



**STUDY OF IMPACT BRUISING OF GUAVA
USING FRACTAL IMAGE ANALYSIS**

THAN HTIKE

**MASTER OF SCIENCE
IN
POSTHARVEST TECHNOLOGY AND INNOVATION**


**SCHOOL OF AGRO-INDUSTRY
MAE FAH LUANG UNIVERSITY**

2021

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EXAMINATION COMMITTEE



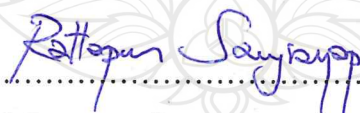
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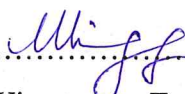
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“Namo tassa bhagavato arahato sammāsam buddhassa.”

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Than Htike

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Degree	Master of Science (Postharvest Technology and Innovation)
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ABSTRACT

Guava fruits were examined under different impact test conditions such as drop height and either number of drops or storage temperature. In the first trial, the completely randomized design (CRD) was assigned two drop heights (0.3 and 0.6 m) for once and five times compared with control as no drop for impact testing of ‘Gim Ju’ guava. The peeled color of bruising guava and fractal dimension (FD) value using image analysis was analyzed at the bruise region. Results show five drops from a height of 0.3 and 0.6 m gave greater bruise area (BA), bruise volume (BV), bruise susceptibility (BS), bruise score and a^* value, with lower L^* , b^* , C and FD values than those of the other three treatments, as indicated by browning level. The FD value gradually declined with increasing of storage time and bruise severity. The correlation coefficient (r) values of FD between BA and BV were more significant than those L^* and a^* values. It was the first report to exhibit FD potential for assessment of bruise impact damage in guava and other fruits.

In the second trial, the experimental design was response surface methodology (RSM) using central composite design (CCF) with three independent variables. The aim of this trial was to investigate factor analysis of impact testing in ‘Glom Sali’ guava. The drop heights (0.2, 0.4 and 0.6 m), number of drops (1, 3 and 5) and storage temperature (10, 20 and 30 °C) were employed for impact testing of ‘Glom Sali’ guava.

Twenty simulated impact treatments were conducted for 48 h to assess and analyze bruise area (BA), bruise volume (BV), browning index (BI), the total color difference (ΔE), bruise area by image analysis (BAI) and fractal dimension (FD) for impact bruise damage on guava. The correlation coefficient ($r = -0.6055$) between ΔE and FD value was higher than that of ΔE and either BA ($r = 0.3132$) or BV ($r = 0.2095$). The FD variable exhibited the highest R^2_{adj} value (81.69%) among the other five variables which represented the highest precision model with a high determination coefficient value (R^2_{adj}) (>0.8) for impact bruising prediction. A lower FD value performed a higher impact bruising severity level of guava and supported conventional bruise determination. The optimization of impact testing to minimize its bruising was drop height of 0.53 m for five drops with a storage at 30 °C under simulated impact test. Overall conclusion in this thesis research, it was the first study to apply RSM design in impact bruising of guava and other fruits. RSM was performed in drop heights, the number of drops and storage temperature factor that had an individual effect on impact bruising of guava. A lower FD value exhibited a higher impact bruising severity level of guava and supported conventional bruise determination. The fractal dimension of peeled guava assessed by image analysis was achieved a highly capable measurement to determine impact bruising susceptibility in guava fruit.

Keywords: Bruise Susceptibility, Fractal Dimension, Impact Bruise, Mechanical Injury

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ABBREVIATION AND SYMBOL



BA	Bruise Area
BV	Bruise Volume
BS	Bruise Susceptibility
SBS	Specific Bruise Susceptibility
BI	Browning Index
ΔE	Total Color Difference
BAI	Bruise Area by Image Analysis
FD	Fractal Dimension
RSM	Response Surface Methodology
CCF	Central Composite Design
kg	Kilogram
$^{\circ}\text{C}$	Degree Celsius
RH	Relative Humidity
L^*	Lightness
a^*	Red/green opponent color
b^*	Yellow/blue opponent color
C	Chroma
Hue	Hue Angle
TSS	Total Soluble Solid
CRD	Completely Randomized Design
SE	Standard Error
r	Correlation coefficient
R^2	Coefficient of Determination
$R^2_{(\text{adj})}$	The determination coefficient of prediction
$R^2_{(\text{val})}$	The determination coefficient of validation

CHAPTER 1

INTRODUCTION

1.1 Background

Guava (*Psidium guajava* L.) is all-year-round production and belongs to the family Myrtaceae. There are around 150 species in this genus, it is the most popular fruit of this genus. Guava is an important commercial fruit in the global market in Thailand [1]. Recently, Thailand was the sixth-ranked of global production, and export value had significantly increased within 5 years (2013-2017) [2]. The export value of guava increased from around 5 to 9 million tons of exported guava around 170 million Baht from 2015 to 2019. The major export markets were Singapore, Myanmar, United Arab Emirates, Malaysia, and Qatar [3]. To maintain quality, prolong the shelf life, and decrease losses after harvesting, guava requires suitable post-harvest handling. The guava can be damage by improper handling, which can produce in bruises and injuries making it susceptible to microbial spoilage [4]. The bruising effects that occur can be distinguished from changes that occur in the guava such as, browning and softening of the fruit peel. The water released from the ruptured cells causes cell destruction at the time of the bruise, and the intercellular air spaces are decreased [5]. Postharvest losses of guava at different levels such as farm, trader, and retailer showed an aggregate loss of 50.17%. The highest loss was at the retailer level (3.99%) and the lowest at the trader level (0.3%). Therefore, transport and distribution from the retailer level affect the guava bruise [6].

Fresh fruits are very susceptible to mechanical damages such as impact, vibration, compression, and puncture damage through harvesting, packaging, and transportation, and bruising is one of the most important mechanical damages. Bruising is the most common type of mechanical damage caused by the fruit dropping due to inappropriate handling and packing, poor design and harvesting, and packaging that

affects fresh horticultural product, and reduces quality to the consumers [7]. Impact damage to fruit is highly severer than vibration and compression damages to fruit. The dynamic impact of single fruit happens in fruit-to-fruit impact and between packing, whereas impact damage occurs when the fruit drops with adequate force onto a surface. Free drops of fruit from trees to ground during harvesting cause impact damage, as does dynamic impact between single fruit and between them and packaging and containers [8].

There are various factors affecting impact bruise damage testing such as drop heights, number of drops, storage temperature, and impact surface by using free-fall drop test. Based on fruit size and spherical shape as guava, a range of drop height for impact testing was 0.04 to 0.75 m in various fruits such as guava [5], apple [9-11], pomegranate [12-14], peach [15] and pear [16-17]. Increasing the drop height (or impact energy) elevated the potential for bruise damage in apple [11, 18], pomegranate [13, 19], kiwifruit [20] and peach [21]. Therefore, a higher drop height gave more bruise susceptibility than a lower drop height. Fruit quality is also affected by the number of drops onto an impact surface. There have been less studies in the number of drops to investigate the quality of fruit impact bruise. The number of drops of one to three times were used for impact testing in apple, pomegranate, peach and pear [10, 13, 15-16, 18]. The fourth drop of apple resulted in an irreversible change in cell damage which stabilized the bruise energy [22]. Regarding storage temperature, the optimal storage temperature for guava was recommended at 10°C [23]. Different storage temperatures range from 0 to 25 °C were investigated impact bruise damage of the fruit such as apple [11], pomegranate [13, 19, 24] and pear [16-17]. After fruit impact damage, higher storage temperature influenced higher increase in bruise area (BA), bruise volume (BV) and color changes resulting browning symptom on apple [11], pomegranate [24] and pear fruit [16-17]. However, no report has studied impact bruise damage of guava fruit with different drop heights and number of drops.

Previous studies on impact bruise damage investigated two independent variables as drop height and storage temperature, with a fixed number of drops in apple [11], pomegranate [19] and pear [16-17]. Most studies on simulated impact bruise damage only focused on experimental designs by fixing one or two variables as completely randomized design (CRD) or factorial in CRD. Thus, no clear factor

analysis has demonstrated impact fruit bruising under three independent variables. RSM design was not previously applied for impact bruising in guava and other fruits. Until recently, no RSM experiments investigating free fall or impact testing had been conducted for impact bruising susceptibility at various drop heights, number of drops and temperature conditions to simulate the effects on guava and other soft fruit. Only two RSM studies on vibration testing used two independent variables to design vibration conditions of tomato [25] and guava bruising [26]. No previous studies have applied RSM to examine three independent variables as drop height, number of drops and storage temperature, with three factor levels for free fall drop testing in guava and other fruits.

On the other hand, for non-destructive fruit quality evaluation, image analysis is a beneficial automatic tool. It is rapid, effective, low cost, and more consistent than a human evaluation that suffers from fatigue and lack of attention [27]. Conventional method is time-consuming and vulnerable to error measurement for investigation of impact bruise damage such as BA and BV [28]. As various previous reports, most researcher used textural features of image as FD to assess fruit internal browning, color change in the flesh, and fruit defects in pear [29], banana [30], apple [31], and cucumber [32] to evaluate different fruit quality. It was the first report that FD analysis exhibited high potential and accuracy for vibration bruising guava under simulated transportation [26]. The efficiency of FD analysis showed potential when comparing BA with BV (conventional technique) for impact and vibration bruising assessments. However, there has been no report in application for impact bruising determination of guava or other fruits.

According to above reviews provide background, impact bruise damage of guava fruit factors was focused on drop heights, the number of drops, and storage temperature which related to occur impact bruise damage in postharvest loss of the guava. Also, image analysis will be applied in this study for impact bruising determination of guava due to having a high potential determination of vibration bruising of guava [26]. However, there was no report of employing image analysis to assess the impact bruise damage of guava fruit or other fruits. Thus, using image analysis and RSM for impact bruise damage of guava in this study will provide more

information for developing impact bruising assessment under various impact testing conditions.

1.2 Objectives

The main objectives of this research were:

1.2.1 To study and investigate the impact bruising susceptibility of guava sustained from different drop heights and number of drops using fractal image analysis.

1.2.2 To determine the optimal impact test conditions from the combination variables among drop height, the number of drops, and storage temperature, and to apply image analysis to evaluate guava impact bruising in applying RSM design.

1.3 Hypotheses

This study was aimed to address the following research questions from the experiment which can be conducted in the different drop heights and number of drops by free-fall drop test:

1.3.1 It is expected that higher drop heights for five times dropping will have more bruise susceptibility than lower drop height level for a single drop.

1.3.2 It is expected that the optimal impact testing condition will be received from RSM design among the three independent variables of drop height, number of drops and storage temperature.

1.3.3 It is expected that the fractal dimension through image analysis will have a potential parameter to determine the impact bruise damage in guava than color measurement under different impact testing conditions.

1.4 Scope of Studies

1.4.1 To study the impact test from different drop heights (0, 0.3, and 0.6 m) and the number of drops (single and five drops) on impact bruising and quality of ‘Gim Ju’ guava to reduce the postharvest loss.

1.4.2 To study the optimal impact test condition from utilizing response surface methodology (RSM) among drop heights, the number of drops, and storage temperature affecting impact bruising of ‘Glom Sali’ guava.

1.4.3 To study the efficiency of fractal image analysis to determine impact bruising of ‘Gim Ju’ and ‘Glom Sali’ guava fruits from various drop heights and either number of drops or storage temperature as compared with color measurement and bruise susceptibility assessment.

1.5 Expected Benefits

1.5.1 To obtain the impact bruise susceptibility and quality changes of ‘Gim Ju’ guava under different drop heights and number of drops.

1.5.2 To obtain optimal impact test condition from utilizing RSM design among drop height, the number of drops, and storage temperature on impact bruising of ‘Glom Sali’ guava.

1.5.3 To obtain FD potential for impact bruising determination of guava under various impact testing conditions.

1.6 Research Location

Postharvest Technology Laboratory, Scientific and Technological Instruments Center, Mae Fah Luang University, Chiang Rai, Thailand.

CHAPTER 2

LITERATURE REVIEWS

2.1 Guava

Guava (*Psidium guajava* L.) belongs to the Myrtaceae family. There are around 150 species in this genus, it is the most popular fruit of this genus. The initial guava travels from southern Mexico across or into Central America. Guava is one of the most widely grown fruit in tropical and subtropical regions of the world. More than 400 guava cultivars and hybrids are available in the world, many of these cultivars and hybrids are a high yielding variety of good quality and soft seeded but only around ten varieties are responsible for the majority of commercially cultivated [33].

Guava fruit shape differs from round to pear shape. Skin color for mature ripe fruit varies from green to yellow color. The pulp color varies from white to yellow to pink and red color. Texture and taste of different guava cultivars as well as different seed content. Mature green fruits with a sweet and attractive flavor, when plucked from a tree, vary in shape and size depending on the varieties. Due to the soft thin-skinned guava fruit with a short postharvest life, it is subjected to mechanical injury, insect attack, and fungal rots. The fruit is mostly used in jellies and jams, although it is also used in other ways [34].

2.1.1 Guava Production and Export

Guava is grown commercially in many tropical and subtropical countries throughout the world. As shown in Table 2.1, guava production was 6.75 billion tons, including 5.52 billion tons (81.7% of the global production) from Asia countries from 2015 to 2017. India (57.5% of the global production) was the greatest leading country of guava production followed by Pakistan, China, Brazil, Thailand, Malaysia, Vietnam, and the Philippines, respectively [2]. Guava fruit is marked in the fourth of fruit

production in India and about 160 guava cultivars are available. It is growing in almost all-state. In India, guava is commercially propagated by air-layering during monsoon [35].

In Thailand, guava is an important commercial fruit in the global market [1]. Recently, Thailand was the sixth-ranked of global production (Table 2.1) and export value had significantly increased within 5 years (2013-2017) [2]. Nakhonpathom and Ratchaburi provinces are the main guavas production in central Thailand [36]. White guavas are the most common commercial varieties grown in Thailand. In the mature-green stage, they are picked and consumed. Guava characteristics for domestic market in Thailand are bright-green peel color, crispy white flesh, and sweet with slightly sour flavor.

The export value of guava increased from around 5 to 9 million tons with the value of exported guava around 170 million Baht from 2015 to 2019 (Table 2.3). Singapore, Myanmar, United Arab Emirates, Malaysia, and Qatar were the top export markets. Singapore and Myanmar was the first and second ranking of imported guava from Thailand with around 2 to 3 million tons per year as shown in Table 2.4 [37]

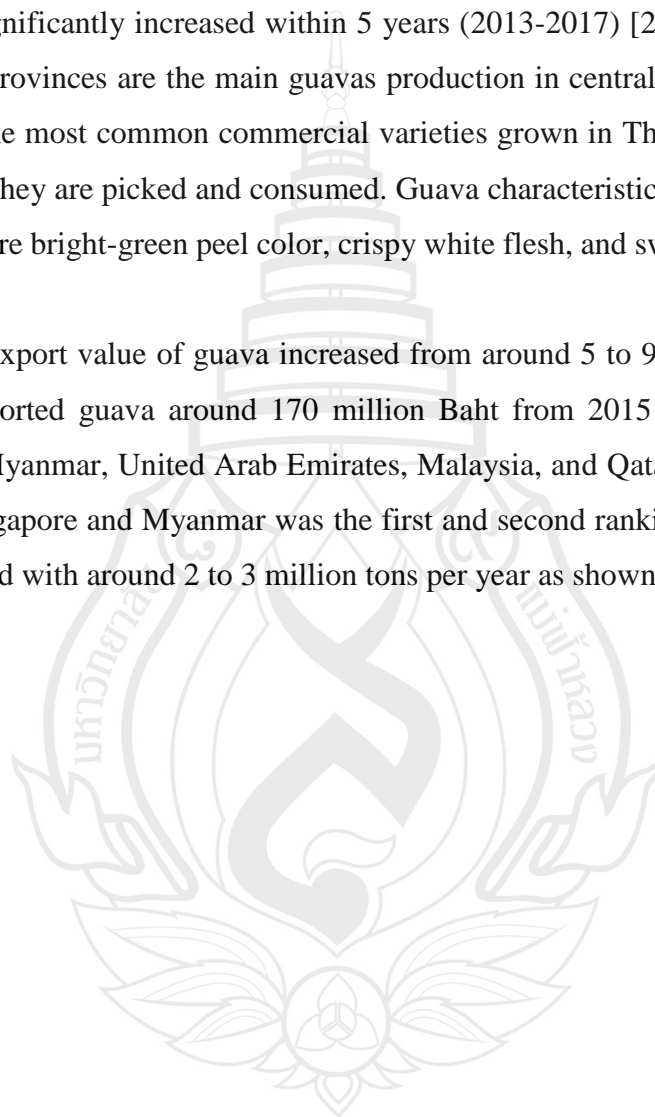


Table 2.1 Guava Average Production in Different Countries

Year (2015-18)	Production (million tons)
World	6,752.5
Asia	5,519.3
1. India	3,885.0
2. Pakistan	474.2
3. China	365.5
4. Brazil	353.2
5. Indonesia	264.8
6. Thailand	218.1
7. Malaysia	49.5
8. Vietnam	24.2
9. Philippine	10.8

Source [2]

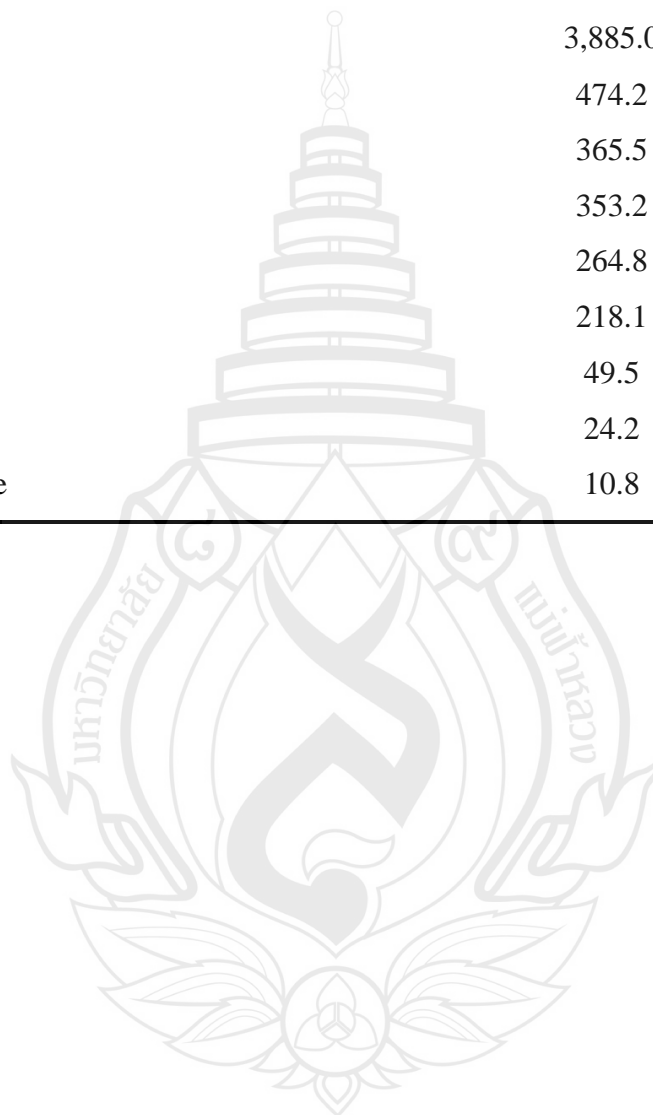


Table 2.2 Evolution of Guava Export from Thailand in 10 years (2007-2017)

Year	Export value (million tons)
2007	2,183
2008	1,567
2009	2,001
2010	2,426
2011	3,034
2012	2,896
2013	4,069
2014	4,985
2015	5,901
2016	6,360
2017	6,625

Source [2]

Table 2.3 The Exported Guava from Thailand from 2015 to 2019

Year	Quantity (million tons)	Value (million Baht)
2015	5.901	135.6
2016	6.360	146.7
2017	6.625	149.3
2018	8.117	206.3
2019	8.986	211.4

Source [37]

Table 2.4 Guava Exported Countries from Thailand from 2015 to 2019 (million tons)

Country	2015	2016	2017	2018	2019
Singapore	2.871	3.075	3.234	4.111	3.263
Myanmar	2.690	3.062	3.124	2.258	3.284
United Arab Emirates	0.163	0.109	0.101	0.088	0.078
Malaysia	0.101	0.064	0.089	1.570	2.240
Qatar	0.011	0.009	0.018	0.028	0.026
India	0.034	0.010	0.009	0.011	0.008
Maldives	0.006	0.007	0.009	0.008	0.011
Oman	0.006	0.010	0.008	0.012	0.027
Bahrain	0.006	0.008	0.014	0.016	0.012
Saudi Arabia	0.003	0.004	0.004	0.009	0.004
Others	0.007	0.002	0.014	0.006	0.032

Source [37]

2.1.2 Guava Characteristics

Since consumers prefer other fruits and can plant guava trees nearly everywhere, especially in their home gardens, only around 29% of total production is marketed domestically. The guava production prices are 10-20 Baht/kg, whereas seedless varieties cost 35-50 Baht/kg. ‘Glom Sali’, ‘Paen Srithong’, ‘Gim Ju’, ‘Sam See Krob’, ‘Phet Pu Thon’, ‘Lai Ma Lat’ and ‘Den Khun’ varieties are some of the most well-known commercial varieties of Thai guava [36]. However, the main issue affecting Thai guava marketing is its high perishability due to moisture loss, browning of the flesh, softness and fruit decay [1]. Among these commercial cultivars, guavas such as ‘Gim Ju’ and ‘Glom Sali’ have become the most appropriate variety for the market and are in high demand. ‘Gim Ju’ cultivar has a globular shape, weights 200-500 g, has a light green skin color, white flesh and aromatic [38].

2.2 Postharvest Handling and Losses from Guava Fruit Bruise

2.2.1 Postharvest Handling of Guava

To control quality, reduce losses and extend the shelf life after harvesting guava requires proper postharvest handling. The guava can be damaged by improper handling, which can produce bruising and wounds, making it susceptible to microbial deterioration [4].

Maturity indices: Changes in skin color from dark green to light green, as well as when mixed with achieved are useful indices to assess harvest maturity. Other maturity indices, including specific gravity, chemical attributes, and fruit detachment strength, have also been shown to be useful in determining harvesting stage. Guava harvested at specific gravity (less mature) <1.0 were higher than those harvested at 1.00-1.02 (high mature). Guava harvesting time is determined by subjective factors such as fruit size, skin color and hand firmness, which may vary depending on cultivar, season, plant age and management procedure for the same location [39]. As a result, the best indicator of maturity is the visual appearance of fruit, which is based on the color and size of the skin.

Harvesting: Harvesting should be done in early morning to avoid sun light exposure that alleviates a higher respiration rate under hot temperature and lead to a shorter shelf life. Guava is a delicate fruit requiring careful handling during harvesting and transportation. Hand harvesting is simply done from the plant and putting them into an appropriate container, while careful to impact damage by dropping the fruit. Guava and soft fruits for processing may be harvested using tractors with combining fingers that shake the stems, and pulling off the fruit bunches [40].

Sorting and grading: Bruised, injured, and rotting guavas are isolated from good fruits. Sorting is performed immediately after the products arrived at the packing house and the next step will be decided. Treatments in sorting as soon as possible can reduce damage, crop loss, prevent microbial infection and other foreign materials. Grading is the process of sorting fruit by grade/class criteria, color, weight, shape and size. Grading can be done by hand or with the assistance of machinery [41].

Washing: Washing can reduce the bacteria and the initial temperature of the product or absorb field heat (precooling) and impurities. Treatment with 100-150 ppm chlorine can assist to reduce infections and prevent them from activity [4]. Ismail, et al [42] investigated the application of many treatments on guava to reduce or eliminate microorganisms. Hot water, hydrogen peroxide, calcium chloride, lemongrass oil fumigation, and many combinations were used on guava.

Packing: The use of proper packaging and cushioning material free from any harmful residual effects on human health is always performed since they help in prolonging the shelf life to a great extent [43]. Guava that had been packaged with Styrofoam net and placed in a cardboard box had mechanical damage of 39.59% damage, which has less than carton packing without filler material, which had 84.03% damage. To protect the fruit from mechanical damage (pressure, impact, and friction), exported fruits are generally packaged with cardboard boxes (fiberboard) with a filler substance. Fiber board is normally covered with wax, protects the product from water or high humidity for a long time [44].

Storage: Guava should be stored at a temperature of 8-10 °C with a relative humidity of 90% [45]. Guava may be shipped to distant markets in 2-3 weeks if stored at low temperatures [46]. Depending on the temperature and relative humidity of the surroundings, the fruit may lose 10-20% of its weight at initial during the course of 6-8 days on the shelf [47].

Transportation: Transportation refers to the process of collecting fresh fruits from orchards and transporting them to a collection and packing facility, as well as attempts to convey or market fresh fruits to consumers. On the shelf life of fruit, this step takes a long time, about 50-75 % of spent in transport and distribution. In other words, the cold chain must be maintained throughout transportation and distribution process. Transportation should be handled with caution during loading and unloading and it is a simple yet efficient method of reducing guava losses [48].

2.2.2 Postharvest Losses of Guava

Most guava fruits are susceptible to different types of damage during harvesting, handling and transportation, resulted in bruising and a loss of the quality [18]. Guava is a climacteric fruit, meaning it ripens to the point when it produces the most

respiration and ethylene. Guava fruit is often damaged due to its rapid respiration rate. Guava damage can occur as a result of rough handling, which can result in bruising and cuts, making it susceptible to microbial deterioration. Therefore, good practices in postharvest handling need to keep the quality of guava [42].

Guava fruit has a postharvest shelf life because it is fragile when ripe and readily damage during storage and transportation [49-50]. The guava fruit is fragile and vulnerable to mechanical problems including cuts, punctures, bruising and compression damage. These injuries is determined by the harvesting method and postharvest handling treatments [47]. Guava postharvest losses were 5.17% on average across all handling levels, including farm, trader, and retailer stages. The retailer stage had the highest postharvest losses (3.99%), while the trader had the lowest postharvest losses (0.3%). Total damaged fruits, on the other hand, were as high as 14.48% throughout guava supply chain [6].

Table 2.5 The Percentage of the Postharvest Losses in Marketing System of Guava

Levels	Sound fruits	Damage density	Total damaged	Aggregate loss
Farm	100.0	1.10	3.29	0.88
Trader	96.71	3.01	3.01	0.30
Retailer	93.70	2.73	8.18	3.99
Total	85.52	4.83	14.48	5.17

Source [6]

2.3 Fruit Bruise

Bruising is described as external forces causing damage to plant tissue, resulting in a physical change in texture as well as chemical color, taste, and texture deviations [51]. Fruit bruising is one of the most important factors during harvesting, sorting and transportation of fruits and vegetables [20]. During harvesting, grading, packaging and

transportation, structural, tissue, and cell damage to fruits is caused by impact, compression, abrasion, puncturing, testing, or combination of these facts. Mechanical injury to the skin that causes browning can also penetrate the produce. Mechanically injury accelerates physiological processes that lead to senescence, breakdown, and the loss nutritional content. The enzyme polyphenol oxidase (PPO) is responsible for tissue browning in mechanically damaged fruits. PPO catalyzes the conversion of phenolic chemicals into o-quinones, which polymerize to create dark color [52]. The enzyme PPO, which was found in the guava fruit, catalyzes the browning of most fresh foods [53]. Skin browning caused a lower visual appearance of the fruit, which should be practically avoided of these defects to fulfill high-quality criteria. Individual fruit is commonly wrapped in paper for long-distance shipment to minimized physical damage [47].

Bruising and other types of mechanical damages (cut, puncture, silt, abrasion) cause a significant economic loss of fresh fruits due to reduction or rejection of the visual quality by the consumer [54]. The main mechanical damage causes by impact, vibration and compression forces. Firstly, the impact force during fruit harvesting, postharvest handling, packaging, transportation and distribution and it is the main cause of mechanical injury to fruits. During postharvest operations, the impact might occur between two fruits or against other surfaces (the hard surface of the storage container or packaging). Most of the impacts use to occur during the transfer of fruit. When this fact happens a surface with adequate force to even isolated cells, impact damage happens. A bruise or crack on the skin is an outward indication. Impact damage is commonly caused by free fall of fruits from trees to the ground during harvesting, as well as dynamic impacts between single fruits and packaging or containers [8]. In pomegranate fruits, impact and vibration loads that may occur during harvesting or sorting, storage and transportation [12]. Impact bruises can occur at any step in the fruit handling system, from harvesting to consumption, and they are still the most mechanical damage [55].

Vibration damage happens when fruit moves inside the packaging as a result of vibration during the transportation. Fruit can be damage by bumping other fruit or packaging, resulting in an impact injury, or by rubbing the fruit against another surface, producing abrasion. Vibration damage was produced by only one of these forms of

damage [56]. Abrasion is less serious type of postharvest mechanical damage than impact or compression, although it is well-known as a pre-harvest damage caused by wind. Pre-harvesting can also be reduced by rubbing the fruit against branches or leaves [56]. Some injuries such as abrasion on watermelon, and rolling on pear appeared on only one type of fruit. The effect of static weight or pressure acting on a product is compression forces. Compression often led in creeping due to the duration and static loads involved. Creep occurs when the fruit load remains constant, but the tissue deforms with time. The duration and the severity of the load are both important factors in determining the compression injuries. Punctures and cuts were found on roughly half of the fruit, with the prevalence being lower than bruising [57].

2.4 Factors Affecting Impact Bruise and Quality Changes of Fruits

The quality changes of fruits are affected by impact bruise damage due to internal and external factors that can be postharvest factors such as fruit characteristics of fruit shape, size (weight), packaging, drop height, number of drops and temperature. Fruit size and firmness affecting impact bruising was studied in apples and peach [58], pomegranate [12], tomato [54] and kiwifruit [20]. Postharvest factors such as drop heights, number of drops, impact surface and packaging influence the incidence and severity of bruising [54]. The bruising effects can be distinguished from changes that occurring such as, browning and softening of the fruit peel. Cell destruction occurs at the time of the bruise and the intercellular air spaces are reduced with the water released from rupture cells [5]. In this study, there are various factors affecting impact damage testing such as drop heights, number of drops and storage temperature [24, 59, 13, 19, 11, 16-17].

2.4.1 Radius of Curvature

First of all, the bruise damage from the impact test depended on the fruit shape, which is an internal factor affecting impact bruising. The most research studies were investigated in the influence of radius of curvature on the impact bruise in fruits such as apple [54, 60], pomegranate [24], tomato [61] and peach [21]. A small radius of curvature caused greater bruise area and volume of damage than a high radius of

curvature. As a result, a larger radius of curvature resulted in greater bruise damage whereas a smaller radius resulted in less bruise damage.

2.4.2 Firmness

In terms of the ripening stage or fruit firmness as an internal factor, absorbed energy increases with more ripeness of tomato fruit which determined by acoustic stiffness and color indicating a higher bruise susceptibility [54]. Also, a reduction of fruit firmness increased a higher fruit bruise in apple by higher impact damage fruit [62-64]. In the case of higher impact in apple, there was a clear difference between the value of BV for the fruit with higher and lower firmness [64]. However, a firmness reduction were not related with the reduce in bruise color of persimmon [65]. Less bruises in pomegranate due to its increased firmness, as well as fruit temperature, appear to influence bruise susceptibility [62]. Therefore, a higher firmness caused less bruising, while lower firmness happened more bruising damage in fruits.

2.4.3 Drop Height

Table 2.6 summarizes the impact condition of a free fall drop test from various heights and number of drops. Overall drop height was examined in a range of 0.01 to 0.75 m in various fruits. It can be concluded a drop height range of 0.01 to 0.75 m in apple [11, 66, 28], a drop height range of 0.05 to 0.60 m in pomegranate [24, 13-14], a drop height range of 0.20 to 0.60 m in pear [16-17] and a drop height range of 0.05 to 0.15 m in peach [15]. Regarding bruise determination, an increase of BA and BV caused a higher drop height. For example, several research examining a drop test have been conducted in apple. The increase of drop height at 0.4 m for the apple increased the BV due to the increasing of impact energy [18]. In pomegranate bruising, the BA increased significantly from the lower impact level (0.2 m) to the higher impact level (0.6 m) [19]. Bruise assessment such as BA, BV and BS were the highest at higher drop impact (0.6 m) and lower drop impact (0.2 m) on pear fruit [16]. Therefore, an increase of the drop height provided more bruise susceptibility than a reduction of drop height.

2.4.4 The Number of Drops

Fruit quality was also affected by the number of drops onto an impact surface also effects such as steel plate, sponge cushion, sandpaper, (fine) mesh, corrugated fiber

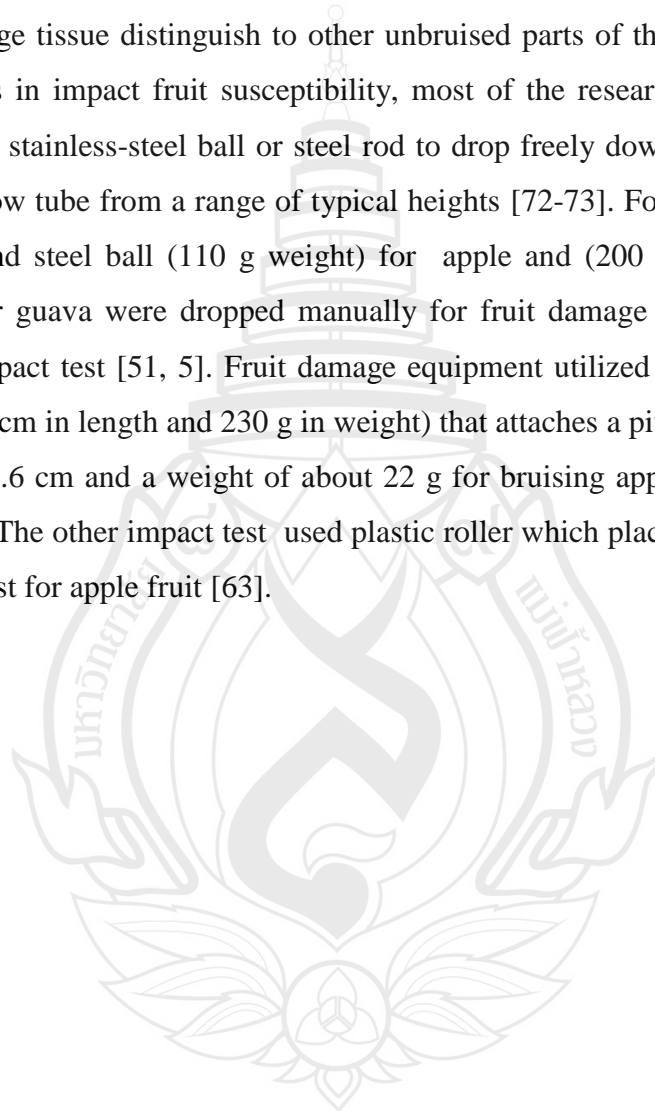
board and silicon rubber sheet [67-69]. Proper selection of the thickness of cushion materials during the handling process is important for reducing bruise [67]. There was less reported in the number of drop study to evaluate the quality of fruit impact bruise damage. Some studies investigating the number of drops from a range of 1 to 3 times in spherical shape fruit such as apple, pomegranate, peach and pear [18, 13, 16, 15, 70]. In the apple bruise investigation, the fourth drop resulted in an irreversible change in cell damage which stabilized the bruise energy [22]. Most of the researcher used fixed number of drops with other drop heights and storage temperature to investigate impact bruise [24, 13, 11, 16]. However, there was no study to investigate impact bruise damage in guava fruit with different drop heights and number of drops.

2.4.5 Storage Temperature

The impact condition of a free fall drop test from the different drop heights, number of drops and storage temperature conditions were exhibited as shown in Table 2.6. One of the most significant influence on fruit bruise evaluation is storage temperature level [62]. After drop testing, different storage temperatures range from 0 to 25 °C were kept the fruit for impact bruise damage such as apple [11], pomegranate [24, 13, 19] and pear [16-17]. For two apple storage temperatures (3 and 20 °C) at three levels of impact energy, it was happened that an increase of temperature resulted in less BV [60]. In tomato, low temperature reduced metabolic activity and modified the texture of the tomato, and the interaction of the both processes was occurred on the bruise susceptibility in the final effect of temperature [71]. However, more bruise damages were appeared at storage temperature 5 °C than at 25 °C, particularly at high impact level in pomegranate [24]. Pomegranate fruit stored at temperature 5°C had higher BV and BS than at 20 °C [19]. The recommended optimum storage temperature for guava was at 10 °C [23]. Based on above review, higher storage temperature alleviated impact bruising in most of fruits more than lower temperature, excepted pomegranate. Interestingly, no research has been examined that effect of storage temperature on the impact bruising in guava.

2.5 Free Fall Drop Test

As illustrated in Figure 2.1, a free fall drop test was followed by dropping a ball impactor of known mass (260.45 g) on the pomegranate fruit from a range of preset heights through a perforated tube. The bruise region were marked by the present of visible damage tissue distinguish to other unbruised parts of the same fruits [59]. In many studies in impact fruit susceptibility, most of the researchers generally allow using mass a stainless-steel ball or steel rod to drop freely down from the top of the guiding hollow tube from a range of typical heights [72-73]. For example, the size of uniform round steel ball (110 g weight) for apple and (200 g weight and 3.5 cm diameter) for guava were dropped manually for fruit damage to investigate impact bruise on impact test [51, 5]. Fruit damage equipment utilized pendulum test with a steel rod (80 cm in length and 230 g in weight) that attaches a pivot to a steel ball with diameter is 1.6 cm and a weight of about 22 g for bruising apple for another impact testing [73]. The other impact test used plastic roller which placed in this position for the impact test for apple fruit [63].





Source [59]

Figure 2.1 Schematic of Impact Testing with a Free Fall Steel Ball Through a Hollow Pipe (A), Stainless-Steel Ball Impactor (B) and Bruised Pomegranate Fruit (C)

Table 2.6 The Height, Number of Drops and Storage Temperature Affect Impact Bruising of Guava, Apple, Pomegranate, Peach and Pear

No.	Commodities	Height (m)	No. of Drops	Temp (°C)	Results	Ref.
Guava						
1	Guava	0.20, 0.50	1	RT	The bruise depth of a height of 0.2 m was smaller than that of a height of 0.5 m.	[5]
2	'Gim Ju' guava	0.30, 0.60	1, 5	20	The highest BS in a height of 0.3 and 0.6 m.	[74]
Apple						
3	'Golden Delicious' apples	0.15, 0.20, 0.25, 0.30, 0.40, 0.50	1	n/a	BV increased linearly with increasing of drop height and apple mass.	[18]
4	'Gala' apple	0.04, 0.05, 0.08, 0.10, 0.20, 0.30, 0.40, 0.50, 0.60	1	n/a	BV increased with an increase of drop height.	[9]

Table 2.6 (continued)

No.	Commodities	Height (m)	No. of Drops	Temp (°C)	Results	Ref.
5	Apple	0.50, 0.80, 1.10, 1.40,	1	n/a	Bruise diameter was highly influenced by the drop height. Fruits dropped from the height of 1.4 m absorbed the greatest impact energies.	[75]
6	'Gala' apple	0.01- 0.15	1	25	An increase of drop height accompanied an increase of surface pressure and occurred tissue destruction process, which resulted in contact area increase.	[28]
7	'Sannfugi' apple	0.05, 0.10, 0.15, 0.20, 0.30, 0.40, 0.50	3	n/a	The BA and BV increased by the drop height.	[10]
8	'Golden Delicious' apples	0.30, 0.50	2	n/a	Both incidence and impact bruise resulted in drop heights.	[66]

Table 2.6 (continued)

No.	Commodities	Height (m)	No. of Drops	Temp (°C)	Results	Ref.
9	'Pink Lady' Apple	0.25, 0.50, 0.75	1	0, 20	An increase of drop height and storage period affected impact bruising. BA measurement was more accuracy than BV measurement.	[11]
	Pomegranate					
10	Pomegranate	0.05, 0.10, 0.15, 0.30, 0.60, 1.00	1	21	BA and BV values resulted in increasing of drop heights.	[12]
11	Pomegranate	0.15, 0.30, 0.60	1	5, 15, 25	Impact energy and impact region played a major part in BS Fruits stored at 5 °C suffered more bruise damage than at 25°C	[24]
12	Pomegranate	0.25, 0.40, 0.60	2	19-22	An increase of drop height increased the potential for bruise damage.	[59]

Table 2.6 (continued)

No.	Commodities	Height (m)	No. of Drops	Temp (°C)	Results	Ref.
13	Pomegranate	0.20, 0.40, 0.60	1		A higher incidence of was observed at drop heights of 0.4 m and 0.6 m. TSS and TA contents were induced by bruising at drop heights of 0.4 m and 0.6 m.	[14]
14	Pomegranate	0.20, 0.40, 0.60	2	5, 20	The highest browning was observed in fruit bruised at 0.4 m and 0.6 m after 48 h at 5 and 20 °C. An increase of browning intensity was associated with both temperature and time after impact.	[13]
15	Pomegranate	0.20, 0.40, 0.60	2	5, 20	BA increased from 0.2 m to 0.6 m. Fruit stored at 5°C had higher BV and BS than at 20°C.	[19]

Table 2.6 (continued)

No.	Commodities	Height (m)	No. of Drops	Temp (°C)	Results	Ref.
Peach						
16	Peach	0.05, 0.10, 0.15	1	22	An increase of drop height from 0.05 to 0.15 m considerably increased the BA. When the drop height was increased from 0.05 to 0.15 m, the mean value of BA increased around 15%	[15]
Pear						
17	Pear	0.2, 0.4, 0.6	1	10, 22	Highest drop heights and storage temperature showed increase in bruise and color measurements	[16]

Note n/a means not available

2.6 Bruise Measurement, Detection, and Analysis of Fruits

The bruise was evaluated in two ways: by visually, by removing and measuring the damage in cross-section and marked tissue [50]. Due to the first approach as non-destructive method, the visual rating was done at the start and throughout the process, whereas the second method was destructive and only done at the storage time [62]. For impact bruise evaluation, several biochemical, physical analysis and visual detection method were used to detect the bruising. Several biochemical methods applied for analysis of enzymes and metabolites and physical methods for analysis of firmness, weight loss, and color measurements. Optical detection method used for the analysis of ultrasonics, X-rays, magnetic resonance and image analysis [7]. There were three types of bruise measurements namely; (1) using manual measurements of bruise detection such as BA, BV and BS [72], (2) imaging of bruise tissue by image analysis to evaluate bruised tissue [76], and (3) applying non-destructive technique for bruise measurement and analysis [77].

2.6.1 Bruise Area (BA) and Volume (BV)

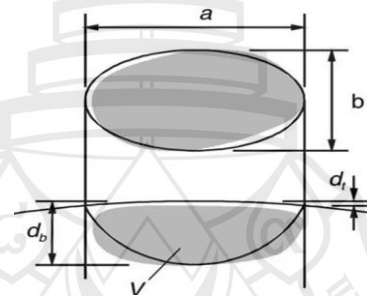
Both the major and minor axes can be used to determine the bruise depth. For a small impact, the bruise thickness approach provides the most accurate volume estimation. By measuring the widths and depth of the damaged fruit, it is difficult to estimate the bruising. The bruise thickness method, which is based on a bruise section parallel to the stem-calyx axis, is the most accurate way for estimating the bruising apple across a wide range of impacts [68]. Although BV is an important, BA is a more significant due to determines the quality of the fruit [12]. The most widely used methods for determining the extent of bruise damage to a fruit are the BA and BV. The BA and BV were determined for fruit damage in many fruits such as apples [51, 10] and pomegranate fruits [12]. The BV increased linearly with higher of drop height and the mass of fruits. In BV determination, there was a high correlation between the drop height and the mass of the apple [18], pomegranate [59] and peach [78]. The apple fruit made contact with impact surface and turned pale after 24 hours [51], whereas the pomegranate fruits storage for 48 hours caused turning pale color [24]. Fruits were cut

along the surface of bruising deformation. The following equation was used to determine the (BA) and (BV) of each fruit [42].

$$BA = \pi/4 (ab) \quad (1)$$

$$BV = \pi d/24 (3ab + 4d^2) \quad (2)$$

Where, a and b are major axes of the bruise elliptical and d is bruise depth in mm as shown in Figure 2.1.



Source [21]

Figure 2.2 Elliptical Method for Determining Bruise Area and Volume

2.6.2 Bruise Susceptibility (BS) and Specific Bruise Susceptibility (SBS)

General bruise susceptibility ($\text{mm}^3 \text{J}^{-1}$) is defined as the ratio of bruise volume to the impact energy, E (J) and specific bruise susceptibility (SBS, $\text{mm}^3 \text{J}^{-1} \text{kg}^{-1}$) [51, 19, 66] that is already been calculated by the equation:

$$BS = BV/E \quad (3)$$

$$E = mgh \quad (4)$$

$$SBS = BS/m \quad (5)$$

Where m is the mass of the impacting object (kg), g is the acceleration due to the gravity (ms^{-2}), and h is the drop height (m).

2.6.3 Bruising Scoring

In the previous studies, there has no study in evaluating the bruise score of guava fruit from impact bruise damage. Most of the studies in the evaluation of bruise scoring were reported in apple fruits. The apple fruit was measured for changes in bruising using a visual score by a direct measure of the bruised area in cross-section and varied in the range of score. Firstly, the visual evaluation of the apple was based on a four-point scale, with 1, no obvious bruising; 2, light brown bruise with no defined edge; 3, moderately dark brown bruise with a well-defined edge; 4, dark brown bruise [62]. The other bruise scores in ‘McIntosh’ apple used to classified bruising in the range of 0-absent, 1- slight, 2- moderate, and 3- severe [50]. Moreover, the visible damage to the bruised site was recorded for the ‘McIntosh’ apples: 0 = absent, and 3 = severe [79]. On the hedonic scale: 1 = unusable, 3 = poor, 5 = good, 7 = very good, 9 = excellent, it was reported to measure the appearance of impact damage in pomegranate fruit including rotting, bruising and scald [80].

2.6.4 Pulp Color at Bruise Area

After impact test, the color changes progressed to browning and were found to be related to fruit bruise damage [81]. The mathematical equation from the CIE *Lab* coordinates value in pomegranate fruit was used to determine the differences in color on the fruit peel [13]. Banks [82] reported the discoloration of bruised ‘Granny Smith’ apple tissue happened within a few hours of the impact being applied to the fruit surface. After bruising the fruit, the tissue becomes darker (L^* value is decreased), browner (Hue value is decreased), and increased in color (C value is an increase). According to [13], the value of the pomegranate fruit is determined by utilizing the mathematical formulation of the CIE *Lab* coordinates to determine the difference in color on the fruit surface. The total color difference (ΔE) was calculated to compare the color difference between the first (immediately after impact) and 4 to 48 hours after impact bruising (incubation period) at a damaged region of the fruit. Drop impact as well as temperature and incubation time had an effect on ΔE . The lightness (L^*) and the values of hue of damaged tissue reduced as the color changes of bruise apples progressed, but the

chroma (C) value increased gradually [83]. The color variations in hue value may induce anthocyanin leaking from cell breakdown in the drop test of lowbrush blueberries from drop height (1.59 m) [27]. Therefore, the color measurement of the L^* , a^* , b^* , C and Hue parameters was influenced by the bruising level and commodity.

2.7 Image Analysis and Fractal Dimension

For non-destructive fruit quality evaluation, image analysis is a beneficial and useful automatic tool. It is faster, more effective, less expensive and more consistent than human assessment which suffers from fatigue and lack of attention [27]. Color, texture, shape and size are image assessment that may be utilized to judge fruit qualities [84]. Generally, colour assessment can be applied to assess physical, chemical and mechanical properties of fruit such as ripening, maturity index and damage fruit [27]. Several studies have been carried out to evaluate the quality evaluation and major color changes in various fruits including apple [85], pineapple [86], orange [87] and mango [88]. Images include various information that may be utilized to perform high accuracy single or combination feature assessments for food quality evaluation [89]. Bruise damage was analyzed using the ImageJ software which includes: image segmentation, extraction of region of interest image: red, blue, green (RGB) color channel and pixel intensity extraction and image threshold. Bruise signs in the form of dark spot were observed at minimum 24.70 and maximum 39.88 threshold values in mango fruit [90].

For a single bruising point, manual measurements are suitable solution, but size feature-based image analysis techniques approaches are a better choice for obtaining acceptable accuracy than the time-consuming of manual measuring procedure. Several studies used advance image analysis techniques such as hyperspectral imaging [73, 91-92], thermal imaging [63, 93] and X-ray computed tomography [94] to assess impact bruising on apple. Impact bruising was determined in guavas using vis-NIR imaging [5]. To our knowledge, no studies have assessed impact bruising damage of guava using simple fractal image analysis. Previous techniques for image analysis of impact bruising required advanced instruments and complex data interpretation procedures,

while to our knowledge, no studies have assessed impact bruise damage of guava using simple fractal image analysis.

Image analysis approach called fractal dimension (FD) assessment is used to assess the browning of fresh products. In postharvest technology, the fractal method is an essential tool for determining fresh appearance. Avocado fruit browning was assessed during storage using fruit images, and the fractal technique proved to be accurate [95]. Fractal modelling can be used effectively to evaluate the severity of fresh browning, its color change and to gain a better knowledge of enzymatic, chemical changes in fruit fresh [96]. FD is a textural features description approach in image analysis [89]. Surface intensity is computed and assumed to be a fractal, which is defined as an object that keeps its shape regardless of the scale. Texture measurements (surface intensity in images) are obtained using the FD [97]. The FD value may be calculated using a logarithmic regression against the observation scale. When different unit measures are employed, such as the blanket method, box counting method and frequency domain method, different fractal methods develop [98]. Previously, most studies conducted different fruit quality evaluation using image textural feature in Table 2.7, e.g. FD analysis to estimate fruit browning in inside the fruit, flesh color changes and fruit damage such as banana [30], pears [29], apple [31] and cucumber [32]. Recently, FD analysis has exhibited higher potential and accuracy for vibration bruise damage in guava fruit under simulated transportation [26]. Thus, there are currently no study for impact bruising evaluation of guava by using image analysis technique, particularly FD assessment.

Table 2.7 Fruit Quality Assessment by Image Analysis

Fruit	Determination	Ref.
Apple slice	For enzymatic browning rates, fractal browning index (FBI) method were 14.3-23.2 times higher than the rates calculated with L mean values.	[31]
Banana slice	Enzymatic browning rates, FBI method was between 8.5 and 35 times higher than rates calculated with the L^* , a^* and b^* value.	[30]
Pear slice	The lightness decreased when FD increased, indicating a greater complexity in distribution of the L^* value.	[29]
Cucumber	The length, width and neck shape features were extracted accurately.	[32]
Pineapple	Maturity classification based on fruit yellowish shell color with 94.29% accuracy.	[99]

Cropped RGB images were converted to 8-bit images to determine the FD value. The intensity of fruit bruise damage was estimated using a surface plot of 8-bit images. Using the threshold method, the bruise damage region was then separated. Before determining FD values, the selected of the bruise was converted into a binary image. Using a calibrated grayscale, the size and gray scale values of all images were standardized. The Box Counting plug-in on ImageJ program was then used to calculate FD value. when plotting the value of $\log(N)$ on the Y-axis against the value of $\log(r)$ on the X-axis using the box counting method, fractal dimension is again the slope of the line. FD is calculated using the same equation. This time, N represents the number of boxes covering the pattern, and r represents magnification, or the inverse of the box size. The formular for FD was $FD = \log(Nr) / \log(1/r)$ [100]. A greater FD indicates surface roughness [101]. This method has been used to track changes in the mottled skin of banana [30], apples [31] and pears [29], among other fruits.

2.8 Response Surface Methodology and Correlation Coefficients

The response surface methodology (RSM) was developed using the graphical representation generated after fitting the mathematical model was fitted. RSM is used to show the effect of the independent factors on the dependent variables. In addition, the method provides an empirical model [102]. RSM is a set of statistical and mathematical approaches for characterizing data performance and generating statistical inferences based on fitting a polynomial equation to experimental data. The preservation procedure was optimized using RSM, and an incomplete three-level and three factor designs was accepted [103]. RSM is reported to have several advantages, including a decrease in the number of experimental runs needed to evaluate numerous variables and the capacity to identify interaction [104-105]. By creating a model equation, RSM can analyze the relationship as well as interactions among the various factors using quantitative data [106]. RSM is a statistical technique for assessing the effects of various factors and their interactions on response variables [107].

RSM was applied with central composite design (CCD), central composite face-centered design (CCF) and Box-Behnken design (BBD), as shown in Figure 2.3 and Table 2.8. RSM is a collection of mathematical and statistical techniques which are useful for the modeling and analysis of problems in which a response variable of interest is influenced by several independent variables [108]. Face centered designs are a type of CCD design with an alpha of 1 (α of 1). The axial points are located at the center of each face of the factorial space in this design. This type of design involves 3 levels of each factor. CCD of RSM can offer a sufficient quantity of data for lack-of-fit testing without requiring an excessive amount of design points [109]. BBD has the benefit of using a limited number of factors to determine the complicated response function and avoiding experiments conducted under severe conditions [110].

Input variables that may be modified independently of one another are known as independent variables or factors. A run is a sequence of tests that make up an experiment. Out variables that are impacted by numerous independent factors are known as dependent variables or response. Converting each studies value into a range without dimensions (-1 to +1) is required for factor level coding. Using the model

equation and regression coefficients, the estimated response may be determined. However, the model adequacy must be tested in order to decide if it is acceptable or not. The initial derivative of the mathematical equation that shows the response surface and equals it to zero may be used to compute the optimal point (maximum or minimum point). These numbers indicate the variables' coded values, which determine whether the result is maximum or minimum. The goal point for process optimization must be identified within the ranges of examined factors [111]. Because of good $R^2_{(adj)}$ values, lack of fit value, and good predicted and experimental values, RSM was effectively employed to improve extraction conditions by different approaches. Therefore, the RSM algorithm could accurately estimate the results.

The α value can be defined as the calculated distance of each individual axial point from the center of the design. If α is less than 1, which indicates the axial point must be a cube and if it is greater than 1, it indicates it is outside the cube. The α value is determining by using the equation $\alpha = 2(k-p)/4$. The α value depends on the number of variables or experimental runs in the factorial portion of the CCD design. It is 1.41, 1.68, and 2.00 for 2, 3, and 4 variables respectively [112]. The result of the central composite design values of α was given and for the three independent variables, the optimum value of α was 1.682 for both coded and uncoded forms of the design [109]. CCF designs produce generally high-quality predictions over the whole design space without requiring the use of points outside the original factor range and non-rotatable design. For all factors, the star points produce new extremes for the low and high settings. The star points are at the center of each face of the factorial space for the CCF, so $\alpha = \pm 1$ (Figure 2.3). Each component must have three levels in this design [113]. Although BBD design has low prediction quality region, it is rotatable. When the researcher has to prevent combination factor extremes, its 'missing corners' may be beneficial. The α value for BBD designs is 1.00. [112].

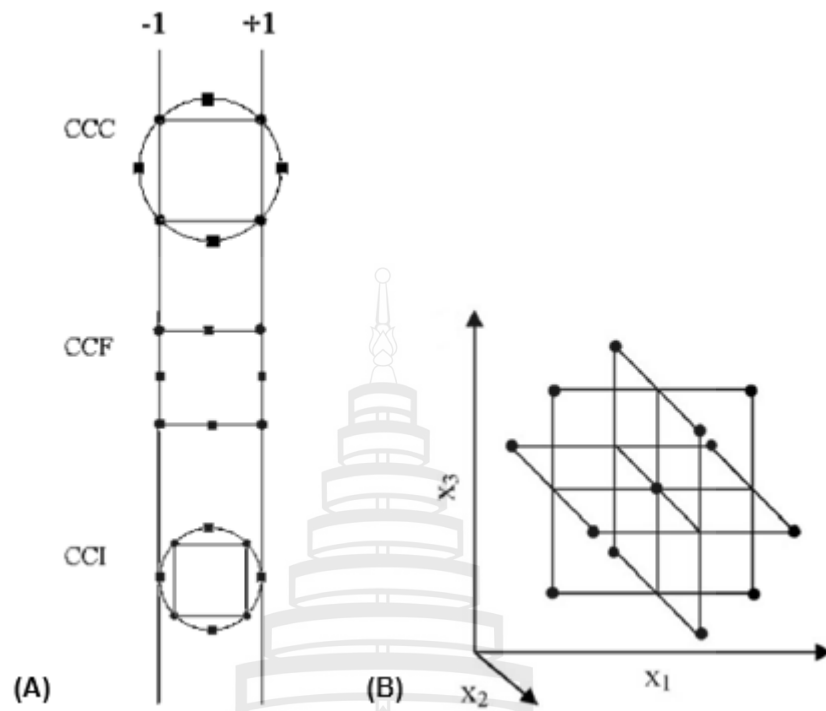


Figure 2.3 Types of Central Composite Design CCC-circumscribed, CCF-face-centered and CCI-inscribed (A) and Three Factors Box-Behnken Design (B)

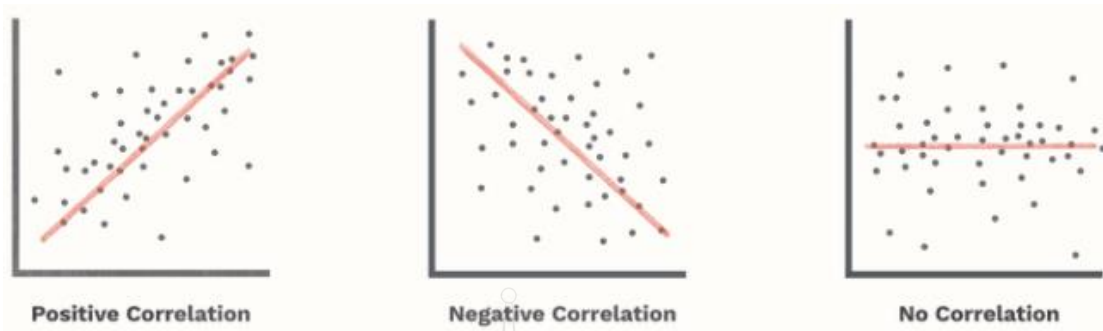
Table 2.8 Structural Comparisons of CCC (CCI), CCF and Box-Behnken Designs for Three Factors

CCC (CCI)				CCF				Box-Behnken			
Rep	X ₁	X ₂	X ₃	Rep	X ₁	X ₂	X ₃	Rep	X ₁	X ₂	X ₃
1	-1	-1	-1	1	-1	-1	-1	1	-1	-1	0
1	+1	-1	-1	1	+1	-1	-1	1	+1	-1	0
1	-1	+1	-1	1	-1	+1	-1	1	-1	+1	0
1	+1	+1	-1	1	+1	+1	-1	1	+1	+1	0
1	-1	-1	+1	1	-1	-1	+1	1	-1	0	-1
1	+1	-1	+1	1	+1	-1	+1	1	+1	0	-1
1	-1	+1	+1	1	-1	+1	+1	1	-1	0	+1
1	+1	+1	+1	1	+1	+1	+1	1	+1	0	+1
1	-1.682	0	0	1	-1	0	0	1	0	-1	-1
1	1.682	0	0	1	+1	0	0	1	0	+1	-1
1	0	-1.682	0	1	0	-1	0	1	0	-1	+1
1	0	1.682	0	1	0	+1	0	1	0	+1	+1
1	0	0	-1.682	1	0	0	-1	3	0	0	0
1	0	0	1.682	1	0	0	+1				
6	0	0	0	6	0	0	0				
Total Runs = 20				Total Runs = 20				Total Runs = 15			

The approach was employed by several researchers for developing and has been used in numerous experimental designs including extraction processes, food preservation, fruit coating and other engineering correction [106]. RSM was used to optimized postharvest treatment in lichee [114], fruit coating in pear and apple [45, 115], guava [116] and Berangan banana [117]. Regarding RSM design in fruit bruising study, as shown in the Table 2.9, the previous studies focused on two independent variable factors such as drop heights and number of drops with a fixed number of drops by using completely randomized design (CRD) design on bruise determination of apple [11], pomegranate [24, 13, 19] and pear [16-17]. In the simulated vibration study,

tomato fruits were analyzed for vibration bruise damage by using RSM design with two independent variable factors such as frequency and duration [25]. Recently, vibration bruising of guava exhibited FD analysis high potential and accuracy for under simulated transportation utilizing RSM design with three independent variable factors of frequency, duration and acceleration [26]. Therefore, no studies have assessed impact bruise damage of guava fruit using RSM to analyze the result data. In this study, RSM was used to determine the optimal conditions for image analysis and impact bruise damage in guava. Optimization using RSM in which the experiment was designed using a central composite face-centered design (CCF).

The strength of the linear link between two variables, x and y , is measured by correlation coefficients (r). A positive relationship is shown by a linear correlation coefficient larger than zero. A negative relationship is indicated by a value smaller than zero. Finally, a value zero implies that the two variables x and y have no relationship. The correlation coefficient can have a range of value from -1.0 to +1.0. In other words, the values cannot more than +1.0 (positive correlation) or be less than -1.0 (negative correlation). A perfect negative correlation is shown by a correlation of -1.0, whereas a perfect positive correlation is indicated by a correlation of +1.0. It is a positive relationship if the correlation coefficient is larger than zero. A negative relationship exists when the value is less than zero. There is no relationship between the two variables, if the value is zero indicates. The term 'positive correlation' refers to a relationship that increase in the same direction. When one variable increases, the other decrease. A negative correlation is the inverse of a opposite correlation. When one variable increases, so does the other, and vice versa. When there is no relationship between the two variables, such that the value of one variable changes while the value of other variable remains constant, this is referred to as no or zero correlation as shown in Figure 2.4 [118].



Source [119]

Figure 2.4 Schematic Presentation of Positive, Negative and No Correlation

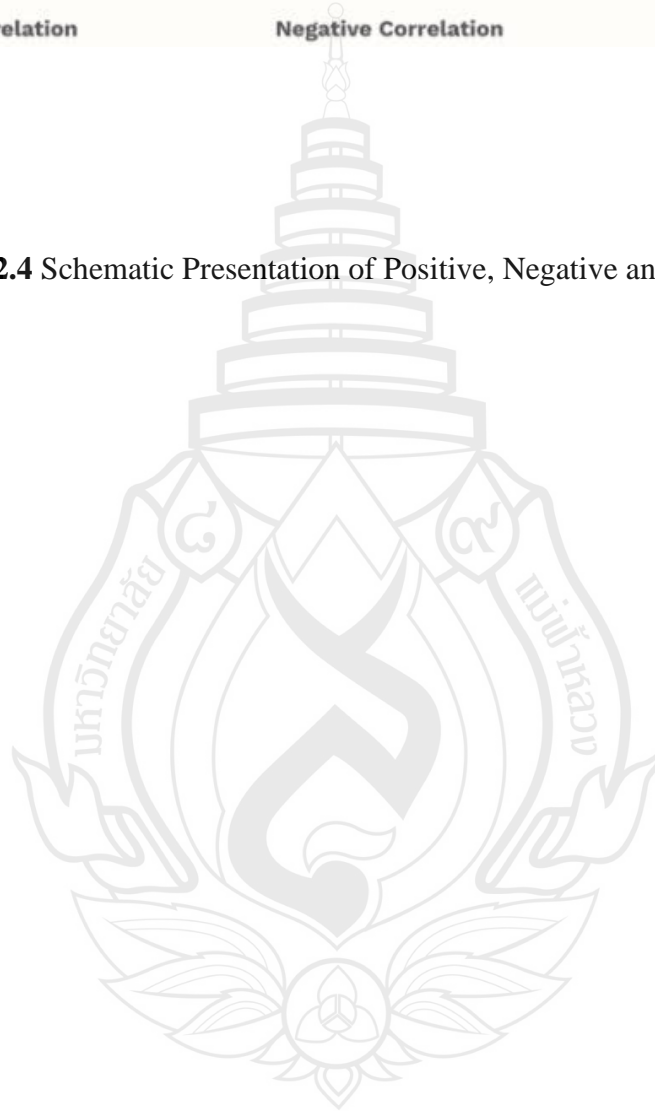


Table 2.9 CRD Experimental Design for Impact Test and RSM Design for Vibration Test with Different Independent Variables

Impact bruise damage					
Fruits	Drop heights (m)	Number of drops (times)	Temperature (°C)	Design	Ref.
Apple	0.25, 0.50, 0.75	1	0,20	CRD	[11]
Pomegranate	0.20, 0.40, 0.60	2	5, 20	CRD	[13]
Pomegranate	0.20, 0.40, 0.60	2	5, 20	CRD	[19]
Pear	0.2, 0.4, 0.6	1	10, 22	CRD	[16]
Pear	0.2, 0.4, 0.6	1	10, 22	CRD	[17]
Vibration bruise damage					
Fruits	Frequency (Hz)	Duration (min)	Acceleration (ms⁻²)	Design	References
Tomato	3, 6, 9	120, 150, 180	n/a	RSM (CCD)	[117]
Guava	7, 13.5, 20	15, 30, 45	2.942, 5.884, 8.826	RSM (CCF)	[26]

Note n/a means not available

CHAPTER 3

METHODOLOGIES

In this chapter, there were two main experiments. The first experiment was entitled “Fractal image analysis and bruise susceptibility evaluation of impact damage in ‘Gim Ju’ guava” that was harvested from Pa-sak village, Chiang Sean District, Chiang Rai Province. Also the second experiment was entitled “Investigation and evaluation of impact bruising in ‘Glom Sali’ guava using image processing and response surface methodology. ‘Glom Sali’ guava fruit was harvested from Pangha Homestay Orchard, Mae Sai District, Chiang Rai Province.

3.1 Fractal Image Analysis and Bruise Susceptibility Evaluation of Impact Damage in ‘Gim Ju’ Guava

3.1.1 Sample Preparation

The ‘Gim Ju’ guava fruit was randomly collected from ‘Poncharoen’ orchard (latitude $20^{\circ} 18' 01.5''N$ and longitude $100^{\circ} 01' 04.9''E$) Pa-sak village, Chiang Sean District, Chiang Rai Province, Thailand. Guava fruits were harvested in the morning, safely packed with foam net and polythene bags and transported to the laboratory S7 building at Mae Fah Luang University within 3 hours. The random ‘Gim Ju’ guava fruit (15 fruit) were selected and examined for fruit properties by checking uniformity, guava maturity, fruit weight, volume, density, diameter, firmness, total soluble solids (TSS) and dry matter. Average weight of guava and density fruit were 250.29 g and 0.73 g mL^{-1} respectively (Table 3.1). Fruit density at <1.0 indicates top quality at the mature stage for harvesting and optimal consumer acceptability [120]. The fruits were selected for uniformity of size as horizontal width 7.76 cm and vertical length 8.03 cm as shown in Table 3.1. All samples were free from defects, diseases and mechanical damages.

Table 3.1 Fruit Characteristics of ‘Gim Ju’ Guava Subjected to Simulated Impact Testing

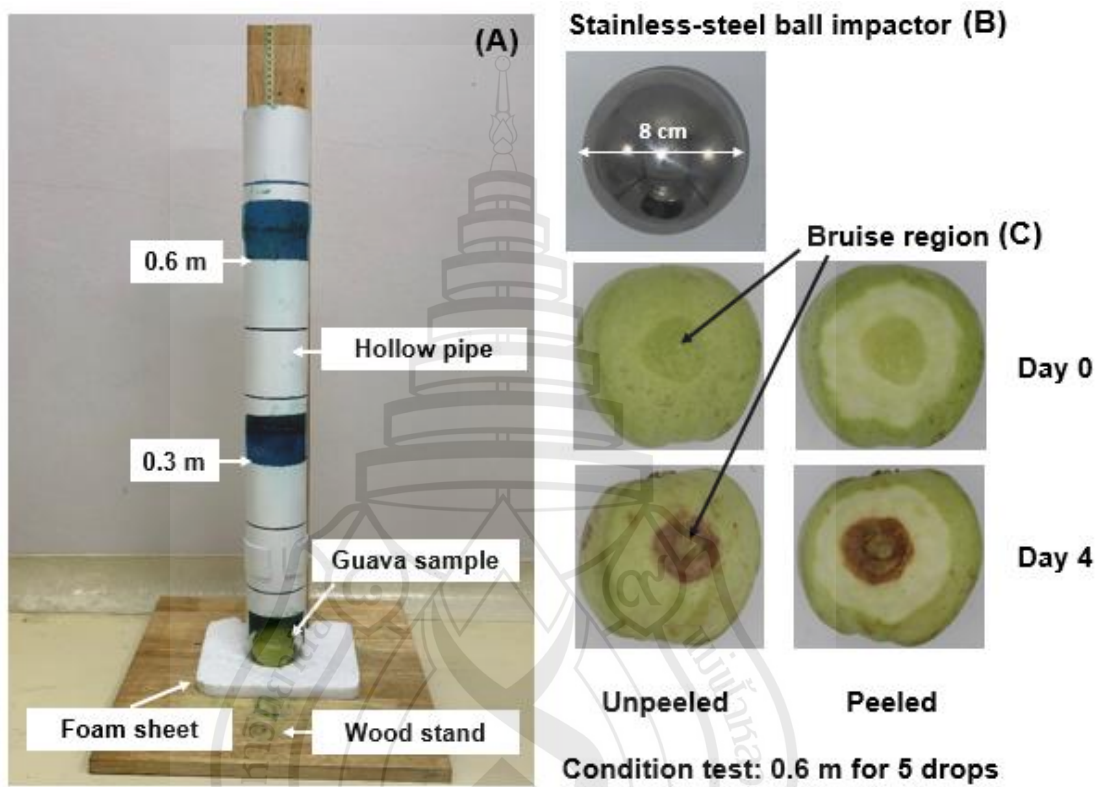
Fruit characteristics	Mean ± SE
Fruit weight (g)	250.29 ± 6.79
Volume (mL)	322.0 ± 7.53
Density (g mL ⁻¹)	0.73 ± 0.02
Horizontal diameter (cm)	7.76 ± 0.10
Vertical length (cm)	8.03 ± 0.11
Firmness (N)	6.12 ± 0.20
Total Soluble Solids (TSS) (%)	6.83 ± 0.11
Dry matter (%)	9.22 ± 0.20

Note Results of fifteen random fruit samples before impact testing (n = 15).

3.1.2 Bruise Susceptibility Testing of Guava Fruit

A simulated impact test was set up using a pipe with a diameter of 9 cm adapted from Hussein [59] for impact testing of pomegranate. Each fruit was placed over a shallow depression (a diameter of 8 cm) in the middle of the foam pad with dimensions 20 × 23 × 2 cm, as shown in Figure 3.1. A stainless-steel ball (diameter 8 cm and weight 250 g) with similar size and weight of a guava fruit was dropped on each guava fruit from different heights and number of times. A completely randomized design (CRD) with five treatments was used with five replicates per treatment. The five treatments involved different drop heights and number of drops as (1) no drop (control), (2) height of 0.3 m for one drop, (3) height of 0.3 m for five drops, (4) height of 0.6 m for one drop and (5) height of 0.6 m for five drops. Five drops were assumed as the probable number during postharvest handling of guava throughout the supply chain [4]. Impact energy (E) from the drop impact was calculated by $E = mgh$, where m is the guava mass, g is the gravitational constant (9.81 m s⁻²) and h is the drop height (m) [59]. The impact energy level of a stainless-steel ball for five treatments was calculated as shown in Table 3.2. The highest impact energy value from a height of 0.6 m for five drops was 7357.50 J, followed by 3678.75 J (0.3 m + 5 drops), 1471.50 (0.6 m + 1 drop) and

735.75 J (0.3 m + 1 drop). After impact testing, the fruits were stored at room temperature (25 °C under 70% RH) and checked for quality measurement every day (24 hours) for four days.



Source [59]

Figure 3.1 Experimental Setup of Simulated Laboratory Bruise Impact Test (A), Stainless-Steel Impactor (250 g, Diameter 8 cm) (B) and Bruise Region of Guava on Day 0 and 4 (C)

Table 3.2 Impact Energy for Different Drop Heights and Number of Drops

Treatment	Impact energy (J)
Control (no drop)	0.00
0.3 m + 1 drop	735.75
0.3 m + 5 drops	3678.75
0.6 m + 1 drop	1471.50
0.6 m + 5 drops	7357.50

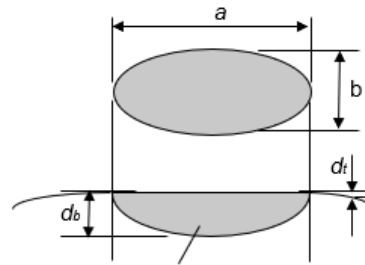
Before impact bruising determination, the bruised guava sample from the simulated impact test was peeled using a sharp knife with peel-off thickness of approximately 0.9 mm to reveal browning of the flesh (Figure 3.1).

Bruise area (BA) and bruise volume (BV): BA and BV are commonly used to measure the amount of fruit bruise damage. The BA and BV of each fruit were determined based on impact bruising in apple [51] and calculated by equations (6) and (7).

$$BA = \pi/4 (ab) \quad (6)$$

$$BV = \pi d/24 (3ab + 4d^2) \quad (7)$$

where a and b are the major axes of the bruise elliptical and d is bruise depth measured from peel thickness [12]. These bruise parameters, introduced in Figure 3.2, were measured with a digital caliper (RS PRO 150 mm, RS Components Pte Ltd Robinson Road, Singapore) with ± 0.01 (mm) accuracy.



Source [21]

Figure 3.2 Elliptical Methods for Determining BA and BV

Bruise susceptibility (BS): BS (mm^3J^{-1}) was calculated as the ratio of BV (mm^3) to the impact energy E (J) by equation (8) [66, 19, 72].





$$\text{BS} = \text{BV}/E \quad (8)$$

Specific bruise susceptibility (SBS): To measure the index of BS, SBS ($\text{mm}^3\text{J}^{-1}\text{g}^{-1}$) was determined by equation (9) [72].

$$\text{SBS} = \text{BS}/ \text{fruit weight} \quad (9)$$

Bruise score: The peeled guava was visually assessed for bruise score adapted from the bruise score of apple [62]. Based on a 4 point scale as 1 No apparent bruise, 2 Light bruise with no defined edge, 3 Moderately dark brown bruise with a well-defined edge, and 4 Dark brown bruise, as shown in Table 3.3.

Table 3.3 Bruise Damage Visual Rating Score

Rating score	Description	Image
1	No apparent bruising	
2	Light brown bruise with no defined edge	
3	Moderately dark brown bruise with a well-defined edge	
4	Dark brown bruise	

3.1.3 Pulp Color at Bruising Region

A colorimeter (model: CR-10 color reader, Osaka, Japan) was used to measure bruise lightness (L^*), redness (a^*), yellowish (b^*) and chroma (C) based on CIELab; L^* (lightness) as black (0) and white (100), a^* (red to green) as red (positive value) and green (negative value) and b^* (yellow to blue) as yellow (positive value) and blue (negative value). The chroma (C) defines color saturation [62].

3.1.4 Image Analysis

The bruise region on the fruit surface after impact testing was placed under a square light box (UDI OBIZ 40D), size $40 \times 40 \times 40$ cm, adjustable light, pocket studio with 4 rows of LED as light source intensity 9.6×10^5 lux using a light meter (Tenmars TM-204, Taipei, Taiwan). Guava images were taken at a uniform distance of 40 cm, with camera settings on manual mode, autofocus, lens capture f 7.1, 1/250 shutter speed, and ISO 100 using a digital mirrorless camera (Canon EOS M50, 15–45 mm, Tokyo, Japan). The original image files (6000×3368 pixels) were saved in JPEG format with 72-dpi resolution. Guava bruise damage images were analyzed by ImageJ software (version 1.51j8, NIH, Bethesda, MD, USA) following the method of

Chaiwong et al. [26]. All original images were pre-processed and resized. The image analysis procedure is shown in Figure 3.3. The resized image was cropped around the bruising area, which is the region of interest (ROI). After that, the RGB image was converted into an 8-bit image (grayscale), then the surface plot of bruising image was determined by plotting the grayscale values (z-axis) of each pixel in the image (x- and y-axis). The surface plot showed the roughness of bruise surface texture due to the variation of grayscale level in each pixel of the image. Moreover, the 8-bit image was thresholded to convert to be the binary image. Then, the binary image of bruising was computed as a percentage of the bruise region corresponding to the entire flesh area.

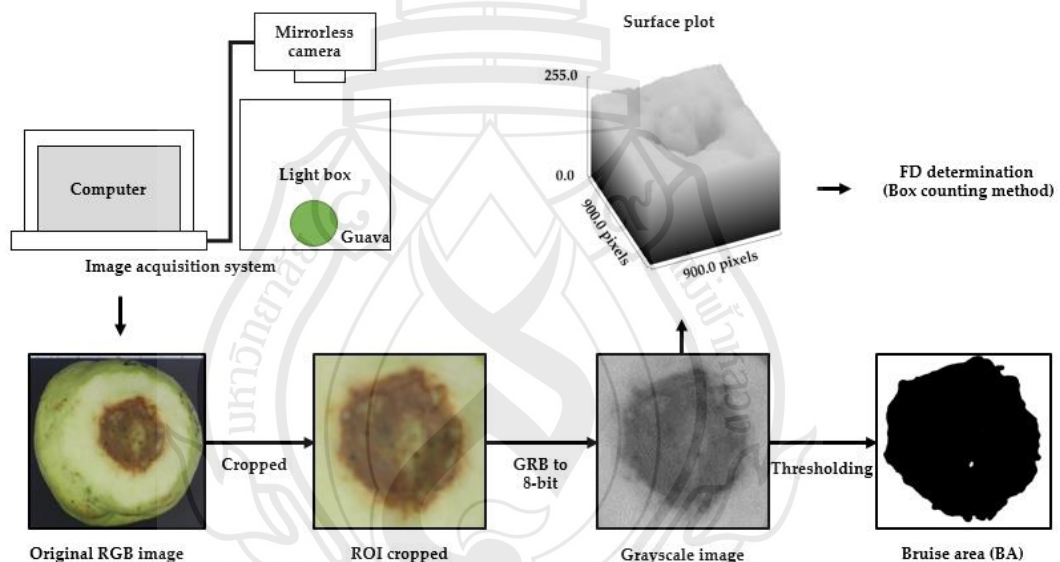


Figure 3.3 Image Acquisition System and Image Analysis Procedure for Impact Bruise Determination

The intensity of fruit bruise damage was estimated using the binary image. The Fractal dimension (FD) values of the bruise images were then determined using the fractal box counting technique by intercepting N number of various box size r (2–128 pixels) with the binary image. FD value was calculated as $FD = \log(Nr)/\log(1/r)$ [26].

3.1.5 Model Prediction and Validation Testing

FD value was determined and polynomial fitting with coefficient determination value (R^2) for five treatments followed equation (10).

$$Y = b_0X^2 + b_1X + b_3 \quad (10)$$

where Y is the predicted FD value, X is storage time (day), X^2 is the square of X and b_0 and b_1 are constants.

Fruit samples with three impact bruises (replicates) in each treatment were used to validate testing for five polynomial equations with R^2 validation (R^2_{val}). Validation of impact bruising in guava focused on the FD values from image analysis.

3.1.6 Data Analysis

SPSS for Windows version 20 (SPSS Inc., Chicago, IL, USA) was used for statistical analysis. Data analyses for impact bruising parameters, color indices and FD values were compared by means ($\alpha = 0.05$) using Tukey's HSD post hoc test. Fractal image analysis of impact bruise incidence was investigated using ImageJ software (version, 1.51j8). Impact bruising parameters, color indices and FD values were analyzed using Pearson's correlation coefficient (r) at significant difference $p < 0.01$. All data variables were compared by a heatmap of Pearson correlation analysis on the web interface for MetaboAnalyst 5.0 Online, freely available at <https://www.metaboanalyst.ca/>. Red and blue color tones indicate positive and negative correlation, respectively.

3.2 Investigation and Evaluation of Impact Bruising in 'Glom Sali' Guava Using Image Processing and Response Surface Methodology

3.2.1 Plant Materials and Sample Preparation

Guava fruit cv. 'Glom Sali' were randomly collected from Pangha Homestay Orchard, Mae Sai District, Chiang Rai Province, Thailand (latitude 20°24'22.7"N

longitude 100°00'23.9"E). The guava fruit were harvested at the mature green stage (approximately 100 days after fruit set) having a light green skin color and carefully handled to avoid mechanical injuries. The fruit were packed into plastic baskets and covered with two material layers of foam net and polythene bags as cushioning materials to protect against mechanical injuries during transportation to the laboratory S7 building at Mae Fah Luang University within 3 h. The fruit were sorted for uniformity of size (approximately 250 to 300 g), color, and if they were free of distinct signs of bruising and disease with a smooth and clean peduncle cut surface.

The random 'Glom Sali' guava fruit (15 fruits) were selected and examined for fruit properties by checking uniformity, guava maturity, fruit weight, volume, density, diameter, radius of curvature, firmness, total soluble solids (TSS), and dry matter. Average weight of guava and density fruit were 250.26 g and 0.98 g mL⁻¹ (Table 3.4). Fruit density at <1.0 indicates top quality at the mature stage for harvesting and optimal consumer acceptability. The highest bruise susceptibility was occurred in the cheek region of the fruit [24], while the middle and top regions of apple showed higher bruise sensitivity than the bottom region for fruit-to-fruit bruising [121]. In this study, the cheek region was exposed to the highest impact under free fall drop test. The averaged radius of curvature at the cheek region of this guava fruit was 50.94 mm (Table 3.4).

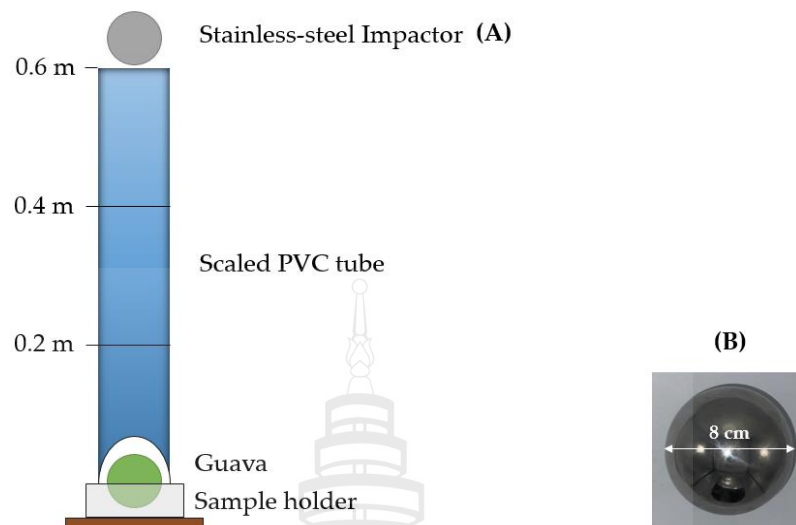
Table 3.4 Fruit Characteristics of ‘Glom Sali’ Guava Subjected to Simulated Impact Testing

Fruit Characteristics	Mean ± SE
Fruit weight (g)	250.26 ± 1.50
Volume (mL)	235.33 ± 7.81
Density (g mL ⁻¹)	0.98 ± 0.02
Horizontal diameter (cm)	7.65 ± 0.07
Height (cm)	8.72 ± 0.12
Radius of curvature (mm)	50.94 ± 1.44
Firmness (N)	5.09 ± 0.31
Total Soluble Solids (TSS) (%)	11.51 ± 0.30
Dry matter (%)	13.32 ± 0.30

Note Results of fifteen random fruit samples before impact testing (n = 15).

3.2.2 Bruise Susceptibility Testing of Guava Fruit

Simulated impact testing was set up utilizing a 9 cm diameter pipe, following the method of Hussein et al. [59] for pomegranate impact testing. As shown in Figure 3.4, each fruit was located over a shallow depression (8 cm diameter) in a foam sheet material with dimensions 20 × 23 × 2 cm. A stainless-steel ball (diameter 8 cm and weight 250 g) of equal size and weight to a guava fruit was dropped on each guava fruit from three different drop heights (0.2, 0.4, and 0.6 m) for 1, 3, and 5 drops at each drop height.



Source [59]

Figure 3.4 Experimental Setup of Simulated Impact Testing in Guava (A) with Stainless-Steel Ball (250 g, Diameter 8 cm) (B)

The impact energy (E) was calculated using the equation of $E = mgh$, where m is the guava mass, g is the gravitational constant (9.81 m s^{-2}), and h is the drop height (m) [59]. As shown in Table 3.5, the impact energy of a stainless-steel ball for 1, 3, and 5 drops at each of the three drop heights ranged from 507.05 to 7273.60 J. After impact testing, the fruit were stored at three temperature conditions (10, 20, and 30 °C under 90% RH) and checked for quality measurement after 48 h (see Section 3.2.3 for more information).

Table 3.5 Impact Energy for Three Drop Heights (0.2, 0.4, and 0.6 m) and Number of Drops (1, 3 and 5 Times)

Treatment	Impact Energy (J)
0.2 m + 1 drop	507.05
0.2 m + 3 drops	1374.96
0.2 m + 5 drops	2529.00
0.4 m + 1 drop	1141.49
0.4 m + 3 drops	2950.29
0.4 m + 5 drops	4995.25
0.6 m + 1 drop	1590.40
0.6 m + 3 drops	4040.16
0.6 m + 5 drops	7273.60

After simulated impact testing, the bruised guava sample was peeled using a sharp knife with peel thickness of 0.9 mm to reveal browning of the flesh before impact bruising was determined.

3.2.3 Experimental Design by Response Surface Methodology (RSM)

RSM with central composite face-centered design (CCF) was examined to evaluate the main interaction and quadratic effects of drop height, number of drops, and temperature on impact bruising and color change of the guava fruit (Table 3.6).

Table 3.6 Coded Independent Variables of Drop Heights, Number of Drops, and Storage Temperature for RSM with Central Composite Face-Centered Design (CCF)

Independent Variable	Code	Coded Variable		
		-1	0	+1
Drop height (m)	X ₁	0.2	0.4	0.6
Number of drops (times)	X ₂	1	3	5
Temperature (°C)	X ₃	10	20	30

As shown in Table 3.7, the three independent variables of CCF design with 20 treatments (runs) were coded as -1 (lowest level), 0 (middle level), and +1 (highest level), and applied with the cube point for 8 times, axial point for 6 times, and the repeated center point (0.0.0) for 6 times. The three independent variables were drop height (X₁) (0.2, 0.4 and 0.6 m), number of drops (X₂) (1, 3 and 5 times), and storage temperature (X₃) (10, 20 and 30 °C). In this study, drop heights (0.2, 0.4 and 0.6 m) were compared with a range of drop heights (0.04 to 0.6 m) onto a rigid impact surface in previous studies on pomegranate [13-14, 19], apple [9] and pear [16-17]. Five drops were assumed to be the maximum number throughout the guava supply chain [4]. The optimal storage temperature for guava was recommended at 10 °C [23], while temperature during road transportation in open trucks was estimated to average 30 °C. The response function was determined as bruise area (BA), bruise volume (BV), browning index (BI), total color difference (ΔE), bruise area by image analysis (BAI), and fractal dimension (FD). The response function value (Y) was related to the coded three independent variables (X_i, 1, 2 and 3) following Equation (11).

Table 3.7 RSM with Central Composite Face-Centered Design (CCF) to Optimize Simulated Impact Conditions of ‘Glom Sali’ Guava

Run	Code			Treatment Factors		
	X ₁	X ₂	X ₃	Drop heights (m)	No. of Drops (times)	Temperature (°C)
1	-1	-1	-1	0.2	1	10
2	-1	-1	+1	0.2	1	20
3	-1	+1	-1	0.2	1	30
4	-1	+1	+1	0.2	5	30
5	+1	-1	-1	0.6	1	10
6	+1	-1	+1	0.6	1	30
7	+1	+1	-1	0.6	5	10
8	+1	+1	+1	0.6	5	30
9	0	0	-1	0.4	3	10
10	0	0	+1	0.4	3	30
11	0	-1	0	0.4	1	20
12	0	+1	0	0.4	5	20
13	-1	0	0	0.2	3	20
14	+1	0	0	0.6	3	20
15	0	0	0	0.4	3	20
16	0	0	0	0.4	3	20
17	0	0	0	0.4	3	20
18	0	0	0	0.4	3	20
19	0	0	0	0.4	3	20
20	0	0	0	0.4	3	20

Response data of the three independent variables to the six dependent variables as BA, BV, BI, ΔE , BAI, and FD were examined, and the quadratic polynomial fitting was accomplished. The equation of the prediction model is shown as polynomial Equation (11).

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_{11}X_1^2 + b_{22}X_2^2 + b_{33}X_3^2 + b_{12}X_1X_2 + b_{23}X_2X_3 + b_{13}X_1X_3 \quad (11)$$

Where, Y is the representation of the predicted response functions (BA, BV, BI, ΔE , BAI and FD), X_1 , X_2 , and X_3 are the three independent variables (drop heights, number of drops, and storage temperature), X_1^2 , X_2^2 and X_3^2 are the square of each independent variables. The coefficients of the polynomial were denoted by b_0 (constant term), b_1 , b_2 and b_3 (linear effects), b_{11} , b_{22} and b_{33} (quadratic effects), and b_{12} , b_{13} and b_{23} (interaction effects).

3.2.4 Quality Measurements

3.2.4.1 Bruise Determination of the Guava Fruit

After impact testing, the guava fruit were stored at 10, 20 and 30 °C with 90% RH (Constant climate chamber, HPP750, Memmert GmbH + Co. KG, Schwabach, Germany) and checked for quality measurement after 48 h when the bruised tissues had turned pale [28]. BA and BV, commonly used to measure the amount of fruit bruise damage, were determined for fruit damage in apple [51]. BA and BV of each fruit were calculated by Equations (6) and (7).

3.2.4.2 Pulp Color at Bruise Area

The guava was peeled to reveal either wet bruising (translucent) or browning incidence at four positions with three positions at no bruising region and one position at middle of bruising region that were measured in the equation zone for each fruit by a CIE-Lab color colorimeter (Konica Minolta, Color Reader CR-10, Osaka, Japan) to determine bruise lightness (L^*), redness (a^*), yellowish (b^*). Browning index (BI) was calculated to represent purity of the brown color [122] using Equations (12) and (13). Total color difference (ΔE) was calculated using different color levels on fruit peel (L^* , a^* , and b^* values) between initial (before impact bruise) [88] and after impact damage for 48 h at the bruise area using Equation (14).

$$BI = [100 * (X-0.31)]/0.172 \quad (12)$$

$$\text{When, } X = (a^* + 1.75L^*)/(5.645L^* + a^* - 3.012b^*) \quad (13)$$

$$\Delta E = ((L_0^* - L^*)^2 + (a_0^* - a^*)^2 + (b_0^* - b^*)^2)^{1/2} \quad (14)$$

Where, L_0^* , a_0^* , and b_0^* are the initial color values of the peeled fruit, L^* , a^* , and b^* are the final values at bruised area after storage at 10, 20, and 30 °C with 90% RH for 48 h.

3.2.4.3 Image Analysis

Image analysis and FD value for ‘Glom Sali’ guava impact bruise damage was done according to the method described in section 3.1.4.

3.2.5 Statistical Analysis

The effect of each of the three independent factors, lack-of-fit, and regression coefficients (R^2) in individual linear, quadratic, and interaction terms was determined in analysis of variance (ANOVA) tables and generated to establish the model equation by Minitab version 19 (Minitab, LLC, State college, PA, USA). Three-dimensional response surface plots were created using the overlaid contour plot feature in MATLAB software version R2018a (MathWorks Inc., Natick, MA, USA). Predictive equations for the thirteen treatments within the experimental range conditions were checked for correctness (Table 3.8). To generate a matrix plot using PAST 4.05 software, all variables were evaluated using Pear-son’s correlation ($p < 0.05$).

Table 3.8 Thirteen Treatments to Verify the Predictive Model in ‘Glom Sali’ Guava Impact Bruising by Simulated Impact Testing

Run	Code			Treatment Factors		
	X ₁	X ₂	X ₃	Drop Heights (m)	No. of Drops	Temperature (°C)
1	-1	-1	-1	0.2	1	10
2	-1	+1	-1	0.2	1	30
3	-1	+1	+1	0.2	5	30
4	+1	+1	+1	0.6	5	30
5	0	0	-1	0.4	3	10
6	0	0	+1	0.4	3	30
7	0	-1	0	0.4	1	20
8	0	+1	0	0.4	5	20
9	-1	0	0	0.2	3	20
10	+1	0	0	0.4	3	30
11	0	0	0	0.4	3	20
12	0	0	0	0.4	3	20
13	0	0	0	0.4	3	20

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Fractal Image Analysis and Bruise Susceptibility Evaluation of Impact Damage in ‘Gim Ju’ Guava

4.1.1 Pulp Color at Bruising Region

Results in Figure 4.1 show that an increase in drop height and number of drops significantly affected the values of all color attributes (L^* , a^* , b^* and C) ($p < 0.05$). Control treatment (no drop) had significantly higher values of L^* , a^* , b^* and C than the other four treatments after storage at 25 °C under 70% RH for four days ($p < 0.05$). After fruit impact damage, enzymatic browning reaction and resulting discoloration occurred inside cells, following cell membrane damage and release of cell contents into intercellular spaces [123]. In this study, five drops from a height of 0.3 and 0.6 m showed the lowest color values (L^* , a^* , b^* and C) concurring with results of highest impact BS (Table 3.3). Major bruise damage to guava was caused by the number of drops, resulting in the highest peel color changes compared with the control (no impact bruise). The number of drops was also a major factor affecting impact damage in pomegranate fruit. Drop heights of 0.2, 0.4, 0.6 m were tested twice and compared with the control (no drop). No significant difference in bruise score was recorded between the height of 0.2 m (twice) and no drop [13]. Drop conditions in ‘Montenegrina’ tangerines were tested at heights of 0.4 and 0.6 m or 0.8 and 1 m, twice at each height. No significant differences in hue value were found at drop heights of 40 and 60 cm or drop heights of 0.8 and 1 m [81]. Impact damage browning of guava fruit was investigated by L^* , a^* and b^* values (Figure 4.1). A higher correlation coefficient (r) was determined between bruise parameters and L^* , a^* and b^* color values than the C value (Table 4.2). In this study, major and minor factors of impact guava bruising were

the number of drops and drop heights, respectively. Therefore, postharvest handling of guava should be conducted with care.

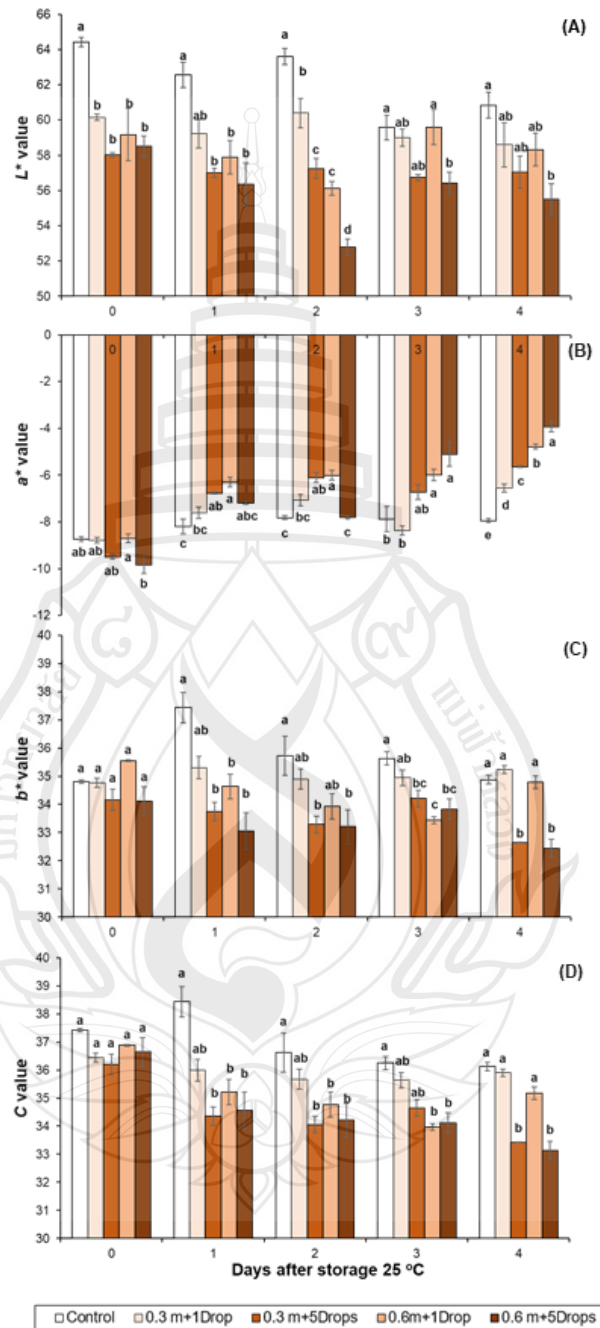


Figure 4.1 Pulp Color (A) L^* , (B) a^* , (C) b^* and (D) C Values of Bruise Damage of Guava Fruit from Day One to Day Four After Storage at 25 °C Under 70% RH

4.1.2 Bruise Susceptibility of Guava Fruit

The accumulation of impact energy by dropping the steel ball from different heights and number of drops at one or five significantly affected the increase of BA and BV in guava fruit (Tables 3.2 and 4.1). Most drop height tests in round fruit (apple and pomegranate) were examined in a range of 0.05 to 0.3 m [51, 59, 61]. In this study, guava fruit weight for sample testing showed no significant difference. The drop test from 0.6 m drop height with five drops had the highest BA, BV, BS, SBS values and bruise score, followed by dropping from 0.3 m with five drops, dropping from 0.6 m with one drop and from 0.3 m with one drop (Table 4.1). Bruise results indicated the highest impact energy at 0.6 m for five drops (Table 3.2). This finding concurred with previous impact fruit studies by dropping from different heights and number of drops [51, 59, 61]. Increasing the drop height (or impact energy) elevated the potential for bruise damage in pomegranate [19], kiwifruit [20] and peach [21]. Drop height and number of drops also influenced the increase of bruise susceptibility in apple [51], pomegranate [59] and tomatoes [124].

4.1.3 Image Analysis for Bruise Determination

The FD values were analyzed by converting cropped RGB images and calculated to view the intensity of fruit bruise damage. The bruise damage region was cropped and then separated using the threshold method. The selected BA was converted into a binary image before determining FD values using a box-counting method. Image analysis for impact damage of guava after the drop test and storage at 25 °C under 70% RH for two days and four days is shown in Figure 4.2. Impact damage of the pulp surface was visibly presented in BA and BV by image analysis. A greater impact bruise showed as a deeper surface plot image (Figure 4.2), relating to increased impact energy (Table 3.2) and period of storage (Figure 4.2).

Using image analysis, FD values were determined for lightness and darkness of the surface [30] to assess the bruising or browning of the fruit. Fractal modeling was effectively applied to determine the intensity of flesh browning and its color change to provide a better understanding of enzymatic chemical changes and their location within fruit flesh [31].

The FD value of impact bruising in guava gradually decreased with the increase of all bruise parameters and time duration after storage, except for the control (no drop) (Figure 4.3 and Table 4.1). The control exhibited the highest FD values (1.944 and 1.942) than the other four impact bruise treatments. The lowest FD value was observed at the two height levels of 0.3 m (1.930) and 0.6 m (1.932) with five drops (Figure 4.3). Most previous researchers conducted fruit quality evaluation using the FD method to assess internal browning, color change in flesh and defects in pear [29], banana [30] and apple [31]. The FD method showed accuracy for avocado fruit browning [95], while increased uniformity of the browning area resulted in decreased FD value in pineapple [125]. A non-homogenous change in color intensity distribution resulted in a higher FD value, while homogenous change resulted in a lower FD value in avocado fruit [95]. However, no studies of the FD method for impact bruise evaluation have been conducted for guava. To the best of our knowledge, this is the first study reporting the advantage of FD techniques for impact bruise analysis in guava. Results indicated that FD analysis has the potential for the determination of impact bruising in guava for different bruise severities and storage times.

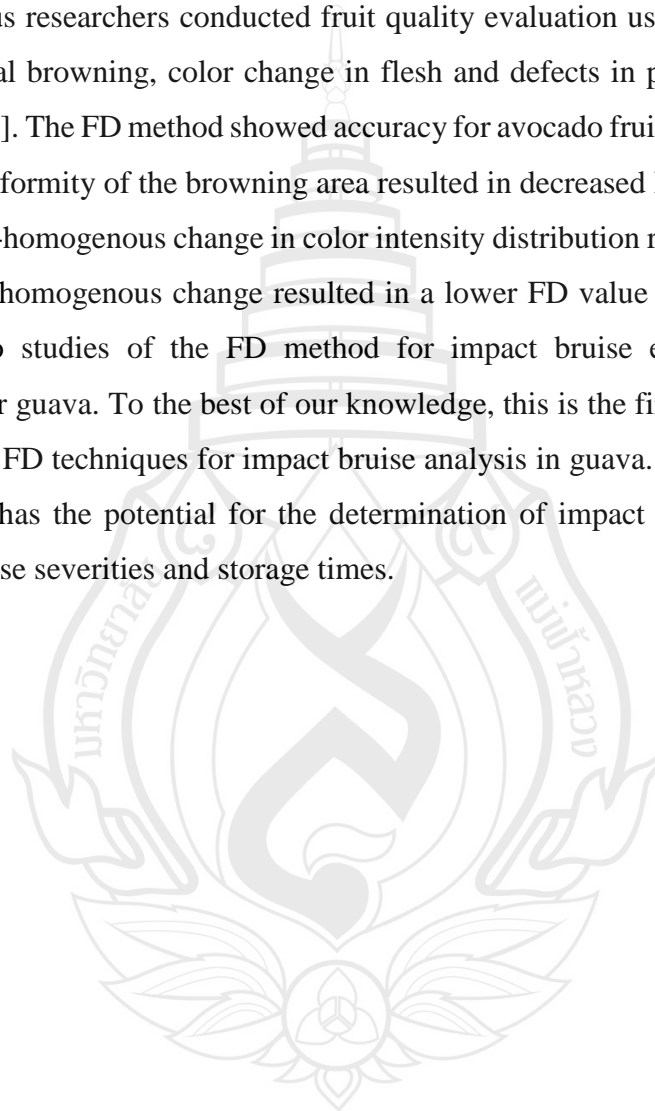


Table 4.1 Bruise Assessment Parameters of Guava Fruit Stored at 25 °C under 70% RH for Four Days

Days after storage at 25 °C	Treatment	Fruit mass (g)	Bruise area (BA) (mm ²)	Bruise volume (BV) (mm ³)	Bruise susceptibility (BS) (mm ³ J ⁻¹)	Specific bruise susceptibility (SBS) (mm ³ J ⁻¹ kg ⁻¹)	Bruise score
1	Control (no drop)	255.32±19.50 ^a	0.0 ± 0.0 ^d	0.0 ± 0.0 ^c	0.00±0.00 ^d	0.000±0.000 ^d	1.0 ± 0.0 ^d
	0.3 m + 1 drop	247.04±10.13 ^a	149.3 ± 4.4 ^c	712.0 ± 74.9 ^c	0.97±0.10 ^{cd}	0.004±0.000 ^{cd}	2.4 ± 0.2 ^c
	0.3 m + 5 drops	237.36±8.54 ^a	281.0 ± 55.3 ^b	1,906.2 ± 374.3 ^b	2.57±0.51 ^b	0.011±0.002 ^b	3.2 ± 0.2 ^b
	0.6 m + 1 drop	257.68±15.18 ^a	329.9 ± 18.4 ^b	2,069.5 ± 205.1 ^b	1.41±0.14 ^c	0.006±0.001 ^c	3.4 ± 0.2 ^{ab}
	0.6 m + 5 drops	266.72±9.08 ^a	633.0 ± 24.1 ^a	6,131.2 ± 281.7 ^a	4.17±0.19 ^a	0.016±0.001 ^a	4.0 ± 0.0 ^a
2	Control (no drop)	246.8±14.02 ^a	0.0 ± 0.0 ^d	0.0 ± 0.0 ^d	0.00±0.00 ^d	0.000±0.000 ^b	1.0 ± 0.0 ^c
	0.3 m + 1 drop	247.16±7.49 ^a	151.0 ± 15.9 ^c	790.3 ± 113.8 ^d	1.07±0.15 ^c	0.004±0.001 ^b	2.4 ± 0.2 ^b
	0.3 m + 5 drops	249.7±15.13 ^a	406.0 ± 36.0 ^b	3,116.9 ± 325.4 ^b	4.24±0.44 ^a	0.017±0.003 ^a	3.4 ± 0.2 ^a
	0.6 m + 1 drop	278.68±9.77 ^a	315.4 ± 27.9 ^b	2,164.5 ± 151.9 ^c	1.47±0.10 ^c	0.005±0.001 ^b	3.6 ± 0.2 ^a
	0.6 m + 5 drops	227.76±10.75 ^a	596.3 ± 50.4 ^a	4,483.3 ± 305.0 ^a	3.05±0.21 ^b	0.014±0.001 ^a	4.0 ± 0.0 ^a
3	Control (no drop)	279.16±9.59 ^a	0.0 ± 0.0 ^d	0.0 ± 0.0 ^c	0.00±0.00 ^c	0.000±0.000 ^c	1.0 ± 0.0 ^c
	0.3 m + 1 drop	252.4±12.58 ^a	137.4 ± 5.9 ^c	700.2 ± 74.4 ^c	0.95±0.10 ^b	0.004±0.000 ^b	2.2 ± 0.2 ^b
	0.3 m + 5 drops	247.14±14.79 ^a	310.5 ± 35.8 ^b	1,993.5 ± 295.7 ^b	2.71±0.40 ^a	0.011±0.001 ^a	3.4 ± 0.2 ^a
	0.6 m + 1 drop	241.7±18.27 ^a	279.8 ± 28.0 ^b	1,515.8 ± 228.4 ^b	1.03±0.16 ^b	0.004±0.001 ^b	3.6 ± 0.2 ^a
	0.6 m + 5 drops	247.08±6.10 ^a	562.7 ± 20.1 ^a	3,896.1 ± 197.7 ^a	2.65±0.13 ^a	0.011±0.001 ^a	4.0 ± 0.0 ^a
4	Control (no drop)	268.24±7.36 ^a	0.0 ± 0.0 ^c	0.0 ± 0.0 ^b	0.00±0.00 ^c	0.000±0.000 ^c	1.0 ± 0.0 ^c
	0.3 m + 1 drop	264.52±11.31 ^a	119.0 ± 9.2 ^c	546.0 ± 46 ^{cd}	0.74±0.06 ^{bc}	0.003±0.000 ^c	2.4 ± 0.2 ^b
	0.3 m + 5 drops	248.18±12.83 ^a	334.9 ± 37.1 ^b	1,937.6 ± 280.1 ^b	2.63±0.38 ^a	0.011±0.002 ^{ab}	3.4 ± 0.2 ^a
	0.6 m + 1 drop	251.78±12.63 ^a	315.1 ± 27.1 ^b	1,847.6 ± 64.0 ^{bc}	1.26±0.04 ^b	0.005±0.000 ^{bc}	3.6 ± 0.2 ^a
	0.6 m + 5 drops	235.26±17.0 ^a	615.6 ± 74.5 ^a	4,982.6 ± 631.8 ^a	3.39±0.43 ^a	0.015±0.002 ^a	4.0 ± 0.2 ^a

Note Mean values presented as mean ± SE in the same column followed by a different superscript letter is significantly different ($p < 0.05$) using

Tukey's HSD post hoc test.

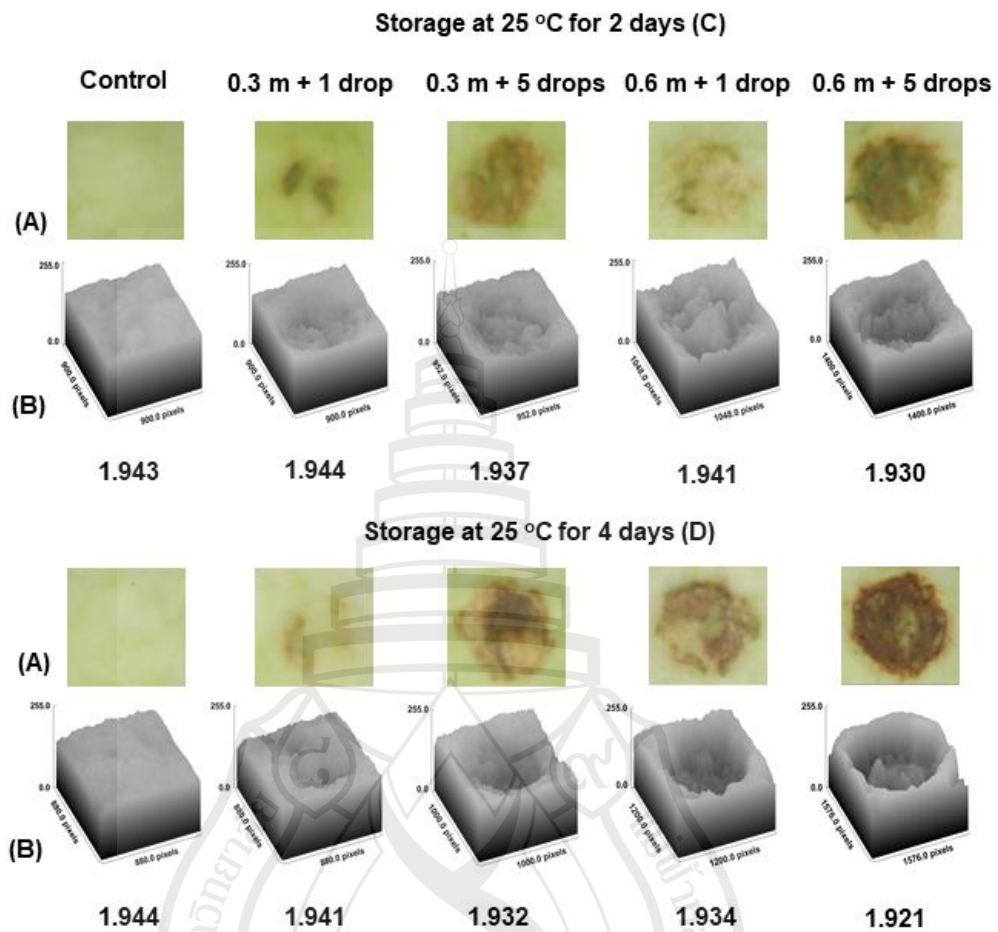


Figure 4.2 Image Analysis for FD Value of the Cropped Image (A) and Surface Plot (B) of Impact Damage to Guava Fruit After Drop Test and Storage at 25 °C Under 70% RH for Two Days (C) and Four Days (D)

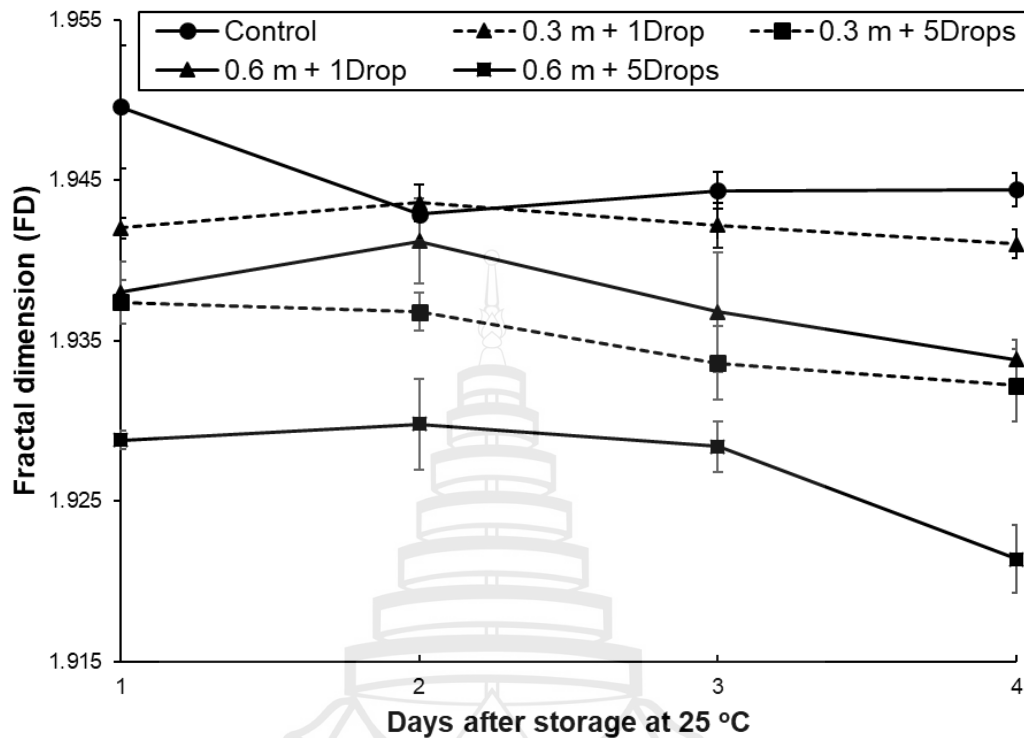


Figure 4.3 Impact Bruise Damage of Guava Fruit Based on FD

4.1.4 Correlation between Color Features and Bruise Indices

Results of the correlational analysis are compared in Figure 4.4. The correlation coefficient (r) between FD and bruise parameters was higher than for all color values (L^* , a^* , b^* and C), particularly BA ($r = -0.794$) and BV ($r = -0.745$), while L^* , a^* and b^* values showed the greater correlation between bruise parameters than C values. FD value indicated a good correlation between impact bruise damage in guava than all color parameters. This finding supported previous research into the relationship between FD, L^* and environmental exposure, showing patterns of redness spreading related to non-homogenous color changes on the surface of papaya slice, clearly exhibited by 65% ripe papaya [126]. Lightness parameters decreased when FD value increased, indicating greater complexity in the distribution of L^* values in the selected area analyzed during enzymatic browning in apples [31]. Thus, more research in the impact bruising of guava or other fruit is required to determine the efficacy of FD techniques for mechanical damage caused by different external factors in fruit bruise sensitivity.

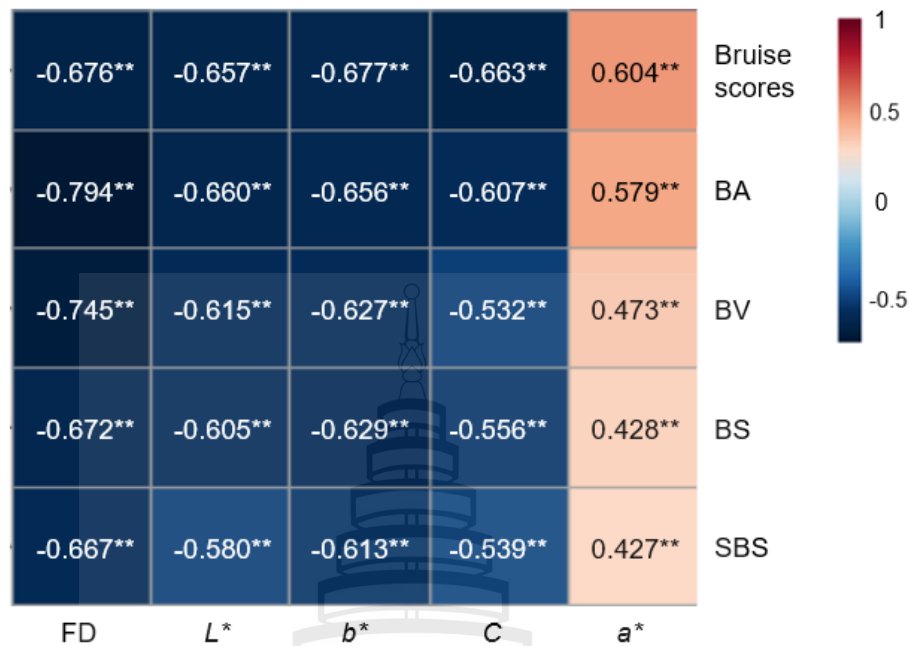


Figure 4.4 Heatmap of Pearson Correlation Coefficients (r) between Bruise Assessment Parameters of Guava Fruit and Image Analysis of FD and L^* , a^* , b^* and C Values with a Significant Difference Set at ($p < 0.01$)

4.1.5 Model Prediction and Validation Testing of FD

A good polynomial equation relationship was obtained between FD for impact bruise assessment of guava fruit in each treatment during storage for four days (Figure 4.3 and Table 4.2). The lowest R^2 value (0.800) occurred in the control (no drop) due to no impact bruise damage on guava pulp. Among the other four treatments, five drops from heights of 0.3 and 0.6 m (0.948 and 0.988) had higher R^2 values than a single drop (0.851 and 0.855), respectively. The validation results of five R^2_{val} values were 0.87 (control), 0.83 (0.3 m + 1 drop), 0.88 (0.3 m + 5 drops), 0.76 (0.6 m + 1 drop) and 0.92 (0.6 m + 5 drops). Thus, a higher both R^2 and R^2_{val} at five drops exhibited a higher accuracy for bruise prediction by FD value with greater bruise susceptibility (Table 4.2). For the type of prediction modeling in fruit bruising, linear regression showed assessment and prediction with a high R^2 value (>0.91) in apple [31], kiwifruit [20] and tomato [61]. A good linear regression equation for bruise assessments was also

observed [61, 20, 127]. However, no reports are available showing the polynomial equation relationship between FD and impact bruise damage assessment in other fruit.

Table 4.2 Coefficient of Determination (R^2 and R^2_{val}) and Polynomial Equations of Guava Fruit for Different Drop Heights and Numbers of Drops

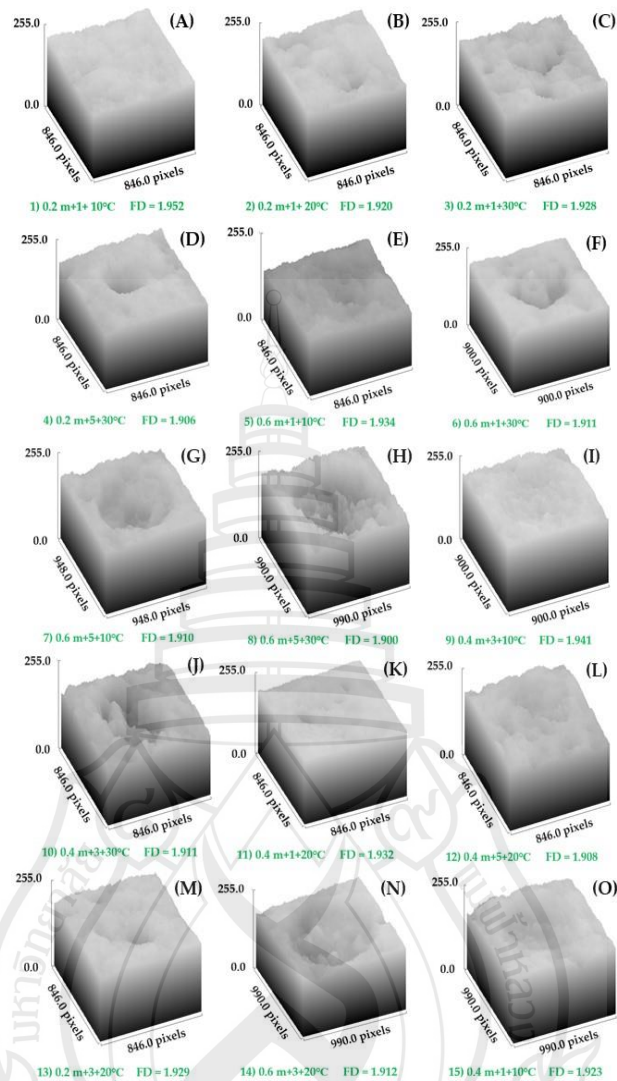
Treatment	Polynomial equation	R^2	R^2_{val}
Control (no drop)	$Y = -0.0014X^2 - 0.0089X + 1.9565$	0.80	0.87
0.3 m + 1 drop	$Y = -0.0007X^2 + 0.0031X + 1.9398$	0.85	0.83
0.3 m + 5 drops	$Y = -0.0002X^2 - 0.0009X + 1.9387$	0.95	0.88
0.6 m + 1 drop	$Y = -0.0015X^2 + 0.0060X + 1.9340$	0.85	0.76
0.6 m + 5 drops	$Y = -0.0020X^2 + 0.0076X + 1.9230$	0.99	0.92

4.2 Investigation and Evaluation of Impact Bruising in ‘Glom Sali’ Guava Using Image Processing and Response Surface Methodology

4.2.1 Fractal Image Analysis and Correlation between Bruise Susceptibility and Color

The intensity of fruit bruise damage was analyzed by converting cropped RGB images and calculating the FD values. Using the threshold method, the bruise damage region was cropped and then separated. Before applying a box counting method to determine FD values, the selected BA was transformed into a binary image (Figure 4.5). Fractal image analysis of guava impact bruising was varied in the drop test under RSM design (Table 3.7). Image analysis revealed visible damage to the peeled guava surface. A greater impact bruise with lower FD value exhibited as a significantly deeper surface plot image, which related to the low value of grayscale that came from the dark brown color of bruising, relating to increase in drop height, number of drops, and storage temperature. For example, the deepest surface plot image with the lowest FD value (1.900) was a drop height of 0.6 m for five drops ($E = 7273.60$ J) (Table 4.2) for storage condition at 30 °C (Figure 4.5H). Lower storage temperature at 10 °C with drop height

(0.6 m) and five drops ($E = 7273.60$ J) (Table 3.5) exhibited the deepest surface with FD value (1.910) (Figure 4.5G), while the shallowest surface with the highest FD value (1.952) was a drop height of 0.2 m for one drop and storage at 10 °C (Figure 4.5A). In previous studies, image analysis was used to obtain FD values for lightness and darkness of the surface to assess bruising or browning of banana [30]. Fractal modeling was used to assess the intensity of flesh browning and its color change to acquire improved knowledge of the enzymatic chemical changes and their location within the apple fruit [31]. Recently, it was found that higher impact bruising for both drop heights of 0.3 and 0.6 m repeated five times. Results showed a deeper surface plot with lower FD values of 1.937 and 1.930, respectively, after storage at 25 °C for 48 h [74]. Therefore, fractal image analysis in this study successfully performed impact bruising severity of guava under different drop test conditions utilizing RSM design. Recently, the advanced techniques, i.e., hyperspectral, computerized, and X-ray imaging had successfully achieved assessing bruise severity accurately; however, these techniques required higher invest in the machine and complex computational processes to interpret the data. Although, application of FD was only suitable for characterizing the external bruising such as vibrational bruising damage [26], this technique using simple equipment and data processing technique that may have a change to apply for various commodities.



Note The fifteen runs were 0.2 m + 1 drop + 10 °C (A), 0.2 m + 1 drop + 20 °C (B), 0.2 m + 1 drop + 30 °C (C), 0.2 m + 5 drops + 30 °C (D), 0.6 m + 1 drop + 10 °C (E), 0.6 m + 1 drop + 20 °C (F), 0.6 m + 5 drops + 10 °C (G), 0.6 m + 5 drops + 30 °C (H), 0.4 m + 3 drops + 10 °C (I), 0.4 m + 3 drops + 30 °C (J), 0.4 m + 1 drop + 20 °C (K), 0.4 m + 5 drops + 20 °C (L), 0.2 m + 3 drops + 20 °C (M), 0.6 m + 3 drops + 20 °C (N), 0.4 m + 1 drop + 10 °C (O).

Figure 4.5 Fifteen Pictures of Fractal Dimension (FD) of Surface Plots in Impact Bruising of Guava for Different Drop Heights, Number of Drops, and Storage Temperature Variables Under RSM Design from Fifteen Runs (Treatments) After Storage for 48 h

Guava fruit peel is thin and delicate and easily damaged by rough handling during harvest and postharvest [47]. In this study, Pearson's correlation was performed to establish the relationship between the six measured dependent variables BA, BV, BI, ΔE , BAI, and FD. The FD variable showed good agreement with the ΔE parameter ($r = -0.6055$) when compared with BA, BV, BI, and BAI, while color measurement and analysis of BI and ΔE parameters for impact guava bruising correlated poorly with bruise damages (BA and BV) as well as FD. Thus, image analysis (FD) was a good indicator to respond to browning incidence of impact guava bruising as the ΔE variable from 20 different impact conditions (Table 4.3). Heterogeneous changes of fruit and vegetable surfaces such as color intensity and enzymatic browning reaction had a strong correlation with FD value variations [128]. For bruise formation of fresh-cut apple, the higher potential of image analysis detected that ΔE value correlated to color changes [129]. Image analysis by the FD method offers great potential for application where color intensity has a non-homogenous color surface [29]. Increase in the FD value in the selected area indicated major complexity in color distribution during the enzymatic browning kinetic for banana [30]. For vibration bruising of guava, FD analysis exhibited high potential and accuracy under frequency, acceleration, and time duration of vibration testing [26]. For impact bruising of guava, FD analysis showed higher potential than color measurements to evaluate impact bruise damage under testing conditions such as drop height, number of drops, and storage temperature [74]. Thus, the FD variable was a good indicator for impact bruising of guava under varying conditions of drop height, number of drops, and storage temperature. Therefore, high efficacy of the FD technique was suggested to assess mechanical damages in guava, with applications on other sensitive fruit from impact and vibration forces.

Table 4.3 Bruise Assessment Parameters of Guava Fruit Stored at 25 °C Under 70% RH for Four Days

	BA	BV	BI	ΔE	BAI	FD
BA	1.0000					
BV	0.2363					
BI	-0.1871	0.1332				
ΔE	0.3132	0.2095	0.4332			
BAI	0.9975 *	0.2372	-0.1837	0.3116		
FD	-0.3854	-0.2366	-0.0448	-0.6055 *	-0.3992	1.0000

Note * Significant at $p < 0.05$: BA = bruise area; BV = bruise volume; BI = browning index; ΔE = total color difference, BAI = bruise area by image analysis; FD = fractal dimension.

In this study, ΔE was a better indicator for impact bruising damage of guava than BI and showed high correlation with the FD variable. Both browning scores and ΔE parameters revealed highest values in pomegranate corresponding to medium and high drop impact bruise damage [13]. At medium and maximum drop levels, a high ΔE value indicated impact bruising of pomegranate [19]. Variations in ΔE value correlated to changes in color of fresh-cut apple over time [129]. Interestingly, the BAI parameter from the image analysis technique showed positive correlation with BA ($r = 0.9975$). Bruise area by image analysis was suggested to apply for BA measurement as a conventional technique, with calculation as Equation (2) to determine the impact bruising area of guava.

4.2.2 Model Fitting and Statistical Analysis of CCF

RSM values utilizing CCF from 20 treatments (runs) performed the correlation of the response data between three independent variables and six dependent variables (BA, BV, BI, ΔE , BAI, and FD) (Table 3.7) by quadratic multiple regression equations as follows (Equations (15) to (20)).

$$Y_1 \text{ (BA)} = 219.8 - 2.3X_1 + 57.7X_2 + 117.1X_3 - 7.1X_1^2 - 107.6X_2^2 + 47.5X_3^2 - 2.0X_1X_2 + 4.3X_1X_3 + 29.0X_2X_3 \quad (15)$$

$$Y_2 \text{ (BV)} = 2559 + 1525X_1 + 386X_2 + 862X_3 + 4895X_1^2 - 3977X_2^2 - 2294X_3^2 + 4X_1X_2 + 25X_1X_3 + 372X_2X_3 \quad (16)$$

$$Y_3 \text{ (BI)} = 97.42 + 4.72X_1 - 3.50X_2 + 1.06X_3 + 4.72X_1^2 + 5.03X_2^2 - 0.35X_3^2 - 5.48X_1X_2 - 0.90X_1X_3 - 1.64X_2X_3 \quad (17)$$

$$Y_4 \text{ (\Delta E)} = 4.159 + 2.183X_1 + 0.491X_2 + 0.775X_3 + 1.140X_1^2 - 0.927X_2^2 + 0.567X_3^2 - 0.106X_1X_2 + 0.666X_1X_3 + 0.007X_2X_3 \quad (18)$$

$$Y_5 \text{ (BAI)} = 233.5 + 0.3X_1 + 56.6X_2 + 117.1X_3 - 7.3X_1^2 - 108.6X_2^2 + 48.5X_3^2 - 4.1X_1X_2 + 0.3X_1X_3 + 28.1X_2X_3 \quad (19)$$

$$Y_6 \text{ (FD)} = 1.9177 - 0.0117X_1 - 0.0097X_2 - 0.0068X_3 + 0.0049X_1^2 - 0.0011X_2^2 - 0.0006X_3^2 + 0.0029X_1X_2 + 0.0026X_1X_3 + 0.0004X_2X_3 \quad (20)$$

Table 4.4 shows the coefficient results of RSM regression equations generated from the ANOVA analysis of BA, BV, BI, ΔE , BAI, and FD models. The predicted six models (BA, BV, BI, ΔE , BAI, and FD) provided the determination coefficient (R^2_{adj}) values of 0.5304, 0.0868, 0.2227, 0.5751, 0.4960, and 0.8169, respectively. These findings indicated that the FD model provided higher response performance than the BA, BV, BI, ΔE , and BAI model predictions. The lack of fit values of the five models (BA, BI, ΔE , BAI, and FD) were not remarkable, except that the BV model showed low levels of reliability and repeatability, with significant lack of fit and R^2_{adj} (8.68%). The FD variable exhibited the highest R^2_{adj} value (81.69%), representing the highest precision model for impact bruising prediction among the other five variables. Also, the ΔE model with R^2_{adj} value (57.51%) exhibited greater liability for impact bruising prediction than BI with R^2_{adj} value (22.27%). In this study, FD exhibited the highest

dependent variable for impact damage of guava due to a significant correlation with ΔE (Table 4.3) as well as the highest levels of reliability and repeatability (Table 4.4).

Until recently, no RSM experiments investigating free fall or impact testing had been conducted for impact bruising susceptibility at various drop heights, number of drops and temperature conditions to simulate the effects on guava and other sensitive fruit. Only two RSM studies on vibration testing for fruit bruising used two independent variables to design vibration conditions of tomato [25] and three independent variables to design vibration conditions of guava [26]. Most studies on simulated impact bruise damage only focused on experimental designs by fixing one or two variables. With their fruit size and spherical shape similar to guava fruit, pomegranate and apple were tested under two variables of three drop heights and two storage temperature conditions with a fixed number of drops [11, 13, 24], while pear bruises were determined using two variables with three drop heights and two storage temperature conditions with a fixed number of drops [16-17]. Recently, impact bruising using two variables of three drop heights and member of drops (one and five times) on bruise assessment in guava fruit was examined [74]. Thus, no clear factor analysis has demonstrated impact fruit bruising under three independent variables.

In this study, an RSM design for simulated impact testing identified three major in-dependent variables in guava bruising with both individual and combined effects. The ANOVA result showed that all linear coefficients (drop height (X_1), number of drops (X_2), and temperature (X_3)) affected the FD model. The linear coefficients (X_1) and (X_3) also affected color changes (ΔE) and both BA and BAI models, respectively. The quadratic drop height (X_1^2) only had an effect on the BV model (Table 4.4). Most previous studies focused on two independent variables (drop height and storage temperature) with a fixed number of drop heights using CRD design in pomegranate [13, 19], 'Pink Lady' apple [11] and pear [16-17]. For example, in the study of pomegranate fruit, storage temperature factors affect-ed impact bruising with higher refrigerated storage temperature reducing bruise damage [24]. The drop height factor combined with impact materials affected bruise area measurement in apples [11], while the combination of drop height and storage temperature showed the highest increase in bruise area, bruise volume, and color measurements in pear fruit [16-17]. Recently, the number of drops (five drops) from different heights (0.3 and 0.6 m) affected impact

bruising of guava more than the same drop height (0.3 and 0.6 m) with a single drop [74]. Therefore, this is the first study to undertake impact test by RSM experiment for this bruising in guava and other fruit. However, this study of RSM design did not exhibit significant cross-product coefficients among the three independent variables from six dependent variables. Therefore, the linear model was suggested as optimal to predict impact bruising of guava compared with the quadratic model (Table 4.4). Previous studies investigated impact bruising volume of apple, with impact energy ranging 0 to 2.25 J. Results showed that linear regression fitted the impact energy for apple sizes of 180 and 240 g, with high coefficient of determination (R^2) values at 0.94 and 0.93, respectively [130]. There was also a high linear relationship ($R^2 = 0.94$) between BV and drop height for BV of apple [18]. Recently, a strong linear regression between impact bruise susceptibility and color parameters in pear at different drop heights and storage temperatures was found by Pathare [16].

The FD model was optimized by setting minimum conditions as drop height of 0.53 m for five drops under storage temperature of 30 °C (Equation (20)). To minimize the FD value, postharvest handling of guava must be gentle to avoid impact bruising, coupled with a cool storage temperature. Response surface analysis of CCF (Figure 4.6) showed that surfaces of the BA, BV, BI, and BAI models for drop height, number of drops, and storage temperature showed no interaction between X_1X_2 , X_2X_3 , and X_1X_3 (Table 4.3), while the ΔE model became steeper with increasing number of drops and storage temperature. The 3D graphs of FD model indicated that increasing number of drops and storage temperature in the slope of the curved surface and lower steepness (Figure 4.6F), giving linear model ($p < 0.05$) (Table 4.4) when compared with ΔE response surface with a non-significant impact in both number of drops and storage temperature ($p > 0.05$) (Figure 4.6C) (Table 4.4).

Table 4.4 ANOVA Results of each Drop Height (X_1) (m), Number of Drops (X_2) (times) and Temperature (X_3) ($^{\circ}\text{C}$) Factors as Response Surface Models for BA, BV, BI, ΔE , BAI and FD Values of Impact Bruise of ‘Glom Sali’ Guava Fruit

Coefficient terms	Y ₁ (BA)		Y ₂ (BV)		Y ₃ (BI)		Y ₄ (ΔE)		Y ₅ (BAI)		Y ₆ (FD)	
	Regression coefficient	p-value	Regression coefficient	p-value	Regression coefficient	p-value	Regression coefficient	p-value	Regression coefficient	p-value	Regression coefficient	p-value
Constant term	219.8	0.000	2559	0.057	97.42	0.000	4.195	0.000	233.5	0.000	1.9177	0.000
X ₁ -Drop heights	-2.3	0.934	1525	0.193	4.72	0.101	2.183	0.001**	0.3	0.993	-0.0117	0.000**
X ₂ -Number of drops	57.7	0.057	386	0.731	-3.50	0.209	0.491	0.288	56.6	0.071	-0.0097	0.000**
X ₃ -Temperature	117.1	0.001**	862	0.449	1.06	0.693	0.775	0.107	117.1	0.002**	-0.0068	0.003**
X ₁ × X ₂	-2.0	0.949	4	0.997	-5.84	0.090	-0.106	0.834	-4.1	0.899	0.0049	0.180
X ₁ × X ₃	4.3	0.888	25	0.984	-0.90	0.764	0.666	0.203	0.3	0.992	0.0011	0.217
X ₂ × X ₃	29.0	0.355	372	0.767	-1.64	0.588	0.007	0.990	28.1	0.391	0.0006	0.854
X ₁ ²	-7.1	0.892	4895	0.041*	4.72	0.365	1.140	0.202	-7.3	0.895	0.0029	0.179
X ₂ ²	-107.6	0.062	-3977	0.085	5.03	0.337	-0.927	0.293	-108.6	0.070	0.0026	0.755
X ₃ ²	47.5	0.375	-2294	0.297	-0.35	0.946	0.567	0.513	48.5	0.386	0.0004	0.865
Lack of fit		0.851		0.001**		0.933		0.403		0.877		0.536
R ²		75.28%		51.93%		59.09%		77.64%		73.47%		90.36%
R ² _{adj}		53.04%		8.68%		22.27%		57.51%		49.60%		81.69%

Note Significant level: ** significant at p-value < 0.01; * significant at p-value < 0.05.

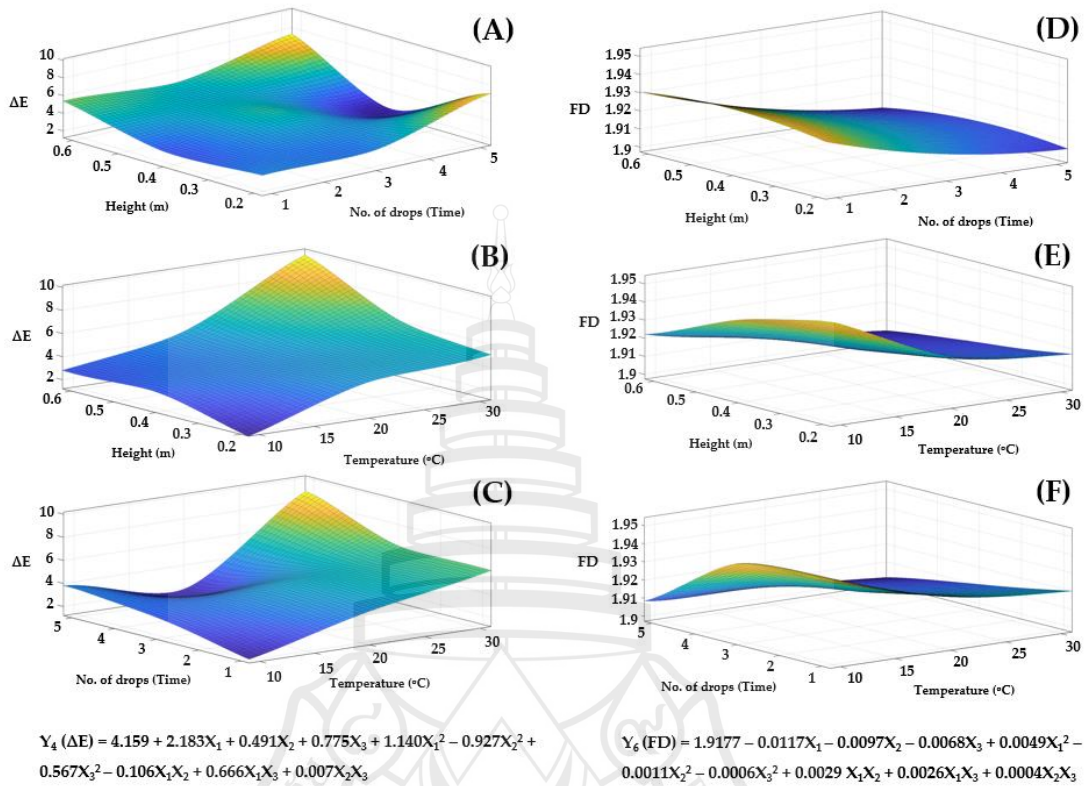


Figure 4.6 Response Surface Plot Graphs of ΔE and FD Variables were Created from Equations (10) and (12), Respectively. Response Surface Plots Demonstrating Effects of Drop Heights, Number of Drops, and Storage Temperature on ΔE and FD (D)–(F) in ‘Glom Sali’ Guava Impact Bruising at Various Drop Heights and Number of Drops (A) and (D), Different Drop Heights and Temperature (B) and (E), and Different Number of Drops and Storage Temperature (C) and (F)

4.2.3 Validation Testing of CCF

Validation of impact bruise damage of guava fruit focused on image analysis by FD (0.82) variables, with higher determination coefficient (R^2_{adj}) value of the predicted model compared to ΔE (0.57), BA (0.53), BAI (0.50), BI (0.2227), and BV (0.09) (Table 4.4). To evaluate and confirm the predicted FD model value for impact bruising (Equation (20), the model was verified using thirteen treatments in a range of 20 RSM

conditions with three independent variables. As shown in Figure 4.7, the predicted value of FD exhibited a high linear correlation with the observed value of FD ($R^2 = 0.83$) for impact bruise assessment of guava. Thus, FD by image analysis was proven to predict impact bruising of guava with high determination coefficient value (R^2_{adj}) (> 0.8) of the predicted model, with no significant lack of fit compared to the validated model. In a previous study, high values of both R^2 and R^2_{val} were recorded for a polynomial equation (plotting between drop height and storage period) at five drop times, with drop heights of 0.3 (0.95 and 0.88) and 0.6 m (0.99 and 0.92), respectively. The FD value exhibited a higher accuracy for impact bruise prediction with greater bruise susceptibility in guava fruit [74]. Most previous researchers conducted fruit quality evaluation using the FD method to assess internal browning and color change in the flesh [29-30], including fruit bruising [70]. Classification models based on the FD parameter attained a total accuracy rate of 100%, while the support vector machine model based on RGB values only realized 85.29% for bruising detection on red bayberries [70].

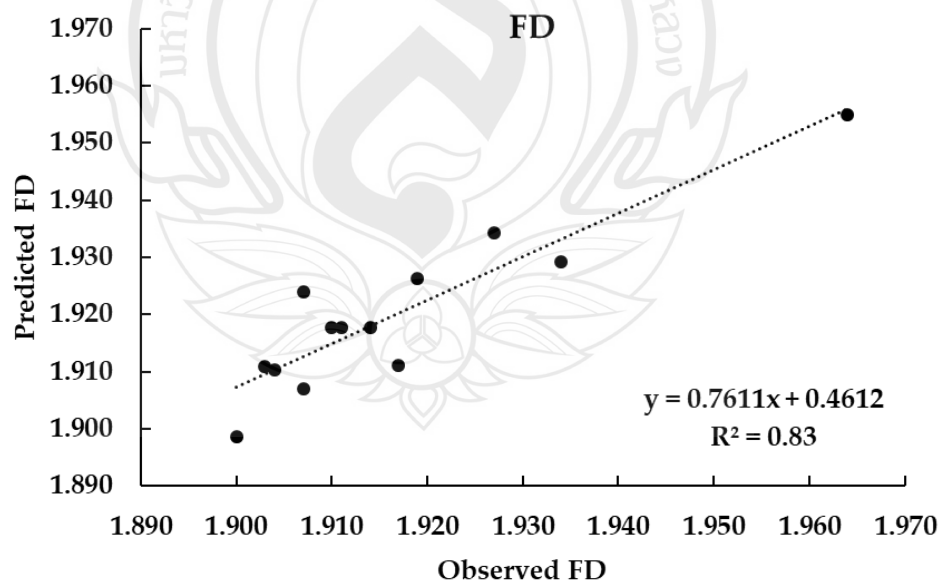


Figure 4.7 Verification of Predictive Model Equations for Fractal Dimension (FD) Values in ‘Glom Sali’ Bruised Guava by Impact Testing for 48 h

CHAPTER 5

CONCLUSIONS

In the first experiment, the completely randomized design (CRD) was assigned two drop heights (0.3 and 0.6 m) with one and five times compared with control as no drop for impact bruise of 'Gim Ju' guava. Results showed that the drop heights (0.3 and 0.6 m) with five times of drop gave greater BA, BV, BS, bruise score and a^* value and lower in L^* , b^* , C and FD value than those of the other three treatments. Image analysis of FD values showed higher potential than color measurement to investigate impact bruise in guava fruit. A higher of both R^2 (0.99) and R^2_{val} (0.92) at five times of drop revealed a higher potential for bruise determination by FD value with greater bruise susceptibility.

In the second experiment, the RSM with CCF design was examined impact bruising damage with different three independent variables of drop heights (0.2, 0.4 and 0.6 m), number of drops (1, 3 and 5 times) and storage temperature (10, 20 and 30 °C) in 'Glom Sali' guava. This is the first report in RSM study for impact bruising in guava or other fruits. Results showed that drop heights, the number of drops and storage temperature factors exhibited an individual effect on impact bruising of 'Glom Sali' guava. FD value also exhibited highest R^2_{adj} value (81.69%) with no remarkable lack of fit among the other five parameters. The optimized condition to minimize impact bruising was a drop height of 0.53 m for five drops coupled with a storage temperature of 30 °C. The predicted value of FD exhibited a high linear correlation with the observed value of FD ($R^2 = 0.83$) for impact bruise assessment of guava. Thus, impact bruising was increased by higher drop heights and increase number of drops on guava damage under higher storage temperature from both experiments.

In addition, the fractal image analysis technique was examined for impact bruising damage of guava under both CRD and RSM designs. The FD value gradually

decreased with the increase of storage time and bruise severity. In RSM design, the FD variable of peeled guava exhibited a good correlation with ΔE to indicate the browning incidence of impact bruising in guava as compared with BI. Thus, FD value results for two experiment was performed be more important parameter and higher potential than color measurements to evaluate impact bruise in guava fruit, particularly BA and BV.

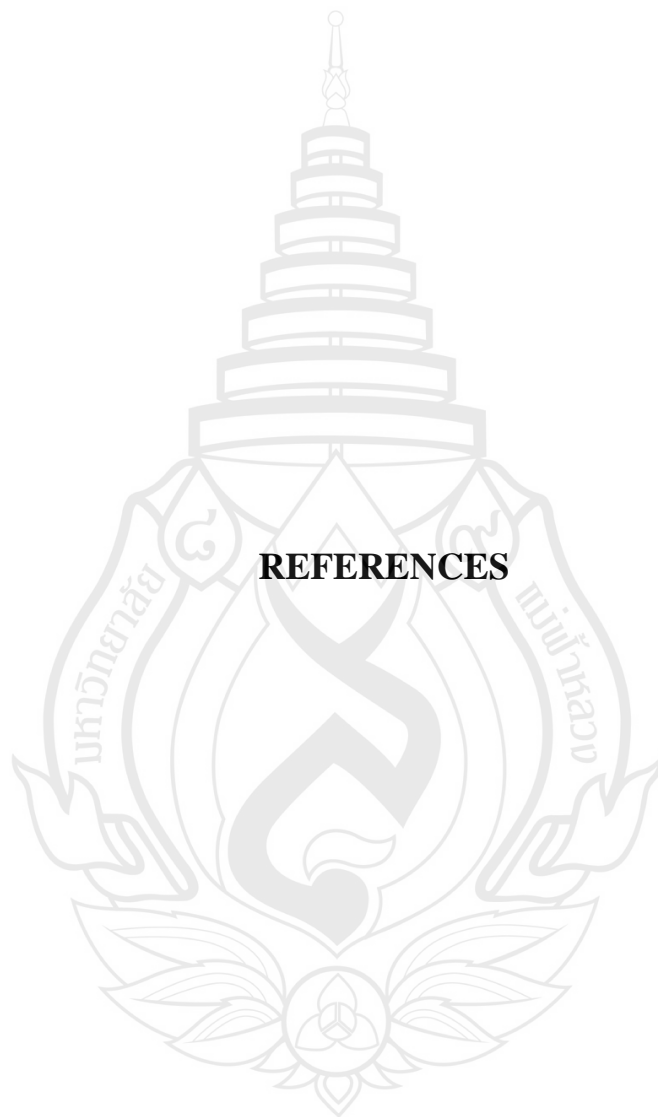


CHAPTER 6

SUGGESTIONS

The recommended condition to minimize impact bruising of guava was a drop height of 0.53 m for five drops coupled with a storage temperature of 30 °C. To reduce impact bruising incidence of guava throughout its supply chain, lower impact energy by a reduction of drop height (<0.5 m) and number of drops (< 5 drops) is suggested to control under cool chain management (<30°C). In this study, impact bruising of guava was examined under simulated drop test as a fruit-to-fruit impact. In the further study, the impact bruising of guava should be suggested to investigate and consider in other factors such as cushioning, packaging types, impact surface material to reduce impact bruising of guava under simulated impact testing or real transportation. The packaging system of guava distribution should be not only considered impact bruising but also should be investigated in vibration and compression forces under simulated transportation or real transportation.

Interestingly, this is the first report in RSM design to undertaken impact test of guava and other fruit. Our results are recommended using RSM design for spherical fruit or delicate fruit from impact bruising as browning incidence. In addition, our finding of fractal image analysis offers science-based tools to assist in improving fruit quality assessment from impact bruising of guava. Further research should be required to investigate and apply FD analysis for BA and BV assessments in spherically shaped fruit such as apple, pomegranate and peach using simulated impact testing.



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