

## **CHAPTER III**

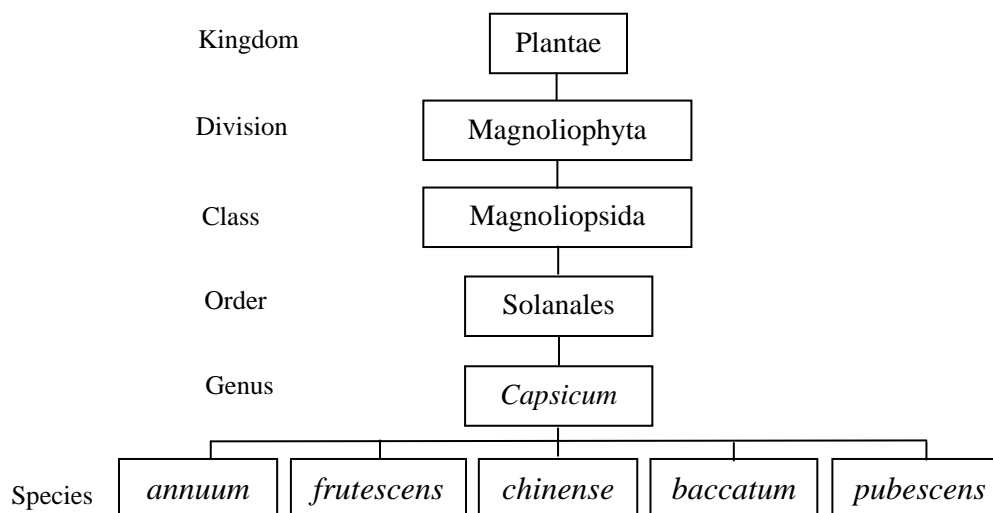
### **LITERATURE REVIEW**

#### **3.1 Chili and It Characterizations**

Chili is a plant in the *Capsicum* genus and Solanaceae family. The origin of the chili cultivation is in America; however, as time passes, it is cultivated around the world. Chili is known under different names in different countries. It is an annual crop, which can grow up to 2 meters in height. Chili normally grows in tropical area with sandy loam and optimum pH of 5.5-6.5. The main characteristic of chili is its spicy favor, which caused by a specific compound in chili called capsaicin. This compound is located in the area of placenta of the fruit. The colors of immature fruits are light yellow, light green, dark green and purple, depending on the particular type of chili. However, the ripping (mature) stage may change fruit colors into red, orange, yellow, brown or purple.

##### **3.1.1 Types of Chili**

Chili is a plant in Solanaceae family. The taxonomic hierarchy of chili can be classified into the genus of *Capsicum* with five domesticated species that can grow around the world, including *C. annuum*, *C. frutescens*, *C. chinense*, *C. baccatum* and *C. pubescens* (Figure 3.1-3.4). The characteristics of these species can be differentiated based on fruit colors, flowers and shapes of calyx.



**Figure 3.1** The scheme shows the taxonomic classification of chilies, which eventually can be divided into five species including *C. annuum*, *C. frutescens*, *C. chinense*, *C. baccatum* and *C. pubescens*.

The species of *C. annuum* is originally cultivated in Central America and is the most recognized commercially available chili in market nowadays. The plants of *C. annuum* have different flowering and fruiting periods from other species. The flower is pure white, and only one flower per node. The corolla is white without spots, and the pedicels decline at anthesis. The seeds are white or cream to yellow, thin and circular. The calyx of mature fruit does not have annular constriction at junction with pedicel. The fruits are variable in size, shape and pungency. Mature fruits have various colors such as green, purple, yellow, red or orange. This species involves commonly consumed sweet peppers and hot chilies. Raw sweet pepper is non-pungent and can be consumed in salads as fresh vegetable or cooked in many dishes. Pungent *C. annuum* is used as spice for seasoning. Common names of *C. annuum* are chili pepper, bell pepper, paprika, Christmas pepper, cayenne, halapenos and chitlepin.

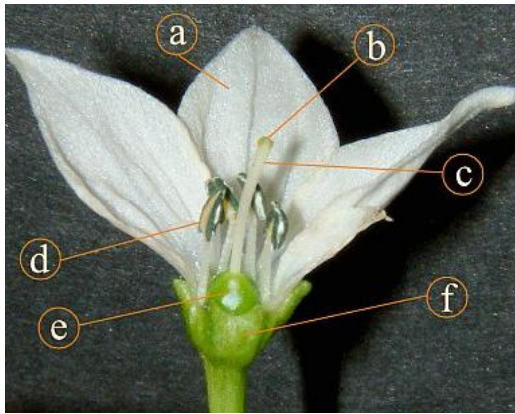
The species of *C. frutescens* is originated in South America and spread quickly throughout the world. This species possesses greenish white, two or more flowers per node. The corolla is greenish white without spots. The pedicels erect at anthesis but flowers nodding. The seeds are creamy to yellow. The calyx of mature fruit does not have annular constriction at junction with pedicel. The fruits have pale

yellow, red, red-orange colors. This species is commonly used to make flavoring in food due to their hot and spicy taste and sometimes is made into chili powder. Common name of *C. frutescens* are bird pepper, hot pepper and tabasco pepper.

The species of *C. chinense* is native to South America and Amazon. The flower characteristics of this species are greenish white color with two or more flowers per node. The corolla is greenish white without spots. The pedicels erect or decline at anthesis, while the calyx of mature fruit has annular constriction at junction with pedicel. The fruit is varied greatly in colors (red, yellow and orange) and shapes. This species provides the hottest chilies such as fatalii habanero, billy goat, rocotillo and scotch bonnet.

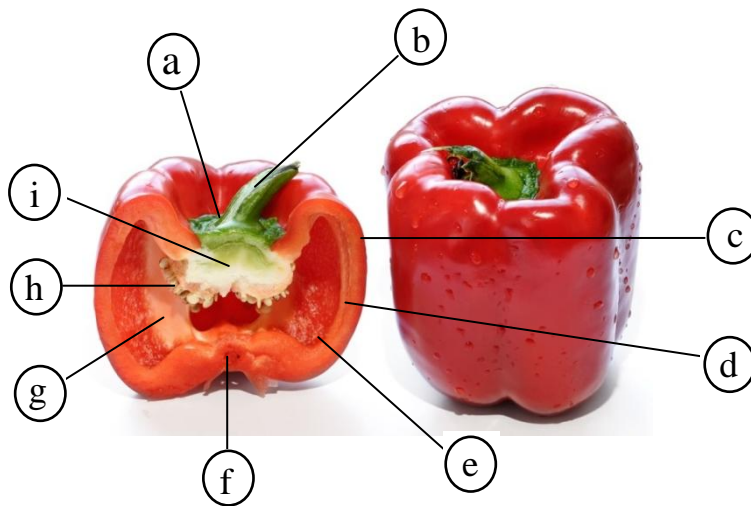
The species of *C. baccatum* is originated in Bolivia and spread quickly through Peru, Argentina and South America. The characteristics consist of white or cream colored flowers, one flower per node and corolla with yellow spots. The pedicels erect or decline at anthesis. The seeds are cream, and the calyx of mature fruit does not have annular constriction at junction with pedicel. The fruits are in green, yellow to red color. *C. baccatum* species are unlike other species that have a wide variety of shapes and sizes. It is distinctive not only use as a hot spice, but also use for decorate bouquet.

The species of *C. pubescens* is primary cultivated in the region of Central and South America. This species is greatly different from domesticated chilies. The characteristic of this species is morphology with purple flowers, one flower per node, furry leaves and black or dark brown seeds. The corolla is purple without spots. The pedicels erect at anthesis but flowers nodding. The calyx of mature fruit does not have annular constriction at junction with pedicel. The fruits have oval shaped like small apples and green, red, yellow, orange and dark brown colors.



(A) Parts of flower

- a = Petal
- b = Stigma
- c = Pistil
- d = Stamens
- e = Sepal
- f = Calyx

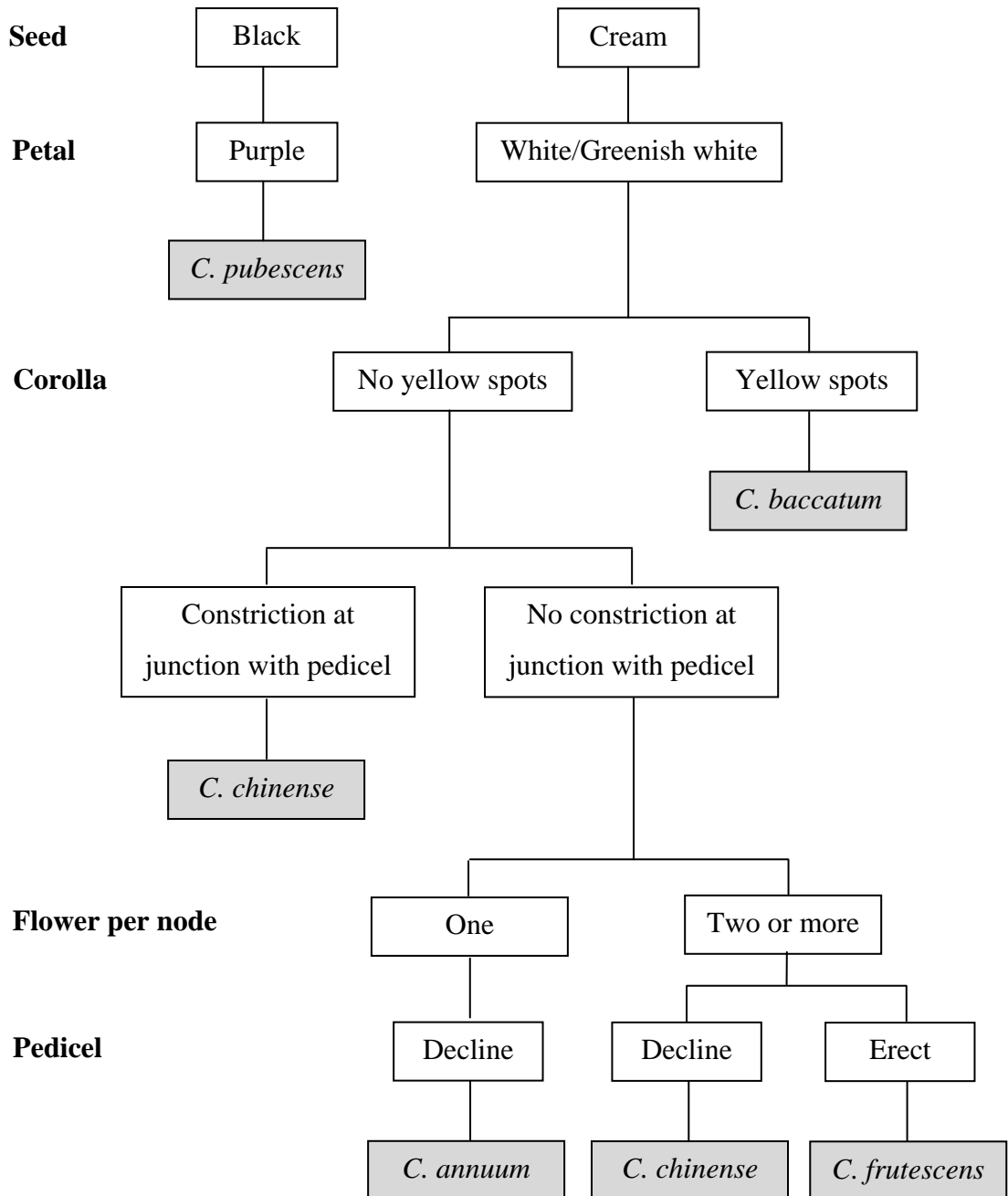


(B) Parts of fruit

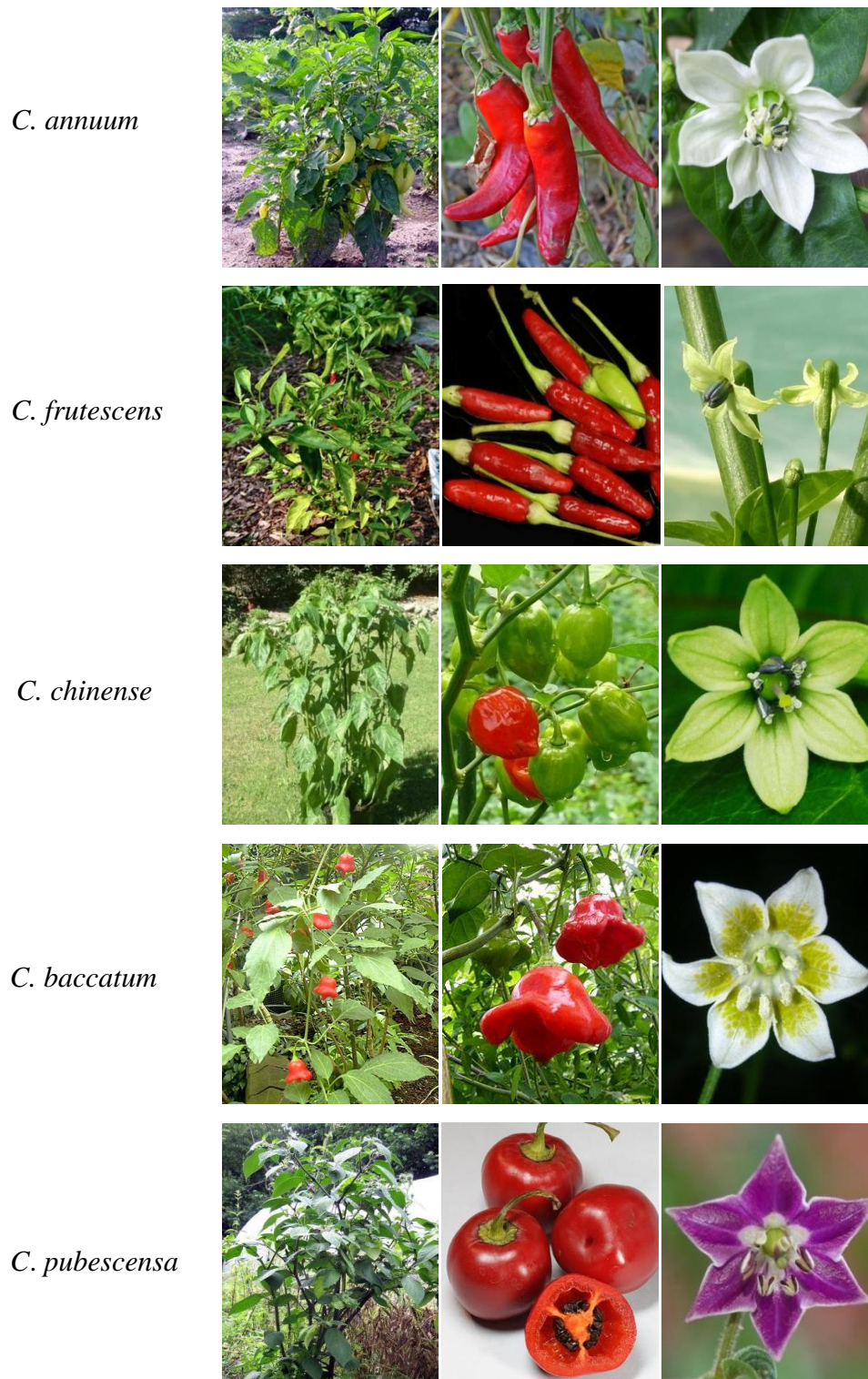
- a = Calyx
- b = Peduncle
- c = Exocarp
- d = Mesocarp
- e = Endocarp
- f = Apex
- g = Capsaicin gland
- h = Seeds
- i = Placenta

**Figure 3.2** The schemes showed (A) parts of flower and (B) parts of fruit to differentiate the species of chili.

### Classification of Chili



**Figure 3.3** The scheme used to classify the species of chili according to their particular physical properties, which eventually can be divided into *C. annuum*, *C. frutescens*, *C. chinense*, *C. baccatum* and *C. pubescens*.



**Figure 3.4** Whole plants, fruits and flowers of five *Capsicum* species including *C. annuum*, *C. frutescens*, *C. chinense*, *C. baccatum* and *C. pubescens*.

The chilies that are commonly cultivated in Thailand consist of two main species including *C. annuum* and *C. frutescens*. These species are consumed both in mature and immature stages. The maturity of chilies can be recognized as fruit color change. The fruit colors of *C. annuum* and *C. frutescens* are found in green, yellow, orange, red and purple. *C. annuum* cultivated in Thailand is included prik-chi-fa, prik-jin-da, prik-wan, prik-yuak and prik-yak. The other species, *C. frutescens*, is prik-khenu-yao and prik-khenu-pol-yai.

### 3.1.2 Phytochemical Compositions

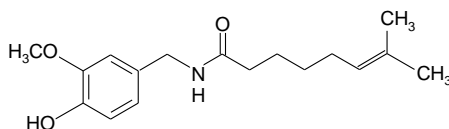
Phytochemicals are bioactive substances that commonly found in plants. These substances cause specific color, odor and taste, thus leading to different characterization in each particular plant. Phytochemicals found in chili are carotenoid, vitamin C, capsaicinoid and flavonoids (Table 3.1) [14]. Some of these compounds exhibit antioxidant activities, thus controlling oxidative stress related diseases such as cancer, inflammation and metabolic syndrome.

**Table 3.1** Phytochemical compositions in chilies (*C. annuum*, *C. frutescens*, *C. chinense* and *C. baccatum*)

<i>Capsicum</i> spp.	Capsaicinoid (mg/100g)	Ascorbic acid (mg/100g)	Total polyphenols (g GAE/100g)	Total flavonoids (mg/100g)
<i>C. frutescens</i>	1560	trace	1.5-2.0	trace
<i>C. annuum</i>	809	25	1.5-2.0	30
<i>C. baccatum</i>	712	60	1.5-2.0	27
<i>C. chinense</i>	1412	295	3.7	28

Chili is reported to contain many nutritional compositions, including macrominerals (Ca, P, Na, Mg and K), microminerals (B, Cu, Mn, Zn and Fe), carbohydrate, protein and lipid [15, 16]. Many researches suggested that the quantity and quality of phytochemicals and antioxidants in chilies are change during maturity [9, 17-19]. The maturity stages of chili could be observed as fruit color changing from immature (green) to mature (red, orange or yellow). As results, phytochemicals and

antioxidant activities of mature chilies are higher than those of immature ones [4, 17, 18]. Besides, substance that makes chilies superior to other plants is capsaicinoid (Figure 3.5). Capsaicinoid is a phenolic compound that causes chili to have spicy and pungent favors. Capsaicin is one of the major capsaicinoid that can be found in chilies, followed by dihydrocapsaicin. Previous research was reported that the highest levels of capsaicinoids, flavonoids and total phenolics were found in chilies extracted under hexane, methanol and ethyl acetate, respectively [20], suggesting that bioactive compounds in chilies likely dissolved in solvent with low polarity index. The data was corresponded to the chemical structure of capsaicinoid, which contains aromatic moiety and long hydrophobic side chain.



**Figure 3.5** The chemical structure of capsaicinoid found in the area of chili placenta. This compound is in a group of phenolics, which causes spicy and pungent favors in chili.

### 3.1.3 Commercial Applications

Commonly, chili is used as a food ingredient to add spicy and as a food decoration due to their varieties of fruit colors (green, yellow, orange, red, purple and etc.). Nowadays, many new products of chili such as chili jam and chili oral spray are also available in market. Chili jam is a product that developed by reducing the pungency of chili with sugar solution. Oral chili spray is a stimulating product, which used to spray in the mouth to be alert (sleep protection). In addition, chili has also used to produce snack. Chili rice cracker is a snack that is produced from sweet chili and rice. The flavor of cracker is blended with a balance of sweet, spicy and crunchy sensation. Moreover, coconut water with chili extract is produced as a beverage. Besides, coco chili oil is natural edible oil that combines natural virgin coconut oil with red chili. Coco chili oil is used for cooking and salad dressing. In addition, chili gel and capsaicin cream are invented for pain relief. Chili cream has produced as

beauty product, which claimed to promote circulation metabolism, moisture, shrinkage in order to achieve weight loss.

### **3.2 Chili and Health Benefits**

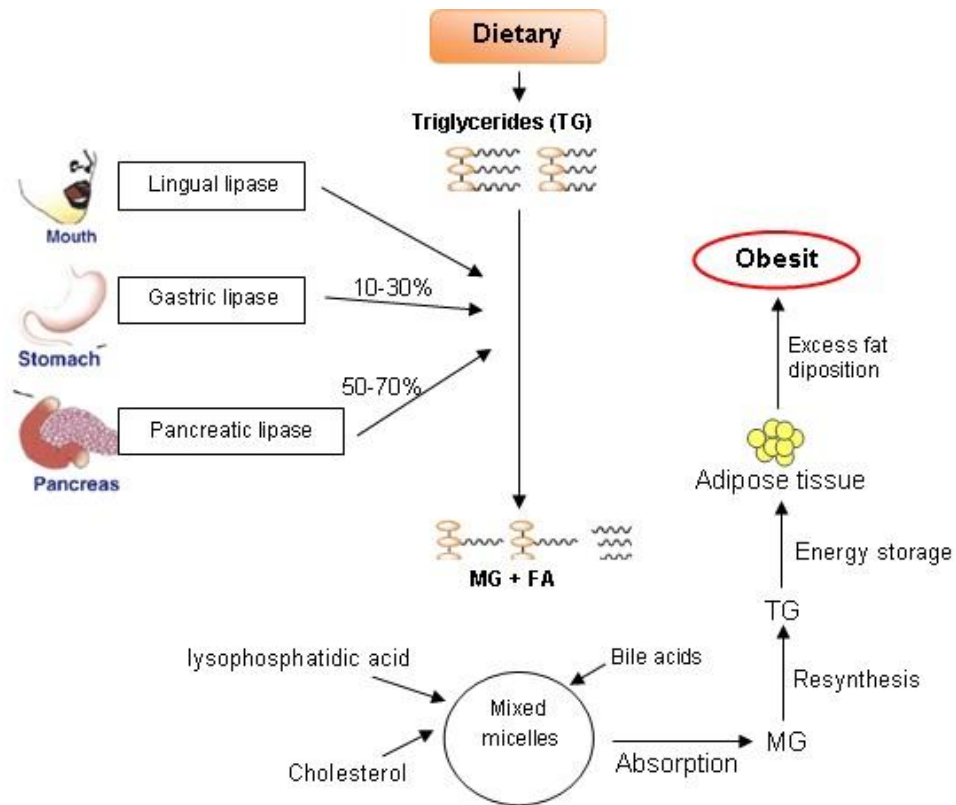
Chili has been studied regarding its health promotions on obesity, diabetes, cancer and atherosclerosis. Chili composes of important substances that promote health benefits, which can reduce risk of some chronic diseases such as atherosclerosis, cancer, diabetes and cardiovascular disease. In this study, the investigation of biochemical properties against obesity, diabetes and hypertension of crude extract from chilies through inhibition of key enzymes that control those particular diseases will be focused.

Fat and sugar are two significant factors that cause obesity, thus the inhibition of fat and sugar digestion is of interest in this research. Controlling of fat digestion involves lipase, the enzyme that hydrolyzes triglyceride into free fatty acids and glycerol before being absorbed into body. Lipase inhibition from plant extracts and lipase inhibitors from natural sources have been widely investigated for potential anti-obesity treatment and prevention. Sugar digestion, on the other hand, is controlled by two crucial enzymes,  $\alpha$ -amylase and  $\alpha$ -glucosidase. These enzymes can degrade carbohydrate to glucose, which is further absorbed into body. Overload of sugar consumption not only causes obesity, but also diabetes, the 6<sup>th</sup> cause of death among Thai public health problems in 2012 [21, 22]. Thus, the inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase is important to prevent carbohydrate digestion, leading to controlling of obese and diabetic occurrences. Besides, being the 8<sup>th</sup> mortal cause of Thais [21, 22], hypertension is controlled by rennin-angiotensin-aldosterone system (RAAS), a group of hormone homeostasis that regulates blood pressure. Occurrence of hypertension involves angiotensin-converting enzyme (ACE), which converts angiotensin I into angiotensin II, leading to an increase in blood pressure. Thus, inhibition of ACE would lead to reduction of hypertensive risk.

### 3.2.1 Anti-obesitic Property

Obesity is a major public health problem that is continually increased in Thailand due to change of lifestyle. The criteria used to predict obesity is Body Mass Index (BMI). The BMI standard for normal individual is 18.5-24.9, while ones with BMI over 30 are obese (World health organization: WHO, 1995). Obesity affects on body systems and is associated with many diseases such as coronary heart disease, gallbladder disease, cirrhosis, cancer, gout and stroke. The causes of obesity include genetics, environmental factors, social lifestyle, illnesses, and medications. Genetic traits inherited from parents may cause obesity such as having a slow metabolism. Human lifestyle can affect obesity such as eating fast food high in fat, drinking too much alcohol, not eating fruits and vegetables. Certain medicines such as corticosteroids, antidepressants, and seizure medicines can cause individual to gain weight. These medicines can slow down metabolism and increase appetite.

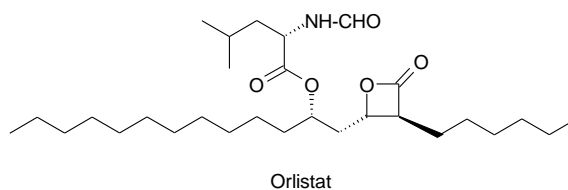
One of several causes on obesity occurrence is overloaded lipid consumption [7]. Lipid is a major nutrient that provides energy for body activity. One gram of lipid provides 9 kilocalories of energy, the highest energy provider among all nutrients. The major human lipases comprise of lingual, gastric and pancreatic lipase (Figure 3.6). Pancreatic lipase hydrolyzes approx. 50–70% of total dietary fat, while gastric lipase exhibits approx. 10–30% digestion. Lingual lipase, on the other hand, exhibits trace activity. The digestion of lipids starts in oral cavity by lingual lipase and continues to stomach by gastric lipase. The lipids are emulsified by bile acids and mixed with pancreatic lipase for hydrolysis. Most dietary lipids are composed of triglycerides (TGs), which are hydrolyzed to fatty acids (FAs) and monoglycerides (MGs). Then, free FAs and MGs form micelles by mixing with bile salts, cholesterol and lysophosphatidic acid (LPA) before being absorbed into enterocytes. Triglycerides are re-formed and stored in adipocytes as main energy source. Thus, eating too much food and lack of exercise will cause excess fat deposition, leading to obesity. Since lipase is an important enzyme that hydrolyzes TGs into glycerol and free FAs, lipase inhibition could decrease rate of fat adsorption.



**Figure 3.6** The scheme showed lipid digestion and absorption. Dietary lipids are composed of triglycerides (TGs), which are hydrolyzed by lipases for fat absorption. Pancreatic lipase hydrolyzes 50–70% of total dietary lipid into free fatty acids (FA) and monoglycerides (MGs). The free FAs and MGs form mixed micelles with bile salts, cholesterol and lysophosphatidic acid (LPA), which eventually absorbed into enterocytes. These micelles act as sources of fat components for the resynthesis of TGs, which are stored in adipocytes as the main energy source.

Currently, numerous options are available for treatment of obesity including lifestyle therapy, pharmacological interventions and surgical operation. The medicinal treatment involves the only commercially available synthesized drug, orlistat, which controls obesity through inhibition of lipase reaction. Orlistat is a semi-synthetic hydrogen derivative (Figure 3.7) that could be isolated from *Streptomyces toxytricini*, a Gram-positive bacterium found mostly in soil. Orlistat could form interactions with a catalytic residue in the active site of lipase, forming the stable enzyme complex and inactivating the enzyme reaction. When this occurred, lipids

cannot enter the catalytic pocket of lipase, thus preventing lipid digestion and decreasing fatty acid absorption rate. Nevertheless, orlistat has harmful side effects, including liquid stool, flatulence, constipation nausea, vomiting and diarrhea. Thus, lipase inhibitors from natural sources that can prevent obesity without causing severe side effects are of interest.



**Figure 3.7** Chemical structure of orlistat, a reversible lipase inhibitor. This compound is able to inhibit lipase reaction by forming interaction with catalytic residue at the active site of lipase. Thus, orlistat can prevent binding of triglyceride and lipase, causing the decrease of fat absorption into body.

Previous studies had been reported many bioactive compounds from plants such as saponins, polyphenolics and terpenes that can inhibit lipase activity (Table 3.2) [7]. Saponins are components of root and rhizomes of various plants. These compounds contain sugars that attached to steroid or triterpene. Flavonoids such as flavones, flavonols and tannins are active against pancreatic lipase. Terpenes are widespread in nature, mainly in plants as constituents of essential oils. These compounds can inhibit lipase activity, thus reducing lipids absorption and preventing obesity.

**Table 3.2** Anti-lipase compounds from different plant sources

Anti-lipase compound	Plant source	Ref
EGCG	<i>Camellia Sinensis</i>	[23]
(-)-4'-Omethylepigallocatechin	<i>Salacia reticulate</i>	[24, 25]
Luteolin	<i>Arachis hypogaea</i> and <i>Eminium spiculatum</i>	[26]
3-Methyl ethergalangin	<i>Alpinia officinarum</i>	[27]
Platycodin saponin	<i>Platycodin grandiflorum</i>	[28]
Sessiloside and chiisanoside	<i>Accanthopanax sessiliflorus</i>	[29]
Teasaponin	<i>Camellia Sinensis</i>	[30]
Scabiosaponin	<i>Scabiosa tschiliensis</i>	[31]
Carnosic acid	<i>Salvia officinalis</i>	[32]
Crocin and crocetin	<i>Gardenia jasminoids</i>	[33]

The effects of chili on obesity have been continually received attention. The mechanisms of actions may be related to certain pathways such as energy balance, lipid accumulation and lipase inhibition. Previous research suggested that red pepper and caffeine consumption could decrease energy intake and increase energy expenditure [34]. A study in rabbits was suggested that chili consumption could reduce the body weight, possibly through the mechanism of capsaicin [35]. Capsaicin, the major phenolic compound in chili, can decrease triglycerides level in serum and reduce adipose tissue mass for weight loss and weight maintenance [36, 37]. Besides, red sweet pepper was investigated regarding its effect on the body fat and body weight in humans [38]. In this study, the subjects were received a repeated intake of sweet pepper for two weeks. The results suggested that intake of sweet pepper could reduce the body fat and body weight.

As for lipid accumulation perspective, it was suggested that chilies are capable of reducing fat accumulated into cells. Ethanolic extract of hot pepper (*Capsicum annum*) seeds were reported to possess anti-obesity through reduction of lipid accumulation in adipocyte [39]. Besides, kochujang (Korean fermented red pepper paste), a Korean sauce that is a mixture of fermented red pepper power, wheat

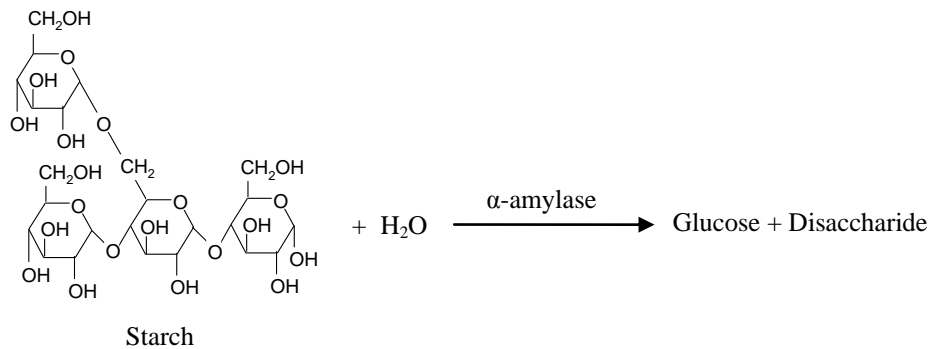
and soybeans, was reported to be able to reduce adipocyte size *in vitro* [40]. Its *in vivo* study also suggested that kochujang could reduce visceral fat [41].

For lipase inhibitory pathway, it was reported that chili is a rich source of bioactive compounds such as  $\beta$ -carotene, capsanthin, myricetin, quercetin and luteolin [3], which possess biological functions against lipase [7]. Myricetin could inhibit lipase with a half maximal inhibitory concentration ( $IC_{50}$ ) of 0.276 mM [42]. Quercetin and luteolin at the concentration of 25  $\mu$ g/mL exhibited approx. 27 and 17% inhibition against lipase [26]. Besides, capsaicin consumption could decrease plasma triglyceride levels in obese diabetic mice [43]. Nevertheless, lipase inhibition of capsaicin has not yet been reported.

### 3.2.2 Anti-diabetic Property

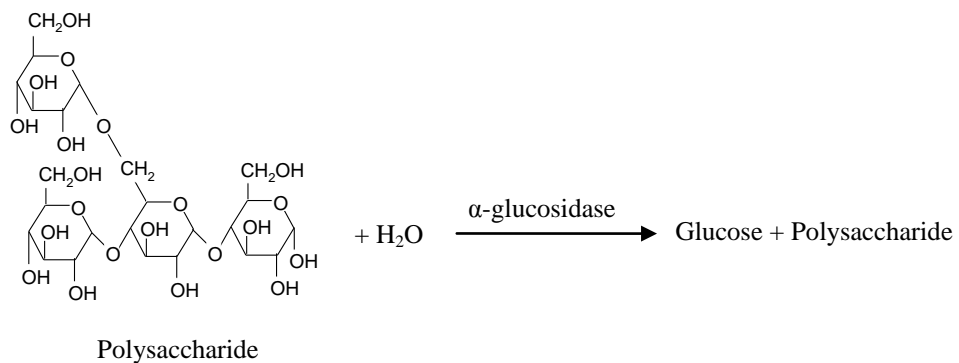
Diabetes is a chronic disease, in which body cannot produce enough insulin or respond to the insulin. In 2004, WHO reported that 376 million people were under diabetes worldwide, and it is continually increased every year (WHO, 2004). Diabetes is a public health problem, because it can cause acute or chronic complications that increased morbidity and mortality rates. Diabetes can be divided into two groups, type 1 and type 2. The former involves immune system, which destroys islet cells that produce insulin in pancreas. As a result, insulin production is terminated. The later involves the body that continues to produce insulin, but it does not function properly due to insulin resistance.

Diabetes can be prevented by reducing sugar absorption through inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase, the key enzymes that degrade carbohydrate into sugar. The  $\alpha$ -amylase is a digestive enzyme that found in saliva and pancreas. It hydrolyzes  $\alpha$ -1-4-glycosidic linkage on polysaccharide (starch), producing smaller sugar units such as disaccharide (maltose) and glucose (Figure 3.8). The digestion of starch is started in oral cavity by salivary  $\alpha$ -amylase and continued to a small intestine by pancreatic  $\alpha$ -amylase. The products of the reaction, glucose units, are absorbed into the wall of intestine. Glucoses are then delivered by blood to targeted cells throughout the body. Thus,  $\alpha$ -amylase inhibition is important for delaying carbohydrate degradation and glucose absorption.



**Figure 3.8** The function of  $\alpha$ -amylase, which hydrolyzed polysaccharide such as starch into smaller sugar units such as glucose and disaccharide, maltose.

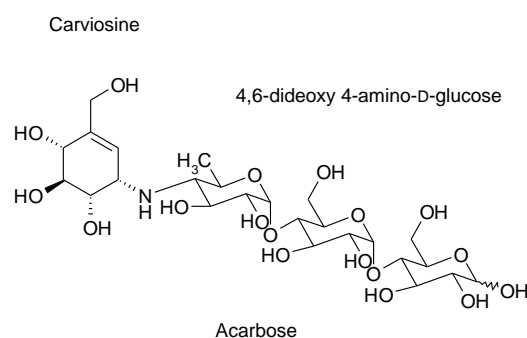
Similarly,  $\alpha$ -glucosidase can hydrolyze polysaccharide into glucose at terminal non-reducing  $\alpha$ -1-4-glycosidic linkage in small intestine, where the enzyme is located. The product from  $\alpha$ -glucosidase is monosaccharide such as glucose, which is absorbed into intestine wall (Figure 3.9). Thus, the inhibition of  $\alpha$ -glucosidase can, as well, control diabetes by delaying carbohydrate degradation and glucose absorption.



**Figure 3.9** The function of  $\alpha$ -glucosidase, which hydrolyzed polysaccharide such as starch into glucose and smaller polysaccharide.

The  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitors can treat hyperglycemia or diabetes by slowing carbohydrates degradation and glucose absorption. Currently, acarbose, a pseudo-oligosaccharide analogue, is used to treat diabetes through inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase [44]. Acarbose is composed of acarviosin moiety, which is an unsaturated cyclitol unit, and 4,6-dideoxy 4-amino-D-glucose at the terminal reducing end (Figure 3.10). The drug possesses the  $IC_{50}$  of 80 and 36

ug/mL for  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitions, respectively [45, 46]. However, acarbose has side effects, including flatulence and diarrhea. Thus, natural plant extracts that can inhibit  $\alpha$ -amylase or  $\alpha$ -glucosidase without or less side effects are of interest.



**Figure 3.10** The chemical structure of acarbose, which consists of acarviosin moiety and maltose at the reducing terminus.

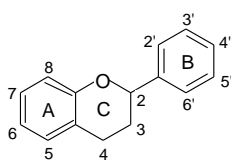
Various researches have been studied on the anti-diabetic property of plant extracts. The studies reported that plant extracts including tea (*Camellia sinensis*), guava (*Psidium guajava*), roselle (*Hibiscus sabdariffa*), apple (*Malus domestica*), chili (*Capsicum* spp.) and purple passion vine (*Gynura divaricata*) could inhibit  $\alpha$ -amylase and  $\alpha$ -glucosidase activity [9, 10, 47-51]. Bioactive compounds that have been reported for their biological functions against lipase  $\alpha$ -amylase and  $\alpha$ -glucosidase are polyphenolic, phenolics and flavonoids [52-56].

Chilies were reported to possess anti-obesity property (Table 3.3). Previous study suggested that the evaluation of *Capsicum annuum* had effect on diabetic enzyme activities, both  $\alpha$ -amylase and  $\alpha$ -glucosidase. It was suggested that mature (red) chilies exhibited higher  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activities than immature (green) ones [9]. Interestingly, these inhibitory activities were correlated to total phenolic content and antioxidant activity, suggesting that some phenolics could inhibit diabetic enzyme.

In addition, *C. chinense*, *C. pubescens* and *C. baccatum* could prevent hyperglycemia, in which their extracts were also found to exhibited similar  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activities [11]. The anti- $\alpha$ -glucosidase activities were

higher than anti- $\alpha$ -amylase activities in mature chilies. However, the opposite results were observed in immature ones.

Capsaicin, a main component in chilies, has a negative effect on plasma glucose level in humans [12]. It was found that capsaicin could decrease plasma glucose levels and maintain insulin levels. However, the directed inhibitory activity against  $\alpha$ -amylase and  $\alpha$ -glucosidase has not yet been reported. Other bioactive compounds such as myricetin, quercetin and luteolin were reported to exhibited  $\alpha$ -amylase inhibitory activity with the  $IC_{50}$  of 0.3-0.5 mM and  $\alpha$ -glucosidase inhibitory activities with the  $IC_{50}$  of 5-21  $\mu$ M [56]. The results suggest that these flavonoids could inhibit  $\alpha$ -amylase more efficiently than  $\alpha$ -glucosidase, which might be due to the structures of bioactive compounds that affect enzyme inhibitory activity. It was previously suggested that the enzyme inhibitory activity could be increased due to an increase of hydroxyl groups on phenyl group (B ring) (Figure 3.11) [56]. The increase of hydroxyl moiety in B ring will imitate the structure of the substrate of the enzyme, polysaccharide, thus making it a strong inhibitor. Besides, the hydroxyl groups at the 3 and 5 positions also improve inhibitory activity [56]. Nevertheless, the double bond between 2 and 3 positions on benzopyran group (C ring) decrease inhibitory activity. Flavonoids, for example, are competitive inhibitor on  $\alpha$ -amylase and act as non-competitive inhibitor on  $\alpha$ -glucosidase [57, 58].



**Figure 3.11** The basic structure of flavonoid found generally in fruits and vegetables. Flavonoids consist of three rings: A ring (benzopyran group), B ring (phenyl group) and C ring (benzopyran group). It can be divided into six groups, including anthocyanidin, flavones, isoflavone, flavonol, flavanone and flavan-3-ol depend on the variation on C ring and the linkage between C and B rings.

**Table 3.3** The summarized data on  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activity of chili extracts

Chili species	Chili types	Solvents	% inhibition			IC <sub>50</sub> (ug/mL)		Ref
			$\alpha$ -amylase	$\alpha$ -glucosidase	$\alpha$ -glucosidase	$\alpha$ -amylase	$\alpha$ -glucosidase	
<i>C. annuum</i>	*Green	DI water	No	40.5	-	-	[10]	
	*Red	DI water	No	14	-	-	[10]	
	*Orange	DI water	9.1	18.5	-	-	[10]	
	*Yellow	DI water	No	20	-	-	[10]	
	*Cubanelle	DI water	34.2	20	-	-	[10]	
	*Red sweet	DI water	75.6	48.5	-	-	[10]	
	*Yellow sweet	DI water	1	36	-	-	[10]	
	*Long hot	DI water	No	38	-	-	[10]	
	*Jalapeno	DI water	7.5	25	-	-	[10]	
	Green (PM)	Methanol	-	-	171.06	133.30	[59]	
	Green (M)	Methanol	-	-	55.88	76.11	[59]	
	Red (M)	Methanol	-	-	>500	142.71	[59]	

PM =Premature, M= Mature, I = Immature

\*Concentration of extracts = 1.5 g/mL

‡ Concentration of extracts = 0.05 g/mL

**Table 3.3** The summarized data on  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activity of chili extracts

Chili species	Chili types	Solvents	% inhibition		IC <sub>50</sub> (ug/mL)		Ref
			$\alpha$ -amylase	$\alpha$ -glucosidase	$\alpha$ -amylase	$\alpha$ -glucosidase	
<i>C. annuum</i>	‡Chili de arbol	DI water	11	31	-	-	[11]
	‡Chili ancho	DI water	5	32	-	-	[11]
	‡Japanese chili	DI water	35	48	-	-	[11]
	‡Paprika pepper	DI water	30	40	-	-	[11]
	Fiesta (I)	Ethanol	-	-	47.8	109.2	[60]
	Fiesta (M)	Ethanol	-	-	>1000	>1000	[60]
	Orange Thai (I)	Ethanol	-	-	98.7	105.2	[60]
	Orange Thai (M)	Ethanol	-	-	437.9	166.5	[60]
	Acuminatum (I)	Ethanol	-	-	>1000	>1000	[60]
	Acuminatum(M)	Ethanol	-	-	118.1	71.5	[60]
	Cayenne golden (I)	Ethanol	-	-	129.6	81.1	[60]
	Cayenne golden (M)	Ethanol	-	-	222.8	63.6	[60]

PM =Premature, M= Mature, I = Immature

\*Concentration of extracts = 1.5 g/mL

‡ Concentration of extracts = 0.05 g/mL

**Table 3.3** The summarized data on  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activity of chili extracts

Chili species	Chili types	Solvents	% inhibition			IC <sub>50</sub> (ug/mL)		Ref
			$\alpha$ -amylase	$\alpha$ -glucosidase	$\alpha$ -amylase	$\alpha$ -glucosidase		
<i>C. chinense</i>	Green (I)	Ethanol	-	-	229	150	[9]	
	Red (M)	Ethanol	-	-	131	265	[9]	
	<sup>‡</sup> Red pepper	Ethanol	31.5	55	-	-	[11]	
<i>C. baccatum</i>	<sup>‡</sup> Yellow pepper	DI water	32	50	-	-	[11]	
	<sup>‡</sup> Rocoto	DI water	28	55	-	-	[11]	
<b>Bioactive compounds</b>								
Myricetin		Ethanol						
Quercetin		Ethanol	-	-	151.12	2.12	[56]	
Luteolin		Ethanol	-	-	120.93	1.59	[56]	
<b>Medicine</b>			-	-	103.05	6	[56]	
Acarbose			-	-	80	36	[44, 45]	

PM =Premature, M= Mature, I = Immature

<sup>\*</sup>Concentration of extracts = 1.5 g/mL<sup>‡</sup> Concentration of extracts = 0.05 g/mL

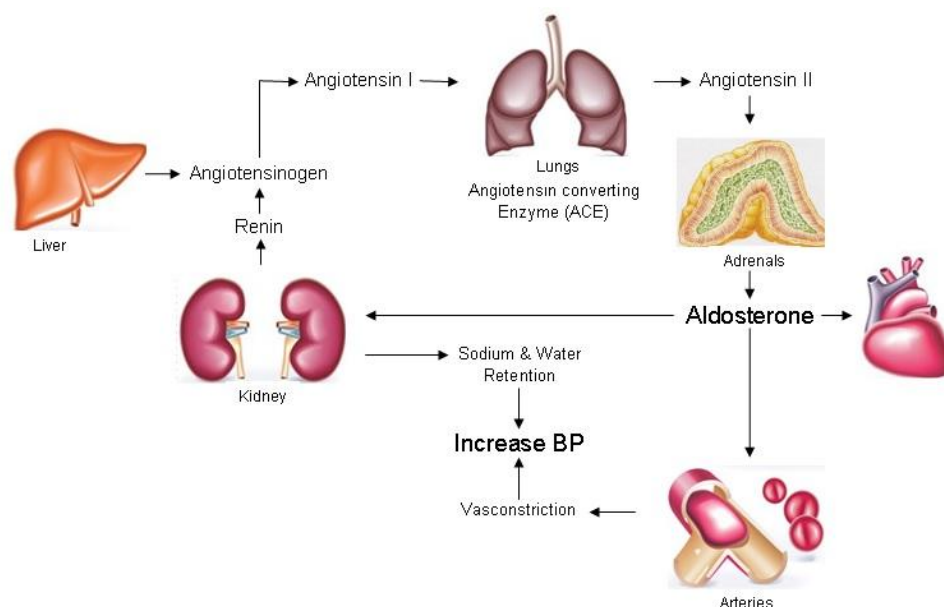
### 3.2.3 Anti-hypertensive Property

High blood pressure (hypertension) is caused by blood that is pumping with more force than normal through the arteries. Thus, the heart is work hard to pump blood through the arteries for circulation. The information from WHO as being determined in 1999 suggested that any individuals with the blood pressure higher than 140/90 mm Hg is considering as having hypertension. Hypertension is a major public health problem in Thailand and is ranked the 8<sup>th</sup> of Thai death from non-communicable diseases [15]. The Ministry of Public Health is reported that Thai individuals with expected high blood pressure are approx. 10 million in 2025. Hypertension increases the risk of many related diseases such as stroke, heart disease and renal failure.

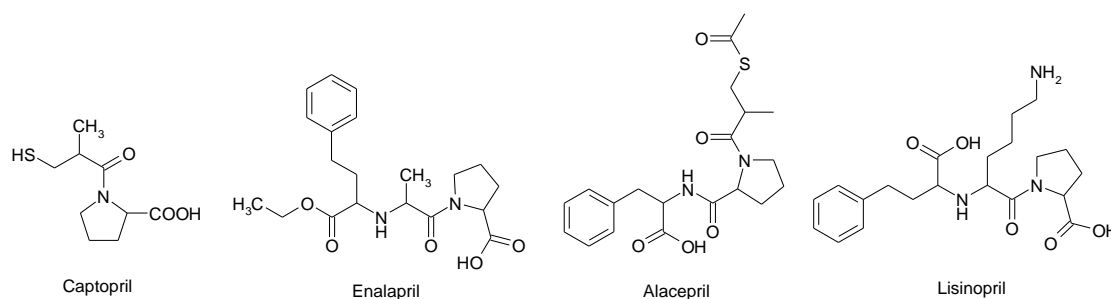
Hypertension is associated with rennin-angiotensin-aldosterone system (RAAS), a group of hormone homeostasis that controls blood pressure and water balance. When blood volume is low, renin, a kidney hormone, is secreted into blood circulation and converted liver angiotensinogen, a precursor hormone with 453 amino acids, into angiotensin I (AngI, Asp-Arg-Val-Tyr-Ile-His-Pro-Phe-His-Leu). This decapeptide is subsequently hydrolyzed into octapeptide angiotensin II (AngII, Asp-Arg-Val-Tyr-Ile-His-Pro-Phe) by angiotensin-converting enzyme (ACE) found in the lungs. AngII is the main hormone of RAAS that affects vascular properties and blood volume directly, resulting in increased blood pressure. Moreover, AngII stimulates the secretion of the hormone aldosterone from the adrenal cortex. Aldosterone causes blood vessels to shrink and kidney tubules to increase sodium, leading to water re-absorption into blood. The volume of fluid in the body is, in turn, elevated, and thus increasing blood pressure (Figure 3.12).

Nowadays, ACE inhibitor is considered as anti-hypertensive drugs due to its ability to restrain AngI from transforming into AngII. Therefore, these drugs are able to decrease the aldosterone that causes blood vessels to shrink. Moreover, the inhibitor can reduce the degradation of bradykinin, the nonapeptide that causes blood vessels to expand. Thus, ACE inhibition is the target for hypertensive prevention and treatment. At present, drugs that inhibit the ACE are captopril, enalapril, alacepril and lisinopril (Figure 3.13). These drugs can react with zinc at active site of enzyme, causing the enzyme to be incapable of reacting with the substrate. However, these

drugs can cause side effects such as low blood pressure, rashes, urticaria, high potassium, liver toxicity disease and kidney dysfunction [61, 62]. Therefore, the development of natural products as potential drugs or nutraceuticals for treatment and prevention of hypertension is alternative choice for patients.



**Figure 3.12** The scheme showed renin-angiotensin-aldosterone system (RAAS). When blood volume is low, renin is secreted into blood circulation and convert angiotensinogen into AngI. ACE hydrolyzes AngI into AngII. Then, aldosteron is stimulated by AngII and cause high blood pressure.



**Figure 3.13** The chemical structures of anti-hypertensive drugs that inhibit ACE and are used for treatment of hypertension including captopril, enalapril, alacepril and lisinopril.

At present, ACE inhibitors from natural sources such as peptides and bioactive compounds from plants (corn, sesame, green beans, rice and tea), animals (milk, eggs and fish) and microorganism are discovered [63-65]. Besides, ACE inhibitors present in roselle are found as anthocyanins (delphinidin-3-O-sambubioside and cyanidin-3-O-sambubioside), which functioned by competing with the substrate at the active site of enzyme (competitive inhibitor) [65]. Polyphenolics in green tea could inhibit ACE activity [66]. In addition, flavonoids and procyanidins from plants that exhibited ACE inhibitory activity are isoquercitrin, vitexin, isovitexin, orientin, isoorientin, (+)-catechin, (-)-epicatechin, procyanidin B<sub>2</sub>, procyanidin C<sub>1</sub>, apigenin, luteolin, kaempferol and quercetin [67, 68]. In addition, potassium intake from fruits (bananas, apples, citrus fruits, pears, plums, peaches and berries) and vegetables (tomatoes, carrots, capsicum peppers and green vegetables including spinach, lettuce, cabbage and green beans) were studied for their hypertensive activities in UK men and women [69]. The results suggested that potassium in fruits and vegetables could lower blood pressure.

Chili has been believed to possess a positive effect on high blood pressure relieving. Previous studies suggested that three species of chilies (*C. annuum*, *C. baccatum* and *C. pubescens*) could function against ACE, and this property had correlation with the total phenolic contents [11]. Furthermore, the ACE inhibitory activities of water extracted peppers (*C. annuum*) including green, red, orange, yellow, cubanelle, red sweet, yellow sweet, long hot and jalapeno peppers were investigated [10]. The results suggested that yellow pepper exhibited the highest ACE inhibitory activity (Table 3.4). Besides, capsaicin in peppers could reduce high blood pressure in hypertensive rat [13]. Nevertheless, the information regarding biological properties against hypertension of chili is limited.

**Table 3.4** The summarized data on angiotensin-converting enzyme inhibitory activity of chili extracts

Chili species	Chili types	Solvents	% inhibition of ACE	Ref
<i>C. annuum</i>	*Green	DI water	23	[10]
	*Red	DI water	76.5	[10]
	*Orange	DI water	59	[10]
	*Yellow	DI water	84.1	[10]
	*Cubanelle	DI water	79.9	[10]
	*Red sweet	DI water	73	[10]
	*Yellow sweet	DI water	60	[10]
	*Long hot	DI water	70	[10]
	*Jalapeno	DI water	62	[10]
	¥Chili de arbol	DI water	43	[11]
	¥Chili ancho	DI water	68	[11]
	¥Japanese chili	DI water	68.5	[11]
	¥Paprika pepper	DI water	92	[11]
<i>C. chinense</i>	¥Red pepper	DI water	84	[11]
<i>C. baccatum</i>	¥Yellow pepper	DI water	41	[11]
<i>C. pubescens</i>	¥Rocoto	DI water	71	[11]

\* Concentration of extracts = 1.5 g/mL

¥ Concentration of extracts = 0.05 g/mL