Eco-immunology: Role of melatonin in avian immune defense

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Summary

Avian immunology is a fascinating and growing field. Ecologists are now taking an interest in measuring immune-competence and determining its importance as a heritable trait for the survival of the wild species. Seasonally breeding animals encounter different environmental challenges throughout the year. In order to survive in specific environments these animals have developed various strategies that help them to find food, protect themselves from the tough environments, and also reproduce accordingly. Indeed, wild birds are continually challenged with different natural stressors such as shortage of food (during winter and rainy season), lack of water during summer, predator pressure in social system, sudden change in weather, season-bound diseases, infection (like avian influenza), etc. Only a few ecological studies have analyzed the stress and general immunity in wild birds that might have a link with the anti-stress hormone melatonin. Therefore, approaches improving our understanding of the stress-induced immune depression and their interrelationship in birds are most desirable. Unfortunately, our knowledge of the immune system has advanced at a much faster pace for mammals while immunology of wild birds received less attention. The purpose of this review is to bring together the current knowledge on the biological effects of melatonin on avian immune defense with special emphasis on new immerging area of neuro-immuno-endocrinology i.e., eco-immunology.

Key words: Eco-immunology, stress, immunity, birds, melatonin.

Introduction

Avian immunology is a fascinating and growing field. Ecologists are now taking interest in measuring immune-competence and determining its importance as a heritable trait for the survival of the wild species. Other than mammals, birds have provided an invaluable model for investigating basic immunological mechanism. The current threat from avian influenza reminds us of the necessity for gaining a thorough understanding of the avian immune system in order to develop novel and effective strategies for control. At the same time, it should be borne in mind that infections provide an important selection pressure and immune-competence to the organism, which is a valuable trait for survival of the species. Birds have many immunological mechanisms in common with mammals but have evolved a number of quite distinct strategies; they achieved the same goal through use of different mechanism. Some of their different physiological characteristics such as their lung ventilation system, significantly different from that of the mammals, one comprising Lung Associated Immune System (LAIS) differentiated into Bronchus Associated Lymphatic Tissue (BALT) and non-BALT other than general immunity. In wild birds, the lung-associated immune system (LAIS) plays a major role in fighting pathogens invading the lungs. Thee BALT nodules, as described by Bienenstock et al. (1973) in chickens, has also been reported for a wild tropical species *Perdicula asiatica* (Kharwar and Haldar, 2011). BALT structures consisting of aggregates of lymphocytes are located at the junctions of the primary and secondary bronchi, while the non-BALT nodules with similar cell types (T-cells and B-cells) are sparsely distributed throughout the lung of chicken (Reese et al., 2006).

Why wild birds?

Birds may be the most attractive species in the world; no other group attracts so many researchers and amateur scientists. For researchers, birds are ideal objects of study for a whole range of fundamental biological questions since birds are abundant in species diversity and rich in adaptation. They share their habitats with humans more so than any other animal group and act as important biological indicators. The consumption of bird meat (Turkey, emu, etc.) other than chicken meat is increasing annually. In view of the growing number of commercially held avian meat (Turkey, emu, chicken, Japanese quail, Indian quail) and its economic importance, these birds need to be protected against pathogens by improving their natural and specific defense mechanisms so that it is least spread in human being. Further, various common small birds like house sparrow, dove, Indian finches, myna, etc., are greatly reduced in urban areas because of the

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destruction of their natural habitats due to fast industrialization, concrete elevations, social pressure by other birds, less cultivation of food grain, excessive night time lights, etc. Therefore, approaches improving our understanding of the stress-induced immune depression are most desirable. Unfortunately, our knowledge of the immune system has advanced at a much faster pace for mammals while immunology of wild birds received less attention.

What is eco-immunology?

From this perspective, how animals respond to man-made insults and what comprise the mechanisms of defense or silence seem fundamentally ecological in character. Eco-immunologists have made many important discoveries pertaining to the immune systems of wild animals. Some such discoveries include (i) immune activity is usually costly, (ii) counter-intuitive decrements in immune activity are often due to trade-offs with other physiological activities or behaviors, and (iii) immune activity is a currency by which sexually selected traits are indices of individual quality (Martin et al., 2006). The use of single assays to characterize "immune-competence," however, as was and is, the common practice in eco-immunology. Recently, it has been suggested that eco-immunologists measure disease resistance or the fitness consequences of immunological insults instead of the immune system itself. We propose that researchers continue to use the techniques that have already been fruitful in eco-immunology, but better incorporate the underlying immune-physiology of such techniques into their study designs and interpretation.

Globally, one in eight bird species may become extinct over the next 100 years-99% of the extinctions owing to human activities such as deforestation and hunting (Bird Life International, 2000). Therefore, there is an urgent need to understand the patterns and processes of avian extinctions. In order to develop novel solutions to avian disease problems, including novel vaccines and/or vaccine adjuvants, and the identification of disease resistance genes which can contribute to conventional breeding programs, it is necessary to gain a more thorough understanding of the avian immune response and how pathogens can subvert that response. Birds occupy the same habitats as mammals, have similar ranges of longevity and body mass, and face similar pathogen challenges, but have a different repertoire of organs, cells, molecules and genes of the immune system compared to mammals (Davison et al., 2008).

Environmental factors, stress and birds

Environmental and biotic inûuences on immune function are certainly complex, but not empirically intractable. Low ambient temperatures, reduced food availability, and other energetically demanding winter conditions compromise immune function, and these factors can be manipulated and controlled in the laboratory. Because many stressors are seasonally recurrent in nature, individuals of some species may have evolved mechanisms to anticipate and counteract these recurring threats to maximize immunity. Short photoperiods have been hypothesized to serve as a cue used by animals to enhance immune function in advance of energy-compromising conditions (Nelson and Drazen, 1999; Yellon et al., 1999). This enhancement appears to be mediated by the pineal hormone, melatonin. Increased duration of melatonin treatment (mimicking long nights) enhances immune function, either directly or indirectly by affecting the secretion of steroid hormones and prolactin (Grossmann, 1985).

The Indian spotted owlet, *Athene brama* is commonly known as "farmer's friend" because of its biological importance in predating on the field rodents, mice, lizards and other small pests of agricultural crops. The frequent use of herbicides and pesticides directly or indirectly influences immunity via the food chain (Rohr et al., 2008). The nature of the food always challenges the immunity, and this could be the reason behind the decreasing number of some predatory birds. Several studies on spotted owlet have inferred that habitat quality, prey abundance and weather are having drastic influence on reproduction (Thomas et al., 1990; Franklin et al., 2000; Millon et al., 2010). However, only few studies dealing with reproduction of birds in relation to immune capacity under natural condition have been carried out (Saino et al., 1997; Sorci et al., 1997; Singh and Haldar, 2005, 2007; Fig. 1). In addition, owlets have to endure many stressors in their natural environments, for e.g., they experience food shortage, predator pressure, high parasite densities with season, and social pressure (Guchhait and Haldar, 1999a, b, 2000; Fig. 2)

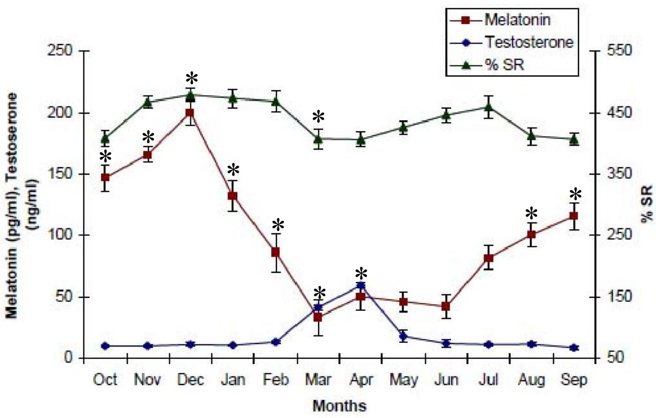

Fig. 1 Annual variation of percent stimulation ratio (%SR) of splenocytes, plasma melatonin (pg/ml) and plasma testosterone (ng/ml) in male *Perdicula asiatica.* Each data point represents mean + S.E.M; N=5. Vertical bar represents standard error. *P < 0.05. In Dunnett t-test, the data from June were treated as control and compared with other months (From Singh and Haldar, 2007).

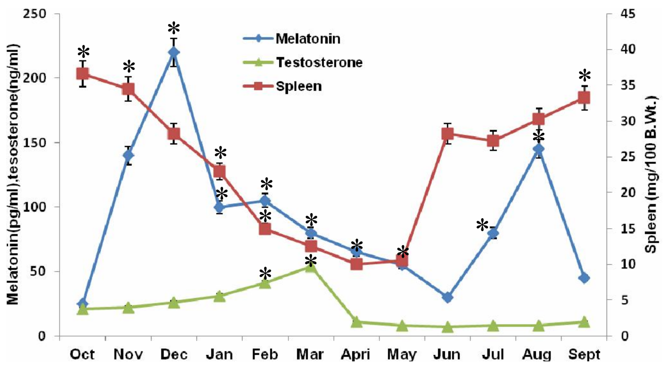

Fig. 2 Annual variation of splenic mass (mg/100 g body mass), plasma melatonin (pg/ml) and plasma testosterone (ng/ml) in male *Athene brama.* Each data point represents mean + S.E.M; N=5. Vertical bar represents standard error. *P < 0.05. In Dunnett t-test, the data from June were treated as control and compared with other months (From Guchhait and Haldar, 1999a).

Ecological implications of stress–immune interactions

A variety of factors may inûuence the effect of stress on immunity in wild animals. Group-living species experience different sorts of stressors than individual or pair-living species. Predominantly monogamous species may experience mate separation as a greater stressor than promiscuous species. In group living species, stressor effects on immune activity may be more affected by conûicts than mate loss. Experimental increase in competition compromised humoral immunity in house ûnches (*Carpdodacus mexicanus*), although these effects were not mediated directly by glucocorticoids (GCs) (Hawley et al., 2006). In zebra ûnches, pair-housing induces subordinate but not dominant individuals to reduce immune function (Gleeson, 2006). Environmental conditions may further inûuence how group-living species adjust their immune systems in response to stressors. In cooperative-breeders, alleviation of reproductive responsibilities can have positive effects (Valencia et al., 2006), but when environmental conditions are demanding, help during breeding may not offset stress-induced immune suppression (Rubenstein et al., 2008). Environmental conditions generally may have broad effects on stress–immune interactions. Incidence of *Mycoplasma gallisepticum* infections and GC responses to stressors in house ûnches vary among populations and years, but not in an obviously intelligible way (Lindström et al., 2005). GC release by birds varies along urbanization gradients (Partecke et al., 2006; Bonier et al., 2007), and songbird populations in urban areas tend to have higher disease prevalence than rural ones (Bradley et al., 2008).

More study in these contexts is needed as different species harbor different pathogens depending on habitat (Fokidis et al., 2008). Proactive coping styles tend to be coupled with efforts to prevent or manipulate stressors, high SNS activity, and a Th1 biased immune system (Koolhaas, 2008). Evidence from wild species supports the generalizations above based on domesticated ones (Roberts et al., 2007). Moreover, GC responses to stressors tend to be repeatable within individuals irrespective of the stressor imposed but potential differential effects of stress on immunity are likewise unstudied (Wada, 2008).

Immune organs and seasonality

Splenic and thymic sizes have been reported to be minimal in several avian species when the gonads begin vernal recrudescence (John, 1994). Mallard ducks (*Anas platyrhynchos*) undergo thymic involution at puberty. Similar observations have been made in house sparrow (*Passer domesticus*) and robin (*Turdus migratorious*) (Hohn, 1956). White-crowned sparrows (*Zonotrichia leucophrys gambelli* and *Zonotrichia leucophrys nuttalli*) undergo a reduction in relative splenic size at the beginning of the breeding season in western North America; splenic regression in this particular case cannot be attributed to the stress of migration because both migratory and non-migratory populations display identical patterns of splenic development (Oakeson, 1956). Migratory pied ûycatchers also display a seasonal pattern of splenic development, with regression occurring prior to the onset of breeding season in Sweden, and splenic regeneration occurring during subsequent incubation and feeding of the hatchlings (Fange and Silverin, 1985).

Seasonal disease and wild birds

In order to develop novel solution of avian disease problem it is necessary to gain a more thorough understanding of the avian neuro-immuno-endocrine system. It is becoming evident that the immune system of birds differs from that of mammals sufficiently and that untested extrapolation from mammalian systems cannot provide the quality of knowledge that is required for understating the host pathogen relationships and the factors determining the outcomes of immune response in birds. Increasing awareness of the zoonotic threat, especially currently from avian influenza, provides new motivation to develop effective controls. Bearing these points in mind, the ultimate aim of our research is, therefore, to generate essential knowledge required in these areas, leading to continued development of novel solutions (vaccines and breeding for disease resistance) to the economic burden and zoonotic threat (e.g., *Alternaria* species) that accompany infectious diseases (Kaiser, 2010; Kharwar and Haldar, 2012). We need to understand these special features of the avian immune system to develop effective control which shares some common similarities with the immune system of other species. This is because birds have complex immune system with different repertoire of organs, cells, molecules and genes of the immune system compared to mammals (Kaiser, 2010; Yadav et al., 2011). To understand host-pathogen interactions in birds it is necessary to have in-depth understanding of those aspects that are unique to birds, as well as those that are shared with other species.

The pineal gland as an ecological sensor for different types of stresses

The only neuroendocrine organ of the brain that responds to ecological variations is the pineal gland. Environmental stresses affect pineal function, impacting overall body alertness, temperature levels, and hormone operations. Stresses that affect pineal function include unusual light and dark rhythms, solar radiation, magnetic fields, nutritional imbalances, temperature swings, high altitude, and overall daily stress patterns. Although each type of stress (physical or psychological) may result in different changes in the central nervous system, the peripheral response is stereotypical as it is mediated by the activation of the HPA axis (Couto-Moraes et al., 2009). We showed that there is an elevation of melatonin in birds under experimental stress, as they experience in nature (lack of food and shelter during monsoon, extreme temperature variation, etc.) (Yadav and Haldar, unpublished). These data suggest that there is a conflict between differential regulations of environmental stresses in terms of the effects on the pineal gland. However, it is important to point out that the stress imposed in this study might have increased the levels of immunity along with melatonin production. Hence, stress may modulate pineal function.

Conclusion

Both the neuroendocrine and immune systems act to protect the internal homeostasis of the organism and it is not surprising, therefore, that they should be so closely intertwined. Infections are perceived by the neuroendocrine system as stressors, just like other stressors such as shortage of food (during winter and rainy season), lack of water during summer, predator pressure in social system, sudden change in weather, season-bound disease, infection (avian influenza) etc. The function of the neuroendocrine hormone, melatonin, in relation to stress, infection, etc., is to protect the homeostasis of the body. In the case of infection this may involve working for or against the immune system. Activation of the immune system poses potential dangers not just to the invading microorganism (e.g., *Alternaria* species) but also to the integrity of the host, for an overly vigorous response) and may kill the host in the process of controlling an infection. The neuroendocrine system must, therefore, constantly monitor and, if necessary, regulate the activities of the immune system to ensure the integrity of the host. Conversely, the immune system depends on the neuroendocrine system in determining the context of a perceived threat and how best to respond. A breakdown in this communication may be responsible for problems such as ecological misbalance in wild bird populations by low immune status in extreme climatic conditions.

Acknowledgments

The authors thank Prof. Krystyna Skwarlo Sonta (Poland), Prof. R. Nelson (USA) and Late Prof. E. Gwinner (Germany) for motivation to write this review. Financial support from CSIR, ICMR (SRF to SKY) and DST, New Delhi, is heartily acknowledged.

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