# Mass transfer and ascorbic acids degradation during osmotic dehydration of ripe mango (*Mangifera indica L.*)


**Supervisors**

Ita Sulistyawati

Ruud Verkerk

Matthijs Dekker

**Lingqi Kong**

930520459050

Master Food Technology

Product design

Wageningen University and Research

Department Food Quality and Design


Wageningen

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# MASS TRANSFER AND ASCORBIC ACIDS DEGRADATION DURING OSMOTIC DEHYDRATION OF RIPE MANGO (MANGIFERA INDICA L.)

LINGQI KONG

WAGENINGEN UNIVERSITY AND RESEARCH

DEPARTMENT FOOD QUALITY AND DESIGN

## Abstract

Mango is one of the most consumed tropical fruit, with a high nutritional value that is loved by many people around the world. However as a perishable fruit, mango has a short shelf life due to high water content. Osmotic dehydration is applied to get a good texture. This project is aimed to study the mass transfer and ascorbic acid degradation during osmotic dehydration of mango at maturity stage 5.

To reach this goal, several analysis was conducted, including pH measurement, dry matter analysis, water activity and vitamin C concentration measurement. Mango cubes was treated under osmotic dehydration condition for 0-29h. due to the vulnerable texture of stage 5 mango, Pectin methyl esterase (PME) and calcium lactate was added to improving product texture during osmotic dehydration.

The water started leaching out of cubes right after the osmotic dehydration took place, while the sugar moved into cubes a little later. With longer time of OD, more water came out of cubes while more sugar leached in. This mass transfer slowed down with time and after reaches certain point, it stopped. However, this exact time point for maximum OD treatment is not yet confirmed, it should not exceed 29 hours. It was found that half of the weight is lost due to osmotic dehydration, the longer time of osmotic dehydration, more water transferred from the mango cubes to OD solution, in the meantime, sugar entered to the mango cubes. The mass transfer of water leach out and sugar gain can be modelled by Peleg's equation, the parameter k1 and k2 were obtained from the non-linear regression analysis from the experiment data, and used for modelling for OD process from 0 to 29h.

The most valuable vitamin in mango: vitamin C, the content was also influenced by the time of OD, in 100g of dry mango, 130mg total ascorbic acids(TAA) is present in fresh mango, while only 25mg is ascorbic acid(Vihakas), the concentration difference can be explained by ascorbic acid oxidation in to dehydroascorbic acids(Bchir et al.) acid during preparation, or ascorbic acid leached out of mango cubes and entered to the OD solution. On the other hand, OD treatment caused rapid degradation and in the first half an hour, TAA content dropped to 60mg in 100g dry mango, this trend of decreasing kept until 29 hours of OD to 3mg/100g dry mango. As for AA, the concentration decreased during OD processing. Water activity for fresh mango is 0.98 and improved to 0.9 after 29h of OD. It is to recommended that earlier maturity stage mango should be used due to stronger tissue structure. And for fruit like mango with high sugar content with sensitive characteristics , protocols should be adjusted for future investigation.

KEYWORDS:

Kent mango; Osmotic dehydration; ascorbic acids degradation; total ascorbic acids; pectin methyl esterase; calcium lactate; mass transfer; Peleg's equation.

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# List of abbreviations

| | | |
|---|---|---|
| OD | = | Osmotic dehydration |
| PME | = | pectin methyl esterase |
| DM | = | Dry matter |
| HPLC | = | High performance liquid chromatography |
| Aw | = | Water activity |
| TSS | = | Total soluble solid |
| TTA | = | Total Titratable acid |
| MPA | = | Metaphosphoric Acid |
| TCEP | = | Tris-2-carboxyethyl phosphine |
| TBHQ | = | Tert-Butylhydroquinone |
| AA | = | Ascorbic acid |
| TAA | = | Total Ascorbic acid |
| DHA | = | L-Dehydroascorbic Acid |
| FQD | = | Department Food Quality and Design |

# 1. Introduction

Mango (*Mangifera indica L.*) is the second most important fruit crop of the tropics with an annual production of 28.22 million tonnes in the world after banana. It is popularly known as 'the king of fruits' and is the choicest fruit due to its delicious taste, pleasant aroma and high nutritional value. The fruit is a large, fleshy drupe with edible mesocarp, and the size and shape vary considerably depending upon the cultivar. There is a great diversity in mango cultivars distributed throughout the world(Singh, 2012).

Mango fruit is considered a good source of dietary antioxidants, and phenolic compounds (Palafox-Carlos, Yahia, & González-Aguilar, 2012). Mango is cultivated over the world, mango production is located in most of the developing countries where an appropriate post-harvest handling infrastructure is still in infancy, Asian continent contributes about 77% of the total world mango production of the world. It is a highly perishable fruit because of high water content(Yadav & Singh, 2014), it keeps well for 9–10 days if harvested at mature green stage. However, the post-harvest behaviour of mango fruit is strongly influenced by the cultivar, harvest maturity, the post-harvest treatments and storage conditions. The chilling sensitive nature of mango fruit limits its long duration storage and transportation at low temperature below 13°C(Singh, 2012). Mango is a climacteric fruit. While ripening, it becomes much more susceptible to pathogen infections, due to the decrease in peel(Mattiuz et al., 2015).

Sensory quality is particularly difficult to control for mangos, colour changes (due to both enzymic and non-enzymic browning) and/or texture defects can occur. Careful control of drying chamber design is critical to achieving optimal product quality(Donald G. Mercer, 2008). Conventional drying methods are normally time and energy consuming and hence most often uneconomical. Osmotic dehydration has been suggested by some previous research as a pre-treatment for reducing the high water content of fresh fruits and vegetables before further processing (Alakali, 2006).

## 1.1.Kent mango

Among mango cultivars, 'Kent' is dominating the production as it is widely accepted by consumers due to its excellent sensory attributes such as flavour, aroma, and attractive colour activity of fresh-cut 'Kent' mango(Robles-Sánchez, Rojas-Graü, Odriozola-Serrano, González-Aguilar, & Martín-Belloso, 2009).

### Maturity stage

The maturity of mango is related to the onset of ripening. Mangos should be harvested at a minimum internal stage of 2, Mangos harvested immature (stage 1) will never develop good flavour(Dea, Brecht, Nunes, & Baldwin, 2010). A mature mango will ripen normally with increasing soluble solids content (degrees Brix) and decreasing firmness (lbs. force) to become ready to eat. The maturity of mango can be indicated by a combination of several factors, internal colour, firmness, degrees brix and fruit shape, for most of cultivar mango, 5 stages are expected according to Mango Maturity and Ripeness Guide(team, 2010). In table 1, 5 stages of maturity were quantitatively compared for 'Kent' mango, firmness and brix are chosen to be the indicator for maturity stage.

**Table 1 Maturity stage index for Kent mango.**

| | Stage 1 | Stage 2 | Stage 3 | Stage 4 | Stage 5 |
|---|---|---|---|---|---|
| **Firmness (lbs. force)** | 19-22 | 14-18 | 11-13 | 5-8 | 2-4 |
| **Brix** | 8-10 | 9-11 | 12-13 | 12-14 | 14-15 |

# 2. Literature review

## 2.1.Osmotic dehydration

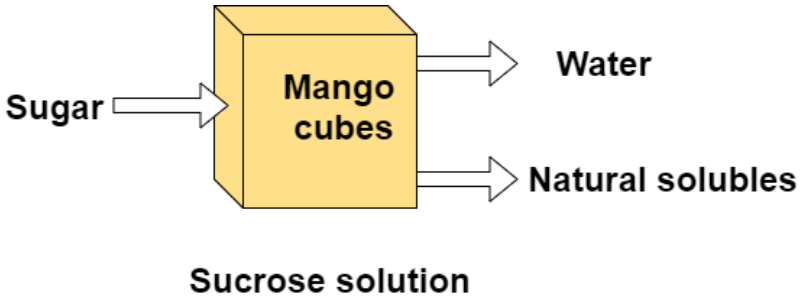


**Figure 1 Schematic demonstration of osmotic dehydration process.**

Osmotic dehydration (OD) is a widely used drying technique for the concentration of fruit and vegetables, realized by placing the solid food, whole or in pieces, in sugars or salts aqueous solutions of high osmotic pressure(Torreggiani, 1993). A result of osmotic dehydration is mass transport. As shown in Figure 1 Mass transfer occurs in three different types, water can flow from the product to the sucrose solution, some natural soluble can leach out of the mango into the solution (vitamins, phenols, minerals, etc.), whereas sugar transfer from the solution into the mango (Nicolaos M. Panagiotou, Vaios T. Karathanos, & Maroulis, 1998). The flows in and out are both due to the water and solute activity gradients across the cell's membrane(Torreggiani, 1993).

Previous studies have shown that mass transport properties of mango tissue in osmotic treatments with sucrose solution were greatly affected by sucrose concentration and by sample vacuum impregnation at the beginning of the process; these give rise to tissue impregnation with sucrose solution(Giraldo, 2003). However different time of osmotic dehydration treatment applied to mango cubes is suggested to be a variable.

Compare to other drying method, osmotic dehydration reduced water to a still relatively high moisture content level, as well as having microbiological stability due to reduced water activity(Nicolaos M. Panagiotou et al., 1998). There are some other major advantages of osmotic dehydration process in the food industry: improvement in terms of colour, flavour, texture, product stability and retention of nutrients during storage, Simple equipment is required for the process(Yadav & Singh, 2014), chemical treatment not required(Akbarian, , & 2014), energy efficiency, cost reduction of packaging and distribution, and greatly enhanced storage life of product(Akbarian et al., 2014).

Some study showed that due to the presence of Pectin methyl esterase (PME) and Calcium ions (Ca++) in the osmotic solutions, weight reduction upon dehydration were slightly decreased, which was correlated to a small positive effect on the net uptake of sugars and depression of the initial freezing point. The added PME and Calcium however positively influenced the volume and shape of the thawed samples, which could be related to slightly higher relative hardness values and, for the Elsanta strawberry fruits(Van Buggenhout, Grauwet, Van Loey, & Hendrickx, 2007). To enhance the osmotic dehydration, PME and Calcium was added to the osmotic solution.

## 2.2.Kinetics

Osmotic dehydration is used to remove water in fruit tissues. The mass transfer kinetics for water loss and solids gain during OD of fruits and vegetable have been studied. The research shows the mass transfer kinetics can be modelling according to Peleg's equation(Ganjloo, Rahman, Bakar, Osman, & Bimakr, 2011; Khin, Zhou, & Perera, 2006; Khoyi & Hesari, 2007; Mercali, 2010; B. Singh, Kumar, & Gupta, 2007). Peleg's model is a two-parameter sorption equation that can be employed to predict successfully, or at least estimate, moisture and solute contents for long exposure times of dehydration, using experimental data that are obtained in a relatively short time(Mercali, 2010).

## 2.3.Pectin methyl esterase

Within the enzyme systems present in the products obtained from fruit, is pectin methyl esterase (PME) which belongs to the pectin esterase group, pectic acid interaction with calcium ions forming precipitates(DÍAz-Cruz et al., 2016). This reaction creates negatively charged carboxyl groups, which can impact cell wall integrity and texture(Matella, 2012).

PME may cause effects before, during, or after processing of fruit and/or fruit products. PME enhances the texture of fruit and vegetable products, and promotes water removal from the tissues on drying, activation of PME is accompanied by the hydrolysis of the methyl ester groups from pectin, leading to the formation of a calcium pectate gel(Ben - Shalom, 1985).

## 2.4.Calcium addition

Structural changes was noticed in the product texture during osmotic dehydration, caused loss of cell turgidity, deformation and/or cell wall rupture, and tissue shrinkage that is not promising for cellular structure of fruits(Phisut, 2013). The addition of salts to the osmotic solution, calcium infiltration as a pre-treatment to the osmotic dehydration process or calcium salt dips in combination with mild heat treatments have resulted in improvements in product texture and have shown a protective effect on tissue structure(Pereira, Carmello-Guerreiro, Bolini, Cunha, & Hubinger, 2007). The addition of calcium in osmotic solutions has been widely used in plant foods as fortifier and to enhance firmness(Mauro et al., 2016). The action of calcium on the cellular structure has been explained by its effect on the pectin matrix present in the cell wall of plant tissues. Treatment with calcium salts has also shown an effect on the mass transfer kinetics of osmotically dehydrated products. An increase in water and sucrose transport was verified when calcium salts were used as a pre-treatment in the osmotic process of tomato quarters. Penetration of calcium ions and formation of bridges between pectin molecules stiffen the tissue, increasing the resistance to deformation. As a result of this, an open structure is formed favoring the mass transport (Pereira et al., 2007).

Calcium chloride is a calcium salt commonly used in the preservation of fruits and vegetables but in some products like apples and cantaloupes, it may impart bitterness or changes in flavor. The use of calcium lactate has been proposed as an alternative source of calcium (Pereira et al., 2007).

## 2.5. Ascorbic acids

Vitamin C is a water-soluble vitamin, thus human body cannot store it. Ascorbic acid (2-(1,2-dihydroxyethyl)-4,5-dihydroxyfuran-3-one) (Vihakas) and dehydroascorbic acid (Bchir et al.) are reduced and oxidized forms of vitamin C, that can be found in various fruits and vegetables(Chebrolu, Jayaprakasha, Yoo, Jifon, & Patil, 2012). Under oxidative stress, ascorbic acid (Vihakas) is converted to dehydroascorbic acid (Bchir et al.) by losing two protons (J.C. Deutsch 1996) as shown in Figure 2. However, as a natural soluble, AA could also leach out from the cubes to the OD solution with water.

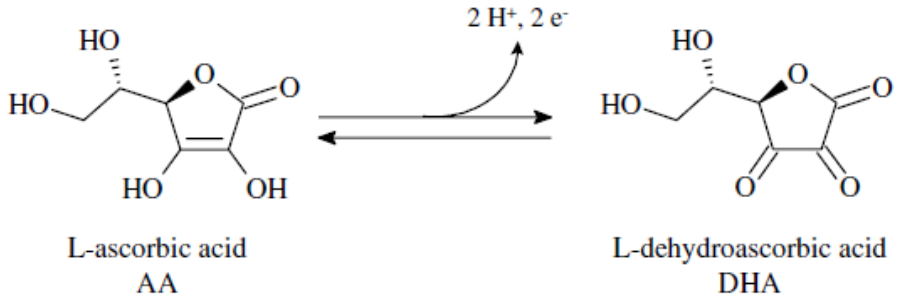


**Figure 2 Oxidation of L-ascorbic acid**

# 3. Research objective

Mass transfer and Ascorbic acids degradation during osmotic dehydration with PME present of ripe mango cubes at maturity stages 5.

- To investigate the mass transfer in mango cubes during osmotic dehydration.
- To model the kinetics and mass transfer of water and sugar during osmotic dehydration.

# 4. Material and method

This preparation of materials and OD treatment was done in FQD food safe lab, later on, moved to other lab for further treatment and HPLC analysis. The OD treatment was done in duplicate to avoid differences between batch, all the measurements were done in duplicate.

## 4.1.Material

The Kent mangoes used are provided by Nature's Pride harvested from Peru, Sucrose solution is prepared with Van Gilse fine crystal sugar purchased from Albert Heijn, PME added into sucrose solution is provided by Novozymes A/S, Copenhagen, Denmark and stored at 5°C before use. Calcium L-lactate pentahydrate added in sucrose solution is from Fluka analytical (BCBN1455V).

## 4.2.Method

In Figure 3, the experiment procedure is shown, kent mangoes was first received from supplier, followed by a maturity stage analysis, those mangoes in maturity stage 5 was used for OD treatment, the mango cubes was prepared and immersed in OD solution for 7 different time of treatment and including another fresh sample. The OD treated mango cubes and OD solution after OD process was collected and used for further experiment.

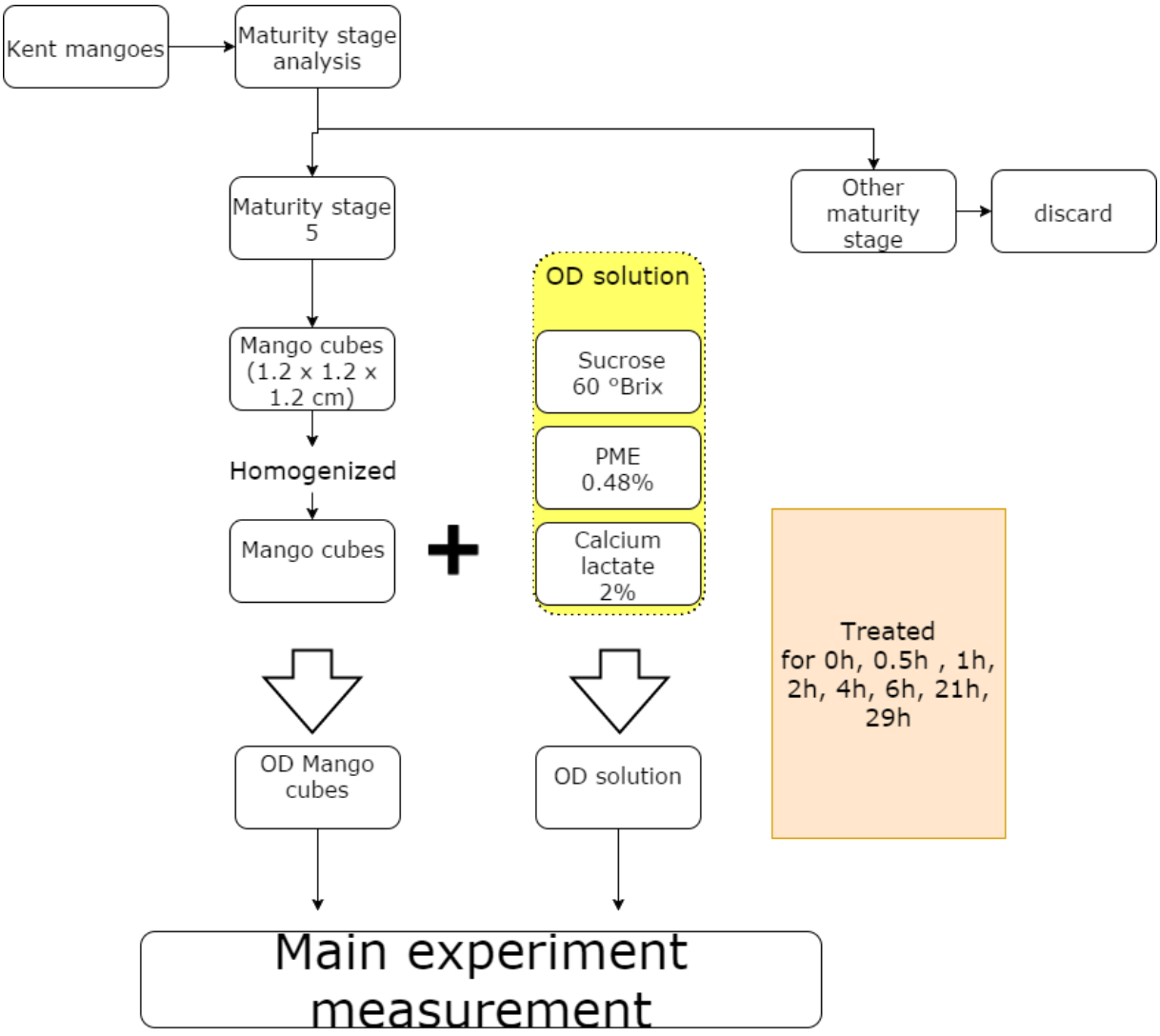


**Figure 3 Processes flow chart for the experiment design.**

### 4.2.1. Preparation method

**Preparing solution**

The sucrose solution was prepared one day in advance, the composition is shown in table 2 , the sucrose solution was made by adding sugar (58.8%), Calcium (0.2%) into demi-water(40%), PME was added later with mango cubes at the same time, because PME is heat sensitive.

**Table 2 composition of OD solution and concentrations.**

| Component | Concentration |
|---|---|
| **Sugar** | 58.8% |
| **Water** | 40% |
| **Calcium lactate** | 0.2% |
| **PME** | 0.048% |

### Preparing mangoes

The maturity stage of mangoes were determined by the firmness according to national mango board(team, 2010) by a penetrometer with an 8mm tip with the skin removed, then mango has firmness between 2-4lbs is kept for further used, other mangoes were discharged.

The weight of qualify mangoes were recorded and cut to 2 slices and cut to cubes of 1.2 x 1.2 x 1.2 cm. The mango cubes are mixed together to be homogenized, about 150g of mango cubes is prepared for one time point. This step was prepared in duplicate, the first batch (batch A) prepared at day one, the second batch (batch B) prepared one day after.

### OD treatment

The OD condition is shown in table 3 the mango cubes were immersed in sucrose solution at 1:4 ratios, and PME (0.048%) was added at this step to the sucrose solution, the mixture was placed in a large beaker in hot water bath at 50 °C for different time period. Including a fresh sample (0h) and 7 treated sample 0.5h, 1h, 2h, 4h, 6h, 21h, 29h samples. This time point is determined by conducting a several trial experiment.

**Table 3 Fixed feature and variable for OD treatment including experiment design reference.**

| | Description | Value | Reference |
|---|---|---|---|
| **Fixed** | **Sample size** | **1.2 x 1.2 x 1.2 cm** | **(Grunsven, 2015)** |
| | **Solute solution** | **60 °Brix** | **(K. S. Silva, Fernandes, & Mauro, 2013)** |
| | **Ratio solution to fruit (w/w)** | **4:1** | **(Super, 2014)** |
| | **Temperature** | **50 °C** | **(Super, 2014)** |
| | **Calcium concentration** | **2%** | **(Torres, Talens, Escriche, & Chiralt, 2006)** |
| **Variable** | **PME added** | **0, 0.48%** | **(Van Buggenhout et al., 2007)** |
| | **OD treatment time** | **0,0.5,1,2,4,6,21,29 hours** | **(Super, 2014)** |

### Storage

After set OD time were reached, the mango cubes were removed from solution and dried by paper towel, the weight recorded to calculated water lose during OD, the weight of sucrose solution is also recorded. The mango cubes were separated to two parts, about 70 grams will be frozen immediately by adding liquid nitrogen, the other 70 grams will be storage in a fridge and use for further analysis.

### 4.2.2. Analysis methods

The methods used in this experiment is listed in table 4.

**Table 4 Analysis methods used for the experiment for mango cubes and OD solution after OD process.**

| | Mango cubes | Remarks | OD solution | Remarks |
|---|---|---|---|---|
| pH | ✓ | / | ✓ | / |
| TSS | ✓ | / | x | / |
| TTA | ✓ | only for fresh | x | / |
| DM | ✓ | / | x | / |
| Aw | ✓ | only for fresh and max time | x | / |
| HPLC | ✓ | / | ✓ | / |
| Picture | ✓ | / | x | / |

**pH analysis**

pH (potential of hydrogen) is a numeric scale used to specify the acidity of mango cubes pulp and OD solution. Contreras mentioned that weight loss in mango was enhanced when the pH of the concentrated sucrose solutions was decreased by the addition of acetic, lactic and citric acids (Contreras & Smyrl, 1981). Citric acids is leaching out from mango cubes to OD solution during OD process. It is obvious that weight loss is related to pH in some level.

pH was measured by pHenomenal 1000L and pH-electrode SenTix Sp at room temperature, mango cubes were blended by a blender. OD solution was direct measured. pH measurement is repeat for three times.

**Total Soluble Solids (TSS) and Total titratable acidity(TTA)**

Total soluble solids is an indicator for maturity stage and quality of consuming. TSS referring to sugar gain during the reaction, TSS is used for know how many grams of sucrose is gained compare to the total weight. Frozen mango cubes are first blended by a blender, the juice will be pressed out by using a cheese cloth TSS was measured for mango cubes using a HANNA refractometer.

Total titratable acidity is also called titratable acidity which decreases with ripeness, citric acid and malic acid are the main organic acids in mango which decreases as titratable acidity. The TTA was used for evaluating for the fresh fruit mixture of mango cubes, this procedure was done with 10mL of mango juice. The juice pressed out of frozen mango pulp of untreated mango cubes is titrated with 0.1N NaOH until pH reaches 8.1.With this procedure, percentage of acid (considering citric acid factor of 0.0064), the percentage of acid and sugar acid ratio can be calculated as below:

$$Acid\ percentage = \frac{(mL\ of\ NaOH)(0.0064)(100)}{10mL\ juice}$$

$$Sugar\ Acid\ Rati0 = \frac{^{\circ}Brix}{Percentage\ Acid}$$

**Dry matter analysis**

Dry matter content in mango has been shown to be a reliable indicator for the quality of mangoes during ripening(Saranwong, Sornsrivichai, & Kawano, 2004), by measuring dry matter to have a better understanding about what is happening during OD.

A small piece of mango will be first weighted out and recorded, and left in an incubator at 105c for overnight and next morning will be moved to exicator to cool down without water attached to the surface, and the final weight will be recorded and used for further calculation.

$$waterLoss = \frac{(M_t)(x_{w,t}) - (M_o)(x_{w,o})}{M_o}$$

$$SolubleSolidGain = \frac{(M_t)\left(x_{s,t}\right) - (M_o)\left(x_{s,o}\right)}{M_o}$$

$$WeightReduction = \frac{M_t - M_o}{M_o}$$

$$ODPerformance = \frac{WaterLoss}{SolubleSolidGain}$$

$M_0$ the initial weight of sample (g)

$M_t$ the weight of sample at time t (g)

$x_{w,0}$ the mass fraction of water initial content

$x_{w,t}$ the mass fraction of water content at time t

$x_{s,0}$ the mass fraction of solids initial content

$x_{s,t}$ the mass fraction of solids content at each sampling times

**Water activity**

One of the purpose for osmotic dehydration is to reduce water activity to increasing storage stability(M. A. d. C. Silva, Silva, Mariani, & Darche, 2012), aw is well related with most degradation reactions of a chemical, enzymatic nature, as well as a determinant for the growth of micro-organisms. Comparing with moisture content, aw is easier to measure (Maltini, Torreggiani, Venir, & Bertolo, 2003).

Water activity of mangoes is measured using Novasine Labmaster-aw for time point fresh and max time 29h. Samples are prepared by cutting the cubes into smaller pieces. The small pieces was placed in a container matched with machine and preheated in the waiting chamber, The measurement is set with 2 minutes stability time for aw measurement and 3 minutes for temperature.

**Vitamin C analysis**

HPLC is used for vitamin c analysis; the preparation method is according to Protocol 35A2. Ascorbic acid peak normally shows on the Figure around at 2.7min.

Weigh approximately 0.25g of fresh frozen mango pulp in a 15 ml Greiner tube. □ Mix this sample with 3.5ml of the MPA,THBQ solution(3% MPA, 1 mM THBQ in Milli-Q water). Homogenize the mixture using a Ultra Turrax with small Turrax tube at the highest speed for 1 minute. Centrifuge the mixture for 5 min 3000 rpm at 4°C.Collect supernatant in pre-weighted 15 ml tubes. Repeat this procedure twice. Combine the supernatants in the pre-weight 15 ml tube. Weigh tube with content after 3 times treatment with MPA THBQ solution. Deposit 2 x 5.0 ml of the supernatant in 5.0 mL reaction tubes. centrifuge at 10.500 rpm for 10 minutes at 4°C. Filter the juices using 0.2 µm filters. Separate juices to two HPLC vials, each around 2 mL, one is for ascorbic acid measurement, another one is for total ascorbic acid, including 1.485mL of solution and 0.015mL of TCEP well mixed, vial with TCEP need at least 20 mins of incubation. In addition, a standard calibration line should be constructed using ascorbic acid stock solution range from 200 µg/ml to 1.56 µg/ml. the running of HPLC should according to method: 20120802 vit C Polaris 2010 nov.

**Visual appearance**

A picture for a whole set of mango cubes are taken in the food safe lab picture station by Canon camera at fixed setup.

# 5. Results and discussion

## 5.1.pH

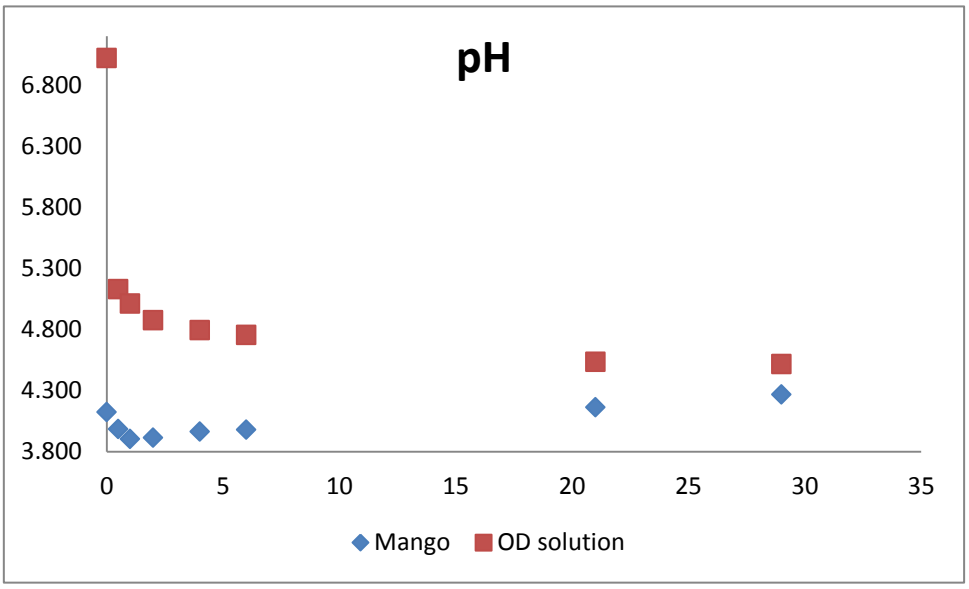


**Figure 4 pH change of mango and OD solution as function of time of OD(h) of Kent mango cubes for average of two batches.**

The Figure 4 the average pH of mango cubes of two batches, the Figure 5 described the average for OD solution after OD. The pH of fresh mango cubes was around 4.1, while the sucrose solution made is around 7. After OD treatment, the pH of mango cubes dropped to around 4.0, and with longer OD treatment time, pH decreased to around 3.9 after one hour of OD. pH started to increase slightly after one hour of treatment, however, the changes of pH was too small which cannot be considered as a significant indicator for OD time.

Unlike mango cubes, the OD solution after OD treatment showed quite a trend, the sucrose solution has pH around 7, and after 29 hours of OD, pH decreased to around 4.5, the biggest change happened during the first half hour of OD treatment from pH 7 to 5. In previous study, the same trend for pH of OD solution was observed, with OD time increasing, pH decreased because of electrolytes (EC) leaching out of mango cubes entered OD solution (Bm, 2013).

## 5.2.TSS & TTA

Total soluble solid content of fresh mango cubes was around 16, with the time of OD increased, the TSS gradually increased to around 54 after 21 hours of OD treatment, after 21h, the growth stopped as shown in Figure 5.

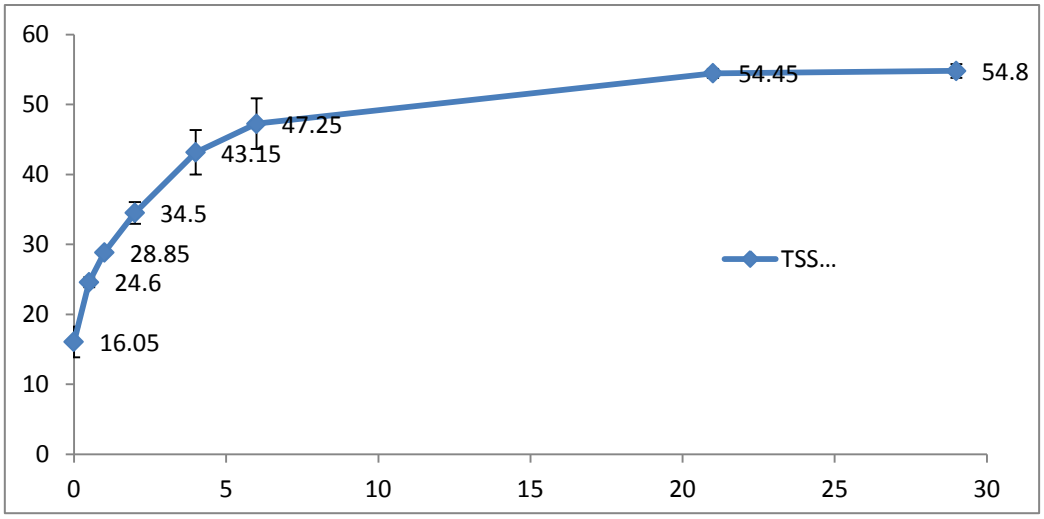


**Figure 5 Total soluble solid as function of time of OD(h) of Kent mango cubes for average of two batches with standard deviation.**

In Figure 6, the sugar acid ratio is presented, even there is a huge difference between two batches, high sugar: low acid could be really different from low sugar: high acid, however it may differ from difference individual.

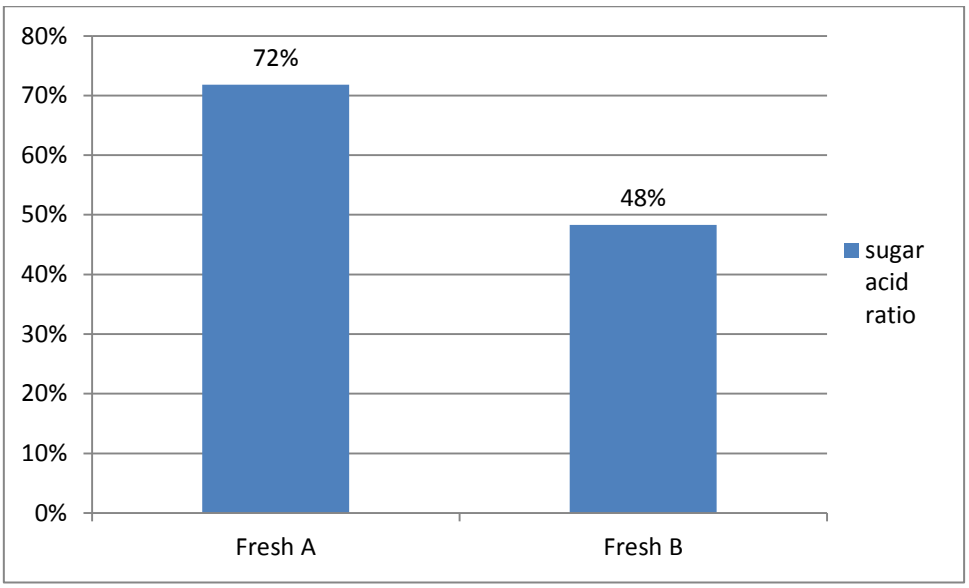


**Figure 6 Sugar acid ratio of two batches in percentage.**

## 5.3.Dry matter

Dry matter is evaluated from four aspects, water loss, soluble solid gain, weight reduction and OD performance index.

### 5.3.1. Water loss

Figure 7 and Figure 8 illustrate water loss as a function of OD time for batch A and batch B. A pattern can be described from the blue dots, at the first half an hour of OD, there is a significant water loss can be noticed, about 35% of water was reduced. According to Contreras and Smyrl's article, rapid removal of water and uptake of solids in the early stages can be observed in OD of apples(Contreras & Smyrl, 1981), due to the large osmotic driving force between the mango cubes and the hypertonic medium in sucrose solution, a rapid loss of water in the beginning is proved by many published article (Azoubel & Elizabeth Xidieh Murr, 2004). With longer time of OD, more water lost into OD solution, at around 6 hours, water loss content reached to a level around 60~65% where the change of water loss rate gradually became to 0~2%. But most likely the massive water loss is stopped at this moment. This trend was also described by many researches (Harris N. Lazarides, 1997; Kowalska; & Lenart).

In order to have a better interpretation for water transfer during osmotic dehydration, a mathematical model is applied. According to Ganjloo et al.(Ganjloo et al., 2011) about kinetics modelling of mass transfer of OD, Peleg's equation is adapted as:

$$X = X_0 \pm \frac{t}{K_1 + K_2 t}$$

X=water loss content

$X_0$= initial water loss content

t= osmotic dehydration time

+= weight gain process (for water loss)

-= weight loss process (for sugar gain)

However Azoubel used redefined equation for soluble solids and moisture content(Azoubel & Elizabeth Xidieh Murr, 2004), by using water content instead of water loss.

$$MC(t) = MC_0 \pm \frac{t}{K_1 + K_2 t}$$

MC=water content at time t

MC0=initial water content

$k_1$ and $k_2$ are model parameters obtained by linear regression from data point for experiment. the values of 1/k1 and 1/k2 are the initial rate of mass transfer and the mass transfer at equilibrium(Ganjloo et al., 2011). 1/k1 is depend on the concentration of OD solution, the high brix of OD solution results in a higher 1/k1 value(Azoubel & Elizabeth Xidieh Murr, 2004). However, the brix of OD solution is not studied in this research and Park mentioned that there is no relation between peleg's equation parameters with the increase of sucrose concentration at constant temperature in the OD of pears(Kil Jin Park , 2002).

The Peleg equation obtained for batch A and batch B fitting into same equation as shown below, the data points scattered near the equation line, Peleg equation presented a good fitting to the experiment data.

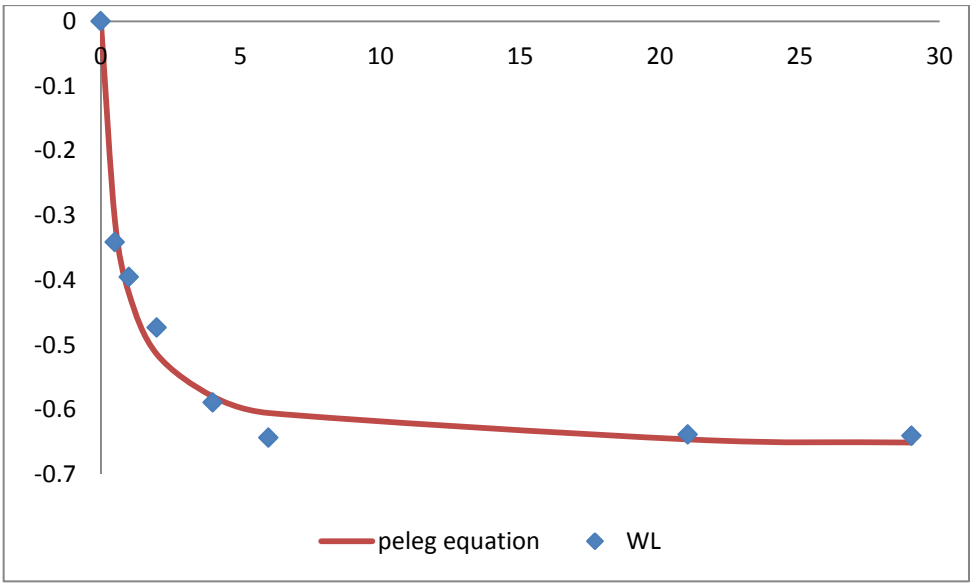


**Figure 7 Water loss (g of water/g of fresh mango cubes) as function of time of OD(h) of Kent mango cubes applied to peleg equation for batch A.**

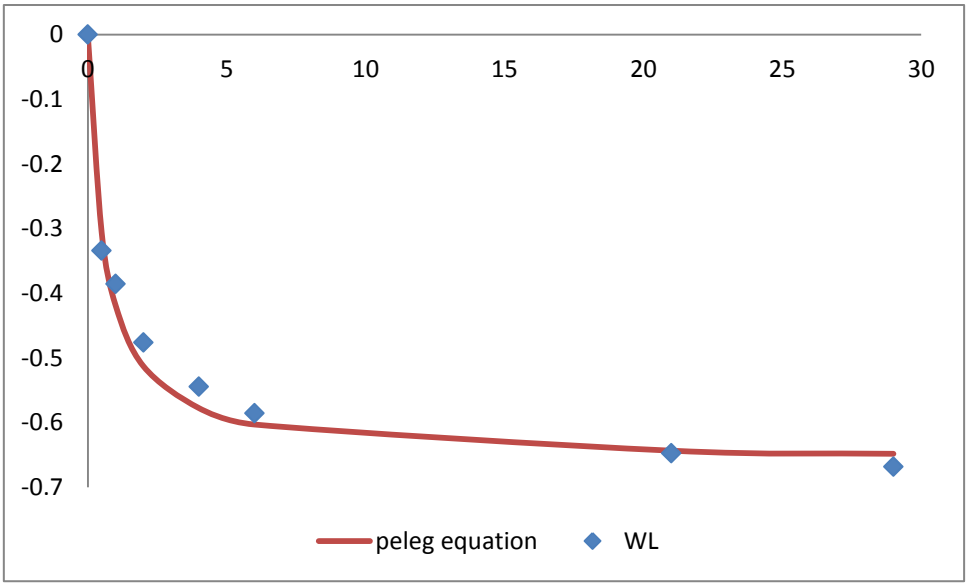


**Figure 8 Water loss (g of water/g of fresh mango cubes) as function of time of OD(h) of Kent mango cubes applied to peleg equation for batch B.**

### 5.3.2. Soluble solid gain

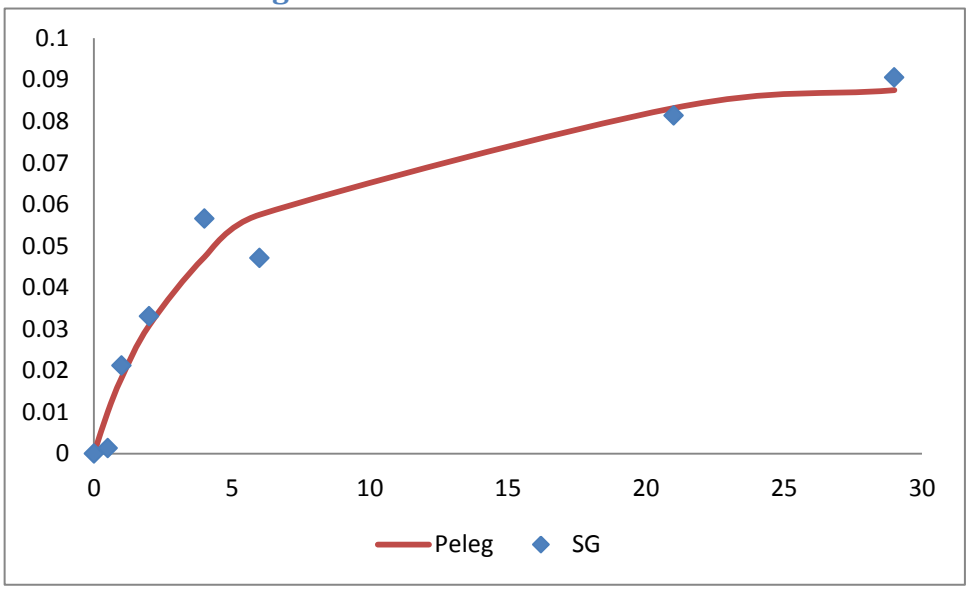


**Figure 9 Soluble solid gain (g of sugar/g of fresh mango cubes) as function of time of OD(h) of Kent mango cubes applied to peleg equation for batch A.**

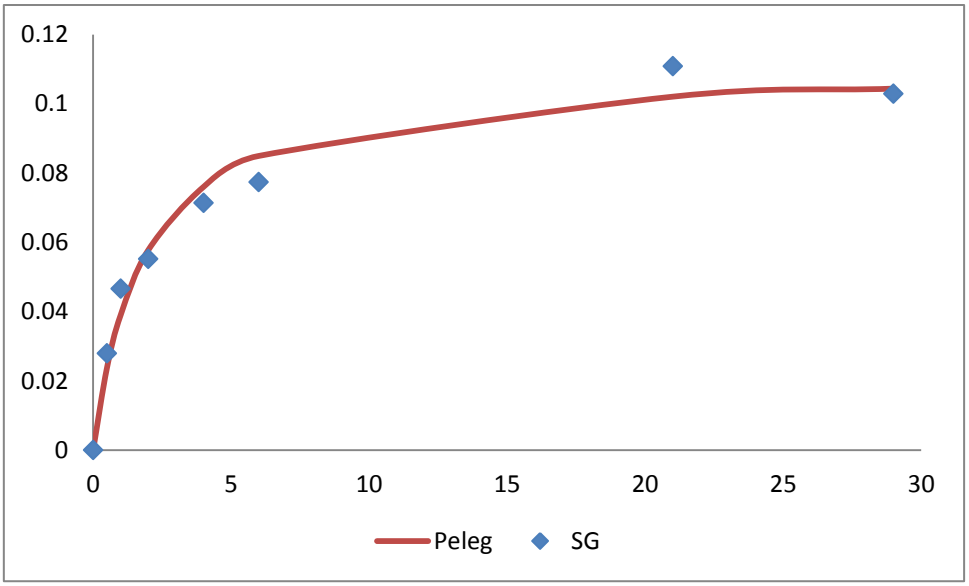


**Figure 10 Sugar gain (g of sugar/g of fresh mango cubes) as function of time of OD(h) of Kent mango cubes applied to peleg equation for batch B.**

From Figure 9 and Figure 10, the soluble solid gain changed with time of OD is given for batch A and batch B. Same as water loss, a pattern can be easily observed, soluble solid gain increased with the time of OD increases. According to Contreras and Smyrl's article, rapid removal of water and uptake of solids in the early stages can be observed in OD of apples(Contreras & Smyrl, 1981). Kowalska concluded that for some fruit and vegetables, the highest rate of water loss and sugar gain took place at the first 5-10 min of the OD process(Kowalska; & Lenart, 2001). Nevertheless, in the first half an hour, sugar has not entered the mango cubes, this could be caused by the Calcium and PME added. While in the first half an hour, no big difference for the solid gain, for batch B, it slight decreased to negative, which means soluble solid actually leached out from mango cubes, however, this only happened to one batch, it cannot reach to an conclusion yet. During 0.5h to 1h, the soluble solid gain increased significantly within half an hour, in the next 3 hour, soluble solid gain keep increasing to around 5% with a slower rate. After 21h, soluble solid has reached a value around 8%, and it seemed like no tread for further changes. However, batch A and B have significant difference at the same time set point, the error cannot be ignored but could be improved in the future. However, a significant difference can be found between two batches, there could be two explanation for this situation. On the one hand only a few cubes is used for measuring TTA, those cubes could came from different mangoes or different part of mango, Padda mentioned that in the center of the fruit, near the seed total soluble solid content is higher since the ripening starts from the middle(Padda, do Amarante, Garcia, Slaughter, & Mitcham, 2011). This could cause not fully homogenised sample used, some exceptional value could appears. On the other hand, Batch B was prepared one day after Batch A, in maturity stage 5, a consistent increasing in total soluble solids content is expected during ripening.

The Peleg's model parameter k1 and k2 is shown in table 19.

### 5.3.3. Weight reduction

From the Figure 11 below, the change of weight reduction of mango cubes is presented, similar to water loss, the first half an hour, the weight reduced at higher speed, and after half an hour, the reduction slowed down gradually, and around 6 hour, it reached to 52% and no big difference can be observed after that.

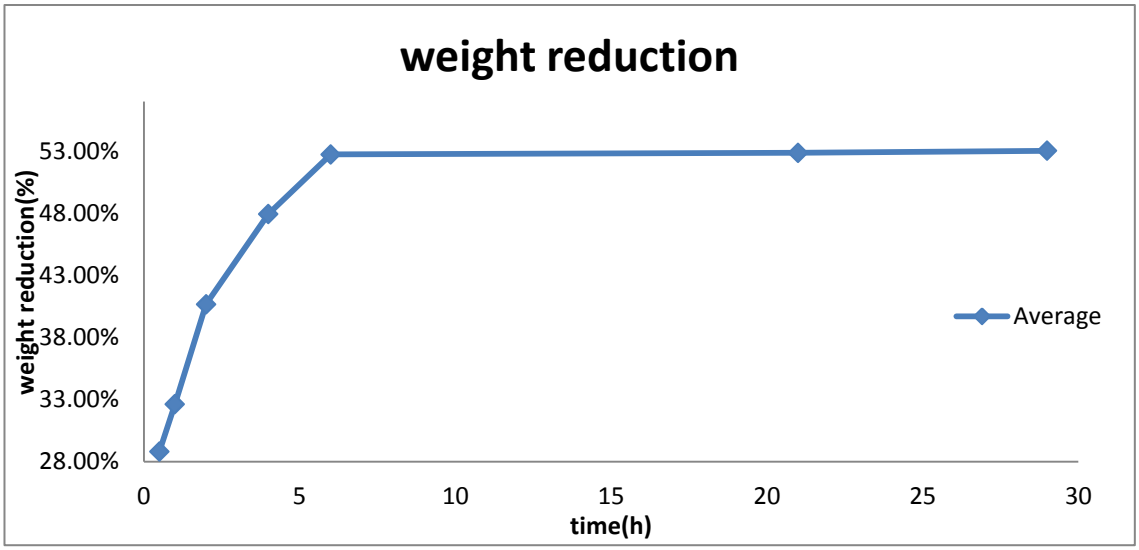


**Figure 11 Weight reduction as function of time of OD(h) of Kent mango cubes for average of two batches with standard deviation.**

### 5.3.4. OD performance

From the Figure 12 below, the OD performances of different OD treatment time is displayed, 0.5h of OD for batch A has an extremely high value around 260 due to low soluble solid contents, the value is excluded. Unlike the other three criteria, OD performance has no trend in general, and between batch A and B, there is nothing in common that could reach to a conclusion. One thing can be observed, shorter OD treatment time has better OD performance index than longer OD treatment.

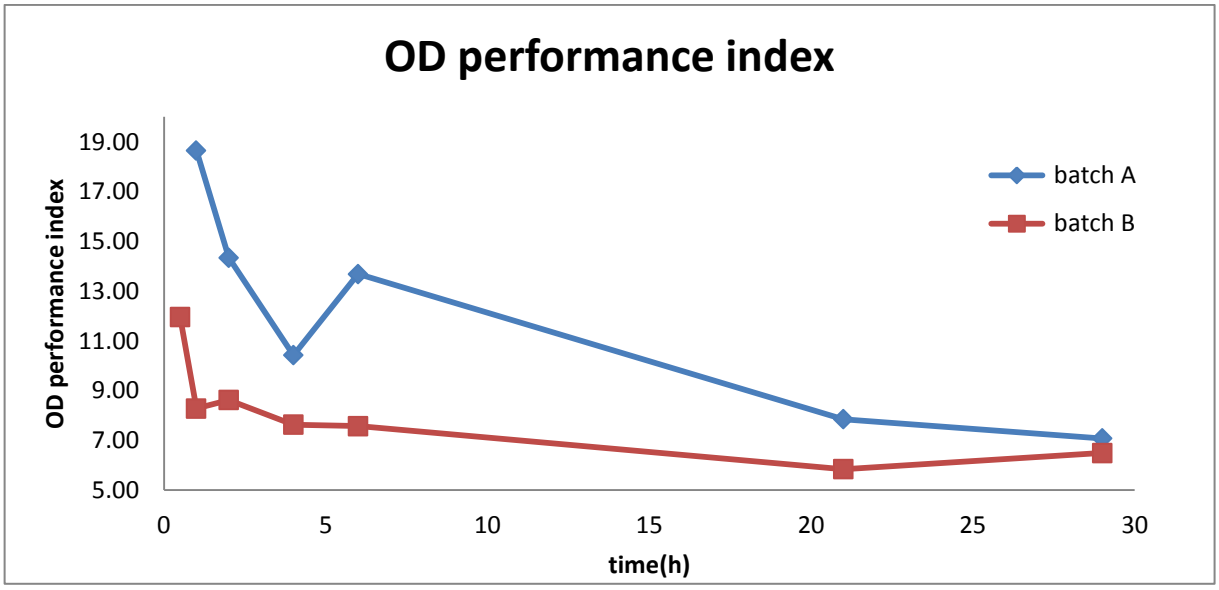


**Figure 12 OD performance Index as function of time of OD(h) of Kent mango cubes for two batches. Blue line, batch A; Red line, batch B.**

## 5.4.Water activity

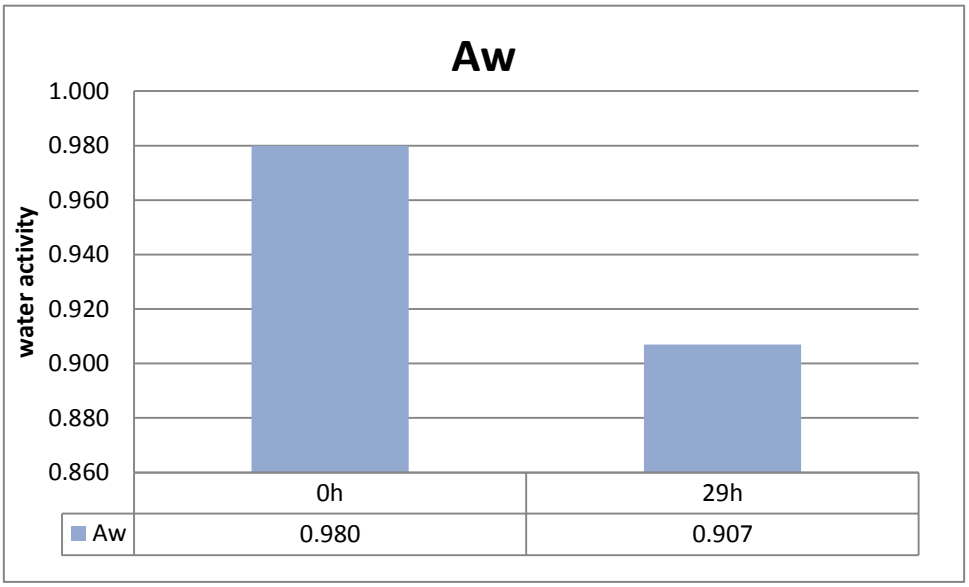


**Figure 13 Water activity average for fresh mango cubes(0h) and maximum OD treated mango cubes(29h).**

Figure 13 about showed that Water activity of fresh mango cubes was around 0.98, after 29 hours of OD treatment, water activity decreases to around 0.9. These shown that with the loss of water also influence the water activity. Water activity is a critical indicator for shelf life of the mango cubes because of microbial stability(Guiamba, 2016).

## 5.5.Vitamin C analysis

The chromatogram for reference mango provided from FQD technical is shown in Figure 19, the peak is clear and there is no shoulder before or between or after, and the peak can be easily observed. This is the example graph for good results. Figure 20 is one of the chromatogram that can be used directly by the auto assigned peaks. And in Figure 21 is an example for most of the chromatograms that is not ideal, however, the peak can still be used, but the area is not accurate for constructed a model. And Figure 22 is one of the chromatogram for OD solution after OD treatment, there are several peaks in the run and the ascorbic acid peak cannot be defined and for this reason, the vitamin C concentration in OD solution was not studied.

The Figure below (Figure 13)is the standard calibration curve made from ascorbic acid stock solution in MPA THBQ. The concentration range chosen for calibration is from 1.56 µg/ml to 25 µg/ml. even the original calibration curve has high concentrated vitamin C solution up to 200 µg/ml, however the sample used in experiment is far lower than 200 µg/ml, to have the best fit, only concentration points below 25 µg/ml were chosen. There were several calibration curve equation available depend on the numbers of parameter and the intercept. Since most of peak area is too small, , the y-intercept was chosen as 0, to avoid negative value for y. the equation obtained was y=1.1828x with coefficient of determination (R2) around 0.9999.

However most of the peak area for experiment sample is smaller than the peak area of 1.56 µg/ml, that means to fit those point to the calibration curve, the sample data will be mostly located in the bottom left area (marked with red ring) which is out of calibration range.

From the calibration curve in Figure 18 in appendix, to plot AA and TAA to a graph, the average for batch A and B was shown in Figure 14, however it is obvious that the AA concentration was very different for replicates, standard deviation for some point exceed 100% for the minimum value. The measurement for AA is unstable due to various reasons.

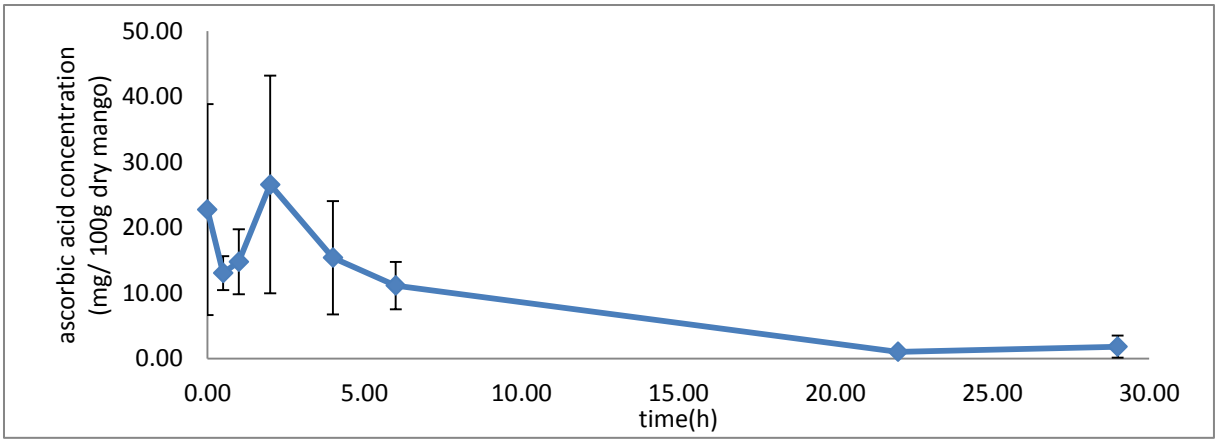


**Figure 14 Ascorbic acid concentration in 100g dry mango as function of time of OD(h) of Kent mango cubes for average of two batches with standard deviation.**

As for the Figure 15 shown quite a trend for TAA concentration below. Unlike AA concentration, the standard deviation for TAA was within the range. The TAA concentration for fresh mango was around 130mg/100g of dry mango, and after 0.5h, the concentration dropped to 60mg/100g, about 50% of TAA leached out of mango cubes, and this massive vitamin C loss can be confirmed from several experiment before(Guiamba, 2016) (Guiamba, Ahrné, Khan, & Svanberg, 2016) ,vitamin C kept leaching out of mango till there is limited amount left after 29h.

However to determine the DHA content, by calculation for TAA minus AA is not valid in this case since the standard deviation of AA content is too significant. About 110mg of DHA was presented in the fresh mango cubes according to the calculation, that has never been recorded in any literature, to be precise, the data obtained from this experiment is not accurate, a conclusion cannot be reached.

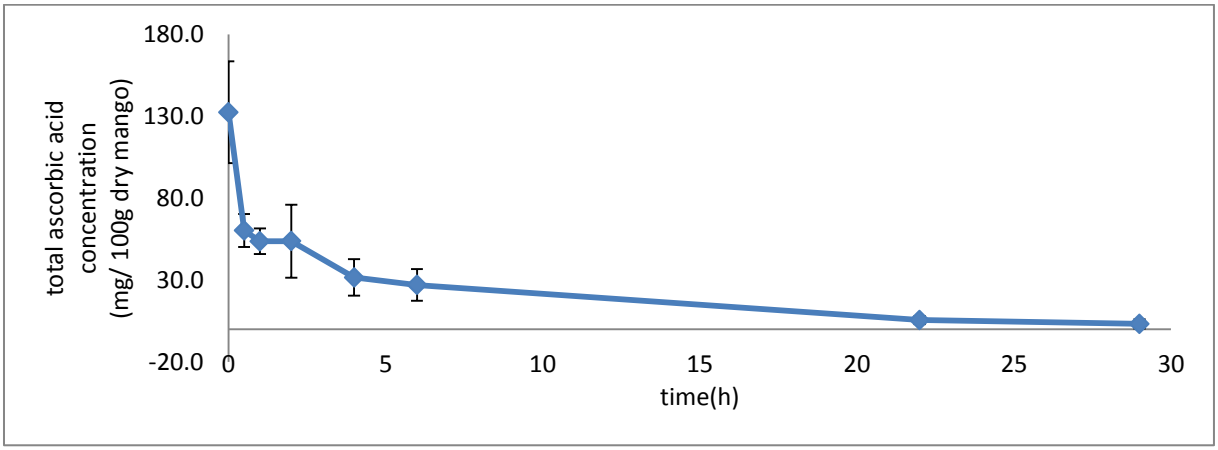


**Figure 15 Total ascorbic acid concentration in 100g dry mango as function of time of OD(h) of Kent mango cubes for average of two batches with standard deviation.**

## 5.6.Visual appearance

It can be observed from the picture of Figure 16 and Figure 17, with OD time increases, mango cubes shrink to a small size with a dark yellowish surface due to water loss. The browning colour occurred is an indicator for the mango cubes has reached the maximum process time. The quality can no longer assured, and 29h(AA or BA) should be the maximum time point to investigate OD treatment influences.

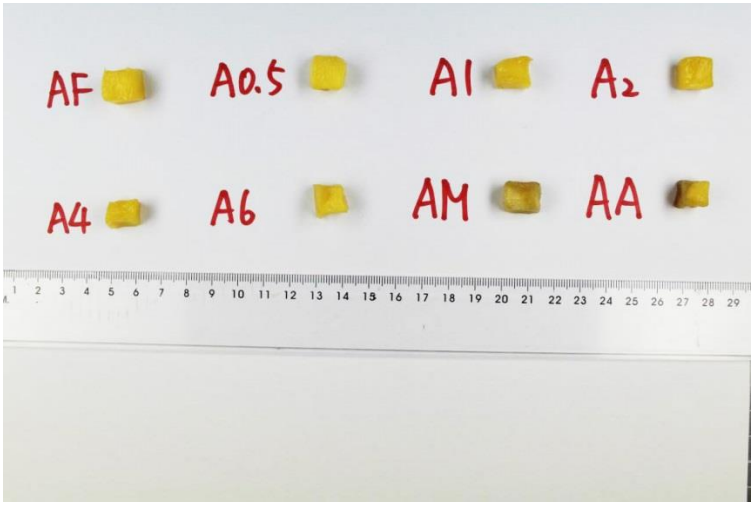


**Figure 16 Picture for appearance for batch A of fresh,0.5h,1h,2h,3h,5h,21h(AM),29h(AA) of OD treated mango cubes.**

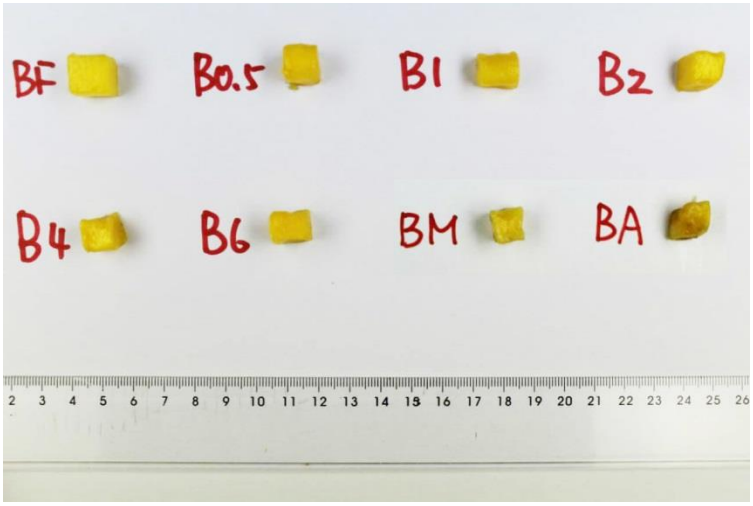


**Figure 17 Picture for appearance for batch A of fresh,0.5h,1h,2h,3h,5h,21h(BM),29h(BA) of OD treated mango cubes.**

# 6. Conclusion

pH of mango cubes decreased with the increased OD treatment time, with longer OD treatment time, the original OD solution became acidic due to the leaching of acid component in mango cubes. The pH value of OD solution did not change significantly.

A large amount of water losses during the first half hour of OD, it stop leaching out of mango cubes after around 6 hours of OD treatment. The soluble solid gain start increases dramatically after 1 hour, the growth slows down and increases to around 9% after 21h. Weight reduces fast in first half an hour, and slows down after that, weight keep loss in the first 6 hour and reaches to a level of 52% and the weight stop changing. No trend can be found for OD performance, while at 1h and 21h, OD performance has some good results, but this phenomenon can be not observed in duplicates.

Despite the significant drop of pH in the first half an hour, and after one hour of OD, the pH became stable. TSS starting from around 16, TSS increased with time even after 29 hours to more than 54. Water activity of fresh mango was quite different from 29 hours treated mango, Aw decreased from 0.980 to 0.907. However as a important characteristic for dry mango, Aw should be considered as part of the design. The water loss during OD raise with OD time and reached to the maximum value after around 4 hours. The same trend for weight reduction, weight reduced with time of OD increases while the rate decreases(Yadav & Singh, 2014).The soluble solid on the contrary dropped with time, it can be concluded that soluble solid in the mango cubes have leaching out with longer treatment time. The OD performance of different time point is in doubt due to high standard deviation. However the trend can be

summarized in the first 4 to 6 hours, longer time of OD treatment gave a better OD performance, after 6 hours, OD performance dropped to minus, and 29 hours has better performance than 22 hours.

From the chromatogram, the vitamin C content showed an odd picture. In 90% of chromatograms, unknown peaks shown in the picture all the time, this could be the impurity in the supernatant, those unknown component could affect the final content of vitamin C, by blocking the way of vitamin C in the column. To compare with the reference mango, those peaks are not shown, it can be conclude that there is no contamination during the preparation of supernatant, since the reference mango was prepared under same condition. In some of the chromatograms, at the last few second of responding time there is a shoulder shown up at ascorbic acid peak, some of those shoulders are too big that can no longer be ignored, due to the presence of another unresolved component, and cause trouble for identification, since it could be split peak judged by retention time. For various reasons mentioned above, the vitamin C content cannot be calculated. The only conclusion could be made is that with the longer time of OD treatment, vitamin C present in mango cubes is decreasing, and vitamin C content is relatively low after OD treatment.

# 7. Recommendation

- In previous studies, the durometer is suggested for low fruit firmness, since the durometer provided better resolution for fruit during later stages of ripening(Padda et al., 2011), for future study this could be used as well.
- The chosen mango maturity stage 5 is the last stage of ripening, the mango are too ripen at this moment with vulnerable structure, in future study, earlier stage mango is recommended for a more stable research outcome.
- To study later stages, more developed method should be applied, late stage mango has soft tissue structure, the structure of cubes is hard to maintain during OD treatment.
- After 4 hours of OD treatment, most of the analyses show out the similar characteristics, to select a better time point should be: choose more time point in the first 4 hours and less in the longer time range. Especially the first 1h, since most of the water loss occurs during this time.
- The trial of QUENCHER method shows that the total antioxidant is too low to be calculated. since the high sugar content present in the cubes made a undesired mixing of dry mango powder with cellulose , this preparation method is not valid for mango cubes treated under high sugar condition. For future experiment, new method should be invented for this particular characteristic.
- As soon the OD treatment is done, mango cubes should be dried by a towel as soon as possible, and freeze by liquid nitrogen, and for further use, it should be separate to different containers since there are multiple analyses need to be done and the frozen sample should not be thawed twice.
- For HPLC analysis, samples should be prepared at the same day, otherwise, it should be kept at a freezer.
- The mango cubes used for HPLC is wet based and because of the high water content, the characteristics might have changed in between preparation, dry mango powder should be used, and freeze drying method is highly recommended for solving this problem.
- The calibration curve is prepared with other students and the concentration range is not suitable for this study, in the next experiment, a low range of concentration of ascorbic acids should be used to make sure that experimented sample are within the calibration range.
- HPLC analysis shown strange peak in the chromatogram, to identify what kind of unresolved component is disturbing should be put more attention on, and try to eliminate those uncertainty.
- It is suggested that the unknown peaks should be study in the future to identify what kind of compound is causing this situation.
- To modelling the leaching kinetics however still need to analysis the vitamin C content in the OD solution after OD treatment, to determine what is the path of vitamin C, whether degradation occurs or leaching into OD solution.
- The chemical used during this experiment could also be the reason of bad chromatograms, the chemical might have been contaminated by others.
- For most of the protocol used during this experiment is valid for our sample, however, some of the protocol is not suitable for sensitive composition and low concentration, to adjust some of the protocol for the use of our experiment should be priority for the study.
- Addition of 1% AA to OD solution is suggested by much research(Michael Robbers, 1997; Nagai, Santos, Faria, Boscolo, & Mauro, 2015), it not only prevented losses during OD treatment, but in some cases could resulted in impregnation of the substance into the mango as well.

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# Appendices

**Table 5 Weight for mango cubes and OD solution before and after OD and PME used in each as 0.048% for batch A and batch B for all the od time points.**

| | Batch A | | | | | Batch B | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| time(h) | mango weight | | solution weight | | PME | mango weight | | solution weight | | PME |
| | before | after | before | after | | before | after | before | after | |
| | (g) | (g) | (g) | (g) | (mL) | (g) | (g) | (g) | (g) | (mL) |
| Fresh | / | / | / | / | / | / | / | / | / | / |
| 0.5 | 209.8 | 148.2 | 839.2 | 857 | 4.03 | 254 | 182.26 | 1016 | 1035.6 | 4.88 |
| 1 | 200.1 | 135.6 | 801 | 838.3 | 3.84 | 224 | 150.06 | 896 | 936.5 | 4.30 |
| 2 | 199.1 | 116.9 | 796.4 | 854.3 | 3.82 | 248.2 | 148.77 | 992.8 | 1051.3 | 4.77 |
| 4 | 164.4 | 84.2 | 657.6 | 710.3 | 3.16 | 229.9 | 121.63 | 919.5 | 980.9 | 4.41 |
| 6 | 189 | 84.66 | 756.4 | 830.4 | 3.63 | 242.5 | 120.62 | 970 | 1055.9 | 4.66 |
| 21 | 173 | 81.1 | 694 | 726.1 | 3.32 | 234.4 | 111.07 | 937.6 | 1030.6 | 4.50 |
| 29 | 170.8 | 84.15 | 683.2 | 736.0 | 3.28 | 239.7 | 107.07 | 958.7 | 1014.3 | 4.60 |

**Table 6 Weight and maturity stage check for raw mango used for experiment.**

| | Batch A | | Batch B | |
|---|---|---|---|---|
| No | weight (g) | maturity stage | weight (g) | maturity stage |
| 1 | 470 | 5 | 408 | 5 |
| 2 | 400 | 5 | 415.66 | 5 |
| 3 | 396 | 5 | 417.76 | 5 |
| 4 | 417.3 | 5 | 428.58 | 5 |
| 5 | 450.6 | 5 | 411.04 | 5 |
| 6 | 341.4 | 5 | 421.62 | 5 |
| 7 | 386.2 | 5 | 408.86 | 5 |
| 8 | 415 | 5 | 415.08 | 5 |
| 9 | 399.1 | 5 | 368.57 | 5 |
| 10 | 435.4 | 5 | 461.33 | 5 |
| 11 | 426.5 | 5 | 407.73 | 5 |
| 12 | 395.4 | 5 | 446.68 | 5 |
| 13 | 437.3 | 5 | 395.04 | 5 |
| 14 | 380.9 | 5 | 443.11 | 5 |
| 15 | 446.6 | 5 | 432.09 | 5 |
| 16 | 391.2 | 5 | 446.86 | 5 |
| 17 | 434 | 5 | 414.43 | 5 |
| 18 | 411 | 5 | 370.03 | 5 |
| 19 | 457.2 | 5 | 388.65 | 5 |
| 20 | 455.5 | 5 | 473.63 | 5 |
| 21 | 425.1 | 5 | 454.16 | 5 |

| | | | | |
|---|---|---|---|---|
| **22** | 387.4 | 5 | 464.02 | 5 |
| **23** | 444.4 | 5 | 417.57 | 5 |
| **24** | 436.5 | 5 | 450.46 | 5 |
| **25** | 450.6 | 5 | 428.22 | 5 |
| **26** | 426.2 | 5 | 461.02 | 5 |
| **27** | 450.5 | 5 | 441.42 | 5 |
| **28** | 477.6 | 5 | 415.78 | 5 |
| **29** | 444.51 | 5 | 426.02 | 5 |
| **30** | 428.76 | 5 | 364.09 | 5 |
| **31** | 429.3 | 5 | 416.03 | 5 |
| **32** | 400.58 | 5 | 350.1 | 5 |
| **33** | 420.3 | 5 | 412.89 | 5 |
| **34** | 467.12 | 5 | 422.89 | 5 |
| **35** | 442.16 | 5 | 411.98 | 5 |
| **36** | 451.29 | 5 | 349.08 | 5 |
| **37** | 392.13 | 5 | 413.76 | 5 |
| **38** | 429.28 | 5 | 429.4 | 5 |
| **39** | 422.56 | 5 | | |
| **40** | 402.54 | 5 | | |
| **41** | 390.75 | 5 | | |

**Table 7 pH, total soluble solid for all the od time points, water activity of fresh mango cubes and 29h of OD, volume of NaOH used for total titratable acid for fresh mango cubes for batch A and B.**

| | Batch A | | | | | Batch B | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **time(h)** | pH | | TSS | Aw | TTA | pH | | TSS | Aw | TTA |
| | mango | sucrose | brix | | | mango | sucrose | brix | | |
| | | | g/100g | | mL | | | g/100g | | mL |
| **Fresh** | 4.174 | 7.019 | 17.6 | 0.986 | 3.83 | 4.068 | 7.021 | 14.5 | 0.974 | 4.69 |
| **0.5** | 4.092 | 5.143 | 25.1 | | | 3.874 | 5.112 | 24.1 | | |
| **1** | 3.907 | 5.025 | 29.1 | | | 3.895 | 4.995 | 28.6 | | |
| **2** | 3.903 | 4.855 | 35.6 | | | 3.924 | 4.895 | 33.4 | | |
| **4** | 3.931 | 4.731 | 45.4 | | | 3.994 | 4.854 | 40.9 | | |
| **6** | 3.953 | 4.687 | 49.8 | | | 4.002 | 4.821 | 44.7 | | |
| **21** | 4.165 | 4.54 | 54.9 | | | 4.156 | 4.526 | 54 | | |
| **29** | 4.264 | 4.534 | 54.1 | 0.913 | | 4.27 | 4.494 | 55.5 | 0.901 | |

**Table 8 Dry matter for duplicates as dry matter 1 and dry matter 2 and calculated water content based on the dry fruit/mango cube pulp for all the OD time points.**

| | Batch A | | | Batch B | | |
|---|---|---|---|---|---|---|
| **time(h)** | dry matter 1 | dry matter 2 | water | dry matter 1 | dry matter 2 | water |

| | | | | | content | | | | | content |
|---|---|---|---|---|---|---|---|---|---|---|
| | before | after | before | after | dry/cubes | before | after | before | after | dry/cubes |
| | (g) | (g) | (g) | (g) | % | (g) | (g) | (g) | (g) | % |
| **Fresh** | 2.335 | 0.314 | 2.017 | 0.285 | 0.134 | 1.720 | 0.239 | 1.818 | 0.271 | 0.134 |
| **0.5** | 1.550 | 0.400 | 1.282 | 0.343 | 0.258 | 1.339 | 0.344 | 1.158 | 0.334 | 0.258 |
| **1** | 1.176 | 0.364 | 1.106 | 0.346 | 0.310 | 1.161 | 0.337 | 1.276 | 0.390 | 0.310 |
| **2** | 1.257 | 0.424 | 1.308 | 0.443 | 0.337 | 1.200 | 0.429 | 1.105 | 0.414 | 0.337 |
| **4** | 1.563 | 0.700 | 1.751 | 0.851 | 0.448 | 0.994 | 0.381 | 0.951 | 0.418 | 0.448 |
| **6** | 1.088 | 0.557 | 0.998 | 0.513 | 0.512 | 1.013 | 0.445 | 0.857 | 0.406 | 0.512 |
| **21** | 0.776 | 0.388 | 1.011 | 0.553 | 0.500 | 0.657 | 0.373 | 0.767 | 0.424 | 0.500 |
| **29** | 1.189 | 0.678 | 0.820 | 0.436 | 0.570 | 0.725 | 0.420 | 0.910 | 0.527 | 0.570 |

**Table 9 pH for mango cubes and OD solution after each OD time points for batch A and Batch B with the average and standard deviation.**

| | **pH** | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Mango | | | | OD solution | | | |
| **time(h)** | Batch A | Batch B | Average | SD | Batch A | Batch B | Average | SD |
| **0** | 4.174 | 4.068 | 4.121 | 0.075 | 7.019 | 7.021 | 7.020 | 0.001 |
| **0.5** | 4.092 | 3.874 | 3.983 | 0.154 | 5.143 | 5.112 | 5.128 | 0.022 |
| **1** | 3.907 | 3.895 | 3.901 | 0.008 | 5.025 | 4.995 | 5.010 | 0.021 |
| **2** | 3.903 | 3.924 | 3.914 | 0.015 | 4.855 | 4.895 | 4.875 | 0.028 |
| **4** | 3.931 | 3.994 | 3.963 | 0.045 | 4.731 | 4.854 | 4.793 | 0.087 |
| **6** | 3.953 | 4.002 | 3.978 | 0.035 | 4.687 | 4.821 | 4.754 | 0.095 |
| **21** | 4.165 | 4.156 | 4.161 | 0.006 | 4.540 | 4.526 | 4.533 | 0.010 |
| **29** | 4.264 | 4.270 | 4.267 | 0.004 | 4.534 | 4.494 | 4.514 | 0.028 |

**Table 10 Total soluble solid measurement by brix for all the OD time points of batch A and Batch B and average and standard deviation.**

| | **Brix** | | | |
|---|---|---|---|---|
| **time(h)** | Batch A | Batch B | average | SD |
| **0** | 17.6 | 14.5 | 16.1 | 2.2 |
| **0.5** | 25.1 | 24.1 | 24.6 | 0.7 |
| **1** | 29.1 | 28.6 | 28.9 | 0.4 |
| **2** | 35.6 | 33.4 | 34.5 | 1.6 |
| **4** | 45.4 | 40.9 | 43.2 | 3.2 |
| **6** | 49.8 | 44.7 | 47.3 | 3.6 |
| **21** | 54.9 | 54.0 | 54.5 | 0.6 |
| **29** | 54.1 | 55.5 | 54.8 | 1.0 |

**Table 11 Water activity for fresh mango cubes and 29h OD treated mango cubes for batch A and Batch B and average and standard deviation.**

| | Aw | | | |
|---|---|---|---|---|
| **time(h)** | Batch A | Batch B | average | SD |
| **0h** | 0.986 | 0.974 | 0.980 | 0.008 |
| **29h** | 0.913 | 0.901 | 0.907 | 0.008 |

**Table 12 Total titratable acid measurement for fresh mango cubes for batch A and batch B.**

| | TTA | |
|---|---|---|
| **Fresh mango cubes** | Batch A | Batch B |
| **NaOH used(mL)** | 3.830 | 4.690 |
| **acid percentage** | 0.245 | 0.300 |
| **sugar acid ratio** | 72% | 48% |

**Table 13 Ascorbic acids concentration and HPLC peak area for batch A.**

| **Batch A** | | **y** | | | | **x** | | |
|---|---|---|---|---|---|---|---|---|
| | Injection Name | Area | mango weight | mango weight | supernatant | Conc of Vit C | content of AAVitC | average |
| | Selected Peak: | mV*min | g | g | ml | µg/ml | mg/ 100g mango | |
| | ascorbic acid | ascorbic acid | fresh mango | dry base | | | | |
| **Reference mango** | REF BA | 26.758 | 0.252 | | 8.847 | 22.623 | 79.518 | 79.518 |
| **fresh** | FAA | 1.874 | 0.278 | 0.037 | 9.742 | 1.584 | 41.299 | |
| **fresh** | FAB | 1.602 | 0.278 | 0.037 | 9.742 | 1.354 | 35.297 | |
| **fresh** | FBA | 1.064 | 0.281 | 0.037 | 10.839 | 0.900 | 26.100 | |
| **fresh** | FBB | 1.379 | 0.281 | 0.037 | 10.839 | 1.166 | 33.809 | 34.126 |
| **0.5h** | 0.5AA | 1.173 | 0.276 | 0.071 | 10.093 | 0.992 | 14.048 | |
| **0.5h** | 0.5AB | 1.326 | 0.276 | 0.071 | 10.093 | 1.121 | 15.878 | |
| **0.5h** | 0.5BA | 1.074 | 0.286 | 0.071 | 9.992 | 0.908 | 12.735 | |
| **0.5h** | 0.5BB | 1.425 | 0.286 | 0.071 | 9.992 | 1.205 | 16.900 | 14.890 |
| **1h** | 1AA | 1.254 | 0.266 | 0.082 | 9.793 | 1.060 | 12.625 | |
| **1h** | 1AB | 1.572 | 0.266 | 0.082 | 9.793 | 1.329 | 15.830 | |
| **1h** | 1BA | 1.717 | 0.296 | 0.082 | 10.547 | 1.452 | 18.620 | |
| **1h** | 1BB | 2.408 | 0.296 | 0.082 | 10.547 | 2.036 | 26.109 | 18.296 |
| **2h** | 2AA | 1.715 | 0.255 | 0.086 | 10.223 | 1.450 | 17.238 | |
| **2h** | 2AB | 1.395 | 0.255 | 0.086 | 10.223 | 1.179 | 14.016 | |
| **2h** | 2BA | 1.509 | 0.258 | 0.086 | 10.061 | 1.276 | 14.925 | |
| **2h** | 2BB | 1.322 | 0.258 | 0.086 | 10.061 | 1.118 | 13.077 | 14.814 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **4h** | 4AA | 0.984 | 0.255 | 0.114 | 10.558 | 0.832 | 7.677 | |
| **4h** | 4AB | 1.287 | 0.255 | 0.114 | 10.558 | 1.088 | 10.045 | |
| **4h** | 4BA | 1.435 | 0.260 | 0.114 | 10.086 | 1.213 | 10.700 | |
| **4h** | 4BB | 1.172 | 0.260 | 0.114 | 10.086 | 0.991 | 8.739 | 9.290 |
| **6h** | 6AA | 1.484 | 0.252 | 0.129 | 9.995 | 1.254 | 9.703 | |
| **6h** | 6AB | 1.270 | 0.252 | 0.129 | 9.995 | 1.073 | 8.303 | |
| **6h** | 6BA | 0.982 | 0.256 | 0.129 | 10.422 | 0.830 | 6.694 | |
| **6h** | 6BB | 1.419 | 0.256 | 0.129 | 10.422 | 1.199 | 9.673 | 8.593 |
| **21h** | MAA | 0.120 | 0.254 | 0.127 | 10.255 | 0.102 | 0.823 | |
| **21h** | MAB | 0.116 | 0.254 | 0.127 | 10.255 | 0.098 | 0.794 | |
| **21h** | MBA | 0.128 | 0.250 | 0.127 | 10.281 | 0.108 | 0.874 | |
| **21h** | MBB | 0.105 | 0.250 | 0.127 | 10.281 | 0.089 | 0.720 | 0.803 |
| **29h** | AAA | 2.608 | 0.508 | 0.290 | 9.633 | 2.205 | 7.329 | |
| **29h** | AAB | 0.964 | 0.508 | 0.290 | 9.633 | 0.815 | 2.707 | |
| **29h** | ABA | 0.329 | 0.510 | 0.290 | 10.061 | 0.278 | 0.964 | |
| **29h** | ABB | 0.360 | 0.510 | 0.290 | 10.061 | 0.304 | 1.057 | 3.014 |

**Table 14 Total ascorbic acids concentration and HPLC peak area for batch A.**

| **Batch A** | | **y** | | | | **x** | | |
|---|---|---|---|---|---|---|---|---|
| | Injection Name | Area | mango weight | mango weight | supernatant | Conc of Vit C | content of TAAVitC | average |
| | Selected Peak: | mV*min | g | g | ml | µg/ml | mg/ 100g mango | |
| | ascorbic acid | ascorbic acid | fresh mango | dry base | | | | |
| | | UV_VIS_1 | | | | | | |
| **Reference mango** | RAAT | 56.153 | 0.263 | 0.263 | 9.514 | 47.474 | 171.794 | |
| **Reference mango** | RABT | 33.105 | 0.263 | 0.263 | 9.514 | 27.989 | 101.283 | |
| **Reference mango** | RBAT | 84.260 | 0.252 | 0.263 | 8.847 | 71.238 | 239.730 | |
| **Reference mango** | RBBT | 27.846 | 0.252 | 0.263 | 8.847 | 23.542 | 79.224 | 148.008 |
| **fresh** | FAAT | 4.188 | 0.278 | 0.037 | 9.742 | 3.541 | 92.302 | |
| **fresh** | FABT | 5.690 | 0.278 | 0.037 | 9.742 | 4.811 | 125.415 | |
| **fresh** | FBAT | 3.959 | 0.281 | 0.037 | 10.839 | 3.347 | 97.070 | |
| **fresh** | FBBT | 5.174 | 0.281 | 0.037 | 10.839 | 4.375 | 126.877 | 110.416 |
| **0.5h** | 0.5AAT | 3.739 | 0.276 | 0.071 | 10.093 | 3.161 | 44.776 | |
| **0.5h** | 0.5ABT | 4.507 | 0.276 | 0.071 | 10.093 | 3.810 | 53.971 | |
| **0.5h** | 0.5BAT | 4.146 | 0.286 | 0.071 | 9.992 | 3.505 | 49.156 | |
| **0.5h** | 0.5BBT | 5.430 | 0.286 | 0.071 | 9.992 | 4.591 | 64.380 | 53.071 |
| **1h** | 1AAT | 3.430 | 0.266 | 0.082 | 9.793 | 2.900 | 34.536 | |
| **1h** | 1ABT | 4.304 | 0.266 | 0.082 | 9.793 | 3.638 | 43.328 | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **1h** | 1BAT | 4.494 | 0.296 | 0.082 | 10.547 | 3.799 | 48.724 | |
| **1h** | 1BBT | 6.073 | 0.296 | 0.082 | 10.547 | 5.134 | 65.842 | 48.107 |
| **2h** | 2AAT | 4.138 | 0.255 | 0.086 | 10.223 | 3.498 | 41.577 | |
| **2h** | 2ABT | 3.572 | 0.255 | 0.086 | 10.223 | 3.020 | 35.897 | |
| **2h** | 2BAT | 4.054 | 0.258 | 0.086 | 10.061 | 3.428 | 40.091 | |
| **2h** | 2BBT | 3.476 | 0.258 | 0.086 | 10.061 | 2.939 | 34.371 | 37.984 |
| **4h** | 4AAT | 2.520 | 0.255 | 0.114 | 10.558 | 2.130 | 19.661 | |
| **4h** | 4ABT | 3.304 | 0.255 | 0.114 | 10.558 | 2.793 | 25.781 | |
| **4h** | 4BAT | 3.518 | 0.260 | 0.114 | 10.086 | 2.974 | 26.225 | |
| **4h** | 4BBT | 3.066 | 0.260 | 0.114 | 10.086 | 2.592 | 22.853 | 23.630 |
| **6h** | 6AAT | 3.484 | 0.252 | 0.129 | 9.995 | 2.945 | 22.782 | |
| **6h** | 6ABT | 2.976 | 0.252 | 0.129 | 9.995 | 2.516 | 19.459 | |
| **6h** | 6BAT | 2.272 | 0.256 | 0.129 | 10.422 | 1.921 | 15.495 | |
| **6h** | 6BBT | 3.316 | 0.256 | 0.129 | 10.422 | 2.803 | 22.610 | 20.086 |
| **21h** | MAAT | 0.504 | 0.254 | 0.127 | 10.255 | 0.426 | 3.446 | |
| **21h** | MABT | 0.719 | 0.254 | 0.127 | 10.255 | 0.608 | 4.918 | |
| **21h** | MBAT | 0.587 | 0.250 | 0.127 | 10.281 | 0.496 | 4.023 | |
| **21h** | MBBT | 0.422 | 0.250 | 0.127 | 10.281 | 0.357 | 2.894 | 3.820 |
| **29h** | AAAT | 2.368 | 0.508 | 0.290 | 9.633 | 2.002 | 6.653 | |
| **29h** | AABT | 2.391 | 0.508 | 0.290 | 9.633 | 2.021 | 6.717 | |
| **29h** | ABAT | 1.146 | 0.510 | 0.290 | 10.061 | 0.969 | 3.362 | |
| **29h** | ABBT | 1.240 | 0.510 | 0.290 | 10.061 | 1.048 | 3.639 | 5.093 |

**Table 15 Ascorbic acids concentration and HPLC peak area for batch B**

| **Batch B** | | **y** | | | | **x** | | |
|---|---|---|---|---|---|---|---|---|
| | Injection Name | Area | mango weight | mango weight | supernatant | Conc of Vit C | content of TAAVitC | average |
| | Selected Peak: | mV*min | g | g | ml | µg/ml | mg/ 100g mango | |
| | ascorbic acid | ascorbic acid | fresh mango | dry base | | | | |
| **Reference mango** | REF BB | 35.134 | 0.247 | 0.247 | 9.227 | 29.704 | 110.872 | 110.872 |
| **fresh** | FCA | 0.536 | 0.264 | 0.037 | 10.079 | 0.453 | 12.436 | |
| **fresh** | FCB | 0.645 | 0.264 | 0.037 | 10.079 | 0.545 | 14.962 | |
| **fresh** | FDA | 0.275 | 0.285 | 0.037 | 10.568 | 0.232 | 6.680 | |
| **fresh** | FDB | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 0.000 | 11.359 |
| **0.5h** | 0.5CA | 0.909 | 0.280 | 0.072 | 10.004 | 0.769 | 10.706 | |
| **0.5h** | 0.5CB | 0.936 | 0.280 | 0.072 | 10.004 | 0.791 | 11.019 | |
| **0.5h** | 0.5DA | 0.946 | 0.268 | 0.072 | 9.970 | 0.800 | 11.104 | |
| **0.5h** | 0.5DB | 1.029 | 0.268 | 0.072 | 9.970 | 0.870 | 12.071 | 11.225 |
| **1h** | 1CA | 1.092 | 0.268 | 0.078 | 9.544 | 0.923 | 11.339 | |
| **1h** | 1CB | 1.149 | 0.268 | 0.078 | 9.544 | 0.971 | 11.927 | |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **1h** | 1DA | 0.971 | 0.255 | 0.078 | 10.484 | 0.821 | 11.074 | |
| **1h** | 1DB | 0.944 | 0.255 | 0.078 | 10.484 | 0.798 | 10.769 | 11.277 |
| **2h** | 2CA | 5.865 | 0.255 | 0.091 | 10.100 | 4.959 | 55.025 | |
| **2h** | 2CB | 5.418 | 0.255 | 0.091 | 10.100 | 4.580 | 50.824 | |
| **2h** | 2DA | 2.744 | 0.260 | 0.091 | 10.527 | 2.319 | 26.826 | |
| **2h** | 2DB | 2.106 | 0.260 | 0.091 | 10.527 | 1.781 | 20.594 | 38.317 |
| **4h** | 4CA | 2.503 | 0.251 | 0.096 | 10.243 | 2.116 | 22.494 | |
| **4h** | 4CB | 3.625 | 0.251 | 0.096 | 10.243 | 3.065 | 32.582 | |
| **4h** | 4DA | 1.551 | 0.252 | 0.096 | 10.166 | 1.311 | 13.831 | |
| **4h** | 4DB | 1.928 | 0.252 | 0.096 | 10.166 | 1.630 | 17.198 | 21.526 |
| **6h** | 6CA | 1.358 | 0.256 | 0.112 | 10.346 | 1.148 | 10.560 | |
| **6h** | 6CB | 1.898 | 0.256 | 0.112 | 10.346 | 1.605 | 14.762 | |
| **6h** | 6DA | 1.645 | 0.254 | 0.112 | 10.178 | 1.391 | 12.585 | |
| **6h** | 6DB | 2.216 | 0.254 | 0.112 | 10.178 | 1.874 | 16.955 | 13.716 |
| **21h** | MCA | 0.200 | 0.253 | 0.143 | 10.107 | 0.169 | 1.193 | |
| **21h** | MCB | 0.259 | 0.253 | 0.143 | 10.107 | 0.219 | 1.540 | |
| **21h** | MDA | 0.161 | 0.252 | 0.143 | 10.754 | 0.136 | 1.023 | |
| **21h** | MDB | 0.216 | 0.252 | 0.143 | 10.754 | 0.183 | 1.368 | 1.281 |
| **29h** | ACA | 0.222 | 0.508 | 0.294 | 9.571 | 0.188 | 0.610 | |
| **29h** | ACB | 0.256 | 0.508 | 0.294 | 9.571 | 0.217 | 0.704 | |
| **29h** | ADA | 0.195 | 0.501 | 0.294 | 9.545 | 0.165 | 0.535 | |
| **29h** | ADB | 0.257 | 0.501 | 0.294 | 9.545 | 0.217 | 0.705 | 0.639 |

**Table 16 Total ascorbic acids concentration and HPLC peak area for batch B.**

| **Batch B** | | **y** | | | | **x** | | |
|---|---|---|---|---|---|---|---|---|
| | Injection Name | Area | mango weight | mango weight | supernatant | Conc of Vit C | content of TAAVitC | average |
| | Selected Peak: | mV*min | g | g | ml | µg/ml | mg/ 100g mango | |
| | ascorbic acid | ascorbic acid | fresh mango | dry base | | | | |
| **Reference mango** | REF CAT | 41.098 | 0.262 | 0.262 | 9.542 | 34.746 | 126.784 | |
| **Reference mango** | REF CBT | 40.769 | 0.262 | 0.263 | 9.542 | 34.468 | 125.100 | |
| **Reference mango** | REF DAT | 40.775 | 0.247 | 0.263 | 9.227 | 34.473 | 120.988 | |
| **Reference mango** | REF DBT | 42.161 | 0.247 | 0.263 | 9.227 | 35.645 | 125.101 | 124.493 |
| **fresh** | FCAT | 6.726 | 0.264 | 0.037 | 10.079 | 5.686 | 155.990 | |
| **fresh** | FCBT | 6.536 | 0.264 | 0.037 | 10.079 | 5.526 | 151.598 | |
| **fresh** | FDAT | 6.351 | 0.285 | 0.037 | 10.568 | 5.370 | 154.449 | |
| **fresh** | FDBT | 6.384 | 0.285 | 0.037 | 10.568 | 5.397 | 155.242 | 154.320 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **0.5h** | 0.5CAT | 5.500 | 0.280 | 0.072 | 10.004 | 4.650 | 64.758 | |
| **0.5h** | 0.5CBT | 5.505 | 0.280 | 0.072 | 10.004 | 4.654 | 64.824 | |
| **0.5h** | 0.5DAT | 5.972 | 0.268 | 0.072 | 9.970 | 5.049 | 70.075 | |
| **0.5h** | 0.5DBT | 5.911 | 0.268 | 0.072 | 9.970 | 4.998 | 69.362 | 67.255 |
| **1h** | 1CAT | 5.671 | 0.268 | 0.078 | 9.544 | 4.795 | 58.889 | |
| **1h** | 1CBT | 5.431 | 0.268 | 0.078 | 9.544 | 4.592 | 56.393 | |
| **1h** | 1DAT | 5.327 | 0.255 | 0.078 | 10.484 | 4.504 | 60.768 | |
| **1h** | 1DBT | 5.322 | 0.255 | 0.078 | 10.484 | 4.499 | 60.710 | 59.190 |
| **2h** | 2CAT | 9.377 | 0.255 | 0.091 | 10.100 | 7.927 | 87.965 | |
| **2h** | 2CBT | 9.116 | 0.255 | 0.091 | 10.100 | 7.707 | 85.523 | |
| **2h** | 2DAT | 4.961 | 0.260 | 0.091 | 10.527 | 4.194 | 48.504 | |
| **2h** | 2DBT | 5.708 | 0.260 | 0.091 | 10.527 | 4.825 | 55.807 | 69.450 |
| **4h** | 4CAT | 3.705 | 0.251 | 0.096 | 10.243 | 3.132 | 33.293 | |
| **4h** | 4CBT | 5.068 | 0.251 | 0.096 | 10.243 | 4.285 | 45.545 | |
| **4h** | 4DAT | 3.923 | 0.252 | 0.096 | 10.166 | 3.317 | 34.993 | |
| **4h** | 4DBT | 4.926 | 0.252 | 0.096 | 10.166 | 4.164 | 43.932 | 39.441 |
| **6h** | 6CAT | 3.568 | 0.256 | 0.112 | 10.346 | 3.017 | 27.753 | |
| **6h** | 6CBT | 4.609 | 0.256 | 0.112 | 10.346 | 3.897 | 35.851 | |
| **6h** | 6DAT | 4.104 | 0.254 | 0.112 | 10.178 | 3.470 | 31.404 | |
| **6h** | 6DBT | 5.284 | 0.254 | 0.112 | 10.178 | 4.467 | 40.426 | 33.859 |
| **21h** | MCAT | 1.081 | 0.253 | 0.143 | 10.107 | 0.914 | 6.439 | |
| **21h** | MCBT | 1.469 | 0.253 | 0.143 | 10.107 | 1.242 | 8.752 | |
| **21h** | MDAT | 0.938 | 0.252 | 0.143 | 10.754 | 0.793 | 5.947 | |
| **21h** | MDBT | 1.207 | 0.252 | 0.143 | 10.754 | 1.021 | 7.650 | 7.197 |
| **29h** | ACAT | 0.357 | 0.508 | 0.294 | 9.571 | 0.301 | 0.980 | |
| **29h** | ACBT | 0.481 | 0.508 | 0.294 | 9.571 | 0.407 | 1.322 | |
| **29h** | ADAT | 0.419 | 0.501 | 0.294 | 9.545 | 0.354 | 1.148 | |
| **29h** | ADBT | 0.473 | 0.501 | 0.294 | 9.545 | 0.400 | 1.296 | 1.187 |

**Table 17 Ascorbic acid concentration for all the OD time point.**

| **AA** | | | | |
|---|---|---|---|---|
| **time(h)** | batch A | batch B | average | sd |
| **ref** | 79.52 | 110.87 | 95.20 | 22.17 |
| **0** | 34.13 | 11.36 | 22.74 | 16.10 |
| **0.5** | 14.89 | 11.22 | 13.06 | 2.59 |
| **1** | 18.30 | 11.28 | 14.79 | 4.96 |
| **2** | 14.81 | 38.32 | 26.57 | 16.62 |
| **4** | 9.29 | 21.53 | 15.41 | 8.65 |
| **6** | 8.59 | 13.72 | 11.15 | 3.62 |
| **21** | 0.80 | 1.28 | 1.04 | 0.34 |
| **29** | 3.01 | 0.64 | 1.83 | 1.68 |

**Table 18 Total ascorbic acid concentration for all the OD time point.**

| TAA | | | | |
|---|---|---|---|---|
| **time(h)** | batch A | batch B | average | sd |
| **ref** | 148.01 | 124.49 | 136.25 | 16.63 |
| **0** | 110.42 | 154.32 | 132.37 | 31.04 |
| **0.5** | 53.07 | 67.25 | 60.16 | 10.03 |
| **1** | 48.11 | 59.19 | 53.65 | 7.84 |
| **2** | 37.98 | 69.45 | 53.72 | 22.25 |
| **4** | 23.63 | 39.44 | 31.54 | 11.18 |
| **6** | 20.09 | 33.86 | 26.97 | 9.74 |
| **21** | 3.82 | 7.20 | 5.51 | 2.39 |
| **29** | 5.09 | 1.19 | 3.14 | 2.76 |

**Table 19 Peleg's equation parameters fit for water loss(WL) and sugar gain(SG) and sum of squared difference (SSD) during osmotic dehydration.**

| | **WL** | | | **SG** | | |
|---|---|---|---|---|---|---|
| | K1 | K2 | SSD | K1 | K2 | SSD |
| **Batch A** | 0.87605 | 1.505304 | 0.005204 | 45.13541 | 9.879048 | 0.000296 |
| **Batch B** | 0.875372 | 1.512346 | 0.005015 | 16.59576 | 9.879048 | 0.00024 |

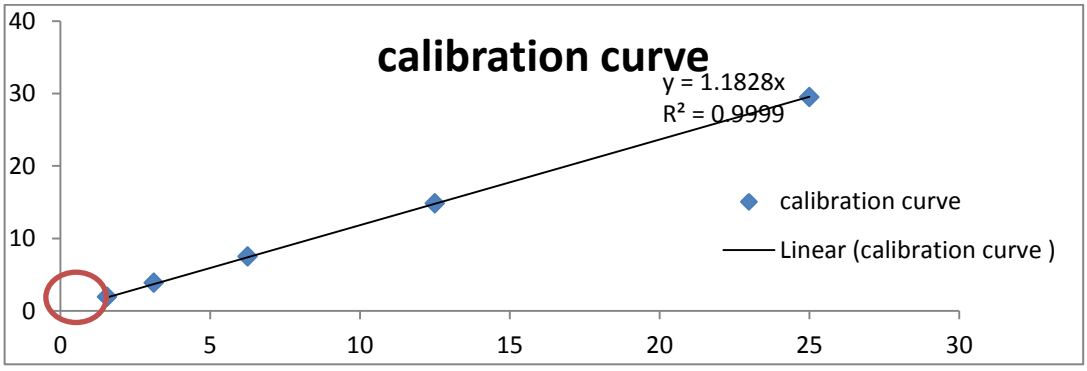
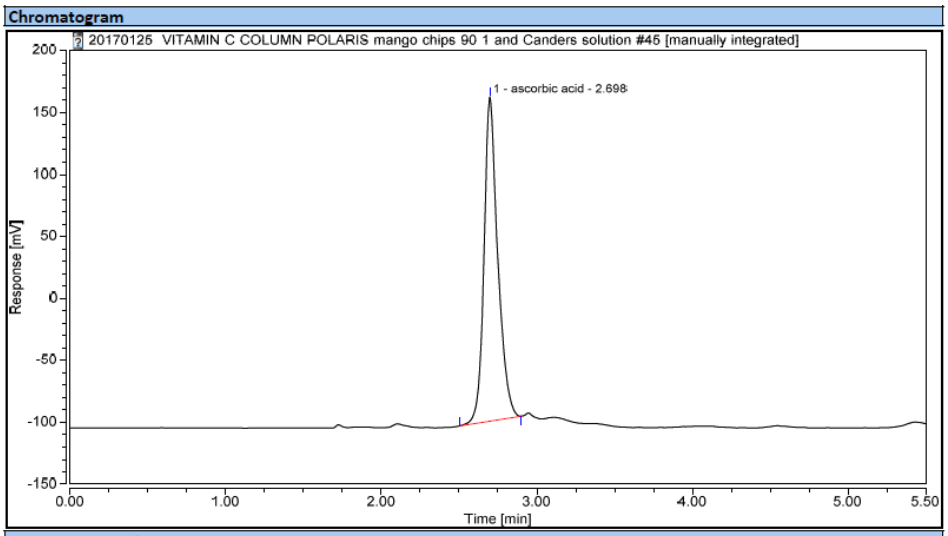


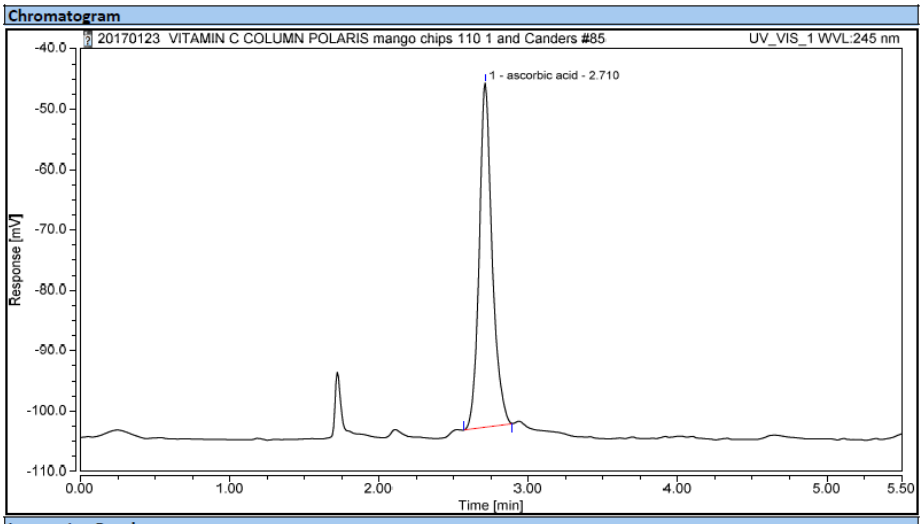

**Figure 18 Calibration curve for vitamin C , the concentration range for vitamin C from 1.56 µg/ml to 25 µg/ml verse the peak area.**




**Integration Results**

| No. | Peak Name | Retention Time min | Area mV*min | Height mV | Relative Area % | Relative Height % | Amount |
|---|---|---|---|---|---|---|---|
| n.a. | MPA | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| 1 | ascorbic acid | 2.698 | 26.758 | 261.717 | 100.00 | 100.00 | n.a. |
| Total: | | | 26.758 | 261.717 | 100.00 | 100.00 | |

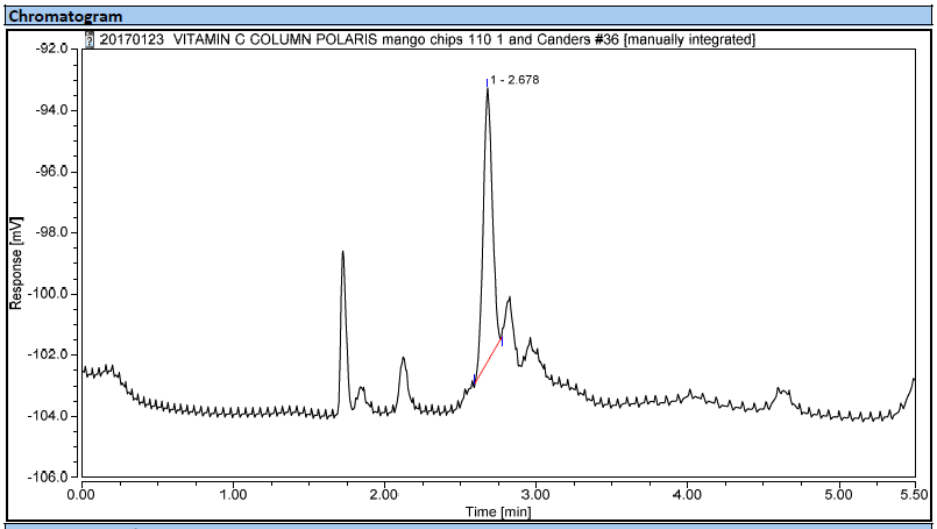
**Figure 19 Chromatogram for HPLC of vitamin C concentration of dry reference mango provided by FQD.**




**Integration Results**

| No. | Peak Name | Retention Time min | Area mV*min | Height mV | Relative Area % | Relative Height % | Amount |
|---|---|---|---|---|---|---|---|
| n.a. | MPA | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| 1 | ascorbic acid | 2.710 | 5.707 | 56.948 | 100.00 | 100.00 | n.a. |
| Total: | | | 5.707 | 56.948 | 100.00 | 100.00 | |

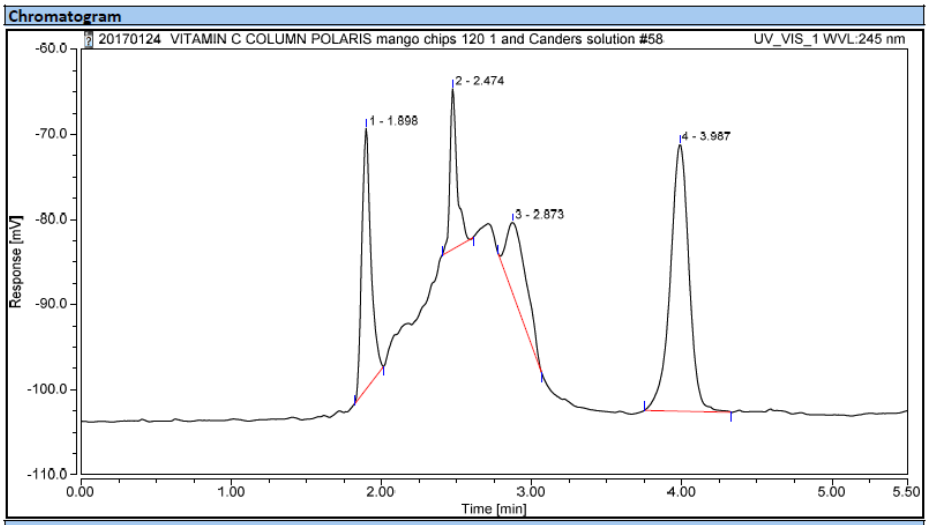
**Figure 20 Chromatogram for HPLC of vitamin C concentration of 2h OD treated mango cubes.**




**Integration Results**

| No. | Peak Name | Retention Time min | Area mV*min | Height mV | Relative Area % | Relative Height % | Amount |
|---|---|---|---|---|---|---|---|
| n.a. | MPA | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| 1 | | 2.678 | 0.617 | 8.954 | 100.00 | 100.00 | n.a. |
| n.a. | ascorbic acid | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| Total: | | | 0.617 | 8.954 | 100.00 | 100.00 | |

**Figure 21 Chromatogram for HPLC of vitamin C concentration of 4h OD treated mango cubes.**




**Integration Results**

| No. | Peak Name | Retention Time min | Area mV*min | Height mV | Relative Area % | Relative Height % | Amount |
|---|---|---|---|---|---|---|---|
| n.a. | MPA | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| 1 | | 1.898 | 2.139 | 30.600 | 23.60 | 34.33 | n.a. |
| 2 | | 2.474 | 1.018 | 18.838 | 11.23 | 21.13 | n.a. |
| n.a. | ascorbic acid | n.a. | n.a. | n.a. | n.a. | n.a. | n.a. |
| 3 | | 2.873 | 1.371 | 8.352 | 15.12 | 9.37 | n.a. |
| 4 | | 3.987 | 4.537 | 31.344 | 50.05 | 35.17 | n.a. |
| Total: | | | 9.065 | 89.135 | 100.00 | 100.00 | |

**Figure 22 Chromatogram for HPLC of vitamin C concentration of OD solution after 0.5h of treated mango cubes.**

**Table 20 Dry matter calculation for water content, soluble solid gain, weight reduction and OD performance index for batch A group 1 of duplicates.**

| Batch A 1 | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **time(h)** | M0 | Mt | cube before | cube after | water content | xw0 | xwt | water loss | xs0 | xst*100 | xst | soluble solid | weight reduction | weight reduction | OD performance |
| **0** | 200.00 | 200.00 | 2.335 | 0.314 | 0.866 | 0.866 | | | 0.176 | | | | | | |
| **0.5** | 209.80 | 148.20 | 1.550 | 0.400 | 0.742 | | 0.742 | 0.341 | | 0.251 | 0.251 | 0.001 | -0.294 | 0.294 | 262.005 |
| **1** | 200.10 | 135.60 | 1.176 | 0.364 | 0.690 | | 0.690 | 0.398 | | 0.291 | 0.291 | 0.021 | -0.322 | 0.322 | 18.756 |
| **2** | 199.10 | 116.90 | 1.257 | 0.424 | 0.663 | | 0.663 | 0.476 | | 0.356 | 0.356 | 0.033 | -0.413 | 0.413 | 14.427 |
| **4** | 164.40 | 84.20 | 1.563 | 0.700 | 0.552 | | 0.552 | 0.583 | | 0.454 | 0.454 | 0.057 | -0.488 | 0.488 | 10.310 |
| **6** | 189.00 | 84.66 | 1.088 | 0.557 | 0.488 | | 0.488 | 0.647 | | 0.498 | 0.498 | 0.047 | -0.552 | 0.552 | 13.743 |
| **21** | 173.00 | 81.10 | 0.776 | 0.388 | 0.500 | | 0.500 | 0.631 | | 0.549 | 0.549 | 0.081 | -0.531 | 0.531 | 7.757 |
| **29** | 170.80 | 84.15 | 1.189 | 0.678 | 0.430 | | 0.430 | 0.654 | | 0.541 | 0.541 | 0.091 | -0.507 | 0.507 | 7.221 |

**Table 21 Dry matter calculation for water content, soluble solid gain, weight reduction and OD performance index for batch A group 2 of duplicates.**

| Batch A 2 | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **time(h)** | M0 | Mt | cube before | cube after | water content | xw0 | xwt | water loss | xs0 | xst*100 | xst | soluble solid | weight reduction | weight reduction | OD performance |
| **0** | 200.00 | 200.00 | 2.017 | 0.285 | 0.859 | 0.859 | | | 0.176 | | | | | | |
| **0.5** | 209.80 | 148.20 | 1.282 | 0.343 | 0.732 | | 0.732 | 0.341 | | 0.251 | 0.251 | 0.001 | -0.294 | 0.294 | 261.911 |
| **1** | 200.10 | 135.60 | 1.106 | 0.346 | 0.687 | | 0.687 | 0.393 | | 0.291 | 0.291 | 0.021 | -0.322 | 0.322 | 18.540 |
| **2** | 199.10 | 116.90 | 1.308 | 0.443 | 0.661 | | 0.661 | 0.470 | | 0.356 | 0.356 | 0.033 | -0.413 | 0.413 | 14.245 |
| **4** | 164.40 | 84.20 | 1.751 | 0.851 | 0.514 | | 0.514 | 0.595 | | 0.454 | 0.454 | 0.057 | -0.488 | 0.488 | 10.535 |
| **6** | 189.00 | 84.66 | 0.998 | 0.513 | 0.486 | | 0.486 | 0.641 | | 0.498 | 0.498 | 0.047 | -0.552 | 0.552 | 13.618 |
| **21** | 173.00 | 81.10 | 1.011 | 0.553 | 0.453 | | 0.453 | 0.646 | | 0.549 | 0.549 | 0.081 | -0.531 | 0.531 | 7.944 |
| **29** | 170.80 | 84.15 | 0.820 | 0.436 | 0.468 | | 0.468 | 0.628 | | 0.541 | 0.541 | 0.091 | -0.507 | 0.507 | 6.936 |

**Table 22 Dry matter calculation for water content, soluble solid gain, weight reduction and OD performance index for batch B group 1 of duplicates.**

| Batch B 1 | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **time(h)** | M0 | Mt | cube before | cube after | water content | xw0 | xwt | water loss | xs0/100 | xst*100 | xst | soluble solid | weight reduction | weight reduction | OD performance |
| **0** | 200.00 | 200.00 | 1.720 | 0.239 | 0.861 | 0.861 | | | 0.145 | | | | | | |
| **0.5** | 254.00 | 182.26 | 1.339 | 0.344 | 0.743 | | 0.743 | 0.328 | | 24.100 | 0.241 | 0.028 | -0.282 | 0.282 | 11.737 |
| **1** | 224.00 | 150.06 | 1.161 | 0.337 | 0.710 | | 0.710 | 0.386 | | 28.600 | 0.286 | 0.047 | -0.330 | 0.330 | 8.275 |
| **2** | 248.20 | 148.77 | 1.200 | 0.429 | 0.643 | | 0.643 | 0.476 | | 33.400 | 0.334 | 0.055 | -0.401 | 0.401 | 8.622 |
| **4** | 229.90 | 121.63 | 0.994 | 0.381 | 0.617 | | 0.617 | 0.535 | | 40.900 | 0.409 | 0.071 | -0.471 | 0.471 | 7.492 |
| **6** | 242.50 | 120.62 | 1.013 | 0.445 | 0.561 | | 0.561 | 0.582 | | 44.700 | 0.447 | 0.077 | -0.503 | 0.503 | 7.527 |
| **21** | 234.40 | 111.07 | 0.657 | 0.373 | 0.432 | | 0.432 | 0.656 | | 54.000 | 0.540 | 0.111 | -0.526 | 0.526 | 5.918 |
| **29** | 239.70 | 107.07 | 0.725 | 0.420 | 0.421 | | 0.421 | 0.673 | | 55.500 | 0.555 | 0.103 | -0.553 | 0.553 | 6.541 |

**Table 23 Dry matter calculation for water content, soluble solid gain, weight reduction and OD performance index for batch B group 2 of duplicates.**

| Batch B 2 | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **time(h)** | M0 | Mt | cube before | cube after | water content | xw0 | xwt | water loss | xs0 | xst | xst | soluble solid | weight reduction | weight reduction | OD performance |
| **0** | 200.00 | 200.00 | 1.818 | 0.271 | 0.851 | 0.851 | | | 0.145 | | | | | | |
| **0.5** | 254.00 | 182.26 | 1.158 | 0.334 | 0.712 | | 0.712 | 0.340 | | 24.100 | 0.241 | 0.028 | -0.282 | 0.282 | 12.185 |
| **1** | 224.00 | 150.06 | 1.276 | 0.390 | 0.694 | | 0.694 | 0.386 | | 28.600 | 0.286 | 0.047 | -0.330 | 0.330 | 8.279 |
| **2** | 248.20 | 148.77 | 1.105 | 0.414 | 0.625 | | 0.625 | 0.476 | | 33.400 | 0.334 | 0.055 | -0.401 | 0.401 | 8.625 |
| **4** | 229.90 | 121.63 | 0.951 | 0.418 | 0.560 | | 0.560 | 0.554 | | 40.900 | 0.409 | 0.071 | -0.471 | 0.471 | 7.767 |
| **6** | 242.50 | 120.62 | 0.857 | 0.406 | 0.526 | | 0.526 | 0.589 | | 44.700 | 0.447 | 0.077 | -0.503 | 0.503 | 7.618 |
| **21** | 234.40 | 111.07 | 0.767 | 0.424 | 0.447 | | 0.447 | 0.639 | | 54.000 | 0.540 | 0.111 | -0.526 | 0.526 | 5.763 |
| **29** | 239.70 | 107.07 | 0.910 | 0.527 | 0.421 | | 0.421 | 0.663 | | 55.500 | 0.555 | 0.103 | -0.553 | 0.553 | 6.442 |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