

Comparative Histological Study of Glycogen Content in Skeletal Muscles of Green-Winged Teal and Domestic Duck

Zahraa Jasim Hussein  , Asraa Abdul Khalq Zegyer 

Department of Biology, College of Science, Wasit University, Wasit, Iraq

Email: std2023204.zzwain@uowasit.edu.iq

Abstract

This study aimed to detect and compare the glycogen content in the skeletal muscles of Green-Winged Teal and domestic ducks. The birds used in this study were purchased from private commercial markets in Kut City and included 10 adult male Green-winged Teal and 10 adult male domestic ducks. Skeletal muscle samples were taken from the chest and leg regions. Tissue sections were prepared from these samples and stained with acid-Schiff's periodic reagent to determine the amount of glycogen in them. The results of the study showed variation in the glycogen content of the two bird species. The different amounts of glycogen in skeletal muscle are related to different species and muscle fiber types.

Keywords: *Green-wingedTeal, muscle, glycogen.*

I. Introduction

Across all countries, customs, and faiths, poultry meat and eggs are among the most widely consumed livestock-based foods worldwide. Over the last several decades, the consumption of poultry meat and eggs has generally increased (Ismoyowati and J Sumarmono, 2019). Ducks are a diverse group of waterfowl that play significant roles in both natural ecosystems such as nutrient cycling and habitat creation and human economies. They are primarily raised for their meat, eggs, and feathers, with the Mallard (*Anas platyrhynchos*) being the most common ancestor of domestic duck breeds (Yang Zhang, 2023).

Meat and egg prices are beyond the purchasing power of the poor. Increasing duck egg and meat production will play an important role in addressing these issues by providing a more affordable source of protein (Krainyk, A., 2018).

Packed with excellent proteins, beneficial unsaturated fats, essential vitamins, and valuable minerals, duck meat presents a significant nutritional profile that yields several health benefits. This includes fortifying the immune system, supporting a healthy cardiovascular system, and assisting in muscle development, according to research by (Cheng, C. in 2024).

The Domestic duck (*Anas platyrhynchos*) is a common and broadly distributed species, especially across the Northern Hemisphere. In someregions, it is an important component of the diet of local inhabitants. (Agnieszka Sujak, 2019)



Teal with green wings All four North American flyways are home to *Anas crecca*, often known as teal, a socioeconomically significant species that is the second most harvested duck in the region (Raftovich et al. 2018).

The teal, which is the smallest type of dabbling duck found in North America, looks for food in shallow water-ranging from 0 to 12 cm deep -as well as on mudflats. Within these environments, other species pose a limited risk to the teal's ability to obtain small seeds and invertebrates (as cited in Baldassarre 2014; Johnson et al. 2020). Skeletal muscle fibres are large, multinucleated cells that mediate voluntary movement and maintain body posture. These fibres are controlled by the somatic nervous system and are essential for precise movements (Morton et al., 2019).

The muscle's structure includes various components like the neuromuscular junction, sarcomere, and cytoskeleton, which are crucial for its function (Brooks et al., 2023).

Animals with a high rate of protein synthesis and energy consumption have skeletal muscle, a large, metabolically active tissue. The main components of muscle tissue are proteins, which also control metabolic functions (Lana and Zolla, 2016). Muscles, essential organs with an amazing capacity for contraction, make up over half of the body's weight. Muscle tissue is categorized based on differences in shape and function (Krause, 2005). Vertebrates have three different types of muscle tissue: cardiac, skeletal, and smooth (Marieb and Hoehn, 2010).

According to Mukund and Subramaniam (2020), skeletal muscle is a well-organized tissue made up of several bundles of muscle fibres, or myofibers. Each myofiber represents a muscle cell, with the sarcomere serving as its fundamental cellular unit. Myofibrillar proteins are composed of structural, contractile, and regulatory proteins. Actin and myosin are contractile proteins forming thin and thick filaments; they regulate the contraction and relaxation of skeletal muscle (Hopkins, 2014). Muscle fibres are categorized using a number of factors, including myoglobin concentration (colour), glycogen, fatigue, and other traits (Lamboley et al., 2014). Glycogen is a complex polysaccharide that can be rapidly mobilized to provide glucose. Glycogen is stored in the liver and skeletal muscle of animals (Moran et al., 2012). In animals of comparable size, glucose levels in the bloodstream of domestic and wild birds are notably elevated compared to those found in mammals (Braun and Sweazea, 2008).

Even in a "limited age family with similar nutrition and genetics," there is a noticeable difference in the reported glucose levels of birds, even when they are under the same physiological settings (Lu et al., 2007). Following the butchering process, the transformation of muscle into edible meat, alongside the transition from an egg to a self-sufficient existence, and the supply of vital energy throughout the early life stages of avian species, are all thought to be largely impacted by the presence of glycogen stored in both the liver and muscle tissues (Christensen et al., 2001).

Skeletal muscle contains around 1.5-1.8% glycogen, which makes up 3-5% of the liver's weight. It is the primary source of blood glucose. Meat quality hinges significantly on the muscle glycogen content at the point of animal processing (Meléndez et al., 1999). This level is known to fluctuate, and is influenced by several elements, which include the muscle type involved, how well-nourished the animal has been, and the amount of stress it experienced before slaughter (Chauhan, S.S. et al., 2019).

Postmortem muscle acidification is necessary to transform muscle into higher-quality meat after slaughter. When blood flow is interrupted, the capacity to buffer pH changes is greatly reduced and anaerobic carbohydrate breakdown serves as a homeostatic mechanism for ATP production. As a result, muscles become more acidic, with pH levels



falling to 5.5. This acidity is influenced by the amount of carbohydrates present and the ability of the muscles to withstand pH changes (Pösö and Puolanne, 2005).

Previous research has shown that one of the primary elements determining variations in the meat's water-holding capacity, final pH (pHu), and other physical features, such as colour, is the quantity of muscle glycogen in birds (Przybylski et al., 2006).

There is a strong, significant negative association between the eventual pH of meat and the amount of muscle glycogen at the time of slaughter (Le Bihan-Duval et al., 2008). Decreased glycogen reserves limit the production of lactic acid during the process of converting muscle into meat after slaughter, resulting in a decrease in pH levels (Rosenvold et al., 2001). When meat's final pH is low, it becomes "acid meat," which appears pale and has a limited ability to hold water. On the other hand, when the ultimate pH is high, it results in DFD (dark, firm, dry) meat that is dark in colour (Sibut et al., 2011).

Stress is the deadliest enemy of muscle glycogen at the time of slaughter. Stress can be physical, psychological or both. The best way to ensure high-quality meat, the well-being of the slaughtered animals and financial stability is to keep the animals from potential stress. Lack of food and water, strange smells, vibrations and changes in acceleration, extremely high or low temperatures, and disintegration of social groups (Immonen et al., 2000) These stressful circumstances can cause an increased expenditure of the reserve, which decreases the concentration of muscle glycogen. Consequently, this may hinder proper meat acidification, potentially negatively impacting its quality (Muchenje et al., 2009).

II. Materials and Methods

Experimental design

The birds for this study were collected from private commercial fields in the city of Al-Kut. These birds included 10 male of adult Domestic duck and 10 male of adult Green-winged Teal.

Through gross examination, it was found that all the birds were healthy, and then all birds were sacrificed by being anesthetized and deadened with chloroform.

The samples collection

The skeletal muscle specimens (5mm×5mm×5mm) were taken from the chest area (pectorals major muscles) and leg (extensor iliotibialis anticus muscles).

The samples were preserved in a 10% formalin solution for 72 hours. They were then rinsed with water for 2-3 hours and subsequently subjected to various histological techniques, such as dehydration, clearing, infiltration, embedding, cutting, and staining. The periodic acid Schiff's reagent stains were used to determine glycogen content in paraffin sections.



Methodology statistic

The slides were captured with a Canon digital camera that was mounted on a Meija microscope equipped with a 1/2 X photo converter and a 40X objective lens. The output photographs were examined on an Intel Core I3 computer using Video Test Morphology software (Russia), which has a dedicated feature for measuring area, percentage area, and counting objects.

III. Results and Discussion

Histochemical results of skeletal muscle were used to estimate the glycogen content in the chest and leg regions of winter mallards and house ducks after staining the muscle sections with PAS...

In Green-winged Teal, microscopic examination of cross sections of the chest muscles revealed the accumulation of glycogen deposits within muscle bundles, which appeared as a dark purple color, as shown in (Figure 1, A).

When examining cross sections of the leg muscles, glycogen was found within muscle bundles, as shown in (Figure 2, A). Statistical results showed average glycogen deposition in the leg and chest muscles (7.897 and 5.590), respectively, as shown in the surface plots of the cross sections of the muscle samples (Figures 1, 2, B).

In domestic ducks, microscopic examination of cross-sections of breast muscles showed a large accumulation of glycogen deposits within the muscle bundles, which appeared as a dark purple color, as shown in (Figure 3, A). Cross-sections of leg muscles showed a small accumulation of glycogen scattered within the muscle bundles, which appeared as a dark purple color, as shown in (Figure 4, A).

Statistical results showed that the average glycogen deposition in the leg and chest muscles was (9.919, 14.275), respectively, as shown in the surface plots of the cross-sections of the muscle samples (Figures. 3, 4, B).

Microscopic examination of skeletal muscles in green-winged ducks revealed moderate glycogen accumulation, while leg muscles showed minimal deposition. The differences are attributed to muscle fiber type: domesticated ducks have more glycogenolytic fibers that store a higher percentage of glycogen, while green-winged ducks have more oxidative fibers suitable for flight (Choi and Kim, 2009).

The results of current study were appeared the different content of glycogen both species of birds in the study, the glycogen content in skeletal muscles of Green-Winged Teal was less than in comparison with skeletal muscle of Domestic duck (chart 1). This finding was similar to previons study were mentioned by (Awan et al. 2017). In all probability, the different quantity of glycogen in skeletal muscle related to species variation and, fibers muscle type, this view was supported with studies of some researchers (Talpur et al.. 2018).



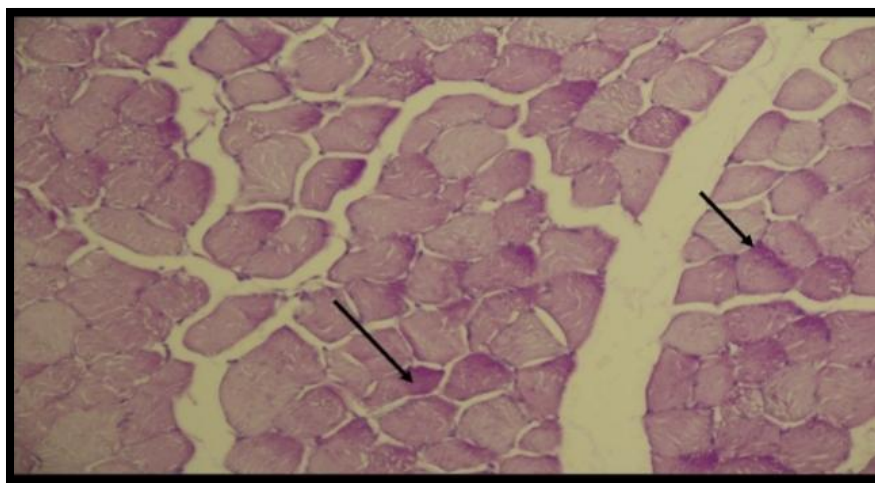


Figure (1-A) : Photomicrograph of cross section in leg skeletal muscles of Green Winged Teal, show the gradual increase in the distribution of glycogen (black arrows). PAS stain, 200 X.

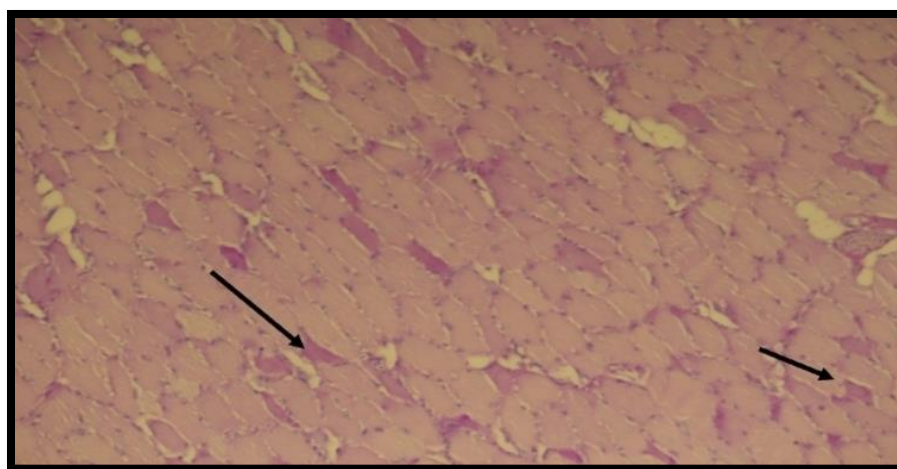
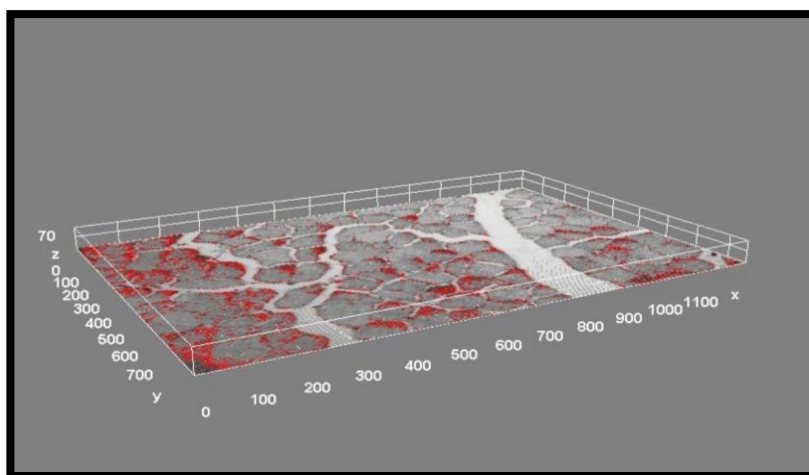
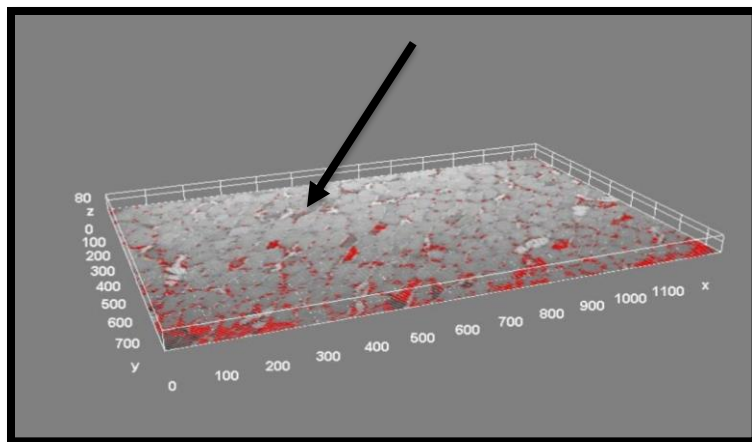


Figure (2-A): Photomicrograph of cross section in chest skeletal muscles of Green Winged Teal, show the gradual decrease in the distribution of glycogen (blue arrows). PAS stain, 200X

Figure (1-B): Surface plot show of cross section in skeletal muscles fibers of the leg area in Green-winged Teal.





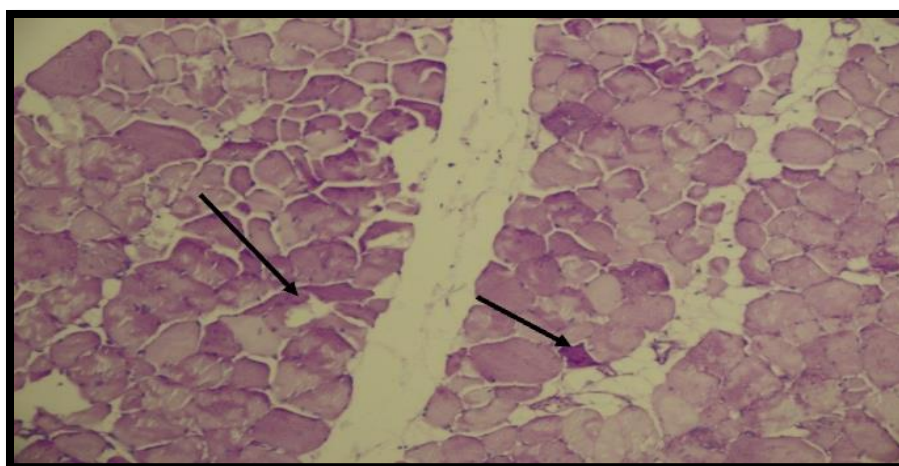
Figur (2-B): Surface plot show of cross section in skeletal muscles fibers of the chest area in Green-winged Teal.

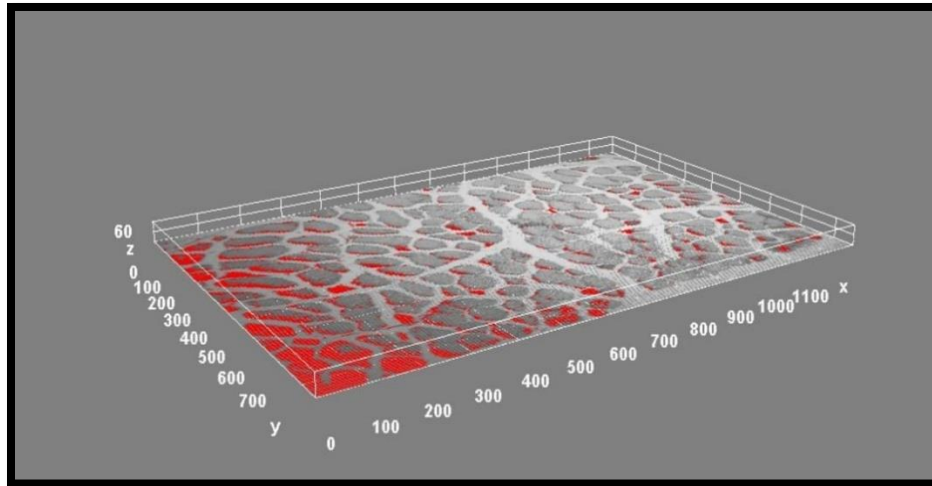
Figure (3-A): Photomicrograph of cross section in leg skeletal muscles of Domestic duck, show the gradual decrease in the distribution of glycogen (blue arrows). PAS stain, 200X.

Figure (3-A): Photomicrograph of cross section in leg skeletal muscles of Domestic duck, show the gradual decrease in the distribution of glycogen (blue arrows). PAS stain, 200X.



Figure (4-A): Photomicrograph of cross section in chest skeletal muscles of Domestic duck, show the gradual increase in the distribution of glycogen (black arrows).PAS stain, 200X.





Figure(3-B): Surface plot show of cross section in skeletal muscles fibers of the leg area in Domestic duck.

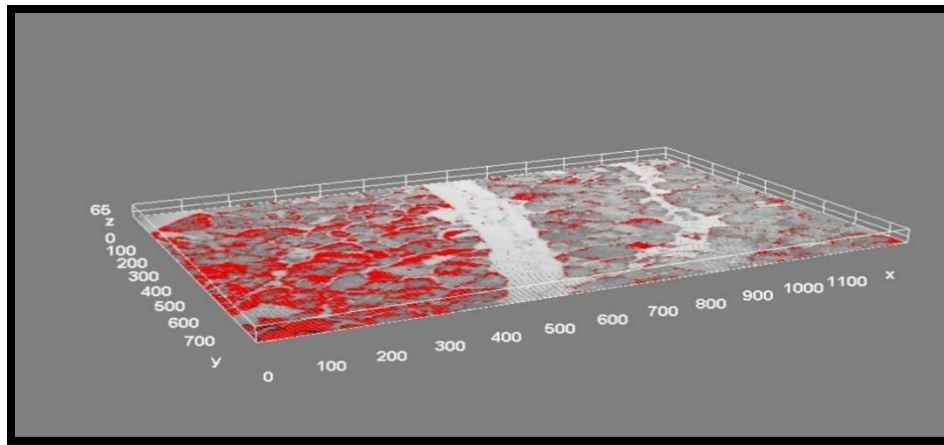
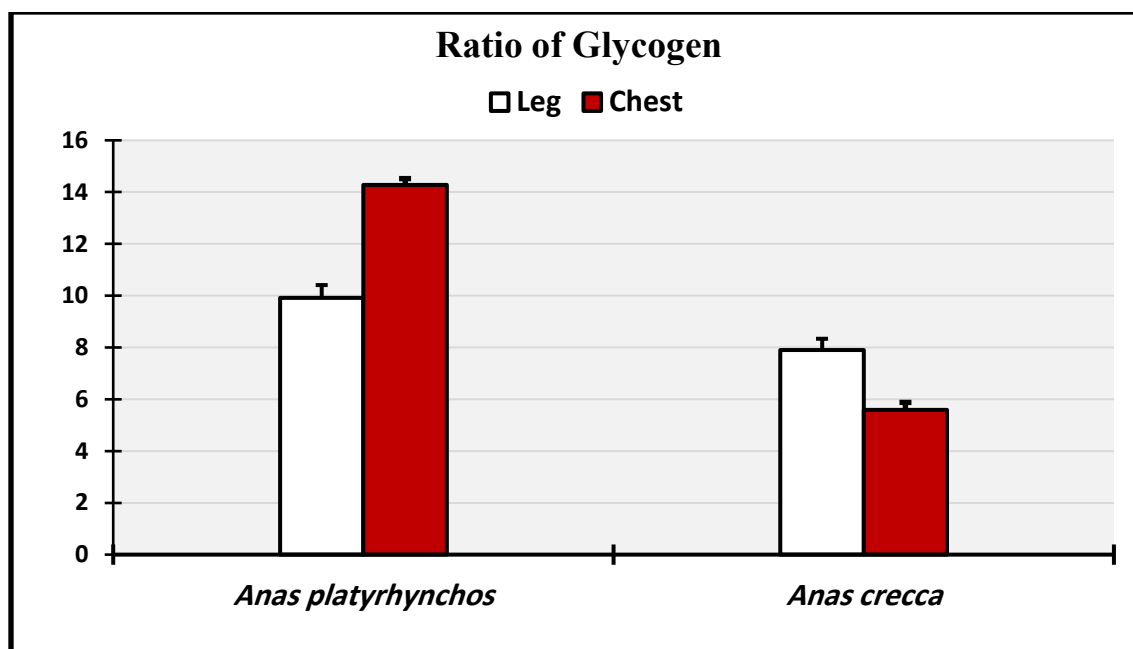


Figure (4-B): Surface plot show of cross section in skeletal muscles fibers of the chest area in Domestic duck.



(Chart 1): The Ratio of glycogen content in chest and leg areas of the skeletal muscles in Domestic duck (*Anas platyrhynchos*) and Green-winged Teal (*Anas crecca*).

IV. Conclusion

The staining of periodic acids Schiff's reagent stain appeared to indicate the glycogen content in muscle samples for both types of birds; the Green Winged Teal muscles showed less glycogen than the Domestic duck muscles. The different quantities of glycogen in skeletal muscle were related to species variation and fiber type.

V. Reference

- 1-Ismoyowati and J Sumarmono, Duck Production for Food Security, IOP Conf. Series: Earth and Environmental Science 372 (2019) 012070.
- 2-Yang Zhang, Q. Bao, Zhi Cao, Chinese Domestic Ducks Evolved from Mallard Duck (*Anas platyrhynchos*) and Spot-Billed Duck (*A. zonorhyncha*), Journal from MDPI Published Mar 24, 2023.
- 3- Krainyk, A., Finger, R. S., Ballard, B. M., Merendino, M. T., Wester, D.B., & Terry, R. H. (2018). Habitat selection by Mottled Duck broods. *Wildfowl*, 68, 104-122 .



4-Cheng, C.; He, X.; Li, H.; Zhang, Y.; Sun, S.; Liu, G.; Li, Y. Study on the antibacterial activity of Litsea essential oil nanoemulsion and its effect on the storage quality of duck meat. *J. Mol. Liq.* 2024, 410, 125610

5-Agnieszka Sujak ORCID Icon, Dariusz Wiącek, Dariusz

Jakubas ORCID Icon, Andrzej Komosa & Ignacy Kitowski, Mallards *Anas platyrhynchos* shot in Eastern Poland: ecological, Human and Ecological Risk Assessment: An International Journal, Volume 25, 2019 Issue 8.

6-Raftovich RV, Chandler SC, Fleming KK. 2018. Migratory bird hunting activity and harvest during the 2016-17 and 2017-18 hunting seasons. Laurel, Maryland: U.S. Fish and Wildlife Service (see Supplemental Material, Reference S7).

7-Baldassarre, G. 2014. Ducks, geese, and swans of North America, Volume 1. Baltimore, Maryland: John Hopkins University Press

8-Johnson K, Carboneras C, Christie DA, Kirwan GM. 2020. Green-winged teal (*Anas crecca*), version 1.0. In Billerman SM, editor. *Birds of the world*. Ithaca, New York: Cornell Lab of Ornithology. Available: <https://doi.org/10.2173/bow.gnwtea.01>

9-Morton, R. W., Sonne, M. W., Farias Zuniga, A., Mohammad, I. Y. Z., Jones, A., McGlory, C., Keir, P. J., Potvin, J. R., & Phillips, S. M. (2019). Muscle fibre activation is unaffected by load and repetition duration when resistance exercise is performed to task failure. *The Journal of Physiology*, 597(17), 4601-4613.

<https://doi.org/10.1113/JP278056>.

10- Brooks, S. V., Guzman, S. D., & Ruiz, L. P. (2023). Skeletal muscle structure, physiology, and function. *Handbook of clinical neurology*, 195, 3–16. <https://doi.org/10.1016/B978-0-323-98818-6.00013-3>.

11-Lana, A., and Zolla, L. (2016). Proteolysis in meat tenderization from the point of view of each single protein: perspective. *Journal of Proteomics*, 147, 85-97

12-Krause, W. J. (2005). *Krause's essential human histology for medical students*. 3rd edition. Universal- Publishers. P.320.

13-Marieb, E. N. and Hoehn, K. (2010). *Human Anatomy and Physiology*. 8th edition. San Francisco: Benjamin Cumming, p.312.

14-Mukund, K., and Subramaniam, S. (2020). Skeletal muscle: A review of molecular structure and function, in health and disease. *Wiley Interdisciplinary Reviews: Systems Biology and Medicine*, 12(1), e1462.

15-Hopkins, D. La. (2014). *Tenderizing mechanisms. Mechanical*. in: Devine, C., Dikeman M. (Eds.), *Encyclopedia of Meat Sciences*, seconded. Academic Press, London: 443-451.



- 16-Lamboleay, C. R., Murphy, R. M., McKenna, M. J., & Lamb, G. D. (2014). Sarcoplasmic reticulum Ca²⁺ uptake and leak properties, and SERCA isoform expression, in type I and type II fibres of human skeletal muscle. *The Journal of physiology*, 592(6), 1381-1395.
- 17-Moran, L. A., H. R. Horton, G. K. Scrimgeour, and M. D. Perry (2012). *Principles of Biochemistry*. 5th ed. Pearson Education Inc., Glenview.
- 18-Braun, E. J., and Sweazea, K. L. (2008). Glucose regulation in birds. *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*. 151(1), 1-9.
- 19-Lu, J. W., McMurtry, J. P., & Coon, C. N. (2007). Developmental changes of plasma insulin, glucagon, insulin-like growth factors, thyroid hormones, and glucose concentrations in chick embryos and hatched chicks. *Poultry science*, 86(4), 673-683.
- 20-Christensen, V. L., Wineland, M. J., Fasenko, G. M., & Donaldson, W. E. (2001). Egg storage effects on plasma glucose and supply and demand tissue glycogen concentrations of broiler embryos. *Poultry Science*, 80(12), 1729-1735.
- 21-Melendez, R., Melendez-Hevia, E., & Canela, E. I. (1999). The fractal structure a clever solution to optimize cell of glycogen: metabolism. *Biophysical journal*, 77(3), 1327-1332.
- 22-Cheng, C.; He, X.; Li, H.; Zhang, Y.; Sun, S.; Liu, G.; Li, Y. Study on the antibacterial activity of Litsea essential oil nanoemulsion and its effect on the storage quality of duck meat. *J. Mol. Liq.* 2024, 410, 125610.
- 23-Pösö, A. R., and Puolanne, E. (2005). Carbohydrate metabolism in meat animals. *Meat science*, 70(3), 423-434.
- 24-Przybylski, W., Monin, G., Kocwin-Podsiadla, M., & Krzecio, E. (2006). Glycogen metabolism in muscle and its effects on meat quality in pigs-a mini review. *Polish journal of food and nutrition sciences*, 15(3), 257.
- 25-Le Bihan-Duval, E., Debut, M., Berri, C. M., Sellier, N., Santé-Lhoutellier, V., Jégo, Y., & Beaumont, C. (2008). Chicken meat quality: genetic variability and relationship with growth and muscle characteristics. *BMC genetics*, 9, 1-6.
- 26-. Rosenvold, K., Petersen, J. S., Lærke, H. N., Jensen, S. K., Therkildsen, M., Karlsson, A. H., & Andersen, H. J. (2001). Muscle glycogen stores and meat quality as affected by strategic finishing feeding of slaughter pigs. *Journal of Animal Science*, 79(2), 382-391.
- 27-Sibut, V., Hennequet-Antier, C., Le Bihan-Duval, E., Marthey, S., Duclos, M. J., & Berri, C. (2011). Identification of differentially



Expressed genes in chickens differing in muscle glycogen content and meat quality. *Bmc Genomics*, 12, 1-13.

28-Immonen, K., Ruusunen, M., Hissa, K., & Puolanne, . (2000).

Bovine muscle glycogen concentration in relation to finishing diet, slaughter and ultimate pH. *Meat Science*, 55(1), 25-31.

29-Muchenje, V., Dzama, K., Chimonyo, M., Strydom, P. E., Hugo, A., & Raats, J. G. (2009). Some biochemical aspects pertaining to beef eating quality and chemistry, 112(2), 279-289.

30-Choi, Y. M., & Kim, B. C. (2009). Muscle fiber characteristics, myofibrillar protein isoforms, and meat quality. *Livestock Science*, 122(2-3), 105-118.

31-Awan, F. N., Shah, A. H., Soomro, A. H., Barahm, G. S., & Tunio, S. G.

(2017). Carcass yield and physico-chemical characteristics of Japanese quail meat. *Pakistan Journal of Agriculture, Agricultural Engineering and Veterinary Sciences*, 33(1), 111-120.

32-Talpur, M. Z., Abdulwahid, A. M., Yan, S., Tao, L., Ahmed, I., Liu, L., & Jia, J. (2018). Chicken meat quality; association with different gene expression, physiochemical properties and muscle glycogen. *Pakistan journal of agricultural sciences*, 55(4).

33-Chauhan, S. S., LeMaster, M. N., Clark, D. L., Foster, M. K., Miller, C. E., & England, E. M. (2019). Glycolysis and pH decline terminate prematurely in oxidative muscles despite the presence of excess glycogen. *Meat and Muscle Biology*, 3(1).

