The neural mechanism of auditory perception is very complex and is influenced by several other mechanisms in vertebrates. In recent years, neuroethological studies have shown evidence for hormone-dependent modulation of the sense of hearing. In this review, I use plainfin midshipman as a neuroethological model system to study the neuro and endocrine mechanism of vocal acoustic social behavior. By putting together work from different researchers, the paper shows how seasonal changes cause fluctuation in hormone levels among female midshipman. This fluctuation in hormonal levels affects their vocal behavior and subsequently mating and reproductive circuits.

Introduction

Hormones have a profound effect on the vertebrate central nervous system that can ultimately influence the expression of behavior in adults. Hormonal activation produces a suite of morphological and physiological changes in the central nervous system. These changes shape the neuroethological mechanisms that are necessary for the reproduction and survival of vertebrates. One such mechanism is the neuro mechanism underlying the sense of
hearing. There are many vocal-acoustic behaviors expressed during courtship and reproduction that are hormone sensitive. The auditory system of vertebrates is known to be affected by both internal and external factors. One of the internal factors that influence the auditory system is the endocrine system. The endocrine system determines the type of cues that are sent from one vertebrate to another. It further determines how these cues are interpreted by the receiver. In this review, I put together several studies that have been conducted to understand the neuroendocrine mechanism underlying adaptive plasticity in audition and vocal communication in fish. Specifically, I focus on behavioral and neurohormonal characteristics among teleost (a clade of ray-finned fish that are the most species-rich group of vertebrates).

Fish, like tetrapods, have easy accessibility to their peripheral and central auditory systems. This is what makes them easy to use as experimental models. This review looks at various subtopics relating to how acoustic communication is affected by auditory hormones. Firstly, the review presents an overview of the peripheral and central auditory systems on which the whole paper is based. Secondly, I give a broader view of how this hormone-dependent mechanism modulates the auditory sense. The paper then goes on the give a deeper look into experimental results obtained from behavioral observations that show how seasonal variation in female reproductive states influences neurophysiological response properties of the auditory system. This seasonal shift in behavior amongst females is thought to be due to dramatic changes in the circulation of steroid hormones levels in males that bring about a shift in their vocal courtship and territorial aggression from season to season.
Peripheral and the central nervous system

To understand the neuroendocrine mechanism underlying the hormone-dependent plasticity of the auditory system, we first look at the peripheral and central nervous systems of fish. The inner ear of teleosts includes non-otolithic end organs and otolithic end organs. Otolithic organs are organs in the inner ear that are used to detect gravitational force and linear acceleration of the ear. The otolithic organs include the saccule and utricle. Some teleost species also have a non-otolithic sensory epithelium of unknown function called the macula neglecta, which has an auditory function in sharks. According to Corwin JT et al (1977), macula neglecta is composed of two patches of sensory epithelium. In an adult shark, these have sensory hair cells that detect forces acting oppositely and laterally to them. Unlike tetrapods that have a non-otolithic end dedicated to hearing, many species of teleosts have adapted the saccule to serve mainly an auditory function, although the utricle and lagena can also be sensitive to sound. The saccule has been the focus of most recent studies of cellular and molecular mechanisms and, hence, is the focus here as well. The auditory saccule’s sensory epithelium extends along the long axis of the otolith and is innervated by the saccular branch of the VIIIth nerve. In species like the plainfin midshipman fish, the epithelium and nerve are easily accessible because they lie lateral to the brain and just below the surface of the skull. Saccular afferents, originating from VIIIth nerve ganglion cells, and afferents, originating from a rostral hindbrain nucleus known as the octavolateralis efferent nucleus, synapse directly on hair. The efferent nucleus innervates all divisions of the inner ear and the lateral line organs.
The general organization of central auditory pathways is similar among teleosts and tetrapods. The most information is available for brainstem and thalamic targets of the ascending auditory system; telencephalic auditory centers in fishes are the least studied and remain important targets for future investigation.

**Connectivity of Dopaminergic neurons of the inner ear and the auditory system and reproduction**

Dopamine is a modulator of the vertebrate neural network. In their studies, Jonathan T. Perelmutter et al (2017) observes that dopaminergic cells are a source of Innervation in hindbrain auditory nucleus and saccule in the Inner ear. Hormone regulation of dopamine functions in part to modulate behavioral response to conspecific visualization. In a study,
Forlano PM et al (2015) observes that dopaminergic neurons in the diencephalon have widespread to the auditory system including saccule innervation. From the study, reproductive summer females showed differential dopamine innervation of the auditory system as compared to their non-reproductive winter counterparts.

This difference in levels of dopamine innervation in different females does indeed show that dopamine may function to seasonally modulate the sensitivity of the inner ear. This does in turn influence the behavioral response to reproductive acoustic signals.

**Hormone-dependent modulation of the auditory sense**

Gonadal hormone levels are known to fluctuate seasonally in fish, a thing that affects their reproductive behavior and biology. For example, in midshipman, there are four seasonal variations in female fish’s steroid hormone levels. The four seasons include non-reproductive, pre-nesting, nesting and post-nesting. The four reproductive seasons occur during different times of the years and their occurrence is dictated by the levels of hormones that female possess at these times.
Fig 1. Summary diagram of steroid hormone level in female fish during different seasons of the year Adapted from Sisneros et al. (2004a).

During the non-reproductive period, females have low plasma levels of testosterone (T) and 17β-estradiol (E₂) and a corresponding low gonadal somatic index (GSI) with ovaries containing only small undeveloped oocytes. In the pre-nesting period, females exhibit a brief annual peak of T and E₂ levels. In the nesting period, females have low levels of T and E₂ but a high GSI. There are low levels of GSI and plasma levels of E₂ and T during the post-nesting period.

Saccular afferents of vertebrates are known to respond to acoustic stimuli. The frequency of midshipman saccular afferents has been described using measures that include post-stimuli time histograms and intensity response curves based on average evoked spike rate and
synchronization. When auditory nerve neurons fire action potentials, they tend to respond to a peak in the sound pressure waveform. This means that neurons fire near the peak of each cycle of a pure tone. Since no individual neuron can respond to every cycle of the sound cycle, stimuli make the neurons respond based on average evoked spike rates. This fixation of sound frequency is called phase-locking.

A study by Joseph A. Sisneros et al (2004) was done by removing ovaries of non-reproductive females implanted with T or E₂ capsules. This ovary removal led to an increase in phase locking (frequency fixation) accuracy of the saccular afferents within the midshipman’s hearing range at higher frequencies. That is, there was an increase in the frequency that the inner saccules could respond to. This is in contrast to what was thought that T and E₂ can induce increases in the phase-locking accuracy and best frequency of saccular afferents in non-reproductive female midshipman fish

These hormone-induced changes in auditory frequency sensitivity were evident at the higher frequencies that corresponded to the second (~ 200 Hz) and third (~ 300 Hz) harmonics of the hum. Treating non-reproductive midshipman females with T or E₂ increased production of the dominant frequency components of male hum. This male hum mimicked the reproductive female’s auditory phenotype that had higher sensitivity. Midshipman-specific estrogen receptor alpha (ERα) receptor was identified in the saccular epithelium of the inner ear by RT-PCR and the use of midshipman-specific primers from an ERα clone.
This hormone-dependent plasticity in the female fish represents a mechanism that acts to enhance the coupling of sender and receiver in the auditory system. This may increase the chance of detection, recognition and localization of mates during the breeding season.

**Seasonal enhancement of vocal ability**

Studies of toadfishes have correlated elevated circulating steroid levels with seasonal offset of vocal behavior. In particular, more recent studies have correlated elevate steroids with calling behavior. For example, Genova et al. (2013) showed that either plasma or testis levels of the androgens 11-KT and testosterone, respectively, are elevated during advertisement calling (“humming”) in a captive population of midshipman fish. The study shows that non-calling fish that are put in a population of calling fish, instantly start advertising as well while exhibiting significantly higher plasma 11-KT levels with a strong trend for higher cortisol levels.

This seasonal enhancement of vocal ability is also seen in changes in vocal muscles’ ability. There is a significant change in vocal muscle mass consistent with seasonal shifts in circulating androgen levels. To investigate the molecular mechanism underlying the mediation of effects of steroids on vocal muscle function, Genova et al. (2013) compared the abundance of mRNA transcripts coding for different receptors and enzymes involved in steroid signaling pathways between advertisement calling (“humming”) and non-calling male midshipman. The researchers observed that humming males showed higher 11-KT levels whereas non-humming males showed higher cortisol levels. Humming males also had higher testis levels of 11-KT as well as testosterone. The results suggested that higher expression of
androgen implies a specific role in supporting the effects of steroids on vocal muscle physiology during advertisement calling, while higher expression of other steroid-related transcripts in the vocal muscle of non-calling males suggests a preparatory role for the physiological demands of long bouts of advertisement calls.

Studies show that implants of testosterone in juvenile male midshipman can induce increased vocal motor neuron size and increased excitability of the vocal motor system in midshipman fish. This is something that was not looked at by the article. Furthermore, in the study, the authors only use PCR to measure the abundance of the mRNA transcripts and the two enzymes (11β-hydroxysteroid dehydrogenase and 11β-hydroxy (OH)-testosterone) involved in steroid metabolism. They do not, however, set up a control experiment for this. This makes it hard to check the results obtained.

Mediation of seasonal auditory plasticity by Diencephalic Dopaminergic neurons

There is little knowledge as to what is the mechanism that underlies the coordination of behavioral responses to acoustic signals in female fish. This is despite the fact that there is a well-established knowledge of a similar mechanism in males. To understand this, Forlano et al. (2014) conducted a study that looked at neuroendocrine control of seasonal plasticity in the auditory and vocal systems in fish. In the study, the researcher uses the fact that since catecholamines, which include dopamine and noradrenaline, are known to regulate motivation, attention and arousal to hypothesize that they are modulators of acoustic-driven social behavior.
In the study, Forlano et al. (2014) demonstrate direct and robust catecholaminergic (CA) innervation of the auditory system by neurobiotin labeling of the saccule combined with hydroxylase immunoreactivity (TH-ir) as a marker of CA synthesis. They observe that these TH-ir terminals containing saccules are homologous to dopaminergic (DA) neurons in tetrapods. This result does suggest that the DA neurons are in a key position to modulate sensory-motor integration in the context of social auditory cues.

Another study, Petersen et al. (2013) uses double immunofluorescence for TH and c-Fos as a proxy for neural activity to demonstrate that TH-ir TPp neurons are activated in males exposed to advertisement calls of other males.

All these studies do show that indeed, Dopaminergic neurons seasonally modulate the sensitivity of the saccule and facilitate behavioral response to reproductive acoustic signals in midshipman.

**Summary and Concluding Remarks**

As shown in this paper, studies have shown that the plainfin midshipman is a good model system to study the vocal-acoustic behavior, neurophysiology and the endocrine system of fish. The studies have shown the impact of hormones on the auditory system of fish. In particular, there is evidence for adaptive plasticity that underlies seasonal improvements that help in detection of vocal cues during breeding season. In this way, the hormone-dependent plasticity of auditory systems in fish affects their reproductive system.
While this paper only looks at the effects of hormones on vocal acoustic cues in fish, seasonal and hormone medicated plasticity of vocal motor and auditory systems exist in several other vertebrate taxa (Remage-Healey et al (2010)). There is also evidence of behavioral relevance for neuropeptides like gonadotropin-release hormones for modulating acoustic cues (Maruska et al (2011)). This widespread of hormone-depend plasticity across different taxa may suggest that there is an evolutionary element to this plasticity.

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