

Review Article

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The Silkworm, *Bombyx mori*: A Promising Model Organism to Study the Longevity - A Review

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ABSTRACT

The silkworm *Bombyx mori* being a typical representative of lepidopteron insects is of great economic importance in terms of longevity having short generation time, wherein physiological, genetical, molecular and biological aspects are playing an important role. But, the mechanism of longevity has been gradually unraveled due to not realizing in-depth in basic theoretical and practical fundamentals for the several above said aspects of insects in particularly silkworm *Bombyx mori*. In spite of the fact, application of several biotechnological methods have been implementing in the era of advanced biology. Silkworm has to be a model organism to study the longevity, because it is an economically important insect for the silk production, wherein longevity play an important role in metabolic activities during its life cycle and understanding of its durability to reduce still shorter is the need of hour for the benefit of rearer, breeders, physiologist, biochemist, geneticist, farmers, etc in economic & scientific point of view. Regretfully, the application of theoretical and practical based theories are not possible until unless the efforts of converting into the action through proper research investigations and during various movements/programmes to reach the knowledge of longevity in a systematic way for the users. In the light of the above concept, herein this review, summarized the several important aspects are directly or indirectly influencing/affecting on longevity with providing authentically supporting evidences to understand the impact of age-related disorders for the standardization of life span in the insects with special reference to the *Bombyx mori*. The information gathered and discussed herein is a first ever effort as a review and greatly be helps to further research investigations in the field of gerontology with reference to silkworm *Bombyx mori*.

Keywords

Bombyx mori,
Longevity (aging),
Intrinsic and
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Introduction

The word insect originated from Latin, it means “cut into sections” and insects are belong class invertebrates within the arthropod

phylum, those all are having chitinous exoskeleton, dividing its body into three parts namely, head, thorax and abdomen. However, insects are most different classes among the animals on the earth and which have been

identified as more than one million species and representing greater than half of all known living organisms (Chapman, 2006 and Wilson, 2009). It is estimated fact that, potentially greater than 90% of the differing animals living forms on earth (Erwin and Terry, 1982), except few numbers of species reside in ocean but remaining all species found in nearby all environments. Further, life cycles of the insects vary depending upon the species but most of the insects' hatches from eggs and undergo series of moulting, pupal, adult, metamorphosis stages, *etc.*, (Chapman, 2006).

However, the insect silkworm *Bombyx mori* have being a representative group of Lepidoptera insects is of great economic importance in the silkworm industry. Silkworm is a model insect in lepidopteran group for its clear genetic background and having short generation time and it has highly beneficial organism in the life science (Meng *et al.*, 2017). The life cycle of the silkworms depending upon the voltinism (Uni-Bi-Multivoltine races) and it also undergo four moults and five instars. Generally, Culturing of silkworm itself is known as "silk culture or sericulture" which has been practicing since around 5000 years in china (Goldsmith *et al.*, 2004). It is believed fact that, the silk culturing or producing of silk kept many year secret in china for several years. But even though presently china (146,000 MT), India (28,708 MT) and Uzbekistan (1,100 MT) are the three countries have occupied first, second and third positions respectively in the raw silk production and 60% of world silk producing by two countries alone (China & India).

Life is a journey in the journey could not realize the durability in sometimes and other word can be says "longevity of life". It can be defined as some times meant to refer only to especially long live members of a population and also used this term as an aging or "life expectancy" in demography. Aging is a complex process that involves the number of

deleterious changes resulting in overall decline in several important physiological functions *e.g.* Energy imbalance, consumption of high caloric food, causing of several diseases, unbalancing genetic molecular mechanisms and their associated metabolic disorders in totally overall leads to lower physiological activities of the particular organism and eventually causing several diseases and leading to death at ultimate due to misbalancing immune system, which is said to be most important host-defence mechanism in human and also highly conserved in insects particularly in silkworm *Bombyx mori*. Hence, keeping above concept in the mind, a review survey has made to understand the importance of aging mechanism factors for causing longevity, intrinsic, extrinsic and some biochemical factors influences aging in insects and particularly in silkworm *Bombyx mori*.

Origin of ageing and background aspects

Prior to the several decades ago, many groups of insects have evolved with different percentage-wise including lepidopteron silkworm group (Figure 1) and numerous gerontologists have researched on the aging phenomenon in insects too. During 20th century "proximate" (mechanistic) and "ultimate" (evolutionary) are the two theories of aging, which have positively linked among each other, because to understand their interaction and shared questions, which generated the better information comprehension of aging and evolution of life spans in multicellular organisms (Cohn *et al.*, 2010 and McGraw *et al.*, 2010).

But, generally, the definition of aging so far yet to be have solid authentication and does not exist universally in the world. However, an American gerontologist has given basic characteristic phenomenon by means of postulating four associated phenomenon (Fig. 2). Further, Jose Vana *et al.* (2007) have

clearly revealed through presenting different consequences influencing on aging (Fig. 3) and there are more than 300 theories, which are associated with aging and still number is increasing (Mevdevev, 1990). But, Hamilton, (1966) and Charles worth, (1980) have reported that, aging is a biological phenomenon and based on which mainly three important theoretic theories gives in exactly what and how theories of aging originated. These theoretic theories are based on fact of extrinsic mortality which leads to decrease of natural selection with aging.

Mutation accumulation theory

This is the theory which gives some biological clues to think over aging phenomenon in insects. The scientist Medawar, (1952) who has predicted through observing slight effect in genome of an organism, which leads to change in physiological organs associated with the aging.

Antagonistic pleiotropy theory

Several years later, the second theory further associated with first theory, through with slight information on aging, which has investigated by Williams, (1957) who suggested that, it is an active mechanism takes place in insects where how mutation causes aging and introduces a theory called antagonistic pleiotropy theory.

Disposable some theory of aging

Few decade later, some researchers *i.e.*, Kirk wood, (1977), Kirk wood and Rose, (1991) have postulated in their finding that, early reproduction trade off against late survival, but through which optimal resource allocation strategic rather than pleiotropic genes plays an important role in aging.

Apart from above theories, some important contribution have also made by several research investigators during early 50th and

60th decades onwards on the evolution of aging in insects are Medawar, (1952), Williams, (1957), Strehler, (1960 & 1962) and Charles worth and Stephen, (2001). The evolutionary phenomenon of aging gives varies twisted prediction of which highlights the extrinsic mortality should be negatively related with the intrinsic life span (*i.e.*, life unaffected by environmental factors). Moreover, most later on 60th and 70th century onwards several well-known researchers have worked on aging related aspects by utilizing different insects including mulberry silkworm *Bombyx mori* are Gerald *et al.*, (2002) in Bat, Carey and Judge, (2001) in mammals, birds, reptiles, fish, Gavrilob and Gavrilova, (1991) in drosophila melanogaster, mice and dogs, Brooks *et al.*, (1994) in nematodes, Johnson, (1987 & 1990) in mosquitoes, Gavrilov, (1980) in human lice (*Pediculum humanus*) and utilizing mulberry silkworm *Bombyx mori*, Murakami, (1989a and 1989b), Osanai, (2003), Murthy, (2008), Doddaswamy and Subramanya, (2007), Anantha and Subramanya, (2012) and Ramya and Jagadeesh kumar, (2017).

Phenomena of longevity in Silkworm, *Bombyx mori*

The silkworm *Bombyx mori* being monophagous insects has been utilizing one of the popular genetical model organisms to investigate various aspects of biological, genetical, molecular, physiological, biochemical, nutritional and climatic aspects, gerontology *etc.* Further, several group of insects have different lifespan including lepidopteron silkworm *Bombyx mori* (Table 1) and having egg, larva, pupa and adult stages in its whole developmental stages of its life cycle (Fig. 4). There are numerous important steps for selecting silkworm insect has a model organism to study the aging phenomenon are 1) To observe the relatively shorter lifespan phenomenon (lesser than 25 days in case of larvae) depending upon races/strains (Nirmal

kumar and Sreerama Reddy, 1994), 2) To observe the availability of stocks containing altered genes, 3) For maintenance of the silkworms in germplasm rearing stations with easy handling for the rearers (Krishnaswami, 1978), 4) The phenomenon of climatic and genetic manipulations that varies lifespan of silkworms, 5) Influence of molecular genetical mechanism, 6) To observe the phenomenon of sequencing of the silkworm genome, 7) To observe the phenomenon of dissecting complex biological parts such as head, thorax, abdomen, nerve and circulatory systems, silk gland and sex identification *etc*, 8) To observe the silkworm behaviour (moulting, under moulting feeding and diseases affected worms phenomenon, *etc*), 9) To observe in detailed information in regards to pupal and adult longevity (Ramya and Jagadeesh kumar, 2017), 10) In addition, the other aspects of biology of physiological and biochemical of silkworm insect are of particular advantage for aging work including its life history, which has divided into morphological features, duration of growth/development (egg, larvae, pupa and adult, *etc*) can be readily distinguished. In several organisms visually distinguishing of mature aging adults from immature or juvenile phases is not simple. In *drosophila* for example development is restricted only to the embryo, larva, pupa and adult life. However, in silkworm *Bombyx mori* the adult fly is sexually mature and hence, considered as a mature aging adults. Post mitotic fully differentiated cells except for cell gonads and few cells in gut (Bozuck, 1972 and Ito and Hotta, 1997). Other organism such as human being carried many organ system *i.e* gastrointestinal, dermatological and hematopoietic systems are constituted by continuous cell replacement. Further, several other organs, brain and heart have rarely excess of new neurons or cardiac cells (Stephen *et al.*, 2003). Longevity may be said different meaning, in different situation, few reports suggesting that by focusing

primarily on above said phenomenon, then involved in trying to understand the physiological, biochemical, biological, genetical and molecular level and changes that take place in cells and different organs over the time and situation. The insects such as silkworm *Bombyx mori*, which is almost similar to the *drosophila melanogaster* for some aspects as said above being utilised as an excellent model system of aging studies in the different well equipped laboratories and few reports were made by Murakami, (1989a and 1989b), Osanai, (2003), Murthy, (2008), Doddaswamy and Subramanya, (2007), Anantha and Subramanya, (2012) and Ramya and Jagadeesh kumar, (2017) utilizing silkworm as a model organism for the study of ageing related aspects.

Intrinsic factors effects on aging

Intrinsic aging is a term used to describe cutaneous aging of the skin and other parts of integumentary system. In other words intrinsic aging is a phenomenon where influencing of internal physiological factors and this intrinsic aging is popularly known as chronologic aging. Because where occurrence of inherent degenerative process from the process internal physiological factors decreases, such factors are both qualitative and quantitative in nature.

Physiological and biological factors influence on aging

Aging is a process, which involving under influences both intrinsic and extrinsic factors. The intrinsic factors are influences some extends greater than extrinsic factors but both plays coordinated with in an organism. Recently, gerontologist and physiologist are researching on aging of silkworm and other model organism and realizing how the decline physiological and biological activities of silkworm insects continuously to be one of the remarkable question with relevant to aging research. But it has been difficult to address yet in detailed by gerontologists..

Table.1 Longevity of different group of insects including lepidopteran silkworm *Bombyx mori*

Insects group	Male (days)	Female (days)	Reference
Longevity of insects belonging to Dipteran group			
<i>Drosophila melanogaster</i>			
Wild (line 107)	38.1	40.1	Gonzalez (1923)
Vestigial mutant	15.0	21.0	
<i>Drosophila subobscura</i>			
9 inbred lines (average) (1959)	40.0	36.4	Maynard-Smith
4 outbred population (average)	56.8	60.0	
<i>Musca domestica</i> (1959)	17.5	29.0	Rockstein and Lieberman
<i>Musca vicina</i>	20.8	23.3	Feldman-Muhsam and Muhsam (1945)
<i>Calliphora erythrocephala</i>	35.2	24.2	
<i>Aedes aegypti</i>	-	15	Kershaw <i>et al.</i> (1953)
Longevity of insects belonging to Lepidopteran group			
Silkworm <i>Bombyx mori</i> (unmated)	11.9	11.9	Alpatov and Gordeenko (1932)
Silkworm <i>Bombyx mori</i> (mate)	15.2	14.2	
<i>Acrobasis caryae</i>	6.5	7.3	Pearl and Miner (1936)
<i>Fumea Crassiorella</i> (unmated)	-	5.5	Matthes (1951)
<i>Fumea Crassiorella</i> (mated)	-	2.3	
<i>Samia cecropia</i> (1932)	10.4	10.1	MacArthur and Baillie
<i>Samia californica</i>	8.7	8.8	
<i>Tropea luna</i>	5.9	6.0	
<i>Philosamia cynthia</i>	5.9	7.1	
Longevity of insects belonging to Hymenopteran group			
<i>Apis mellifera</i>			
Summer bees	-	35	Ribbands, (1952)
Winter bees	-	350	Maurizio, (1959)
<i>Habrobracon juglandis</i>			
Wild type	24	29	Georgiana, (1949)
Small wings, white eyes mutant	20	24	
Longevity of insects belonging to Orthopteran group			
<i>Blatta orientalis</i>	40.2	43.5	Rau, (1924)
<i>Periplaneta americana</i> (1942)	200	225	Griffiths and Tauber,
<i>Schistocerca gregaria</i>	75	75	Bodenheimer, (1938)
Longevity of insects belonging to Coleopteran group			
<i>Tribolium confusum</i>	178	195	Park, (1945)
<i>Tribolium madens</i>	199	242	
<i>Procrustes</i>	374	338	Labitte, (1916)
<i>Carabus</i>	323	386	
<i>Melolontha vulgaris</i>	19	27	

Fig.1 Evolution of different insects in percentage (Source from Grimaldi and Engel, 2005)

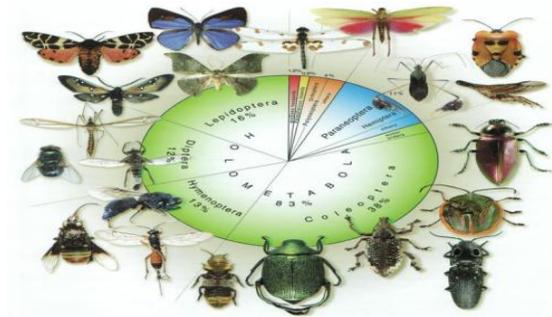


Fig.2 Characteristics of aging process. Fig.3 Theories of aging (Classification) (Source: IUMB Life, 2007)

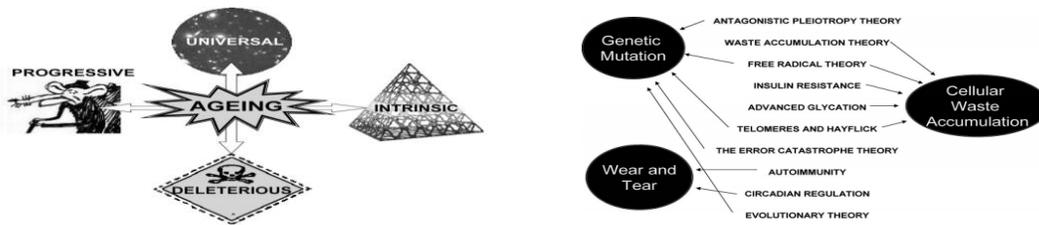


Figure.4 Life cycle of the silkworm *Bombyx mori* (Source: Image belongs to Joanna Rose Tidey)

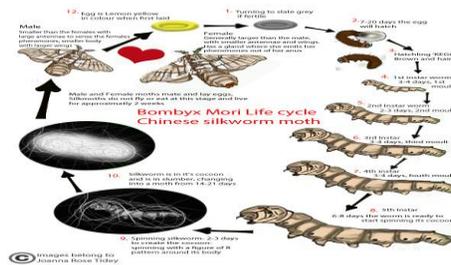
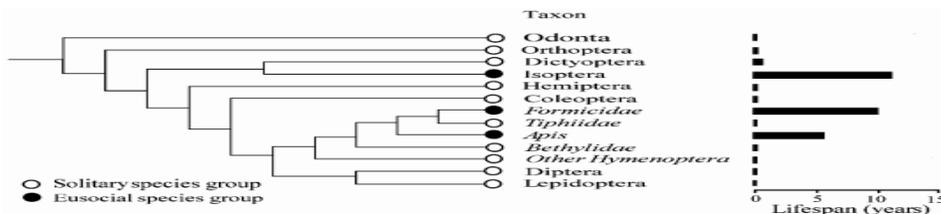


Fig.5 Phylogenetic cluster showing relative reproductive lifespan of various insect groups (taxa), Lepidopteran (*Silkworm, Bombyx mori*) and Dipteran (*Drosophila melanogaster*) groups are showing closely relative reproductive lifespan. (Source: Nature, Keller & Genoud, 1997)



Recently, several gerontologists are concentrating on importance of aging process

with relevant to physiological and biochemical factors in different insects and

also in silkworm *Bombyx mori*. Further, Keller & Genoud, (1997) showed the relationship between lepidopteron (Silkworm, *Bombyx mori*) and Dipteran (*Drosophila melanogaster*) groups were closely related with reproductive lifespan (Fig. 5). In *Drosophila* research was done on complex organ and endocrine system that were allow to study the role of conserved signalling transduction path ways with help of genetic and oxidative stress and metabolic changes along with between signalling systems insulin respecer (Tatar *et al.*, 2003), target of rapamycin (TOR) and Jun N-terminal kinase (JNK) have been reward some of the important documentary evidences on aging by well known researchers.

However, Lim *et al.* (2005) was reviewed on physiological activities of aging in *Drosophila* and suggested that insect sleep and organ specific aging as also been remained vital to understand the aging phenomena in *Drosophila*. Further, Tettweiler *et al.* (2005) were unravel in their research, the changes of several energy compounds with response signalling pathway and amino acids and it is to noted that the size, growth and longevity of insects regulate in *Drosophila*. However, regulation of metabolism as well as lifespan is depends on dTORacts downstream of dFOXO which are inter connected with INR pathway (Luong *et al.* 2006). Earlier study of Wang *et al.* (2005) have focused the cross talk between INR, TOR and JNK signalling in aging. Moreover, another report revealed that decline of lifespan observed due to up-regulating the levels of antioxidants (Bayne *et al.*, 2005). On other hand some evidence by Fridell *et al.* (2005) whose research says the increase of longevity by decreasing in ROS generation because human uncoupling protein 2 (Lucpz) in the mitochondria of adult fly, neurons and human SOD in adult fly motor neurons. Above statement directly supports to the advantage of utilizing *Drosophila* as a

model organism for testing the various physiological functions of human proteins with relevant to aging.

Nevertheless, the study by Partridge *et al.* (2005) revealed not much significant differs of ROS production and metabolic rate compare to controls. In some extent, several researches proved that, Shaw *et al.* (2000) noted insects flies use to showed remarkable sleeping characteristic features, this study revealed the rate of sleep correlated with the fragmentation of longevity and that leads to shortening of lifespan. Further, irregular sleeping causes physiological aging disorders (Koh *et al.* 2006). In some cases, the physical activity changes said to be effects on longevity and that may do so through affecting on metabolic rates, the forced decrease of physical activities in house flies through housing single fly in normal containers that will not allow them to fly hence enhances the longevity (Buchan and Sohal, 1981). Few reports suggested like, in mutants K^+ channels (hyperkinetic, shaker) increases physical activity and shorten life span (Trout and Kaplan, 1970).

Extrinsic factors effects aging

Extrinsic aging is a word which also involves cutaneous aging, which is also part of the integumentary system. The extrinsic aging is also known as photoaging. Because of the radiation which effects more among the all factors which influence on aging. Hence, the term extrinsic aging is often referred as photoaging (Gilchrest, 1998 and Uitto *et al.*, 1998).

In conclusion, different group of insects in general and lepidopteron silkworm *Bombyx mori* in particular are immensely provide a unique system for aging investigations under advanced scientific world. Based on inferences from different phenomenon that is

physiological, biochemical, ecological, cytological, genetical and molecular studies, it seems like that, the striking longevity play an important role on over all the benefits/income from highly economical important insects like, *Bombyx mori* in silk industry. A dramatic progress have been achieved right from the early years to modern year through identifying specific physiological, biochemical, genetical, molecular knowledge, etc., which extends/decrease the aging pattern of different group of insects including silkworm *Bombyx mori* by utilizing different research procedures and techniques in the life science. Hence, it is noteworthy that, the said phenomenon of aging viz., intrinsic and extrinsic factors are basically helps to understand the longevity and dramatic life span differences existed between each and every races/breeds/hybrids of the silkworm *Bombyx mori*, which are fundamental base to further research investigations to be explored several evolutionary information relevant to shortened the lifespan. The gathered information herein this review paper will glance a great promise and encourage to shape fundamental answer for micro or macro level researchers to both gerontologists and evolutionary biologist from a view point of economical and scientific growth & development of the silk industry in India and other part of the world.

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