles of birds but only if they exercise. PLoS One 16(6), e0253264.

Cooper-Mullin, C., Carter, W.A., McWilliams, S.R., 2019. Acute effects of intense exercise on the antioxidant system in birds: does exercise training help? J. Exp. Biol. 222.

Cooper-Mullin, C., McWilliams, S.R., 2016. The role of the antioxidant system during intense endurance exercise: lessons from migrating birds. J. Exp. Biol. 219, 3684–3695.

Corder, K.R., Demoranville, K.J., Russell, D.E., Huss, J.M., Schaeffer, P.J., 2016. Annual life-stage regulation of lipid metabolism and storage and associated with PPARs in a migrant species: the gray catbird (*Dumetella carolinensis*). J. Exp. Biol. 219, 3391–3398.

Cornelius, J.M., Boswell, T., Jenni-Eiermann, S., Breuner, C., Ramenofsky, M., 2013. Contribution of endocrinology to the migration life history of birds. Gen. Comp. Endocrinol. 190, 47–60.

Costantini, D., 2008. Oxidative stress in ecology and evolution: lessons from avian studies. Ecol. Lett. 11, 1238–1251.

Costantini, D., 2014. Oxidative Stress and Hormesis in Evolutionary Ecology and Physiology: A Marriage between Mechanistic and Evolutionary Approaches. Springer-Verlag, Berlin, Heidelberg.

Costantini, D., 2016. Oxidative stress ecology and the d-ROMs test: facts, misfacts and an appraisal of a decade's work. Behav. Ecol. Sociobiol. 70, 809–820.

Costantini, D., 2019. Understanding diversity in oxidative status and oxidative stress: the opportunities and challenges ahead. J. Exp. Biol. 222.

Costantini, D., Cardinale, M., Carere, C., 2007. Oxidative damage and anti-oxidant capacity in two migratory bird species at a stop-over site. Comp. Biochem. Physiol. 144C, 363–371.

Costantini, D., Dell'Ariccia, G., Lipp, H.-P., 2008. Long flights and age affect oxidative status of homing pigeons (*Columba livia*). J. Exp. Biol. 211, 377–381.

Costantini, D., Metcalfe, N.B., Monaghan, P., 2010a. Ecological processes in a hormetic framework. Ecol. Lett. 13, 1435–1447.

Costantini, D., Rowe, M., Butler, M.W., Mcgraw, K.J., 2010b. From molecules to living systems: historical and contemporary issues in oxidative stress and antioxidant ecology. Funct. Ecol. 24, 950–959.

Covino, K.M., Holberton, R.L., 2011. The influence of energetic condition on flight initiation and orientation of migratory songbirds in the Gulf of Maine region. Auk 128, 313–320.

Covino, K.M., Jawor, J.M., Kelly, J.F., Moore, F.R., 2017. Overlapping life-history stages in migrating songbirds: variation in circulating testosterone and testosterone production capacity. J. Ornithol. 158, 203–212.

Covino, K.M., Morris, S.R., Moore, F.R., 2015. Patterns of testosterone in three Nearctic–Neotropical migratory songbirds during spring passage. Gen. Comp. Endocrinol. 224, 186–193.

Dawson, A., King, V.M., Bentley, G.E., Ball, G.F., 2001. Photoperiodic control of seasonality in birds. J. Biol. Rhythm. 16, 365–380.

Dekinga, A., Dietz, M.W., Koolhaas, A., Piersma, T., 2001. Time course and reversibility of changes in the gizzards of red knots alternately eating hard and soft food. J. Exp. Biol. 204, 2167–2173.

Delingat, J., Dierschke, V., Schmaljohann, H., Bairlein, F., 2009. Diurnal patterns of body mass change during stopover in a migrating songbird, the northern wheatear *Oenanthe oenanthe*. J. Avian Biol. 40, 625–634.

Delingat, J., Dierschke, V., Schmaljohann, H., Mendel, B., Bairlein, F., 2006. Daily stopovers as optimal migration strategy in a long-distance migrating passerine: the Northern Wheatear *Oenanthe oenanthe*. Ardea 94, 593–605.

DeMoranville, K.J., Carter, W.A., Pierce, B.J., McWilliams, S.R., 2020. Flight training in a migratory bird drives metabolic gene expression in the flight muscle but not liver, and dietary fat quality influences select genes. Am. J. Physiol. Regul. Integr. Comp. Physiol. 319, R637–R652.

DeMoranville, K.J., Corder, K.R., Hamilton, A., Russell, D.E., Huss, J.M., Schaeffer, P.J., 2019. PPAR expression, muscle size and metabolic rates across the gray catbird's annual cycle are greatest in preparation for fall migration. J. Exp. Biol. 222, 1–11.

DeSimone, J.G., Ramirez, M.G., Elowe, C.R., Griego, M.S., Breuner, C.W., Gerson, A.R., 2020. Developing a Stopover-CORT hypothesis: corticosterone predicts body composition and refueling rate in Gray Catbirds during migratory stopover. Horm. Behav. 124, 104776.

Desvergne, B., Wahli, W., 1999. Peroxisome proliferator-activated receptors: nuclear control of metabolism. Endocr. Rev. 20, 649–688.

Deviche, P., 1995. Androgen regulation of avian premigratory hyperphagia and fattening: from eco-physiology to neuroendocrinology. Am. Zool. 35, 234–245.

Diamond, J., 1991. Evolutionary design of intestinal nutrient absorption: enough but not too much. News Physiol. Sci. 6, 92–96.

Diamond, J.M., Karasov, W.H., Phan, D., Carpenter, F.L., 1986. Digestive physiology is a determinant of foraging bout frequency in hummingbirds. Nature 320, 62–63.

Diano, S., Naftolin, F., Horvath, T.L., 1997. Gonadal steroids target AMPA glutamate receptor-containing neurons in the rat hypothalamus, septum and amygdala: a morphological and biochemical study. Endocrinology 138, 778–789.

Dick, M.F., 2017. The Long Haul: Migratory Flight Preparation and Performance in Songbirds (Ph.D. dissertation). University of Western Ontario.

Dick, M.F., Guglielmo, C.G., 2019. Dietary polyunsaturated fatty acids influence flight muscle oxidative capacity but not endurance flight performance in a migratory songbird. Am. J. Physiol. Regul. Integr. Comp. Physiol. 316, R362–R375.

Dietz, M.W., Piersma, T., Dekinga, A., 1999. Body-building without power training: endogenously regulated pectoral muscle hypertrophy in confined shorebirds. J. Exp. Biol. 202, 2831–2837.

Dingle, H., 1996. Migration: The Biology of Life on the Move. Oxford University Press, New York.

Dingle, H., 2014. Migration, the Biology of Life on the Move. Oxford University Press, New York.

Dohm, G.L., 1986. 5 protein as a fuel far endurance exercise. Exerc. Sport Sci. Rev. 14, 143–174.

Dolnik, V.R., 1980. Regulyatsiya okonchaniya linki inachala osennei migratsii u molodykh zyablikov (*Fringilla coelebs*). Zool. Zh. 59, 91–98.

Drake, A., Rock, C., Quinlan, S.P., Green, D.J., 2013. Carry-over effects of winter habitat vary with age and sex in yellow warblers *Setophaga petechia*. J. Avian Biol. 44, 321–330.

Drent, R., Daan, S., 1980. Prudent parent: energetic adjustments in avian breeding. Ardea 68, 225–252.

Driedzic, W.R., Crowe, H.L., Hicklin, P.W., Sephton, D.H., 1993. Adaptations in pectoralis muscle, heart mass, and energy metabolism during premigratory fattening in semipalmated sandpipers (*Calidris pusilla*). Can. J. Zool. 71, 1602–1608.

Dudley, R., Vermeij, G.J., 1992. Do the power requirements of flapping flight constrain folivory in flying animals? Funct. Ecol. 6, 101–104.

Dykstra, C.R., Karasov, W.H., 1992. Changes in gut structure and function of house wrens (*Troglodytes aedon*) in response to increased energy demands. Physiol. Zool. 65, 422–442.

Dzialowski, E.M., Crossley II, D.A., 2015. The cardiovascular system. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Egeler, O., Williams, T.D., 2000. Seasonal, age, and sex-related variation in fatty-acid composition of depot fat in relation to migration in western sandpipers. Auk 117, 110–119.

Egevang, C., Stenhouse, I.J., Phillips, R.A., Petersen, A., Fox, J.W., Silk, J.R., 2010. Tracking of Arctic terns *Sterna paradisaea* reveals longest animal migration. Proc. Natl. Acad. Sci. U.S.A. 107, 2078–2081.

Egginton, S., 1996. Effect of temperature on optimal susbtrate for B-oxidation. J. Fish. Biol. 49.

Eikenaar, C., 2017. Endocrine regulation of fueling by hyperphagia in migratory birds. J. Comp. Physiol. 203, 439–445.

Eikenaar, C., Ballstaedt, E., Hessler, S., Klinner, T., Muller, F., Schmaljohann, H., 2018a. Cues, corticosterone and departure decisions in a partial migrant. Gen. Comp. Endocrinol. 261, 59–66.

Eikenaar, C., Fritzsch, A., Bairlein, F., 2013. Corticosterone and migratory fueling in Northern wheatears facing different barrier crossings. Gen. Comp. Endocrinol. 186, 181–186.

Eikenaar, C., Hegemann, A., 2016. Migratory common blackbirds have lower innate immune function during autumn migration than resident conspecifics. Biol. Lett. 12, 20160078.

Eikenaar, C., Hesslera, S., Ballstaedta, E., Schmaljohann, H., Kaiya, H., 2018b. Ghrelin, corticosterone and the resumption of migration from stopover, an automated telemetry study. Physiol. Behav. 194, 450–455.

Eikenaar, C., Jonsson, J., Fritzsch, A., Wang, H.-L., Isaksson, C., 2016. Migratory refueling affects non-enzymatic antioxidant capacity, but does not increase lipid peroxidation. Physiol. Behav. 158, 26–32.

Eikenaar, C., Kallstig, E., Andersson, M.N., Herrera-Duenas, A., Isaksson, C., 2017a. Oxidative challenges of avian migration: a comparative field study on a partial migrant. Physiol. Biochem. Zool. 90, 223–229.

Eikenaar, C., Klinner, T., Stowe, M., 2014. Corticosterone predicts nocturnal restlessness in a long-distance migrant. Horm. Behav. 66, 324–329.

Eikenaar, C., Muller, F., Leutgeb, C., Hessler, S., Lebus, K., Taylor, P.D., Schmaljohann, H., 2017b. Corticosterone and timing of migratory departure in a songbird. Proc. R. Soc. B 284, 20162300.

Eikenaar, C., Schmaljohann, H., 2015. Wind conditions experienced during the day predict nocturnal restleness in a migratory songbird. Ibis 157, 125–132.

Engel, S., Biebach, H., Visser, G.H., 2006. Water and heat balance during flight in the rose-colored starling (*Sturnus roseus*). Physiol. Biochem. Zool. 79, 763–774.

Evans, P.R., Davidson, N.C., Uttley, J.D., Evans, R.D., 1992. Premigratory hypertrophy of flight muscle: an ultrastructure study. Ornis Scand. 23, 238–243.

Falsone, K., Jenni-Eiermann, S., Jenni, L., 2009. Corticosterone in migrating songbirds during endurance flight. Horm. Behav. 56, 548–556.

Faraci, F.M., 1991. Adaptations to hypoxia in birds: how to fly high. Annu. Rev. Physiol. 53, 59–70.

Farner, D.S., Follett, B.K., 1966. Light and other environmental factors affecting avian reproduction. J. Anim. Sci. (Sofia) 25, 90–115.

Feige, J.N., Gelman, L., Michalik, L., Devergne, B., Wahli, W., 2006. From molecular action to physiological outputs: peroxisome proliferator-activated receptors are nuclear receptors at the crossroads of key cellular functions. Prog. Lipid Res. 45, 120–159.

Follett, B.K., King, V.M., Meddle, S.L., 1998. Rhythms and photoperiodsm in birds. In: Lumsden, P.J., Millar, A.J. (Eds.), Biological Rhythms and Photperiodsm in Plants. BIOS Scientiic Publishers Ltd, Oxford.

Follett, B.K., Mattocks JR., P.W., Farner, D.S., 1974. Circadian function in the photoperiodic induction of gonadotropin secretion in the white-crowned sparrow, *Zonotrichia leucophrys gambelii*. Proc. Natl. Acad. Sci. U.S.A. 71, 1666–1669.

Frawley, A.E., DeMoranville, K.J., Carbeck, K.M., Trost, L., Bryla, A., Dzialo, M., Sadowska, E.T., Bauchinger, U., Pierce, B.J., McWilliams, S.R., 2021. Season, anthocyanin supplementation, and flight training have mixed effects on the antioxidant system of migratory European Starlings. Ornithology 138, 1–16.

Friedman-Einat, M., Cogburn, L.A., Yosefi, S., Hen, G., Shinder, D., Shirak, A., Seroussi, E., 2014. Discovery and characterization of the first genuine avian leptin gene in the rock dove (*Columba livia*). Endocrinology 155, 3376–3384.

Friedman-Einat, M., Seroussi, E., 2019. Avian leptin: bird's-eye view of the evolution of vertebrate energy-balance control. Trends Endocrinol. Metabol. 30, 819–832.

Fry, C.H., Ferguson-Lees, I.J., Dowsett, R.J., 1972. Flight muscle hypertrophy and ecophysiological variation of yellow Wagtail *motacilla-Flava* races at lake Chad. J. Zool. 167, 293–306.

Fusani, L., Cardinale, M., Carere, C., Goymann, W., 2009. Stopover decision during migration: physiological conditions predict nocturnal restlessness in wild passerines. Biol. Lett. 5, 302–305.

Fuxjager, M.J., Barske, J., Du, S., Day, L.B., Schlinger, B.A., 2012. Androgens regulate gene expression in avian skeletal muscles. PLoS One 7, e51482.

Gannes, L.Z., 2002. Mass change pattern of blackcaps refueling during spring migration: evidence for physiological limitations to food assimilation. Condor 104, 231–239.

Gaunt, A.S., Hikida, R.S., Jehl, J.R., Fenbert, L., 1990. Rapid atrophy and hypertrophy of an avian flight muscle. Auk 107, 649–659.

Gavrilov, V.M., Dolnik, V.R., 1974. Bioenergetika i regulyatsiya poslebrachnoi i postyuvenaloi linek u zyablikov (*Fringilla coelebs coelebs* L.). Tr. Zool. Inst. Akad. Nauk SSSR 55, 254–267.

Geiser, F., Learmonth, R.P., 1994. Dietary fats, selected body temperature and tissue fatty acid composition of agamid lizards (*Amphibolurus nuchalis*). J. Comp. Physiol. 164B, 55–61.

Gerson, A., Brown, J.C., Thomas, R., Bernards, M.A., Staples, J.F., 2008. Effects of dietary polyunsaturated fatty acids on mitochondrial metabolism in mammalian hibernation. J. Exp. Biol. 211, 2689–2699.

Gerson, A.R., Guglielmo, C.G., 2011. Flight at low ambient humidity increases protein catabolism in migratory birds. Science 333, 1434–1436.

Giladi, I., Pinshow, B., 1999. Evaporative and excretory water loss during free flight in pigeons. J. Comp. Physiol. B 169, 311–318.

Goodridge, A.G., 1964. The effect of insulin, glucagon and prolactin on lipid synthesis and related metabolic activity in migratory and non-migratory finches. Comp. Biochem. Physiol. A 13, 1–26.

Goymann, W., Lupi, S., Kaiya, H., Cardinale, M., Fusani, L., 2017. Ghrelin affects stop-over decisions and food-intake in a long-distance migrant. Proc. Natl. Acad. Sci. Unit. States Am. 114, 1946–1951.

Goymann, W., Spina, F., Ferri, A., Fusani, L., 2010. Body fat influences departure from stopover sites in migratory birds: evidence from whole-island telemetry. Biol. Lett. 6, 478–481.

Greenewalt, C.H., 1960. Hummingbirds. Dover Publications, New York.

Groom, D.J.E., Deakin, J.E., Lauzau, M.C., Gerson, A.R., 2019. The role of humidity and metabolic status on lean mass catabolism in migratory Swainson's thrushes (*Catharus ustulatus*). Proc. Biol. Sci. 286, 20190859.

Groscolas, R., Leloup, J., 1986. The endocrine control of reproduction oand moult in male and female Empereor *Aptenodytes forsteri* and Adelie *Pygoscelis adeliae* Penguins. Gen. Comp. Endocrinol. 63, 264–274.

Guglielmo, C., 2010. Move that fatty acid: fuel selection and transport in migratory birds and bats. Integr. Comp. Biol. 50, 336–345.

Guglielmo, C., 2018. Obese super athletes: fat-fueled migration in birds and bats. J. Exp. Biol. 221, jeb165753.

Guglielmo, C., Haunerland, N.H., Williams, T.D., 1998. Fatty acid binding protein, a major protein in the flight muscle of migrating western sandpipers. Comp. Biochem. Physiol. 119, 549–555.

Guglielmo, C.G., Gerson, A.R., Price, E.R., Hays, Q.R., 2017. The effects of dietary macronutrients on flight ability, energetics, and fuel metabolism of yellow-rumped warblers *Setophaga coronata*. J. Avian Biol. 48, 133–148.

Guglielmo, C.G., Haunerland, N.H., Hochachka, P.W., Williams, T.D., 2002a. Seasonal dynamics of flight muscle fatty acid binding protein and catabolic enzymes in a long-distance migrant shorebird. Am. J. Physiol. 282, R1405–R1413.

Guglielmo, C.G., O'hara, P.D., Williams, T.D., 2002b. Extrinsic and intrinsic sources of variation in plasma lipid metabolites of free-living western sandpipers (*Calidris mauri*). Auk 119, 437–445.

Guglielmo, C.G., Piersma, T., Williams, T.D., 2001. A sport-physiological perspective on bird migration: evidence for flight-induced muscle damage. J. Exp. Biol. 204, 2683–2690.

Guglielmo, C.G., Williams, T.D., Zwingelstein, G., Brichon, G., Weber, J.-M., 2002c. Plasma and muscle phospholipids are involved in the metabolic response to long-distance migration in a shorebird. J. Comp. Physiol. 172B, 409–417.

Guillemette, M., 1994. Digestive-rate constraint in wintering Common Eider (*Somateria mollissim*a): implications for flying capabilities. Auk 111, 900–909.

Guillemette, M., Ouellet, J.-F., 2005. Temporary flightlessness in pre-laying Common Eiders *Somateria mollissima*: are females constrained by excessive wing-loading or by minimal flight muscle ratio? Ibis 147, 293–300.

Guillemette, M., Polymeropoulos, E.T., Portugal, S.J., Pelletier, D., 2017. It takes time to be cool: on the relationship between hyperthermia and body cooling in a migrating seaduck. Front. Physiol. 8, 532.

Guillemette, M., Richman, S.E., Portugal, S.J., Butler, P.J., 2012. Behavioural compensation reduces energy expenditure during migration hyperphagia in a large bird. Funct. Ecol. 26, 876–883.

Guillemette, M., Woakes, A.J., Larochelle, J., Polymeropoulos, E.T., Granbois, J.-M., Butler, P.J., Pelletier, D., Frappell, P.B., Portugal, S.J., 2016. Does hyperthermia constrain flight duration in a short-distance migrant? Phil. Trans. Biol. Sci. 371, 20150386.

Gutiérrez, J.S., Sabat, P., Castañeda, L.E., Contreras, C., Navarrete, L., Peña-Villalobos, I., Navedo, J.G., 2019. Oxidative status and metabolic profile in a long-lived bird preparing for extreme endurance migration. Sci. Rep. 9, 17616.

Gwinner, E., Zeman, M., Schwabl-Benzinger, I., Jenni-Eiermann, S., Jenni, L., Schwabl, H., 1992. Corticosterone levels of passerine birds during migratory flight. Naturwissenschaften 79, 276–278.

Hahn, T.P., Brazeal, K.R., Schultz, E.M., Chmura, H.E., Cornelius, J.M., Watts, H.E., Macdougall-Shackelton, S.A., 2015. Annual schedules. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Elsevier, Boston.

Hahn, T.P., Sockman, K.W., Breuner, C.W., Morton, M.L., 2004. Facultatiive altitudinal movements by mountain white-crowned sparrows (*Zonotrichia leucophrys oriantha*) in the Sierra Nevada. Auk 121, 1269–1281.

Hahn, T.P., Wingfield, J.C., Mullen, R., Deviche, P.J., 1995. Endocrine bases of spatial and temporal opportunism in arctic-breeding birds. Am. Zool. 35, 259–273.

Hall, Z.J., Bauchinger, U., Gerson, A.R., Price, E.R., Langlois, L.A., Boyles, M., Pierce, B., McWilliams, S.R., Sherry, D.F., MacDougal-Shackleton, S.A., 2013. Site-specific regulation of adult neurogenesis by dietary fatty acid content, vitamin E and flight exercise in European starlings. Eur. J. Neurosci. 2013, 1–8.

Hall, M.R., Gwinner, E., 1987. Annual cycles in moult, body mass, luteininzing hormone, prolactinand gonadal steroids during the development of sexual maturity in the white stork (*Ciconia ciconia*). J. Zool, Lond 211, 467–486.

Halliwell, B., Gutteridge, J.M., 1999. Free Radicals in Biology and Medicine. Clarendon Press, Oxford.

Hamiliton, A., Ly, J., Robinson, J.R., Corder, K.R., Demoranville, K.J., Schaeffer, P.J., Huss, J.M., 2018. Conserved transcriptional activity and ligand responsiveness of avian PPARs: potential role in regulating lipid metabolism in migratory birds. Gen. Comp. Endocrinol. 268, 110–120.

Hammond, K.A., Diamond, J., 1997. Maximal sustained energy budgets in humans and animals. Nature 386, 457–462.

Harman, D., 1956. Aging: a theory based on free radical and radiation chemistry. J. Gerontol. 11, 298–300.

Harrison, X.A., Blount, J.D., Inger, R., Norris, D.R., Bearhop, S., 2011. Carry-over effects as drivers of fitness differences in animals. J. Anim. Ecol. 80, 4–18.

Hasselquest, D., 2007. Comparative immunoecology in birds: hypotheses and tests. J. Ornithol. 148, S571–S582.

Hasselquest, D., Nilsson, J.A., 2012. Physiological mechanisms mediating costs of immune responses: what can we learn from studies of birds? Anim. Behav. 83, 1303–1312.

Hazelwood, R.L., 2000. Pancreas. In: Whittlow, G. (Ed.), Sturkie's Avian Physiology. Academic Press, New York.

Hegemann, A., Matson, K.D., Both, C., Tielman, B.I., 2012. Immune function in a free-living bird varies over the annual cycle, but seasonal patterns differ between years. Oecologia 170, 605–618.

Heitmeyer, M.E., Fredrickson, L.H., 1990. Fatty acid composition of wintering female mallards in relation to nutrient use. J. Wildl. Manag. 54, 54–61.

Helms, C.W., 1963. The annual cycle and Zugunruhe in birds. Proc. Int. Ornithol. Congr. 13, 925–939.

Herborn, K.A., Daunt, F., Heidinger, B.J., Granroth-Wildling, H.M.V., Burthe, S.J., Newell, M.A., Monaghan, P., 2016. Age, oxidative stress exposure and fitness in a long-lived seabird. Funct. Ecol. 30, 913–921.

Herrera, C.M., 1984. Selective Pressures on Fruit Seediness: Differential Predation of Fly Larvae on the Fruits of Berberis Hispanica. Oikos, pp. 166–170.

Hintz, J.V., 2000. The hormonal regulation of premigratory fat deposition and winter fattening in red-winged blackbirds. Mol. Integr. Physiol. 125, 239–249.

Hochachka, P.W., Somero, G.N., 2002. Biochemical Adaptation - Mechanisms and Process in Physiological Evolution. Oxford University Press, New York.

Holberton, R., Dufty, A.M., 2005. Hormones and variation in life history strategies of migratory and nonmigratory birds. In: Greenberg, R., Marra, P.P. (Eds.), Birds of Two Worlds, the Ecology and Evolution of Migration. Johns Hopkins University Press, Baltimore, London.

Holberton, R.L., Parrish, J.D., Wingfield, J.C., 1996. Modulation of the adrenocortical stress response in neotropical migrants during autumn migration. Auk 113, 558–564.

Holmes, R.T., 2007. Understanding population change in migratory songbirds: long-term and experimental studies of Neotropical migrants in breeding and wintering areas. Ibis 149, 2–13.

Hou, L., Welch, K.C., 2016. Premigratory ruby-throated hummingbirds, Archilochus colubris, exhibit multiple strategies for fuelling migration. Anim. Behav. 121, 87–99.

Hudson, D.M., Bernstein, M.H., 1981. Temperature regulation and heat balance in flying white-necked ravens, *Corvus cryptoleucus*. J. Exp. Biol. 90, 267–281.

Hulbert, A.J., Else, P.L., 1999. Membranes as possible pacemakers of metabolism. J. Theor. Biol. 199, 257–274.

Hulbert, A.J., Else, P.L., 2000. Mechanisms underlying the cost of living in animals. Annu. Rev. Physiol. 62, 207–235.

Hulbert, A.J., Pamplona, R., Buffenstein, R., Buttemer, W.A., 2007. Life and death: metabolic rate, membrane composition, and life span of animals. Physiol. Rev. 87, 1175–1213.

Hulbert, A.J., Turner, N., Storlien, L.H., Else, P.L., 2005. Dietary fats and membrane function: implications for metabolism and disease. Biol. Rev. 80, 155–169.

Infante, J.P., Kirwan, R.C., Brenna, J.T., 2001. High levels of docosahexaenoic acid (22:6n-3)-containing phospholipids in high-frequency contraction muscles of hummingbirds and rattlesnakes. Comp. Biochem. Physiol. 130B, 291–298.

Jacobs, J.D., Wingfield, J.C., 2000. Endocrine control of life-cycle stages: a constraint on response to the environment? Condor 102, 35–51.

Jehl, J.R., 1997. Cyclical changes in body composition in the annual cycle and migration of the Eared Grebe *Podiceps nigricollis*. J. Avian Biol. 28, 132–142.

Jenni-Eiermann, S., 2017. Energy metabolism during endurance flight and the post-flight recovery phase. J. Comp. Physiol. 203, 431–438.

Jenni-Eiermann, S., Jenni, L., 1991. Metabolic responses to flight and fasting in night migrating passerines. J. Comp. Physiol. 161, 465–474.

Jenni-Eiermann, S., Jenni, L., Piersma, T., 2002. Temporal uncoupling of thyroid hormones in Red Knots: T3 peaks in cold weather, T4 during moult. J. Ornithol. 143, 331–340.

Jenni-Eiermann, S., Jenni, L., Smith, S., Costantini, D., 2014. Oxidative stress in endurance flight: an unconsidered factor in bird migration. PLoS One 9, e97650.

Jenni, L., Jenni-Eiermann, S., 1992. Metabolic patterns of feeding overnight fasted and flying night migrants during autumn migration. Ornis Scand. 23, 251–259.

Jenni, L., Jenni-Eiermann, S., 1998. Fuel supply and metabolic constraints in migrating birds. J. Avian Biol. 29, 521–528.

Jenni, L., Jenni-Eiermann, S., Spina, F., Schwabl, H., 2000. Regulation of protein breakdown and adrenocortical response to stress in birds during migratory flight. Am. J. Physiol. Regul. Integr. Comp. Physiol. 278, R1182–R1189.

Jenni, L., Schaub, M., 2003. Behavioural and Physiological Reactions to Environmental Variation in Bird Migration: A Review. *Avian migration*. Springer.

Jensen, J.K., Isaksson, C., Eikenaar, C., Andersson, M.N., 2020. Migrant blackbirds, *Turdus merula*, have higher plasma levels of polyunsaturated fatty acids compared to residents, but not enhanced fatty acid unsaturation index. Ecol. Evol. 2020, 1–11.

Jimenez, A.G., O'Connor, E.S., Elliott, K.H., 2019. Muscle myonuclear domain, but not oxidative stress, decreases with age in a long-lived seabird with high activity costs. J. Exp. Biol. 222 (18), jeb211185.

Kaiser, P., Balic, A., 2015. The Avian Immune System. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Kaiya, H., Furuse, M., Miyazato, M., Kangawa, K., 2009. Current knowledge of the roles of ghrelin in regulating food intake and energy balance in birds. Gen. Comp. Endocrinol. 163, 33–38.

Karasov, W.H., Hume, I., 1997. Vertebrate gastrointestinal system. In: Dantzler, W. (Ed.), Handbook of Physiology Section 12: Comparative Physiology. Oxford University Press, New York, NY.

Karasov, W.H., McWilliams, S.R., 2005. Digestive constraints in mammalian and avian ecology. In: Starck, J.M., Wang, T. (Eds.), Physiological and Ecological Adaptations to Feeding in Vertebrates. Science Publishers Inc, Enfield, New Hampshire, pp. 87–112.

Karasov, W.H., Pinshow, B., 2000. Test for physiological limitation to nutrient assimilation in a long-distance passerine migrant at a springtime stopover site. Physiol. Biochem. Zool. 73, 335–343.

Ketterson, E.D., Nolan Jr., V., 1976. Geographic variation and its climatic correlates in the sex ratio of eastern-wintering dark-eyed Juncos (*Junco hyemalis hyemalis*). Ecology 57, 679–693.

King, J.S., Farner, D.S., 1965. Studies of fat deposition in migratory birds. Ann. N. Y. Acad. Sci. 131, 422–440.

Kirby, J.S., Stattersfield, A.J., Butchart, S.H.M., Evans, M.I., Grimmett, R.F.A., Jones, V.R., O'Sullivan, J., Tucker, G.M., Newton, I., 2008. Key conservation issues for migratory land- and waterbird species on the world's major flyways. Bird Conserv. Int. 18, S49–S73.

Klaassen, M., 1996. Metabolic constraints on long-distance migration in birds. J. Exp. Biol. 199, 57–64.

Klaassen, M., Biebach, H., 1994. Energetics of fattening and starvation in the long-distance migratory garden warbler, *Sylvia borin*, during the migratory phase. J. Comp. Physiol. B 164, 362–371.

Klaassen, M., Lindström, Å., Zijlstra, R., 1997. Composition of fuel stores and digestive limitations to fuel deposition rate in the long-distance migratory thrush nightingale, *Luscinia luscinia*. Physiol. Zool. 70, 125–133.

Klaiman, J.M., Price, E.R., Guglielmo, C.G., 2009. Fatty acid composition of pectoralis muscle membrane, intramuscular fat stores and adipose tissue of migrant and wintering white-throated sparrows (*Zonotrichia albicollis*). J. Exp. Biol. 212, 3865–3872.

Klasing, K., 2004. The costs of immunity. Acta Zool. Sin. 50, 961–969.

Klasing, K.C., 1998. Comparative Avian Nutrition. CAB International, New York.

Koutsos, E.A., Clifford, A.J., Calvert, C.C., Klasing, K.C., 2003. Maternal carotenoid status modifies the incorporation of dietary carotenoids into immune tissues of growing chickens (*Gallus gallus domesticus*). J. Nutr. 133, 1132–1138.

Krause, J., Nemeth, Z., Perez, J., Chmura, H.E., Ramenofsky, M., Wingfield, J.C., 2016. Annual hematocrit profiles in two sub-species of White-crowned Sparrows: a migrant and resident comparison. Physiol. Biochem. Zool. 89, 51–60.

Krause, J.S., Németh, Z., Pérez, J.H., Chmura, H.E., Word, K.R., Lau, H.J., Swanson, R.E., Cheah, J.C., Quach, L.N., Meddle, S.L., Wingfield, J.C., Ramenofsky, M., 2021. Annual regulation of adrenocortical function in migrant and resident subspecies of white-crowned sparrow. Horm. Behav. 127, 104884.

Kuenzel, W.J., Helms, C.W., 1967. Obesity produced in a migratory bird by hypothalamic lesions. Bioscience 17, 395–396.

Lack, D., 1954. The Natural Regulation of Animal Numbers. Oxford University Press, London.

Landys-Ciannelli, M.M., Ramenofsky, M., Piersma, T., Jukema, J., Wingfield, J.C., Group, C.R., 2002. Baseline and stress-induced plasma corticosterone during long-distance migration in the bar-tailed godwit, Limosa lapponica. Physiol. Biochem. Zool. 75, 101–110.

Landys, M.M., Ramenofsky, M., Guglielmo, C.G., Wingfield, J.C., 2004a. The low-affinity glucocorticoid receptor regulates feeding and lipid breakdown in the migratory Gambel's white-crowned sparrow *Zonotrichia leucophrys gambelii*. J. Exp. Biol. 207, 143–154.

Landys, M.M., Ramenofsky, M., Wingfield, J.C., 2006. Actions of glucocorticoids at a seasonal baseline as compared to stress-related levels in the regulation of periodic life processes. Gen. Comp. Endocrinol. 148, 132–149.

Landys, M.M., Wingfield, J.C., Ramenofsky, M., 2004b. Plasma corticosterone increases during migratory restlessness in the captive white-crowned sparrow *Zonotrichia leucophrys gambelii*. Horm. Behav. 46, 574–581.

Lee, K.A., 2006. Linking immune defenses and life history at the levels of the individual and the species. Integr. Comp. Biol. 46, 1000–1015.

Levey, D.J., Cipollini, M.L., 1998. Glycoalkaloid in ripe fruit deters consumption by cedar waxwings. Auk 115, 359–367.

Leyton, J., Drury, P.J., Crawford, M.A., 1987. Differential oxidation of saturated and unsaturated fatty acids in vivo in the rat. Br. J. Nutr. 57, 383–393.

Lindström, Å., 2003. Fuel Deposition Rates in Migrating Birds: Causes, Constraints and Consequences. In: Berthold, P., Gwinner, E., Sonnenschein, E. (Eds.), Avian Migration. Springer, Berlin, pp. 307–320.

Lindström, Å., Piersma, T., 1993. Mass changes in migrating birds: the evidence for fat and protein storage re-examined. Ibis 135, 70–78.

Livezey, B.C., Humphrey, P.S., 1986. Flightlessness in steamer-ducks (Anatidae: *Tachyeres*): its morphological bases and a probable evolution. Evolution 40, 540–558.

Lohmus, M., Sandberg, R., Holberton, R.L., Moore, F.R., 2003. Corticosterone levels in relation to migratory readiness in Red-Eyed Vireos (*Vireo olivaceus*). Behav. Ecol. Sociobiol. 54, 233–239.

Long, J.A., Holberton, R.L., 2004. Corticosterone secretion energetic condition and a test of the migration modulation hypothesis in the hermit thrush (*Catharus guttatus*) a short disttance migrant. Auk 1094–1102.

Loshchagina, J., Tsveya, A., Naidenkoc, S., 2018. Baseline and stress-induced corticosterone levels are higher during spring than autumn migration in European robins. Horm. Behav. 98, 96–101.

Lovvorn, J.R., Jones, D.R., 1994. Biomechanical conflicts between adaptations for diving and aerial flight in estuarine birds. Estuaries 17, 62–75.

Lundgren, B.O., Kiessling, K.-H., 1985. Seasonal variation in catabolic enzyme activities in breast muscle of some migratory birds. Oecologia 66, 468–471.

Lymburnera, A.L., Kellya, T.R., Hobsona, K.A., MacDougall-Shackleton, E.A., MacDougall-Shackleton, S.A., 2016. Testosterone, migration distance, and migratory timing in song sparrows. Horm. Behav. 85, 102–107.

Maillet, M., Weber, J.-M., 2006. Performance-enhancing role of dietary fatty acids in a long-distance migrant shorebird: the semipalmated sandpiper. J. Exp. Biol. 209, 2686–2695.

Maillet, M., Weber, J.-M., 2007. Relationship between n-3 PUFA content and energy metabolism in the flight muscles of a migrating shorebird: evidence for natural doping. J. Exp. Biol. 210, 413–420.

Marden, J.H., 1994. From damselflies to pterosaurs: how burst and sustainable flight performance scale with size. Am. J. Physiol. 266R, 1077–1084.

Marra, P.P., Hobson, K.A., Holmes, R.T., 1998. Linking winter and summer events in a migratory bird by using stable-carbon isotopes. Science 282, 1884–1886.

Marsh, R.L., 1981. Catabolic enzyme activities in relation to premigratory fattening and muscle hypertrophy in the gray catbird (*Dumetella carolinensis*). J. Comp. Physiol. 141, 417–423.

Marsh, R.L., 1984. Adaptations of the gray catbird, *Dumetella carolinensis* to long-distance migration: flight muscle hypertophy associated with elevated body mass. Physiol. Zool. 57, 105–117.

Marsh, R.L., Storer, R.W., 1981. Correlation of flight-muscle size and body mass in Cooper's hawks: a natural analogue of power training. J. Exp. Biol. 91, 363–368.

Marshall, T.J., Dick, M.F., Guglielmo, C.G., 2016. Seasonal dietary shifting in yellow-rumped warblers is unrelated to macronutrient targets. Comp. Biochem. Physiol. Mol. Integr. Physiol. 192, 57–63.

Martin, T.E., Karr, J.R., 1986. Patch utilization by migrating birds: resource oriented? Ornis Scand. 165–174.

McClelland, G.B., 2004. Fat to the fire: the regulation of lipid oxidation with exercise and environmental stress. Comp. Biochem. Physiol. 139B, 443–460.

McCue, M.D., Amitai, O., Khozin-Goldberg, I., McWilliams, S.R., Pinshow, B., 2009. Effect of dietary fatty acid composition on fatty acid profiles of polar and neutral lipid tissue fractions in zebra finches, *Taeniopygia guttata*. Comp. Biochem. Physiol. 154A, 165–172.

McCue, M.D., Sivan, O., McWilliams, S.R., Pinshow, B., 2010. Tracking the oxidative kinetics of carbohydrates, amino acids, and fatty acids in the house sparrow using exhaled $13CO_2$. J. Exp. Biol. 213, 782–789.

McFarlan, J.T., Bonen, A., Guglielmo, C.G., 2009. Seasonal upregulation of fatty acid transporters in flight muscles of migratory white-throated sparrows (*Zonotrichia albicollis*). J. Exp. Biol. 212, 2934–2940.

McGraw, K.J., Adkins-Regan, E., Parker, R.S., 2005. Maternally derived carotenoid pigments affect offspring survival, sex ratio, and sexual attractiveness in a colorful songbird. Naturwissenschaften 92, 375–380.

McKenzie, D.J., 2001. Effects of dietary fatty acids on the respiratory and cardiovascular physiology of fish. Comp. Biochem. Physiol. 128A, 607–621.

McKenzie, D.J., Higgs, D.A., Dosanjh, B.S., Deacon, G., Randall, D.J., 1998. Dietary fatty acid composition influences swimming performance in Atlantic salmon (*Salmo salar*) in seawater. Fish Physiol. Biochem. 19, 111–122.

McWilliams, S.R., 1999. Digestive strategies of avian herbivores. In: Adams, N., Slotow, R. (Eds.), Proceedings of the 22nd International Ornithological Congress. University of Natal, Durban, South Africa.

McWilliams, S.R., Caviedes-Vidal, E., Karasov, W.H., 1999. Digestive adjustments in cedar waxwings to high feeding rate. J. Exp. Zool. 283, 394–407.

McWilliams, S.R., Guglielmo, C., Pierce, B.J., Klaassen, M., 2004. Flying, fasting, and feeding in birds during migration: a nutritional and physiological ecology perspective. J. Avian Biol. 35, 377–393.

McWilliams, S.R., Karasov, W.H., 2001. Phenotypic flexibility in digestive system structure and function in migratory birds and its ecological significance. Comp. Biochem. Physiol. Mol. Integr. Physiol. 128, 577–591.

McWilliams, S.R., Karasov, W.H., 2005. Migration takes guts. In: Birds of Two Worlds: The Ecology and Evolution of Migration. Smithsonian Institution Press, Washington, DC, pp. 67–78.

McWilliams, S.R., Karasov, W.H., 2014. Spare capacity and phenotypic flexibility in the digestive system of a migratory bird: defining the limits of animal design. Proc. Roy. Soc. B 281, 20150308.

McWilliams, S., Pierce, B.J., 2006. Diet, body composition, and exercise performance: why birds during migration should be careful about what they eat. Physiologist 49, C1–C57 (Comparative Physiology 2006: Integrating Diversity) Abstract.

McWilliams, S.R., Pierce, B.J., Witenzellner, A., Langlois, L., Engel, S., Speakman, J., Fatica, O., DeMoranville, K.J., Goymann, W., Trost, L., Bryla, A., Dzialo, M., Sadowska, E.T., Bauchinger, U., 2020. The energy savings-oxidative cost trade-off for migratory birds during endurance flight. eLife 9:e60626.

Meddle, S.L., King, V.M., Follett, B.K., Wingfield, J.C., Ramenofsky, M., Foidart, A., Balthazart, J., 1997. Copulation activates Fos-like immunoreactivity in the male quail forebrain. Behav. Brain Res. 85, 143–159.

Meier, A.H., Farner, D.S., 1964. A possible endocrine basis for premigratory fattening in the white-crowned sparrow, *Zonotrichia leucophrys gambelii* (nuttall). Gen. Comp. Endocrinol. 102, 584–595.

Minias, P., Kaczmarek, K., Włodarczyk, R., Janiszewski, T., 2013. Hemoglobin concentrations in waders vary with their strategies of migration: a comparative analysis. Comp. Biochem. Physiol. Mol. Integr. Physiol. 165, 7–12.

Mishra, I., Singh, D., Kumar, V., 2017. Daily levels and rhythm in circulating corticosterone and insulin are altered with photostimulated seasonal states in night-migratory blackheaded buntings. Horm. Behav. 94, 114–123.

Mitchell, G.W., Guglielmo, C.G., Wheelwright, N.T., Freeman-Gallant, C.R., Norris, D.R., 2011. Early life events carry over to influence pre-migratory condition in a free-living songbird. PLoS One 6, e28838.

Mizrahy, O., Bauchinger, U., Aamidor, S., McWilliams, S.R., Pinshow, B., 2011. Availability of water affects renewal of tissues in migratory blackcaps during stopover. Integr. Comp. Biol. 51, 374–384.

Møller, A.P., Erritzøe, J., 1998. Host immune defence and migration in birds. Evol. Ecol. 12, 945–953.

Montgomery, M.K., Hulbert, A.J., Buttemer, W.A., 2012. Metabolic rate and membrane fatty acid composition in birds: a comparison between long-living parrots and short-living fowl. J. Comp. Physiol. 182B, 127–137.

Moore, M.C., Donham, R.S., Farner, D.S., 1982. Physiological preparation for autumnal migration in white-crowned sparrows. Condor 84, 410–419.

Moore, M.C., Farner, D.S., Donham, R.S., Matt, K.S., 1983. Endocrine and photoperiodic relationships during photorefractoriness postnuptial molt and onset of migration in *Zonotrichia leucophrys gambelii*. In: Proceedings XVIII International Ornithological Congress, Moscow, vol. 2, pp. 946–952.

Morton, M.L., 2002. The mountain white-crowned sparrow: migration and reproduction at high altitude. In: Rottenberry, J.T. (Ed.), Studies in Avian Biol. Cooper Ornithological Society, Lawrence, Kansas.

Morton, M.L., Liebman, H.A., 1974. Seasonal variations in fatty acids of a migratory bird with and without a controlled diet. Comp. Biochem. Physiol. Part A: Physiol. 48 (2), 329–335.

Morton, M.L., Pereyra, M.E., 1994. Autumnal migration departure schedules in mountain white-crowned sparrows. Condor 96, 1020–1029.

Mouritsen, H., 2015. Magnetoreception in birds and its use for long-distance migration. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Mouritsen, H., 2018. Long-distance navigation and magnetoreception in migratory animals. Nature 558, 50–59.

Mrosovsky, N., 1990. Rheostasis: The Physiology of Change. Oxford University Press, New York.

Muheim, R., Schmaljohann, H., Alerstam, T., 2018. Feasibility of sun and magnetic compass mechanisms in avian long-distance migration. Movement Ecol. 6, 8.

Muñoz-Garcia, A., Aamidor, S.E., McCue, M.D., McWilliams, S.R., Pinshow, B., 2012. Allocation of endogenous and dietary protein in the reconstitution of the gastrointestinal tract in migratory blackcaps at stopover sites. J. Exp. Biol. 215, 1069–1075.

Munshi-South, J., Wilkinson, G.S., 2010. Bats and birds: exceptional longevity despite high metabolic rates. Ageing Res. Rev. 9, 12–19.

Murphy, M.E., 1994. Dietary complementation by wild birds: considerations for field studies. J. Biosci. 19, 355–368.

Murphy, M.E., King, J.R., 1987. Dietary discrimination by molting white-crowned sparrows given diets differing only in sulfur amino acid concentration. Physiol. Zool. 60, 279–289.

Nagahuedi, S., Popesku, J.T., Trudeau, V.L., Weber, J.-M., 2009. Mimicking the natural doping of migrant sandpipers in sedentary quails: effects of dietary n-3 fatty acids on muscle membranes and PPAR expression. J. Exp. Biol. 212, 1106–1114.

Nakazato, M., Murakami, N., Date, Y., Kojima, M., Matsuo, H., Kangawa, K., 2001. A role for ghrelin in the central regulation of feeding. Nature 409, 194e198.

Napolitano, G.E., Ackman, R.G., 1990. Anatomical distribution of lipids and their fatty acids in the semipalmated sandpiper *Calidris pusilla* L. from Shepody Bay, New Brunswick, Canada. J. Exp. Mar. Biol. Ecol. 114, 113–124.

Narkar, V.A., Downes, M., Yu, R.T., Embler, E., Wang, Y.-X., Banayo, E., Mihaylova, M.M., Nelson, M.C., Zou, Y., Juguilon, H., Kang, H., Shaw, R.J., Evans, R.M., 2008. AMPK and PPAR-delta agonists are exercise mimetics. Cell 134, 405–415.

Nebel, S., Bauchinger, U., Buehler, D.M., Langlois, L.A., Boyles, M., Gerson, A.R., Price, E.R., McWilliams, S.R., Guglielmo, C.G., 2012. Constitutive immune function in European starlings *Sturnus vulgaris* is decreased in response to endurance flight in a wind tunnel. J. Exp. Biol. 215, 272–278.

Newton, I., 2008. The Migration Ecology of Birds. Academic Press, London.

Newton, I., Dale, L., 1996a. Relationship between migration and latitude among west European birds. J. Animal Ecol. 65, 137–146.

Newton, I., Dale, L., 1996. Bird migration at different latitudes in Eastern North America. The Auk 113 (3), 626–635.

Nishiwaki-Ohkawa, T., Yoshimura, T., 2016. Molecular basis for regulating seasonal reproduction in vertebrates. J. Endocrinol. 229, R117–R127.

Norberg, U.M., 1990. Vertebrate Flight. Springer-Verlag, Berlin, Germany.

O'Reilly, K.M., Wingfield, J.C., 1995. Spring and autumn migration in arctic shorebirds - same distance, different strategies. Am. Zool. 35, 222–233.

O'Neal, B.J., Stafford, J.D., Larkin, R.P., Michel, E.S., 2018. The effect of weather on the decision to migrate from stopover sites by autumn-migrating ducks. Movement Ecol. 6, 23.

Odum, E.P., 1960. Premigratory hyperphagia in birds. Am. J. Clin. Nutr. 8, 621–629.

Ogoh, S., Ainslie, P.N., 2009. Cerebral blood flow during exercise: mechanisms of regulation. J. Appl. Physiol. 107, 1370–1380.

O'Reilly, K.M., Wingfield, J.C., 2003. Seasonal, age, and sex differences in weight, fat reserves, and plasma corticosterone in Western Sandpipers. Condor 105, 13–26.

Owen, J.C., Moore, F.R., 2006. Seasonal differences in immunological condition of three species of thrushes. Condor 108, 389–398.

Pamplona, R., Costantini, D., 2011. Molecular and structural antioxidant defenses against oxidative stress in animals. Am. J. Physiol. Integr. Compar. Physiol. 301, R843–R863.

Pant, K., Chandola-Saklani, A., 1993. A role for thyroid hormones in the development of premigratory disposition in Redheaded Bunting, *Emberiza bruniceps*. J. Comp. Physiol. 163B, 389–394.

Parrish, J.D., 1997. Patterns of frugivory and energetic condition in Nearctic landbirds during autumn migration. Condor 99, 681–697.

Parrish, J.D., 2000. Behavioral, energetic, and conservation implications of foraging plasticity during migration. Stud. Avian Biol. 20, 53–70.

Pawar, A., Xu, J., Jerks, E., Mangelsdorf, D.J., Jump, D.B., 2002. Fatty acid regulation of liver X receptors (LXR) and peroxisome proliferator-activated receptor α (PPARα) in HEK293 cells. J. Biol. Chem. 277, 39243–39250.

Paxton, K.L., Moore, F.R., 2015. Carry-over effects of winter habitat quality on en route timing and condition of a migratory passerine during spring migration. J. Avian Biol. 46, 495–506.

Pennycuick, C., 1998. Computer simulation of fat and muscle burn in long-distance bird migration. J. Theor. Biol. 191, 47–61.

Pennycuick, C.J., 1975. Mechanics of flight. In: Farner, D.S., King, J.R. (Eds.), Avian Biology. Academic Press, London.

Pérez, J.H., Furlow, J.D., Wingfield, J.C., Ramenofsky, M., 2016. Regulation of vernal migration in Gambel's white-crowned sparrows: role of thyroxine and triiodothyronine. Horm. Behav. 84, 50–56.

Pérez, J.P., Meddle, S.L., Wingfield, J.C., Ramenofsky, M., 2018. Effects of thyroid hormone manipulation on pre-nuptial molt, luteinizing hormone

and testicular growth in male white-crowned sparrows (*Zonotrichia leuchophrys gambelii*). Gen. Comp. Endocrinol. 255, 12–18.

Perrins, C.M., 1970. The timing of birds' breeding seasons. Ibis 112, 242–255.

Petersson, A., Pickova, J., Brannas, E., 2010. Swimming performance at different temperatures and fatty acid composition of Arctic charr (*Salvelinus alpinus*) fed palm and rapeseed oils. Aquaculture 300, 176–181.

Pierce, B.J., McWilliams, S.R., 2004. Diet quality and food limitation affect the dynamics of body composition and digestive organs in a migratory songbird (*Zonotrichia albicollis*). Physiol. Biochem. Zool. 77, 471–483.

Pierce, B.J., McWilliams, S.R., 2005. Seasonal changes in composition of lipid stores in migratory birds: causes and consequences. Condor 107, 271–281.

Pierce, B., McWilliams, S.R., 2014. The fat of the matter: how dietary fatty acids can affect exercise performance. Integr. Comp. Biol. 54, 903–912.

Pierce, B.J., McWilliams, S.R., O'Connor, T., Place, A.R., Guglielmo, C., 2005. Effect of dietary fatty acid composition on depot fat and exercise performance in a migrating songbird, the red-eyed vireo. J. Exp. Biol. 208, 1277–1285.

Pierce, B.J., McWilliams, S.R., Place, A.R., Huguenin, M.A., 2004. Diet preferences for specific fatty acids and their effect on composition of fat reserves in migratory Red-eyed Vireos (*Vireo olivaceous*). Comp. Biochem. Physiol. 138A, 503–514.

Piersma, T., 1998. Phenotypic flexibility during migration: optimization of organ size contingent on the risks and rewards of fueling and flight? J. Avian Biol. 29, 511–520.

Piersma, T., Gils, J.A.V., 2011. The Flexible Phenotype: A Body-Centred Integration of Ecology, Physiology, and Behaviour. Oxford University Press, New York.

Piersma, T., Perez-Tris, J., Mouritsen, H., Bauchinger, U., Bairlein, F., 2005. Is there a "migratory syndrome" common to all migrant birds?. In: Bird Hormones and Bird Migrations: Analyzing Hormones in Droppings and Egg Yolks and Assessing Adaptations in Long-Distance Migration, vol. 1046, pp. 282–293.

Powell, F., 2015. Respiration. In: Scanes, C.G. (Ed.), Sturkie's Avian Physiology, sixth ed. Academic Press, London.

Pradhan, D.S., Ma, C., Schlinger, B.A., Soma, K.K., Ramenofsky, M., 2018. Preparing to migrate: expression of androgen signaling molecules and insulin-like growth factor-1 in skeletal muscles of Gambel's white-crowend sparrows. J. Comp. Physiol. 205, 113–123.

Pradhan, D.S., Newman, A.E.M., Wacker, D.W., Wingfield, J.C., Schlinger, B.A., Soma, K.K., 2010. Aggressive interactions rapidly increase androgen synthesis in the brain during the non-breeding season. Horm. Behav. 57, 381–389.

Pradhan, D.S., Van Ness, R., Jalabert, C., Hamden, J.E., Austin, S.H., Soma, K.K., Ramenofsky, M., Schlinger, B.A., 2019. Phenotypic flexibility of glucocorticoid signaling in skeletal muscles of a songbird preparing to migrate. Horm. Behav. 116, 104586.

Price, E.R., McFarlan, J.T., Guglielmo, C.G., 2010. Preparing for migration: the effects of photoperiod and exercise on muscle oxidative enzymes, lipid transporters, and phospholipids in White-crowned Sparrows. Physiol. Biochem. Zool. 83, 252–262.

Price, E., Staples, J.F., Milligan, C.L., Guglielmo, C.G., 2011. Carnitine palmitoyl transferase activity and whole muscle oxidation rates vary with fatty acid substrate in avian flight muscles. J. Comp. Physiol. 181B, 565–573.

Price, E.R., 2010. Dietary lipid composition and avian migratory flight performance: development of a theoretical framework for avian fat storage. Comp. Biochem. Physiol. 157A, 297–309.

Price, E.R., Guglielmo, C.G., 2009. The effect of muscle phospholipid fatty acid composition on exercise performance: a direct test in the migratory white-throated sparrow. Am. J. Physiol. 297R, R775–R782.

Price, E.R., Krokfors, A., Guglielmo, C.G., 2008. Selective mobilization of fatty acids from adipose tissue in migratory birds. J. Exp. Biol. 211, 29–34.

Raclot, T., 2003. Selective mobilization of fatty acids from adipose tissue triacylglycerols. Prog. Lipid Res. 42, 257–288.

Raclot, T., Groscolas, R., 1995. Selective mobilization of adipose tissue fatty acids during energy depletion in the rat. J. Lipid Res. 36, 2164–2173.

Raja-Aho, S., Lehikoinen, E., Suorsa, P., Nikinmaa, M.V., Vosloo, D., Eeva, T., 2013. Corticosterone secretion patterns prior to spring and autumn migration differ in free-living barn swallows (*Hirundo rustica* L.). Oecologia 173, 689–697.

Ramenofsky, M., 1990. Fat storage and fat metabolism in relation to bird migration. In: Gwinner, E. (Ed.), Physiology and Ecophysiological Aspects of Bird Migration. Springer, Berlin.

Ramenofsky, M., 2011. Hormones in migration and reproductive cycles of birds. In: Norris, D., Lopez, K.H. (Eds.), Hormones and Reproduction of Vertebrates. Academic Press.

Ramenofsky, M., Agatsuma, R., Barga, M., Cameron, R., Harm, J., Landys, M., Ramfar, T., 2003. Migratory behavior: new insights from captive studies. In: Berthold, P., Gwinner, E., Sonnenschein, E. (Eds.), Avian Migration. Springer, Berlin.

Ramenofsky, M., Cornelius, J.M., Helm, B., 2012. Physiological and behavioral responses of migrants to environmental cues. J. Ornithol. 153, S181–S191.

Ramenofsky, M., Nemeth, Z., 2014. Regulatory mechanisms for the development of the migratory phenotype: roles for photoperiod and the gonad. Horm. Behav. 66, 148–158.

Ramenofsky, M., Savard, R., Greenwood, M.R.C., 1999. Seasonal and diel transitions in physiology and behavior in the migratory dark-eyed junco. Compar. Biochem. Physiol.-Mol. Integr. Physiol. 122, 385–397.

Ramenofsky, M., Wingfield, J.C., 2006. Behavioral and Physiological Conflicts in Migrants: The Transition between Migration and Breeding, pp. 135–145.

Ramenofsky, M., Wingfield, J.C., 2007. Regulation of migration. Bioscience 57, 135–143.

Ramenofsky, M., Wingfield, J.C., 2017. Regulation of complex behavioral transitions: migration to breeding. Anim. Behav. 124, 299–306.

Rani, S., Kumar, V., 2014. Photoperiodic regulation of seasonal reproduction in higher vertebrates. IJEB 52, 413–419.

Rastogi, A., Kumari, A., Rani, S., Kumar, V., 2013. Neural Correlates of migration: activation of hypothalamic clock(s) in and out of migratory state in the Blackheaded Bunting (*Emberiza melanocephal*). PLoS One 8, e70065.

Raveling, D.G., Heitmeyer, M.E., 1989. Relationships of population size and recruitment of pintails to habitat conditions and harvest. J. Wildl. Manag. 53, 1088–1103.

Reiser, P.J., Greaser, M.L., Moss, R.L., 1996. Contractile properties and protein isoforms of single fibres from the chicken pectoralis red strip muscle. J. Physiol. (Lond.) 493, 553–562.

Remage-Healey, L., Romero, L.M., 2001. Corticosterone and insulin interact to regulate glucose and triglyceride levels during stress in a bird. Am. J. Physiol. 281, R994–R1003.

Reneerkens, J., Morrison, R.I.G., Ramenofsky, M., Piersma, T., Wingfield, J.C., 2002a. Baseline and stress-induced levels of corticosterone during different life cycle substages in a shorebird on the high arctic breeding grounds. Physiol. Biochem. Zool. 75, 200–208.

Reneerkens, J., Piersma, T., Ramenofsky, M., 2002b. An experimental test of the relationship between temporal variability of feeding opportunities and baseline levels of corticosterone in a shorebird. J. Exp. Zool. 293, 81–88.

Richardson, R.D., Boswell, T., Weatherford, S.C., Wingfield, J.C., Woods, S.C., 1992. Central and peripheral regulators of food-intake in the white-crowned sparrow. Appetite 19, 212.

Richardson, R.D., Boswell, T., Weatherford, S.C., Wingfield, J.C., Woods, S.C., 1993. Cholecystokinin octapeptide decreases food-intake in white-crowned sparrows. Am. J. Physiol. 264, R852–R856.

Rios, J.M., Barcelo, G.F., Narvaez, C., Maldonado, K., Sabat, P., 2014. Feeding and digestive responses to fatty acid intake in two South American passerines with different food habits. J. Comp. Physiol. B 184, 729–739.

Romero, L.M., Dickens, M.J., Cyr, N.E., et al., 2009. The reactive scope model - A new model integrating homeostasis, allostasis, and stress. Horm. Behav. 55.

Romero, L.M., Ramenofsky, M., Wingfield, J.C., 1997. Season and migration alters the corticosterone response to capture and handling in an Arctic migrant, the white-crowned sparrow (*Zonotrichia leucophrys gambelii*). Compar. Biochem. Physiol. C-Pharmacol. Toxicol. Endocrinol. 116, 171–177.

Romero, L.M., Wingfield, J.C., 2016. Tempests, Poxes, Predators and People: Stress in Wild Animals and How They Cope. Oxford University Press, Oxford, U.K.

Royle, N.J., Surai, P.F., Hartley, I.R., 2003. The effect of variation in dietary intake on maternal deposition of antioxidants in zebra finch eggs. Funct. Ecol. 17, 472–481.

Ruf, T., Valencak, T., Tataruch, F., Arnold, W., 2006. Running speed in mammals increases with muscle n-6 polyunsaturated fatty acid content. PLoS One 1, e65.

Runfeldt, S., Wingfield, J.C., 1985. Experimentally prolonged sexual-activity in female sparrows delays termination of reproductive activity in their untreated mates. Anim. Behav. 33, 403–410.

Ryder, J.P., 1970. A possible factor in the evolution of clutch size in Ross's Goose. Wilson Bull. 82, 5–13.

Sandberg, R., Moore, F.R., Backman, J., Lohmus, M., 2002. Orientation of nocturnally migrating Swainson's thrush at dawn and dusk: importance of energetic condition and geomagnetic cues. Auk 119, 201–209.

Savard, R., Ramenofsky, M., Greenwood, M.R.C., 1991. A north-temperate migratory bird - a model for the fate of lipids during exercise of long duration. Can. J. Physiol. Pharmacol. 69, 1443–1447.

Schaefer, H.M., Schmidt, V., Bairlein, F., 2003. Discrimination abilities for nutrients: which difference matters for choosy birds and why? Anim. Behav. 65, 531–541.

Schat, K.A., Kaspers, B., Kaiser, P. (Eds.), 2014. Avian Immunology. Elsevier, New York, NY.

Schaub, M., Jenni, L., 2001. Stopover durations of three warbler species along their autumn migration route. Oecologia 128, 217–227.

Schmaljohann, H., Bruderer, B., Liechti, F., 2008. Sustained bird flights occur at temperatures far beyond expected limits. Anim. Behav. 76, 1133–1138.

Schmaljohann, H., Liechti, F., Bruderer, B., 2009. Trans-Sahara migrants select flight altitudes to minimize energy costs rather than water loss. Behav. Ecol. Sociobiol. 63, 1609–1619.

Schmidt-Nielsen, K., 1972. Locomotion: energy cost of swimming, flying, and running. Science 177, 222–228.

Schwabl, H., Bairlein, F., Gwinner, E., 1991. Basal and stress-induced corticosterone levels of garden warblers, *Sylvia borin*, during migration. J. Comp. Physiol. B Biochem. Syst. Environ. Physiol. 161, 576–580.

Schwabl, H., Farner, D.S., 1989. Endocrine and environmental control of vernal migration in male White-crowned Sparrows, *Zonotrichia leucophrys gambelii*. Physol. Zool. 62, 1–10.

Schwabl, H., Schwabl-Benzinger, I., Goldsmith, A.R., Farner, D.S., 1988. Effects of ovariectomy on long-day-induced premigratory fat deposition, plasma levels of luteinizing hormone and prolactin, and molt in White-crowned Sparows. *Zonotrichia leucophrys gambelii*. Gen. Comp. Endocrinol. 71, 398–405.

Schwemmer, P., Voigt, C.C., Corman, A.M., Adler, S., Garthe, S., 2016. Body mass change and diet switch tracked by stable isotopes indicate time spent at a stopover site during autumn migration in dunlins *Calidris alpina alpina*. J. Avian Biol. 47, 806–814.

Schwilch, R., Grattarola, A., Spina, F., Jenni, L., 2002. Protein loss during long-distance migratory flight in passerine birds: adaptation and constraint. J. Exp. Biol. 205, 687–695.

Scott, G.R., Hawkes, L.A., Frappell, P.B., Butler, P.J., Bishop, C.M., Milsom, W.K., 2015. How bar-headed geese fly over the Himalayas. Physiology 30, 107–115.

Scott, G.R., Milsom, W.K., 2006. Flying high: a theoretical analysis of the factors limiting exercise performance in birds at altitude. Respir. Physiol. Neurobiol. 154, 284–301.

Scott, G.R., Schulte, P.M., Egginton, S., Scott, A.L.M., Richards, J.G., Milsom, W.K., 2011. Molecular evolution of the cytochrome C oxidase underlies high-altitude adaptation in the bar-headed goose. Mol. Biol. Evol. 28, 351–363.

Sedinger, J.S., Alisauskas, R.T., 2014. Cross-seasonal effect and the dynamics of waterfowl populations. Wildfowl 99, 836–840.

Sharma, A., Singh, D., Malik, S., Gupta, N.J., Rani, S., Kumar, V., 2018. Difference in control between spring and autumn migration in birds: insight from seasonal changes in hypothalamic gene expression in captive buntings. Proc. R. Soc. B 285.

Sidell, B.D., Crockett, E.L., Driedzic, W.R., 1995. Antarctic fish tissues preferentially catabolize monoenoic fatty acids. J. Exp. Zool. 271, 73–81.

Sjoberg, S., Alerstam, T., Akesson, S., Schulz, A., Weidauer, A., Coppack, T., Muheim, R., 2015. Weather and fuel reserves determine departure and flight decitions in passerines migrating across the Baltic Sea. Anim. Behav. 104, 59–68.

Skrip, M., Bauchinger, U., Goymann, W., Fusani, L., Cardinale, M., Alan, R., Mcwilliams, S.R., 2015a. Migrating songbirds on stopover prepare for, and recover from, oxidative challenges posed by long-distance flight. Ecol. Evol. 5, 3198–3209.

Skrip, M.M., Bauchinger, U., Goymann, W., Fusani, L., Mcwilliams, S.R., 2015b. Access to water affects the condition dependency of nocturnal restlessness in Garden Warblers on a Mediterranean island stopover. J. Ornithol. 156, 425–432.

Skrip, M., McWilliams, S.R., 2016. Oxidative balance in birds: an atoms-to-organisms-to-ecology primer for ornithologists. J. Field Ornithol. 87, 1–20.

Skrip, M.M., Seeram, N.P., Yuan, T., Ma, H., McWilliams, S.R., 2016. Dietary antioxidants and flight exercise in female birds affect allocation of nutrients to eggs: how carry-over effects work. J. Exp. Biol. 219, 2716–2725.

Smith, J.P., 1982. Changes in blood levels of thyroid hormones in two species of passerine birds. Condor 84, 160–167.

Smith, A.D., McWilliams, S.R., 2014. What to do when stopping over: behavioral decisions of a migrating songbird during stopover are dictated by initial change in their body condition and mediated by key environmental conditions. Behav. Ecol. 25, 1423–1435.

Smith, R.J., Moore, F.R., 2005. Arrival timing and seasonal reproductive performance in a long-distance migratory landbird. Behav. Ecol. Sociobiol. 57, 231–239.

Soma, K.K., Wissman, A.M., Brenowitz, E.A., Wingfield, J.C., 2002. Dehydroepiandrosterone (DHEA) increases territorial song and the size of an associated brain region in a male songbird. Horm. Behav. 41, 203–212.

Speakman, J.R., 2005. Body size, energy metabolism and lifespan. J. Exp. Biol. 208, 1717–1730.

Speakman, J.R., Garratt, M., 2013. Oxidative stress as a cost of reproduction: beyond the simplistic trade-off model. Bioessays 36, 93–106.

Speakman, J.R., Selman, C., 2011. The free-radical damage theory: accumulating evidence against a simple link of oxidative stress to ageing and lifespan. Bioessays 33, 255–259.

Stafford, J.D., Janke, A.K., Anteau, M.J., Pearse, A.T., Fox, A.D., Elmberg, J., Straub, J.N., Eichholz, M.W., Arzel, C., 2014. Spring migration of waterfowl in the northern hemisphere: a conservation perspective. Wildfowl 4, 70–85.

Stetson, M.H., Erickson, J.E., 1972. Hormonal control of photoperiodically induced fat deposition in White-crowned Sparrows. Gen. Comp. Endocrinol. 19, 355–362.

Stillwell, W., Wassall, S.R., 2003. Docosahexaenoic acid: membrane properties of a unique fatty acid. Chem. Phys. Lipids 126, 1–27.

Sweazea, K.L., McMurtry, J.P., Braun, E.J., 2006. Inhibition of lipolysis does not affect insulin sensitivity to glucose uptake in the mourning dove. Comp. Biochem. Physiol. B 144, 387–394.

Terrill, S.B., 1990. Ecophysiological aspects of movements by migrants in the wintering quarters. In: Gwinner, E. (Ed.), Bird Migration: Physiology and Ecophysiology. Springer-verlag, Berlin.

Thomas, V.G., George, J.C., 1975. Plasma and depot fat fatty acids in Canada Geese in relation to diet, migration, and reproduction. Physiol. Zool. 48, 157–167.

Tobalske, B.W., 2016. Avian flight. In: Lovette, I.J., Fitzpatrick, J.W. (Eds.), Handbook of Bird Biology, third ed. John Wiley & Sons, Ltd, West Sussex, U.K.

Torre-Bueno, J.R., 1976. Temperature regulation and heat dissipation during flight in birds. J. Exp. Biol. 65, 471–482.

Torre-Bueno, J.R., 1978. Evaporative cooling and water balance during flight in birds. J. Exp. Biol. 75, 231–236.

Totzke, U., Hubinger, A., Bairlein, F., 1997. A role for pancreatic hormones in the regulation of autumnal fat deposition of the garden warbler (*Sylvia borin*)? Gen. Comp. Endocrinol. 107, 166–171.

Tsipoura, N., Burger, J., 1999. Shorebird diet during spring migration stopover on Delaware Bay. Condor 101, 635–644.

Tsipoura, N., Scanes, C.G., Burger, J., 1999. Corticosterone and growth hormone levels in shorebirds during spring and fall migration stopover. J Exp BIology 284, 645–651.

Tsutsui, K., Saigoh, E., Ukena, K., Teranishi, H., Fujisawa, Y., Kikuchi, M., Ishii, S., Sharp, P.J., 2000. A novel avian hypothalamic peptide inhibiting gonadotropin release. Biochem. Biophys. Comm. 275, 661–667.

Tucker, V.A., 1970. Energetic cost of locomotion in animals. Comp. Biochem. Physiol. 34, 841–846.

Turner, N., Else, P.L., Hulbert, A.J., 2003. Docosahexaenoic acid (DHA) content of membranes determines molecular activity of the sodium pump: implications for disease states and metabolism. Naturwissenschaften 90, 521–523.

Valencak, T.G., Arnold, W., Tataruch, F., Ruf, T., 2003. High content of polyunsaturated fatty acids in muscle phospholipids of a fast runner, the European brown hare (*Lepus europaeus*). J. Comp. Physiol. 173B, 695–702.

Velten, B.P., Welch JR., K.C., Ramenofsky, M., 2016. Altered expression of pectoral myosin heavy chain isoforms corresponds to migration status in the White-crowned Sparrow (*Zonotrichia leucophrys gambelii*). Roy. Soc. Open Sci. 3, 160775.

Wagner, D.N., Green, D.J., Cooper, J.M., Love, O.P., Williams, T.D., 2014. Variation in plasma corticosterone in migratory songbirds: a test of the migration-modulation hypothesis. Physiol. Biochem. Zool. 87, 695–703.

Vezina, F., O'Connor, R.S., Le Pogam, A., De Jesus, A.D., Love, O.P., Jimenez, G., 2021. Snow buntings preparing for migration increase muscle fiber size and myonuclear domain in parallel with a major gain in fat mass. J Av Biol 52 (5).

Wagner, G.N., Balfry, S.K., Higgs, D.A., Lall, S.P., Farrell, A.P., 2004. Dietary fatty acid composition affects the repeat swimming performance of Atlantic salmon in seawater. Comp. Biochem. Physiol. 137A, 567–576.

Wang, G., Ramenofsky, M., Wingfield, J.C., 2013. Apparent dissociation of photoperiodic time measurement between vernal migration and breeding under dim green light conditions in Gambel's white-crowned sparrow *Zonotrichia leucophrys gambelii*. Curr. Zool. 59, 349–359.

Warren, J.M., Cutting, K.A., Koons, D.A., 2013. Body condition dynamics and the cost-of-delay hypothesis in a temperate-breeding duck. J. Avian Biol. 44, 1–8.

Watson, H., Salmon, P., Isaksson, C., 2018. Maternally derived yolk antioxidants buffer the developing avian embryo against oxidatives stress induced by hyperoxia. J. Exp. Biol. 221, jeb179465.

Watts, H.E., Cornelius, J.M., Fudickar, A.M., Pérez, J., Ramenofsky, M., 2018. Understanding variation in migratory movements: a mechanistic approach. Gen. Comp. Endocrinol. 256, 112–122.

Weber, J.-M., 2009. The physiology of long-distance migration: extending the limits of endurance metabolism. J. Exp. Biol. 212, 593–597.

Weise, G.M., 1967. Castration and spring migration in the white-throated sparrow. Condor 69, 49–68.

Wikelski, M., Tarlow, E.M., Raim, A., Diehl, R.H., Larkin, R.P., Visser, G.H., 2003. Costs of migration in free-flying songbirds. Nature 423, 704.

Williamson, K.A., Surai, P.F., Graves, J.A., 2006. Yolk antioxidants and mate attractiveness in the Zebra Finch. Funct. Ecol. 20, 354–359.

Wilson, F.E., Reinert, B.D., 1996. Programming in seasonally breeding male American tree sparrows (*Spizella arborea*). Gen. Comp. Endocrinol. 103, 82–92.

Wilson, F.E., Reinert, B.D., 1999. Time of thyroidectomy variably affects seasonality in female American tree sparrows (*Spizella arborea*). Gen. Comp. Endocrinol. 114, 425–430.

Wiltschko, R., Wiltschko, W., 2009. Avian navigation. Auk 126, 717–743.

Wiltschko, W., Wiltschko, R., Emlen, S.R., Demong, N.J., 1980. Nocturnal activity and orientation behavior during spring migration and early summer in the indigo bunting, *Passerina cyanea*. J. Comp. Physiol. 137, 4740.

Wingfield, J.C., Farner, D.S., 1978a. The annual cycle of plasma irLH and steroid hormones in feral populations of the white-crowned sparrow, *Zonotrichia leucophrys gambelii*. Biol. Reprod. 19, 1046–1056.

Wingfield, J.C., Farner, D.S., 1978b. Endocrinology of a natural breeding population of the White-crowned sparrow, *Zonotrichia leucophrys pugetensis*. Physiol. Zool. 51, 188–205.

Wingfield, J.C., Owen-Ashley, N.T., Benowitz-Fredericks, Z.M., Lynn, S., Hahn, T.P., Wada, H., Breuner, C.M., Meddle, S.L., Romero, L.M., 2004. Arctic Spring: the arrival biology of migrant birds. Acta Zool. Sin. 50, 948–960.

Wingfield, J.C., Perfito, N., Calisi, R., Bentley, G.E., Ubuka, T., Mukai, M., O'Brien, S., Tsutsui, K., 2016. Putting the brakes on reproduction: implications for conservation, global climate change and biomedicine. Gen. Comp. Endocrinol. 227, 16–26.

Wingfield, J.C., Schwabl, H., Mattocks Jr., P.W., 1990. Endocrine mechanisms of migration. In: Gwinner, E. (Ed.), Bird Migration. Springer-Verlag, Berlin.

Witmer, M.C., 2001. Nutritional interactions and fruit removal: cedar waxwing consumption of *Viburnum opulus* fruits in spring. Ecology 82, 3120–3130.

Wojciechowski, M.S., Pinshow, B., 2009. Heterothermy in small, migrating passerine birds during stopover: use of hypothermia at rest accelerates fuel accumulation. J. Exp. Biol. 212, 3068–3075.

Wu, B.J., Else, P.L., Storlien, L.H., Hulbert, A.J., 2001. Molecular activity of Na+/K+-ATPase from different sources is related to the packing of membrane lipids. J. Exp. Biol. 204, 4271–4280.

Wu, B.J., Hulbert, A.J., Storlien, L.H., Else, P.L., 2004. Membrane lipids and sodium pumps of cattle and crocodiles: an experimental test of the membrane pacemaker theory of metabolism. Am. J. Physiol. 287, R633–R641.

Yap, K.N., Dick, M.F., Guglielmo, C.G., Williams, T.D., 2018. Effects of experimental manipulation of hematocrit on avian flight performance in high-and low-altitude conditions. J. Exp. Biol. 221, jeb191056.

Yokoyama, K., 1976. Hypothalamic and hormonal control of photoperiodically induced vernal functions in the White-crowned sparrow, *Zonotrichia leucophrys gambelii*. I. The effects of hypothalamic lesions and exogenous hormones. Cell Tissue Res. 174, 391–416.

Zeng, W.-H., Lin, L., Lui, J.-S., Xu, Z.-J., Li, M., 2013. Geographicaal variation in basal thermogenesis in little buntings: relation to celular thermogenesis and thyroid hormone concentrations. Comp. Biochem. Physiol. A 164, 483–490.

Zhang, F., Basinski, M.B., Beals, J.M., Briggs, S.L., Churgay, L.M., Clawson, D.K., Dimarchi, R.D., Furman, T.C., Hale, J.E., Hsiung, H.M., Schoner, B.E., Smith, D.P., Zhang, X.Y., Wery, J.P., Schevitz, R.W., 1997. Crystal structure of the obese protein leptin-E100. Nature 387, 206–209.

Zhang, J., Phillips, D.I.W., Wang, C., Byrne, C.D., 2004. Human skeletal muscle PPARα expression correlates with fat metabolism gene expression but not BMI or insulin sensitivity. Am. J. Physiol. 286, E168–E175.

Zurovchak, J.G., 1997. Nutritional Role of High-Lipid Fruits in the Diet of Migrant Thrushes (Ph.D. dissertation). Rutgers University.