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Review Article

Prolactin, genetics, and bioinformatics: The trinity for improving duck egg production in Nigeria – A review

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ABSTRACT: Recently, there has been an increase in duck production in Nigeria, meaning that the products obtainable from this animal will experience a concomitant demand. This review provides a holistic review of locally adapted Mallard and Muscovy ducks in Nigeria and their viability as a complement and or replacement for chicken eggs. A brief taxonomic and evolutionary history of ducks and a plausible explanation of how they arrived in Nigeria is explained in this review. The defining phenotypic features and performance under different production systems were discussed. Prolactin (*PRL*) and prolactin receptor (*PRLR*) were the genes of interest in this review because of their association with egg production traits in ducks. Single nucleotide polymorphisms (SNPs) and insertions and deletions (INDELs) associated with egg production traits in ducks were described. Furthermore, the review provides compelling reasons why genetic or biological data and bioinformatic tools are better for unraveling the complexities associated with functional genes and traits. Lastly, this review encourages researchers in Nigeria and Sub-Saharan Africa to use nucleotide and protein sequences when studying genetic variation for possible genetic improvement. Improving the egg production of locally adapted Nigerian ducks will require the use of genetic information for developing breeding strategies specific to the tropics and consumer demands.

Keywords: Genetics, bioinformatics, duck, egg, prolactin, Nigeria.

INTRODUCTION

Duck meat and eggs are recognized for their high nutritional value, particularly in less developed and developing countries (Sabry *et al*., 2020). Additionally, raising ducks can provide a viable source of income for small households. The two main duck breeds in Nigeria are the Mallard (*Anas platyrhynchos*) and the Muscovy (*Cairina moschata*), with the Mallard breed predominantly found in the Northern hemisphere of Nigeria (Oguntunji *et al.,* 2020). The meat of *C. moschata* is known for its unique taste and lower caloric content (Veeramani *et al*., 2016), but it is consumed less compared to other popular duck breeds, possibly due to consumer preferences and unfounded stigmas. In Nigeria, the current Mallard and Muscovy duck populations are considered to be descendants of previous generations that have adapted to environmental changes over the years, making them locally adapted (Oguntunji *et al*., 2020).

In duck production, two economically important traits are egg and meat production. Concomitantly, there has been an upward increase in the demand for duck meat and egg (Indriati *et al*., 2016). It is no news that the genes associated with duck egg production are prolactin receptor (*PRLR*) and prolactin (*PRL*) (Wang *et al*., 2011). Prolactin, a polypeptide hormone present abundantly in mammals, is produced by the anterior region of the pituitary gland, not the placenta or ovary, as it is often erroneously assumed

(Jiang *et al*., 2009; Indriati *et al*., 2016). The duck prolactin gene (*dPRL*) is 6.33 kilobases (kb) in size, with a total collection of four exons and five introns. The coding regions of the *dPRL* encode 229 amino acids (Indriati *et al*., 2016). The expression of *dPRL* is regulated by the 5'-flanking region with the aid of transcription factors (TFs) (Purwantini *et al*., 2020). Quantitative traits such as egg weight, egg production, and egg shell strength in ducks are coded by exons 5, 4, and 2, respectively, of the *dPRL* gene (Wang *et al*., 2011).

PRL nucleotide sequences and the subsequent protein sequences are an inestimable source of biological information regarding the genetic complexities and biological processes of the gene. This information is also useful in detecting polymorphisms within the *PRL* gene that are associated with economic traits, as well as how the resulting protein interacts with other proteins. Undoubtedly, bioinformatics is the appropriate computational tool for *in silico* analysis for acquiring relevant biological information on the *PRL* gene, which will prove useful in candidate gene prioritisation. Nucleotide sequence noise can be removed using bioinformatic methods, producing data-driven hypotheses and conclusions. To speed up bioinformatics research in various niches and shorten the time it takes to analyse genetic data, numerous biological databases have been developed. Principally, this review postulates that bioinformatic methods are useful for clarifying the genetic intricacies related to the *dPRL* gene and the obtained results can be interpreted within the biological context of quantitative genetics.

Studies on the phenotypic variations in Muscovy and Mallard ducks in Nigeria are numerous (Yakubu, 2013; Oguntunji and Ayorinde, 2015; Kadurumba *et al*., 2019; Kadurumba *et al*., 2022). On the use of bioinformatic and computational methods and methodologies in analysing of PRL gene of locally adapted Muscovy, however, there is very little, if any, scholarly literature. The measurement error is one of the drawbacks of heavily relying on morphometric data to estimate genetic diversity. Specifically, the approximation mistakes, parallax errors, and standard errors brought on by inaccurately calibrated measurement devices and human observers. The physical measurement of various body components in animals is laced with human and measurement-related mistakes.

Genetic variation detection through the use of computational methods minimizes the reliance on phenotypic evaluation in estimating genetic merit. A lot of focus has been placed on analysing biological data, particularly that kept on publicly accessible databases, such as DNA, messenger RNA (mRNA), and protein sequences. Unlike morphological data, this type of data is not tainted by measurement errors. Finally, this review advocates for the use of biological data, such as SNPs, diversity indices, evolutionary dynamics, and structural features, in explaining the genetic architectures of the *dPRL* gene. By incorporating the aforementioned biological data, Nigerian duck breeding programs will have access to crucial genetic data for improving egg and meat production. Consequently, this study provides a holistic review of the evolutionary and taxonomic history, morphological features, sexual dimorphism, and polymorphism of ducks, with the sole aim of demonstrating how bioinformatics – alongside the gene of interest and the foundational principles of genetics – can be used to achieve genetic progress for duck egg production.

AN OVERVIEW OF DUCK TAXONOMY

Ducks alongside other waterfowl species, such as geese and swans, all belong to the Anatidae family. However, belonging to the same taxonomic family does not imply that the phylogenetic or evolutionary relationship among these waterfowls is monophyletic. Such a conclusion will be erroneous because it is well-known that species belonging to the same taxonomic families often differ regarding the sub-families they fall under – and the waterfowl species in the Anatidae are no different. Additionally, the morphological differences between ducks and geese or swans suggest that, on a phylogenetic tree, irrespective of the algorithm used, geese, ducks, and swans will not be clustered into a single (monophyletic) clade. In other words, they possess different ancestral origins. Below is a succinct explanation of the duck scientific classification.

Kingdom: Animalia

Organisms in Kingdom Animalia are characteristically not unicellular, rather they are conglomerates of cells, making them multicellular and more evolved. Additionally, organisms within this Kingdom are heterotrophic; that is, they are not self-sustaining and derive nutrients by consuming other organisms. Members of Kingdom Animalia proliferate through sexual reproduction; they are aerobic organisms, requiring oxygen for respiration; and their skeletal structure makes locomotion possible. Members of this Kingdom are also called Metazoans or Animals.

Phylum: Chordata

The defining feature of animals belonging to this phylum is that, at a certain developmental stage, they possess a notochord. The notochord is, in a non-technical term, a straight long rod consisting of cartilage and is situated between the gastrointestinal tract and nerve cord. The notochord is a supportive structure for the nerve cord. Vertebrate animals have a more advanced form of the

notochord, the vertebral column, a more stable structure that replaces the notochord.

Subphylum: Vertebrata

Under Chordata, there are three subphyla, namely Vertebrata, Tunicata, and Cephalochordata. Ducks or birds belong to the subphylum Vertebrata.

Superclass: Gnathostomata

The distinguishing feature of this superclass is the presence of upper and lower well-developed jaws that encompass the mouth opening.

Class: Aves

Birds belong to this class, seeing as they are vertebral column-possessing animals shielded with beta-keratin hairlike extensions. Aves possessed modified skeletons that allow them to be light for flight and, concomitantly, the skeleton is strong enough to support their entire structure. The presence of a four-compartment heart is another defining feature of this class. The jaws of Aves form a beak without teeth, which is used for food sourcing and protection. Animals in this class are oviparous.

Subclass: Neornithes

Ducks are true birds, belonging to the Neornithes subclass. Distinctively, birds in this subclass possess wings, replacing forelimbs; absence of teeth; short tail; properly formed sternum; and horny rhamphotheca, replacing teeth. The evolution of Neornithes occurred around 66 million years ago (mya) during the mid to distal periods of the Cretaceous era. Additionally, Neornithes experienced substantial diversification around the Cretaceous-Paleogene (K-Pg) extinction period. This subclass is divided into Neognathae and Palaeognathae.

Infraclass: Neognathae

Almost all modern birds are Neognathae, with the rest belonging to Palaseognathae. Neognaths can be divided into two orders: waterfowl or Anseriformes and gamefowl or Galliformes. Structural and physiological similarities, with emphasis on the genetic underlying factors, submit that these two orders have a close evolutionary relationship. Phylogenetically, these two orders form a clade otherwise called Galloanserae.

Order: Anseriformes

Anatidae, Anhimidae, and Anseranatidae are the families belonging to this order.

- Anatidae: Waterfowl, such as ducks, swans, and geese, belong to this family. Birds in this family are almost ubiquitous. Anatidae has approximately 43 genera and 146 species. Birds in this family have anatomical features that enable them to swim and float.
- Anseranatidae: The population of this family is quite low; there are only two genera, namely *Eoanseranas* and *Anseranas*, with each genus containing one species.
- Anhimidae: This family is related to Anatidae; however, their bills resemble those of gamefowl (order Galliformes). The genera *Anhima* and *Chauna* are found in this order. Birds in this family are characteristically shouters.

Birds in the Anatidae and Anseranatidae families possess structural features, such as sturdy wings, delicate bills, web-like feet, slender necks and short legs, and distinctive plumage colours in drakes. Muscovy ducks have an extra feature on their head known as the caruncle.

DUCK PRODUCTION IN NIGERIA

Over the years, awareness of the need for the consumption of animal proteins in underdeveloped and developing countries has increased (Kadurumba *et al*., 2022). However, it seems that only the why of the problem has been addressed, leaving out the mechanics and systematics of how the problem will be resolved. Fortunately, Nigeria, a country whose animal protein consumption is vastly lower than some other developing countries, has animal resources that can be used to address its low protein consumption.

There are numerous domestic animals, such as poultry, cattle, goats, sheep, and pigs, that can be reared in Nigeria to enable the production of high-quality animal proteins. Even though there is a litany of macro- and micro-livestock that can be reared in Nigeria, not all of these animals are dual-purpose, socially accepted, and have short generational intervals. Fortunately, ducks are one of the few livestock species in Nigeria that meet the prerequisites above.

As described earlier, duck breeds in Nigeria can be termed locally adapted, indicating that they can, presumably, survive in the tropical climes of the sub-Sahara (Precious, 2020). Ducks are known to possess the genetic potential to be highly-producing poultry species for meat and egg because of their short generational interval, mature body weight, and fecundity (Oguntunji *et al*., 2020;

Kadurumba *et al*., 2022). Additionally, ducks can thrive on agricultural by-products (Arias-Sosa and Rojas, 2021), unlike other poultry species, making them easier to rear for low-income households. Despite the obvious benefits of duck rearing, it is a livestock enterprise that receives less attention, compared to its counterparts, chickens and turkeys.

DUCK RESEARCH IN NIGERIA

In Nigeria, ducks are mostly reared by household farmers for subsistence purposes and small-scale farmers (Oguntunji and Ayorinde, 2014; Kadurumba *et al*., 2019; Arias-Sosa and Rojas, 2021). Aside from duck rearing, academic research on locally adapted ducks in Nigeria is limited. Fairly speaking, notable research includes the genetic and phenotypic characterisation of ducks. These research endeavours have been useful in detecting genetic variations in different duck breeds using morphometric and biochemical parameters. Information obtained from these studies has been useful in genotyping interventions for several duck breeds in different agroecological zones in Nigeria.

Similarly, the use of morphological traits has been vital in detecting phenotypic variations due to selection, both natural and artificial. Characterising ducks based on their morphological features helps in sampling for molecular analysis (Kadurumba *et al*., 2022). Furthermore, morphological traits have high classificatory value (Yakubu, 2013). Two of the most populous duck breeds in Nigeria are Mallard and Muscovy (Oguntunji and Ayorinde, 2015; Yakubu, 2013).

DUCK EVOLUTION AND DIVERSITY: A RETROSPECTIVE AND PROSPECTIVE OVERVIEW

Taxonomically, ducks and geese are relatives because they have the same family (*Anatidae*) and order (*Anseriformes*); however, ducks are distinct from their *Anatidae* ilk due to phenotypic features, such as wider bills, dorsal-ventrally suppressed bodies, lower body sizes, and shorter legs and necks (Grow, 1985). Reportedly, humans domesticated (mallard) ducks circa 1000-1500 Before Christ (BC) (Su, 2022). The domestication history of Muscovy ducks, on the other hand, goes far back to the sixteenth century in South and Central America (Huang *et al*., 2012).

Birds, as we know them today, evolved from a group of theropod *Paraves* dinosaurs during the Jurassic era (Wilford, 2016). Huxley Thomas Henry posited that dinosaurs live their evolutionary life vicariously through birds (Liu and Churchil, 2022). Similarly, the Cretaceous-Paleogene (K-Pg) demarcation signalled the close of the dinosaur epoch, marking the creation, macroevolution and genetic diversification of the Aves class (Feduccia, 1995). Anthropological data and phylogenetic studies both affirm that the extinct bird *Vegavis iaai* existed in Antarctica during the latter parts of the Cretaceous time, about 66-68 mya. Scientific evidence strongly suggests that *V. iaai* was a relative of the Anseriformes order and Anatidae family, the groups from which modern-day ducks emerged (Liu and Churchil, 2022). Furthermore, the divergence patterns within the Aves class indicated that chickens and duck evolutionary ancestors coexisted with dinosaurs of non-avian origins (Clake *et al*., 2005). Even though ducks are flightless birds, their ancestors did, in fact, fly.

The litany of phylogenetic evidence suggests that today's domesticated duck breeds are the filial offspring of the common wild Mallard duck (Su, 2022). The phenotypic and genotypic diversity observed across the breed of ducks is the consequence of selection, mutations, and domestication. This observed diversity does not disprove the Mallard duck as the true parental origin of today's domestic duck breed, seeing as crosses between Mallard ducks and wild ducks yield sex-limited feather plumage in drakes (male ducks) – a feature present only in tamed true male ducks. Therefore, it is unequivocally true that modern-day common duck breeds descended exclusively from the common wild Mallard duck (Grow, 1985), while the domesticated Muscovy ducks are the evolved descendants of wild Muscovies (Liu and Churchil, 2022).

Among the popular poultry species – for example, Japanese quail, turkey, chicken, and guinea fowl – the species with the most polymorphic and diverse 12S ribosomal RNA (rRNA) gene is duck (Gupta *et al*., 2005). According to the Food and Agricultural Organisation's (FAO) Domestic Animal Diversity Information System (DAD-IS) (DAD-IS, 2020), there are fewer genetically diverse groups of domesticated Muscovy ducks (63), compared to those of Mallard ducks (400). However, there has been concern regarding the conservation status of some of these groups, as a few of the groups are one of the following: critical, endangered, or extinct (Liu and Churchil, 2022).

The use of modern computational tools and technologies has extended the frontier of selection and breeding in duck production. The development of pangenome and whole genome sequencing has reduced the time and labour needed for candidate gene prioritisation in quantitative trait loci (QTL). Additionally, the employment of computational algorithms in the determination of nucleotide variations reduces the reliance on phenotypic appraisal in genetic merit estimation. Consequently, genetic diversity indices and single nucleotide polymorphisms (SNPs) can be estimated faster and easier, making it less tedious to achieve genetic progress in a poorly performing duck population.

Although significant strides have been made in molecular and quantitative genetics, the applications of novel technologies in the local duck population will be futile

if sustainable solutions are not implemented to conserve the genetic resources of ducks, preventing genetic erosion and extinction.

Mallard ducks

Mallards lost their flight ability after several years of domestication and *ad libitum* feeding, which caused a commensurate increase in body weight (Holderread, 1978; Su, 2022). Female Mallards possess a good mothering ability, a trait that is particularly useful when feed resources are scarce during brooding (Holderread, 1978).

With the mountain of evolutionary genetic evidence, undoubtedly, the Mallard duck is regarded as the "Adam and Eve" of today's domestic duck breeds, except Muscovy ducks. The Mallard duck uniquely displays plumage dimorphism, unlike the Muscovy duck, which only displays sexual dimorphism in growth parameters. In Mallard, plumage mosaicism is astonishingly more beautiful in drakes than hens (Grow, 1985).

In Nigeria, the prominent plumage colours are black, brown, green, and an admixture of black and water (Oguntunji *et al*., 2020). Mallard ducks are known to have a characteristic ring-shaped colour on their necks. The ring colour often depends on the plumage colours; that is, ducks with brown plumage have brown rings and ducks with white or variations of white plumage colour have white rings.

Plumage colours and sexual dimorphism in Nigerian mallard ducks

The plumage colours of Nigerian Mallard ducks, as reported by Oguntunji *et al*. (2020), suggest unambiguously that the plumage phenotype is a sex-limited or -dependent trait. Furthermore, this observation asserts that the mode of inheritance of plumage colour is autosomal and sexually dichromatic. Similarly, the notable green-coloured head seen in Nigerian Mallard males and females suggests that this specific colour phenotype is sex-influenced (Oguntunji *et al*., 2020). This colour phenotype is sex-influenced because, unlike sex-limited traits, it is present in both sexes. The testosterone-influenced glossy green head seen in Mallard males is a characteristic used in their identification.

It has been argued that the plumage colour variation seen in Mallard ducks could be explained solely by male and female sex hormones (Owens and Short, 1995). The gravamen of the hormone-controlled plumage dichromatism is the presence of bright, glossy head colour in drakes and dull colours in hens. In other words, the estrogen hormone in female ducks is responsible for the dull plumage colours, while testosterone is responsible for the bright plumage colours in male ducks. However, Kimball (2006) posited that the quantitative absence or presence of estrogens is pivotal in the existence of iridescent plumage colours in the sexes of birds belonging to the orders Anseriformes, Galliformes, and Struthioniformes.

Additionally, the presence of shimmering plumage colours in Mallard males – an avian species that belong to the Anseriformes order – has no causal link with the presence of testosterone. In lieu, the bright and rainbow-like plumage colour in Mallard males is the consequential colour phenotype of an ovary-devoid biological system. Plumage colours could be useful tools when determining the sex of day-old ducks.

Muscovy ducks

The locally adapted Muscovy ducks in Nigeria are scientifically classified as *C. moschata domestica*. Without a doubt, it is known that today's locally adapted Muscovy ducks are the offspring of ducks native to South American countries, such as Brazil, Mexico, and Uruguay (Smith, 2022). The etymology of the name Muscovy is tied to the feeding habit of their wild ancestors: they consumed numerous mosquitoes; hence, the name Musco, is a shorthand vernacular rendition of mosquito. It is generally agreed that Muscovy ducks were introduced to Nigeria after colonization (Arias-Sosa and Rojas, 2021). In Nigeria, Muscovy ducks are mainly found in the riverine regions of Southern Nigeria (Yakubu, 2013). Muscovy ducks are reared in rural farming communities because, one, they are a source of meat; two, the breed is resistant to most poultry diseases; three, the breed is relatively easier to rear than chickens (Ramos, 2009; Salgado-Ubeda and López-Mendonza, 2012).

As with other poultry species, the most important economic trait in Muscovy ducks is their slaughter weight. Even though this duck breed has the genetic potential to be a fast-growing bird, the environmental conditions in which it is reared play a crucial role in its growth rate and feed conversion ratio (FCR) (Arias-Sosa and Rojas, 2021). For instance, in Nigeria, depending on the type of production system, the mature body weight of male duck (drake) varied between 2.0 kg and 2.7 kg, while for female ducks (hens), it ranged between 1.5kg and 1.6kg (Yakubu 2011; Omojola, 2007). Although, there are reports of an increase in body weight after force-feeding in France (Larzul *et al*., 2006), Poland (Wawro *et al*., 2004), China (Wen *et al*., 2016) and Egypt (Shamma *et al*., 2011). However, this method of increasing weight gain in ducks and other poultry species is considered unethical, as it does not consider the welfare of the birds. Furthermore, force-feeding could have deleterious effects on the health status of ducks (Skippon, 2013).

In duck rearing, the system of production greatly alters the FCR. Etuk *et al*. (2006) reported that, under an extensive system of production, a high FCR was observed.

On the other hand, under the intensive production system, a low FCR of 2.2-4.7 was observed (Shamma *et al*., 2011) – which is, arguably, within the desirable range for FCR; the lower the FCR, the better. Consequently, the genetic merit of Muscovy ducks will become evident under proper management systems and selective breeding.

Even though Muscovy ducks are relatively more resistant to infectious avian diseases, biosecurity and sanitary measures must be implemented on duck farms to avoid the outbreak of diseases. Infectious diseases can cause weight loss and death in ducks (Arias-Sosa and Rojas, 2021), leading to economic loss for the owner. Muscovy ducks are particularly susceptible to viral diseases, such as duck Viral Enteritis (DVE), Muscovy Duck Reovirus (MDRV), Newcastle Disease (NCD), Muscovy Duck Parvovirus (MDPV), Highly Pathogenic Avian Influenza (HPAI) virus (Arias-Sosa and Rojas, 2021). Consequently, it is requisite for duck farmers to strictly follow vaccination schedules to reduce the spread and outbreak of viral infections. Additionally, the use of probiotics could enhance the performance and health of Muscovy ducks (Sheng-Qiu *et al*., 2013; Xie *et al*., 2015; Kamollerd *et al*., 2016; Anggraeni *et al*., 2018).

Sexual dimorphism in muscovy ducks – A blessing or a curse?

Some of the distinct features of the Muscovy duck, when juxtaposed with its counterpart, the Mallard duck, are its large body size and low caloric meat (Oguntunji and Ayorinde, 2014). A plausible explanation for phenotypic variation is that, in Mallard duck, there is a significant increase in the protein expression pathways associated with lipid metabolism (Arias-Sosa and Rojas, 2021).

Presumably, anything that has advantages or benefits will have disadvantages and the Muscovy duck is no exception. In terms of economic traits, the sexual dimorphism observed in Muscovy ducks irreversibly favours males over females. That is, higher body weight and lower feed conversion ratio have been reported in male Muscovy ducks (Etuk *et al*., 2006; Yakubu, 2011; Oguntunji and Ayorinde, 2014). Concomitantly, the meat obtained from male Muscovy ducks has better sensory properties (Omojola, 2007). Consequently, sexual dimorphism in Muscovy ducks causes an increase in the cost of production and lower profit margins in Muscovy females (Arias-Sosa and Rojas, 2021).

Animal breeders have implemented crossbreeding methods, involving Mallard and Muscovy ducks, to address this unintended curse of sexual dimorphism. The resulting crossbreed is called a mule – a name poignantly describing its infertility – which has better growth performance indices and lower expression of sexual dimorphism (Larzul *et al*., 2006; Chartrin *et al*., 2006; Wen *et al*., 2016).

Caruncle: A distinct feature of muscovy ducks

The caruncle is an anatomical feature present in Muscovy ducks but absent in Mallards. Caruncles are tiny, thick outgrowth found on the head and circumference of the eyes of Muscovy ducks. Different colour phenotypes are observed on the caruncle; these colours are black, red, commixture of red, black, and faint yellow (Oguntunji and Ayorinde, 2015).

Egg production and reproductive performance in muscovy ducks

Typically, *C. moschata* is reared for its meat, however, some farmers rear this duck for their eggs (Yakubu, 2013). It is within reason that some individuals prefer to consume duck eggs; for one, it possess enormous amounts of (animal) protein and considerably low amounts of carbohydrates (Arias-Sosa and Rojas, 2021). Furthermore, duck eggs contain high amounts of minerals, such as iron, zinc, and calcium. Similarly, compared to chicken eggs, duck eggs have more albumen, making them valuable baking ingredients (Oluwafemi and Udeh, 2016).

The production system has considerable effects on the egg production and sexual maturity of ducks. Ducks reared under an extensive system where uncontrolled mating is allowed, female ducks attain sexual maturation between 210-270 days, with an egg production of 60-80 every year and a 34-day incubation time (Salgado-Ubeda and López-Mendonza, 2012; Yakubu 2013).

On the other hand, under an intensive system of production with controlled mating and breeding programmes, Muscovy ducks can lay between 100-125 eggs yearly (Yakubu, 2013). The system of production equally affects the egg weight: eggs produced under intensive management systems have a mean egg weight between 76g and 78g, while less intensive systems produce mean egg weights ranging from 71g to 72g (Salgado-Ubeda and López-Mendonza, 2012; Yakubu 2013). As with chickens, environmental changes associated with dry and rainy seasons affect egg production in Muscovy ducks (Oguntunji *et al*., 2015).

Rashid *et al*. (2013) reported that the reproductive performance of Muscovy ducks is between 86.7%-97%, provided there is a drake-to-hen ratio of 1:6 or 1:5. Using natural incubation, a hatchability of 70% was achieved under semi-intensive farming (Rashid *et al*., 2013). Higher percentages of hatchability (73.3%) have been reported using artificial incubation, with a humidity-temperature ratio of 58%:37.5°C (Arias-Sosa and Rojas, 2021).

PROLACTIN GENE: SNPs AND INDELs

The reported single nucleotide polymorphisms (SNPs) and insertion and deletion (INDELs) of the prolactin (*PRL*)

Table 1. Reported SNPs in the *PRL* gene of ducks.

SNP	Region	Source
C-213T	5' flanking region	Wang *et al*. (2011)
C-381A	intron 1	Wang *et al*. (2011)
A-412G	intron 1	Wang *et al*. (2011); Bai *et al*. (2019)
T-1326C	intron 1	Li *et al*. (2009)
T-2231C	Intron 2	Wang *et al*. (2011)
T-3941G	Intron 4	Chang *et al*. (2012); Indriati *et al*. (2016); Purwantini *et al*. (2020)
C-3975A	Intron 4	Chang *et al*. (2012); Indriati *et al*. (2016); Purwantini *et al*. (2020)
C-3949T	Intron 4	Wang *et al*. (2011)
T-3988G	Intron 4	Wang *et al*. (2011)
T-4009C	Intron 4	Wang *et al*. (2011)
T-4110C	Intron 4	Indriati *et al*. (2016)
C-5796A	Intron 4	Damayanti *et al*. (2022)
T-5817C	Intron 4	Damayanti *et al*. (2022)
A-1842G	Exon 2	Wang *et al*. (2011)
A-3869G	Exon 4	Wang *et al*. (2011)
T-3777C	Exon 4	Wu *et al*. (2008)
A-3785G	Exon 4	Wu *et al*. (2008)
C-5961T	Exon 5	Wang *et al*. (2011)
T-6052A	3' flanking region	Wang *et al*. (2011)

Table 2. Reported INDELs in the *PRL* gene of ducks.

INDEL	Region	Type	Source
3724A	Intron 3	Deletion	Indriati *et al*. (2016)
3939A	Intron 4	Insertion	Indriati *et al*. (2016)
4031A	Intron 4	Deletion	Indriati *et al*. (2016)

gene is evidence for the gene's candidacy in egg reproduction in ducks (Wang *et al*., 2011; Bai *et al*., 2019).

In avian species, the gene coding for *PRL* is found on chromosome 2 (Alipanah *et al*., 2011; Miao *et al*., 1999). *PRL* in birds consists of four introns and five exons (Li *et al*., 2009; Yousefi *et al*., 2012). Two regions of the avian *PRL* gene perform regulatory functions, namely the proximal and distal enhancers (Wilkanowska *et al*., 2014).

After the successful cloning of the avian (for example, turkey, duck, chicken, quail, and pigeon) *PRL* gene, polymorphic regions or sites have been discovered in both exonic and intronic regions. It is not enough to discover SNPs, polymorphic sites, or mutations; discovered polymorphism or polymorphic will be of interest when they have a significant relationship with quantitative traits, such as body weight, egg weight, egg size, and egg production.

In the duck prolactin (*dPRL*) gene, the majority of the polymorphisms reported were in the coding regions and 5'- and 3'-flanking regions (Li *et al*., 2009). Similarly, the SNP C-5961T in exon 5 is reportedly significantly associated with egg weight and production in ducks (Wang *et al*., 2011). Furthermore, an insertion of 24 base pairs (bp) in the promoter region of *PRL* in birds had a positive correlation with increased egg production (Jing *et al*. 2009; Kulibaba and Podstreshnyi, 2012). The highly polymorphic nature of the *PRL* gene in avians and, its significant association with economic traits, suggest that it will be a good genetic marker for egg-laying-related traits. A list of published SNPs and INDELs found in the *PRL* gene is shown in Tables 1 and 2, respectively. Additionally, Oyebanjo *et al*. (2023) reported 21 previously unreported SNPs in the exon 5 and 6 INDELs in both intron 4 and 3' flanking region of *PRL* gene of locally adapted Muscovy and Mallard Nigerian ducks.

The transcription of *PRL* is initiated by a transcription factor (TF) known as pituitary-specific TF 1 (PIT-1) (Ohkubo *et al*., 2000). Furthermore, this TF stimulates the Thyroid Stimulating Hormone Subunit Beta (TSHB) and growth hormone gene promoters (Nie *et al*., 2008). Similarly, prolactin receptor stimulation initiates a torrent of signalling pathways, one of which is the Janus kinase 2-signal transducer and activator of the transcription 5 (JAK2-STAT5) pathway (Bu *et al*., 2013). The PIT-1 plays a vital role in the expression regulation of the *PRL* gene.

In chicken, expressions of the *PRL* gene were observed in the pituitary gland, oviduct ovaries, and hypothalamus; however, the most substantial expression was noted in the pituitary gland (Li *et al*., 2009; Wilkanowska *et al*., 2014). Chu *et al*. (2008), similarly, reported a high expression of the *PRL* gene in the pituitary gland of geese, then in the hypothalamus – and, as expected, the lowest expression was observed in the ovary.

The biological molecule encoded in the *PRL* gene is prolactin, a polypeptide hormone. The partial coding sequence for prolactin hormone has 199 amino acids (Liu *et al*., 2008), while the complete coding sequence has 229 amino acids (Indriati *et al*., 2016). Prolactin hormone has three disulfide bridges interspersed across six highly conserved cysteine residues (Wilkanowska *et al*., 2014). The molecular weight of the prolactin hormone is 23kDa (Kansaku *et al*., 2008).

Prolactin hormone is secreted in the anterior pituitary gland (Wilkanowska *et al*., 2014). In the anterior pituitary gland, lactotroph cells are responsible for the production of the hormone. However, lactotrophs are not the only source of biological manufacturers of prolactin, as considerable amounts of prolactin have been discovered in other organs and cells, such as the spleen, thymus, epithelial cells, and lymphocytes (Wilkanowska *et al*., 2014).

Prolactin releasing factors

Prolactin production in avians is primarily controlled by various releasing factors, such as dopamine, serotonin, and vasoactive intestinal peptide (VIP) (Kagya-Agyemang *et al*., 2012). Additionally, in birds, the hypothalamus is an important factor in prolactin secretion by producing prolactin-releasing factor (PRF) or prolactin-inhibiting factor (PIF) (David *et al*., 2003; Al Kahtane *et al*., 2005).

For instance, VIP is a crucial neuroendocrine chemical secreted by the hypothalamus that heavily affects prolactin production in avians (Kuenzel, 2003). Additionally, VIP has been used to induce the release of prolactin, alongside its expression, in birds, under *in vivo* and *in vitro* conditions (Al Kahtane *et al*., 2005), making it a PRF.

Although VIP plays a role in prolactin release, it is not the main actor, the hypothalamus is. The hypothalamus' tonic inhibitory dopaminergic control enables it to regulate prolactin secretion (Tong *et al*., 1998). Prolactin production is repressed by dopamine, a neurotransmitter, and hormone secreted by the hypothalamus-housed Tuberoinfundibular Dopamine (TIDA) Neurons (Chang and Shin, 1999; Freeman *et al*., 2000). Dopamine regulates prolactin production by inhibiting the work of VIP in the anterior pituitary gland. This inhibition is achieved through the activation of D_2 Dopamine receptors resident in the anterior pituitary gland (Wilkanowska *et al*., 2014). Broodiness in hens was halted after the administration of dopamine receptor antagonists because the prolactin secretion was greatly inhibited (Xu *et al*., 2010).

Serotonin is another neurotransmitter involved in the release of prolactin in birds. A technical name for the type of serotonin involved in prolactin release is 5-hydroxy-tryptamine (5-HT). Stimulation of prolactin production by serotonin is facilitated by VIP. This assertion was confirmed through the work of El Halawani *et al*. (1995), where it was proven that, in Turkey, the assistance of VIP was consequential in serotonin-stimulated prolactin release.

Other non-biological factors influence prolactin secretion in birds, and the environment is one of the factors. Contained in the environment are various stressors and stimuli that can affect prolactin secretion (Dusza and Ciereszko, 2007). For example, in avian species, temporary stressors elicit a corresponding reduction in the level of prolactin (Angelier and Chastel, 2009). Light is an environmental factor that affects prolactin levels in birds. In a controlled experiment involving the application of photostimulation on pigeons, quail, and ducks, prolactin levels in the serum increased (Erdost, 2005). Notably, it is counterproductive to increase the prolactin level, hoping to have highly producing ducks, because an increase in prolactin level is associated with an absolute termination of egg production (Kulibaba and Podstreshnyi, 2012).

BIOINFORMATICS

The field of bioinformatics is an ever-evolving landscape in the biological sciences. From the epochal development of Sanger sequencing in 1977 to the automated second- and third-generation sequencing of 2004, bioinformatics has followed an upward and forward direction. Consequently, it extends our scientific understanding of genetics, genomes, and genes. Animal breeding is not exempted from the benefits of bioinformatics because it is intricately tied to genetics.

Bioinformatics involves using computational techniques or algorithms, to analyse biological information; for example, information encoded in Deoxyribonucleic acid (DNA), Messenger RNA (mRNA), and protein, to understand the biological complexities, evolutionary relationships, and inheritance patterns of complex traits. The widespread use of bioinformatics and computational techniques in biosciences suggests that more researchers are seeing its relevance. At the outset, the conceptual framework for bioinformatics revolved around using it for database creation and maintenance, which will also enable the easy retrieval of DNA and protein sequences.

It was later realised that there would be a need for researchers to update the existing sequences and submit new ones to databases. The application of bioinformatics was revised accordingly. Now, the analysis of protein domains, motif prediction, protein structures, and gene annotation has been incorporated into the applications of bioinformatics.

Bioinformatics: Its nostalgic history

A quick survey of the early frontiers and imprints of bioinformatics suggests that what it is today is lightyears away from what its proponents envisage. Simply put, bioinformatics, in its early days, was a systematic method of processing biological processes (Hesper and Hogeweg, 1970; Hogeweg, 1978; Hogeweg, 2011). This clear definition made bioinformatics at par with biochemistry – the latter was largely concerned with elucidating the chemical processes underlying biological systems (Hogeweg, 2011).

The relevance of computers and computational methods in molecular biology garnered attention after the insulin sequence, determined by Frederick Sanger, was made publicly available. As expected, sequence comparisons were initially done manually. Soon, the inefficiency of this method of sequence comparison became evident, as it was error-prone and tedious. Even though this method is now considered crude, it was, at that time, a starting point and an actionable solution for the interpretation of nucleotide and protein sequences.

Margaret O. Dayhoff pioneered sequence alignments and molecular evolution analysis using manual methods (Eck and Dayhoff, 1966). Dayhoff made substantial contributions to the field of bioinformatics by engineering the transition for sequence comparison from books to public databases (Dayhoff, 1965).

After the development of the Sanger sequencing method in 1977 (Sanger *et al*., 1977), other methods of DNA sequencing (for example, the Maxam-Gilbert sequencing method) were developed and used to sequence bacteriophages ØX714 and MS2 (Maxam and Gilbert, 1977). The generated DNA sequences were analysed using existing knowledge of molecular biology and algorithms (Altman and Erickson, 1981). The use of statistical methods revealed that biological information, such as coding regions or segments and codons, could be identified easily, proving that bioinformatics will be a useful tool in molecular biology (Yang *et al*., 1984).

Bioinformatics: The current clime

Ever since its inception, bioinformatic methods have been used to create several databases, through which researchers can corroborate, exchange, store, retrieve and analyse data within a biological context (Singh *et al*., 2018). Currently, there are several public access databases where nucleotide and protein sequences, among others, of various species, breeds, and strains can be obtained.

Since humans domesticated animals, there have been several successful attempts to select and breed highly-producing animals. Although, then, the genetic intricacies surrounding heredity, selection, and breeding were yet uncovered. The advent of bioinformatic methods and algorithms served as harbingers for the all-encompassing changes that will propel the field of animal breeding and genetics forward.

In animal genetics, various scientific inquiries are performed to illuminate and understand the complexities underlying the inheritance of traits, and what causes the (genetic or phenotypic) variation of said traits within and between populations (Habier *et al*., 2007). The rapid development of novel tools and high-throughput techniques in animal genetics creates a need for advanced computational and or bioinformatic tools in the analysis of biological data. This integration of bioinformatics and animal genetics is useful in the creation of a multi-pronged approach to scientific discovery.

Although a larger aspect of animal genetics sub-disciplines, such as population genetics, greatly utilise (bio)statistics to understand the genetic structure, selection pressures, and genetic progress of a population, there are still roles that bioinformatics can play (Habier *et al*., 2007). The information contained in the DNA sequences of animals is useful to quantitative geneticists to properly elucidate the genetic diversity of a population and selective breeding. For instance, marker-assisted selection (MAS) is useful in quantitative genetics because the genetic markers used encompass the entire genome. Consequently, all the quantitative trait loci (QTL) are automatically in linkage disequilibrium (LD) with one or more genetic markers (Habier *et al*., 2007). The creation of this selection technique was possible due to the use of bioinformatics tools in SNP discovery through deep sequencing and throughput SNP genotyping.

Bioinformatics: Its application to animal breeding and genetics

After the domestication of several animal species, it was only corollary that mankind will select and breed these animals to get better animal products. Initially, phenotypic appraisals were used to select animals that will parent the next generations. The advent of genotyping, sequencing, and bioinformatics reduced the reliance on phenotypic appraisals in selection programmes.

Significant progress was made in the field of quantitative genetics by exploiting the foundational theories and equations proposed by Fisher, Lush, and Wright. These theories allowed for the development of multifactorial models and structured breeding programmes based on Mendelian laws of inheritance. However, there are other aspects of animal genetics, such as gene identification, protein-to-protein interactions, functional annotation, transcriptomics, motif analysis, and mutation prediction (Daetwyler *et al*., 2007) that quantitative genetics models cannot capture. This limitation or, better yet, insufficiency is why bioinformatics is useful to animal breeding and

genetics.

Large sequencing data, such as whole genome sequencing and whole exome sequencing, can be analysed using bioinformatic tools. Such analysis will be useful in the understanding of the effects of SNPs, INDELs, copy number variations (CNVs) mutations, and functional sequence variants on quantitative traits. This knowledge can provide relevant directions for genetic improvement programmes (Daetwyler *et al*., 2007). Additionally, biological data, such as an animal's genome, epigenome, pangenome, transcriptome, and proteome, were created using bioinformatic algorithms. Studies such as genome-wide association studies, especially those that yield genome-wide SNP data, provide relevant information for animal genetic improvement and climate change-conscious livestock production. Undoubtedly, bioinformatics will play a vital role in achieving said goal.

CONCLUSION

Genetically improving duck egg production would have practical implications for the poultry industry in Nigeria, where duck eggs can be utilized as a complementary or replacement option for chicken eggs, contributing to improved nutrition and livelihoods. This genetic improvement can be achieved when the use of genetic or biological information is properly appreciated and duck rearing becomes more commercial. Bioinformatic tools are very important in actualising genetic improvement in Nigerian ducks because they will aid in the understanding of the *PRL* gene in all its complexities. Furthermore, genetic information, which is the basal data for bioinformatic analysis, is not riddled with errors, compared to morphological data. Lastly, coupled with genetic principles, the results obtained from the bioinformatic analysis of the *PRL* gene can be used to design breeding strategies specifically for locally adapted Nigerian ducks.

RECOMMENDATION

The prolactin gene is a critical gene in poultry production and for profitability, therefore, grant bodies, animal geneticists, and bioinformaticians must work together using bioinformatic tools to understand the *PRL* gene and then genetically improve duck egg production in Nigeria. This knowledge will pave the way for targeted breeding strategies, promoting improved nutrition and livelihoods through the utilization of duck eggs.

DISCLOSURE STATEMENT

The authors declare that they have no conflict of interest.

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