



**UNIVERSITI PUTRA MALAYSIA**

***DEVELOPMENT OF DRIED RICE NOODLES FORTIFIED WITH  $\beta$ -  
CAROTENE FROM DIFFERENTLY PROCESSED CARROTS (*Daucus  
carota L.*)***

**HAMIDAH BINTI ISMAIL**

**FSTM 2021 20**



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**By**

**HAMIDAH BINTI ISMAIL**

**Thesis Submitted to the School of Graduate Studies, Universiti  
Putra Malaysia, in Fulfilment of the Requirements for the Degree of  
Master of Science**

**November 2019**

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

**DEVELOPMENT OF DRIED RICE NOODLES FORTIFIED WITH  $\beta$ -CAROTENE FROM DIFFERENTLY PROCESSED CARROTS (*Daucus carota* L.)**

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**November 2019**

**Chair : Prof. Sharifah Kharidah Syed Muhammad, PhD**  
**Faculty : Food Science and Technology**

The trend towards consuming foods fortified with fruits and vegetables is escalating due to the increasing consumer awareness on functional foods which include fortified rice noodles. However, food processing such as dried rice noodle making often involves high temperature and high moisture that can affect the stability of fruit and vegetable nutrients such as  $\beta$ -carotene from carrot. This study was conducted to determine the physico-chemical properties of 3 differently processed carrot preparations namely carrot puree (CP) as a control, oven dried carrot powder (ODCP) and spray dried carrot powder (SDCP), the retention and storage stability of  $\beta$ -carotene in rice noodles fortified with these preparations post extrusion, and  $\beta$ -carotene retention as well as the physico-chemical properties of the aforementioned fortified noodles after cooking. Each processed carrot preparation at various levels (5%, 10% or 15%) was mixed with rice flour and tapioca starch blend (80:20) and the moisture content of the mixture (extrusion feed) was adjusted to 30% before extrusion at 80 °C, 100 °C, 100 °C, 100 °C for zones 1, 2, 3 and 4 of the extruder barrel, respectively. The feed rate was set at 20 rpm with a screw speed of 60 rpm. The fortified rice noodles were dried at 30 °C for 16 h and then analyzed for  $\beta$ -carotene content, retention of  $\beta$ -carotene, and colour. In addition, the physico-chemical properties of CP, ODCP and SDCP were also examined. The results showed that SDCP exhibited the lowest moisture content (4.09%) and the lowest hygroscopicity (15.92%) while ODCP had the lowest water activity (0.37), highest  $\beta$ -carotene content (159.48 mg/100g) and highest redness value.  $\beta$ -carotene content of the noodles increased as level of fortification with CP, ODCP and SDCP increased. Dried rice noodle containing 15% ODCP had the highest  $\beta$ -carotene content (5.74 mg/100g) and redness value (21.06) compared to the other fortified dried rice noodles while 5% CP noodle had a significantly higher  $\beta$ -carotene retention (63.45%) post extrusion compared to the other fortified rice noodles. The redness value of the noodles was positively correlated with their  $\beta$ -carotene content. Storage stability and degradation kinetics of  $\beta$ -carotene in the dried rice noodles were assessed

under conditions of with or without light and oxygen at room temperature or 40 °C. Presence of oxygen, exposure to light and elevated temperature of 40 °C were found to increase  $\beta$ -carotene degradation. Half-life of  $\beta$ -carotene in noodles containing CP was the highest (25 - 103 weeks) followed by that in noodles containing SDCP (8 - 19 weeks) and ODCP (8 - 15 weeks) for all levels of fortification and storage conditions. Cooking quality, textural properties, sensory properties,  $\beta$ -carotene retention,  $\beta$ -carotene content and colour of fortified noodles post cooking were also studied. All noodle rehydration and cooking loss values were in the acceptable range of 167.83% - 207.29% and 5.70% - 11.20%, respectively. In terms of textural qualities, rice noodles containing 15% ODCP and 15% SDCP had significantly lower hardness, gumminess and chewiness compared to the other noodles. For sensory evaluation, both of these samples were not significantly different in terms of their flavour, texture and overall acceptability except for their appearance. However, rice noodles containing 15% ODCP exhibited a significantly higher  $\beta$ -carotene retention (51.90%),  $\beta$ -carotene content (2.09 mg/100g) and redness value (8.03) post cooking. In conclusion, 15% ODCP noodle was chosen to be the best fortified rice noodle due to its high  $\beta$ -carotene content, high retention of  $\beta$ -carotene post cooking, most orange in colour and acceptability by the sensory panelists.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia  
sebagai memenuhi keperluan untuk ijazah Master Sains

## **PEMBANGUNAN MEE BERAS KERING YANG DIPERKAYA DENGAN $\beta$ - KAROTEN DARIPADA LOBAK MERAH YANG DI PROSES SECARA BERBEZA**

Oleh

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Trend ke arah pemakanan yang diperkaya dengan buah-buahan dan sayur-sayuran semakin meningkat disebabkan peningkatan kesedaran pengguna terhadap makanan fungsian termasuk mee beras. Walau bagaimanapun, pemprosesan makanan melibatkan suhu tinggi yang mempengaruhi kestabilan nutrien buah dan sayuran seperti  $\beta$ -karoten dari lobak merah. Kajian ini dijalankan untuk menentukan pengekalan  $\beta$ -karoten dalam mee beras yang diperkaya dengan lobak merah. Kajian ini dijalankan untuk menentukan pengekalan  $\beta$ -karoten dalam mee beras yang diperkaya dengan lobak merah. Puri lobak merah (CP), serbuk kering lobak merah (ODCP) dan semburan kering lobak merah (SDCP) secara berasingan dimasukkan ke dalam mee beras yang disemperit. Setiap lobak merah yang diproses di tambah pada tiga tahap berbeza (5%, 10%, 15%) kemudian dicampurkan dengan tepung beras dan tepung ubi kayu (80:20) dan kandungan kelembapan suapan diselaraskan kepada 30% sebelum penyemperitan pada 80 ° C, 100 ° C, 100 ° C, 100 ° C untuk zon 1, 2, 3 dan 4 barel, dengan 20 rpm kadar suapan dan 60 rpm kelajuan skru. Mee dikeringkan pada suhu 30 ° C selama 16 jam dan dinilai untuk pengekalan  $\beta$ -karoten dan warna. Di samping itu, sifat fiziko-kimia CP, ODCP dan SDCP diperiksa. Ini termasuk kandungan kelembapannya, aktiviti air, hygroscopicity, kandungan  $\beta$ -karoten dan warna. Keputusan menunjukkan SDCP menunjukkan kandungan kelembapan terendah (4.09%) dan hygroscopicity terendah (15.92) manakala ODCP menunjukkan aktiviti air terendah (0.37%), kandungan  $\beta$ -carotene tertinggi (159.48 mg/100g) dan nilai kemerahan tertinggi. Kandungan  $\beta$ -karoten dalam mee beras meningkat dengan pertambahan CP, ODCP dan SDCP. Mie beras kering yang mengandungi 15% ODCP mempunyai kandungan  $\beta$ -karoten yang jauh lebih tinggi (5.74 mg / 100g) dan nilai kemerahan (21.06) berbanding dengan mee beras kering lain manakala 5% CP mempunyai pengekalan  $\beta$ -karotena (63.45%) penyemperitan berbanding mee beras lain. Nilai kemerahan mee

berkait secara positif dengan kandungan  $\beta$ -karoten. Kestabilan penyimpanan dan kinetics degradasi  $\beta$ -karoten dalam mee beras kering telah dinilai di bawah keadaan ada atau tanpa cahaya dan oksigen pada suhu bilik atau 40 °C. Kehadiran oksigen, pendedahan kepada cahaya dan suhu tinggi 40 °C didapati meningkatkan degradasi  $\beta$ -karoten. Mee yang mengandungi CP menunjukkan separuh hayat  $\beta$ -karoten tertinggi (25 - 103 minggu) diikuti dengan mee yang mengandungi SDCP (8 - 19 minggu) dan ODCP (8 - 15 minggu) untuk semua keadaan penyimpanan. Kualiti masakan, tekstur, sensori, pengekal  $\beta$ -karoten, kandungan  $\beta$ -karoten dan warna mee yang diperkaya selepas memasak juga dinilai. Semua rehidrasi mee dan nilai kehilangan memasak berada dalam lingkungan 167.83% - 207.29% dan 5.70% - 11.20%. Dari segi kualiti tekstur, mee beras yang mengandungi 15% ODCP dan 15% SDCP mempunyai kekerasan, kelekatan dan kekenyalan yang lebih rendah daripada berbanding dengan mee lain. Untuk penilaian deria, kedua-dua sampel tidak memberikan perbezaan yang signifikan dari segi rasa, tekstur dan penerimaan keseluruhan kecuali penampilan. Walaubagaimanapun, mee beras yang mengandungi 15% ODCP menunjukkan pencapaian post retensi masakan  $\beta$ -karoten yang lebih tinggi (51.90%), kandungan  $\beta$ -karoten (2.09 mg/100g) dan nilai kemerahan (8.03). Kesimpulannya, 15% ODCP dipilih sebagai mee beras yang terbaik kerana kandungan  $\beta$ -karotena yang tinggi, pengekal penyimpanan makanan yang tinggi, warna paling oren dan diterima oleh panelis deria.

## ACKNOWLEDGEMENTS

Alhamdulillah, praise to Allah s.w.t for the strength and perseverance that he has bestowed upon me throughout my postgraduate studies.

I would like to express my deepest gratitude to my beloved supervisor Assoc. Professor Dr. Sharifah Kharidah Syed Muhammad for the time and effort that she has invested in me, her guidance and constructive criticism, as well as for the unlimited faith that she had in me to complete my research project.

I would like to thank my co-supervisor, Assoc. Professor Dr. Roselina Karim for her aid and support in improving this work.

I am deeply grateful to my fellow postgraduate colleagues who have always been there for me whenever I needed a helping hand and listening ear. Your friendship has been essential in my life and of course maintaining my motivation to finish my thesis writing. I am also grateful to the many staff of UPM-BERNAS Research Laboratory for aiding in my research journey.

In addition, I would like to thank my family for their support, patience and understanding throughout the long years of my postgraduate studies. Acknowledgement is also due to MyBrain15 for covering my tuition fees as well as Universiti Putra Malaysia for allowing me the opportunity to increase my knowledge and potential and also for funding me through the Graduate Research Fund (GRF), of which without, my research would not have been possible.



This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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## LIST OF ABBREVIATIONS

$\mu\text{M}$	Micrometer
$a^*$	Redness
ANOVA	Analysis of variance
$a_w$	Water activity
$b^*$	Yellowness
$^{\circ}\text{C}$	Degres celcius
$\text{C}_3\text{H}_6\text{O}$	Acetone
$\text{C}_6\text{H}_{14}$	Hexane
$\text{CH}_3\text{OH}$	Methanol
CP	Carrot puree
cm	Centimeters
DAD	Diode array detector
$\text{DM}_E$	Dry matter extrudate
$\text{DM}_F$	Dry matter feed
FDA	Food and Drugs Association
G	Gram
g/mg	Gram per mili gram
h	Hour
$k_1$	Reaction rate constant
$L^*$	Lightness
LED	Light emitting diode
M	Mol
MC	Moisture content
mm	Milimeter

mg	Miligram
mg/mL	Miligram per milliliter
mL	Mililiter
mL/min	Mililiter per minute
NaCL	Sodium Chloride
Nm	Nanometer
ODCP	Oven dried carrot powder
pH	Power of hydrogen
rpm	Rotation per minute
S	Seconds
SDCP	Spray dried carrot powder
SEM	Scanning electron microscope
t	Time
$t_{1/2}$	Half life
UV	Ultraviolet
VIS	Visible
VPSEM	Variable pressure scanning electron microscope
v/v	Volume per volume
W	Watt
%WI	Weight of sample after one week – weight of sample
W1	Weight of solids in 10 mL
W0	Weight of powder

## CHAPTER 1

### INTRODUCTION

#### 1.1 Background

The rice noodle market size in Europe and Asia Pacific was estimated at USD 1.69 billion in 2014 and is expected to witness a significant growth over the next seven years owing to increase consumer demands from emerging economies including China and India (Grand View Research, 2018). The positive increment is due to urbanization and changing consumer habits which has resulted in fuelling the demand for convenience food products. Milled rice noodles, however, contains a limited amount of vitamins and minerals due to the removal of bran and lost of germ from rice during its milling. Thus, addition of vegetables such as carrots into milled rice noodle could aid in increasing its nutritional value in terms of its carotenoids content. Addition of other natural pigments such as betalains from red beet (Kim *et al.*, 2015) carotenoids from pumpkin (Lee *et al.*, 2002), chlorophylls from spinach (Ahmed *et al.*, 2010) and lycopene from tomato (Nochai and Pongjanta 2013) to wheat noodle is currently gaining great interest. Durum wheat noodles with added natural pigments from spinach, red beet and purple carrot are even now commercially available, for example, from the brand *San Remo*, *Barilla* and *Eatalian Express*.

Carrot (*Daucus carota*) is a root vegetable, widely found with an orange hue due to its carotenoid content. It is also the most essential source of dietary carotenoids in most western countries (Block 1994; Törrönen *et al.*, 1996). Carrot carotenoids comprised primarily of  $\beta$ -carotene at 60-80%, followed by 10% - 40%  $\alpha$ -carotene, 1-5% lutein and 0.1%-1% of other carotenoids (Chen *et al.*, 1995). Carotenoids are important micronutrients for human health as they are precursors of vitamin A (Castenmiller and West 1998). Additionally, carotenoids have other significant roles in biological functions of humans and animals (Van Vliet, 1996). Based on previous epidemiological studies, an increase in dietary intake of carotenoid-rich food and increase blood levels of  $\beta$ -carotene could eventually reduce incidence of certain cancers (Törrönen *et al.*, 1996), age associated macular degeneration, cataracts, coronary heart disease, cardiovascular disease, and several pathological processes (Kohlmeier and Hasting 1995; Biesalski 1997; Kritchevsky 1999).

Purees from fruits and vegetables have been widely produced commercially for direct consumption or as intermediates in the production of food ingredients (Maceiras *et al.*, 2007). The food industry often use carotenoid containing purees such as mangoes (Ahmed *et al.*, 2002), tomatoes (Sánchez-Moreno *et al.*, 2006), and pumpkins (Gliemmo *et al.*, 2009; Provesi *et al.*, 2011) in their preparations. The fruit and vegetable purees are used as they are or in dehydrated forms. Dehydration is a preservation technique in which moisture

content is reduced to level at which the product is relatively chemically stable. Mechanical drying is among the most common drying techniques employed in food processing. The advantages of this technique are uniform drying and better quality of dried product (Prakash *et al.*, 2004). Spray drying is extensively used in commercial production of milk powder, fruit and vegetable powders (Caparino *et al.*, 2012). It is a process where dried particulates are formed from a fluid feed (solution, dispersion or paste) through a spraying technique. It is a continuous process which involves a combination of several stages, that is, atomization, mixing of spray and air, evaporation and product separation (Rè, 1998). It has several advantages including rapid drying which allows the preservation of the thermal sensitive components in food at high percentages. In this study, milled rice noodle which is fortified with carrot carotenoids will be produced using an extruder. To incorporate the carrot carotenoids into the rice noodles, the most important criteria to note is that the carotenoids in the carrot puree or powder must be able to withstand the hot extrusion during the production of the rice noodles. Extrusion is a highly integrated process at low cost. It boasts several advantages such as being energy efficient, generates low waste, gives wider range of raw material selection, and has the ability to produce products with varied texture and shape (Riaz *et al.*, 2009). In addition to this, extrusion is also a viable method for encapsulation of bioactives, in this case,  $\beta$ -carotene from carrots.

## **1.2 Problem statements**

1. Demand for milled rice noodle is increasing but it is low in nutritional value.
2. Drying method will affect carotenoid content and other physico-chemical properties of carrot powder.
3. Extrusion will degrade the carrot carotenoids incorporated into rice noodle.
4. Storage conditions influence the stability of carrot carotenoids in fortified rice noodle
5. Cooking will affect the  $\beta$ -carotene content and quality of fortified rice noodle.

## **1.3 Objectives**

The current study aims:

1. To investigate the effect of incorporating differently processed carrot preparations on the retention of  $\beta$ -carotene in extruded and dried rice noodles.
2. To determine the degradation kinetics of  $\beta$ -carotene and storage stability of the fortified rice noodles under various conditions.
3. To study the effect of cooking on the fortified rice noodles and its consumer acceptance.

## REFERENCES

- Abdul Manan, S.F. Karim, R. Hashim, D.M. & Muhammad, K. Retention and storage of vitamin A and C in extruded native and pregelatinized starches. Master Thesis. Malaysia: Universiti Putra Malaysia
- Abdul Rahman, H., Zakaria, H., Abdul Rahman, A. O., Wahid, M. A., & Mat Top, O. (1994). Model Perusahaan Makanan Bihun. Institut Penyelidikan dan Kemajuan Pertanian Malaysia. 3-7
- Adegunwa, M. O., Bakare, H. A., & Akinola, O. F. (2012). Enrichment of noodles with soy flour and carrot powder. *Nigerian Food Journal*, 30(1), 74-81.
- Agarwal, S., & Rao, A. V. (1998). Tomato lycopene and low density lipoprotein oxidation: a human dietary intervention study. *Lipids*, 33(10), 981-984.
- Ahmed, H. F., & Sayed, H. S. (2010). Cooking quality, sensory evaluation and nutritional value of instant noodles fortified by spinach and artichoke. *Egyptian Journal of Agricultural Research*.
- Ahmed, J., Shivhare, U. S., & Kaur, M. (2002). Thermal colour degradation kinetics of mango puree. *International Journal of Food Properties*, 5(2), 359-366.
- Ajila, C. M., Aalami, M., Leelavathi, K., & Rao, U. P. (2010). Mango peel powder: A potential source of antioxidant and dietary fiber in macaroni preparations. *Innovative Food Science & Emerging Technologies*, 11(1), 219-224.
- Alavi, S.H., Gogoi, B.K., Khan, M., Bowman, B.J., & Rizvi, S.S.H. (1999). Structural properties of protein-stabilized starch-based supercritical fluid extrudates. *Food Research International*, 32(2), 107-118.
- Albanese, D., Cinquanta, L., Cuccurullo, G., & Di Matteo, M. (2013). Effects of microwave and hot-air drying methods on colour,  $\beta$ -carotene and radical scavenging activity of apricots. *International Journal of Food Science & Technology*, 48(6), 1327-1333.
- Andarwulan, N., Kurniasih, D., Apriady, R. A., Rahmat, H., Roto, A. V., & Bolling, B. W. (2012). Polyphenols, carotenoids, and ascorbic acid in underutilized medicinal vegetables. *Journal of Functional Foods*, 4(1), 339-347.
- Anese, M., Falcone, P., Fogliano, V., Nicoli, M. C., & Massini, R. (2002). Effect of equivalent thermal treatments on the color and the antioxidant activity of tomato puree. *Journal of Food Science*, 67(9), 3442-3446.
- Anon (1952). The wealth of India: raw materials, vol 3. Council of Scientific and Industrial Research, New Delhi, pp 20-21.



- AOAC. (2005) Official Methods of Analysis, (18<sup>th</sup> edition). Washington DC, USA: Association of Official Analytical Chemist.
- Arcscott, S. A., & Tanumihardjo, S. A. (2010). Carrots of many colors provide basic nutrition and bioavailable phytochemicals acting as a functional food. *Comprehensive Reviews in Food Science and Food Safety*, 9(2), 223-239.
- Bao, B., & Chang, K. C. (1994). Carrot pulp chemical composition, color, and water-holding capacity as affected by blanching. *Journal of Food Science*, 59(6), 1159-1161.
- Bauernfeind, J. C., Smith, E. G. and Bunnell, R. H. 1958. Colouring fat-base foods with  $\beta$ carotene. *Food Technol.* 12: 527-535.
- Bechoff, a., Dhuique-Mayer, C., Dornier, M., Tomlins, K. I., Boulanger, R., Dufour, D., & Westby, A. (2010). Relationship between the kinetics of  $\beta$ -carotene degradation and formation of norisoprenoids in the storage of dried sweet potato chips. *Food Chemistry*, 121(2), 348-357.
- Benczedi, D., & Blake, A. (1999). Encapsulation and the controlled release of flavours. *Leatherhead Food RA Food Industry Journal*, 2, 36-48.
- Benczedi, D., Bouquerand, P. E., & Steinboeck, E. (2011). *U.S. Patent No. 8,017,060*. Washington, DC: U.S. Patent and Trademark Office.
- Bhattacharya, M., Zee, S. Y., & Corke, H. (1999). Physicochemical properties related to quality of rice noodles. *Cereal chemistry*, 76(6), 861-867.
- Bielsalki HK. (1997). Bioavailability of vitamin A. *European Journal Clinical Nutrition*, 51, Suppl 1:S71-S75.
- Blake, A. (1994). Flavor encapsulation with carbohydrate glasses. *International Food Ingredient*, 3, 30-34.
- Bleiel, J. (2010). Functional foods from the perspective of the consumer: How to make it a success. *International Dairy Journal*, 20(4), 303-306.
- Block G. (1994), Nutrient sources of provitamin A carotenoids in the American diet. *American Journal Epidemiol* 139(3):290-293.
- Boileau, A. C., Merchen, N. R., Wasson, K., Atkinson, C. A., & Erdman Jr, J. W. (1999). Cis-lycopene is more bioavailable than trans-lycopene in vitro and in vivo in lymph-cannulated ferrets. *The Journal of nutrition*, 129(6), 1176-1181.
- Bonnie, T. P. and Choo, Y. M. 1999. Oxidation and thermal degradation of carotenoids. *Journal of Oil Palm Research.*, 2(1): 62–78.



- Boon, C. S., McClements, D. J., Weiss, J., & Decker, E. A. (2010). Factors influencing the chemical stability of carotenoids in foods. *Critical Reviews in Food Science and Nutrition*, 50(6), 515-532.
- Britton G, Khachik F (2009). *Carotenoid in Food. In: Carotenoids: Nutrition and Health*. 5: 3. Britton, G., Liaaen-Jensen, F, Pfander, H (Eds.). Birkhauser Verlag Basel, ISBN 978-3-7643-7500-3. pp. 45-66.
- Britton, G. (1995). Structure and properties of carotenoids in relation to function. *The FASEB Journal*, 9(15), 1551-1558.
- Bui, L. T., & Small, D. M. (2007). The influence of formulation and processing on stability of thiamin in three styles of Asian noodles. *Food chemistry*, 102(4), 1394-1399.
- Bunell, R. H., Discoll, W. and Bauernfeind, J. C. (1958). Colouring water-base foods with  $\beta$ carotene. *Food Technol.* 12: 536-541.
- Bustos, M. C., Perez, G. T., & Leon, A. E. (2015). Structure and quality of pasta enriched with functional ingredients. *Rsc Advances*, 5(39), 30780-30792.
- Cai, Y. Z., & Corke, H. (2000). Production and properties of spray-dried *Amaranthus betacyanin* pigments. *Journal of Food Science*, 65: 1248–1252.
- Caparino, O. A., Tang, J., Nindo, C. I., Sablani, S. S., Powers, J. R., & Fellman, J. K. (2012). Effect of drying methods on the physical properties and microstructures of mango (Philippine “Carabao” var.) powder. *Journal of Food Engineering*, 111(1), 135–148.
- Castenmiller, J. J. M., & West, C. E. (1995). Bioavailability and bioconversion of carotenoids. *Annual Review of nutrition*, 18, 19-38.
- Chakraborty, S., Rao, P. S., & Mishra, H. N. (2015). Effect of combined high pressure–temperature treatments on color and nutritional quality attributes of pineapple (*Ananas comosus* L.) puree. *Innovative food science & emerging technologies*, 28, 10-21.
- Chakraverty, A., Mujumdar, A. S., & Ramaswamy, H. S. (Eds.). (2003). *Handbook of postharvest technology: cereals, fruits, vegetables, tea, and spices* (Vol. 93). CRC Press.
- Chang, C. H., Lin, H. Y., Chang, C. Y., & Liu, Y. C. (2006). Comparisons on the antioxidant properties of fresh, freeze-dried and hot-air-dried tomatoes. *Journal of Food Engineering*, 77(3), 478-485.
- Chen, B. H., Peng, H. Y., & Chen, H. E. (1995). Changes of carotenoids, color, and vitamin A contents during processing of carrot juice. *Journal of Agricultural and Food Chemistry*, 43(7), 1912-1918.

- Chen, H., Rubenthaler, G. L., Leung, H. K., & Baranowski, J. D. (1988). Chemical, physical, and baking properties of apple fiber compared with wheat and oat bran. *Cereal Chem*, 65(3), 244-247.
- Chen, J. P., Tai, C. Y., & Chen, B. H. (2007). Effects of different drying treatments on the stability of carotenoids in Taiwanese mango (*Mangifera indica* L.). *Food Chemistry*, 100(3), 1005-1010.
- Chirife, J., del Pilar Buera, M., & Labuza, T.P. (1996). Water activity, water glass dynamics, and the control of microbiological growth in foods. *Critical Review in Food Science & Nutrition*, 36(5), 465-513.
- Choo, C. L., & Aziz, N. A. A. (2010). Effects of banana flour and  $\beta$ -glucan on the nutritional and sensory evaluation of noodles. *Food Chemistry*, 119(1), 34-40.
- Choy, A. L., Morrison, P. D., Hughes, J. G., Marriott, P. J., & Small, D. M. (2013). Quality and antioxidant properties of instant noodles enhanced with common buckwheat flour. *Journal of Cereal Science*, 57(3), 281-287.
- Cichello, S. A. (2015). Oxygen absorbers in food preservation: a review. *Journal of food science and technology*, 52(4), 1889-1895.
- Codex Alimentarius Commission. (1991). General principles for the addition of essential nutrients to foods. *CAC/GL*, 91987.
- Collins, J. L., & Pangloli, P. (1997). Chemical , Physical and Sensory Attributes of Noodles with Added Sweetpotato and Soy Flour, 62(8), 622–625.
- Coronel-Aguilera, C. P., & San Martín-González, M. F. (2015). Encapsulation of spray dried  $\beta$ -carotene emulsion by fluidized bed coating technology. *LWT-Food Science and Technology*, 62(1), 187-193.
- Đào, V. T. T. (2015). Optimization of drum drying process parameters for pumpkin powder production and its substitution in rice noodles. *Journal of Science*, 3, 149 – 160.
- De Sousa, P. H., Maia, G. A., De Azeredo, H., De Souza Filho, M. D. S., Garruti, D. D. S., & De Freitas, C. A. (2007). Mixed tropical fruit nectars with added energy components. *International Journal of Food Science & Technology*, 42(11), 1290-1296.
- Desobry, S. A., Netto, F. M., & Labuza, T. P. (1997). Comparison of Spray-drying, Drum-drying and Freeze-drying for  $\beta$ -Carotene Encapsulation and Preservation. *Journal of Food Science*, 62(6), 1158-1162.
- Desobry, Stéphane, and Frédéric Debeaufort. "11 Encapsulation of flavors, nutraceuticals, and antibacterials." *Edible coatings and films to improve food quality* (2011): 333.

- Devine, C. M., Connors, M., Bisogni, C. A., & Sobal, J. (1998). Life-course influences on fruit and vegetable trajectories: qualitative analysis of food choices. *Journal of Nutrition Education*, 30(6), 361-370.
- Dolinsky, A., Maletskaya, K., & Snezhkin, Y. (2000). Fruit and vegetable powders production technology on the bees of spray drying and convective drying methods. *Drying Technology*, 18, 747-758.
- Doymaz, İ. (2017). Drying kinetics, rehydration and colour characteristics of convective hot-air drying of carrot slices. *Heat and Mass Transfer*, 53(1), 25-35.
- Eichler, O., Sies, H., & Stahl, W. (2002). Divergent Optimum Levels of Lycopene,  $\beta$ -Carotene and Lutein Protecting Against UVB Irradiation in Human Fibroblasts. *Photochemistry and photobiology*, 75(5), 503-506.
- El-Tinay, A. H., & Chichester, C. O. (1970). Oxidation of. beta.-carotene. Site of initial attack. *The Journal of organic chemistry*, 35(7), 2290-2293.
- Fang, Z., & Bhandari, B. (2010). Encapsulation of polyphenols—a review. *Trends in Food Science & Technology*, 21(10), 510-523.
- Fellows, P. J. (2009). *Food processing technology: principles and practice*. Elsevier.
- Foy, C. J., Passmore, A. P., Vahidassr, M. D., Young, I. S., & Lawson, J. T. (1999). Plasma chain-breaking antioxidants in Alzheimer's disease, vascular dementia and Parkinson's disease. *Qjm*, 92(1), 39-45.
- Fратиани, A., Albanese, D., Mignogna, R., Cinquanta, L., Panfili, G., & Di Matteo, M. (2013). Degradation of carotenoids in apricot (*Prunus armeniaca* L.) during drying process. *Plant foods for human nutrition*, 68(3), 241-246.
- Fu, B. X. (2008). Asian noodles: History, classification, raw materials, and processing. *Food Research International*, 41(9), 888-902.
- Fuhrman, B., Elis, A., & Aviram, M. (1997). Hypocholesterolemic effect of lycopene and  $\beta$ -carotene is related to suppression of cholesterol synthesis and augmentation of LDL receptor activity in macrophages. *Biochemical and biophysical research communications*, 233(3), 658-662.
- Gama, J. J. T., & de Sylos, C. M. (2007). Effect of thermal pasteurization and concentration on carotenoid composition of Brazilian Valencia orange juice. *Food Chemistry*, 100(4), 1686-1690.
- Garcia, L. G. C., Silva, A. H. S., Cunha, P. C., & Damiani, C. (2016). Preparation of gluten-free noodles incorporated of jaboticaba peel flour. *Journal of Food and Nutrition Research*, 4(2), 82-87.

- Gayathri, G. N., Platel, K., Prakash, J., & Srinivasan, K. (2004). Influence of antioxidant spices on the retention of  $\beta$ -carotene in vegetables during domestic cooking processes. *Food Chemistry*, 84(1), 35-43.
- Ge, Y., Sun, A., Ni, Y., & Cai, T. (2001). Study and development of a defatted wheat germ nutritive noodle. *European Food Research and Technology*, 212(3), 344-348.
- Gill, H. S., & Kataria, A. S. (1974). Some biochemical studies in European and Asiatic varieties of carrot (*Daucus carota*). *Current science*.
- Giovannucci, E. (1999). Tomatoes, tomato-based products, lycopene, and cancer: review of the epidemiologic literature. *Journal of the national cancer institute*, 91(4), 317-331.
- Gliemmo, M. F., Latorre, M. E., Gerschenson, L. N., & Campos, C. A. (2009). Colour stability of pumpkin (*Cucurbita moschata*, Duchesne ex Poiret) puree during storage at room temperature: Effect of pH, potassium sorbate, ascorbic acid and packaging material. *LWT - Food Science and Technology*, 42(1), 196–201.
- Gopalan C, Ramasastry BV, Balasubramanian SC (1991) Nutritive value of Indian foods. National Institute of Nutrition, Hyderabad, p 47
- Grand View Research. (2018). Pasta and Noodle Market Size, Analysis Report by Product (Ambient Noodles, Dried Pasta and Noodles, Chilled Pasta and Noodle), By Region (North America, Europe, Asia Pacific, Latin America, MEA), And Segment Forecasts, 2018-2025. Retrieved from <https://www.grandviewresearch.com/industry-analysis/pasta-noodles-market>
- Guiné, R., Sério, S., Correia, P., & Barroca, M. J. (2014). Effect of pre-treatment on some physical-chemical properties of dried carrots. *Journal of Hygienic Engineering and Design*, 6, 187-191.
- Gull, A., Prasad, K., & Kumar, P. (2015). Effect of millet flours and carrot pomace on cooking qualities, color and texture of developed pasta. *LWT-Food Science and Technology*, 63(1), 470-474.
- Gunning, Y. M., Gunning, P. A., Kemsley, E. K., Parker, R., Ring, S. G., Wilson, R. H., & Blake, A. (1999). Factors affecting the release of flavor encapsulated in carbohydrate matrixes. *Journal of agricultural and food chemistry*, 47(12), 5198-5205.
- Gurpreet Kaur, Sharma S, Nagi HP, Ranote PS. Enrichment of pasta with different plant proteins. *Journal of Food Science and Technology*. 2013; 50(5):1000-1005.
- Guzman-Tello, R., & Cheftel, J. C. (1990). Colour loss during extrusion cooking of  $\beta$ -carotene-wheat flour mixes as an indicator of the intensity of thermal and oxidative processing. *International journal of food science & technology*, 25(4), 420-434.

- Handelman, G. J., van Kuijk, F. J., Chatterjee, A., & Krinsky, N. I. (1991). Characterization of products formed during the autoxidation of  $\beta$ -carotene. *Free Radical Biology and Medicine*, 10(6), 427-437.
- Harper, J. M. (2019). Extrusion of foods. CRC press.
- Henry, L. K., Catignani, G. L., & Schwartz, S. J. (1998). Oxidative degradation kinetics of lycopene, lutein, and 9-cis and all-trans  $\beta$ -carotene. *Journal of the American Oil Chemists' Society*, 75(7), 823-829.
- Henry, L. K., Puspitasari-Nienaber, N. L., Jarén-Galán, M., van Breemen, R. B., Catignani, G. L., & Schwartz, S. J. (2000). Effects of ozone and oxygen on the degradation of carotenoids in an aqueous model system. *Journal of Agricultural and Food Chemistry*, 48(10), 5008-5013.
- Heo, S., Jeon, S., & Lee, S. (2014). Utilization of *Lentinus edodes* mushroom  $\beta$ -glucan to enhance the functional properties of gluten-free rice noodles. *LWT-Food Science and Technology*, 55(2), 627-631.
- Hillian, M. (1995). Functional foods: current and future market developments. *Food Technology International Europe*, 25-31.
- Howard, F. D., MacGillivray, J. H., & Yamaguchi, M. (1962). Nutrient composition of fresh California-grown vegetables. *Bulletin of the California Agricultural Experiment Station*, (788).
- Hsu, K. C. (2008). Evaluation of processing qualities of tomato juice induced by thermal and pressure processing. *LWT-Food Science and Technology*, 41(3), 450-459.
- Hughes, D. A., Wright, A. J., Finglas, P. M., Peerless, A. C., Bailey, A. L., Astley, S. B., ... & Southon, S. (1997). The effect of  $\beta$ -carotene supplementation on the immune function of blood monocytes from healthy male nonsmokers. *Journal of Laboratory and Clinical Medicine*, 129(3), 309-317.
- IFIC Foundation (1995) Functional foods: opening the door to better health. *Food Insight*. November/December
- Ishimi, Y., Ohmura, M., Wang, X., Yamaguchi, M., & Ikegami, S. (1999). Inhibition by carotenoids and retinoic acid of osteoclast-like cell formation induced by bone-resorbing agents in vitro. *Journal of clinical biochemistry and nutrition*, 27(3), 113-122.
- Jabbar, S., Abid, M., Hu, B., Wu, T., Hashim, M. M., Lei, S., ... & Zeng, X. (2014). Quality of carrot juice as influenced by blanching and sonication treatments. *LWT-Food Science and Technology*, 55(1), 16-21.

- Jiwan, M. A., Duane, P., O'Sullivan, L., O'Brien, N. M., & Aherne, S. A. (2010). Content and bioaccessibility of carotenoids from organic and non-organic baby foods. *Journal of food composition and analysis*, 23(4), 346-352.
- Ju, Z. Y., Hettiarachchy, N. S., & Rath, N. (2001). Extraction, denaturation and hydrophobic properties of rice flour proteins. *Journal of food science*, 66(2), 229-232.
- Jyothi, N. V. N., Prasanna, P. M., Sakarkar, S. N., Prabha, K. S., Ramaiah, P. S., & Srawan, G. Y. (2010). Microencapsulation techniques, factors influencing encapsulation efficiency. *Journal of microencapsulation*, 27(3), 187-197.
- Kaisangsri, N., Kowalski, R. J., Wijesekara, I., Kerdchoechuen, O., Laohakunjit, N., & Ganjyal, G. M. (2016). Carrot pomace enhances the expansion and nutritional quality of corn starch extrudates. *LWT-Food Science and Technology*, 68, 391-399.
- Karabulut, I., Topcu, A., Duran, A., Turan, S., & Ozturk, B. (2007). Effect of hot air drying and sun drying on color values and  $\beta$ -carotene content of apricot (*Prunus armenica* L.). *LWT-Food Science and Technology*, 40(5), 753-758.
- Kaur, G., Sharma, S., Nagi, H. P. S., & Dar, B. N. (2012). Functional properties of pasta enriched with variable cereal brans. *Journal of food science and technology*, 49(4), 467-474.
- Khachik, F., Beecher, G. R., Goli, M. B., & Lusby, W. R. (1991). Separation, identification, and quantification of carotenoids in fruits, vegetables and human plasma by high performance liquid chromatography. *Pure and Applied Chemistry*, 63(1), 71-80.
- Khoo, H. E., Prasad, K. N., Kong, K. W., Jiang, Y., & Ismail, A. (2011). Carotenoids and their isomers: color pigments in fruits and vegetables. *Molecules*, 16(2), 1710-1738.
- Kilcast, D., & Subramaniam, P. (Eds.). (2000). *The stability and shelf-life of food*. Elsevier.
- Kim, L., Rao, A. V., & Rao, L. G. (2003). Lycopene II—effect on osteoblasts: the carotenoid lycopene stimulates cell proliferation and alkaline phosphatase activity of SaOS-2 cells. *Journal of medicinal food*, 6(2), 79-86.
- Kim, M. J., Park, J. E., Park, S. H., Han, J. S., Choi, J. H., & Lee, H. (2015). Quality characteristics of noodles supplemented with dried *Beta vulgaris* L. root powder. *Journal of the Korean Society of Food Science and Nutrition*, 44(2), 302-306.



- Knockaert, G., Lemmens, L., Van Buggenhout, S., Hendrickx, M., & Van Loey, A. (2012). Changes in  $\beta$ -carotene bioaccessibility and concentration during processing of carrot puree. *Food Chemistry*, 133(1), 60-67.
- Kobayashi, M., & Sakamoto, Y. (1999). Singlet oxygen quenching ability of astaxanthin esters from the green alga *Haematococcus pluvialis*. *Biotechnology Letters*, 21(4), 265-269.
- Koca, N., Burdurlu, H. S., & Karadeniz, F. (2007). Kinetics of colour changes in dehydrated carrots. *Journal of Food Engineering*, 78(2), 449-4
- Kohlmeier, L., & Hastings, S. B. (1995). Epidemiologic evidence of a role of carotenoids in cardiovascular disease prevention. *The American journal of clinical nutrition*, 62(6), 1370S-1376S.
- Kohlmeier, L., Kark, J. D., Gomez-Gracia, E., Martin, B. C., Steck, S. E., Kardinaal, A. F., Martin-Moreno, J. M. (1997). Lycopene and myocardial infarction risk in the EURAMIC Study. *American Journal of Epidemiology*, 146(8), 618-626.
- Kongkachuichai, R., Kounhaweij, A., Chavasit, V., & Charoensiri, R. (2007). Effects of various iron fortificants on sensory acceptability and shelf-life stability of instant noodles. *Food and nutrition bulletin*, 28(2), 165-172.
- Konovalov, V., & Kispert, L. (1999). AM1, INDO/S and optical studies of carbocations of carotenoid molecules. Acid induced isomerization. *Journal of the Chemical Society, Perkin Transactions 2*, (4), 901-910.
- Konovalova, T. A., Kispert, L. D., Polyakov, N. E., & Leshina, T. V. (2000). EPR spin trapping detection of carbon-centered carotenoid and  $\beta$ -ionone radicals. *Free Radical Biology and Medicine*, 28(7), 1030-1038.
- Kramer, A., & Szczesniak, A. S. (Eds.). (2012). *Texture Measurement of Foods: Psychophysical Fundamentals; Sensory, Mechanical, and Chemical Procedures, and their interrelationships*. Springer Science & Business Media.
- Krinsky, N. I. (1998). The Antioxidant and Biological Properties of the Carotenoids a. *Annals of the New York Academy of Sciences*, 854(1), 443-447.
- Krinsky, N. I., & Yeum, K. J. (2003). Carotenoid-radical interactions. *Biochemical and biophysical research communications*, 305(3), 754-760.
- Kritchevsky, S. B. (1999).  $\beta$ -Carotene, carotenoids and the prevention of coronary heart disease. *The journal of nutrition*, 129(1), 5-8.

- Kulthe, A. A., Thorat, S. S., & Khapre, A. P. (2018). Effects of different packaging materials on the sensory characteristics of  $\beta$ -carotene enriched pearl millet based cookies during storage.
- Kumar, S., & Aalbersberg, B. (2006). Nutrient retention in foods after earth-oven cooking compared to other forms of domestic cooking: 2. Vitamins. *Journal of Food Composition and Analysis*, 19(4), 311-320.
- Leach, G., Oliveira, G., & Morais, R. (1998). Spray-drying of *Dunaliella salina* to produce a  $\beta$ -carotene rich powder. *Journal of industrial Microbiology and Biotechnology*, 20(2), 82-85.
- Lee, C. H., Cho, J. K., Lee, S. J., Koh, W., Park, W., & Kim, C. H. (2002). Enhancing  $\beta$ -carotene content in Asian noodles by adding pumpkin powder. *Cereal Chemistry*, 79(4), 593–595.
- Lee, H. S., & Coates, G. A. (2003). Effect of thermal pasteurization on Valencia orange juice color and pigments. *LWT-Food Science and Technology*, 36(1), 153-156.
- Lee, L., Baik, B. K., & Czuchajowska, Z. (1998). Garbanzo bean flour usage in Cantonese noodles. *Journal of food science*, 63(3), 552-558.
- Lee, T. C., Chen, T., Alid, G., & Chichester, C. O. (1978). Stability of vitamin A and pro-vitamin A (carotenoids) in extrusion cooking processing. In *AICHE Symp Ser* (Vol. 74, pp. 172-192).
- Lewicki, P. P. (2006). Design of hot air drying for better foods. *Trends in Food Science & Technology*, 17(4), 153-163.
- Liavoga, A., & Matella, N. J. (2012). Enzymes in quality and processing of tropical and subtropical fruits. *Tropical and subtropical fruits: postharvest physiology, processing and packaging*, 35.
- Lila, M. A. (2004). Anthocyanins and human health: an in vitro investigative approach. *BioMed Research International*, 2004(5), 306-313.
- Lin, C. H., & Chen, B. H. (2005). Stability of carotenoids in tomato juice during storage. *Food Chemistry*, 90(4), 837-846.
- Lin, T. M., Durance, T. D., & Scaman, C. H. (1998). Characterization of vacuum microwave, air and freeze dried carrot slices. *Food Research International*, 31(2), 111-117.
- Loksuwan, J. (2007). Characteristics of microencapsulated  $\beta$ -carotene formed by spray drying with modified tapioca starch, native tapioca starch and maltodextrin. *Food hydrocolloids*, 21(5-6), 928-935.
- Maceiras, R., Álvarez, E., & Cancela, M. A. (2007). Rheological properties of fruit purees: Effect of cooking. *Journal of Food Engineering*, 80(3), 763–769.



- Mader, I. (1964). Beta-carotene: Thermal degradation. *Science*, 144(3618), 533-534.
- Malahayati, N., Muhammad, K., Bakar, J., & Karim, R. (2015). Quality and fortificant retention of rice noodles as affected by flour particle size. *Cereal Chemistry*, 92(2), 211-217.
- Man, D. (2002). *Food Industry briefing series: Shelf life*. Blachwell Science Ltd Ed. UK.
- Martin, K. R., Failla, M. L., & Smith Jr, J. C. (1996).  $\beta$ -Carotene and lutein protect HepG2 human liver cells against oxidant-induced damage. *The Journal of nutrition*, 126(9), 2098-2106.
- Marty, C., & Berset, C. (1990). Factors affecting the thermal degradation of all-trans-  $\beta$ -carotene. *Journal of Agricultural and Food Chemistry*, 38(4), 1063-1067.
- Mearin, M. L., Ivarsson, A., & Dickey, W. (2005). Coeliac disease: is it time for mass screening?. *Best practice & research Clinical gastroenterology*, 19(3), 441-452.
- Ministry of Health, (2018). Recommended Nutrient Intake for Malaysia. Retrieved from <http://nutrition.moh.gov.my/wp-content/uploads/2017/05/FA-Buku-RNI.pdf>
- Miskelly, D. M. (1993). Noodles: a new look at an old food. *Food Australia*, 45(10), 496-500.
- Mordi, R. C. (1993). Mechanism of beta-carotene degradation. *Biochemical journal*, 292(Pt 1), 310.
- Mordi, R. C., Walton, J. C., Burton, G. W., Hughes, L., Keith, I. U., David, L. A., & Douglas, M. J. (1993). Oxidative degradation of  $\beta$ -carotene and  $\beta$ -apo-8'-carotenal. *Tetrahedron*, 49(4), 911-928.
- Mortensen, A. (2006). Carotenoids and other pigments as natural colorants. *Pure and applied chemistry*, 78(8), 1477-1491.
- Mortensen, A., & Skibsted, L. H. (1996). Kinetics of parallel electron transfer from  $\beta$ -carotene to phenoxyl radical and adduct formation between phenoxyl radical and  $\beta$ -carotene. *Free radical research*, 25(6), 515-523.
- Mossel, D. A. A., & Westerdijk, J. (1949). The physiology of microbial spoilage in foods. *Antonie van Leeuwenhoek*, 15(1), 190-202.
- Mudgil, D., Barak, S., & Khatkar, B. S. (2018). Development and characterization of soluble fiber enriched noodles via fortification with partially hydrolyzed guar gum. *Journal of Food Measurement and Characterization*, 12(1), 156-163.

- Muhammad, S. K. S., Amin, H., & Bakar, J. (2015). *U.S. Patent No. 9,028,891*. Washington, DC: U.S. Patent and Trademark Office.
- Muthayya, S., Sugimoto, J. D., Montgomery, S., & Maberly, G. F. (2014). An overview of global rice production, supply, trade, and consumption. *Annals of the new york Academy of Sciences*, 1324(1), 7-14.
- Namitha, K. K., & Negi, P. S. (2010). Chemistry and biotechnology of carotenoids. *Critical reviews in food science and nutrition*, 50(8), 728-760.
- Nath, S., & Satpathy, G. R. (1998). A systematic approach for investigation of spray drying processes. *Drying Technology*, 16(6), 1173-1193. National Academy of Science CoDaH, 1989
- Nawirska, A., Figiel, A., Kucharska, A. Z., Sokół-Łętowska, A., & Biesiada, A. (2009). Drying kinetics and quality parameters of pumpkin slices dehydrated using different methods. *Journal of Food Engineering*, 94(1), 14-20.
- Nhung, D. T. T., Bung, P. N., Ha, N. T., & Phong, T. K. (2010). Changes in lycopene and beta carotene contents in aril and oil of gac fruit during storage. *Food Chemistry*, 121(2), 326-331.
- Nochai, K., & Pongjanta, J. (2013). Physicochemical properties of dried noodle with tomato lycopene supplement. *RMUTP Research Journal*, 1, 211-221.
- Nunes, I. L., & Mercadante, A. Z. (2007). Encapsulation of lycopene using spray-drying and molecular inclusion processes. *Brazilian Archives of Biology and Technology*, 50(5), 893-900.
- Odriozola-Serrano, I., Soliva-Fortuny, R., & Martín-Belloso, O. (2008). Changes of health-related compounds throughout cold storage of tomato juice stabilized by thermal or high intensity pulsed electric field treatments. *Innovative Food Science & Emerging Technologies*, 9(3), 272-279.
- Olson, J. A., & Hayaishi, O. (1965). The enzymatic cleavage of beta-carotene into vitamin A by soluble enzymes of rat liver and intestine. *Proceedings of the National Academy of Sciences of the United States of America*, 54(5), 1364.
- Paiva, S. A., & Russell, R. M. (1999).  $\beta$ -carotene and other carotenoids as antioxidants. *Journal of the American college of nutrition*, 18(5), 426-433.
- Pakhare, K. N., Dagadkhair, A. C., & Udachan, I. S. (2018). Enhancement of Nutritional and Functional Characteristics of Noodles by Fortification

with Protein and Fiber: A Review. *Journal of Pharmacognosy and Phytochemistry*, 7(1), 351-357.

- Panlasigui, L. N., Thomson, L. U., Jenkins, D. J. A., Juliano, B. O., Perez, C. O., & Yiu, S. (1990). Starch digestibility and glycemic response to extruded high amylose and rice noodles. *Trans Nat Acad Sci Technol*, 12, 109-127.
- Paran, E. (2006). Reducing hypertension with tomato lycopene. *Tomatoes, Lycopene and Human Health*, 169-182.
- Park, J. H., Choi, J. E., & Lee, J. H. (2015). Selected physicochemical and consumer preference characteristics of noodles incorporated with sweet pumpkin powder. *Journal of the Korean Society of Food Science and Nutrition*, 44(2), 291-295.
- Parthasarathy, S., Steinberg, D., & Witztum, J. L. (1992). The role of oxidized low-density lipoproteins in the pathogenesis of atherosclerosis. *Annual review of medicine*, 43(1), 219-225.
- Pérez-Conesa, D., García-Alonso, J., García-Valverde, V., Iniesta, M. D., Jacob, K., Sánchez-Siles, L. M., ... & Periago, M. J. (2009). Changes in bioactive compounds and antioxidant activity during homogenization and thermal processing of tomato puree. *Innovative food science & emerging technologies*, 10(2), 179-188.
- Pesek, C. A., & Warthesen, J. J. (1990). Kinetic model for photoisomerization and concomitant photodegradation of beta-carotenes. *Journal of Agricultural and Food Chemistry*, 38(6), 1313-1315.
- Piwińska, M., Wyrwisz, J., Kurek, M., & Wierzbicka, A. (2015). Hydration and physical properties of vacuum-dried durum wheat semolina pasta with high-fiber oat powder. *LWT-Food Science and Technology*, 63(1), 647-653.
- Polyakov, N. E., and Leshina, T. V. (2006). Certain aspects of the reactivity of carotenoids. Redox processes and complexation. *Russ Chem Rev.* 75(12):1049–1064.
- Prakash, S., Jha, S. K., & Datta, N. (2004). Performance evaluation of blanched carrots dried by three different driers. *Journal of Food Engineering*, 62(3), 305–313.
- Priyadarshani, A. M. B., & Jansz, E. R. (2014). A critical review on carotenoid research in Sri Lankan context and its outcomes. *Critical reviews in food science and nutrition*, 54(5), 561-571.
- Provesi, J. G., Dias, C. O., & Amante, E. R. (2011). Changes in carotenoids during processing and storage of pumpkin puree. *Food Chemistry*, 128(1), 195–202.

- Qi, Z. H., & Xu, A. (1999). Starch-based ingredients for flavor encapsulation. *Cereal Foods World*.
- Que, F., Hou, X. L., Wang, G. L., Xu, Z. S., Tan, G. F., Li, T., ... & Xiong, A. S. (2019). Advances in research on the carrot, an important root vegetable in the Apiaceae family. *Horticulture research*, 6(1), 1-15.
- Que, F., Mao, L., Fang, X., & Wu, T. (2008). Comparison of hot air-drying and freeze-drying on the physicochemical properties and antioxidant activities of pumpkin (*Cucurbita moschata* Duch.) flours. *International journal of food science & technology*, 43(7), 1195-1201.
- Rao, A. V., & Rao, L. G. (2007). Carotenoids and human health. *Pharmacological research*, 55(3), 207-216.
- Rao, L. G., Guns, E., & Rao, A. V. (2003). Lycopene: its role in human health and disease. *Agro Food*, 7, 25-30.
- Ratti, C. (2001). Hot air and freeze-drying of high-value foods: a review. *Journal of food engineering*, 49(4), 311-319.
- Ratti, C. (Ed.). (2008). *Advances in food dehydration*. CRC Press.
- Ratti, Cristina. "Freeze and vacuum drying of foods." *Drying technologies in food processing* (2008): 225-251.
- Ray, B., & Bhunia, A. (2013). *Fundamental food microbiology*. CRC press.
- Raymond, N., Heap, J., & Case, S. (2006). The gluten-free diet: An update for health professionals. *Practical Gastroenterology*, 30(9), 67.
- Re, M. I. (1998). Microencapsulation by spray drying. *Drying Technology*, 16(6):1195–1236.
- Reineccius, G. A. (1991). Role of carbohydrates in flavor encapsulation. *J. Dairy Sci.*, 45, 144-146.
- Riaz, M. N., Asif, M., & Ali, R. (2009). Stability of vitamins during extrusion. *Critical Reviews in Food Science and Nutrition*, 49(4), 361-368
- Rocha, G. A., Fávaro-Trindade, C. S., & Grosso, C. R. F. (2012). Microencapsulation of lycopene by spray drying: characterization, stability and application of microcapsules. *Food and Bioprocess Processing*, 90(1), 37-42.
- Rodriguez-Amaya, D. B. (1997). *Carotenoids and food preparation: the retention of provitamin A carotenoids in prepared, processed and stored foods* (pp. 1-93). Arlington, VA: John Snow Incorporated/OMNI Project. Rodriguez-Hernández *et al.*, 2005

- Rodríguez-Roque, M. J., de Ancos, B., Sánchez-Vega, R., Sánchez-Moreno, C., Cano, M. P., Elez-Martínez, P., & Martín-Belloso, O. (2016). Food matrix and processing influence on carotenoid bioaccessibility and lipophilic antioxidant activity of fruit juice-based beverages. *Food & function*, 7(1), 380-389.
- Rogers, D. E. Malouf. R.B.7, Langemeier. J., Gelroth, J. A., and Ranhotra. G. S., (1993). Stability and nutrients contribution of  $\beta$ -carotene added to selected bakery products. *Cereal Chemistry*. (70), 558-561
- Sagar, V. R., & Kumar, P. S. (2010). Recent advances in drying and dehydration of fruits and vegetables: a review. *Journal of food science and technology*, 47(1), 15-26.
- Saifullah, R., Abbas, F. M. A., Yeoh, S. Y., & Azhar, M. E. (2009). Utilization of green banana flour as a functional ingredient in yellow noodle. *International Food Research Journal*, 16(3), 373-379.
- Sánchez-Moreno, C., Plaza, L., De Ancos, B., & Cano, M. P. (2006). Impact of high-pressure and traditional thermal processing of tomato purée on carotenoids, vitamin C and antioxidant activity. *Journal of the Science of Food and Agriculture*, 86(2), 171–179.
- Schieber, A., & Carle, R. (2005). Occurrence of carotenoid cis-isomers in food: technological, analytical, and nutritional implications. *Trends in Food Science & Technology*, 16(9), 416-422.
- Schieber, A., Ullrich, W., & Carle, R. (2000). Characterization of polyphenols in mango puree concentrate by HPLC with diode array and mass spectrometric detection. *Innovative Food Science & Emerging Technologies*, 1(2), 161-166.
- Scita, G. (1992). The stability of  $\beta$ -carotene under different laboratory conditions. *The Journal of Nutritional Biochemistry*, 3(3), 124-128.
- Shaaruddin, S., Mahmood, Z., Ismail, H., Ghazali, H. M., Hamzah, M. Y., & Muhammad, K. (2019). Stability of  $\beta$ -carotene in carrot powder and sugar confection as affected by resistant maltodextrin and octenyl succinate anhydride (OSA) starches. *Journal of food science and technology*, 56(7), 3461-3470.
- Silva, E., Birkenhake, M., Scholten, E., Sagis, L. M. C., & Van der Linden, E. (2013). Controlling rheology and structure of sweet potato starch noodles with high broccoli powder content by hydrocolloids. *Food Hydrocolloids*, 30(1), 42-52.
- Sims, C. A., Balaban, M. O., & Malthews, R. F. (1993). Optimization of carrot juice color and cloud stability. *Journal of Food Science*, 58(5), 1129-1131.
- Singha, P., & Muthukumarappan, K. (2017). Effects of processing conditions on

- the system parameters during single screw extrusion of blend containing apple pomace. *Journal of Food Process Engineering*, 40(4), e12513.
- Singh, G., Singh, B., Sharma, S., & Singh, A. (2017). Development and Storage Study of Maize and Chickpea Based Extruded Snacks. *International Journal of Current Microbiology and Applied Sciences*, 6(10), 4798-4804.
- Siro, I., Kápolna, E., Kápolna, B., & Lugasi, A. (2008). Functional food. Product development, marketing and consumer acceptance—A review. *Appetite*, 51(3), 456-467.
- Sogi, D. S., Siddiq, M., & Dolan, K. D. (2015). Total phenolics, carotenoids and antioxidant properties of Tommy Atkin mango cubes as affected by drying techniques. *LWT-Food Science and Technology*, 62(1), 564-568.
- Soto-Zamora, G., Yahia, E. M., Brecht, J. K., & Gardea, A. (2005). Effects of postharvest hot air treatments on the quality and antioxidant levels in tomato fruit. *LWT-Food Science and Technology*, 38(6), 657-663.
- Stahl, W., & Sies, H. (2001). Effects of carotenoids and retinoids on gap junctional communication. *Biofactors*, 15(2-4), 95-98.
- Steele, R. (2004). *Understanding and measuring the shelf-life of food*. Woodhead Publishing.
- Suknark, K., Lee, J., Eitenmiller, R. R., & Phillips, R. D. (2001). Stability of tocopherols and retinyl palmitate in snack extrudates. *Journal of food science*, 66(6), 897-902.
- Suvarnakuta, P., Devahastin, S., & Mujumdar, A. S. (2005). Drying kinetics and  $\beta$ -carotene degradation in carrot undergoing different drying processes. *Journal of Food Science*, 70(8), s520-s526.
- Törrönen, R., Lehmusaho, M., Häkkinen, S., Hänninen, O., & Mykkänen, H. (1996). Serum  $\beta$ -carotene response to supplementation with raw carrots, carrot juice or purified  $\beta$ -carotene in healthy non-smoking women. *Nutrition Research*, 16(4), 565-575.
- Tsotsas, E., & Mujumdar, A. S. (Eds.). (2007). *Modern drying technology* (Vol. 5). Wiley-VCH.
- Van Vliet, T. (1996). Absorption of beta-carotene and other carotenoids in humans and animal models. *European Journal of Clinical Nutrition*, 50, S32. Wagner & Warthesen, 1995
- Wagner, L. A., & Warthesen, J. J. (1995). Stability of Spray-Dried Encapsulated Carrot Carotenes. *Journal of Food Science*, 60(5), 1048-1053.



- Wani, T. A., Monica, S., Amin, Q. A., Nuzhat, W., & Harleen, K. (2013). Nutritional and organoleptic evaluation of noodles prepared by supplementation with cauliflower leaves. *Asian Journal of Horticulture*, 8(1), 304-312.
- Watzl, B., Bub, A., Briviba, K., & Rechkemmer, G. (2003). Supplementation of a low-carotenoid diet with tomato or carrot juice modulates immune functions in healthy men. *Annals of nutrition and metabolism*, 47(6), 255-261.
- West, C. E., Eilander, A., & van Lieshout, M. (2002). Consequences of revised estimates of carotenoid bioefficacy for dietary control of vitamin A deficiency in developing countries. *The Journal of nutrition*, 132(9), 2920S-2926S.
- Witztum, J. L. (1994). The oxidation hypothesis of atherosclerosis. *The Lancet*, 344(8925), 793-795.
- Wootton-Beard, P. C., & Ryan, L. (2011). Improving public health?: The role of antioxidant-rich fruit and vegetable beverages. *Food Research International*, 44(10), 3135-3148.
- Wootton-Beard, P. C., Moran, A., & Ryan, L. (2011). Stability of the total antioxidant capacity and total polyphenol content of 23 commercially available vegetable juices before and after in vitro digestion measured by FRAP, DPPH, ABTS and Folin–Ciocalteu methods. *Food Research International*, 44(1), 217-224.
- Xiao HW, Gao ZJ, Lin H, Yang WX (2010) Air Impingement drying characteristics and quality of carrot cubes. *J Food Process Eng* 33:899–918
- Yahia, E. M., & Ornelas-Paz, J. D. J. (2010). Chemistry, stability, and biological actions of carotenoids. In *Fruit and vegetable phytochemicals: Chemistry, nutritional value and stability* (pp. 177-222). Wiley-Blackwell, Ames.
- Yamasaki, H. (1990). Gap junctional intercellular communication and carcinogenesis. In *Parallels in Cell to Cell Junctions in Plants and Animals* (pp. 115-127). Springer, Berlin, Heidelberg.
- Ying, D., Cheng, L. J., Chibracq, G., Sanguansri, L., Oiseth, S. K., & Augustin, M. A. (2015). The format of  $\beta$ -carotene delivery affects its stability during extrusion. *LWT-Food Science and Technology*, 60(1), 1-7.
- Ziegler, A., Leffell, D. J., Kunala, S., Sharma, H. W., Gailani, M., Simon, J. A., ... & Bale, A. E. (1993). Mutation hotspots due to sunlight in the p53 gene of nonmelanoma skin cancers. *Proceedings of the National Academy of Sciences*, 90(9), 4216-4220.