CHAPTER II
LITERATURE REVIEW

2.1 General reviews of Antioxidant

2.1.1 Antioxidant

In chemical term, antioxidant is chemical component that can able to prevent and slow down the oxidation reaction. Oxidation reactions can occur in many ways, such as oxidation which make steel becomes rusty, change the colors of apple to becomes browns or make vegetable oil to becomes rancid (Songchitsomboon, 2005).

In addition, oxidation reaction can occur in human body. For example, digestion process from food to protein and fat, respiration process which can intake air pollution such as smoke from cigarettes and UV radiation. All of these processes can make free radicals and causes many effects to human body. In fact, one chemical component cannot prevent all of the oxidation reactions. Some oxidation reactions need different antioxidant to prevent the reaction.

In other ways, oxidation reactions are very important to our human body. For instance, human use oxygen from respiratory system for digesting and use energy from this process for working cells in our body. However, free radical which is one of side product from this process will make reaction with other important molecules in the body and cause side negative effect to these molecules, such as fat, protein and DNA.

For example, when free radical cause oxidant reaction with LDL (Low density lipoprotein) is negative cholesterol make oxidized LDL. There is the study that proof this chemical can cause Atherosclerosis which is one of the reasons to hearth attack.

Free radicals occurred in the body are caused of oxygen, it is called Reactive Oxygen Species (ROS). The important free radicals are superoxide anion (O_2-l), hydrogen peroxide (H_2O_2) and hydroxyl radical (-OH)

2.1.2 Role of antioxidant

There are many researches show the antioxidant can reduce the probability of many diseases, especially diseases that came from food habit, such as cancer, diabetes, heart diseases and Alzheimer (Songchitsomboon, 2005).

Moreover, it can slow down the aging processes. Normally, human body has capability to destroy free radicals before it can harm our body. However, when the creating of free radical is more than our capability to destroy, it will cause damage to our cell and tissue then it can harm to our healthy.

The way of reducing free radicals damage by antioxidant

1. Reducing the making of free radicals
2. Reducing damage from free radicals

Even though, antioxidant cannot fix the damage which is happened before but it can slow down the damaging, especially damaging from chronic diseases which are cells and tissues were damage for long time (mostly more than ten years). Due to this reason, most of the people who have chronic diseases are middle age and elder. Consequently, everyone should have enough antioxidant in everyday in order to control the free radicals in our body.

The example of antioxidants are vitamin C, vitamin E, Silinium, Beta-carotene, vitamin A and phytochemical, such as polyphenol from teas and herbs or isoflavones from soybeans

In order to have enough antioxidant, we should eat vegetable by washing it every times. In addition, vegetables are contained dietary fibers which can help digestion system and prevent constipation. Moreover, it can accelerate the process of removing the toxic which can cause cancer from our body.

2.1.3 Phenolic compounds

Phenolic compounds are the compound that structural formulas have more than 1 of -OH group on aromatic ring. These compounds group qualify to dissolve in water well and found in generally plants, vegetables and fruits (Laopatarakasem, 2004). Divided into three groups as follow;

1. Simple phenols/phenolic acid and derivatives such as gallic acid, ellagic acid, tannic acid, vanillin, catechol, resorcinol and salicylic acid. These compounds group found in many types of fruits such as raspberry and blackberry.

2. Phenylpropanoids such as phenolic compound that aromatic ring has three-carbon side chain binding. Separated into several groups such as hydroxycinnamic acids (ferulic acid, caffeic acid or coumaric acid), coumarins (umbelliferone, scopoletin, aesculetin or psoralen), lignans (pinoresinol, eugenol or myristicin) and can found in apple pear and coffee.

3. Flavonoids are an important group of phenolic compounds including structural formulas C6-C3-C6. Separated into several groups including catechins, proanthocyanins, anthocyanidins, flavones, flavonols, flavonones and isoflavones. From the flavonoids that are founded widely in both plants, vegetable, fruits including drinks prepared from plants such as tea. In the tea leaves found the catechins up to 30% of tea leaves dry weight and believe that is very important to act as antioxidant and chemoprevention. Anthocyanins is pigments in plants so flavones, flavonols and isoflavones are commonly found and believe that are beneficial to the health.

Table 2.1 Various flavonoids from natural source (Hertog, 1998)

Class	Typical sources	Representative
Flavonols	Tea, onions, red wine, fruit	Quercetin
Flavones	Vegetables, citrus fruits	Apigenin
Flavanones	Citrus fruit	Hesperitin
Anthocyanidins	Berries, colored fruit	Cyanidin
Catechins	Tea, wine	Epigallocatechin
Isoflavonoids	Legumes	Genistein

2.2 Antioxidant Analysis

There are 3 generally methods include (Duangnak, 2004)

1. Antioxidant activity is the method that to determine the effectiveness of antioxidants to against the occurrence free radicals of linoleic acid.

2. Reducing power is the method that to determine the reducing ability in antioxidant.

3. Scavenging effect on 1,1-diphenyl-2-picrylhydrazyl radicals (DPPH) is the method that to determine the antioxidant ability to remove the DPPH which is a kind of free radical.

2.2.1 The analysis for the ability of the antioxidant activity to prevent the occurred of hydroperoxide.

Linoleic acid is one of the fatty acid which is a double bond composition. It can pair with other radical in system that's made free radical the oxidized made hydroperoxide which is a stable conjugated diene. The antioxidant with double bond composition can provide the electron for a free radical which decrease the possibilities the creation of free radical of Linoleic acid as well. Therefore to evaluate the ability of this antioxidant for the creation of the free radical of Linoleic acid can be made by add the antioxidant in the solution which contain Linoleic acid then leave for a while. After that use UV Spectrophotometer measure the solution at 234 nanometer. The result varies with the concentrate of hydroperoxide which occurred. Decrease of the result can indicated the ability of the antioxidant to prevent the creation of hydroperoxide.

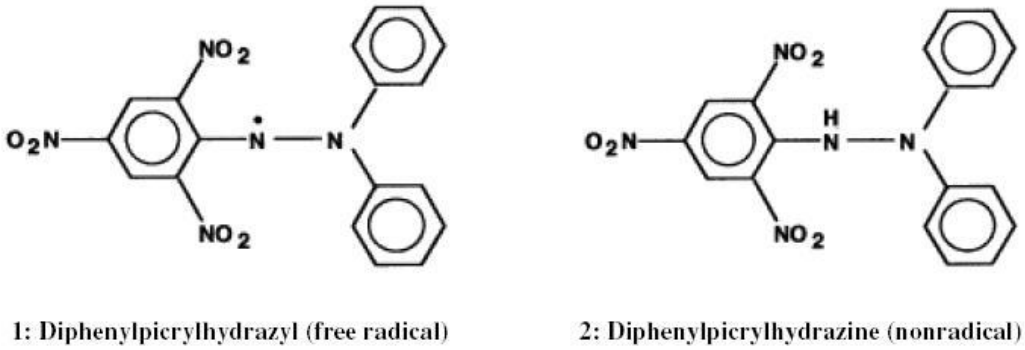
2.2.2 The analysis the ability to reducing power of antioxidant.

The reducing agent substance can provide the electron to the atom or molecule of metal which can be split to ion (such as metal, copper) (Halliwell and Gutteridge, 1984). The Ferric ion (Fe^{3+}) has good ability to extract the electron from other substance. In term of biochemistry, mostly found free radical is sensitive reaction oxygen (ROS). The free-ion from metal catalysis the occurrence of ROS such as superoxide and ion radical, hydrogenperoxide and hydroxine radical. To compare the abilities of electron's provider from each extraction substance, the period of the reaction between Ferric acid and each extraction substance must stable and the result

of the absorbance by measure at 700 nanometer from UV Spectrophotometer will result varies by the concentration from Ferrus ion which occurred. The increase of the absorbance can indicate the abilities of the reducing agent.

2.2.3 The analysis the ability of free radical substance by DPPH (Scavenging effect on 1,1- diphenyl-2-picrylhydrazyl radical (DPPH)

DPPH is a stable free particle and can obtain electron to change the molecule which is not free radical and after obtain hydrogen atom from other molecule will not change those substance to free radical as the picture below;



1: Diphenylpicrylhydrazyl (free radical) 2: Diphenylpicrylhydrazine (nonradical)

Figure 2.1 The transformation of DPPH when obtained electron

Therefore, the abilities of this antioxidant substance is the research for the efficiency of the condense between antioxidant substance and DPPH in terms of stabilized free radical inside the solution (Mostly use DPPH in terms of free radical due to the easily used). The test will use DPPH (dark purple color) react with the antioxidant substance in the specific time range. The absorbance measure will set at 517 nanometer of Spectrophotometer will varies with the concentration of DPPH. Consequently the decrease of a concentration DPPH (lower purple color) indicated the abilities to eliminated a free radical of antioxidant substance.

2.3 General information of Mango

The scientific name of mango is *Mangifera indica* Linn. which the family name is Anacardiaceae. They have the origin in Tropical Asia region. Mango is the perennial plant that high 10 – 15 meters (Suwanalert, 2009). Mango in Thailand has many varieties which are separated into 3 groups of the consumption characteristic as follow;

1. Raw mango such as Namdokmai Mun, Pimsaen Mun, Rad, Keaw savoy, Nong sang, Fah lun, Mun Wan Pak chong, Bao songkla.

2. Ripe mango such as Ok-rong, Ok-rong pikulthong, Namdokmai, Nang klangwan, Thong dam.

3. Mango processing industry;

- Pickled mango such as Kaew, Chok anan.
- Canned mango, fruit juice, compote mango such as Sam pee, Mahachanok.

2.4 General description for mango cultivation

Thailand plants mango for a long time which carried more than 100 varieties. Mango can adapt itself to Thailand weather very well and mango is the most popular fruit cultivated in Thailand. To choose the suitable planting area should be considered as below (Department of Agriculture, 2008);

The varieties that the market has a demand and high productivity according to the recommendation of Department of Agriculture, 2008 are as follow;

- Kaew Srisaket 007 variety

Well grown in a various condition especially low fertility and dry. The average productivity is 300 mango fruit per tree per year and average weight is 252 grams per fruit that include 81% mango pulp which is good quality to the processing.

- Keaw morrakot variety

It is a mixed gene between Kaew Keaw and Sam-pee. They have hard pulp, thick peel, good smell and good taste. Percentage of pulp is 59% which is high sugar rate.

- Talab nark variety

The average productivity is 841.5 mango fruit per tree per year and average weight is 395.40 grams per fruit which is quite a big size of mango.

- Chok anan

The origin was grown by seed from Chinese grave, Mae rim district, Chiangmai province. They has hard pulp and thick peel which is sweet taste.

- Sam pee

Good singular smell. Orange and fine pulp with little fiber. Highly productivity but small size.

2.5 Composition of mango seed

The recently researches only focus on mango seed genus M.indica from India, Egypt and Nigeria. From this researches, it shows that the composition of mango seed is different depends on the location and weather. The mango seed kernel is white, sour and the moisture is around 51%. When dissolve in 50 times of water, it will be acidity (pH 5) and the colors will change to browns. This shows the oxidation of phenolic component by polyphenol oxidase. After drying with different methods, such as sun or steaming, the moisture will be around 9-12 %. In addition, the dry mango seed kernel is contained protein 5-6%, fat 9-16%, carbohydrate 73-80%, fiber 2-3%, ash 2-3% and tannin 1% (Inprakhon, 2009).

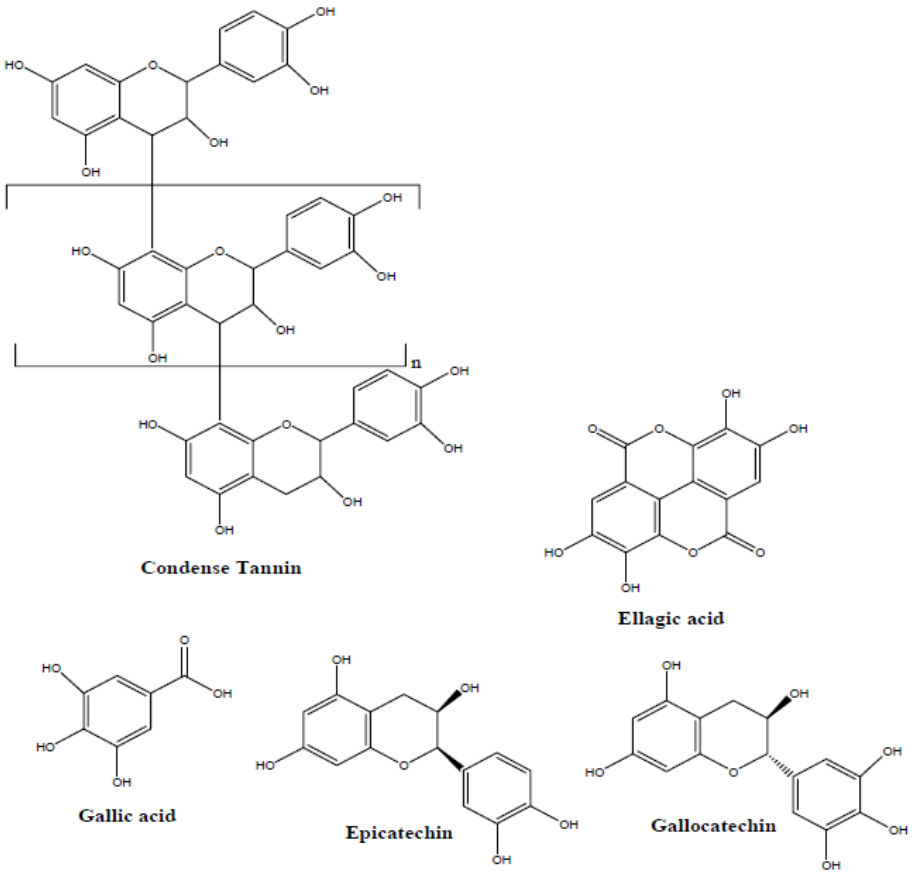


Figure 2.2 Antioxidant type polyphenol and phenolic compound found in mango seed kernel (Arogba, 2000 ; Puravankara, 2000)

2.6 Applied using mango seed kernel in food products

The mango seed kernel is high nutrition because it is contained protein, fat, starch and fiber. The local and poor people in India and Africa consume this seed instead of starch food by roast or boil since the ancient times and some Indian food use the mango seed kernel as ingredients, such as Chapati and Supari (Inprakhon, 2009).

In some places, the starch is made from the mango seed kernel. This starch has resistance starch in very high amount (75-80%). Therefore, consuming this starch will low glycemic index.

The components of this starch are amylose 40% and amylopectin 61%. The quality of this starch, such as dissolve performance, viscosity and molding is similar to other starch from grain, wheat or rice. Due to these qualities, using of mango

seed starch instead of wheat starch will be made more profit because wheat starch which is important ingredient in many desserts are imported from another countries.

Using of cheaper mango seed starch is very interesting because the capacity of water in this starch is high and this quality is properly for making bread. From the studies show that the participants cannot told the different, such as taste, smell or crispy, between crisp bread using the mango seed starch 50% and crisp bread using wheat starch 100%. Moreover, the nutrients of these breads are very similar as it shows in table 2.2 (Arogba, 1999).

Table 2.2 Compositions of mango seed kernel flour, wheat flour, mix flour (mango seed kernel:wheat 50:50) and biscuits made with mix flour (Arogba,1999)

Compositions (g/kg)	Type of flour			Biscuits made with mix flour
	Mango seed kernel	Wheat flour	Mix flour	
Moisture	91.0	136.8	122.9	77.1
Protein	66.1	134.0	93.0	68.1
Fatty	94.0	10.0	65.1	182.0
Fiber	28.0	3.8	13.8	10.3
Soluble carbohydrate (as Sucrose)	27.5	31.9	57.0	179.0
Flour	500.0	657.0	594.7	336.1
Carbohydrate	182.6	20.4	46.3	136.5
Calories (kJ/kg)	16.6	14.5	15.4	18.9
pH	4.5	4.4	4.4	4.4
Acidity (against citric acid)	17.5	7	10.5	10.5
Ascorbic acid	0.17	0.08	0.09	0.00
Tannin (against cathechin)	23.4	0.6	15.1	1.3

For some types of starch with low nutritional value for example cassava starch, corn starch, millet starch, the mixture with mango seed kernel starch will increase the amount of fat and protein of these dough (Arogba, 1997). The high amount of carbohydrates make this starch suitable to be used as substrate in the digestion by α-amylase and glucoamylase for produce glucose syrub (Velan, 1995).

Although the amount of protein in mango seed kernel starch is relatively lower than protein in other general consumtion starch (Table 2.2), but it is importance as a source of protein that contained high amino acid in ratio 32% of all amino acid which higher than FAO/WHO standard (Table 2.3) (Abdalla, 2007) except methionine, threonine and tyrosine.

Table 2.3 The compositions of amino acid of mango seed kernel starch (mean ± SD, n=4) compare with standard protein that specified by FAO/WHO (g amino acid/100 g protein) (Abdalla, 2007)

Amino acid	**Mango Seed Kernel starch**	**FAO/WHO Standard**
Leucine	6.9 ± 0.2	4.8
Isoleucine	4.4 ± 0.2	4.2
Methionine	1.2 ± 0.1	2.2
Phenylalanine	3.4 ± 0.1	2.8
Lysine	4.3 ± 0.2	4.2
Threonine	3.4 ± 0.2	4
Tyrosine	2.7 ± 0.1	4.1
Valine	5.8 ± 0.3	4.2
All necessary amino acid 32.1 ± 2.2		
Aspartic acid	6.5 ± 0.4	
Glutamic acid	18.2 ± 0.8	
Serine	3.3 ± 0.1	
Proline	3.5 ± 0.2	
Glycine	4.0 ± 0.4	
Alanine	4.2 ± 0.4	
Histidine	5.5 ± 0.6	
Arginine	7.3 ± 0.8	
All necessary amino acid 52.2 ± 2.1		

The mango seed oil has the potential to produce in factory scale because it is contained fat around 11-18%. This is higher than other kinds of oil which is produced in factory, such as corn, soybean and cotton seed which has fat around 5%, 18% and 15%, respectively (Gunstone, 2002).

The process of oil refining is not complicated and low costs because mango seed oil is tolerance to oxidation reaction and has a low amount of lipase and low acidity. In contrast, vegetable oil which is produced from tropical countries is contained long chain fatty acid and fatty acid, such as lauric acid that high in coconut oil or palmitic acid in palm oil.

However, mango seed oil has high nutrition and different from vegetable oil because it has important fatty acids which are oleic acid and linoleic acid and has high amount as olive oil (Table 2.4). In addition, antioxidants, such as phospholipids and phytosterol (sualene sterol and tocopherol around 0.34%), can be found in this oil. It is higher amount than other oil which has phytosterol around 0.1-0.5% (Lercker, 2000).

The component of tri-glyceride in mango seed oil is similar to rarity oil which is cocoa butter. In case of type of fatty acid and position of fatty acid in tri-glyceride composition as it shows in table 2.4 (Gunstone, 1997).

Table 2.4 The amount of fatty acid (%) of mango seed kernel oils, cocoa butter and olive oils (Gunstone, 1997)

Types of fatty acid	**Amount of fatty acid (%) of oils**		
	Mango Seed Kernel	**Cocoa butter**	**Olive oils**
Myristic acid (C14:0)	0.5 ±0.1	-	0.0-1.5
Palmitic acid (C16:0)	5.8 ±0.3	23-26	7.5-20
Steric acid (C18:0)	38.3±1.2	35-37	0.5-5.0
Oleic acid (C18:1)	46.1 ±2.3	34-37	55.0-83.0
Linoleic acid (C18:2)	8.2 ±2.3	1-3	3.5-21.0
Linolenic acid (C18:3)	1.2 ±0.2	-	0.0-1.5

Cocoa butter which is important ingredient for making chocolates has one special characteristic that hardly found in other kinds of oil. The melting point of this oil is around 35^{o}C that can easily melt in human body.

The reason of this characteristic comes from the composition of palmitic acid, steric acid and oleic acid in optimal ratio. Moreover, the position of olaic acid in tri-glyceride composition is sn-2 chain.

Therefore, mango seed oil is one of some kinds of oils that have palmitric acid, steric acid and oleic acid in sn-2 position. It is very similar to the cocoa butter. Due to this factor, it is easy to use this oil instead of cocoa butter and very difficult to examine.

In these days, African countries are the only exporter of cocoa butter and the products is also depends on the weather. Therefore, the price of cocoa butter is very high. As a result, using of mango seed oil is very interesting and challenging in other counties that cannot make cocoa butter, such as Thailand. It also reduces cost for making chocolate.

In Asia, the using of mango seed oil in factory scale can easily applied in India. The product of these factories is shortening which is ingredient for making bread, crisp bread and cake. It is contained both fatty acid and long chain acid, such as palmitic acid and steric acid. However, it has drawback from hydrogenation process. This process can make high amount of trans fatty acid around 20-40%.

Medical research shows the trans fatty acid is major cause of heart diseases. It can reduce the quantity of HDL fat. In contrast, the quantity of LDL fat is increase. Consuming of trans fatty acid in 4% of total calories will increase this negative fat in blood.

Due to this flaw, mango seed oil that has high steric acid is the possible way to make free trans fatty acid shortening.

The important process of shortening by using mango seed oil is extraction of steric acid in solvent. This free trans fatty acid composition can able to mixture in shortening processes more than 55%. However, this shortening will have higher cost than the shortening that make from hydrogenation. The benefit of this shortening are safety and healthy for the consumer (Reddy, 2001).

The taste and food safety are the fact that need to consider for consuming of mango seed. Normally, the mango seed kernel is sourness because tannin in this seed. However, tannin component is easily to dissolve in water or steaming. These methods can reduce the amount of tannin around 48%. In addition, the consuming of the mango seed kernel is very safety because human consume this food since ancient times. Moreover, science research shows the normal weight people (70 kilograms) can consume the mango seed kernel 400 grams or mango seed starch 800 grams without harm.

2.7 The adaption of product which is not food (Inprakhon, 2009).

2.7.1 Natural preservatives.

Mango seed kernel is fully with antioxidant substance class Polyphenols such as tannin which can easily disintegrate by water (hydrolysable tannin) and Gallotannin which hardly disintegrate by water (condensed tannin). In the other hand proanthocyanidin and phenolic such as gallic acid, ellagic acid and epicatechin (Figure 2.1) (Arogba, 2000; Puravankara, 2000).

Gallic acid which found in mango seed kernel has about 23 – 838 mg/100 grams of dried weight and ellagic acid has about 3 – 126 mg/100 gram depends on extraction method, mango's gene and source. Gallic acid and ellagic acid may found as free substance or compound which create ester bond with sugar in terms of gallotannin (Masibo, 2008).

Polyphenol and phenolic are both strange in antioxidant and stop activities of microorganism (Ahmed, 2006). Therefore, the substance which extract from mango seed will be both antioxidants and bio preservatives. Moreover, the oil from mango seed kernel found phospholipid substance more 8 types; phosphatidyl serine, lyso-phosphatidyle choline, phosphatidylinositol, sphingomyelin, phosphatidyl choline, phosphatidyl etanolamine, phosphatidic acid and a compound substance of glycerol-phosphatidyl which has an antioxidant abilities as well.

In India, the extraction experiment of mango seed kernel both has phenolic and phospholipid substance which were the compound in buffalo gee. According to the research found out that in 5% of substance has a better preserved ability than

butlylated hydroxyanisole (BHA) which allowed to add 0.02% by w/v. The extract substance from mango's seed 5% can extended a buffalo gee for 928 hours, by the way BHA 0.02% can extended only 210.6 hours (Puravankara, 2000).

In many country used the extract substance from mango's seed as the preservative substance. In Japan allowed ellagic acid as the preservative substance in food and gallo tannin classified as a safe additive in food (GRAS; generally recognized as safe) because the gallo tannin can degraded easily. The research by extract polyphenol substance with 95% alcohol got a compound of polyphenol substance about 80%. This extract substance got the abilities to stop the grown of pathogen in both gram plus and minus group totally 43 genes which MIC (minimum inhibitory concentrations) from less than 50 to 2500 ppm but against the grown of gram plus bacteria better than gram minus bacteria. So, the usage of extract substance from mango's seed for preserved is necessary to use with other preservative substance to increase the abilities to stop the grown of gram minus bacteria.

2.8 Applied using mango seed kernel in cosmetics

The mango seed kernel can use in cosmetics that help slow down the aging process because it is contained the moisture oil such as polyphenol and phenolic which are the strong antioxidant.

The oil from mango seed is contained oleic acid. When this component mixes in cosmetics, it can provide the moisture to skin as well as cocoa butter and olive oil. Furthermore, using this seed as the raw material of soap is possible because it has high steric acid and the surface of this soap is hard and shiny. It is the same appearances as soap that makes from palm oil but this soap which has high oleic acid can make small soap bubble, more purify, and cream state. All of these differences reduce irritated to our skin more than soap that make from palmitic acid.

In fact, free radicles are occurrence all the times from pollution and stress. These free radicles can damage cells and modify the chemical compound of biomolecule, such as protein, fat, nucleic acid. It can cause chronic effect to human body and tissues, such as amnesia, coronary thrombosis, more aging skin or skin losing moisture (Gutterridge, 1996).

Strong antioxidant which can found in mango seed can easily make reaction with free radicles. Therefore, the cosmetic which is contained mango seed can help slow down the aging as skincare (Fox, 1997).

In addition, it can use as supplementary food to increase immune system and prevent diseases that comes from damage of free radicles, such as amnesia, heart disease or cancer.

Snow Brand Milk Products, Japan has licensed the extraction of mango seed for using in their products to prevent the growth of *Propionibacterium acnes* which can cause acnes and *Streptococcus sanguinis* which can found in tooth plaque. Therefore, soap, shampoo and lotion cream that are contained mango seed can prevent and cure acnes. Moreover, the supplementary food and mouthwash that have mango seed as one of the component can prevent tooth plaque. The optimal ratio in these products is 0.001-5% (Nakajima, 2000).

The using of this extraction in products has to consider the stability of the extraction after the processes because it can lose stability when in high temperature or very low-high pH. The study of stability of mango seed shows that the mango seed kernel can use in the processes of food products and cosmetics (pH 3-9, temperature -20 – 121 ^{o}C)

2.9 Applied using mango seed kernel in Animal foods

The mango seed kernel is contained with starch protein fat and polyphenol. This can use as main food source and increase immune system in livestock. There are many experiment of using mango seed as food for small tilapia fish *(Oreochromis niloticus)* (Omoregie, 1991), chicken (Joseph, 1997) or chick (Ravindran, 1996).

The result of these experiments shows the optimal quantity for using raw mango seed is 10% and cooked mango seed is 20% as main food sources. The other research is mixing the mango seed kernel in food in order to increase immune system in high density aquatic animal's livestock. This type of livestock has the problem from diseases and has to use high amount of antibiotics. These high amount antibiotics can leftover in food and environment. This is unhealthy and unacceptable from consumers.

Due to this reason, the using of biological antibiotics, such as probiotics, chemical from plants or herbal plants are more acceptable.

The mango seed kernel has many benefits. It is cheap because it is waste. It also has the capability to inhibit the growth of bacteria, fungus and virus. In addition, it can reduce inflame, cure cholera and prevent tumor. From the experiment of mixing 0.5% of mango seed kernel in rohu fish (*Labeo rohita*) food for 60 days.

The result illustrates the immune system (innate response) in the fish's blood is increase by creating superoxide anion and lysosome which has capability to destroy the bacteria, as well as, the amount of albumin protein and globurin in serum is increase.

When rohu fish was receive *Aeomonas hydrophilia*, this fish will die by motile aeromonas disease. However, rohu fish which has 0.5% of mango seed kernel in food after receive this bacteria for 10 days. The surviving rate is higher than 98%. Consequently, the mango seed kernel is more suitable for mixing in rohu fish food to increase immune system instead of using antibiotics. The benefit of this seed is cheap, degradable and good for environment and consumers.

2.10 The information of Grape seed

Grape in scientific name is *Vitis vinifera*. Grape is the subtropical fruit which mostly product in warm climate country. Around the year 1956, Professor Pavin Punsri and his researcher team of Department of Horticulture, Kasetsart University, Bangkok, studied about the grape culture and developed grape production in Thailand until the grape production in Thailand has been a success at that time. The initial commercial grape production areas were located in the Central Plain region of Thailand at Nakhon Pathom, Ratchaburi, Samut Sakhon and Samut Songkhram provinces, which are near Bangkok. Nowadays, the grape industry has expanded to the Northern, Northeastern and the Western regions of Thailand (Nilnond, 2001).

In the present, antioxidant compounds are commonly used as a supplement. The most widely known was the grape seed extracts that quite expensive. The compounds in grape seed extracts were rich in various kinds of phenolic compounds as reported

that the phenolic compounds can inhibit several activities such as anti-oxidation, inhibit inflammation, inhibit cancer, anti-microbial and so on (Lee, 2004; Evans, 1997).

About 80% of grapes are used in wine manufactures and the byproduct of grape in the winery process consisted 20% of whole weight (Samavardhana, 2015; Maier, 2009; Lafka, 2007). In Thailand, grapes are usually processed into various products for examples, wine juice raisin through the ingredient in food and bakery. The byproducts or waste from grape processed such as the pomace including grape pulp, peels and seeds were obtained in large quantity. There are many studies that showing the valuable of grape byproducts which has many useful substances. Studies have found that grape seed could be natural source of antioxidant and antimicrobial agents.

Typically, the compositions of grape seeds (w/w) consists of 40% Fiber, 16% oil, 11% protein and 7% complex phenolic compounds as tannin, sugar, minerals and others compounds (Campos, 2008). And the grape peels are also the source of anthocyanidins, anthocyanins and natural pigments which have an anti-oxidation activity by inhibiting the oxidation of oils and can be inhibited by the mutation (Pedreschi & Cisneros-Zevallos, 2006). Ribereau-Gayon (2000) found that about 60 – 70% of phenolic compounds has contained in grape seeds, including catechin epicatechin and procyanidin were found to be the major antioxidant (Maier, 2009; Katalinic´, 2010; Chedea, 2010).