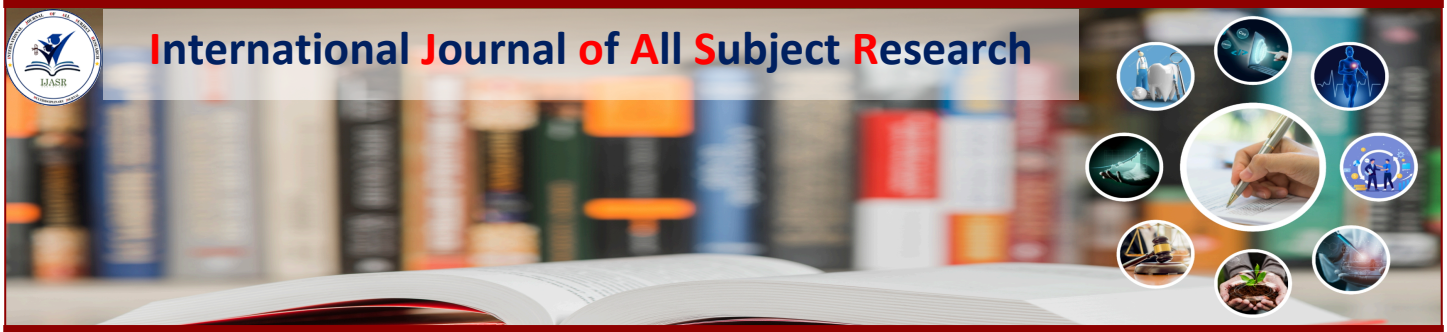
Impact Factor (SJIF): 5.158 **E-ISSN: 2583-7206**

International Journal of All Subject Research



Received: 08/June/2024 **IJASR:** 2024; 3(4):01-05 **Accepted:** 13/July/2024

Effects of *Terminalia chebula* (*Haritaki*) on the Sex Ratio of the *Drosophila melanogaster*

[1]Brinda R and [*2]V Shakuntala

[1, *2]Department of Zoology, University of Mysore, Manasagangotri, Mysuru, Karnataka, India.

Abstract

Terminalia chebula is a herbal product which is widely used for various medicinal purposes and *Drosophila melanogaster* is a model organism by using which we can able to do our various studies. In our studies we analysed the effect of *Terminalia chebula* on sex ratio in Drosophila we treated the flies in wheat cream agar media, 250 mg concentration of *Terminalia chebula* and 300mg concentration of *Terminalia chebula*. According to our studies we found that female offspring were more than male offspring in all the three medias but we found that the number of female offspring was more in 300 mg *Terminalia chebula* media compared to wheat cream agar media and 250 mg *Terminalia chebula* media . This we can state that *Terminalia chebula* alters the sex ratio in *Drosophila melanogaster*.

Keywords: *Drosophila melanogaster, Terminalia chebula,* sex ratio

Introduction

Any plant that has chemical in one or more of its organs that have medical value or that serve as building blocks for the creation of effective medications is considered medicinal .With the help of this description, it will be easy to discern between plants that are considered medicinal but not have not yet undergone extensive scientific investigation, and plants whose therapeutic qualities and ingredients have been shown by science. Since the beginning of time, medicinal plants have been utilized for medical purposes. Through the "hit and trial" method, the use of plants to create medicine has been realized, and man has increasingly been able to meet his demand from his environment. The Japanese were the first to use medicinal plants for rituals, food flavouring, and diseases treatment. Global research has been done to confirm their promise, and some of the results have let to the development of what are known as plant-based medications. These plants have always been extremely essential in almost all cultures since they are valuable source of medicine. In addition to being valuable source for many contemporary medications, medicinal plants are also thought to be abundant sources of traditional medicines. Many of the herbs and species that people use to season their meals also contain beneficial therapeutic ingredients.

In India, medicinal plants have been used for five thousand years as part of Ayurvedic medical system. Customers perceive herbal items as being more "natural" than synthetic," and they also consider that they are more likely to be safe than synthetic pharmaceuticals. These attitudes are shared by customers across the globe. They started avoiding touch with traditional "western" medicine and began to think about leading a healthy life style. According to reports, herbal medications and the ingredients in them can effectively heal human illness and have positive long-term fitness consequences (Luqman et al., 2014).

Phytochemicals presents in medicinal plants: plants materials often have positive medical benefits because of the combination of secondary compounds they contain. Consumer interest in phytonutrients, also known as phytoceuticals, which are naturally occurring compounds derived from plants and believed to have curative, preventative, or nutritive value, has increased due to growing concerns about aging, chronic disease prevention, and general health (Guhr and LA chance, 1997).

Medicinal plants are thought to provide a special and renewable resource for the discovery of possible novel medications and biological entities due to the structural and biological diversity of its elements (Lahlou, 2007). It is therefore reasonable to assume the plants based metabolites will continue to play a crucial role in the hunt for novel, affordable, and safe medications. Pharmaceutically active compounds are primarily found in plants for commercial importance, such as the Apiaceae, Apocynaceae, Araliaceae, Araceae, Asphodelaceae, Asteraceae, Ginkgoaceae, Hypericaceae, Lamiaceae, Papaveraceae, Piperaceae, Rhamnaceae, Rubiaceae, Rutaceae, and Zingiberaceae (Gurib-Fakim, 2006). Numerous human ailments, such as diabetes, cancer, hepato-renal diseases, heart disease, and neurological disorders, have been linked to decreased risk of using phytochemicals.

***Corresponding Author:** V Shakuntala

Medicinal Plants Importance

These plants synthetic chemicals can help protect both human and animal health. These include compounds that are aromatic, like phenols or their derivatives that have had their oxygen replaced, or tannins. The capacity of plants to synthesise aromatic compounds is nearly infinite. These compounds are primarily secondary metabolites, of which at least 12000 have been isolated and are thought to make up less than 10% of the total. These substances often function as the plants defence chemicals against herbivores, insects, and microbes. Many of the herbs and spices that people use to season their meals also contain beneficial therapeutic ingredients. For a variety of reasons, the demand for medicinal plants is currently rising in both developed and developing nations.

For some, however, the availability and reasonable prices of natural products would be the deciding factor, for others, it would be the increasing awareness that these items have few, if any, negative consequences. Distillation can be used to turn medicinal and fragrant plants into essential oils, but market demand is for the plants components and cut flowers. Among other industries, they are utilized in the food, cosmetics, medicine, and perfume industries. Many significant plant species that were previously common have become rare in locations where there is a greater demand for the limited resources.

Since ancient times, medicinal plants, also known as medicinal herbs, have been utilized in traditional medical practices. These uses have been known for decades. Plants produce hundreds of different chemical and biological chemicals for a variety of purposes, such as defence against pathogens, insects, fungus, and herbivorous mammals. Plants include a wide range of phytochemicals that have been found to have biological activity, either known or unknown. The effectiveness of using an entire plant as medication is uncertain, though, because a single plant contains a vast variety of phytochemicals. Furthermore, in order to definitively establish safety and efficacy, rigorous scientific study is still lacking in the assessment of the pharmacological effects and phytochemicals contains, if any, of various plants with therapeutic potentials. (Ahn 2017).

The use of chemical analyses in the nineteenth century significantly changed the place and functions of plants in medicine. A series of medicinal plants were used to extract and isolate alkaloids, starting with morphine from the poppy and moving quickly on to strychnos and ipecacuanha, quinine from the cinchona tree, and a few more. More classes of pharmacologically active chemicals and substances were found in medicinal plants as chemistry progressed (Atanasov et al., 2015).

The fruit *Drosophila melanogaster* is a versatile model organism that has been used in biomedical research for over a century to study a broad range of phenomena. There are many technical advantages of using Drosophila over vertebrate models; they are easy and inexpensive to culture in laboratory conditions, have a much shorter life cycle, they produce large numbers of externally laid embryos and they can be genetically modified in numerous ways. Research using Drosophila has made key advances in our understanding of regenerative biology and will no doubt contribute to the future of regenerative medicine in many different ways.

Many obvious practical and ethical obstacles severely limit the scope for experiments using humans in biomedical science, thus much of what we know about the underlying biology of cells and tissues comes from studies using model organisms such as mice, and the focus of this article, the fruit fly *Drosophila melanogaster*. Drosophila has been used productively as a model organism for over a century to study a diverse range of biological processes including genetics and inheritance, embryonic development, learning, behavior, and aging. Although humans and fruit flies may not look very similar, it has become well established that most of the fundamental biological mechanisms and pathways that control development and survival are conserved across evolution between these species.

Over the past four decades, Drosophila has become a predominant model used to understand how genes direct the development of an embryo from a single cell to a mature multicellular organism. In 1995, Christiane Nusslein-Volhard, Eric Wieschaus, and Ed Lewis won the Nobel Prize in physiology or Medicine “for their discoveries concerning the genetic control of early embryonic development”. Many of the genes that they defined as being important for fly development have since been shown to be critical for all animal development, including humans. Although the final architecture of a fly and a human differs greatly, many of the underlying building blocks and engineering processes have been conserved through evolution and are strikingly similar.

In 1999, Craig Venter and colleagues used the Drosophila genome as a test bed to prove the practicality of the “shot-gun” approach for sequencing the human genome. This approach worked well and the first release of the sequence of the Drosophila genome was released in March 2000, just 11 months ahead of the human genome. The sequence and annotation of the Drosophila genome is freely available to all and can be accessed via “Fly base”, the outstanding online database dedicated to Drosophila. Comparisons between the fully sequenced Drosophila and human genomes revealed that approximately 75% of known human disease genes have a recognizable match in the genome of fruit flies consolidating its legitimacy as a model organism for medical research. Currently, it is estimated that there are around 140000 genes in Drosophila and each of these has a dedicated page on Fly base that contains links to practically everything known about that gene including sequence, gene product sequence, known mutations, and related literature.

Owing to the successful results in the use of Drosophila in experiment studies, it has met the standard of the European Centre for the validation of Alternative Methods (ECVAM): Reduction, Refinement and Replacement (3Rs) of laboratory animal usage (Festing et al., 1999). D. melanogaster as a model raises few ethical concerns and its genome can be easily manipulated in the study of a particular gene of interest under a defined condition. Interestingly, the post-genomic sequencing of *D. melanogaster* generated a great deal of attention to biomedical researches because it revealed functional conservation of the majority of the genes present in mammals. As a result of this, it has been used to obtain mechanistic insights into human diseases. Thus, its use in toxicological studies will continue to generate valuable data.

D. melanogaster shows anatomical characteristic features such as compound eyes and wings. It has a lifecycle of between 40-120 days depending on environmental stress conditions (e.g. population density and temperature) and diet. A diet such as commeal extends the life expectancy of the fly, while diets with high quantities of cholesterol and saccharides (free available carbohydrates) can reduce lifespan (Hirth, 2010). Additionally, overpopulation has been shown to reduce the lifespan of the insect (Joshi and Mueller, 1997). The resemblances of molecular processes involved in the control

of lifespan and aging between *Drosophila melanogaster* and human, as well as a good degree of genetic homology between this species, makes the fruit fly a fascinating model system for human's diseases of particular significance, significance, over 65-70% of human disease genes can be found in Drosophila (Reiter et al., 2001; Pandey and Nichols, 2011; Poddighe et al., 2013, which makes it an important model to understand not only how the genes induce diseases, but also the finding of the relation of such genes to diseases (Fortini et al., 2000; Fortini and Bonini, 2000). When compared with other models, the fly undergoes rapid generation time, easy to use, and easy to conserve in the laboratory in large quantity due to its short lifecycle and tiny body size.

Fruit fly has a rapid life cycle of single fertile mating pair which can produce hundreds of genetically alike offspring's within 10 to 12 days at 25 degree Celsius. Another fascinating feature of Drosophila is that it can be used as a multiple-model organisms. Hence, its adult, pupa, larvae, and embryo can be used model in different human diseases settings. As such, the embryo and pupa can be used as models in developmental human diseases studies, whereas the larvae can be used to study behavioral and physiological processes. Fascinatingly, the adult of *Drosophila melanogaster* processes, complex and sophisticated systems. It has some structures that can mimic the corresponding functions of the mammalian lung, heart, gut, kidney and reproductive tract.

Additionally, the fruit flies brain possesses more than 100, 000 neurons that are vital in circadian rhythms, feeding, memory, courtship, aggression, and flight navigation. Essentially *Drosophila melanogaster* responds to numerous central nervous system drugs in a similar way to mammals (Nichols et al., 2002; Rothenfluh and Heberlein, 2002; Satta et al., 2003; Wolf and Heberlein, 2003; Andretic et al., 2008). This review aimed at concisely showing *Drosophila melanogaster* as a potential model for human diseases.

Benefits and Chemical Composition of *Terminalia chebula*

In *Terminalia chebula*, 33% of the total phytoconstituents are hydrolysable tannis (which may vary from 20-50%) and are responsible for pharmacological activity. These tannins contain phenolic carboxylic acid like Gallic acid, ellagic acid, chebulic acid and Gallo tannins such as 1, 6 di-O-galloyl-Beta-D-glucose, 3,4,6 tri-O-galloyl-Beta D-glucose, 2,3,4,6 tetra-O-galloy-Beta-D-glucose, 1,2,3,4,6 penta-Ogalloyl-Beta-D-glucose. Ellagitannin such as punicalagin, casuarinin, corilagin and terchebulin and others such as chebulanin, neo chebulinic acid, and chebulagic acid reported in literature. The tannin content varies with the geological variation. Flavonol glycosides, triterpenoids, coumarin conjugated with Gallic acid called chebulin, as well as phenolic compounds were also isolated.

Various methods have been reported for extraction of phytoconstituents from *Terminalia chebula* for studying their pharmacological activities, total eight compounds viz. Gallic acid, methyl gallate, ethyl gallate, chebulagic acid, tetra-O-galloy-Beta-D-glucose, ellagic acid, chebulinic acid and penta-O-galloy-Beta-D-glucose from *Terminalia chebula* were isolated on reverse phase chromatography. There are seven varieties of *Terminalia chebula* all of which are more or less used in similar fashion but vary in specific usages and qualities.

Benefits

Charaka Samhita and Sushrusha Samhita, though, extensively describe various medicinal plants, T. chebula (haritaki) enjoys the prime place among medicinal plants not only in India but also in other countries like Asia and Africa. It is extensively used in Ayurveda, Siddha, Unani and homeopathic medicines in India. It is a top listed plant in Ayurvedic Materia medica for treatment of asthma bleeding piles, sore throat, vomiting and gout. It is used in Thai traditional medicine as a carminative, astringent and expectorant (Malik et al., 2012). According to Vagbhata, it is the drug of choice in the therapy of 'vata-kapha' diseases. The 'Triphala' a herbal preparation of 'three fruits' from plants *Terminalia chebula*, *Terminalia bellerica, Emblica officinalis*, is used as laxative in chronic constipation, detoxifying agent of the colon, food digestive problems (poor digestion and assimilation) and rejuvenator of the body. Certain studies have shown that 'Triphala stimulates appetite, and is useful in treating cancer and detoxification. Triphala is considered as the most versatile of all herbal formulations and is prescribed as a cardio tonic and for candid infection (Kadian, 2016)

The fruits of haritaki are used both externally as well as internally for medicinal purposes. Externally, the paste of fruits effectively reduces the swelling, hastens the healing and cleanses the wounds and ulcers. In erysipelas and other skin hemanta ritu (November-December) with sunthi, in sisira ritu (January-February) with pippali, in vasanta ritu (March-April) with honey and in grisma ritu (May-June) with jiggery. According to Vagbhata, when haritaki powder fried in ghee is regularly consumed with sufficient ghee in food, it promotes longevity and boosts energy. Common gastrointestinal ailments, tumours, ascites, piles, enlargement of liver and spleen, worms, colitis can be treated well with haritaki. The bark of haritaki, if eaten after chewing, improves digestion. 'Bala haritaki' is useful in haemorrhoids and in clearing the bowels. The mixture of Triphala powder and haridra is a well known adjunct in diabetes. Bronchospasm is mitigated effectively with the combination of haritaki and bibhitaka powders with honey. In abdominal pain due to flatulence, it is given with jaggery and ghee. The most popular combination of haritaki, musta, sunthi and jaggery is an effective panacea for diarrhoea, dysentery, and flatulence etc. 'Haritaki siddha ghrta' is beneficial in chronic fever. The decoction of haritaki or Triphala is given along with honey in hepatitis. Haritaki powder with honey and ghee is also effective remedy for anemia. In obesity, its decoction with honey reduces the excessive body fats. Regular use of haritaki improves memory due to beneficial effects on the nerves of brain. It is also valuable in dysuria and urinary stones. Its paste with water is found to be anti-inflammatory analgesic and having purifying and healing capacity for wounds. They are used as astringent in hemorrhoids. It has a ability to increase the appetite as digestive aid. It is used as supplement to cholesterol normalizing drugs (Usha, 2007).

Work done to understand its medicinal value in different model.

Evaluated on excision and incision model, in albino rats, in the form of an ointment with two concentration. Both concentration of ethanolic extract showed significant response in wound healing.

Studied on 30 rabbits in wound model. Herbal paste application showed significant improvement on maturation, wound concentration and epithelialization on rabbits the wound healing experiments suggests that tannin extracts from dried immature fruits of *Terminalia chebula* fructus retz. Can promote cutaneous wound healing in rats, probably resulting

from powerful anti-bacterial and angiogenic activity of the extracts.

All the tested extracts of *T. Chebula* were highly effective against two of the tested dental caries causing bacteria. They suggests as an alternative antimicrobial agent dental caries causing microorganisms.

Animal sex ratio are known to be impacted by a variety of causes, including environmental and genetic influences. Factors affecting the primary sex ratio or those acting after fertilization and affecting the secondary sex ratio can be the source of deviations from the theoretical 1:1ratio of men the females. One of the key areas of evolutionary biology has been the study of insect sex ratio (Leigh et al .,1985) .When it comes to insects that have distinct semesters, the sex ratio-the percentage of females-indicates the populations capacity for reproduction. Because sex chromosomes segregate during gametogenesis, the sex ratio in randomly mated population typically fluctuates between 1:1(Rawlings and Maudlin, 1984; Werren and Godfray, 1995; Hoy, 2004) and this generally indicates stabilizing selection on males and females (schowalterl, 1996).

In numerous creatures where the sexes are distinct, females and men are generated in roughly equal quantities (Hardy, 2002).Therefore, in many organisms, such as insects, the sex ratio is 1:1 prakahs, (2008). This ratio optimizes genetic heterogeneity by optimizing the availability of males to females Schowalter, (2016). However, a number of environmental elements, whether they be chemical, biological, or physical, can change the sex ratio. Schowalter (2016) and Hardy (2002) .In the several study, the effect of some physical factors on sex ratio in *D. melanogaster* were studied.

In nature , there are many animals that create a biased sex ratio among its offspring due to many different reasons and thus male or female-biased sex ratio if found in the wild this affects the life history traits of males and females. Organism change their mating behavior to survive in adverse conditions when a biased sex ratio raises this affective their efficiency of gender roles in both males and females which they modify in order to achieve fitness in adverse conditions.

Materials and Methodologies

Drosophila melanogaster Stock Culture

Experimental Oregon k strain of *Drosophila melanogaster* which is utilized in the experiment was extracted from, Drosophila stock centre, department of studies in zoology, University of Mysore. The stock was cultured in the glass bottles consisting of yeast agar media was prepared by adding 100g of jaggery, 100g of Sooji and 10g of agar into 1000ml of distilled water and boiled, then 7.5ml propionic acid was added. After it got dried pinch of yeast was added. Flies were transferred to the media containing cultured bottle and maintain at optimum laboratory conditions such as 70% humidity, 12:12 dark light cycle and 22±2 degree Celsius.

Diet preparation: 3 different kinds of Medias were prepared. They are as follows,

i). **Control:** The yeast agar media was prepared as previously stated.

ii). **Treatment 1:** 250 mg of powder of *Terminalia chebula* extract was mixed in 100 ml of yeast agar media.

iii). **Treatment 2:** 300 mg of powder of *Terminalia chebula* extract was mixed in 100 ml of yeast agar media.

Establishment of Experimental Stock

10 pair of male and female flies were isolated form the stock and transferred to each bottles containing three different media and are tagged as control, treatment 1 and treatment 2 respectively. Such that 10 replicates of each group were established. They are maintained in the laboratory conditions and were allowed to mate and lay egg. For every 2 days the flies were transferred to new culture bottles of respective media to avoid overcrowding of flies. After a week of transfer, overall, 40 bottles of flies from each group were obtained. These flies were used for the further experimentation.

Experimental Procedure

Before carrying out the sex ratio assay. The flies were allowed to feed on control, 250mg and 300mg chebula containing media for 5days. 5 males and 5 females from control, test 1, and test 2 groups were transferred to new bottles. 2 replicates were maintained and named as replicate numbers. These flies were maintained at 25 degree Celsius under 12 hours light and 12 hours dark condition. And male and female offspring's emerged from each diet and experiments continued for 2 weeks. The data obtained from the assessment by using chi square analysis.

Result and Analysis

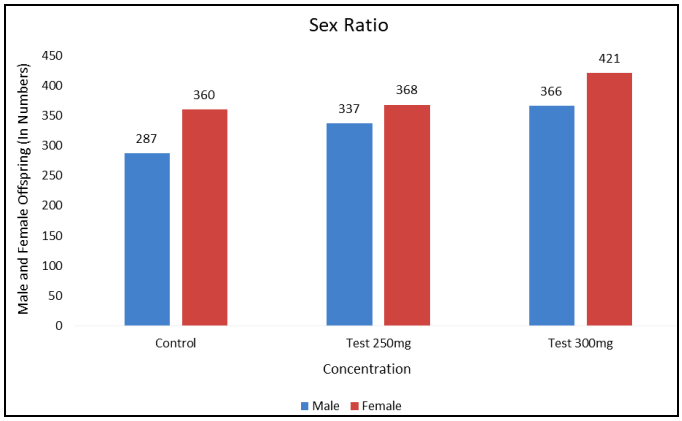


Fig 1: The graph showed the comparison between the Male and Female adult filial numbers in D. *Melanogaster* raised on the different concentration. (Yeast agar media, *Terminalia chebula)*

The figure 1 suggests that the female offspring's are produced more than the male offspring in all the 3 concentration. When the data subjected to the Chi square value analysis showed there is significant value in concentration 250mg and 300mg, insignificant in control.

Table 1: The effect of yeast agar media, *Terminalia chebula* Diet on the male offspring's sex ratio of *Drosophila melanogaster.*

Treatment	Number of Adults	Number of Females	Number of Males	F:M Ratio
Control	647	360	287	1:0.797
Test 250mg	705	368	337	1:0.915
Test 300mg	787	421	366	1:0.869

Table 1: The effect of yeast agar media, *Terminalia chebula* on the female offspring's sex ratio of *Drosophila melanogaster.*

Treatment	Number of Adults	Number of Females	Number of Males	M:F Ratio
Control	647	360	287	1:1.254
Test 250mg	705	368	337	1:1.091
Test 300mg	787	421	366	1:1.150

Table 1: The effect of yeast agar media, *Terminalia chebula* on the Female and Male offspring's Sex ratio of *Drosophila melanogaster* and Chi square analysis value.

Treatment	F:M Ratio	Chi Square Value	Significant Value
Control	1.254:0.797	4.118	P >0.05
Test 250mg	1.091:0.915	0.681	P <0.05
Test 300mg	1.150:0.869	1.921	P <0.05

Discussion

There are several studies reveals that the physical factors like temperature, photoperiod effects on the sex ratio in insects including *Drosophila melanogaster* but as per our Knowledge of information our study is the 1st report on the effects of *Terminalia chebula* diet on the sex ratio in *Drosophila melanogaster.*

Terminalia chebula is of great use in Ayurveda as it as many medicinal properties. The aim of the present study was to study the reproductive assay on *Terminalia chebula* using *Drosophila melanogaster* as a model organism. In the present experiment *Drosophila* flies were allowed to feed on the media containing different concentration such as 250mg/100ml and 300mg/100ml water of *Terminalia chebula* 5 males and 5 females from control, test 1, and test 2 groups were transferred to new bottles. 2 replicates were maintained and the male and female offspring's which were emerging from the bottles were counted for 2 weeks and the data was collected.

In the present study, the (Fig-1) Results revealed that the female's offspring's were produced more than the male off springs in all the 3 Concentration. This is because the quality and quantity of the diet is influence on the variation in the sex of the offspring's. Several studies have been demonstrated that diet quantity and quality influences maternal reproductive output and sex of the organism. According to the (Yazgan 1972), the increase in amino acids in the diet increases the number of female individuals of *Pimple thirunallar* (endoparasitoid). Parent flies prefer the production of a particular sex in their progeny that increases the possibilities of survival for that species, by assisting them in reproduction or decreasing the competition for mates, habitats, resources. Besides these factors, sometimes the sex of the offspring also depend upon the health of the mother (Trivers and Willard, 1973).

According to the above study, Table 1A and Table 1B Shows that the male and female sex ratio should be 1:1 but our study showed that female are more than male.

Table 1C shows that in control P>0.05 which is insignificant, in 250mg and 300mg P<0.05 which is significant according to Chi square value analysis.

There are several research showed that the variation in the environmental factors such as the temperature, light, variation in the age also alter the sex ratio of in the several insects including the *Drosophila melanogaster*. However, in our experiments we use the same aged flies raised on the different diets which maintained under same environmental condition were used to study the sex ratio hence the observed variation in experimental results is due to the variation in the quality and quantity of the diet.

According to my studies from the above data, i.e., Females are more because of the below reasons,

i). **Hormonal Effects:** Phytoestrogens, *Terminalia chebula* contains phytoestrogens, which might influence hormonal balances in developing larvae. So this lead to the Sex ratio towards Females.

ii). **Differential Viability:** The compounds in *T. chebula* may enhance the survival of female larvae over male larvae. This could be due to females having a better physiological response to the active components of *T. chebula.*

iii). **Gene Regulation:** *T. chebula* might affect the expression of genes related to sex determination and differentiation, possibly favouring the development of female traits.

iv). **Oxidative Stress Reduction:** If *T. chebula* reduces oxidative stress through its antioxidant properties, it might improve the overall health and survival of female larvae more effectively than males.

v). **Developmental Timing:** Sex-Specific Developmental Rates, The compounds in *T. chebula* could influence the developmental rates of male and female larvae differently, potentially leading to a higher number of females reaching maturity under the conditions provided.

References

1. Abolaji AO, Kamdem JP, Farombi EO, Rocha J.B. *Drosophila melanogaster* as a Promising Model Organism in Toxicological Studies. Arch. Bas. App. Med. 2013; 1:33-38.
2. Aritakula A, Ramasamy A. Drosophila-based in vivo assay for the validation of inhibitors of the epidermal growth factor receptor/Ras pathway. *J Biosci*. 2008; 33:731-742.
3. Barbara H. Jennings, Drosophila a versatile model in biology and Medicine transcriptional regulation group, UCL Cancer Institute Paul o' Gorman Building 72 Huntley st, London WC1E6BT, United Kingdom.
4. Braun T, Wool lard A. RUNX factors in development: lessons from invertebrate model Systems. Blood Cells Mol Dis. 2009; 43:43-48.
5. Choma MA, Izatt SD, Wessel's RJ, Bodmer R, Izatt JA. Images in cardiovascular medicine: in vivo imaging of the adult *Drosophila melanogaster* heart with real-time optical coherence tomography. Circulation. 2006; 114:e35-e36.
6. DiAngelo JR, Birnbaum MJ. Regulation of fat cell mass by insulin in *Drosophila melanogaster*. Mol Cell Biol. 2009; 29:6341-6352.
7. Fatemeh Jashidi-Kia, Zahra Lorigooini, Hossein-Amini- Medicinal plants: Past history and future perspective Journal of Herbmed Pharmacology. 2018; 7(1):1-7.
8. Kiran K and Krishna MS, The effects of Jeeni millets traditional mix on the sex ratio of *Drosophila melanogaster* 2023; 2(9):93-96.
9. Kohler RE. Lords of the fly: Drosophila genetics and the experimental life. University of Chicago Press, Chicago, 1994.
10. Lin dsley DL. & Zimm GG. The genome of *Drosophila melanogaster*. Academic Press, San Diego, 1992.
11. Puneeta singh and Hitesh Malhotra, *Terminalia chebula*: A Review Pharmacognistic and Phytochemical studies, *International Journal of recent scientific research* Vol 8, 2017 Issue, 11, PP.
12. Rid wan olamilekan adesola, Jadalhaq Taiwo Lawal and Oluwatobi Emmanuel oladele, *Drosophila melanogaster* a potential model for human diseases, *World News of Natural Sciences an International Scientific Journal*, 2021, 42-59.
13. Rathinamoorthy R and Thilagavathi G, *Terminalia chebula*-Review on Pharmacological and Biomedical studies. *International Journal of Pharmtech Research*, 6(1), 97-116.