

The experimental evidences of steamed and freeze-dried mature silkworm powder as the calorie restriction mimetics

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Abstract

Steamed and freeze-dried mature silkworm powder (SMSP) is a natural food containing a large amount of various functional materials and has various health promoting effects. SMSP is known to increase the life expectancy and healthspan, simultaneously. The accomplishment of extension of healthspan should be possible to achieve by activating various signaling pathways delaying aging in various tissues, not by regulating only a few signaling pathways. Consistent with this notion, SMSP increased the resistant to Parkinson disease by enhancing olfaction and mitochondrial activity in neurons of animal models. In addition, SMSP could enhance the gastrointestinal functions. The animals consumed SMSP showed enhanced alcohol metabolisms, reduced cholesterols in bloods, increased resistance to carcinogens causing liver cancers, and protective effects in alcohol induced stomach ulcers. Furthermore, SMSP was also effective in appearance. The SMSP consumed animals showed reduced skin pigmentations and more hair growth compared with control animals. Taken together, the functional enhancement effects of SMSPs in various tissues and organs, which have been discovered to date, are combined to extend healthspan. Therefore, SMSP can be regarded as calorie restriction mimetics. Further studies in the health promoting effects of SMSP will contribute to identifying new applicable diseases, resulted in increased sales of SMSP and incomes of sericulture farmers.

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Introduction

The annual fertility rate (AFR) of Korea (ARFK) has fallen sharply from 1.297 in 2012 to 0.97 in 2018 (Fig. 1). The ARFK was the lowest among those of OECD member countries and only 57.8% of OECD average AFR (1.68) in 2015. In contrast,

the number of deaths in 2018 is 298,900 and the natural population increase in Korea was only 28,000 (e Country Index, 2019; Statics Korea, 2019)

The life expectancy of Korean has been rapidly extended over the past several decades due to the improvement of the living environment and hygiene of society and individuals as well as

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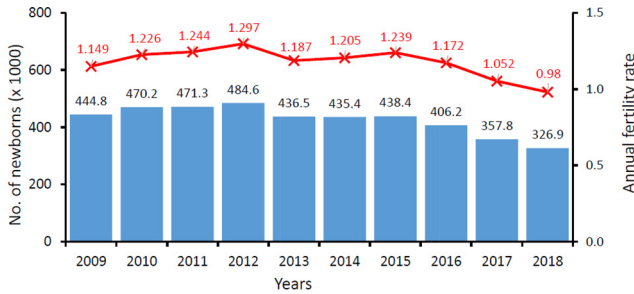


Fig. 1. The trends of annual fertility rates and numbers of newborns in Korea from 2009 to 2018.

The numbers of newborns in Korea from 2009 to 2018 were represented by blue bars. The red lines with x-shaped bullets were representing the annual fertility rates. After 2012, annual fertility ratio was continuously decreased.

the development of bio-medical sciences and technologies due to economic development (e Country Index, 2019). The life expectancy in 2008 for Korean men, women and total which were 76.25, 82.96 and 79.66 year, respectively, were increased to 79.3, 85.41 and 82.36 years in 2016 (Statics Korea, 2019). In addition, a recent study that forecasted the life expectancy of 2030 births in 35 industrialized countries predicted that those of Korean women and man would be the longest in the world with 90.82 and 84.07, respectively (Kontis *et al.*, 2017).

The world's highest expectation of life expectancy and the lowest ARF in Korea are expected to pose a serious threat to its development. A serious problem will occur if the KARF remain below 1.0 as similar as 2018 for next several years and the life expectancy increased as expected. The time when the population of Korea will start to decline in 2027, which was 4 years earlier than a prediction which was made in 2016 (Statics Korea, 2019). And the proportion of producible population (population between 15 and 64 year old) of Korea will be reduced more rapidly, resulted in a social and national economic problem.

The common national socioeconomical problems faced by industrialized countries are a sharp increase in the medical care expenditures of the state and individuals as the proportion of the elderly population increases (Knickman and Snell, 2002). Most of elderly older than 65 years suffer from one or more chronic and/or degenerative diseases. Even though bio-medical sciences and technologies have developed rapidly, the number of diseases that can be cured to date is very limited (Niccoli and Partridge, 2012). Reflecting these facts, different from expected lifespan which rapidly increased, healthspan, a period of life without diseases, was 64.9 years in 2016 which was 0.8 year decreased

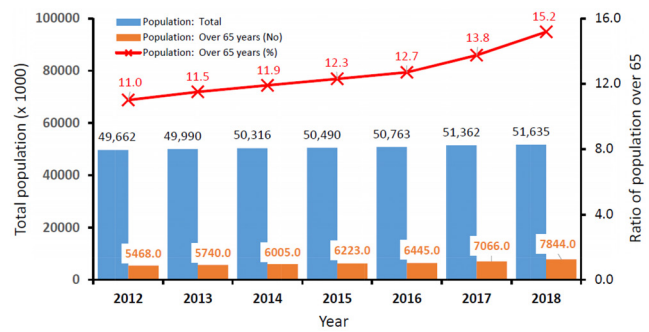


Fig. 2. The changes of total population or population over 65 Total population in Korea indicated by blue bars were increased from 49,662,000 in 2012 to 51,635,000 in 2018. The population over 65 years old represented by orange bras were more rapidly increased 546,800 in 2012 to 784,400 in 2018. The ratios of population over 65 in total population depicted by red lines with x-shaped bullets were changed from 11.0% in 2012 to 15.2% in 2018.

from 65.7 years in 2012 in Korea. The discrepancy between expected life span and healthspan are common phenomena and getting wider and wider in all developed countries, resulted in increase in nursing time and medical care costs. The proportion of the elderly population's medical expenditure (271,357 billion won) accounted for 38.7% in the national medical expenditure (696,623 million won) in 2017, and 39.9% in 2018 (national medical expenditure = 311,173 million won, elderly population's medical expenditure = 779,141 million won) in Korea (Figs. 2 and 3) (Health Insurance Review & Assessment Service, 2019). Since the proportion of the elderly health care expenditure in total healthcare expenditure was 39.9%, 2.62 time more than the raito of the elderly population (15.2%) in 2018, if the proportion of elderly population increases continuously, most of the national budget and individual incomes will be used for elderly health care expenditure, which will become a national socioeconomic problem. Thus, in order to solve these problems, it is urgently needed to dramatically increase the healthspan as much as life expectancy.

The ways to extend healthspan

According to the data from Static Korea for 2016, the healthspan is 64.8 years, which is 17.46 years shorter than the life expectancy. These data suggested that almost all the elderly suffer from one or more diseases after 65 year of age. Currently, the prevention of chronic and/or degenerative diseases has been

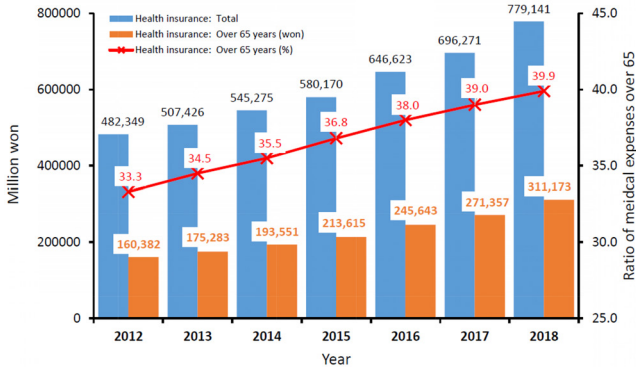


Fig. 3. Trends in health insurance expenditures for the country and only for the elderly population aged more than 65. Total medical expenditures in Korea were increased from 482,349 billion won in 2012 to 779,141 billion won in 2018. The medical expenditures for elderly aged more than 65 were increased from 160,383 billion won in 2012 to 311,173 billion won in 2018. The ratios of medical expenditures over 65 compared to total medical expenditure were increased from 33.3% in 2012 to 39.9% in 2018.

largely dependent on personal preventive efforts. On Oct 2nd, 2016, the senior citizen's day in Korea, the Korean Geriatrics Society announced the 7 methods that improve healthspan (The Korean Geriatrics Society, 2016). Briefly, 1. Reduce the salt intake by half, 2. To maintain a clean dry indoor environment, 3. Quit smoking and drinking alcohol, 4. Exercise a little bit more than 30 minutes each day, 5. Maintain a good interpersonal relationship, 6. If you have metabolic diseases, actively manage them with healthy lifestyles and medication, and 7. Consult with the physician periodically. In addition, preventing inhaling toxic substances such as fine dusts or smokes, maintaining important hormone levels and metabolism through periodic sunbathing, and controlling weight may be necessary efforts. If you have a strong will, you can follow those instructions. However, those restricted life style may cause some problems to maintain smooth interpersonal relations and social life.

In particular, calorie restriction (CR), which limits food intake, has been shown to slow down senescence and increase healthspan in many model animal studies (Fernández-Ruiz, 2017). Therefore, since the 20th century, many researchers of CR have been continuously investigating to achieve the effect of CR by taking a certain amount of medication or food per day. Those foods or drugs can induce the beneficial effects of CR was defined as CR mimetics (CRM). As a result of plethora researches, various types of foods and drugs have been proposed as candidates for CRM (Ingram and Roth, 2015). Although recent

studies have suggested the possibility of incorrect statistical analyses, the most well-known CRM candidates are resveratrol, which is famous for its relationship to French paradox (Catalgol *et al.*, 2012). The French paradox is that French people who consume high-fat, high-calorie foods, similar to other people in the Western countries, have a relatively low cardiovascular mortality rate compared to people in other countries because their consumption of wine is relatively larger than those of other countries. Thus, resveratrol, a plant-derived material with high antioxidant power, which is abundant in wine, has been proposed as a CRM candidate. However, there were various problems such as failure to reproduce the extension of life expectancy and healthspan results in animal model experiments to confirm it as CRM. In addition, metformin (an inhibitor of citric acid cycle) oxaloacetate (an anti-obesity drug), rimonabant, 2-deoxyglucose, and rapamycin (a signaling inhibitor of mTOR signaling) were suggested as CRM candidates, but fail to prove the effects of CR in the experiment (Ingram and Roth, 2015).

The effect of CR is summarized as delayed aging and its accompanying various physiological dysfunctions, thereby lowering the risks of onset and progression of various diseases, resulted in simultaneous extension of life expectancy and healthspan (Fernández-Ruiz, 2017). Therefore, CRM should be able to prolong life expectancy and healthspan when people or animals consume it (Ingram and Roth, 2015). The condition for the increase in the life expectancy together with healthspan in humans or animal models is that the function of all organs should be maintained at a constant level while aging (Fig. 4).

To summarize the results of the current investigations on CRM, the prerequisites for becoming a CRM that increases healthspan by delaying aging might be summarized as follows (Fig. 4). 1. Uptake of CRM will prevent senescence of nervous systems including brains, which regulates the sensory,

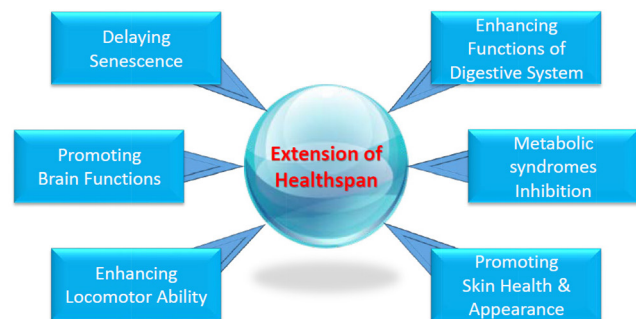


Fig. 4. The prerequisites for the CRM

Table 1. Reported health benefits of various silkworm products

Developmental stage	Larvae		Chrysalis	Silk fibers (cocoons)
	5 th instar 3 rd day	Mature		
Product formulation	Freeze-dried powder	Steamed and freeze-dried powder	Powder/Extracts	Fibroin hydrolysate
Health improvement effects	- Hypoglycemic effects (Ryu <i>et al.</i> , 1997; Ryu <i>et al.</i> , 2002b)	- Extension life expectancy and healthspan (Nguyen <i>et al.</i> , 2016) - Prevention Parkinson's disease (Choi <i>et al.</i> , 2017b; Nguyen <i>et al.</i> , 2016) - Improving liver functions (Hong <i>et al.</i> , 2018) - Prevent liver cancers (Cho <i>et al.</i> , 2016) - Gastrointestinal protective effect (Lee <i>et al.</i> , 2017; Yun <i>et al.</i> , 2017) - Improvement of skin health (Kim <i>et al.</i> , 2017) - Promoting hair growth (Ji <i>et al.</i> , 2019)	- Promoting sexual functions (Oh <i>et al.</i> , 2012; Ryu <i>et al.</i> , 2002a)	- Improvement of memory in human children and adults (Kang <i>et al.</i> , 2018; Kim <i>et al.</i> , 2005)

cognitive, and locomotive ability, the most important body functions for the survival of the individual, 2. The function of the digestive system responsible for nutrient intake and metabolism should be maintained normally, 3. There should be prevention and suppression effects on metabolic diseases such as hypertension, hyperglycemia, hyperlipidemia and obesity, 4. Improving individual's appearance by promoting skin health and/or preventing hair loss will give confidence and stability in interpersonal and social relationships. As such, in order to be CRM, it is not only to simply regulate one signal transduction pathway, but also to control the various signal transduction pathways that can suppress aging in various organs, simultaneously (Testa *et al.*, 2014). Therefore, it is difficult for one single substance to meet the requirements of CRM, and it is highly possible that functional foods contained various functional substances can meet the requirements.

The experimental evidences of steamed and freeze-dried mature silkworm powder as the CRM

Currently, the most closely related substance (food) to CRM is assumed to be steamed and freeze-dried mature silkworm powder (SMSP). First, it has been reported that animals fed with SMSP containing food (SMSPf) showed significantly extended life expectancy and healthspan compared to the control animals fed with normal food (Nf) or experimental groups fed with high nutrient foods (Table 1) (Nguyen *et al.*, 2016). The results

of the comparative analysis of gene expression between the SMSPf fed group and the Nf intake group to ascertain how the SMSP increase the life expectancy and healthspan result in an increase in the expression of the olfactory genes. Consistent with these gene expression differences, SMSPf fed animals showed enhanced olfactory responses compared to normal food fed animals. In addition, the SMSPf fed animals showed resistance to rotenone, which is one of known causes of Parkinson's disease (PD)(Coulom and Birman, 2004; Tanner *et al.*, 2011). One of the molecular and cellular pathologies of PD is mitochondrial dysfunction (Ferretta *et al.*, 2014; Jin *et al.*, 2014), and the mitochondria activity of SMSPf fed animals was found to be higher than that of Nf fed animals (Nguyen *et al.*, 2016). These results suggest that SMSP promotes the activity of mitochondria of nerve cells that regulate olfactory and locomotor ability at the individual level, thereby preventing the onset of PD and delaying the loss of locomotor ability due to aging or rotenone (Choi *et al.*, 2017b; Nguyen *et al.*, 2016).

The next important health improving effect of SMSP is to improve the function of digestive organs responsible for absorption and metabolism of nutrients. Experimental animals receiving SMSP showed resistance to alcoholic gastritis (Table 1) (Lee *et al.*, 2017; Yun *et al.*, 2017). Ethanol-induced gastric mucosal ulceration and hemorrhagic injury were significantly reduced in the rats fed with SMSP before alcohol administration. This reduction in gastric mucosal damage by SMSP was found to be accomplished by regulating the proinflammatory response through the reduction of recruiting immune cells, increased anti-oxidant capacity and mucus secretion (Yun *et al.*, 2017).

Table 2. Summary of crude nutrients and phytochemical compositions in SMSPs

Crude nutrients (%)	Sizes of SMSP particles	
	Fine (average 10 μ M)*	Ultra fine (average 1.1 μ M)**
water	1.71~4.29	1.72~2.87
protein	69.36~73.13	67.22~73.00
lipid	13.57~15.59	9.88~16.00
fiber	2.78~3.50	1.27~1.91
ash	3.43~3.96	3.38~4.30
Phytochemicals (mg/100g)	-	-
Total flavonoids	423.59~552.64	298.78~630.62
Total phenolic compounds	744.45~822.15	587.70~817.78
GABA	1.96~4.59	2.62~4.49
Vitamins (mg/100g)	-	-
Vitamin B1	0.53~0.63	0.46~0.48
Vitamin B2	4.63~6.04	3.43~4.14
Vitamin B3	4.06~4.94	1.63~2.47
Vitamin B6	0.51~0.67	0.43~0.59
Folic acid	0.46~0.72	0.43~0.59
Vitamin C	9.26~12.33	14.54~21.46
β -carotene	1.11~7.30	1.86~5.71

Modified from Ji *et al.* (2017)

** Modified from Ji *et al.* (2019)

Interestingly, these gastric mucosal protective effects were found to be the most effective in the SMSP generated with the silkworm varieties that make white cocoons than other varieties spinning color cocoons (Table 1) (Lee *et al.*, 2017).

In addition, SMSP had the effect of improving the functions of the liver which is a known major organ for energy metabolisms, detoxifications, and hematopoiesis (Table 1) (Cho *et al.*, 2016; Hong *et al.*, 2018). The rats consumed SMSP showed improvement of liver alcohol decomposition ability compared with that of control rats and also significantly reduced the incidence of fatty liver caused by long-term alcohol administration (Hong *et al.*, 2018). SMSP has also shown to reduce fatty liver by increasing the expression of β -oxidation-related genes that burn fat, and alleviating the oxidative stress and inflammatory response that are exacerbated by alcohol consumption. In addition, the amounts of cholesterol in bloods of rats fed with SMSP showed significant reduction compared with control rats (Hong *et al.*, 2018). Interestingly, the intake of

SMSP in rats has been reported to inhibit the progression of liver cancer by carcinogens such as Diethylnitrosamine (DEN) (Cho *et al.*, 2016). The size and number of cancer masses induced by DEN were significantly reduced by SMSP supplementation in rats. These results show that SMSP improves the liver function, inhibits the progression of cancer and the metabolic syndrome by lowering blood cholesterol levels.

The final health improvement effect of SMSP so far is appearance enhancement (Table 1). Since skin is the primary immune-related organ that protects the human body and animals from invading foreign physical and harmful substances and microorganisms, maintaining health of skin is very important for human and animal health (Di Meglio *et al.*, 2011). And skin whitening is one of the reasons why people do makeup. As a result of studying UV-irradiated mouse models of eating SMSP or applied to the skin, there was a dose-dependent superior whitening effect in the group receiving the SMSP compared to the untreated control group (Kim *et al.*, 2017). The anatomical

analysis of skins revealed that these skin whitening effects of SMSP was accomplished by decrease formation of melanocytes after UV radiation. It was also confirmed that when the fine or ultrafine SMSPs were applied on epidermis of epilating mouse, hair growth enhancement effect was observed compared to the control (Ji *et al.*, 2019). These results show that SMSP improves appearance by preventing skin darkening and hair loss.

Discussion

As of 2019, it's been only about six years since SMSP was invented, but as already mentioned above, versatile functionality of it has already been verified (Table 1). Therefore, it is presumed that new functionalities of SMSP could be discovered in future studies. In order to overcome the national crisis caused by the rapid increase of medical expenditure and the decrease of economically active population due to the increase of the elderly population and the decrease of numbers of newborn babies, it is necessary to significantly increase healthspan. To summarize recent researches, extension of healthspan may not be accomplished by controlling one signal transduction pathway, rather than many signal transduction pathways involved with increasing functions and activities of various cells and organs and inhibiting senescence of them must be regulated. Therefore, SMSP with brain function and locomotor improvement effects (Choi *et al.*, 2017b; Nguyen *et al.*, 2016), improving liver function and inhibiting liver cancer development at the same time (Cho *et al.*, 2016; Hong *et al.*, 2018), suppressing the onset of gastrointestinal disease (Lee *et al.*, 2017; Yun *et al.*, 2017), and promoting skin health (Kim *et al.*, 2017) and hair growth (Ji *et al.*, 2019) can be considered to be the best candidate for CRM at present. The reason why SMSP can have such various functions can be explained by its nutritional compositions (Table 2). Humans have been feeding silk moth chrysalis for a long time (Mishra *et al.*, 2003; Tomotake *et al.*, 2010). In addition, improvement of cognition of human children and adults by the hydrolysates of fibroin (Kang *et al.*, 2018; Kim *et al.*, 2005), hyperglycemic effects of freeze-dried the 5th instar 3rd day silkworm powders (Ryu *et al.*, 1997; Ryu *et al.*, 2002b), and enhancing sexual functions by extracts or powders of male silk moth chrysalis (Oh *et al.*, 2012; Ryu *et al.*, 2002a) were well known health improvement effects of mulberry silkworms. These previous reports suggested that the silkworm contains

various functional materials that have various health promoting effects depending on its developmental stage. Thus, the mature silkworms, which contain enlarged silk glands, must retain all of the nutrients present in silkworm larvae, chrysalis, and silk fibers in cocoons (Ji *et al.*, 2019; Ji *et al.*, 2017; Ji *et al.*, 2016a; Ji *et al.*, 2016b; Ji *et al.*, 2015). Nutrient composition analysis of SMSP showed that although protein contents of SMSP were differed by silkworm varieties, they were ranged between 69.3~73.13% (Ji *et al.*, 2019; Ji *et al.*, 2017; Ji *et al.*, 2016a; Ji *et al.*, 2016b), which is the highest rate of among all foods. The second common nutrient components in SMSPs were fatty acids ranged between 13.6~15.6% in diverse SMSPs (Ji *et al.*, 2019; Ji *et al.*, 2017; Ji *et al.*, 2016a; Ji *et al.*, 2016b). The important characteristic of fatty acids in SMSPs was that the amounts of Omega-3 fatty acids in SMSPs were 3.8~5.7 times more than those of Omega-6 fatty acids. It has been reported that consuming higher contents of Omega-6 fatty acid foods increased the probability of developing cardiovascular diseases (Simopoulos, 2002). Thus, consuming SMSPs with high contents of Omega-3 fatty acids may decrease the possibility of onset of cardiovascular diseases. The third component is inorganic salts, accounting for 3.43 to 3.97% of SMSPs (Table 2) (Ji *et al.*, 2019; Ji *et al.*, 2017; Ji *et al.*, 2016a; Ji *et al.*, 2016b). While a large amount of essential inorganic salts such as calcium, copper, iron, magnesium, manganese, phosphorous, zinc, sulfur, etc were enriched in SMSPs, but heavy metals such as chromium, lead, cadmium, mercury, and arsenic were not detected in SMSPs. Interestingly, the amount of potassium was 14.82~18.18 times more abundant than that of sodium in SMSPs. Since high sodium foods are known to cause hypertension and high potassium foods are known to lower blood pressure (Burnier, 2018), the intake of SMSP is presumed to have a blood pressure lowering effect. The last nutrient components present large quantities in SMSPs are phytochemicals such as vitamins, polyphenols, flavonoids, GABA, etc, originated mainly from the mulberry leave (Choi *et al.*, 2017a; Choi *et al.*, 2017b; Ji *et al.*, 2019; Ji *et al.*, 2017; Ji *et al.*, 2016a). These mulberry-originated materials are known to have strong anti-oxidant, resulted in delaying senescence in cells, organs, and individuals and contributing on extension of healthspan. We compared the effects of plant-derived compounds and small molecules extracted from SMSPs and the rest of the substances after extractions on healthspan extending effect. As a result, the SMSP-extracted plant-derived compounds and small molecules and the rest of the extracts did not have significant

healthspan extending effects (Choi *et al.*, 2017b). These results indicate that various substances involved in SMSP need to regulate various signal transduction mechanisms in order to increase healthspan.

Taken together, the results of SMSP nutrient composition analysis suggested that combinatorial interactions of various nutrients and functional materials abundant in SMSPs may delay senescence and/or improve physiological function of cells, tissues, and organs, resulted in promoting functions of brains, locomotor controls, gastro-intestinal, skin health, appearance, etc. The summation of promoted functions of organs may be resulted in extension of healthspan.

In conclusion, SMSP includes a wide variety of functionality that is not currently unidentified. Therefore, in the future, it is necessary to study the novel functionalities of SMSP and to study the mechanisms at the molecular level of the functions revealed to date. These efforts will be of great help in curbing the sharp rise in medical costs due to the increasing elderly population facing developed countries, including Korea, and will enable SMSP to be developed as a new natural medicine.

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References

- Burnier M (2018) Should we eat more potassium to better control blood pressure in hypertension? *Nephrol Dial Transplant* 34, 184-193.
- Catalgol B, Batirel S, Taga Y, Ozer N (2012) Resveratrol: French paradox revisited. *ront Pharmacol* 3, 141.
- Cho JM, Kim KY, Ji SD, Kim EH (2016) Protective effect of boiled and freeze-dried mature silkworm larval powder against diethylnitrosamine-induced hepatotoxicity in mice. *J Cancer Prev* 21, 173-181.
- Choi BH, Ji SD, Jeong JH, Kim KY, Koh YH, (2017a) Quantification and comparison of functional phytochemicals in steamed and freeze-dried mature silkworm powders and freeze-dried mulberry leaves. *Int J Indust Entomol* 35, 89-96.
- Choi BH, Ji SD, Son JG, Nguyen P, Kim KY, Park YH, *et al.* (2017b) Phytochemicals and silk proteins in mature silkworm powders responsible for extended life expectancy and enhanced resistances to Parkinson's disease. *J Asia-Pac Entomol* 20, 1425-1433.
- Coulom H, Birman S (2004) Chronic Exposure to Rotenone Models Sporadic Parkinson's Disease in *Drosophila melanogaster*. *J Neurosci* 24, 10993-10998.
- Di Meglio P, Perera GK, Nestle FO (2011) The multitasking organ: recent insights into skin immune function. *Immunity* 35, 857-869.
- e Country Index (2019) http://www.index.go.kr/potal/main/EachDtlPageDetail.do?idx_cd=1399.
- Fernández-Ruiz I (2017) Calorie restriction for healthy ageing. *Nat Rev Cardiol* 14, 190.
- Ferretta A, Gaballo A, Tanzarella P, Piccoli C, Capitanio N, Nico B, *et al.* T (2014) Effect of resveratrol on mitochondrial function: Implications in parkin-associated familiar Parkinson's disease. *Biochim Biophys Acta* 1842, 902-915.
- Health Insurance Review & Assessment Service (2019) <https://www.hira.or.kr/main.do>
- Hong KS, Yun SM, Cho JM, Lee DY, Ji SD, Son JG, *et al.* (2018) Silkworm (*Bombyx mori*) powder supplementation alleviates alcoholic fatty liver disease in rats. *J Funct Foods* 43, 29-36.
- Ingram DK, Roth GS (2015) Calorie restriction mimetics: Can you have your cake and eat it, too? *Ageing Res Rev* 20, 46-62.
- Ji SD, Kim SB, Kim KY, Kim NS, Kim SW, Jo YY, *et al.* (2019) Contents of nutrients in ultra-fine powders of steamed and lyophilized mature silkworms generated by four silkworm varieties. *J Asia-Pac Entomol*, in print.
- Ji SD, Nguyen P, Yoo SM, Kim KY, Son JG, Kweon HY, *et al.* (2017) Comparison of nutrient compositions and pharmacological effects of steamed and freeze-dried mature silkworm powders generated by four silkworm varieties. *J Asia-Pac Entomol* 20, 1410-1418.
- Ji SD, Kim NS, Kweon HY, Choi BH, Kim KY, Koh YH (2016a) Nutrition composition differences among steamed freeze-dried mature silkworm larval powders made from 3 *Bombyx mori* varieties weaving different colored cocoons. *Int J Indust Entomol* 33, 6-14.
- Ji SD, Kim NS, Kweon HY, Choi BH, Yoon SM, Kim KY, *et al.* (2016b) Nutrient compositions of *Bombyx mori* mature silkworm larval powders suggest their possible health improvement effects in humans. *J Asia-Pac Entomol* 19, 1027-1033.
- Ji SD, Kim NS, Lee JY, Kim MJ, Kweon H, Sung G, *et al.* (2015) Development of processing technology for edible mature silkworm. *J Seric Entomol Sci* 53, 38-43.
- Jin H, Kanthasamy A, Ghosh A, Anantharam V, Kalyanaraman B, Kanthasamy AG (2014) Mitochondria-targeted antioxidants for

- treatment of Parkinson's disease: Preclinical and clinical outcomes. *Biochim Biophys Acta* 1842, 1282-1294.
- Kang YK, Lee BY, Bucci LR, Stohs SJ (2018) Effect of a Fibroin enzymatic hydrolysate on memory improvement: a placebo-controlled, double-blind study. *Nutrients* 10, 233.
- Kim DK, Kang YK, Lee MY, Lee KG, Yeo JH, Lee WB, *et al.* (2005) Neuroprotection and enhancement of learning and memory by BF-7. *J Health Sci* 5, 317-324.
- Kim HJ, Kim KY, Ji SD, Lee HT (2017) Anti-melanogenic activity of steamed and freeze-dried mature silkworm powder. *J Asia-Pac Entomol* 20, 1001-1006.
- Knickman JR, Snell EK (2002) The 2030 problem: caring for aging baby boomers. *Health Serv Res* 37, 849-884.
- Kontis V, Bennett JE, Mathers CD, Li G, Foreman K, Ezzati M (2017) Future life expectancy in 35 industrialised countries: projections with a Bayesian model ensemble. *Lancet* 389, 1323-1335.
- Lee DY, Cho JM, Yun SM, Hong KS, Ji SD, Son JG, *et al.* (2017) Comparative effect of silkworm powder from 3 *Bombyx mori* varieties on ethanol-induced gastric injury in rat model. *Int J Indust Entomol* 35, 14-21.
- Mishra N, Hazarika NC, Narain K, Mahanta J (2003) Nutritive value of non-mulberry and mulberry silkworm pupae and consumption pattern in Assam, India. *Nutr Res* 23, 1303-1311.
- Nguyen P, Kim KY, Kim AY, Kim NS, Kweon HY, Ji SD, *et al.* (2016) Increased healthspan and resistance to Parkinson's disease in *Drosophila* by boiled and freeze-dried mature silk worm larval powder. *J Asia-Pac Entomol* 19, 551-561.
- Niccoli T, Partridge L (2012) Ageing as a risk factor for disease. *Curr Biol* 22, R741-R752.
- Oh HG, Lee HY, Kim JH, Kang YR, Moon DI, Seo MY, *et al.* (2012) Effects of male silkworm pupa powder on the erectile dysfunction by chronic ethanol consumption in rats. *Lab Anim Res* 28, 83-90.
- Ryu KS, Ahn MY, Lee HS, Kim I, Kim JW, Kim SH, *et al.* (2002a) The tonic effect of the extract from male silkworm (*Bombyx mori* L.) pupae on rats. *Int J Indust Entomol* 5, 123-126.
- Ryu KS, Lee HS, Chung SH, Kang PD (1997) An activity of lowering blood-glucose levels according to preparative conditions of silkworm powder. *J Seric Entomol Sci* 39, 79-85.
- Ryu KS, Lee HS, Kim IS (2002b) Effects and mechanisms of silkworm powder as a blood glucose-lowering agent. *Int J Indust Entomol* 4, 93-100.
- Simopoulos AP (2002) The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomed Pharmacother* 56, 365-379.
- Statics Korea (2019) <http://kostat.go.kr/portal/eng/index.action>
- Tanner CM, Kamel F, Ross GW, Hoppin JA, Goldman SM, Korell M, *et al.* (2011) Rotenone, Paraquat, and Parkinson's Disease. *Environ Health Perspect* 119, 866-872.
- Testa G, Biasi F, Poli G, Chiarpotto E (2014) Calorie restriction and dietary restriction mimetics: a strategy for improving healthy aging and longevity. *Curr Pharm Des* 20, 2950-2977.
- The Korean Geriatrics Society (2016) <http://www.geriatrics.or.kr/>
- Tomotake H, Katagiri M, Yamato M (2010) Silkworm pupae (*Bombyx mori*) are new sources of high quality protein and lipid. *J Nutr Sci Vitaminol (Tokyo)* 56, 446-448.
- Yun SM, Cho JM, Hong KS, Lee DY, Ji SD, Son JG, *et al.* (2017) Gastroprotective effect of mature silkworm, *Bombyx mori* against ethanol-induced gastric mucosal injuries in rats. *J Funct Foods* 39, 279-286.